PROJECT NO: BJKU12, BJKU35, BJKU13, BJKU67

TITLE: Evaluating Ecological and Chemical Approaches to Manage Wireworms, With Respect to Genetic Variations Among Populations and Microbial Associations

PERSONNEL: A. Rashed, C. W. Rogers, J. M. Marshall, X. Liang

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JUSTIFICATION: Wireworms remain to be one of the major pest concerns in Idaho cereal production. It is of a particular importance, due to the lack of effective pesticides registered for application in cereals. Therefore, we continued our research to better understand their ecology and population genetics, in order to develop more effective and sustainable management options.

Variations in the effectiveness of insecticides had been, in part, attributed to the among-species differences in susceptibility. To date, our area-wide survey of wireworms in approximately 130 fields across southern Idaho resulted in identification of 10 pest species of wireworm in barley and wheat fields. We have also collected relevant soil physical, chemical, and biological data to evaluate their impact on wireworm activity and to investigate species-specific habitat preferences. The impact of various cultural practices including tillage, crop rotation and seed treatment on individual species is currently being analyzed.

Throughout our studies, we have received an exemplary support from Idaho growers. Currently, more than 36 growers are participating in our monitoring program; the largest ongoing wireworm study in the Pacific Northwest region. We have also established new collaborations with colleagues on-campus, and in Washington state (WSU and USDA-ARS) and Oregon State University. In addition, we are collaborating with colleagues in Canada to understand genetic variations both within and among species in the region.

The sugar beet wireworm *Limonius californicus* is now confirmed as the most prevalent species in Idaho. As such, this species has become the focus of our management and population genetics studies. The other two species largely populating eastern counties were identified as *Hyponoidus bicolor* and the western field wireworm *L. infuscatus*. We have also shown that multiple species may be present within a field and that the time of activity of each species dependents on mostly abiotic environmental variables. We reported significant relationships between soil bulk density/soil texture and wireworm numbers, pointing to the importance of seed-bed preparation in minimizing losses due to wireworm damage.

Our preliminary evaluations of the microbes associated with the sugar beet wireworms, revealed significant variations across southern Idaho; the presence and density of certain microbes (i.e., endosymbionts) is known to impact insect susceptibility to insecticides. Moreover, our results indicated that the higher tillering barley is relatively more tolerant to wireworm damage than the lower tillering wheat. Our findings within the past four years have opened new avenues of research, which includes the efficacy trials with biological control agents as a component of IPM approach. We are also continuing our research on short-term management solutions by evaluating efficacy of different insecticides against wireworms, as well as cultivar tolerance to wireworm damage.

The continuous support from the Idaho Barley Commission as well as growers' involvement in our research have been precursors to the success of this collaborative effort,

which also attracted support from competitive federal sources of funding. We propose to continue this well-established interdisciplinary study by addressing the following hypothesis and objectives.

Hypothesis: The degree of wireworm damage is species-dependent and a clear understanding of their interaction with the surrounding biotic and abiotic environment will assist with developing integrated management practices to minimize their negative impact. To evaluate this hypothesis the following objectives are proposed:

- I) Continue wireworm monitoring to collect sufficient number of samples for genetic analysis of wireworm populations (focusing on *L. californicus*) across southern Idaho, and to provide growers with updates of wireworm species and numbers in their fields,
- II) Quantify L. californicus susceptibility to neonicotinoids with respect to their associated microbe species and densities,
- III) Study the impact of crop rotation options, and inter-cropping, on wireworm pressure and damage,
- IV) Quantify among-cultivar differences in susceptibility to wireworms, in relation to tiller number and root structure,
- V) Quantify variations in wireworm damage in response to heat/drought stress in relation to soil organic matter content.

