

Camouflage by counter-brightness: the blue wings of Morpho dragonflies *Zenithoptera lanei* (Anisoptera: Libellulidae) match the water background

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Abstract

Bright iridescent colours are widespread in several aquatic and terrestrial animal taxa and are usually involved in intraspecific communication and/or predator avoidance. Camouflage by iridescence may be one strategy to avoid predators when the animal exhibits bright colours that match the brightness of its surroundings. Hence, animal structural colouration may have a “brightness matching” or “counter-brightness” function when observed against bright or glossy backgrounds. Here, we addressed the role of such counter-brightness effect of the iridescent wings of the Morpho dragonfly *Zenithoptera lanei* for avoiding detection. We hypothesized that the bright reflectance of the dragonfly wings is cryptic against the bright water surface and the glossy vegetation where they naturally occur, protecting the dragonfly from visually oriented predators, deceiving prey and signalling to conspecifics when desired. We addressed whether (1) the iridescent colours of *Z. lanei* wings function as a visual strategy to reduce their wing detectability by brightness matching the background and (2) the detectability of wings against vegetation and water varies according to the observer. For this, we modelled how conspecifics, dipteran prey and predatory birds see the odonate wings against the vegetation of the Neotropical Savannah and against the water surface where the dragonflies perch. Our results suggest that *Z. lanei* dragonflies can avoid detection by predators, prey and conspecifics when perched on their natural habitats (i.e., ponds) against the bright background of the water surface. Here, we add evidence to the multifunctionality of structural colours in animals and the function of iridescence in camouflage. The bright iridescence and ultraviolet reflective nanostructures of *Z. lanei* wings when coupled with striking behavioural displays may provide a dynamic and safe intraspecific communication channel.

Introduction

Iridescent colours exhibit multiple adaptive functions in animals, not only to ward off competitors (Guillermo-Ferreira et al., 2019) but also to court females (Guillermo-Ferreira et al., 2014; Schultz & Fincke, 2009) or even to avoid predators (Doucet & Meadows, 2009; Kjærsmo et al., 2020). For instance, the iridescent elytra of the Asian jewel beetle *Sternocera aequisignata* suffer less predation attempts when compared to non-iridescent elytra, especially against glossier visual backgrounds (Kjærsmo et al., 2020). The pupae of *Euploea* and *Amauris ochlea* butterflies are striking examples of iridescence as camouflage (Steinbrecht et al., 1985). The pupae of these danaine butterflies exhibit a metallic multilayer structure that reflects the bright background of forest leaves, matching its surroundings and breaking their silhouette (Steinbrecht

et al., 1985). In these cases, one might suggest that iridescent animals adopt a “brightness matching” or a “counter-brightness” strategy for camouflage.

By matching the brightness of the visual background, animals may be able to remain hidden via silhouette disruption and camouflage from predators (Schnell et al., 2018). When iridescent signals are displayed against bright backgrounds, such bright colours may allow animals to avoid predators, a unique case of camouflage dependent on habitat illumination and background contrast (Doucet & Meadows, 2009; Kjærsmo et al., 2020). An analogous visual strategy called “counterillumination” is exhibited by a myriad of mesopelagic taxa (e.g., cephalopods, crustaceans and fishes). These animals may exhibit multilayer angle-dependent reflectors and bioluminescence that match the downwelling light in marine habitats, masking the animal’s silhouette when observed from below (Denton,

1970; Herring, 2002). Other marine animals, such as cuttlefish, can use brightness matching to camouflage on the bottom background when observed from above (Schnell *et al.*, 2018).

Iridescent visual signals may be brighter or duller when associated with specific light conditions and visual backgrounds (Doucet & Meadows, 2009; Kjernsmo *et al.*, 2020). Some damselflies are known to flash bright iridescent wing signals to communicate with conspecifics (Guillermo-Ferreira *et al.*, 2019; Schultz & Fincke, 2009). By alternating the wing orientation towards the receiver under specific visual background and light conditions (e.g., under the direct sunlight of treefall gaps), males of helicopter damselflies *Megaloprepus caerulatus* can maximize the brightness of their iridescent colours and improve perception from females and rivals (Schultz & Fincke, 2009). In other words, animals are usually able to control the brightness of such iridescent colours using body postures and spatiotemporal dynamics to increase or decrease detection (Hutton *et al.*, 2015). Although several odonate species exhibit iridescent wings, ultraviolet (UV) reflecting nanostructures and complex behavioural displays, only one study shows evidence of a possible role of such dynamic colours for camouflage in odonates (Cezário *et al.*, 2021).

