

Iridescent (angle-dependent reflectance) properties of dorsal coloration in *Podarcis muralis* (Laurenti, 1768)

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Abstract. Iridescence is a visual property of those surfaces that change in colour with viewing angle. Iridescence has been rarely reported in reptiles, but some snakes and lizards show this type of coloration. Here we study the effect of different angles of light incidence and observation on the spectrophotometrically assessed reflectance of dorsal coloration in the lizard *Podarcis muralis*. The results demonstrate clear angle dependence of several colour parameters. In particular, different angles of light incidence and observation result in changes in hue of more than 30 nm. This suggests that lizard dorsal coloration may be perceived, depending on viewing geometry, as being of different colours by a wide range of potential observers. Functional implications of iridescence in dorsal coloration are discussed.

Keywords: colour, iridescence, lizard, spectrophotometry.

Iridescence is a visual characteristic attributed to surfaces that change in colour with viewing geometry (Doucet and Meadows, 2009). Iridescence in animals is structurally produced, resulting from the interaction between incident light and nanostructures present in the integument (Kemp, Herberstein and Grether, 2012), and is responsible for some of the most striking animal colours (Meadows et al., 2009). The hallmark of iridescence coloration is that changes in the angle of illumination and/or observation produce changes in perceived hue, brightness (i.e. intensity) or both (Doucet and Meadows, 2009). Iridescent colorations have been attributed several functions relating to sexual selection and communication, but may also have non-communicative functions (e.g. camouflage; reviewed in Doucet and Meadows, 2009).

Iridescence has evolved independently in many taxa and is relatively common in certain invertebrate groups, particularly arthropods (e.g. butterflies, Pegram, Lillo and Rutowski, 2013; Pegram, Han and Rutowski, 2015; hemipters, Fabricant et al., 2014) and molluscs (Brink, van der Berg and Botha, 2002; Mäthger et al., 2009). Examples of iridescent coloration in vertebrates are scarce, with most of the available information concerning fish and birds (Kodric-Brown, 1998; Osorio and Ham, 2002; Marshall et al. 2003; Prum, 2006; Santos et al., 2007). Studies with these clades have shown that iridescent colours can act as social signals, particularly in the context of mate choice (Loyau et al., 2007). Iridescence of these colour signals provides a mechanism for honest signalling (McGraw et al., 2002; Hill et al., 2005), and may enhance signal detection (Loyau et al., 2007). Among reptiles, iridescence appears to be relatively rare but a few lizards and snakes are notorious for their iridescent coloration (Rohrlich and Porter, 1972; Morrison, 1995; reviewed in Doucet and Meadows, 2009). A remarkable example is the snake *Epicrates cenchria*, whose common name, Brazilian rainbow boa, refers to its iridescent coloration.

Information on iridescence in the Lacertidae, a family comprising close to 300 species of

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diurnal and generally colourful lizards, is restricted to a single study (Pérez i de Lanuza and Font, 2014b). However, there are anecdotal reports of angle-dependent changes in the apparent colour of different body regions in some lacertid species. For example, in *Podarcis liolepis* (formerly *P. hispanica*) ventral coloration varies with viewing conditions (EF, personal observation). During extensive field work with a population of *Podarcis muralis* from the Pyrenees we noticed that their dorsal coloration may also change dramatically with changes in viewing geometry. This often complicates censuses and focal animal observations because the perceived colour of a given individual goes from brown to green and back depending on the position of the lizard relative to the observer and the prevailing illumination. The present report is an attempt to confirm and characterize the angle-dependence of *P. muralis* dorsal reflectance using objective colour measurements.

We captured 14 adult *P. muralis* (5 females and 9 males) in the Cerdanya Valley (Eastern Pyrenees) and measured the reflectance of the background dorsal coloration in the centre of the dorsum, avoiding any black spots, with a USB2000 portable spectrophotometer and a PX-2 xenon strobe light source (Ocean Optics, Dunedin, Florida). We used two independent emissive and recording probes mounted on a goniometer specially designed for taking spectrophotometric measurements that allows probes to be rotated to a precise angular position. This device allows us to vary the angle of emissive and recording probes relative to each other and relative to the lizard's body surface to simulate changes due to differences in incident light and/or the position of the observer (for a detailed description see Pérez i de Lanuza and Font, 2014b). We first took a set of measurements placing the emissive and recording probes at a 60° angle from the lizards' dorsal surface, with 60° between the two probes. We then took a second set of measurements placing the probes at 45° from the lizards' surface (resulting in a 90° angle between the emissive and recording probes). In both cases we took two subsets of measurements, first placing the emissive probe facing antero-posteriorly (i.e. head to tail) and then placing it postero-anteriorly (i.e. tail to head). We adopted these setups with the aim to simulate viewing conditions that may be realistic in a natural context for lizards, avoiding extreme angles that might artificially increase reflectance differences.

