

My research program examines the evolutionary dynamics of natural genetic variation in fitness related traits. We ask questions about the provenance and stability of polymorphisms that underlie functional genetic variation, and test how these polymorphisms adaptively track to spatial and temporal variation in selection pressures. This work seeks to assess the nature of balancing selection, and the importance of balancing selection as an evolutionary force which promotes diversity within populations and species. We address these questions using *Drosophila* and *Daphnia*. Due to their rapid development time, these assemblages of broadly distributed species have multiple generations per year (~10-20), enabling us to study their evolutionary dynamics in real-time and across a varied spatial landscape. Our work on *Drosophila* focuses on the genetic basis of local adaptation to seasonal and latitudinal variation in climate and nutrition. Our work on *Daphnia* examines the recurrent seasonal evolution of sexual investment, the mutational variance of predator induced plasticity. In our research, we combine field-work, genomic analysis, and large-scale phenotyping efforts along with computational and experimental tools that we develop to gain insight into the basic evolutionary forces that maintain diversity.

Drosophila. *D. melanogaster* living in orchards throughout the world experience seasonal fluctuations in selection pressure driven by changes in temperature and resource abundance (amongst other factors). Genetic variation in starvation tolerance, thermal tolerance, longevity, fecundity, etc., enables some individuals to be more likely to survive winter, and others better able to exploit the favorable growing season. Genetically based seasonal variation in phenotype is, in part, generated by polymorphisms that fluctuate in frequency between seasons. Ongoing work in my lab seeks to (1) further document genetic variation across time and space in *Drosophila* and to use this genomic data to infer the seasonal evolutionary dynamics of natural populations; and, (2) to uncover the genetic architecture, molecular function, and evolutionary history of natural polymorphisms that underlie local adaptation.

To study the nature of genetic variation across time and space, we are engaged in several large-scale collection efforts to sample and sequence *D. melanogaster*, and related species, collected throughout their range and over multiple years. This work is broken into consortium work (that the PI is mainly involved in; <http://droseu.net>), fine-grained sampling efforts of local field sites (PhD student), and experimental outdoor mesocosm work (UG, Post-doc). My work in the consortium (along with Martin Kapun, Univ. of Zurich) is to build a pipeline to generate the joint data-set of population-based allele frequencies from samples of wild-derived *D. melanogaster* from ~250 localities across Africa, Europe, the Americas, Australia, and Asia. Many of these samples are collected from the same locality ~2 times per year, and, for some cases, for nearly a decade, enabling us to examine the temporal dynamics of seasonal adaptation across multiple populations (Machado*, Bergland*, et al; 2nd round, [eLife](#)).

The seasonal end-point analysis of pool-seq'd flies has been an informative approach to identify the presence and generality of seasonal adaptation. However, a more informed interpretation of that data requires knowledge of short-term population dynamics and the specific selection pressures acting on flies living in orchards. Alyssa Black (5th year PhD) has been working to measure some of these key parameters, including short-term N_e and kinship across seasons using individually sequenced, wild-caught flies from a local orchard. These samples show an excess of homozygosity (elevated F_{IS}) and we observe related individuals in our samples, suggesting that populations of flies living in orchards might be structured at a family level due to short-distance dispersal and re-colonization dynamics. Having knowledge of this metapopulation dynamic will allow us to build better null models to test for adaptive vs. demographic events in the time-series data (in prep). In a second major project, Alyssa is analyzing genome-wide allele frequency data from fly samples collected every two weeks, from May to December, over 3 years (pool-seq) at a local orchard. She will be using this data to test whether any specific abiotic environmental factors appear to be stronger drivers of allele frequency change across time.

Finally, we are using outdoor mesocosms to track captive populations through time. This work has examined the extent of seasonal plasticity and adaptive tracking in thermal tolerance (Helen Stone, fmr. UG: Stone et al, Ecology and Evolution) and diapause (Priscilla Erickson, post-doc: Erickson et al, in review PLoS Genetics, 2nd round).

The second major way that we study rapid adaptation in *Drosophila* is to examine the genetic architecture of the traits that are thought to be subject to seasonally and clinally varying selection pressures. To advance our ability to accurately map genetic variation in these fitness traits, we have developed a multi-parent mapping approach which we call the Hybrid Swarm. In this approach, between 32-128 inbred founder lines are intercrossed for ~5 generations to generate an outbred population. We have developed a pipeline for reconstructing fully phased genomes from ~0.05X sequencing and have shown that for quantitative traits, association mapping using the Hybrid Swarm has a higher true positive rate, and lower false positive rate, than the DGRP; this is largely due to breakdown of long-distance linkage disequilibrium present in inbred panels which

causes inflated signals of association even on unlinked chromosomes (Cory Weller fmr. PhD student: Weller & Bergland, in revision, Genetics).