Perhaps, one of the most remarkable cases among odonates is the bright iridescent wings of Morpho dragonflies, *Zenithoptera lanei* (Anisoptera: Libellulidae). These dragonflies exhibit a remarkable bright iridescent colouration on the wings, which are UV-bluish on the dorsal surface and dark metallic-red on the ventral surface (Fig. 1; Guillermo-Ferreira *et al.*, 2015). Notably, the iridescent UV-bluish colour of *Z. lanei* wings affects the outcome of male–male agonistic interactions, where males with reduced blue colouration face less aggression from rivals (Guillermo-Ferreira *et al.*, 2015). *Zenithoptera lanei* dragonflies also exhibit a unique behavioural trait among anisopterans. They can close their wings dorsally, like their zygopteran relatives, and open the wings laterally to produce a flickering display (Jurzitza, 1982) (the only other case is the Australian *Cordulephya*). The union of such atypical behaviour, complex ultrastructure (with multilayer interference, nanospheres and thick wax coverage) and unique adaptations (a tracheal system and living tissue in the wing membrane) allow Morpho dragonflies to create a multicomponent and multifunctional wing structure and visual signal (Guillermo-Ferreira *et al.*, 2015, 2017). For instance, not only males use wings to fly, attract females and fight rivals but wings also create a gradient of heat loss from the overheated thorax to the rare wing tracheal system and membrane (Guillermo-Ferreira *et al.*, 2017; Guillermo-Ferreira & Gorb, 2021). At high environmental temperatures, males expose the bright UV-reflective dorsal wing surface when intend to cool down the body temperature and close their wings to expose the dark metallic ventral surface and reduce heat loss by convection (Guillermo-Ferreira & Gorb, 2021).

In their natural habitats (i.e., ponds with exposed water surface in the Neotropical savannah), males of *Z. lanei* might be attacked mainly by tyrant flycatchers near or above the water surface (e.g., Freehling & Johnson, 2018), and prey on mosquitoes, and flies (Corbet, 1999). Since most libellulid dragonflies perch on ponds and often exhibit UV-reflecting colour patterns (see Futahashi, 2020), Harris *et al.* (2011)

hypothesized that the bright UV reflectance of dragonflies could be cryptic against the bright water surface, protecting them from visually oriented predatory birds. If true, theory predicts that *Z. lanei* would match the visual properties (i.e., brightness or colour) of the habitat in which they are found when perceived by a predatory bird but also exhibit traits (e.g., behaviours) that would allow them to reduce their detectability (Stevens & Merilaita, 2009). *Zenithoptera lanei* dragonflies display wing claps, as mentioned above, which consist of a slow opening of the wings and a rapid closing movement (Bick & Bick, 1978; Guillermo-Ferreira, 2021), where the individual alternates the exposure of ventral and dorsal wing surfaces to create a dynamic, angle-dependent iridescent signal (Cezário *et al.*, 2021; Guillermo-Ferreira *et al.*, 2015). The bright wing colours may be undetectable or confusing, enabling *Z. lanei* to choose when and where to signal to conspecifics to avoid unwanted intraspecific interactions (see Hutton *et al.*, 2015). The complex ultrastructure of the bright wings of *Z. lanei* wings may allow these dragonflies to match the background (in particular, the water visual background), where brightness and colour matching would grant them the ability to hide from visually oriented observers.

Therefore, here we addressed whether the iridescent bright UV-bluish and/or the dark metallic-red colours exhibited by the wings of *Z. lanei* males allow them to match the brightness of the background and stay hidden in the habitats they are naturally observed: the Neotropical Savannah vegetation and the water surface. For this, we modelled the visual system of different predatory birds, dipteran prey and conspecifics, as we aimed to answer if: (1) the iridescent colours of *Z. lanei* wings function as a visual strategy to reduce wing detection by brightness matching the background and (2) the detection of wings against vegetation and water varies according to the observer (i.e., predators, prey, and conspecifics). We expected that the bright wings would be cryptic against the glossy vegetation (e.g., Kjernsmo *et al.*, 2020) and/or the water surface (Harris *et al.*, 2011) by brightness matching. To the best of our knowledge, this is the first study addressing the role of iridescence by counter-brightness in terrestrial species that exhibit diffuse reflection on their wings, which allow them to brightness match the background.

Materials and methods

Males of *Z. lanei* ($n = 10$) were collected in Assis, São Paulo state, Brazil, (22°38'S, 50°27' W; altitude 522 m). These dragonflies were euthanized by twisting their heads and had their wings removed with scissors under laboratory conditions. The wings were kept in glass vials until reflectance measurements were carried out.

Reflectance spectra of wings

The spectral data were used to assess the detectability of wings by birds, dipteran prey and conspecifics, against the spectral reflectance of different visual backgrounds. Reflectance spectra of the ventral ($n = 24$) and dorsal ($n = 30$) forewing surfaces in three standard wing cells of 10 *Z. lanei* males were obtained



Figure 1 A *Zenithoptera lanei* male perched in its territory showing the iridescent wings with a bright UV-bluish dorsal surface (a) and a dark metallic-red ventral surface (b). A female perched and exhibited its bluish dorsal wing surface (c) and the darker ventral surface (d) against the Brazilian savannah visual background

with a deuterium and tungsten halogen light source (DH-2000-BAL, Ocean Optics Inc, Dunedin, FL, USA). The spectral range of the light source ranged from 200 to 1100 nm. The light was guided through a 200 μm glass fibre positioned 2 mm above the sample. The reflected light was collected by the same fibre and guided to a spectrometer (HR-4000, Ocean Optics). Spectral data were processed by the Spectral Suite software (Ocean Optics). Electrical noise was smoothed with a span of 0.25 using the pavo package (Maia *et al.*, 2019) in R environment (R Core Team, 2021).