For analyses, we restricted the obtained reflectance spectra to the 300-700 nm range to encompass the entire spectrum of visual sensitivity described in lacertids (Pérez i de Lanuza and Font, 2014a; Martin et al., 2015). Colours

were characterized according to the three variables traditionally used to explain colour, i.e. hue, chroma and brightness (i.e. intensity) (Montgomery, 2006). As hue we calculated the wavelength location of the peak of maximum reflectance. As brightness we considered the sum of the reflectance at each wavelength throughout the entire spectrum. As chroma we calculated the relative amount of reflectance in the long-wavelength portion of the spectrum (i.e. the sum of the reflectance in the 600-700 nm range divided by the sum of the entire spectrum). We checked the spectral variables for normality and homoscedasticity (for the three variables, Shapiro-Wilk normality test: $P \geq 0.06$; Levene test for equality of variances: $P \geq 0.127$). To test for differences between angle setups, we used repeated measures linear models in R (R Core Team, 2014), using the angle of the emissive and recording probes and the antero-posterior orientation as within-subject factors, and considering the interaction between these two factors.

Figure 1 illustrates the chromatic variation of the dorsal coloration of *P. muralis* depending on viewing geometry, and the mean spectra of the different angle setups. Table 1 summarizes the values of hue, chroma and brightness for the four setups. The spectral location of the reflectance peak (i.e. hue) varies depending on the angle of the emissive and recording probes ($F_{1,13} = 410.21$; $P < 0.001$) and the antero-posterior orientation ($F_{1,13} = 13.82$; $P < 0.01$), but not on the interaction between these two factors ($F_{1,13} = 1.01$; $P = 0.33$). Chroma also changes depending on angle ($F_{1,13} = 98.59$; $P < 0.001$) and orientation ($F_{1,13} = 22.38$; $P < 0.001$), but the interaction is not significant ($F_{1,13} = 1.55$; $P = 0.24$). Brightness varies slightly depending on angle ($F_{1,13} = 6.91$; $P < 0.05$), but not on orientation ($F_{1,13} = 2.92$; $P = 0.11$), or on the interaction between angle and orientation ($F_{1,13} = 1.27$; $P = 0.73$). Therefore, we confirmed that the dorsal coloration of *P. muralis* has iridescent features, especially for the chromatic variables (i.e. hue and chroma). In fact, variation in angle setup results in a change in hue of more than 30 nm (mean \pm SEM considering both probe setups together: 35.75 ± 1.80 nm). This variation probably causes the different appearance of the dorsal coloration of *P. muralis*, at least to human observers, and it should cause the perception of different colours also by lizards (potentially more sensitive to chromatic changes than

