At face value: Analyzing species and sex differences in facial colorations of two co-occurring species of jumping spiders using microspectrophotometry

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INTRODUCTION

In nature, visual signals may communicate important information such as species identity. Studies have found that organisms from diverse lineages sometimes signal such information through facial color, pattern, and ornamentations[1,2]. To investigate this concept, we studied differences in facial coloration of two co-occurring jumping spiders: *Habronattus orbus* and *Habronattus decorus* (Fig. 1).

Figure 1. Co-occurring jumping spiders show different facial patterns and colorations. From left to right, top to bottom: male *H. decorus,* female *H. decorus,* male *H. orbus,* and female *H. orbus***.** PC: Thomas Shahan

Jumping spiders act as an excellent model to investigate this concept in invertebrates due to their color vision capabilities, high visual acuity, and diversity of facial coloration exhibited by male spiders.

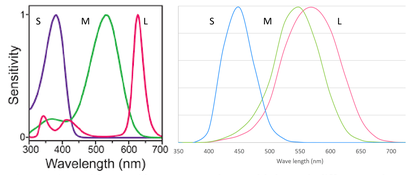


Figure 2. Jumping spiders and humans see the world through different colors. Jumping spider color vision[3] (left) ranges between 300nm-700nm and has peak sensitivities in the UV, green, and red wavelengths. Human visual range[4] (right) falls between 400nm-700nm and has peaks in the blue, green, and red wavelengths. Labels S, M, and L indicate short-, medium-, and long-wavelengths.

We previously found quantitative differences in facial color between sexes and species in two related, co-occurring jumping spider species[6].

We hypothesized that spiders of difference sex and species use differences in facial coloration as a visual courtship signal.

- We aimed to characterize differences between facial colorations in males and females in *H. decorus* and *H. orbus* using microspectrophotometry
- We predict that (i) facial colorations will differ significantly both by sex and by species, and (ii) male faces exhibit greater color diversity between the two species than female faces.

METHODS

Specimen Collection: *Habronattus decorus* and *H. orbus* jumping spiders were collected from the Edge of Appalachia Nature Preserve, died naturally in lab, and were preserved in individual vials at -20°C to 80°C. All specimen were collected between 2020 and 2022.

Data Sampling: Reflectance data was collected using a CRAIC Minerva microspectrophotometer. Six spiders were sampled from each sex and focal species. Each spider was measured at 6 different focal points (Fig. 2) with two replicant measurements at each point. Facial symmetry was assumed, so focal points were all located on the right side of the face.

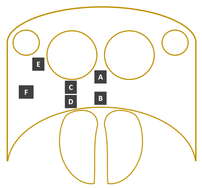


Fig 3. Example spider face with focal regions mapped.

Data Analysis: Spectral data were modeled into the *Habronattus* color space and visualized using the R coding package 'pavo'[5]. Spectral sensitivities were based on previous measurements[3].

