AGRICULTURAL RESEARCH FOUNDATION
INTERIM REPORT, FUNDING CYCLE 2020 – 2022

TITLE: Evaluation of UV-C Seed Treatments for Controlling Seedborne Pathogens

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EXECUTIVE SUMMARY: Numerous fungi are known to be seedborne, including plant pathogenic strains of *Fusarium* and *Leptosphaeria* species. *Fusarium* affects a wide range of plant hosts and incites numerous types of diseases including wilts, seed rots, root rots, and crown diseases. *Leptosphaeria* is a well-known pathogen on vegetable crops, especially brassicas, and is a regulated seedborne pathogen of crucifer seed stocks. Seedborne pathogens are poised to infect plants at very young stages, when plants are more delicate and more vulnerable to disease caused by seedborne and soilborne pathogens. Fungal pathogens can be transported to unaffected farm fields via a seedborne presence, and those pathogens with capabilities of surviving in the soil, such as *Fusarium* spp., can then establish as a long term threat for susceptible crops in infested fields. Although scientists are relatively well-informed about the problem with seedborne *Fusarium* and *Leptosphaeria* in a subset of crops, such as corn, control of seedborne fungi is still an unmet challenge. Fungicide seed treatments do not eradicate the pathogens present but rather only delay infection of plants that develop from infected seeds. Physical treatments such as hot water and steam, or chemical disinfestation such as bleach treatments, can be used to minimize the effect of seedborne pathogens, but seed treatments that require wetting of the seed are limited in application due to logistical and practical considerations. Treatment of seed with UV-C would allow keeping seed dry and could be spun off into a technologically-viable alternative if UV-C is shown to kill off seedborne fungal pathogens while maintaining seed viability. Research during the initial of this project was abbreviated due to Covid-19 curtailment of university research activities and then futher delayed in 2021 as the research group was catching up on multiple projects. Evaluations of UV-C treatment for a small array of crops resulted in a mixed bag of findings. Seed viability generally remained unchanged, except for chickpea, but sometimes the percentage of seed germination improved, as was seen with snap bean. The general fungal presence was not killed by 60-minute UV-C exposures. Longer UV-C treatment periods showed that the wheat varieties Ovation and Duet tolerated 3-hour UV-C exposures without a loss in seed viability but the seed still contained a large percentage of general fungi after 3 hours so longer treatment intervals need to be investigated still. Sweet corn (variety Bodacious) that has *Fusarium* on the majority of seeds saw a drastic decrease after 1 and 2 hours of UV-C treatment but longer treatment intervals need to be investigated for this crop as well. The creation of *Leptosphaeria*-infected seed is underway, and *Brassica* seed testing will commence in 2022.

OBJECTIVES: Evaluate the effect of UV treatment on *Brassica*, corn, and wheat seed for mitigation of seedborne pathogens.

PROCEDURES: UV-C seed treatment will be evaluated to determine if UV-C seed treatment effectively mitigates fungi, including *Leptosphaeria* in *Brassica* seeds and *Fusarium* in seed of corn and wheat. UV-C seed treatments will be done using an OSU prototype UV seed treatment machine. A time series of UV-C treatments will be done to maximize pathogen while maintaining acceptable seed germination levels. Non-fungicide-treated seeds will be used to compare the UV-C treatment effect on seedborne pathogen levels and seed/seedling vigor.

Subsamples of the UV-treated seed and nontreated seed will be tested by the conventional seed assay tests to determine fungal presence.

SIGNIFICANT ACCOMPLISHMENTS TO DATE: The Covid-19 pandemic slowed our progress during 2020 as our research had to be postponed during the initial shutdown as it was deemed new research that could not be conducted under the university's Covid-19 guidelines that were instituted during the spring quarter 2020. However, we were able to start on this research project after the research resumption was approved by OSU during the summer of 2021 but we were also catching up on other funded projects where field samples that required processing took the priority, so our progress has not reached the point of concluding this ARF project and we need an additional year to complete the studies and write a final report.

In initial studies, 60-minute UV-C treatment periods were conducted to evaluate the effect on fungi in general as well as seed vigor as measured by germination. In each of two experimental runs, three replicates of 50 count seed samples were exposed to UV-C treatment for 60 minutes. A nontreated control was included for each run of each crop variety. Seed were then placed in moist chambers (50 seeds per crop replicate per run) for germination and evaluations of general fungal growth. This preliminary study included chickpea and snap bean along with sweet corn and wheat in initial evaluations. Wheat and sweet corn seed germination levels were unaffected by the UV treatment but treated wheat had an increased percentage of seed that grew-out fungi compared to nontreated seed, and it was markedly higher for 'Duet' (Table 1). Snap bean seed after UV treatment had an increase in germination compared to the nontreated control seed but it was accompanied by an increased fungal growth on treated seed as well as increased seed rot counts. Chickpea germination rate was reduced after UV treatment but there was a slight decrease in the percentage of seed with fungal growth or seed rot.

