Title: Developing mutant resources for pennycress to domesticate and improve crop resilience

Ratan Chopra[1,2*] (rchopra@umn.edu), M. David Marks[2], **John Sedbrook[3]**

[1]Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN 55108
[2]Department of Plant and Microbial Biology, University of Minnesota, St. Paul, MN 55108
[3]School of Biological Sciences, Illinois State University, Normal, IL 61790 USA

Project Goals: First-generation domesticated pennycress varieties have limited genetic variation, which hampers their adaptability and resilience against abiotic and biotic challenges. Therefore, crucial work remains to identify genetic variants conferring stress tolerance and resilience for incorporation into next generation elite pennycress varieties. Future pennycress varieties will also require optimized lifespans for a range of latitudes and cropping systems, and improved root architectures and physiologies to maximize water and nutrient scavenging as well as carbon sequestration. To attain these goals, we are developing mutant resources to screen for resilience traits or variants in candidate genes identified using eco-evolutionary computational genomics.

Abstract: The oilseed species *Thlaspi arvense* (pennycress)—a weed that was only recently removed from the wild—has the potential to provide new sources of food and bioproducts when grown as a winter cover crop without requiring new land. By taking advantage of extensive gene and phenotype knowledge in the related plant *Arabidopsis*[1], progress has been made to rapidly identify and stack crucial traits needed to domesticate the plant, allowing it to fit within current crop cycles and to have improved seed harvestability and nutritional content[2]. The first-generation domestication varieties have limited genetic variation, which hampers their adaptability and resilience against abiotic and biotic challenges. Much work has been performed using *Arabidopsis* as a model to identify gene mutations that may improve resilience. We have used classical mutagenesis and whole genome sequencing to recreate and characterize such mutations in pennycress. To date, we have whole-genome re-sequenced ~500 EMS mutagenized pennycress lines. A total of ~2.7 million mutations were identified and the mutation rate per line was estimated to be an average of 14.09 variants/Mb in the EMS population. Functional annotation of SNPs generated by EMS treatments suggested 82% of the pennycress genes had one or more alleles. Of these, 5,000 genes contained a mutation-introduced stop codon. We are on track to sequence an additional 500 mutants to obtain more than two alleles in each of the predicted genes of pennycress. We anticipate that this dataset will contain a spectrum of weak to strong alleles for genes previously identified in Arabidopsis that are predicted to improve resilience. We will be presenting the progress on the development of this mutant resource.

References
1. Chopra, R. *et al.* Translational genomics using Arabidopsis as a model enables the characterization of pennycress genes through forward and reverse genetics. *Plant J.* (2018). doi:10.1111/tpj.14147
2. Chopra, R. *et al.* Identification and stacking of crucial traits required for the domestication of pennycress. *Nat. Food* **1**, 84–91 (2020).