REPORT TO THE AGRICULTURAL RESEARCH FOUNDATION FOR THE OREGON PROCESSED VEGETABLE COMMISSION December 2011

Project Title: Management of Fusarium diseases of sweet corn in the PNW: Seed microflora influence on disease and development of *Fusarium*-free seed

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Background: Sweet corn growers in the Willamette Valley of Oregon reported declining yields during the early 1990s. The decline in yields was associated with leaf "firing", where the leaves die prematurely starting at the base of the plant and then progress up the plant. Initially the firing and associated yield loss was thought to be caused by root rot. However, investigations by my lab group have shown that Fusarium crown rot, accompanied by a stalk node rot, appears to explain the concomitant loss in yield. A complex of *Fusarium* spp. (*F. oxysporum, F. verticillioides*, and *F. proliferatum*) has been found to be associated with Fusarium crown and stalk node rot. Factors such as characteristics of specific corn genotypes, environmental conditions which stress plants or favor *Fusarium*, can all modulate disease incidence or severity. Also, the microbial population structure surrounding or within infection points can influence pathogen behavior and resulting disease.

Commercial sweet corn seed lots have been found to contain high percentages of seed infested with *Fusarium* and both *F. verticillioides* and *F. proliferatum* being the principal *Fusarium* species recovered. Both of these species can cause seedling blight and rot of seed, stalk, root, or ear. Thus, sweet corn seed is usually treated with fungicides to prevent seedling rot and damping-off. However, fungicidal seed treatments may not prevent seedlings from becoming infected by seed-borne *Fusarium*. Not all strains of specific *Fusarium* species are equal in virulence and some strains are reported non-pathogenic, especially *F. verticillioides*. Different species of *Fusarium* can cohabitate the same corn plant and the interaction among strains on seed and with populations of *Fusarium* in the soil is not well understood. Removal of all *Fusarium* from seed and planting of these seeds does not necessarily reduce losses from Fusarium diseases if pathogenic populations are present in the soil, but removal of seed-borne *Fusarium* and subsequent biological seed treatment may lead to reductions in crown rot as well as root rot severity and incidence and can improve sweet corn ear yields. Suggesting that *Fusarium* strains present on seed, even if only weakly pathogenic, can prime the developing plant for subsequent infection by more aggressive soilborne strains of *Fusarium*.

Objectives for 2011 and Accomplishments:

1. Examine the yield and disease levels of sweet corn plants grown from seeds treated with germicidal light.

Treating sweet corn seed with UV-C for 1 hr generally didn't have much effect on disease levels or recovery of Fusarium from subsequent plants grown in either a Fusarium-infested field or a field rarely rotated to corn. There was a trend to a lower proportional of stalk ear nodes to yield Fusarium when seeds were treated with UV-C, and an associated yield increase, but it was a slight and non-significant improvement.

An experimental field on the OSU-Botany Farm was inoculated during several growing seasons since 2000 with a complex of *Fusarium* species, and has be repeatedly shown to have severe disease pressure

for Fusarium crown and stalk node rot of sweet corn. A second corn field was newly-established nearby and remained non-inoculated except for naturally-occurring Fusarium populations. Seeds of sweet corn hybrid HM2390 were exposed to UV-C for 1 hr, or were left non-treated. Treated and nontreated seeds were sown in the "new" field and "Fusarium" field in 4- to 7-replicates of 20-ft long, 1-row plots in each fields. Fields were planted two weeks apart; the Fusarium field was planted 2 July and the new field on 15 July. Every other week, plants were sampled from each field site, starting 23 Aug and 31 Aug for the Fusarium field (8/31, 9/7, 9/22, 10/7/2011) and new field (8/31, 9/13, 9/30, 10/13/2011), respectively. The primary root, sub-crown internode, crown, and lower stalk nodes were sampled on earlier sampling dates while on later sample dates, stalk nodes were examined progressively higher up the plant, up to the apical ear node. On each sampling date, 5 plants were dug from each plot, soil was washed from the root balls, and each plant's crown as well as the lower stalk portion was split open longitudinally. Crown grayscale was measured as an evaluation of crown rot and was done with a grayscale analysis (ImageJ, NIH) after each crown was digitally-captured on a flatbed scanner. Corn tissues were examined for the presence of *Fusarium* species by placing tissue bits from each plant sampled onto a *Fusarium*-selective medium (amended Nash medium). Fusarium verticillioides colonies were enumerated on the isolation plates as a representative *Fusarium* sp. Ear weights were determined at the end of the season but only in the Fusarium field as the other lacked remaining plants for ear yield measurements.

