Research Statement Joseph Gage

My research group at NCSU will be focused on learning how genome sequence drives allelic differences in gene expression and, ultimately, adaptation and GxE in crop species. We will accomplish this by scaling up and applying emerging technologies to ask and answer questions that were previously inaccessible. When I refer to scale, I mean not only physical scale, but also the phenotypic space we can evaluate, the number of genotypes, and temporal scale. Foundational research in model organisms has given us a wealth of exciting sequencing techniques; these can be scaled up to learn about natural allelic variation for biological processes. Foundational research has also revealed biological mechanisms that were gleaned from detailed studies on a small number of genes or genotypes; these discoveries can inspire hypotheses that we will subsequently test on a genomewide scale across diverse individuals. It is important to me that these projects in my research group can ultimately be applied to improving crops for changing and unpredictable climates. In that regard, I foresee my research group as bridging the space between foundational and applied science.

Previous & current work

During my Ph.D., I wrote image analysis software to increase the speed at which we could phenotype maize tassel morphology. Not only could I use images to quantify traits that were previously measured by hand, but also aspects of tassel morphology that could not easily be quantified by manual measurement, such as curvature, compactness, and complexity. By increasing the phenotypic space, I had sufficient power to identify both phenotypic and genetic signatures of selection left behind by breeders, who had selected for improved tassel morphology over the preceding century. I also leveraged the increased scale of the largest coordinated public hybrid maize trials, Genomes to Fields, to show that selection by breeders has reduced GxE effects and that stability is disproportionately controlled by allelic variation in regulatory regions of the genome.

As a postdoc, I have led our group in adopting and scaling up our use of autonomous rovers for under-canopy phenotyping. I have developed machine learning algorithms that utilize proximal-sensing data to describe overall plant architecture without the constraints imposed by human-defined traits. An ongoing project, led by an undergraduate with my supervision, is using the increased temporal resolution afforded by longitudinal phenotyping to study the genetic basis of differential growth rates.

My current projects, funded by an NSF fellowship that I wrote, are using proteomics and circadian transcriptomics across diverse maize inbreds to study allelic regulation of gene expression. Until now, proteomics have been under-utilized for studying natural variation between maize lines at population scale. By studying protein abundance across individuals, I have found that SNPs in the 5' UTR of genes can cause allelic variation in upstream open reading frames (uORFs), which subsequently cause differences in translation and protein accumulation. Similarly, circadian transcriptomic studies typically focus on a limited number of genotypes, but have not been applied to study natural variation. Circadian transcriptomics across diverse inbreds have revealed that presence or absence of transcription factor binding sites in the promoters of target genes appears to be predictive of gene expression patterns throughout the day. Both of these projects have generated more questions than they have answered, which will help initiate my research program at NCSU.

Future work

Allelic variation is the foundation of plant breeding. Understanding the mechanisms by which allelic variation occurs will be essential to building more targeted predictive models and engineering crop varieties. How does sequence variation between alleles control differences in gene expression? How do these differences in gene expression modulate phenotype in response to environmental conditions? These questions will guide my research program, initiated by the two projects described below.

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Project: How does sequence variation control post-transcriptional regulation and environmental response?

Upstream open reading frames (uORFs) are regulatory sequences in the 5' UTR that are sometimes preferentially translated instead of the main open reading frame (mORF). This mechanism of translational regulation has been described in single-gene studies and using reporter assays in single-celled model organisms. I have found that in maize, SNPs throughout the genome that strengthen or weaken the start codon of uORFs are associated with predictable changes in protein abundance and have enriched effects on plant-scale phenotypes. I have also shown that genes in Arabidopsis which show circadian rhythms for translation are more likely to have uORFs than genes which do not have cyclical translation. I hypothesize that these changes in translation between individuals and across time help control responses to variable environmental conditions. Ribosome footprinting is a method that allows sequencing of short segments of mRNA bound by translating ribosomes. By performing circadian ribosome footprinting on diverse maize inbred lines that have high quality genome assemblies, we will be able to answer the following questions:

- Are uORFs differentially translated between inbred lines?
- Are there sequence features of the uORF that affect whether it is translated?
- Are patterns of uORF translation over time inversely correlated with patterns of mORF translation? Are these patterns consistent between individuals?
- Are genes that are regulated by uORFs enriched for functions that require plants to fine-tune gene expression? For example: transcription factors, metabolite production, or biotic/abiotic stress response?
- Can natural variation in uORFs be harnessed to predict, explain, or engineer GxE interactions?