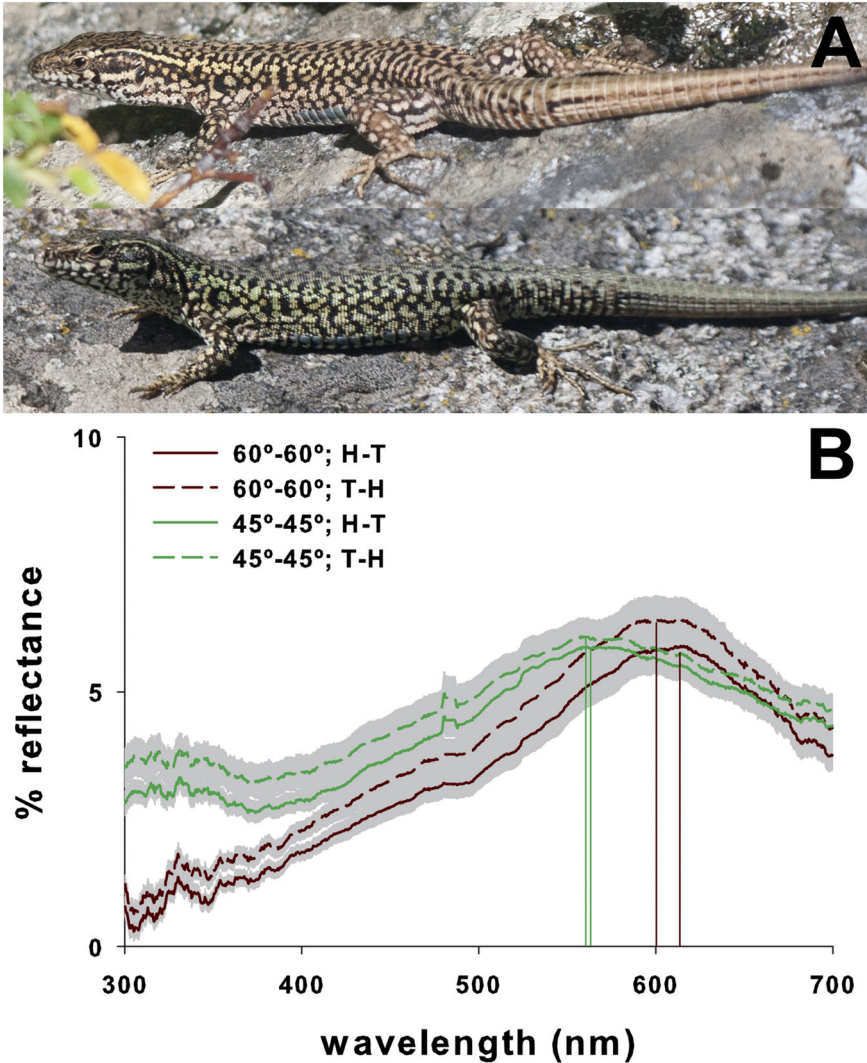


Figure 1. A) Two photographs of the same adult male *Podarcis muralis* taken with different viewing geometries (the photographs have not been manipulated, whether digitally or otherwise, to alter perceived hue). The picture above corresponds to a $<90^\circ$ angle between incident sunlight and the photographic camera (i.e. in the horizontal plane, the camera was located between the sun and the lizard); the picture below corresponds to a $>90^\circ$ angle between incident sunlight and the photographic camera (i.e. the lizard was located between the sun and the camera). B) Reflectance spectra obtained with the different setups. Angles indicate the position of the emissive and recording probes. H-T (i.e. head to tail) indicates spectra obtained with the antero-posterior setup, and T-H (i.e. tail to head) indicates spectra obtained with the postero-anterior setup. Vertical arrows indicate the position of the reflectance peak (i.e. hue) corresponding to each spectrum. Grey bands around the spectra represent error bars (± 1 SEM) for each wavelength. $N = 14$ (5 females, 9 males). This figure is published in colour in the online version.

humans; Pérez i de Lanuza and Font, 2014a; Martin et al., 2015) and other animals (Osorio, Vorobyev and Jones, 1999; Hart, 2001; Macedonia et al., 2009).

Angle-dependent coloration in lizards may have functional implications for thermoregula-

tion or for interactions with conspecifics and heterospecifics. For example, iridescence may theoretically allow signalling animals to be conspicuous to their intended receivers while remaining relatively inconspicuous to predators (Doucet and Meadows, 2009). The blue head of

Table 1. Mean \pm SEM hue, brightness, and chroma (calculated as the sum of the reflectance between 500 and 600 nm divided by the sum of the entire spectrum) for the four recording setups used in this study. H-T (i.e. head to tail) indicates values extracted from spectra obtained with the antero-posterior setup, and T-H (i.e. tail to head) indicates values extracted from spectra obtained with the postero-anterior setup. $N = 14$ (5 females, 9 males).