There was very little difference in rot levels of sweet corn plants growing from nontreated or UV-C treated seeds (data not shown). There was no significant difference in crown grayscale between plants growing from UV-treated and nontreated seed within either field (Figure 1). Plants in the new field had relatively healthier crown tissues compared to plants growing in the Fusarium field (indicated by higher grayscale values), which isn't unexpected since the new field is not *Fusarium*-inoculated nor continuously cropped with corn.

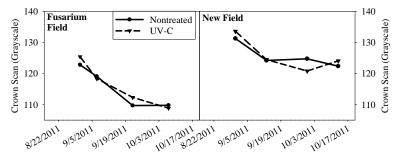


Figure 1. Average crown grayscale measurements for sweet corn 'HM2390' plants treated with 1-hr of UV-C and planted in a *Fusarium*-infested field or a field relatively new to corn.

Fusarium verticillioides was commonly isolated from plant portions throughout the sampling period but there were generally no significant differences in the recovery of *F. verticillioides* from samples of the crowns, primary root, sub-crown internode, or stalk nodes of plants growing from seed treated with UV-C for 1 hour relative to plants produced from nontreated seed (Figures 2 -5). When ear nodes were examined on the final sampling date, the proportion with *F. verticillioides* was 56 and 26 % for plants in the Fusarium field and new field, respectively, but statistically this was non-significant (P=0.05). There was a slight improvement in ear yield with the use of UV-C treated seed for sowing. The primary ear weight averaged 249 and 216 g for plants in the Fusarium field and new field, respectively, averaging about a 15 % yield improvement in primary ear weights with UV-C, but the effect was non-significant (P=0.05).

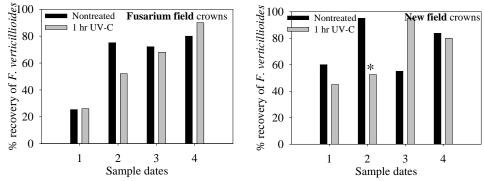


Figure 2. % crown tissue samples from which *F. verticillioides* was recovered when plants were grown from UV-treated seed in a *Fusarium*-infested or new field. * indicates a significant difference (p = 0.05) between recovery levels found in nontreated relative to UV-C treated seed on that sampling date.

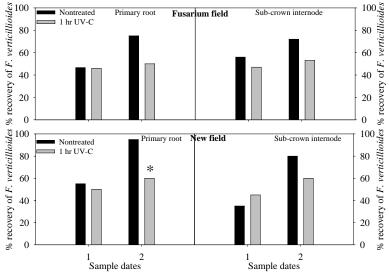


Figure 3. % primary root and sub-crown internode samples from which *F. verticillioides* was recovered when plants were grown from UV-treated seed in a *Fusarium*-infested or new field. * indicates significant (p = 0.05) difference between recovery levels found in nontreated relative to UV-C treated seed on that sampling date.

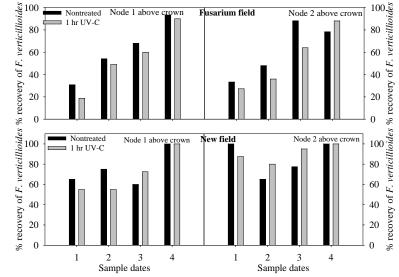


Figure 4. % lower stalk node samples from which *F. verticillioides* was recovered when plants were grown from UV-treated seed in a *Fusarium*-infested or new field.