Project: How does variation in transcription factor binding sites contribute to adaptation?

Core genes of the plant circadian clock, such as CCA1 and LHY, are transcription factors (TFs) with numerous targets genomewide. We know that variation in TF binding sites (TFBS) can affect gene transcription, which is usually measured at a single timepoint. I have scaled up measurements of gene transcription by adding a temporal dimension, sampling mRNA every two hours for 24 hours across 24 diverse inbred lines. Preliminary results indicate that variation in presence/absence of TFBS between individuals is also correlated with diurnal patterns of mRNA abundance. Because the circadian clock and flowering time pathways are closely intertwined with each other, I hypothesize that these differences in TFBS are related to daylength sensitivity, flowering time, and adaptation to northern latitudes (longer days). I would like to continue working with these data and existing ATAC-seq, as well as potentially work with collaborators to generate new ChIP-seq or DAP-seq datasets, to answer the following questions:

- How does allelic sequence variation alter the presence or absence of TFBS between individuals?
- Does allelic variation in TFBS contribute to differences in temporal patterns of target gene transcription?
- Does differential chromatin accessibility contribute to temporal patterns of target gene transcription?
- Are genes with differential TFBS or chromatin accessibility, and differential temporal transcription patterns, important for processes related to the circadian clock, such as day length sensitivity?
- Has selection acted on targets of the circadian clock TFs to help maize adapt to northern latitudes?

These initial projects will be performed using maize, which has a wealth of available genomic resources and rich phenotypic datasets. However, because the questions I am asking deal with conserved, fundamental aspects of gene regulation, adaptation, and GxE, findings from these studies are likely to extend to other crop species. Learning to identify regions of the genome where certain mechanisms of regulation are likely taking place is the first step to building models of gene expression regulation between individuals. In the long term, my research group will work to piece together mechanisms into cohesive models of allelic regulation, GxE, and adaptation that can be applied across economically important crop species.

I love my career as a scientist. My path has been enabled by patient and supportive mentors, a friendly professional network, financial stability, speaking english as a primary language, privileged demographics, and an understanding of the 'hidden curriculum' — unspoken norms and customs within the scientific community. From my position of privilege, it is my responsibility — as a teacher, mentor, member of the community, and leader of a research group — to enable others to also love their scientific education and careers. This means doing my absolute best to create a supportive environment while also trying to recognize and remove obstacles to success.

The leaders of my current lab have put visible effort into creating and maintaining a safe, inclusive lab culture, which has taught me a lot about working toward an equitable and inclusive work environment. The largest lesson that I've taken away is that creating a safe and equitable lab is an *active* and *ongoing* process. This process starts with simple things, like being flexible around remote work and working hours so that members can attend to their own health and family needs first. It continues with more difficult aims that take sustained effort, such as creating an atmosphere of psychological safety, where members feel comfortable asking questions and challenging norms. All members of the lab have rotated through leading weekly discussions on a self-chosen topic related to diversity, equity, and inclusion in STEM. These have been eye-opening for me with regard to the scope and high dimensionality of the inequities that exist in our field of work. I have also had the opportunity to participate in a multi-departmental diversity and inclusion working group, during which I gained an appreciation for the extent of inequities and discrimination that exist at the administative/organizational levels in universities.

At NCSU, I will *continue* to learn how I can help combat discrimination and inequity, while also taking *active* steps to make my research group and classroom safe, equitable spaces. My first step will be to make expectations exceedingly clear so that all members of the lab and/or classroom know my expectations of them, as well as what they can expect of me. Living, evolving documents that outline expectations within the classroom and research group are becoming common, providing examples that I can take inspiration from and improve upon. Certain expectations will vary between undergraduates, graduate students, postdocs, and technicians, such as hours worked, leadership, and independence, whereas other expectations will be the same for everyone: mutual respect, accountability, integrity, and honesty.

My second step will be to try and set the hiring process for my group in such a way as to reduce discrimination. This starts with thinking carefully about the text in job descriptions and the avenues by which they are distributed. When reviewing applications, I would like to try a 'reversed' process, where the first screening step is of de-identified, short (<200 words) written responses to standardized questions, which are scored against a detailed rubric. After applicants are winnowed based on their responses, CVs are collected and evaluated. I hope that this will reduce my own implicit biases toward gender, ethnicity, publication record, experience, or professional network, and instead allow applicants to make a case for themselves through short, written statements. This will be a first step toward building a research group with diverse backgrounds, not only demographically, but also in terms of experience and expertise.