	60°-60°		45°-45°	
	H-T	T-H	H-T	T-H
Hue (nm)	608.6 \pm 4.8 (558.8-626.9)	600.8 \pm 4.9 (557.1-622.6)	571.0 \pm 4.3 (529.7-587.7)	566.9 \pm 4.1 (529.7-584.7)
Brightness	3965.1 \pm 302.1 (2321.7-5980.3)	4570.5 \pm 311.1 (2853.8-7513.9)	4971.9 \pm 284.2 (3380.0-6509.1)	5419.0 \pm 398.6 (2793.1-8267.8)
Chroma	0.383 \pm 0.012 (0.276-0.454)	0.365 \pm 0.011 (0.274-0.440)	0.303 \pm 0.005 (0.265-0.330)	0.295 \pm 0.005 (0.255-0.331)

male *Lacerta schreiberi* is iridescent, enhancing signal detection by conspecifics which observe signallers laterally, but simultaneously decreasing the detectability to avian predators, which more often view lizards from above (Pérez i de Lanuza and Font, 2014b). A similar explanation is unlikely to apply to the dorsal coloration of *Podarcis muralis*. As *P. muralis* often perch on vertical surfaces (e.g. rock walls), conspecifics and predators may, depending on their relative positions, view the lizards' dorsal surface from a similar perspective. However, iridescence may afford some type of visual camouflage, for example by hindering the acquisition of a search image by potential predators (Bond and Riley, 1991). There is evidence that avian predators become less efficient feeders when they have to look for several types of prey at once (Dukas and Kamil, 2001), and a prey item that changes its perceived hue with viewing geometry may make it more difficult for predators to form a search image. Avian and human visual systems differ in important ways (Cuthill et al., 2000) but a difference of 30 nm should affect hue perception in both cases (Lind, Chavez and Kelber, 2014; Olsson, Lind and Kelber, 2015). Alternatively, it is possible that the iridescence of the dorsal coloration of *P. muralis* has no biological function and is simply a non-selected by-product of the structural properties of the integumental chromatophores or of the surface of the dorsal scales that alter reflectance in this way. However, our results give new evidence that iridescence is not exclusive of insects and

birds, and is in fact also present in lizards as suggested by previous work (Rohrlich and Porter, 1972; Morrison, 1995; Pérez i de Lanuza and Font, 2014b). This peculiarity of lizard coloration should be taken into account when comparing colour parameters of studies using spectrophotometry with different angle geometries.

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References

- Bond, A.B., Riley, D.A. (1991): Searching image in the pigeon: a test of three hypothetical mechanisms. *Ethology* **87**: 203-224.
- Brink, D.J., van der Berg, N.G., Botha, A.J. (2002): Iridescent colors on seashells: an optical and structural investigation of *Helcion pruinosus*. *Appl. Opt.* **41**: 717-722.
- Cuthill, I.C., Partridge, J.C., Bennett, A.T.D., Church, S.C., Hart, N.S., Hunt, S. (2000): Ultraviolet vision in birds. *Adv. Stud. Behav.* **29**: 159-214.
- Doucet, S.M., Meadows, M.G. (2009): Iridescence: a functional perspective. *J. R. Soc. Interface* **6**: S115-S132.
- Dukas, R., Kamil, A. (2001): Limited attention: the constraint underlining search image. *Behav. Ecol.* **12**: 192-199.
- Fabricant, S.A., Exnerová, A., Ježová, D., Štys, P. (2014): Scared by shiny? The value of iridescence in aposomatic

