Grant Code:	AN2638
Title:	Grassy weeds, aphid population and BYDV spread, with a particular emphasis on perennial hosts
Personnel:	Arash Rashed, Xi Liang, Nilsa Bosque-Perez, and Juliet Marshall
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Justification: *Barley yellow dwarf virus* (BYDV) is an important pathogen of cereal crops worldwide. BYDV has a wide host range that include many species of wild grasses. Such alternative hosts may serve as important virus reservoirs in the environment. BYDV can only be transmitted by cereal aphids. The bird cherry-oat aphid is known to be one of the most efficient vectors of BYDV; this aphid has also been associated with the outbreaks in Idaho (Marshall and Rashed, 2014). With only one exception in south eastern Idaho, all of the positive samples in southern Idaho were determined to be of the BYDV-PAV strain.

Recently there has been an increase in corn acreage in central and eastern Idaho. As corn is known to be a silent host of BYDV, expressing minimal to no symptom upon infection, it may also be a candidate host plant, perhaps explaining frequent BYDV outbreaks in southern Idaho. Between 2015 and 2017, we reported considerable presence of BYDV in corn, green foxtail and foxtail barley, which supported previous findings of BYDV prevalence in grassy weeds in northern Idaho pastures (Ingwell and Bosque-Perez, 2015). With support from IWC and through a series of experiments we also demonstrated that the bird cherry-oat aphids can successfully inoculate downy brome, green foxtail, foxtail barley, and corn with BYDV. The aphids were also able to effectively acquire the virus from these hosts and inoculate winter wheat seedlings. Throughout our studies we observed differences in aphid survival, and there appeared to be variations in aphid fecundity across different host species. Furthermore, the presence of BYDV in the perennial foxtail barley, raised the question as to whether this host plant can serve as overwintering hosts for BYDV, especially since the virus colonizes roots. The bird cherry-oat aphid also exhibits behavioral responses that can facilitate pathogen spread within a field; while they initially show preference towards infected hosts, they switch their preference toward uninfected hosts, after acquiring BYDV. The dynamic nature of behavioral responses to BYDV infection highlights the importance of also investigating aphid preferences between wild grassy hosts and cultivated wheat with respect to their BYDV infection status, and in the presence of common environmental stress factors, to explain vector and pathogen spillover and movement from reservoirs into the cultivated crop, and vice versa.

The persistence of BYDV outbreaks in the region may be due to an increase in the number of host plants that would support its overwintering, as well as multiplication of its aphid vectors, in the absence of wheat host. Thus, while we are planning to continue, and increase, aphid monitoring in the region. We are proposing a combination of research and service objectives to examine our overall hypothesis. First-year results are available for the objectives I, III and IV. Objectives II and V are currently ongoing.

Hypothesis & Objectives: Wild grasses may serve as both preferred host of the bird cherry-oat aphids and the overwintering host of BYDV and therefore contribute to frequent disease outbreaks in the region.

I. To determine cereal aphid species and their movement from wild BYDV reservoirs to winter wheat (service),

II. To evaluate the perennial foxtail barley as a potential overwintering host for the BYDV (service/research),

III. To evaluate aphid survival and reproduction on predominant Idaho grassy weed species in Idaho (research/service),

IV. To examine whether the infection status of the bird cherry oat aphids impacts their host preference (winter wheat vs wild hosts) (primarily research),

V. To determine the effect of abiotic environmental stress (i.e., drought) on aphid preference and survival on grassy weed hosts (research).