PROCEDURES: To address Objective I, we will continue to monitor wireworm populations in ~130 fields in southern Idaho. Baits will be placed at 6 inches deep (minimum of three traps in each field), throughout the growing season, in every field starting 2 weeks before planting until after harvest in October or November, depending on the location. Traps will be replaced every 4 weeks. The DNA of the trapped wireworms will be extracted. Subsamples will be provided to IBEST for genetic variability analysis. Another subset will be analyzed for the presence of endosymbiont DNAs, using available universal primers for bacterial endosymbionts. As a part of Objective II, sugar beet wireworms hosting different endosymbiotic communities will be evaluated for their susceptibility to the commonly used seed treatments. A wireworm-infested field located at the University of Idaho, Kimberly R&E Center will be used to setup study plots to compare the impact of recommended rotation crops for dryland and irrigated cropping systems (i.e. peas, quinoa, mustard, radish) on wireworm pressure as well as to evaluate their role as trap crops to divert wireworm damage away from barley and wheat at their susceptible seedling stage (Objective III). Since anecdotal observations suggested a greater wireworm damage to cereals following beans, we also propose to evaluate the effectiveness of its use as an inter-crop to divert potential damage away from the seeded barley (or wheat, depending on the trial) at the initial stages of its growth. The first-year trial of intercropping is now completed. To address Objective IV, a selection of wheat and barley varieties will be obtained from the USDA National Small Grains Collection in Aberdeen, ID. This objective is to evaluate whether losses to wireworms could be reduced by using varieties with relatively more robust tiller production and root system. Initial evaluations did not detect reduced damage in high-tiller cultivars. As such, the second year of our study will be focused on evaluating the potential relationship between the extent of root system and yield-loss to L. californicus. We have now equipped our labs with gas chromatography and high-performance liquid chromatograph to quantify any potential effects in relation to root volatiles. In the face of our ever-changing climate and continuous shortage of water, Objective V is set to quantify losses to the sugar beet wireworm in drought/heat stressed plants, a key step towards providing an objective measure to predict the extend of yield loss. This assessment will be conducted in relation to soil organic matter content starting 2018; organic matter may not only reduce wireworm damage, it may also assist with increasing water-holding capacity of the soil.

DURATION: Spring 2018 through Spring 2019.

COOPERATION: This is a cooperative project between Drs. Rashed, Rogers, Marshall and Liang at the Aberdeen R & E Center. In established collaborations with IBEST, USDA-ARS Wapato, and WSU, we will continue to provide DNAs of the collected wireworm species for genetic assessments and to quantify endosymbionts in various soil types. Our work and success would not have been possible without strong support and active collaboration from Idaho cereal producer community.

ANTICIPATED BENEFITS/EXPECTED OUTCOMES/INFORMATION TRANSFER:

This proposal is built upon our most recent findings of wireworm ecology and species composition, based on our 2014-2017 efforts, and is now expanded to address new objectives that are directly relevant to efficacy of the currently available seed treatments, and developing cultural and biological control practices that may limit wireworm damage. To date, our collaborative efforts have resulted in the first-time publication of the full mitochondrial sequence of the sugar beet wireworm, the first published comparison of wireworm damage in wheat and barley in relation to seeding depth and soil texture, and a comprehensive visual key to wireworms and management options in Idaho. We are currently working one manuscript in final stages of preparation in collaboration with WSU collaborators. We also co-authored one new wireworm bulletin published by the Oregon State University.

LITERATURE REVIEW:

Historically, wireworms were controlled by using environmentally persistent chemicals that are now banned due to environmental and health concerns (Vernon et al 2009). Although substitute chemicals provided some level of protection, wireworm susceptibility to the new generation of chemicals varied depending on the species (Van Herk et al. 2007) and the insect pressure. Moreover, anecdotal observations suggest potential among-population variability in response to neonicotinoid seed treatments within species.

Recently, more emphasis has been placed on alternative control measures to achieve more successful and sustainable damage control in conjunction with seed treatments. The effectiveness of any IPM program in controlling wireworms depends on a clear knowledge of the present species and their ecology (Furlan 2005). The sugar beet wireworm *Limonius californicus* is known to be the most damaging species to cereals in Idaho and is believed to show inconsistent degrees of susceptibility to neonicotinoid seed treatments (Rashed et al. 2015). Relatively recent studies have indicated that the susceptibility to imidacloprid and other insecticides can be affected by pest association with its microbial communities (Ghanim and Kontsedalov 2009).

Unfortunately, knowledge of wireworm ecology and control is generic without taking among-species variations into account. For instance, wireworm species may vary in the range of plant species they feed on (Pazmand and Traugott 2005) and the soil characteristics (such as pH, texture, organic matter, and moisture) where they reside (Barsics et al. 2013). It has been also shown that soil type (Rashed et al. 2016), CO₂ (Barsics et al 2013; Gfeller et al. 2013), and volatile organic compounds released from roots (Gfeller et al. 2013) can affect wireworm movement and survival. Such ecological traits can be used in trap cropping and baiting various wireworm species as a part of a sustainable IPM program.