- signalling of the hibiscus harlequin bug. *Anim. Behav.* **90**: 315-325.
- Hart, N.S. (2001): The visual ecology of avian photoreceptors. *Prog. Retin. Eye Res.* **20**: 675-703.
- Hill, G.E., Doucet, S.M., Buchholz, R. (2005): The effect of coccidial infection on iridescent plumage coloration in wild turkeys. *Anim. Behav.* **69**: 387-394.
- Kemp, D.J., Herberstein, M.E., Grether, G.F. (2012): Unraveling the true complexity of costly color signaling. *Behav. Ecol.* **23**: 233-236.
- Kodric-Brown, A. (1998): Sexual dichromatism and temporary color changes in the reproduction of fishes. *Am. Zool.* **38**: 70-81.
- Lind, O., Chavez, J., Kelber, A. (2014): The contribution of single and double cones to spectral sensitivity in budgerigars during changing light conditions. *J. Comp. Physiol. A* **200**: 197-207.
- Loyau, A., Gomez, D., Moureau, B.T., Théry, M., Hart, N.S., Saint Jalme, M., Bennett, A.T.D., Sorci, G. (2007): Iridescent structurally based coloration of eyespots correlates with mating success in the peacock. *Behav. Ecol.* **18**: 1123-1131.
- Macedonia, J.M., Lappin, A.K., Loew, E.R., McGuire, J.A., Hamilton, P.S., Plasman, M., Brandt, Y., Lemos-Espinal, J.A., Kemp, D.J. (2009): Conspicuousness of Dickerson's collared lizard (*Crotaphytus dickersonae*) through the eyes of conspecifics and predators. *Biol. J. Linn. Soc.* **97**: 749-765.
- Marshall, N.J., Jennings, K., McFarland, W.N., Loew, E.R., Losey, G.S. (2003): Visual biology of Hawaiian coral reef fishes. II. Colors of Hawaiian coral reef fish. *Copeia* **2003**: 455-466.
- Martin, M., Le Galliard, J.F., Meylan, S., Loew, E.R. (2015): The importance of short and near infrared wavelength sensitivity for visual discrimination in two species of lacertid lizards. *J. Exp. Biol.* **218**: 458-465.
- Mäthger, L.M., Denton, E.J., Marshall, N.J., Hanlon, R.T. (2009): Mechanisms and behavioral functions of structural coloration in cephalopods. *J. R. Soc.* **6**: S149-S163.
- McGraw, K.J., Mackillop, E.A., Dale, J., Hauber, M.E. (2002): Different colors reveal different information: how nutritional stress affects the expression of melanin and structurally based ornamental plumage. *J. Exp. Biol.* **205**: 3747-3755.
- Meadows, M.G., Butler, M.W., Morehouse, N.I., Taylor, L.A., Toomey, M.B., McGraw, K.J., Rutowski, R.L. (2009): Iridescence: views from many angles. *J. R. Soc. Interface* **6**: S107-S113.
- Montgomerie, R. (2006): Analyzing colors. In: *Bird Coloration, Vol. 1: Mechanisms and Measurements*, pp. 90-147. Hill, G.E., McGraw, K.J., Eds, Harvard University Press, Cambridge.
- Morrison, R.L. (1995): A transmission electron microscopic (TEM) method for determining structural colors reflected by lizard iridophores. *Pigm. Cell. Res.* **8**: 28-36.
- Olsson, P., Lind, O., Kelber, A. (2015): Bird colour vision: behavioural thresholds reveal receptor noise. *J. Exp. Biol.* **218**: 184-193.
- Osorio, D., Ham, A.D. (2002): Spectral reflectance and directional properties of structural coloration in bird plumage. *J. Exp. Biol.* **205**: 2017-2027.
- Osorio, D., Vorobyev, M., Jones, C.D. (1999): Colour vision of domestic chicks. *J. Exp. Biol.* **202**: 2951-2959.
- Pegram, K.V., Han, H.A., Rutowski, R.L. (2015): Warning signal efficacy: assessing the effects of color, iridescence, and time of day in the field. *Ethology* **121**: 861-873.
- Pegram, K.V., Lillo, M.J., Rutowski, R.L. (2013): Iridescent blue and orange components contribute to the recognition of a multicomponent warning signal. *Behaviour* **150**: 321-336.
- Pérez i de Lanuza, G., Font, E. (2014a): Ultraviolet vision in lacertid lizards: evidence from retinal structure, eye transmittance, SWS1 visual pigment genes, and behaviour. *J. Exp. Biol.* **217**: 2899-2909.
- Pérez i de Lanuza, G., Font, E. (2014b): Now you see me, now you don't: iridescence increases the efficacy of lizard chromatic signals. *Naturwissenschaften* **101**: 831-837.
- Prum, R.O. (2006): Anatomy, physics, and evolution of avian structural colors. In: *Bird Coloration, Vol. 1: Mechanisms and Measurements*, pp. 295-355. Hill, G.E., McGraw, K.J., Eds, Harvard University Press, Cambridge.
- R Core Team (2014): R: a Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria. Available at <http://www.R-project.org/>.
- Rohrlich, S.T., Porter, K.R. (1972): Fine structural observations relating to the production of color by the iridophores of a lizard, *Anolis carolinensis*. *J. Cell. Biol.* **53**: 38-52.
- Santos, S.I.C.O., De Neve, L., Lumeij, J.T., Förschler, M.I. (2007): Strong effects of various incidence and observation angles on spectrometric assessment of plumage colouration in birds. *Behav. Ecol. Sociobiol.* **61**: 1499-1506.

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