Caltech Graduate Innovator Grant Proposal **Daniel J. Pollak**
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Background: A core question in visual ecology lies in how organisms overcome constraints on visual processing in varying light conditions, specifically how temporal integration of light trades off against signal gain in retinal processing (Stowasser, Mohr, Buschbeck, & Vilinsky, 2015). The purview of neuroscience to answer this question continues to be limited by the small number of established organisms of study. While these organisms of study (e.g., *Drosophila*, *C. elegans*, mice, rats, and monkeys) span their kingdom, many more offer tractable, inexpensive, and fertile ground for investigation. Insects are freely available in the environment, making them uniquely available to scientists and amateurs alike. *Insecta* is by far the most diverse class of animals and therefore an obvious pool from which to draw, yet only *Drosophila* is regarded as a highly tractable organism of study from it.

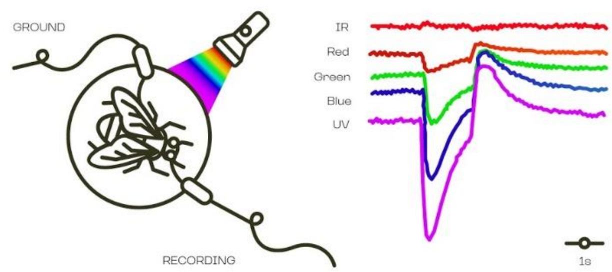

Figure 1. Diurnal bee ERG, comprising on/off transients and holding potentials.

Species within *Insecta* could be added to the pantheon of canonical organisms of study if sorting and screening were distributed among many workers with citizen science (in which members of the public collaborate with supervising scientists). Promising new organisms of study can be expected to emerge within a matter of years. For all the insights they have yielded, most established organisms of study require advanced training to extract basic information and are not readily available. This unavailability reflects science as an institution writ-large: its use of exclusionary gatekeeping mechanisms is intended to guarantee excellent science; however, the benefits of this excellence are often inaccessible to people outside of it. Citizen science represents an opportunity to overcome barriers to accessing and participating in science, yielding new species of study while diversifying voices in science beyond those of privileged few that have always dominated.

Using electroretinograms (ERGs), diel activity in closely related species and light level-dependent shifts can be used as standpoints to investigate tradeoffs in temporal integration and signal gain. Cockroaches dynamically adjust retinal processing to fit light levels (Honkanen, Takalo, Heimonen, Vähäsöyrinki, & Weckström, 2014), and closely related species of hummingbird hawkmoths with differing diel activities process light retinally in ways that match their niches (Stöckl, O'Carroll, & Warrant, 2020). For both cockroaches and hummingbird hawkmoths, temporal integration is slower and signal gain higher in low light conditions, likely due to compensation for low photonic flux, and the reverse is true in high light conditions. Is light level a universal predictor of retinal processing parameters, and does diel activity vs rapid retinal plasticity account for these parameters? This question is well suited as the first of many addressed by a citizen science initiative around ERGs.

Action Plan I will achieve two goals to establish an ERG-based citizen science initiative to include a diverse coalition of citizen scientists in the project of identifying promising organisms of study. For **Goal 1**, I will establish the viability of ERG for investigating visual ecology by performing a pilot study to test **Hypotheses 1** and **2** regarding temporal integration and signal gain, respectively. For **Goal 2**, I will release this ERG platform as a citizen science initiative called ERGo! in collaboration with Backyard Brains and Hirnkastl in two phases. For **Phase 1**, I will develop pedagogy and a user base for ERGo!, and for **Phase 2**, ERGo! will be released to the public as a kit for purchase, discounted for qualifying schools and families.

Materials All materials will be open sourced. The recording rig consists of a Backyard Brains Spikerbox, silver wire electrodes, and light stimulator (Figure 2), both of which are powered by Arduino sketches. The Spikerbox integrates physiological signal data and stimulus delivery timing and identity, and the light stimulator delivers montages of light wavelengths (red, blue, green, infrared, and ultraviolet) with varying flashing frequencies. Data acquisition is performed by the Backyard Brains Spike Recorder software, and data analysis will be performed with annotated Jupyter notebooks in Python. Blu-tack, sewing thread, and cardboard will be used for insect restraint, and all physical materials will be packaged together as an ERGo! kit. The Spikerbox will go through three prototypes: one over the course of **Goal 1** and two over the course of **Goal 2**.