Procedures/Plan of work: As a part of **Objective I**, we will continue to identify aphid species and monitor their numbers in relation to the presence of corn and grassy weeds in the region. Weeds will be sampled and tested for the presence of BYDV using ELISA. To address **Objective II,** foxtail barley seeds have been obtained and will be planted in the field. Through a series of field cage experiments at the Kimberly and Aberdeen R&E Centers, plants will be inoculated with BYDV, using the bird cherry-oat aphids from an infected colony. All plants will be tested for BYDV in subsequent year(s) to evaluate BYDV persistence. To address **Objective III**, seeds of grassy weed species green foxtail, foxtail barley, downy brome, wild oat, and reed canary grass will be collected and planted in the greenhouse. Additional species may be considered as our flora survey continues. Aphids from BYDV-infected and clean colonies will be individually placed on wheat and weed seedlings (10 plants/species) and their survival and daily reproduction will be recorded over a 10-day period. This experiment will be repeated (over two years) to identify host plants that best support aphid survival and reproduction. For **Objective IV**, the winter wheat cv. SY Ovation, and the grassy weed species green foxtail, foxtail barley, downy brome, wild oat, and reed canary grass will be planted in the greenhouse. In a pairwise approach, aphids will be presented with a choice between wheat and each of the grassy weed species. Aphid choice for each plant will be recorded after 24 hours. Eight replications of this experiment will be conducted to determine whether aphids show a nonrandom preference toward various host plant species and whether host preference can be influenced by infection status of the vectors. To address **Objective V**, SY Ovation, and the above-listed grasses will be planted in the greenhouse. After emergence, some plants (8 plants/species) will be maintained at 50% of field capacity, and some (8 plants/species) will be watered to 100% field capacity. Assays described in Objectives III and IV will be used to evaluate the impact of environmental stress on the vector host preference as well as survival/reproduction.

Duration: The proposal is intended to provide preliminary date for pursuing federal funds. It would be the 2nd year of our 2-year proposal.

Cooperation: Proposed research is being carried out collaboratively among Dr. Rashed, Dr. Liang, Dr. Marshall and Dr. Bosque-Pérez's programs in cooperation with several regional cereal growers.

Anticipated Benefits, Expected Outcomes and Impacts, and Transfer of Information: In our previous proposal, we identified corn and several grassy weed species that can effectively act as sources of BYDV-PAV in the region. Here, we continue our survey and studies focusing on perennials and their ability to support bird cherry-oat aphid populations. We are investigating

aphid preference to explain patterns of aphid movement into cultivated fields under optimal and stress conditions. As a part of this proposal we will also provide growers with information on aphid numbers and BYDV incidence. This project is expected to help growers in BYDV management by identifying BYDV overwintering sites and host plants that may support large vector populations. Acknowledging the continuous support from Idaho Wheat Commission, our findings will be communicated to growers and the scientific community through cereal schools, conferences and, at least, one scientific and/or extension publication.

Literature Review:

Barley yellow dwarf is one of the most widespread and damaging viral diseases of cereal crops worldwide. In addition to small grains, corn and many other annual and perennial grass species (Mastari et al. 1998; Malmstrom et al. 2005; Catherall et al. 1966; D'Arcy, 1995) have been suggested as important BYDV reservoirs. Studies have also reported wild oat (*Avena fatua* L.), and several other exotic annual grasses as preferred hosts of cereal aphids such as *Rhopalosiphum padi*, *R. maidis* and *Sitobion avenae* (Malmstrom et al. 2005; Borer et al. 2009). Knowledge of potential BYDV reservoirs that facilitate its overwintering (i.e. perennial hosts) and information on the preferred host plant species of the cereal aphids that can support increases in aphid numbers will help to explain both the persistence of BYDV presence and factors that prompt virus/vector movement between patches (e.g., pasture to field). This information is not only critical in adapting effective cultural management practices (i.e. weed management) but will also assist in predicting frequent regional outbreaks of BYDV. Determining cereal aphid choice of cultivated or uncultivated hosts and its potential dependence on host plant and aphid infection status (see Ingwell et al. 2012) in the presence of abiotic stress factors has direct implications in explaining patterns of BYDV movement and spillover between reservoirs and wheat crop.

References:

Borer, E.T., Adams, V.T., Engler, G.A., Adams, A.L., Schumann, C.B., Seabloom. E.W. 2009. Aphid fecundity and grassland invasion: invader life history is the key. Ecological Applications 19: 1187–96.

Catherall, P. L. 1966. Effects of barley yellow dwarf virus on the growth and yield of single plants and simulated swards of perennial rye-grass. Annals of Applied Biology 57:155–162.

D'Arcy, C.J. 1995. Symptomology and host range of barley yellow dwarf. In: D'Arcy CJ, Burnett PA, eds. Barley Yellow Dwarf: 40 Years of Progress. St Paul, MN, USA: The American Phytopathological Society, 9–28.

Ingwell, L.L., Eigenbrode S.D., Bosque-Pérez, N.A. 2012. Plant viruses alter insect behavior to enhance their spread. Scientific Reports 2: 1-6.

Ingwell, L.L., Bosque-Pérez, N.A. 2015. New experimental hosts of Barley yellow dwarf virus among wild grasses, with implications for grassland habitats. Plant Pathology 64: 1300-1307.