We have applied this mapping approach to the study of diapause and of gene-expression. Our work on diapause (Erickson et al, in review PLoS Genetics) demonstrated that standing genetic variation in diapause is highly polygenic, old (predating colonization of higher latitudes), and shows contrasting patterns of variation across space and time. Signals of seasonal change at these loci suggest idiosyncratic change between populations. We have found a similar pattern at eQTL (Yang Yu, PhD student; Yu and Bergland, in prep). We argue that this idiosyncratic pattern of allele frequency change between seasons across populations is a basic outcome of strong selection on quantitative traits. Whether these selective dynamics, coupled with metapopulation structure, enhances the stability of functional variation remains an interesting avenue of future study.

Daphnia. *Daphnia pulex*, a small aquatic crustacean living in ponds and lakes across the temperate zone, is a facultative parthenogen and an important keystone species through its role as grazer and prey. Clonal isolates of *D. pulex* sampled within and among ponds, and across seasons, show extensive genetic variation in life-history traits, predator defense capacity, and sexual dynamics. Our work in *Daphnia* examines the molecular evolution of loci controlling variation in these traits and works to identify the evolutionary forces maintaining variation.

Central to addressing these basic questions is an understanding of the seasonal life-history of *D. pulex*. The classic view is that daphnia hatch in the spring from resting eggs, which are the product of sex the previous fall; these newly hatched daphnia then undergo asexual reproduction and clonal selection, leading to a rapid decline of clonal diversity; sex ensues in the fall and the population overwinters as resting eggs. In the absence of any diversifying force, such a population would quickly become monomorphic and highly inbred. To assess the importance of different diversifying forces on the maintenance and generation of variation, we have been examining (1) reproductive niche partitioning and (2) the mutational variance of predator induced plasticity.

Our *Daphnia* research is situated in a series of intermittently connected ponds in southern England (Dorset), where we have been sampling over the last 4 years. These populations largely adhere to the classical model, described above, but the duration of time between bouts of sex varies among ponds: shallow ponds go dry every year and sex is enforced on an annual basis; deeper ponds dry on decadal time-scales, and do not freeze, therefore sex is less frequent. The degree of ephemerality affects the genealogical structure of *Daphnia* living in these ponds, but individuals within and among ponds are closely related, resembling something between a 2- to 8-way intercross. These populations resemble a natural QTL mapping panel.

We have observed heritable variation in reproductive allocation among co-existing clonal lineages. As facultative parthenogens, a female daphnid is capable of three modes of reproduction, and she will likely experience all three during her life: she can produce a brood of ~1-20 clonal female progeny; a brood of ~1-10 male progeny; or she can produce an ephippial case which, if fertilized, will contain no more than two embryos which overwinter ("resting eggs"). Two dominant clonal isolates sampled from one pond in 2017 exhibit variation male production rate (~2% vs 15%), which leads to a significant difference in clonal growth rates as measured in lab mesocosms. Given their different life-histories the coexistence of these clones suggests some form of balancing selection caused by unidentified tradeoffs. Moreover, we have identified that variation in male production rate is polygenic, with upwards of ~8 QTL on separate chromosomes segregating within these natural populations. By examining patterns of haplotypic diversity within and among clones using ~500 individually sequenced field isolates, mapped to a high-quality reference genome that we generated (130Mb, 1 scaffold/chr, BUSCO ~95%), we are currently testing several key predictions about the evolutionary forces maintaining and generating polygenic variation in reproductive allocation (Karen Barnard-Kubow, post-doc; Barnard-Kubow et al. in prep).

These *Daphnia* populations live in a dynamically changing environment. One dramatic fluctuation is the presence of predatory midge larvae, which are abundant only during some parts of the growing season. Daphnids are well known to grow defensive structures in response to predators, and *D. pulex* often grows a thickened dorsal carapace and several small spikes as a juvenile. These structures increase handling time of the predatory midge. Using an automated image analysis pipeline that we developed, we have characterized genetic variation in induction across ~200 clones sampled from the Dorset field site. Many of these clones are genetically unique, the product of recent sex in an ephemeral pond, but others are field derived sub-clones from several of the dominant clonal lineages. These contrasting sets of clones, coupled with high-quality and unbiased phenotyping and whole-genome sequencing, allows us to test the role of mutational input - generated via mutation accumulation over the course of 20-100 generations - on phenotypic variation of predator response (Dörthe Becker, fmr. post-doc; Becker et al, in prep).