RESULTS

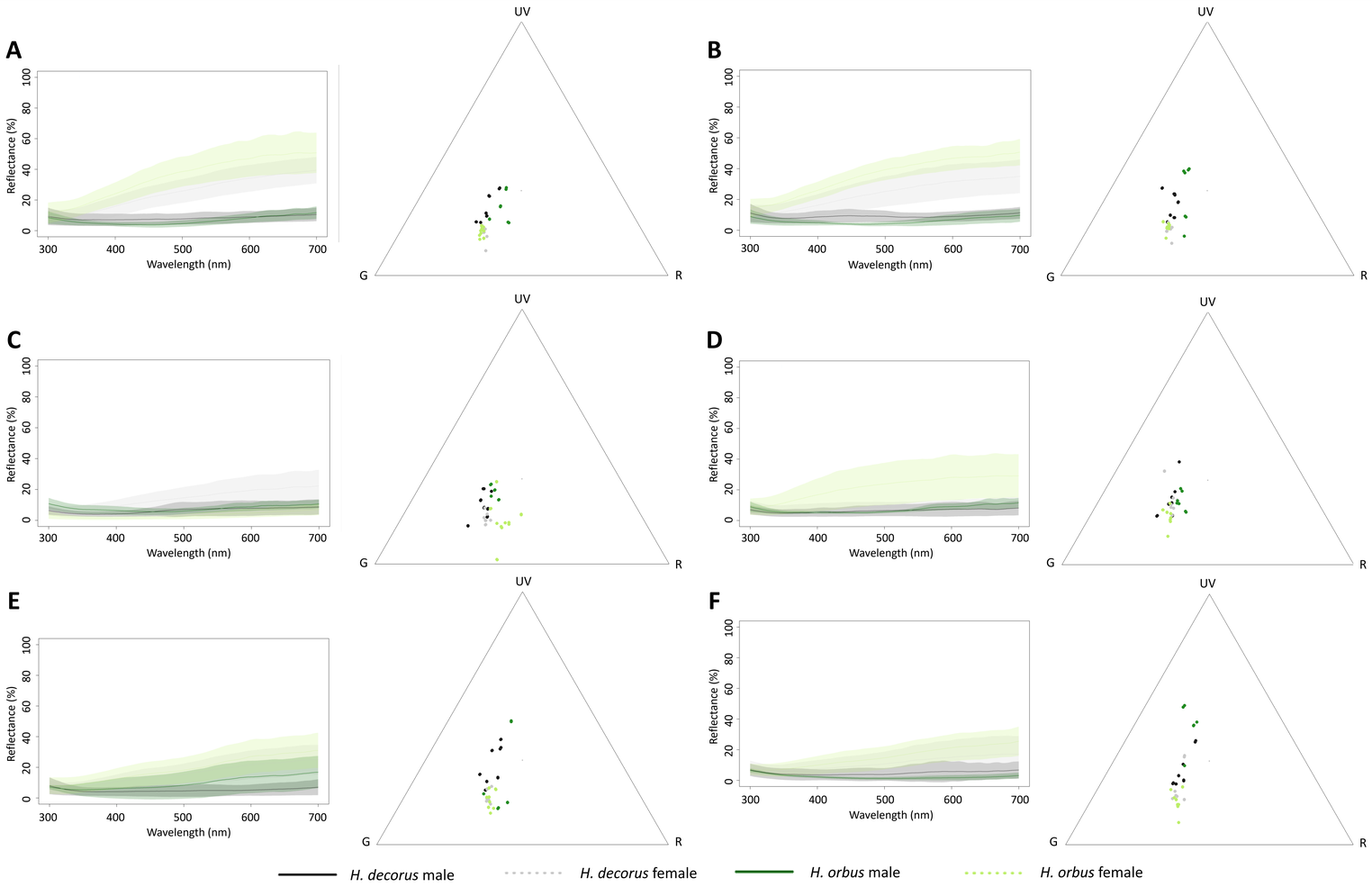


Fig 4. Reflectance spectra (left) and trichromatic color space plots (right) show some differences in facial coloration. Aggregate reflectance spectra graphs and trichromatic color space plots with data from each measured focal point (A-F) from each sex and species. Central lines on the reflectance spectra graphs show the average reflectance for each species and sex, with the shaded regions surrounding those lines showing the variance of each group.

Reflectance Spectra Graph

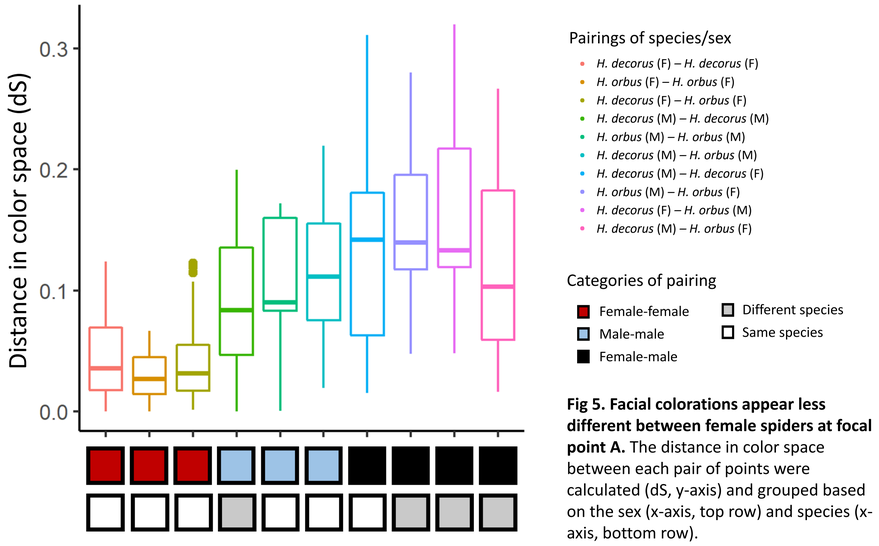
Shows the amount of light reflected at each wavelength for each focal point of the spider's face.