Third, I would like to compile a list of on-campus groups and resources for underrepresented minorities and encourage lab members to engage with those groups. My understanding is that a large contributor to underrepresented minorities leaving academic research is a feeling of isolation and lack of community. I want my research group to provide a support network and community for all of its members. However, I also realize

that external groups can help provide that support network and sense of identity in a way that a research group, particularly a new one, may not be able to.

These actions are small and will not solve the problem of pervasive, institutionalized inequity. However, they will set the tone for my research group and hopefully serve as a model for others. As researchers, we know that large problems cannot be solved in a single leap. Small, local steps and incremental progress pave the way towards larger change. I am committed to continually educating and improving myself throughout my tenure at NCSU; to making a safe, equitable, and enjoyable space for others to work; and to see the department and university evolve into an even better place than when I arrived.

Teaching Statement Joseph Gage

My teaching philosophy is built on the idea that a passionate and open-minded teacher can change the trajectory of a student's career and life. This is informed by my own experiences as an undergraduate: during my third year of college, I found myself (then a communications major) in a plant breeding and food production class that captivated my interest. Though I had little scientific background and no research experience, the teacher of that class cultivated an interest in science that led me to my career as a plant geneticist. My goal as a teacher is to create an environment in which students of all backgrounds are able to find joy and excitement through learning. I recognize that my own trajectory as a scientist has been enabled by patient, supportive teachers and mentors, and I want to pay that investment forward to the next generation of scientists.

Classroom teaching

During graduate school, I spent four semesters as a volunteer TA for graduate level plant breeding courses. During those semesters, I developed an appreciation for how varied each class is with regard to prerequisite knowledge, experience applying concepts, modes of learning, and roadblocks to success. Informed by that experience, as a faculty member I will design my classes in such a way as to accommodate the wide array of students that will sit in my classroom each semester: this means making lectures and notes available before and after topics are covered in class; designing assignments or exams in such a way as to ensure students have the time and resources necessary to succeed; making myself and TAs available for individual tutoring or help outside of the classroom; and remaining open to feedback and being willing to change.

I plan to design classes that incorporate hands-on work with real datasets. Having students do reading outside of class and work on projects together during class hours will enable them to study background material at their own pace and then bring questions about unclear concepts or modes of application to myself and their peers in the classroom. Recently I have seen assignments in which students are asked to write a common program or algorithm (e.g., GWAS, Fst, or a sequence aligner) from scratch, using the original publication and code (if available) as a guide. This type of assignment is a great way for students to become familiar with the tools that they will be using frequently during their tenure as graduate students and possibly the rest of their careers.

In addition to my time as a TA, I was fortunate to spend one semester as a member of the Statistical Consulting Lab at the University of Wisconsin. During my one-on-one advising sessions, I learned to listen patiently to background information, identify roadblocks that the researcher explicitly stated or had not identified yet, and guide them to a solution by asking questions rather than imposing my own idea of the 'right answer'. That experience gave me an appreciation for one-on-one work that I believe will translate well to office hours, tutoring, and mentoring students in my research group.

Mentoring researchers

I have had the opportunity to supervise and mentor undergraduates during both my graduate and postdoctoral positions. My graduate studies had a large field component and during the summers I managed 2-5 undergraduates daily to plant experiments, complete pollinations, collect samples, phenotype populations, and harvest seed. As a postdoc, I was fortunate to mentor an engineering student who maintained and operated a phenotyping robot during the summer. Our work together resulted in a publication, and he is now the founder of a remote sensing startup company. I was also able to work with several computer science students who were interested in field-based robotics, and am currently mentoring a sophomore math, computer science, and statistics major who is working on a project to map genotype-by-environment and genotype-by-time interactions.

Teaching Statement Joseph Gage

Participation in the Cornell Postdoctoral Leadership program, a 10-module course, taught me about managing conflict, recognizing biases, and promoting diversity within my research group. My experiences working with mathematicians, physicists, computer scientists, biologists, and engineers have shown me the value of recruiting and training a diverse lab group. As leader of a research group, I will strive to be available and supportive while also helping members achieve independence and self-worth through ownership of their research. Teaching and mentoring have always been a pleasure for me, and I look forward to training many more scientists yet to come.