



Minnesota Wheat Research and Promotion Council

RESEARCH PROPOSAL GRANT APPLICATION

1. NAME AND ADDRESS OF ORGANIZATION TO WHICH AWARD SHOULD BE MADE Name: Regents of the University of Minnesota Address: Sponsored Projects Administration 450 McNamara Alumni Center, 200 Oak Street SE Minneapolis, MN 55455-2070		
2. TITLE OF PROPOSAL Identification of Bacterial Leaf Streak (BLS) resistance in Minnesota Germplasm		
3. PRINCIPAL INVESTIGATOR(S) Rebecca Curland <hr/> PI# 2 Name: James Anderson <hr/> PI# 3 Name: Ruth Dill-Macky	4. PI #1 BUSINESS ADDRESS Department of Plant Pathology 495 Borlaug Hall, 1991 Buford Circle University of Minnesota St. Paul, MN 55108	
5. PROPOSED PROJECT DATES (calendar years) January 1, 2024 – December 31, 2024 <small>Note: Research Reports are Due November 15th of Each Year</small>	6. TOTAL PROJECT COST \$17,546	7. PI #1 PHONE NO. 612-301-1107
8. RESEARCH OBJECTIVES: (List objectives to be accomplished by research grant) <ul style="list-style-type: none"> Screen 74 hard red spring (HRS) wheat lines in inoculated bacterial leaf streak (BLS) nurseries at two locations (St. Paul and Crookston) to complete phenotyping for genome wide association study (GWAS). Perform GWAS on 197 HRS wheat lines to predict genes associated with resistance to BLS. Screen additional bi-parental mapping population plus two parents (97 lines total) in two locations (St. Paul and Crookston) and perform bi-parental trait mapping. <p>Attach a 2-page detailed discussion of importance of the proposal to wheat profitability; how study complements previous research in area; procedures to be used; and competency of the research group in achieving research objectives. (Please keep the proposal concise, only 2 pages will be provided reviewers).</p>		
Signature Of Principal Investigator 	Date 1/4/24	Phone Number 612-301-1107
Signature Of Authorized Representative 	Title PGCO	Date 1/8/2024
Address Of Authorized Representative Sponsored Projects Administration 450 McNamara Alumni Center, 200 Oak Street SE Minneapolis, MN 55455-2070 awards@umn.edu		Phone Number (612) 624-5599

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RESEARCH PROPOSAL GRANT APPLICATION

(2-pages maximum)

Project Title: Identification of Bacterial Leaf Streak (BLS) resistance in Minnesota Germplasm

Importance of this project to the profitability of wheat producers:

This project focuses on efforts to control bacterial leaf streak (BLS) by identifying sources of resistance in Minnesota wheat. We propose to perform a genome-wide association study (GWAS) and bi-parental mapping on an array of wheat lines from the UMN wheat program. We will generate phenotypic and genotypic data that will be used in analyses to identify potential sources of resistance to bacterial leaf streak (BLS) within the wheat genome. The results of these studies will inform the UMN wheat program in making selections that contain genes that affect increased resistance to BLS. Ultimately, our goal is to provide Minnesota producers with wheat lines that demonstrate strong resistance to BLS to mitigate the economic impacts of this disease.

Procedures:

Wheat seeds will be planted in irrigated nurseries consisting of two-row plots, 3 feet in length and replicated twice at each location. Plots will be inoculated at the boot stage using our standardized protocol of applying a bacterial suspension using a gas-powered backpack sprayer. We will apply a well-characterized bacterial strain (CIX40), which was originally collected in MN in 2010 and has been consistently used in all our inoculated nurseries (Curland et al. 2018). Disease assessments will be collected after two weeks, with additional ratings collected as the disease is monitored over the growing season.

Genotyping by sequencing (GBS; Elshire et al. 2011) uses next-generation DNA sequence technology (Illumina) to obtain single nucleotide polymorphism (SNP) markers across the entire genome. These SNP markers can be used in genome-wide association studies (GWAS; Yu and Buckler 2006) to discover SNP markers that can be used in marker-assisted selection for traits of interest. A panel of 74 advanced experimental breeding lines that were phenotyped in the BLS cooperative nurseries in a minimum of 4 environments will be genotyped using GBS. The samples will first be prepared for DNA sequencing in our lab, then submitted to the University of Minnesota's Genomics Center (UMGC) to run on the Illumina sequencer. The short DNA sequence reads from each individual line will be compared to the wheat reference genome (Zhu et al. 2021) to find the SNP markers and determine their physical positions in the genome. Data from these 74 lines will be combined with the GBS data from the 123 cultivars and advanced breeding lines we previously genotyped and phenotyped. GWAS will be performed using the phenotypic and genotypic data for the complete panel of 197 lines using a specialized software called GAPIT 3.0.1 (Lipka et al., 2012) to find SNP markers that have a significant effect on BLS severity in the field.