Detectability by predators, prey, and conspecifics

To assess the detectability of *Z. lanei* wings by different receivers, we modelled the visual systems of predatory birds, dipteran prey and conspecifics according to the Receptor Noise-Limited (RNL) model (Vorobyev & Osorio, 1998). The RNL model assumes that hue and saturation (the chromatic components of colours) are processed independently of brightness (the achromatic component) (Renoult *et al.*, 2017). The process of phototransduction is linear, the colour differences are

independent of the intensity of the stimulus and the discriminability threshold of a given visual system is limited by the noise (random fluctuations in the rate that photons arrive at photoreceptors) of the chromatic channel at the photoreceptor level (Renoult *et al.*, 2017; Vorobyev & Osorio, 1998). Hence, we started by quantifying the quantum capture efficiency (Q_i) for each photoreceptor (i) using the formula:

$$Q_i = \int_{300}^{700} R_i(\lambda)S(\lambda)I(\lambda)d\lambda, \quad (1)$$

where $R_i(\lambda)$ is the spectral sensitivity of the photoreceptor i , $S(\lambda)$ is the reflectance spectrum, and $I(\lambda)$ is the illuminant spectrum for each wavelength $d\lambda$. The quantum capture efficiency was calculated in the 300–700 nm spectral range. To model the chromatic discriminability by both ultraviolet sensitive (UVS) and violet sensitive (VS) birds, we used the default settings for averaged tetrachromatic ultraviolet sensitive birds (“avg.uv”) and tetrachromatic violet sensitive birds (“avg.v”). The chromatic discriminability by the dipteran prey visual system was modelled with the spectral sensitivities of the tetrachromatic Housefly *Musca domestica* (λ_{max} values: 360, 460, 490 and 620).

Zenithoptera lanei conspecific visual system was modelled as tetrachromatic, although some odonates exhibit tri- and even pentachromatic visual systems (Bybee et al., 2012; van der Kooi et al., 2021). To model the chromatic discriminability of conspecifics, we customized the visual system of *Z. lanei* based on the spectral sensitivities of the pentachromatic *Sympetrum rubicundulum* (λ_{\max} values: 340, 410, 490, 530 and 620), a closely related libellulid with available spectral sensitivity data (Meinertzhagen et al., 1983). *Sympetrum rubicundulum* exhibits two subgroups of green photoreceptors (λ_{\max} values: 490 and 530). Because of the lack of electrophysiological data for *Z. lanei*, here we assumed only one subgroup of green photoreceptors (λ_{\max} values of 340, 410, 490 and 620). By adopting λ_{\max} 490 instead of 530 as the third photoreceptor type, we assume there would be less overlap with the LWS photoreceptors. Therefore, we expected a lower discriminability in that spectral range, a conservative approach in face of the potential for dragonflies to detect LW (Bybee et al., 2012; Futahashi et al., 2015; van der Kooi et al., 2021). Odonata

photoreceptors and the signals that arise from opponent channels (Kemp et al., 2015), n_i is the relative number of photoreceptor cells, and a and b the spectral data being contrasted. For UVS birds, we used the photoreceptor density 1:2:2:4 (SWS1: SWS2:MWS:LWS) available for the Red-billed *Leiothrix lutea* (Maier & Bowmaker, 1993), while we used 0.12:0.30:0.30:0.26 (SWS1:SWS2:MWS:LWS) of the House Sparrow *Passer domesticus* for the VS birds (Ensminger & Fernández-Juricic, 2014). A signal-to-noise ratio of 0.1 was used for both predators (Olsson et al., 2018). For the dipteran prey visual system, we used a proportion of photoreceptors of 1:2.2:1:2.2 (R7p: R7y:R8p:R8y) (Kirschfeld et al., 1978) and a signal-to-noise ratio of 0.2 (e.g., Outomuro et al., 2017). For the conspecific visual system, we used a proportion of photoreceptor cells of 2:2.5:2.5:1 (UV:SW:MW:LW) (Meinertzhagen et al., 1983) and a signal-to-noise ratio of 0.2 (e.g., Outomuro et al., 2017). Finally, the chromatic (formula (3)) and achromatic (formula (4)) discriminability of each receiver (predatory birds, dipteran prey and conspecifics) were assessed as follows:

$$\Delta S = \sqrt{\frac{(w_1 w_2)^2 (\Delta f_4 - \Delta f_3)^2 + (w_1 w_3)^2 (\Delta f_4 - \Delta f_2)^2 + (w_1 w_4)^2 (\Delta f_3 - \Delta f_2)^2 + (w_2 w_3)^2 (\Delta f_4 - \Delta f_1)^2 + (w_2 w_4)^2 (\Delta f_3 - \Delta f_1)^2 + (w_3 w_4)^2 (\Delta f_2 - \Delta f_1)^2}{((w_1 w_2 w_3)^2 + (w_1 w_2 w_4)^2 + (w_1 w_3 w_4)^2 + (w_2 w_3 w_4)^2)}} \quad (3)$$

species present up to 33 opsin genes and the tetrachromacy is a widespread trait within the order, therefore, the tetrachromatic values assumed here may well represent the diversity of visual systems exhibited by these insects (Bybee et al., 2012; Futahashi et al., 2015; van der Kooi et al., 2021).