- Data seem to show a sex difference between male versus female spiders of each species.
- Qualitatively, males seem to exhibit darker colors closer to black and females tend to exhibit a "classic melanin profile", which shows a gradual increase towards the longer wavelength reflectance (e.g., red).
- Female *H. orbus* exhibits the greatest reflectance at all focal points except at point C.
- Male *H. orbus* seems to exhibit similar brightness values across the focal points except at point E.

Trichromatic Color Space Plot

Demonstrates how much the red, green, and UV photoreceptors of the jumping spiders are engaged. Data points that are further apart represent greater color differences, whereas points closer together indicate more similar colors.

- There may be a species difference in male facial colorations at some focal points (Fig 6A, B).
- Females of the two species have high overlap in coloration at almost all focal points (Fig 6A, B, D, E, F).
- Reflectance values seem to overall engage the most with the green photoreceptor.
- There seems to be low engagement of the red sensitive photoreceptors
- Compared to the other focal points, points E and F seem to show a greater spread of colors along the green-UV axis.



Pairings of species/sex

- *H. decorus* (F) – *H. decorus* (F)
- *H. orbus* (F) – *H. orbus* (F)
- *H. decorus* (F) – *H. orbus* (F)
- *H. decorus* (M) – *H. decorus* (M)
- *H. orbus* (M) – *H. orbus* (M)
- *H. decorus* (M) – *H. orbus* (M)
- *H. decorus* (M) – *H. decorus* (F)
- *H. orbus* (M) – *H. orbus* (F)
- *H. decorus* (F) – *H. orbus* (M)
- *H. decorus* (M) – *H. orbus* (F)

Categories of pairing

- Female-female
- Male-male
- Female-male
- Different species
- Same species

Fig 5. Facial colorations appear less different between female spiders at focal point A. The distance in color space between each pair of points were calculated (dS, y-axis) and grouped based on the sex (x-axis, top row) and species (x-axis, bottom row).

DISCUSSION

Preliminary results in color space distances indicate that female of the two species have very similar colorations, perhaps suggesting that facial colorations is not a female signal used by males to differentiate species identity

The findings in this study serve to further our understanding of potential mechanisms by which these jumping spiders discern different sexes and species. The difference in facial colors between males and females of both species indicates that faces may act as a visual signal to communicate information about identity.

This study serves as a continuation of a pilot study previously conducted on related species *Habronattus calcaratus* and *H. coecatus*[6]. Measurements obtained from *H. calcaratus* and *H. coecatus* also showed sex-differences in facial coloration within species and a greater difference in color between males of different species than of females. This study shows a similar trend, suggesting a similar type of sexual selection pressure. By shedding light on the use of faces as a means of communication for *Habronattus* jumping spiders, we may be able to better understand the role they play in reproduction when searching for potential mates.

Future directions

Calculation of color space distances for the other five focal points measured

Statistical analysis of distance in color space

Increase sample size and comparison of all 4 co-occurring species of *Habronattus* spiders at the Edge of Appalachia

Previous study indicated that leg colors of preserved *H. calcaratus* may change over time[6]. We may compare freshly-measured specimen to specimen preserved over time to see if any facial coloration are lost over time.

The MSP allowed for robust measurements of small, focal points. However, this does not for allow complex pattern analyses. Multispectral or hyperspectral imaging techniques may allow for more advance analyses of facial patterns.

REFERENCES

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