Analyzing Negative Phonotaxis in Gryllus bimaculatus Following Injury-induced Deafferentation

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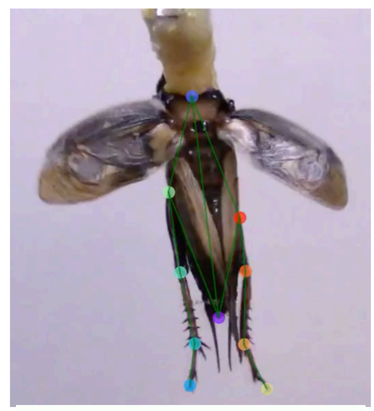
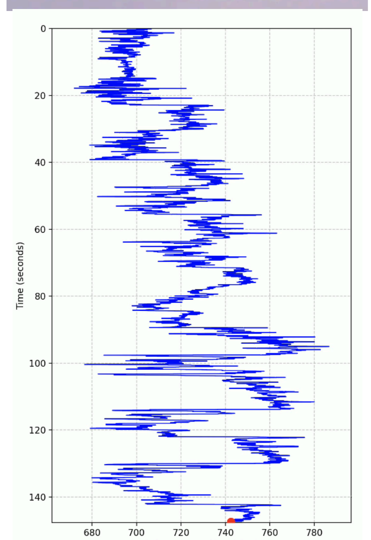
The cricket *Gryllus bimaculatus* is a valuable model for studying the flexibility of the nervous system, known as compensatory neuroplasticity. Uncommon in animals, the cricket auditory system contains various neurons that—when deprived of sensory information following ear loss—aberrantly sprout new branches to receive auditory signals from the opposite, intact ear.[1] Fascinatingly, function is restored to the neuron 4–6 days following ear loss and presumably this growth leads to the cricket recovering some ability to localize sounds.[2,3] Hence, deafferentation, the loss of sensory information to a neuron, is thought to provoke a repair or compensatory mechanism in the cricket. While much work has been done to understand this phenomenon morphologically and physiologically, it is not certain how this instance of compensatory neuroplasticity manifests behaviorally. Thus, my project sought to develop a method for testing and analyzing cricket behavior as to determine what advantages this plasticity affords to the cricket, if any.

To assess behavior following deafferentation, I investigated the cricket's readiness to avoid the sounds of a predatory bat—a behavior known as negative phonotaxis—during flight. Crickets were suspended in a sound-insulated box, whereupon a stream of air compels the crickets to adopt a flight posture. Once in flight posture, audio speakers on either side of the box would reproduce high-frequency bat sounds, eliciting negative phonotaxis. Once a diverse set of cricket footage was collected, 500 images of cricket flight were manually labeled for body parts (abdomen, legs, etc; see attached image). This allowed me to train DeepLabCut (DLC), an AI engine, to expedite the data collection from 300 cricket flight videos recorded this summer. Data were collected in the form of over 50 million x- and y-coordinates for each body part per video frame.





Crucial to analysis—I dedicated a portion of my summer toward finding powerful ways to visualize and present these data. By graphing the abdomen's position over time, I highlighted the cricket's negative phonotaxis away from the audio stimuli (see the attached chart, and note the abdomen's graphed movements left and right). With the assistance of generative AI, I created an analysis app that would produce several key statistics from these charts, such as turning frequency and abdominal activity (measured as the amount the abdomen moves during a flight). Though time constraints kept me from finding why crickets exhibit this compensatory plasticity—my analysis pipeline is a useful starting point for future research.

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[1] Horch, H. W., Mito, T., Popadić, A., Ohuchi, H., & Noji, S. (2018). Plasticity in the Cricket Central Nervous System: Development, Regeneration, and Behavior. In *The cricket as a model organism*. Springer.
[2] Brodfuehrer, P. D., & Hoy, R. R. (1988). Effect of auditory deafferentation on the synaptic connectivity of a pair of identified interneurons in adult field crickets. *Journal of Neurobiology*, *19*(1), 17–38.
[3] Scholes, J. (2020). *Characterization of negative phonotactic behavior in the adult cricket, Gryllus bimaculatus* [Honors Project]. Bowdoin College.