For the achromatic channel of the UVS birds, we used data available for the Blue Tit *Cyanistes caeruleus* double-cones. We used the data for chicken *Gallus gallus* double-cones to represent the VS birds. The achromatic discriminability by dipteran prey was based on data for *M. domestica*. For the achromatic discriminability by the customized conspecific visual model, we used the green photoreceptor sensitivity as a vector, since these photoreceptors predict the discriminability of brightness (Giurfa et al., 1996). Spectral sensitivities are available in the pavo package (Maia et al., 2019) for R virtual environment (R Core Team, 2021). A “bluesky” illuminant vector with the scale parameter set at 10.000 (for bright conditions) was used as the illuminant spectrum since *Z. lanei* inhabits open areas of the Brazilian savannah with a high solar radiance.

The perceptual difference between the wing surfaces and the spectra of the visual background was calculated using Weber’s fraction (w_i) formula, considering that the stimuli intensity contributes to the photoreceptor noise:

$$w_i = \sqrt{\frac{v_i^2}{\sqrt{n_i} + Q_a + Q_b}}, \quad (2)$$

where v_i is the signal-to-noise ratio, that is, the ratio between the random fluctuations in the rate that photons arrive at

$$\Delta L = \frac{\Delta f}{w} \quad (4)$$

Chromatic (ΔS) and achromatic (ΔL) contrasts represent the just noticeable differences (JNDs) that a given visual system discerns two stationary visual stimuli in natural settings.

Detectability against the background

We also measured the colour difference between the wing surfaces and the Brazilian savannah (Gawryszewski, 2018) and inland-water visual backgrounds (Kokaly et al., 2013). The Brazilian savannah visual background was obtained by averaging 401 reflectance measures of leaves, leaf litter, tree bark and twigs (Gawryszewski, 2018). The inland-water visual background was obtained from the USGS Spectral Library Version 7 by averaging 12 reflectance measures of shallow water at the marsh edge (Kokaly et al., 2013). To proceed with the detectability analysis, the spectra range of 300–700 nm is necessary. Because the spectra range of the inland-water background was 350–700 nm, we estimated the values between 300 and 349 nm using the imputeTS package in R environment. The imputeTS package is used to estimate missing values in univariate data series (Moritz & Bartz-Beielstein, 2017). The JNDs were obtained using the above-mentioned (3) and (4) formulae.

Statistical analysis

The reflectance spectra of wing surfaces (dorsal and ventral) and visual backgrounds were standardized using the Z-Score

Standardization method. Unlike Min-Max Normalization, Z-Score Standardization does not tend to bring data towards the mean (Becker *et al.*, 1988).

Under natural bright light, the chromatic and achromatic components of two contrasting colours with less than 3.0 JND are unlikely to be discerned (Thurman & Seymoure, 2016). However, because of the lack of behavioural essays for odonate and other visual systems, we assumed a conservative threshold value of 2.0 JND. Therefore, values under this threshold were cryptic against the visual background, while values higher than 2.0 were conspicuous. After we obtained the JND values for each visual system against the Brazilian savannah and water visual background, we performed the Shapiro–Wilk normality test to determine data distribution. We performed a Wilcoxon's Signed Rank test (considering data were not normally distributed) on the JND values with 2.0 set as the test threshold value. The effect size was calculated using the Rank-Biserial Correlation. We also report JND mean and standard deviation for *Z. lanei* dorsal and

ventral wing surfaces. Statistical analyses were all performed in R environment (R Core Team, 2021) using the *jmv* package (The Jamovi Project, 2021).

Results

Results show that conspecifics can discern the chromatic components (i.e., hue and saturation) of the dorsal surface of *Z. lanei* wings against the savannah visual background (Wilcoxon; dorsal: $W = 465$, d.f. = 29, $p < 0.001$; ventral: $W = 378$, d.f. = 26, $p < 0.001$) (Table 1; Fig. 2a), as can dipteran prey (Wilcoxon; dorsal: $W = 465$, d.f. = 29, $p < 0.001$; ventral: $W = 378$, d.f. = 26, $p < 0.001$; Table 1; Fig. 2b). Dragonflies and dipteran prey are unlikely to discern the colours of wing surfaces of *Z. lanei* males against the water surface (Table 1; Fig. 2a,b).

