Major Activities: In the first year of this MIRA grant, I have been building up my lab group and establishing several major research projects.

- In terms of **personnel**, there are three post-docs, three PhD students, two full time research technicians, three undergraduate researchers, and 7 undergraduate assistants (currently, 1 post-doc is being paid by the MIRA grant). I mention the size of the lab group to highlight that I have built up a critical mass of people who are working together in a cohesive way. Personnel lab are roughly split 50:50 between Daphnia and Drosophila projects.
- We have built several important **infrastructural** components for our Daphnia and Drosophila work.
  - We planted our experimental orchard in April 2016 at Morven Estates. This orchard is composed of 32 mixed fruit trees in a 1/2 acre lot, fully equipped with deer fencing and irrigation. We will likely begin our first experiments out there this year.
  - We have built experimental chambers for Drosophila work that enable us to manipulate temperature and photoperiod. These chambers will allow us perform GWAS on experimental populations of flies with the goal of identifying temperature and photoperiodic responsive polymorphisms. We have built 48 chambers in total, each equipped with a RasberryPi computer, fully control of red, green, blue, and white LEDs, temperature manipulation over a 5°C range, activity monitors, and redundant temperature, humidity, and light sensors. These chambers will initially be used to study overwintering biology of Drosophila.
  - We have established a large scale Daphnia husbandry facility. We are capable of maintaining 500 clonal lineages in triplicate. We have established protocols to extract *Chaoborus* kairamone and trained undergraduate students to conduct phenotyping experiments using Daphnia.

Specific Objectives: During the first year of this MIRA grant we have been focusing on building a lab group and building the infrastructure to conduct experiments using Daphnia and Drosophila. Over the course of the summer 2016, we will begin our first major phenotyping efforts for both species. For Drosophila, we will be studying photoperiodic and temperature dependent induction of diapause and associated gene expression profiles; for Daphnia, we will be studying development of defensive neck-teeth as well as transcriptional profiling in response to predation cues.

Significant Results: We have been conducting extensive simulations to examine optimal experimental designs for association mapping in experimental hybrid swarm populations founded with inbred lines of *Drosophila melanogaster.* In the MIRA grant, I had suggested that such an approach will enable us to perform GWA studies in a cost and time efficient manner. To this end, we have found that:

- individual sequencing to 0.05X coverage (i.e., 1 of every 200 bp sequenced) is sufficient to recover a fully phased, 99% accurate genome for flies that come from hybrid swarms of <200 inbred lines. We have conducted preliminary analyses of this genome-reconstruction approach using the a hybrid swarm derived from the DGRP. Analysis of shallow and high-coverage samples empirically confirms this prediction.

- simulations of GWA experiment demonstrates that hybrid swarms have higher resolution and greater accuracy than simple mapping in the DGRP. Generating swarms with ~32-64 founding lines is sufficient to maximize power.

These results are being pulled together in a MS now. This MS is being written by a PhD student in my lab and has been invited for review at Genetics in their new Multiparental Population series.

Key outcomes or other achievements.

- We have initiated 2 hybrid swarms of D. melanogaster composed of inbred lines collected along the East Coast of the US. These hybrid swarms will be ready for phenotyping in our photoperiod-tempature controlled chambers by early summer.
- We have collected and established 500 clonal lineages of Daphnia pulex for phenotyping this summer. Whole-genome resequencing for these clones will being in May.
- We are generating a new Daphnia pulex reference genome for British D. pulex (where our focal populations reside) using 10X Genomics technology
- We have optimized protocols on UVA's Biomek liquid handling robot for high-throughput DNA extraction and Illumina library preparation.