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IDAHO WHEAT COMMISSION - BUDGET FORM Principal Investigator: Arash Rashed \$ 14,615 Idaho Wheat Commission during FY 2017 Allocated by \$ 15,444 during FY 2018 Idaho Wheat Commission Allocated by REQUESTED FY2019 SUPPORT: Salary (staff, Graduate post-docs, Temporary Tuition/Fees **TOTALS** OE etc.) Help Fringe Travel 3,100 \$ S 15,268 4,200 \$ 6,000 1,968 Idaho Wheat Commission \$ 15,268 **TOTAL BUDGET REQUEST FOR FY 2019:** BREAKDOWN FOR MULTIPLE SUB-BUDGETS: Marshall Chris Rogers Xi Liang Arash Rashed \$ S \$ Salary \$ 1,000 S 4,000 \$ 1,000 \$ Temporary Help \$ S 328 328 S 1,312 \$ Fringe Benefits \$ 700 S 500 \$ 500 \$ 2,500 S Travel 500 1,000 \$ 500 \$ 1,100 S \$ **Operating Expenses** \$ S

3,128 S 1,000

15,268

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Total Sub-budgets S

\$

8,312 \$

Explanatory Comments: (see FY2019 RFP for definition)

Fall 2017 Version

TOTALS

Graduate Student Tultion/Fees

ANNUAL REPORT

PROJECT NO: BJKU12, BJKU35, BJKU13, BJKU67

Evaluating ecological and chemical approaches to manage wireworms, with TITLE:

respect to genetic variations among populations and microbial associations

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ACCOMPLISHMENTS:

More than 130 irrigated and dryland cereal fields have been monitored in 14 counties across southern Idaho over the past four growing seasons. Wireworm traps were continuously replaced throughout the year (with the exception of December and January) on a 3 to 4-week schedule. Traps remained in the soil for approximately 2 weeks, starting 2 weeks before planting. Thirty-five growers in southern Idaho participated in our 2017 monitoring program. So far we have identified 10 species of wireworms; 8 of those species were also observed in 2017. At each sampling date, soil moisture and temperature data were recorded. Soil bulk density, texture (i.e., sand, silt, clay), inorganic carbon (i.e., carbonates) and total nitrogen and carbon contents were characterized on a single sample per field per year, as these properties fluctuate minimally within a short period of time (i.e., a few weeks or months). Findings were presented and will be updated at the 2018 cereal schools. We also continued our collaborations with the IBEST, by providing DNA samples from species collected throughout the 2017 season, and currently a student is hired to continue this effort for the 2018 to 2019 season. In this report, we briefly list our efforts related to each of the proposed objectives, and will focus on one of our latest studies that is in preparation for submission for publication.

Objective 1: Species survey

Soil samples C, N, soil texture, and pH for the 2017 season has been collected and currently are being organized and analyzed in Dr. Rogers laboratory. A combined 2015 to 2017 dataset will be analyzed and presented at the 2018 cereal schools.

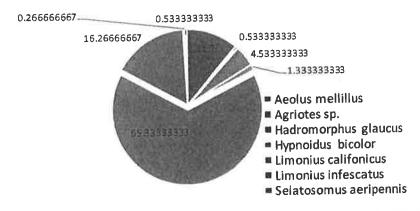


Figure 1: Eight species of damaging wireworms were collected during the 2017 season. Majority of the collected species (65.33%) were sugar beet wireworms. Other species collected and their prevalence are illustrated in the figure.

Objective 2: Wireworm endosymbionts and susceptibility to neonicotinoids (L. califonicus)

The DNA from sugar beet wireworms samples, collected across central and eastern Idaho, have been extracted and sent to IBEST for detection and quantification of microbial communities with respect to the collection site. A meeting was organized with colleagues at IBEST. Dr. William R. Cooper from USDA-ARS, Wapato, WA, was invited to join our meeting as an expert on endosymbionts. Approaches have been finalized, and Atoosa Nikoukar, Dr. Rashed's new MS student, will be conducting the molecular analyses of the collected samples.