Ultraviolet and violet sensitive predatory birds (Table 1; Fig. 2c,d) are unlikely to discern hue and saturation of the

Table 1 The chromatic (ΔS) and achromatic contrasts (ΔL) resume the just noticeable distance (JND, mean \pm sd) between *Zenithoptera lanei* wings (dorsal and ventral surfaces) and the visual background: the Brazilian savannah and Inland water surface. Results are shown as Wilcoxon's Signed Rank Test (W), degrees of freedom (d.f.) and Rank-Biserial Correlation effect size

Visual model	ΔS					ΔL				
	JND Mean \pm sd	W	d.f.	p	Effect size	Mean \pm sd	W	d.f.	p	Effect size
Conspecific										
Dorsal										
Savannah	3.552 \pm 0.171	465	29	<0.001	1.000	0.514 \pm 0.153	0	29	1.000	-1.000
Inland-water	1.584 \pm 0.171	0	29	1.000	-1.000	0.178 \pm 0.121	0	29	1.000	-1.000
Ventral										
Savannah	3.405 \pm 0.284	378	26	<0.001	1.000	0.243 \pm 0.119	0	26	1.000	-1.000
Inland-water	1.470 \pm 0.271	0	26	1.000	-1.000	0.479 \pm 0.198	0	26	1.000	-1.000
Prey										
Dorsal										
Savannah	3.026 \pm 0.166	465	29	<0.001	1.000	1.322 \pm 0.056	0	29	1.000	-1.000
Inland-water	1.303 \pm 0.169	0	29	1.000	-1.000	0.338 \pm 0.056	0	29	1.000	-1.000
Ventral										
Savannah	3.006 \pm 0.225	378	26	<0.001	1.000	1.064 \pm 0.186	0	26	1.000	-1.000
Inland-water	1.300 \pm 0.230	0	26	1.000	-1.000	0.174 \pm 0.099	0	26	1.000	-1.000
UVS										
predator										
Dorsal										
Savannah	4.585 \pm 0.237	465	29	<0.001	1.000	1.231 \pm 0.225	0	29	1.000	-1.000
Inland-water	2.392 \pm 0.245	462	29	<0.001	0.987	1.134 \pm 0.225	0	29	1.000	-1.000
Ventral										
Savannah	4.085 \pm 0.457	378	26	<0.001	1.000	1.186 \pm 0.248	0	26	1.000	-1.000
Inland-water	1.949 \pm 0.418	184	26	0.552	-0.026	1.089 \pm 0.248	0	26	1.000	-1.000
VS										
predator										
Dorsal										
Savannah	3.575 \pm 0.192	465	29	<0.001	1.000	1.695 \pm 0.277	27	29	1.000	-0.884
Inland-water	2.029 \pm 0.205	258	29	0.306	0.110	1.468 \pm 0.277	0	29	1.000	-1.000
Ventral										
Savannah	3.079 \pm 0.446	377	26	<0.001	0.995	1.554 \pm 0.317	6	26	1.000	-0.968
Inland-water	1.672 \pm 0.392	22	26	1.000	-0.884	1.327 \pm 0.317	0	26	1.000	-1.000

Significant results in bold ($\alpha = 0.05$) show when wings are discerned from the visual background (JND < 2.0) by conspecific dragonflies, dipteran prey and predatory birds (UVS – ultraviolet sensitive and VS – violet sensitive).

