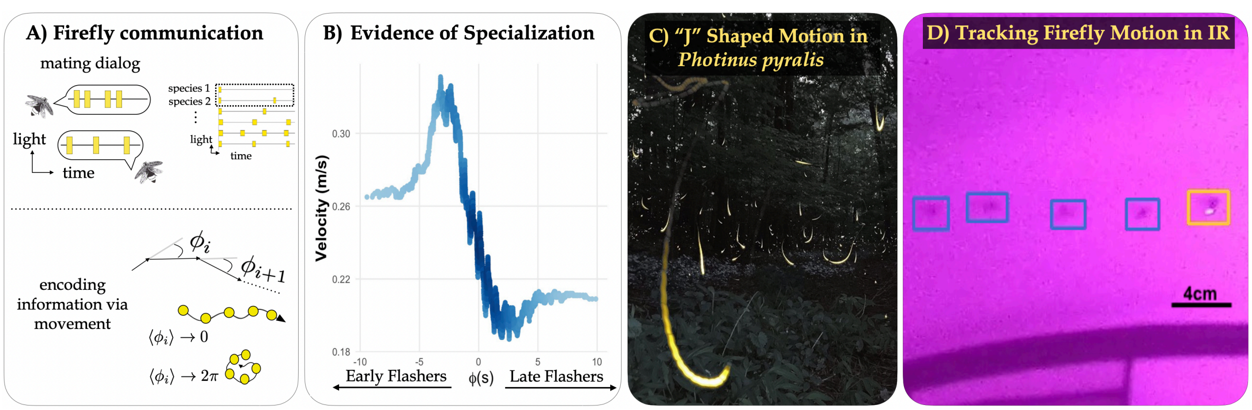
Analyzing Firefly Movement to Discover Complex Communication and Intra-Swarm Specialization

Biological systems have developed unique communication methods over millions of years of evolution. Firefly swarms are one example of a biological system that engages in communication through the use of bioluminescence. Understanding communication mechanisms found in nature is an important challenge which could greatly benefit from modern computational tools[5,6]. While the temporal nature of firefly bioluminescence has been studied in detail[1,4], there has not yet been any research into the importance of spatial positioning. However, there is growing evidence of encoded information in fireflies and other bioluminescent species. For example, *Photinus pyralis* fireflies (also known colloquially as the “Big Dipper”), exhibit very unique “J” shaped movements while flashing (Fig 1C). Additionally, *Anomalops katoptron* have been found to use movement and bioluminescence to coordinate swimming dynamics in swarms[2]. **I will study how firefly movement plays a role in swarm communication and specialization.** My research will address two main questions concerning: the data encoded in spatiotemporal movement during a flash, and the occurrence of intra-swarm specialization. My interdisciplinary research will combine fundamental topics in physics and ecology with modern computational tools, which will result in an understanding of how spatial information contributes to optimal swarm communication in fireflies.

Research Question 1: When fireflies are flashing, is there any communication data encoded into their movement? Firefly communication through bioluminescence can be thought of as a binary state, on or off. However, three-dimensional movement could add a new level of informational complexity that has not yet been understood (Fig 1A). **Hypothesis:** Based off the growing evidence we have seen concerning the importance of movement in fireflies and other bioluminescent species, we hypothesize that there is communication data encoded in firefly movement. Currently, there is a lot of firefly behavior that we can not explain with the simplicity of temporal flashing[3], meaning that there likely is another layer of information that has not been discovered. Moreover, if fireflies are synchronizing their flashes, they might also be synchronizing their movement.

Research Plan: Our lab has already collected large amounts of data for several firefly species found in the U.S., so I have started developing analysis tools to track bioluminescent trajectories in three-dimensional space. My extensive background in data-engineering places me in an ideal position to quantify firefly movement, which will produce valuable metrics such as: spatial coordinates, velocity, angular velocity, and movement vectors. I will then analyze time-series swarm data to extract useful insights on the collective movement properties. I anticipate finding signs of movement synchronization (fireflies moving in unison), repeating trajectory patterns (similar general movement shapes that fireflies repeatedly make overtime), or propagating motion (movement patterns that spread to nearby neighbors like swarming birds). The results from these experiments will allow us to understand if seemingly simple forms of communication have evolved to allow for greater complexity, teaching us how information is optimally communicated in swarms.