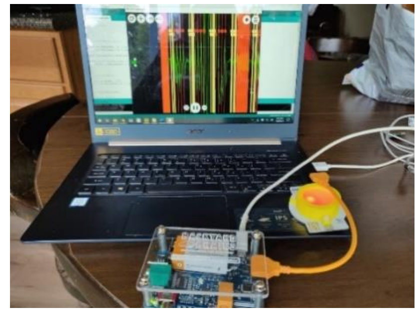

Figure 2. ERG setup. Light stimulator flashing red light (right), Spikerbox (bottom), and ERG data acquisition (screen).

Goal 1 I will investigate two hypotheses relating to retinal processing in *Dolichovespula maculata* and *Vespa crabro*, diurnal and nocturnal wasp species, respectively. I will publish the findings and methods and as a teaching laboratory exercise in the Journal of Undergraduate Neuroscience Education (Pollak, Feller, Serbe, Mircic, & Gage, 2019). We will use insights from this publishing process to generate one of the three projected prototypes for the Spikerbox to refine the process of data acquisition, including simplifying the interface. **Hypothesis 1** Nocturnal insects have a longer "image refresh rate" than diurnal insects. Since light levels are lower at night, nocturnal insects must find ways to respond to behaviorally relevant visual stimuli. I expect *Vespa crabro* to use longer temporal integration than *Dolichovespula maculata* to construct an image with fewer photons. **Hypothesis 2** Nocturnal insects use higher signal gain than diurnal insects. I expect *Vespa crabro* to have stronger ERG responses to white light than *Dolichovespula maculata* to compensate for low overall signal strength. **Methods** To measure temporal integration, ERG responses will be quantified at the frequency of stimulation. The frequency at which the signal strength falls below -3 dB relative to the maximal response strength will be taken as the critical flicker fusion frequency, or the limit of the insect's ability to integrate a visual scene in time. To measure signal gain, each light wavelength will be presented at intensities on the log scale from -3 to 0 dB relative to the maximal intensity. Sensitivity will be measured as the intensity that produces half of the maximal response.

Goal 2 Citizen scientists will send data to a database in Germany, from which broad patterns in visual ecology will be deduced.

Phase 1 Using the outreach network cultivated by Caltechs' Visiting Scientists and Interaxon groups, I will lead preliminary workshops in public school classrooms in Pasadena. These workshops will introduce participants to the citizen science initiative by addressing the three main components of science identity (competence, having the ability of a scientist; performance, using these abilities; recognition, acknowledgement of one's science identity from one's immediate environment and the larger community, Carlone & Johnson, 2007) and build a base of future ERGo! users. I will facilitate competency with a brief interactive lecture to build a knowledge base. To build performance, students will do their own experiments, including catching insects, cooling them, and recording from them. To build recognition, I will emphasize a definition of scientist that includes them. In addition, I will include their names in the online data repository and publications (with participants capable of remaining anonymous by opting out of public recognition). I will collect survey data from students and teachers, with which I will iteratively tune the workshop to achieve its goals of science identity formation and mustering a future user base. Feedback from these workshops will inform the last two prototypes, which will enhance usability of the Spikerbox for lay people by further simplifying the interface and adding visual cues to the board.

Phase 2 In the second phase, ERGo! kits will be made available for purchase by Backyard Brains. To disseminate the project, I will continue to lead workshops, finely tuned based on previous survey data, and release video tutorials on YouTube with Backyard Brains on each topic covered in the workshops. Backyard Brains has 10s of thousands of subscribers and 10 million views on YouTube. *To increase the accessibility of this procedure, part of this grant's funds will go to providing kits to schools and families at dramatically lowered prices or for free.* Eligible schools and families will be identified through the Pasadena Unified School District using outreach networks previously cultivated by Caltech's science outreach groups, Visiting Scientists, and Interaxon. I will collaborate with Hirnkastl to make a database platform for submitting user-acquired data from the field. The submission process will emphasize acknowledgement of citizen scientists while prioritizing privacy, with users being able to opt out of providing identifying characteristics, including as locale and name. Like the Sloan Sky Survey, this database will be made available to the public; however, we will impose conditions of acknowledgement of every citizen scientist whose data contributed to their insights for publishing with the dataset.