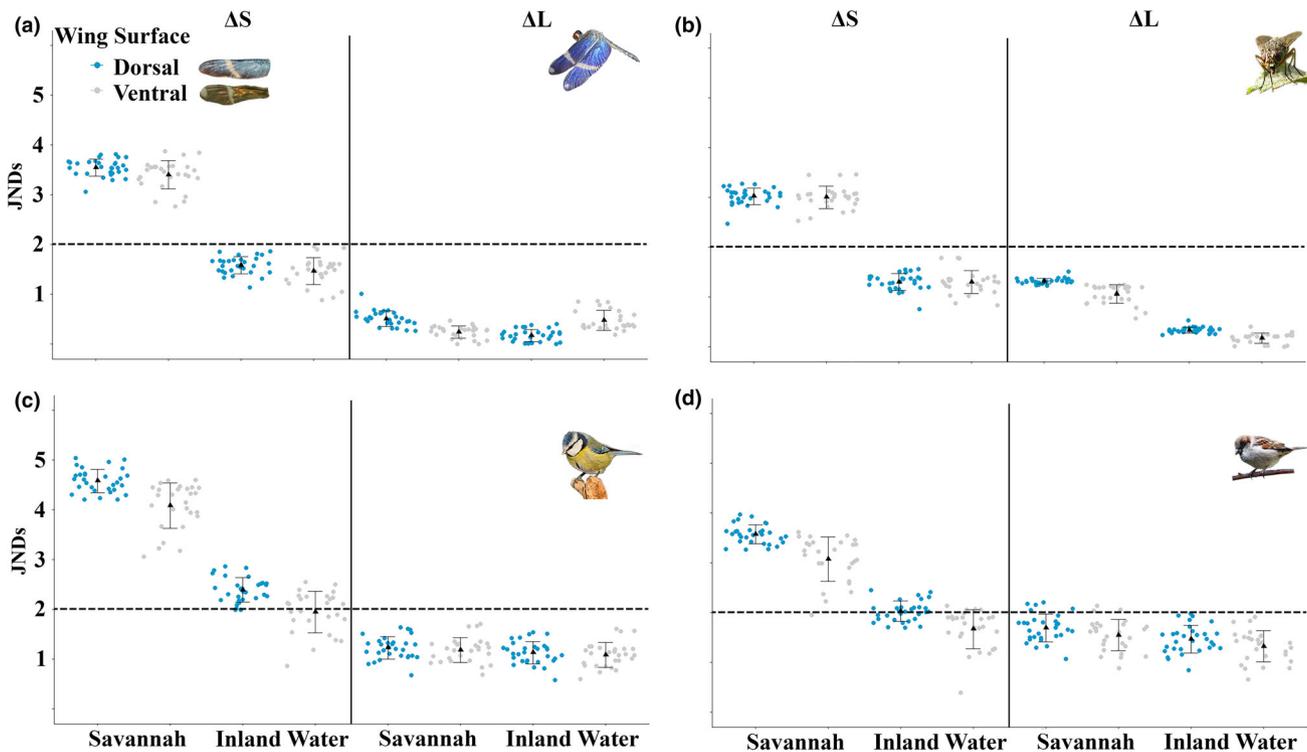


Figure 2 Just noticeable differences (JNDs) values of dorsal and ventral forewing surfaces of *Zenithoptera lanei* males against Brazilian savannah and inland-water visual backgrounds by four types of the visual system. Results are shown according to the visual systems of conspecifics (a), dipteran prey (b), ultraviolet sensitive predatory birds (UVS) (c) and violet sensitive predatory birds (VS) (d). Mean (black triangles), error bars (in black) and individual data points (blue dots = dorsal and grey dots = ventral) for chromatic (ΔS) and achromatic (ΔL) contrasts are shown. The dashed line indicates the threshold of 2.0 JNDs where a visual system discerns a colour patch from the background in bright light conditions

ventral wing surface from the water surface (Wilcoxon; UVS: $W = 184$, d.f. = 26, $p = 0.552$; VS: $W = 22$, d.f. = 26, $p = 1.000$; Table 1; Fig. 2c,d). Nevertheless, UVS birds might be able to perceive the dorsal wing surface against the inland waters (Wilcoxon; $W = 462$, d.f. = 29; $p < 0.001$). Predators might also discern hue and saturation of the dorsal and ventral surfaces against the vegetation (Wilcoxon; UVS: Dorsal: $W = 465$, d.f. = 29, $p < 0.001$; ventral: $W = 378$, d.f. = 26, $p < 0.001$; VS: dorsal: $W = 465$, d.f. = 29, $p < 0.001$; ventral: $W = 377$, d.f. = 26, $p < 0.001$; Table 1; Fig. 2c and 2d). Predators, prey and conspecifics are unable to discern the brightness of wings from the brightness of the water surface and the glossy vegetation (Table 1; Fig. 2).

Discussion

Our results suggest that the iridescent wings of Morpho dragonflies, *Z. lanei*, may camouflage against the water surface using a counter-brightness strategy. By “brightness matching” the background visual properties of inland waters, males may hamper detection by conspecifics, prey, and visually oriented predatory birds, thus avoiding unwanted interactions. Iridescent colours might have a similar function in butterflies and beetles, which may use their bright colours to match the brightness of

the background and disrupt their silhouette (Kjernsmo *et al.*, 2020; Steinbrecht *et al.*, 1985; Vulinec, 1997). Furthermore, besides the multilayer interference and nanostructures that confer iridescent colours to the wings, both dorsal and ventral wing surfaces of *Z. lanei* also exhibit diffuse reflection due to complex wax crystals (Guillermo-Ferreira *et al.*, 2015), which allow *Z. lanei* to match the brightness of the background.

Several marine and terrestrial animals reduce their contrast against specific visual backgrounds and light conditions by matching the luminance of the background (Denton, 1970; Doucet & Meadows, 2009; Herring, 2002; Kjærsmo *et al.*, 2020; Schnell *et al.*, 2018; Steinbrecht *et al.*, 1985). However, in mesopelagic animals, this is often a case of counterillumination by photophores. These marine animals are known to hide from prey and predators by matching the brightness above them, disrupting their silhouette when observed from below (Claes *et al.*, 2010; Denton, 1970; Herring, 2002). The multi-layer structure and the complex wax crystals of *Z. lanei* wings may allow these dragonflies to match the brightness from the inland-water background below, hiding from prey and predators that observe them from above – a possible case of counter-brightness, analogous to counterillumination. To the best of our knowledge, this is the first report of a terrestrial insect that exhibits brightness matching against the water surface.