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Marshall, J., Rashed, A. 2014. Barley yellow dwarf virus in Idaho cereal crops. University of Idaho, extension. CIS 1210. pp: 1-4.

Mastari, J., Lapierre, H., Dessens, J.T. 1998. Asymmetrical Distribution of Barley Yellow Dwarf Virus PAV Variants Between Host Plant Species. Phytopathology 88: 818-821.

FY2020

IDAHO WHEAT COMMISSION - BUDGET FORM

Principal Investigator: Arash Rashed

Allocated by	Idaho Wheat Commission	during FY 2018	$	-
Allocated by	Idaho Wheat Commission	during FY 2019	$	16,024

REQUESTED FY2020 SUPPORT:

Budget Categories	(10) Salaries (*staff, post-docs, etc.*)	(12) Temp Help	(11) Fringe	(20) Travel	(30) OE	(70) Graduate Tuition/ Fees	TOTALS
Idaho Wheat Commission	$ -	$ 4,800	$ 1,589	$ 3,200	$ 5,850	$ -	$ 15,439

TOTAL BUDGET REQUEST FOR FY 2020: $ 15,439

BREAKDOWN FOR MULTIPLE SUB-BUDGETS:

Budget Categories	*Rashed*	*Liang*	*Bosque-Perez*	*Marshall*
(10) Salaries	$ -	$ -	$ -	$ -
(12) Temp Help	$ 2,500	$ 2,300	$ -	$ -
(11) Fringe Benefits	$ 828	$ 761	$ -	$ -
(20) Travel	$ 2,200		$ 500	$ 500
(30) Other Expenses	$ 4,650	$ 1,200	$	$ -
(70) Graduate Student Tuition/F	$ -	$ -	$ -	$ -
TOTALS	$ 10,178	$ 4,261	$ 500	$ 500

Total Sub-budgets $ 15,439

Explanatory Comments:
Some adjustment in travel budgets resulted in reduced budget request

Fall 2018 Version

ANNUAL REPORT

Grant Code: AN2638

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Personnel: Arash Rashed, Xi Liang, Nilsa Bosque-Perez, and Juliet Marshall
Address: University of Idaho, Moscow/Aberdeen R & E Center, 875 Perimeter Dr., Moscow, ID 83844-2329; arashed@uidaho.edu

Accomplishments:

In the initial year of our study, we have started addressing all of our five proposed objectives. While some of our experiments are ongoing we have final results for some of the objectives and those are presented in this report. Based on results from our previously funded proposal, we showed that barley foxtail (*Hordeum jubatum*), green foxtail (*Setaria viridis*), downy brome (*Bromus tectorum*) and corn can all host *Barley yellow dwarf virus* (BYDV) and that the bird cherry-oat aphids are capable of both inoculating and acquiring BYDV from these hosts. Successful transmission into winter wheat (cv. SY Ovation) ranged between 45 and 95%. Aphid monitoring is currently ongoing in corn and winter wheat with only a small fraction of fields (and collected weeds) tested positive in 2018. Evaluations of BYDV status in southern Idaho will be conducted in April 2019.We have also caged patches of naturally present foxtail barley and infested them with the BYDV-infective aphids. These patches will be inspected next year to determine aphid survival and BYDV status. The detection of BYDV within the perennial foxtail barley would be indicative of the role of this plant as an overwintering host for the virus. Results of host choice and reproduction success experiments with the infective aphids have been completed and are presented below. Experiments to evaluate the effects of water stress on host choice and aphid reproduction are currently ongoing and results are expected to be analyzed by the time of our research review.

Results from the first experiments have been presented at the Joint Entomological Society of America/Entomological Society of Canada in Vancouver, Canada in 2018. Findings will also be presented in 2019 cereal schools and departmental seminars. In this report, we are focusing on our completed host choice and reproduction studies on BYDV- carrying aphids.

Aphid population dynamics and BYDV status

Aphid numbers were monitored on yellow sticky traps placed in corn, winter wheat and open pasture/natural vegetation between August and November of 2018. Figure 1 represents aphid numbers collected in September and October in Bingham, Power and Twin Falls counties. According to our yellow pan trap observations cereal aphids, specially the bird cherry-oat aphid *Rhopalosiphum padi* and the English grain aphid *Sitobion avenae* were abundantly represented in the collected samples from Magic Valley. Samples collected in Eastern Idaho, especially Swan Valley, were dominated by *R. rufiabdominale*, among other species.