Table 1. Effect of a 60-minute UV-C treatment on seed germination and fungal presence of seed

Crop	Treatment	% germination	% seed with fungi	% seed with rot
Chickpea 'Sierra'	Nontreated control	68	96	4
	UV-C	56	85	1
Snap bean '94-G'	Nontreated control	75	8	0
	UV-C	90	52	4
Sweet corn 'Jubilee'	Nontreated control	97	99	2
	UV-C	97	99	0
Wheat 'Duet'	Nontreated control	100	60	0
	UV-C	99	91	0
Wheat 'Ovation'	Nontreated control	95	95	7
	UV-C	97	100	6

In the next phase, we began looking at longer time periods of UV-C and effects on wheat seed viability. Wheat seeds of two varieties were treated for 1, 2, and 3 hours of UV-C. In each of 6 runs, 50-seed replicates were treated and then seed were placed in moist chambers to evaluate germination. Wheat 'Duet' exhibited a decrease in germination after the 3-hr UV treatment while 'Ovation' maintained 98% germination across the 1 to 3-hr UV exposure (Table 2).

Table 2. Effect of UV-C treatment on germination of wheat seed

	% germination		
Crop	**1-hr UV-C**	**2-hr UV-C**	**3-hr UV-C**
Wheat 'Duet'	99	100	90
Wheat 'Ovation'	98	98	98

The UV-C treatments were repeated four times on 'Duet' seed for 0, 1, 2, and 3 hours to evaluate the level of seedborne fungi in general. Fifty seeds per wheat variety and treatment time were plated onto a *Fusarium*-selective medium for each experimental run, and then plates were inspected 10 days later for general fungal growth. Fungal growth was rated using the following scale: 0 – little to no growth; 1 – limited amount of growth, little influence on wheat seeds; 2 – semi aggressive growth covering almost entirety of seed; and 3 – aggressive growth covering entirety of seed and rotting seeds. Fungal growth was generally reduced on seeds that were treated for 1, 2, or 3 hours with UV-C compared to nontreated seeds (Table 3). However, the majority of treated wheat seed were found to still have general fungal growth, so we feel that longer intervals of UV-C treatment must be evaluated. It may be that by the very nature of wheat kernels, with a relative abundance of endosperm and a seed husk rather than a hardened seed coat, that wheat kernels are easily colonized by fungi in general. So it may be ultimately difficult to clean up wheat seed via UV-C treatments but additional studies will shed more light on this situation.

Table 3. Effect of UV-C treatment on fungal presence in wheat 'Duet' seed

UV-C treatments	Overall fungal growth rating
nontreated	2.5
1 hour	0.50
2 hours	0.75
3 hours	0.50

Baseline germination rates were determined for a new collection of seeds during 2021 (Table 4). Sets of 100 nontreated seeds per variety in each experimental run were placed in moist chambers and then incubated at room temperature for evaluation of germination. After 10 to 14 days, the number of seeds that were germinated (when the radicle was twice the length of the seed at its widest point) was recorded. Except for one run with the kale variety, seed germination percentages were generally good (≥ 95%).

Table 4. Baseline seed germination rates of potential crop varieties for UV-C studies

Crop seed type	Exp. Run	% germination
'Stephens' Wheat	1	100
	2	100
'Peaches & Cream' sweet corn	1	96
	2	100
'Bodacious' sweet corn	1	95
	2	98
'Purple Top White Globe' turnip	1	98
	2	100
Red radish	1	100
	2	100
Red Russian kale	1	95
	2	91

We next determined the baseline of *Fusarium* spp. present on the two sweet corn varieties and *Fusarium* was detected on approximately 67% of the seeds in both varieties in a single

evaluation run of 90 seeds of each variety plated onto a *Fusarium*-selective medium. We then selected to continue the UV-C work with the sweet corn variety Bodacious as the supply chain appeared more stable at the time for attaining additional seeds if needed.

Sweet corn (Bodacious) was treated with UV-C for 1 and 2 hours, and then seeds were evaluated for the presence of *Fusarium* species. One hundred seeds per treatment in each run were plated onto a *Fusarium*-selective medium (5 seeds/100-mm diameter Petri dish) and then seeds were inspected 10 days later for the presence of *Fusarium*. None of the nontreated sweet corn seed were without fungal presence, and *Fusarium* occurred on nearly three-quarters of the nontreated sweet corn (Table 5) with the predominate *Fusarium* species being *F. proliferatum* and *F. verticillioides*. *Aspergillus* sp., a well-known, serious pathogen of corn, peanut, and bean was recovered from nearly one quarter of the nontreated sweet corn seed. The presence of *Fusarium* and *Aspergillus* decreased when seeds were treated with UV-C for 1 and 2 hours. The proportion of seed that had no fungal growth after plating increased greatly with 1 and 2 hours of UV-C treatment. However, 6% of the seed still yielded *Fusarium* after 2 hours of UV-C treatment, so we will investigate longer treatment periods in order to see if the seedborne *Fusarium* percentage can be made much closer to zero.

Table 5. Effect of UV-C treatment on seedborne *Fusarium* levels of sweet corn 'Bodacious'

UV-C treatment	**% seeds with *Fusarium***	**% seeds with *Aspergillus***	**% seeds with other fungi**	**% seeds with NO fungi**
nontreated	74	24	13	0
1 hours	10	15	18	57
2 hours	6	8	11	75

So additional studies are planned to examine longer treatment intervals of UV-C for wheat and sweet corn seed, and the development of *Leptosphaeria*-infected seed is also underway for the *Brassica* seed testing portion, and we anticipate concluding these study portions during 2022.

ADDITIONAL FUNDING RECEIVED DURING PROJECT TERM: $0

FUTURE FUNDING POSSIBILITIES: NIFA program areas such as Crop Protection and Pest Management grant program and regional federal grant programs such as the Western Integrated Pest Management Center's program.