- the odonate eye. *Organisms, Diversity, and Evolution*, **12**, 241–250.
- Cezário, R. R., Lopez, V. M., Gorb, S., & Guillermo-Ferreira, R. (2021). Dynamic iridescent signals of male copperwing damselflies coupled with wing-clapping displays: The perspective of different receivers. *Biological Journal of the Linnean Society*, **134**, 229–239.
- Claes, J. M., Aksnes, D. L., & Malfet, J. (2010). Phantom hunter of the fjords: Camouflage by counterillumination in a shark (*Etmopterus spinax*). *Journal of Experimental Marine Biology and Ecology*, **388**, 28–32.
- Corbet, P. S. (1999). *Dragonflies: Behaviours and ecology of Odonata*. Harley books.
- Denton, E. J. (1970). Review lecture: On the organization of reflecting surfaces in some marine animals. *Philosophical Transactions of the Royal Society B: Biological Sciences*, **258**, 285–313.
- Doucet, S. M., & Meadows, M. G. (2009). Iridescence: A functional perspective. *Journal of the Royal Society, Interface*, **6**, S115–S132.
- Ensminger, A. L., & Fernández-Juricic, E. (2014). Individual variation in cone photoreceptor density in house sparrows: Implications for between-individual differences in visual resolution and chromatic contrast. *PLoS One*, **9**, e111854.
- Fitzpatrick, J. W. (1980). Foraging behavior of Neotropical tyrant flycatchers. *Condor*, **82**, 43–57.
- Fitzpatrick, J. W. (1981). Search strategies of tyrant flycatchers. *Animal Behavior*, **29**, 810–821.
- Freehling, M., & Johnson, K. (2018). Black Phoebe (*Sayornis nigricans*) predation on Variegated Meadowhawk (*Sympetrum corruptum*). *Argia*, **30**, 17–18.
- Futahashi, R. (2020). Diversity of UV reflection patterns in Odonata. *Frontiers in Ecology and Evolution*, **8**, 201.
- Futahashi, R., Kawahara-Miki, R., Kinoshita, M., Yoshitake, K., Yajima, S., Arikawa, K., & Fukatsu, T. (2015). Extraordinary diversity of visual opsin genes in dragonflies. *Proceedings of the National Academy of Sciences of the United States of America*, **112**, E1247–E1256.
- Gawryszewski, F. M. (2018). Color vision models: Some simulations, a general n-dimensional model, and the colourvision R package. *International Journal of Business Innovation and Research*, **17**, 8159–8170.
- Giurfá, M., Vorobyev, M., Kevan, P., & Menzel, R. (1996). Detection of coloured stimuli by honeybees: Minimum visual angles and receptor specific contrasts. *Journal of Comparative Physiology A*, **178**, 699–709.
- Guillermo-Ferreira, R. (2021). Wing-clapping in *Mnesarete pudica* - a mating call? *Odonatologica*, **50**, 43–54.
- Guillermo-Ferreira, R., Appel, E., Urban, P., Bispo, P. C., & Gorb, S. N. (2017). The unusual tracheal system within the wing membrane of a dragonfly. *Biology Letters*, **13**, 20160960.
- Guillermo-Ferreira, R., Bispo, P. C., Appel, E., Kovalev, A., & Gorb, S. N. (2015). Mechanism of the wing colouration in the dragonfly *Zenithoptera lanei* (Odonata: Libellulidae) and its role in intraspecific communication. *Journal of Insect Physiology*, **81**, 129–136.
- Guillermo-Ferreira, R., Bispo, P. C., Appel, E., Kovalev, A., & Gorb, S. N. (2019). Structural coloration predicts the outcome of male contests in the Amazonian damselfly *Chalcopteryx scintillans* (Odonata: Polythoridae). *Arthropod Structure & Development*, **53**, 100884.
- Guillermo-Ferreira, R., & Gorb, S. N. (2021). Heat-distribution in the body and wings of the morpho dragonfly *Zenithoptera lanei* (Anisoptera: Libellulidae) and a possible mechanism of thermoregulation. *Biological Journal of the Linnean Society*, **133**, 179–186.
- Guillermo-Ferreira, R., Neiss, U. G., Hamada, N., & Bispo, P. C. (2014). Behavior of the Amazonian damselfly *Chalcopteryx scintillans* McLachlan (Zygoptera: Polythoridae) and comments on its morphological distinction from *C. rutilans* (Rambur). *International Journal of Odonatology*, **17**, 251–258.
- Harris, W. E., Forman, D. W., Battell, R. D., Battell, M. T. R., Nelson, A. K., & Brain, P. F. (2011). Odonata colour: More than meets the eye? *International Journal of Odonatology*, **14**, 281–289.
- Herring, P. (2002). *The biology of the deep ocean*. Oxford University Press.
- Hutton, P., Seymoure, B. M., McGraw, K. J., Ligon, R. A., & Simpson, R. K. (2015). Dynamic color communication. *Current Opinion in Behavioral Sciences*, **6**, 41–49.
- Jurzitza, G. (1982). Die Unterscheidung der Männchen von *Zenithoptera fasciata* (Linnaeus, 1758), *Z. viola* Ris, 1910, und *Z. lanei* Santos, 1941 (Anisoptera: Libellulidae). *Odonatologica*, **11**, 331–338.
- Kemp, D. J., Herberstein, M. E., Fleishman, L. J., Endler, J. A., Bennett, A. T. D., Dyer, A. G., Hart, N. S., Marshall, J., & Whiting, M. J. (2015). An integrative framework for the appraisal of coloration in nature. *American Naturalist*, **185**, 705–724.
- Kirschfeld, K., Feiler, R., & Franceschini, N. (1978). A photostable pigment within the rhabdomere of fly photoreceptors no. 7. *Journal of Comparative Physiology A*, **125**, 275–284.
- Kjernsmo, K., Whitney, H. M., Scott-Samuel, N. E., Hall, J. R., Knowles, H., Talas, L., & Cuthill, I. C. (2020). Iridescence as camouflage. *Current Biology*, **30**, 551–555.e3.
- Kokaly, R. F., Couvillion, B. R., Holloway, J. M., Roberts, D. A., Ustin, S. L., Peterson, S. H., Khanna, S., & Piazza, S. C. (2013). Spectroscopic remote sensing of the distribution and persistence of oil from the Deepwater Horizon spill in Barataria Bay marshes. *Remote Sensing of Environment*, **129**, 210–230.
- Labhart T., & Nilsson D.-E. (1995) The dorsal eye of the dragonfly *Sympetrum*: Specializations for prey detection against the blue sky. *Journal of Comparative Physiology A*, **176**(4), 437–453. <https://doi.org/10.1007/bf00196410>
- Lancer, B. H., Evans, B. J. E., & Wiederman, S. D. (2020). The visual neuroecology of Anisoptera. *Current Opinion in Insect Science*, **42**, 14–22.