A) Model of the 2 main aspects of firefly communication: temporal flashing, and motion. B) Preliminary data showing firefly velocities relative to their phase time. C) Long exposure shot of *Photinus pyralis* flash movement. D) Preliminary computer-vision results showing 1 firefly being tracked across multiple frames. A yellow box indicates that the firefly is flashing.

Research Question 2: Can we use movement data of fireflies that are not flashing to answer questions about intra-swarm specialization? Little is known about the non-flashing behavior of fireflies due to the limitations of field recording techniques. However, new advancements in experimental methods using infrared (IR) imaging are allowing us for the first time to capture firefly movement between flashes. **Hypothesis:** Certain fireflies in a swarm might have specific roles. Our lab has found evidence of this in *Photinus carolinus*, discovering that fireflies which flash earlier in a swarm exhibit higher movement velocities in localized firefly clusters[4] (Fig 1B). However, we do not know if these early flashers are always the same firefly, or if a different individual takes the role each time the swarm flashes. By continuously tracking every firefly in a swarm over time, we could identify early flashers through multiple swarm flashes. My results would either provide the first evidence of global intra-swarm specialization in fireflies, or indicate that role specialization is spontaneous.

Preliminary Work: Current imaging techniques are able to temporally track precise locations of fireflies within a swarm while the fireflies are flashing[4]. However, observing fireflies between flashes is challenging given the size of the insects and the darkness of the environment. To address this, our lab has developed an experimental approach to continuously image the fireflies utilizing IR light. We have collected multiple spherical IR videos of fireflies filmed inside a tent, which has been shown to not change firefly behavior[10]. As a proof of concept, I have developed a computer vision model that can accurately track a firefly's movement in this environment (Fig 1D).

Research Plan: Tracking fireflies with IR poses several challenges that I intend to tackle in my graduate work. Specifically, I will continue improving our tracking techniques through the use of neural networks, while also developing new field methods to increase the scale of our experiments. I will use the developed tracking techniques with the use of multiple cameras to record firefly positions in three-dimensional space. <u>Computational Model:</u> Developing computational models can help us understand how emergent behavior arises from the simple communication behavior we observe. By utilizing the experimental observations, we can construct and validate accurate simulations of firefly communication and movement. Our lab has leveraged this model-experiment approach to discover new behaviors in many different species. My results will provide key insight into the existence of swarm specialization and optimal localized communication.

Intellectual Merit: I will be conducting interdisciplinary research, combining topics from physics, ecology, and computer science to understand the complexities of swarm communication. Due to my experience as a researcher at Sandia National Laboratories, and my affiliations with the computer science and Interdisciplinary Quantitative Biology programs at CU Boulder, I am well positioned to answer fundamental questions about swarm specialization and communication. Oftentimes, a computational analysis of complex biological systems has revealed new physics of animal communication, allowing us to learn from nature's ability to optimize through evolution[8,9]. The results of my research will lead to a deeper understanding of the physics, and extent of complexity in optimal animal communication.

Broader Impacts: My research will have the capability to make meaningful impacts in several emerging technologies, such as multi-agent AI systems and swarm robotics[5]. Moreover, the success of systems like 5G, autonomous driving networks, and the internet of things (IoT) will depend heavily on our ability to optimize local communication networks. By utilizing fireflies as a representative system, we can understand how nature has optimized local communication through millions of years of evolution. Humans have a special connection with fireflies; they inspire awe and wonder, and spark curiosity about their synchronous nature. I will take an active role in my lab's educational outreach, where we host workshops at some of the firefly experiment sites to answer various questions about the fireflies and discuss our research. According to the International Union for Conservation of Nature, one in three of all Northern American firefly species are at risk for extinction, and half of the species found in the region are at an unknown threat status due to insufficient data[7]. The research and data our lab produces will directly assist in the assessment and conservation of Northern American fireflies. **References:** [1] S. Lewis, Silent Sparks, Princeton University Press (2016).[2] D. Gruber et al., PLoS ONE 14.8 (2019). [3] J. Lloyd, Science 187.4175 (1975). [4] R. Sarfati et al., Science Advances 7.28(2021). [5] G. de Croon et al., Science Robotics 7.67 (2022). [6] T. Li et al., IEEE Communications 24.2 (2022). [7] C.E. Fallon et al., PLoS ONE 16.11 (2021). [8] N. Ouellette, Physical Biology 19.2 (2022). [9] N. Ouellette et al., Frontiers in Physics 9:687823(2021). [10] R. Sarfati et al., Royal Society Interface 17.170 (2020).