Despite the large number of collected aphids in Twin Falls county (Figure 1), no BYDV was detected in that area in the Fall 2019 (ELISA). Fields will be sampled and tested again for the virus in the Spring. Only fields in Power county tested positive for BYDV. In Power county, one out of three corn fields, and one winter wheat field adjacent to corn tested positive for BYDV-PAV. Creeping bent grass (*Agrostis stolonifera*) and bristly foxtail (*Setaria parviflora*) were the

two weed species sampled from natural vegetation surrounding two fields, that tested positive for BYDV-PAV in Power county.

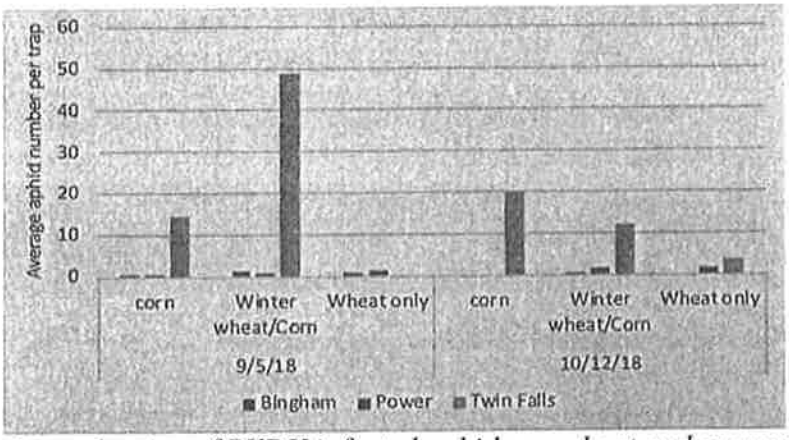

Figure 1. Average number of aphids collected per trap from corn, winter wheat adjacent to corn (winter wheat/corn) and wheat planted away from corn. Corn was monitored using three sticky traps. Wheat was monitored with traps arranged in two rows at 50 (three traps) and 150 (three traps) feet from the edge of the corn field.

Reproduction of BYDV infected aphids on wheat and grassy weeds

Field collected seeds were obtained from weed management program at the University of Idaho, Aberdeen R&E Center. Seeds were planted in flat trays and seedings were transplanted into 3*3*8-inch pots. A similar approach was used to transplant SY Ovation winter wheat. All plants were between 7 and 8 inches in height at the initiation of the experiment. One one-inch leaf cage was placed on a single leaf of each plant (weeds and winter wheat). Two adult aphids were released into each cage, and the number of nymphs were estimated 5 days after the release. The experiment was repeated twice and there were between 8 and 10 plants/species in each of the repeats. Generalized linear mixed model was used to analyze the data.

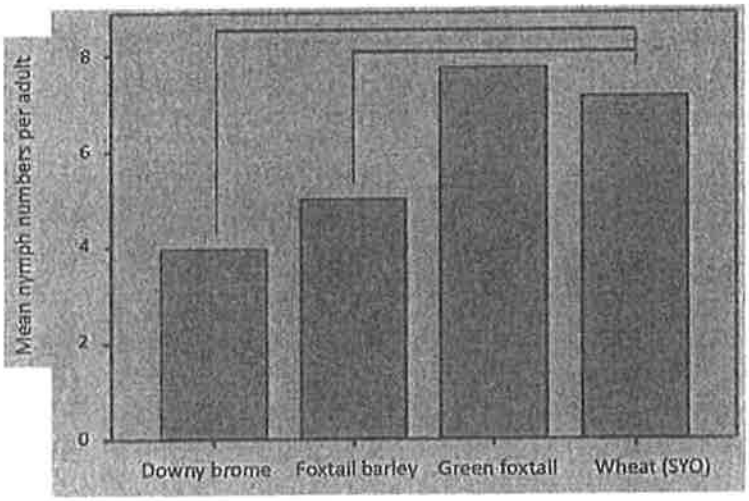

Figure 2: There was a significant difference in the number of the bird cherry-oat aphids produced on each host species ($F_{3,54} = 2.77$; $P = 0.050$). Average number of nymphs per female was significantly lower on Downy brome and Foxtail barley compared to winter wheat.