- Lind, O., & Kelber, A. (2011). The spatial tuning of achromatic and chromatic vision in budgerigars. *Journal of Vision*, **11**(7), 2.
- Lunau, K. (2014). Visual ecology of flies with particular reference to colour vision and colour preferences. *Journal of Comparative Physiology A*, **200**, 497–512.
- Maia, R., Gruson, H., Endler, J. A., & White, T. E. (2019). pavo 2: New tools for the spectral and spatial analysis of colour in R. *Methods in Ecology and Evolution*, **10**, 1097–1107.
- Maier, E. J., & Bowmaker, J. K. (1993). Colour vision in the passeriform bird, *Leiothrix lutea*: Correlation of visual pigment absorbance and oil droplet transmission with spectral sensitivity. *Journal of Comparative Physiology A*, **172**, 295–301.
- Meinertzhagen, I. A., Menzel, R., & Kahle, G. (1983). The identification of spectral receptor types in the retina and lamina of the dragonfly *Sympetrum rubicundulum*. *Journal of Comparative Physiology A*, **151**, 295–310.
- Moritz, S., & Bartz-Beielstein, T. (2017). imputeTS: Time series missing value imputation in R. *R J*, **9**, 207.
- Olsson, P., Lind, O., & Kelber, A. (2018). Chromatic and achromatic vision: Parameter choice and limitations for reliable model predictions. *Behavioral Ecology*, **29**, 273–282.
- Outomuro, D., Söderquist, L., Johansson, F., Ödeen, A., & Nordström, K. (2017). The price of looking sexy: Visual ecology of a three-level predator–prey system. *Functional Ecology*, **31**, 707–718.
- Potier, S., Mitkus, M., & Kelber, A. (2018). High resolution of colour vision, but low contrast sensitivity in a diurnal raptor. *Proceedings of the Royal Society B-Biological Sciences*, **285**, 20181036.
- R Core Team (2021). *R: A language and environment for statistical computing*. R Foundation for Statistical Computing. <https://www.R-project.org/>.
- Renoult, J. P., Kelber, A., & Schaefer, H. M. (2017). Colour spaces in ecology and evolutionary biology. *Biological Reviews*, **92**, 292–315.
- Schnell, A. K., Bellanger, C., Vallortigara, G., & Joze-Alves, C. (2018). Visual asymmetries in cuttlefish during brightness matching for camouflage. *Current Biology*, **28**, R925–R926.
- Schultz, T. D., & Fincke, O. M. (2009). Structural colours create a flashing cue for sexual recognition and male quality in a Neotropical giant damselfly. *Functional Ecology*, **23**, 724–732.
- Steinbrecht, R. A., Mohren, W., Pulker, H. K., & Schneider, D. (1985). Cuticular interference reflectors in the golden pupae of danaine butterflies. *Proceedings of the Royal Society of London. Series B, Biological Sciences*, **226**, 367–390.
- Stevens, M., & Merilaita, S. (2009). Animal camouflage: Current issues and new perspectives. *Philosophical Transactions of the Royal Society of London. Series B*, **364**, 423–427.
- Suvorov, A., Jensen, N. O., Sharkey, C. R., Fujimoto, M. S., Bodily, P., Wightman, H. M. C., Ogden, T. H., Clement, M. J., & Bybee, S. M. (2017). Opsins have evolved under the permanent heterozygote model: Insights from phylotranscriptomics of Odonata. *Molecular Ecology*, **26**, 1306–1322.
- The Jamovi Project (2021). *jamovi*. (Version 1.6) [Computer Software]. <https://www.jamovi.org>
- Thurman, T. J., & Seymoure, B. M. (2016). A bird's eye view of two mimetic tropical butterflies: Coloration matches predator's sensitivity. *Journal of Zoology*, **298**, 159–168.
- van der Kooij, C. J., Stavenga, D. G., Arikawa, K., Belušič, G., & Kelber, A. (2021). Evolution of insect color vision: From spectral sensitivity to visual ecology. *Annual Review of Entomology*, **66**, 435–461.
- Vorobyev, M., & Osorio, D. (1998). Receptor noise as a determinant of colour thresholds. *Proceedings of the Royal Society B-Biological Sciences*, **265**, 351–358.
- Vulinec, K. (1997). Iridescent dung beetles: A different angle. *Florida Entomol*, **80**, 132.