Additionally, a genetic mapping population of 95 lines plus their two parents (MN11394-6 (R) X MS Barracuda (S)) has been screened in two environments. These lines will be screened again in Crookston and St. Paul in 2024 and genotyped using GBS. Biparental trait mapping will be performed in R using the 'qtl' software (Arends et al., 2010; Broman et al., 2003).

Regional linkages to other research activities:

Since 2013, the University of Minnesota's Small Grains Pathology lab has coordinated a cooperative BLS screening nursery. This inoculated nursery is run at four locations each growing season: St. Paul and Crookston (MN), Fargo (ND), and Brookings (SD). Dr. Shaukat Ali (South Dakota State University) manages the Brookings nursery and Dr. Zhaohui Liu (North Dakota State University) oversees the Fargo nursery. The historical data from these nurseries will be used in the GWAS analysis.

List any other secured, pending, or planned submissions to outside funding sources for this work:

Support for the Bacterial Leaf Streak Cooperative Nursery (BLSCN) is provided by the Small Grains Initiative grant "Evaluation of small grains cereals for multiple disease resistance" (\$99,015 for 7/23-6/24). The Dill-Macky lab has additional funds that will cover the GBS (genotype by sequencing) costs (estimated at \$5, 211.44).

Research Group:

Rebecca Curland (Research Scientist, UMN Plant Pathology) is the PI who will lead the project and oversee the field trials and data collection.

Ruth Dill-Macky (Professor and Plant Pathology Department Head) is a co-PI who will advise this project and whose lab will support the pathology work.

James Anderson (Professor, Agronomy and Plant Genetics) is a co-PI who will advise this project and whose lab will

support the GWAS and bi-parental mapping analyses.

Emily Conley (Research Scientist, Agronomy and Plant Genetics) will complete the GWAS and bi-parental mapping analyses.

Relationship to past projects:

This project builds on methods and data from previous projects. The MNWRPC has provided funding that has allowed us to make significant progress in developing screening methods for BLS, generating phenotype data for Minnesota wheat lines, and exploring management options for BLS control. The goals of this project represent a culmination of previous work that will enhance UMN's wheat breeding program's ability to select for BLS resistance.

Estimate the budget requirements:

Wages and fringe benefits: \$10,136 salary and fringe for the support of Rebecca Curland, Plant Pathology technician.

\$5,210 salary and fringe to support Emily Conley, Agronomy technician. \$1,200 to support undergraduate technicians to manage the field plots.

Materials and Supplies: \$300 for lab supplies for culturing bacteria and inoculum production.

Other Direct Costs: \$250 for land rental and \$450 for publication costs.

References:

Arends, Danny, et al. "R/qt1: high-throughput multiple QTL mapping." *Bioinformatics* 26.23 (2010): 2990-2992.

Broman, Karl W., et al. "R/qt1: QTL mapping in experimental crosses." *bioinformatics* 19.7 (2003): 889-890.

Curland, R.D., Gao, L., Bull, C.T., Vinatzer, B., Dill-Macky, R., Von Eck, L., and Ishimaru, C.A. 2018. Genetic diversity and virulence of wheat and barley strains of *Xanthomonas translucens* from the Upper Midwestern United States. *Phytopathology*, 108:443-453.

Elshire, Robert J., et al. "A robust, simple genotyping-by-sequencing (GBS) approach for high diversity species." *PloS one* 6.5 (2011): e19379.

Lipka, Alexander E., et al. "GAPIT: genome association and prediction integrated tool." *Bioinformatics* 28.18 (2012): 2397-2399.

Yu, Jianming, and Edward S. Buckler. "Genetic association mapping and genome organization of maize." *Current opinion in biotechnology* 17.2 (2006): 155-160.

Zhu, Tingting, et al. "Optical maps refine the bread wheat *Triticum aestivum* cv. Chinese Spring genome assembly." *The Plant Journal* 107.1 (2021): 303-314.

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RESEARCH PROJECT PROPOSAL BUDGET

Project Title: Bacterial Leaf Streak (BLS) resistance in Minnesota Germplasm			
Principal Investigator(s) / Project Director(s) Rebecca Curland	Funds Requested For		
	Year 1 (2024)	Year 2 (2025)	Year 3 (2026)
A. Salaries and Wages	\$12,496	\$	\$
1. Co-principal Investigator(s)	\$7,393		
2. Senior Associates			
3. Research Associates – Post Doctorate			
4. Other Professionals	\$3,903		
5. Graduate Students			
6. Prebaccalaureate Students	\$1,200		
7. Secretarial - Clerical			
8. Technical, Shop and Other			
B. Fringe Benefits	\$4,050		
C. Consulting and Professional Services			
D. Supplies and Services	\$300		
E. Travel			
F. Sub-Contracts			
G. Repairs & Maintenance			
H. Rentals & Lease			
I. Other Expenses	\$700		
TOTAL AMOUNT OF THIS REQUEST (per year)	\$ 17,546	\$	\$