Objectives 3 and 4: To address the effectiveness of intercropping we have completed our first-year field study and data will be presented at the 2018 cereal schools. The purchase of a new planter by the University of Idaho would facilitate our intercropping research in the 2018 season; this was a major limiting factor in the first year. We also continued our screening of the susceptibility of high-tiller and low-tiller number wheat varieties to sugar beet wireworms. Four replicated experiments have been completed where no significant difference has been detected between high- and low-tiller varieties to date. It appears that both high-tiller and low-tiller wheat varieties are equally susceptible to wireworms and that relatively higher tolerance of barley to wireworms is unlikely related to tiller numbers. This indicates that root oxidases perhaps play a critical role in rendering barley relatively more tolerant to wireworms. A detailed analysis will be presented at the research review. Our research on alternative management approaches in 2017 also included evaluations of biological control approaches against *L. californicus*.

The entomopathogenic nematode Steinernema carpocapsae and fungus Metarhizium anisopliae both showed potential to reduce damage to wheat seedlings, in some cases providing better protection than the CruiserMaxx seed treatment. Interestingly, the efficacy of the biological control agents depended on the soil type. Nematodes proved to be relatively more effective in sand-dominated soil, while entomopathogenic fungus showed relatively higher efficacy in peatmoss dominated soil (Figure 2). This indicates that recommendation of biological control agent must be made with the knowledge of field soil type. We are proceeding with field studies (to be conducted by WSU) to evaluate the persistence of these biocontrol agents in soil over time.

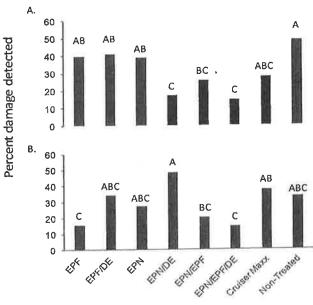


Figure 2: Percent feeding damage observed in sand-dominated (A) and peat-moss-rich media (B). While treatments that include entomopathogenic nematodes show effectiveness reducing wireworm damage in sand-dominated soil, the entomopathogenic fungus is more effective in protecting plants in a peat-moss dominated soil. Treatments include: non-infested (no wireworm), non-treated, seed treatment (CruiserMaxx), entomopathogenic fungus (EPF), entomopathogenic nematode (EPN), and the combination of the two, applied with (EPF/EPN/DE) and without (EPF/EPN)

Objective 5: This objective is new to this proposal and is set to quantify variations in wireworm damage under heat/drought stress conditions, in relation to soil organic matter content. Experimental assays to address this objective is scheduled for the spring of 2018.

Next Steps

Due to continuous need of wireworm samples by our program as well as our collaborative research with IBEST, we will continue our sampling program (wireworms cannot be reared in the laboratory). Monitoring data is being communicated to our cereal growers. Data collection for our phylogenetic studies have been completed and we are hoping to have this data finalized in the upcoming year. The role of genetic variations among populations in susceptibility to seed treatments will be evaluated through this collaborative research. We will continue our research on developing more effective and sustainable cultural control measures that are specific to dryland and irrigated cropping systems. We have developed and tested several new objectives. For the first time, we are identifying and quantifying endosymbionts (microbes) associated with wireworms, in collaboration with colleagues at the USDA-ARS in Wapato (WA) and IBEST. Variations in endosymbiotic microbes have been observed in samples collected across southern Idaho; endosymbionts are known to influence insect susceptibility to pesticides. We will also continue our research on alternative management approaches, including cultural and biological management. Our research on cover crops and trap cropping was started last year (in the field) and will continue in 2018. The impact of water and heat stress on wireworm damage is set to be evaluated in 2018.

PROJECTIONS:

The ongoing support of wireworm survey research provided resources to continuously collect samples for the initiation of original research topics in Idaho. This includes studies of endosymbionts, phylogenetic analyses of wireworm populations, and array of various experiments to evaluate the efficacy of cultural and biological control practices. Our survey is now the largest study in the US and results will soon be finalized. Dr. Rashed's program is hoping to hire a postdoctoral scientist dedicated to analyze and finalize survey data. We now have one manuscript in the final stage of preparation and one published extension bulletin based on our activities in 2017. This trend will continue in 2018-2019 season.

PUBLICATIONS:

- Ensafi, P., Crowder, D., Esser, A., Marshall, J., Rashed, A. Natural enemies in wireworm management: Evaluating the effectiveness of the entomophagous fungus *Metarhizium anisopliae* and the parasitic nematode *Steinernema carpocapsae* in reducing damage by the Sugar beet wireworm *Limonius californicus* (Col., Elateridae). *In final stages of preparation*.
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