Linear and nonlinear optical response from *Cicadas* transparent wings

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Abbreviated abstract: The wings of some insect species are known to fluoresce under illumination by ultraviolet light. Their fluorescence properties are, however, not comprehensively documented. In this presentation, the optical properties of unknown fluorophores naturally occurring within *Cicadas* transparent wing, were investigated using both linear and nonlinear optical (NLO) methods, including one- and two-photon fluorescence and second harmonic generation (SHG).

The Challenge & Motivation

Revealing **the optical response of Cicadas and the link between dye (chemical) structure and photonic environment**, including:

- The stability of excited states and excited states dynamics as a function of emission wavelength and incident polarization, symmetry, etc.;
- The connection between biophotonic structures and optical patterns;
- The combination of linear and nonlinear optical methods to obtain a comprehensive and detailed view of the optical response from Insects.

Previous work and Approach

Work presented here is a continuation of **the preceding study** related to:

- Optics of insects in the visible and infrared regime;
- The connection between elytra+ structures and optical responses;
- Heat management in the biological world;
- Connections between optical and surface properties linked by photonic architectures;

Related publications:

- Sébastien R. Mouchet, Branko Kolaric, et al., Interface Focus 9:20180052 (2018)
- Sébastien R. Mouchet, et al., J. Biophotonics, e201800470 (2019)

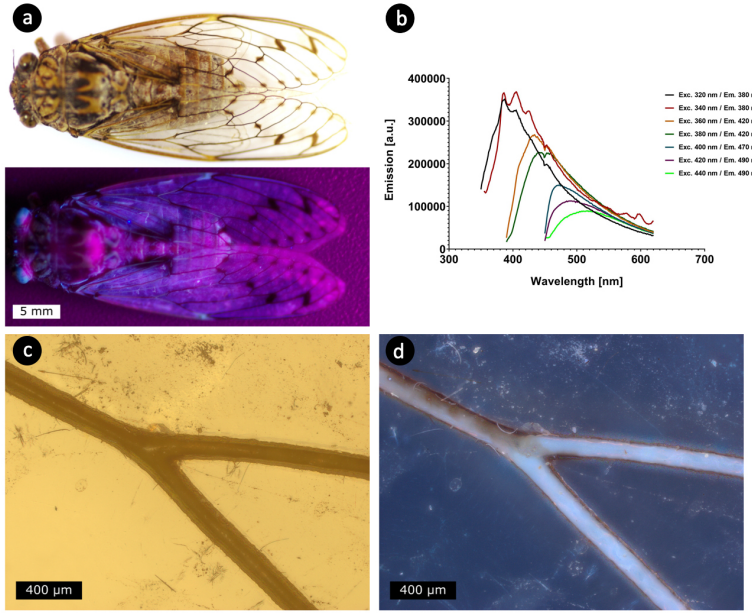
Sample collection

Adult specimens of ***Cicada orni*** and ***Lyristes plebejus*** were collected in Fournès in the Gard Department (France) on the 22nd of June 2014. Adult specimens of ***Hemaris fuciformis*** and ***Vestalis amabilis*** were bought from licensed vendors. All the analyses were performed on the insects' wings without additional sample preparation.

Techniques & Methods

- **Optical and Fluorescence Microscopy**
- **Spectrofluorimetry** - *The sample was excited at different excitation wavelengths: 320 nm, 340 nm, 360 nm, 380 nm, 400 nm, 420 nm, and 440 nm.*
- **Nonlinear Optical Microscopy** - *The excitation was at 800 nm, 900 nm, 1000 nm, and 1100 nm - fundamental wavelengths.*
  - Two-photon Fluorescence (**2PF**)
  - Second Harmonic Generation (**SHG**)

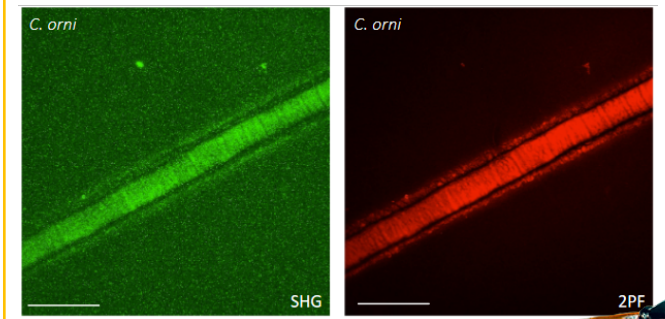
Results



a) Optical and fluorescence image of *Cicada orni*; **b)** emission spectra recorded from the *Cicada orni* wing; **c)** optical image of the veins of transparent wing of *Cicada orni*, and **d)** fluorescence image of the veins of transparent wing of *Cicada orni*.

***Cicada orni* (fluorescence decays in [ns]):**

[exc 340 nm/em 385 nm] t_1 = 1271.23 t_2 = 8933.32
[exc 340 nm/em 403 nm] t_1 = 1309.57 t_2 = 9272.42
[exc 360 nm/em 388 nm] t_1 = 1192.96 t_2 = 8508.55
[exc 360 nm/em 406 nm] t_1 = 1237.31 t_2 = 8831.80



SHG and **2PF** from *Cicadas* wing.

Conclusion

Transparent wings of investigated cicada species emit light under UV excitation and show **complex radiation dynamics** as a function of excitation wavelength. **(Decay profiles)**
Additionally, the wings of both cicada species and, more specifically, their veins showed strong nonlinear response **SHG and 2PF** - probably due to the higher concentration of resilin or chitin in the veins with respect to the membrane that makes up most of the wing.
We reveal by **Linear spectroscopy and NLO microscopy** complex fluorescence patterns from unknown naturally occurring fluorophores within transparent *Cicadas* wings

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