Host preference by the BYDV-infected bird cherry-oat aphids

To evaluate the aphid preference for weed species in the presence of winter wheat, using the one-inch leaf cages two BYDV-infected aphids were presented with a choice of a weed and a wheat leaf. The preference was estimated after 24 hrs. The presence of one aphid on each host, or both on the cage surface, was recorded as no preference.

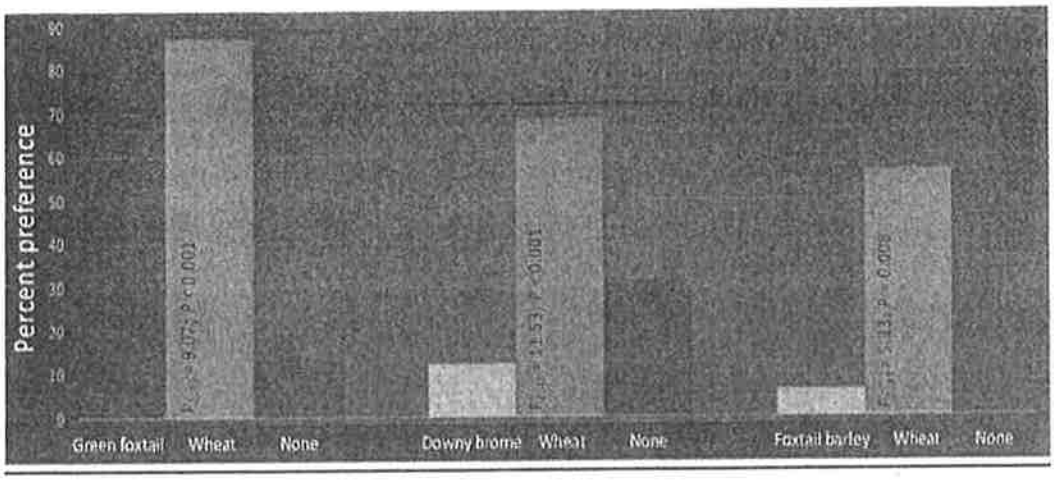

Figure 3: Overall, the bird cherry-oat aphids preferred winter wheat (cv. SY Ovation) over Green foxtail ($P < 0.001$), Downy brome ($P < 0.001$) and Foxtail barley ($P = 0.008$).

Through a previous proposal we demonstrated that Green foxtail, Foxtail barley and Downy brome are all reservoirs of BYDV in the region. This year we showed that the aphids are capable of reproducing on weedy hosts however reproduction was relatively higher on winter wheat and Green foxtail than Foxtail barley and downy brome (Figure 2). Moreover, the observed significant preference for winter wheat seedlings explains the movement of aphids into young winter wheat fields in the fall (Figure 3).

Projections:

An area wide sampling of wild grassy hosts will be continued for the spring of 2019. These include sampling approximately 20 field sites from southern Idaho. Although our first-year data indicates that high aphid numbers in winter wheat in the fall may be associated with corn, at least one more year of data is needed to confirm this pattern and to confirm any potential relationships between corn presence and the increase in BYDV incidence. So far, we have verified that five species of grassy weeds and corn are indeed reservoirs of the pathogen and can play a role in BYDV spread. We have also showed that aphids can survive and reproduce on three species of wild hosts, even though these three wild species were less preferred by the bird cherry oat aphids than winter wheat. This may explain why aphids start to move into winter wheat upon its emergence, and that the preference may be condition dependent. Studies are ongoing and will continue through 2019 to examine this possibility.

Publications:

Rashidi, M., Cruzado, R.K., Nikoukar, A., Hutchinson, P.J.S., Marshall, J., Bosque-Perez, N.A., Rashed, A. Barley yellow dwarf virus transmission by the bird cherry-oat aphid *Rhopalosiphum padi* L. (Hem., Aphididae): Can corn and grassy weeds contribute to regional BYDV outbreaks? Entomological Society of America, Entomological society of America Annual Meeting. Vancouver, BC, November 2018.

Rashidi, M., Cruzado, R.K., Nikoukar, A., Hutchinson, P.J.S., Marshall, J., Bosque-Perez, N.A., Rashed, A. Barley yellow dwarf virus transmission from wild grassy weeds to winter wheat. *In final stages of preparation for* Plant Disease.