Distinctness from ongoing work DJP is an incoming PhD student in Neurobiology and is not currently working in a laboratory, nor is he doing a laboratory rotation until Winter 2021. This proposal is thus separate from ongoing laboratory work.

Works Cited

Carlone, H. B., & Johnson, A. (2007). Understanding the Science Experiences of Successful Women of Color: Science Identity as an Analytic Lens. Journal of Research in Science Teaching, 44(8), 1087–1218. https://doi.org/10.1002/tea.20237

Honkanen, A., Takalo, J., Heimonen, K., Vähäsöyrinki, M., & Weckström, M. (2014). Cockroach optomotor responses below single photon level. *Journal of Experimental Biology*, *217*(23), 4262–4268. https://doi.org/10.1242/jeb.112425

Pollak, D. J., Feller, K. D., Serbe, É., Mircic, S., & Gage, G. J. (2019). An Electrophysiological Investigation of Power-Amplification in the Ballistic Mantis Shrimp Punch. *JUNE*, *17*(2), T12–T18.

Stöckl, A. L., O'Carroll, D. C., & Warrant, E. J. (2020). Hawkmoth lamina monopolar cells act as dynamic spatial filters to optimize vision at different light levels. *Science Advances*, *6*(16), eaaz8645. https://doi.org/10.1126/sciadv.aaz8645

Stowasser, A., Mohr, S., Buschbeck, E., & Vilinsky, I. (2015). Electrophysiology Meets Ecology: Investigating How Vision is Tuned to the Life Style of an Animal using Electroretinography. *Journal of Undergraduate Neuroscience Education*, *13*(3), A234–A243. Retrieved from www.funjournal.org

Budget Justification

Expenses 2021	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	*Total*
Monthly totals:	$1,776	$0	$0	$0	$0	$1,350	$0	$0	$3,200	$1,000	$1,000	$1,500	$9,826
Prototype assembly	**$100**	$0	$0	$0	$0	**$100**	$0	$0	**$800**	$0	$0	$0	**$1,000**
Prototype 1 print	**$100**	$0	$0	$0	$0	$0	$0	$0	$0	$0	$0	$0	**$100**
Prototype 2 print	$0	$0	$0	$0	$0	**$100**	$0	$0	$0	$0	$0	$0	**$100**
Prototype 3 print	$0	$0	$0	$0	$0	$0	$0	$0	**$700**	$0	$0	$0	**$700**
AM systems silver wire (# 786500)	**$105**	$0	$0	$0	$0	$0	$0	$0	$0	$0	$0	$0	**$105**
Soldering Iron Station Kit	**$51**	$0	$0	$0	$0	$0	$0	$0	$0	$0	$0	$0	**$51**
Liquid electrical tape	**$20**	$0	$0	$0	$0	$0	$0	$0	$0	$0	$0	$0	**$20**
Software engineering (Stanislav Mircic)	**$500**	$0	$0	$0	$0	**$500**	$0	$0	**$500**	$0	$0	$0	**$1,500**
Database hosting	**$500**	$0	$0	$0	$0	$0	$0	$0	$0	$0	$0	**$500**	**$1,000**
Classroom kit subsidies	$0	$0	$0	$0	$0	$0	$0	$0	**$500**	**$500**	**$500**	**$500**	**$2,000**
Family kit subsidies	$0	$0	$0	$0	$0	$0	$0	$0	**$500**	**$500**	**$500**	**$500**	**$2,000**
Hardware engineering (Wenbo Gong, Stanislav Mircic)	**$500**	$0	$0	$0	$0	**$500**	$0	$0	**$500**	$0	$0	$0	**$1,500**