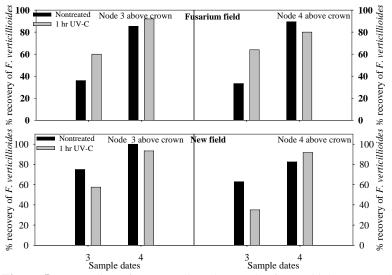


Figure 5. Proportion of upper stalk node samples from which *F. verticillioides* was recovered when plants were grown from UV-treated seed in a *Fusarium*-infested or new field.

Objective 2. Evaluate biological applications to sweet corn seed parents and subsequent *Fusarium* presence on silks and seed infection/contamination.

This year, we planted in a new field as an isolation plot to obtain treated seed. Biocontrol applications resulted in a significant increase in ear weights, both for the primary and secondary ear, as well as a reduction in the presence of Fusarium on silks and kernels. Seed still have to be grown up during 2012 to look for an enhanced vigor in the subsequent plants relative to the plants that remained nontreated.

Kernels of Jubilee's parents were seed-treated with Apron Maxx RTA and then sown with a hand-pushed belt planter into a field not commonly rotated to corn on the OSU Botany Field Lab (Electric Rd., Corvallis) on 15 July 2011. Planting was done later than usual due to wet soil conditions during the spring and the start of a new farmer manager, who filled the vacancy in late May. Each corn line x biocontrol treatment was replicated in four 20-ft long 1-row plots. There were two biocontrol treatments: nontreated or MicroAF (an experimental biocontrol formulation) at 12.8 fl oz/A as a soil drench at planting and later as three additional applications to silks on 3- to 7-day intervals until the silks turned brown. Plants were irrigated every week with approximately 1.5" of water, depending on the weather conditions. Other plants (cv. HM2390 in Objective #1) planted in the same field were evaluated at regular intervals for rot of roots, crown, and stalk nodes as well as larval root feeding to provide a baseline for disease and *Fusarium* pressure in the field.

On 8 Nov 2011, the primary ear was harvested from each of 10 plants per plot of the seed parent of Jubilee. Individual, un-husked ears were placed in paper bags and placed in a drier set at 100°F for 48 hrs. Silks pieces (2" in length) were excised in the laboratory and plated onto a *Fusarium*-selective medium. Five kernels were excised from each ear from the same area that the silks were sampled from and then plated onto *Fusarium*-selective medium. Ears were returned to their respective paper bags and ears were returned to the drier for several more days of drying. On 9 Nov 2011, ears were harvested from 10 additional plants and husked ear weight was recorded. Additional kernels will be assayed this winter and weights of these ears will be determined at that time, as seed is collected for evaluation in the field next season.

The application of MicroAF resulted in a significant decrease in the average number of Fusarium colonies obtained from the silk portions and ear kernels relative to the respective plant parts that were assayed from nontreated plants (Table 1). There was a significant increase in yield in the plants that were treated with MicroAF (Table 2), they yielded about 45 % more in average yield per plant and a 30 % greater primary ear weight compared to the nontreated plants.

Table 1. Effect of biocontrol t	Biocontrol	<i># Fusarium</i> colonies per 2''	% seed tested that grew
Sweet corn inbred	treatment	piece of silk	Fusarium
Female parent of Jubilee	nontreated	1.83 a	63 a
Female parent of Jubilee	MicroAF	0.06 b	4 b

Table 1. Effect of biocontrol treatment on Fusarium r	recovery from silks and kernels
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¹Within each column, means labeled with the same letters are not significantly different (P=0.05) as determined by Fisher's LSD test.

Sweet corn inbred	Biocontrol treatment	Average ear yield per plant (g) ¹	Primary ear wt (g) ¹	Secondary ear wt (g) ¹
Female parent of Jubilee	nontreated	110 b	90 b	20 b
Female parent of Jubilee	MicroAF	160 a	120 a	30 a

¹Within each column, means labeled with the same letters are not significantly different (P=0.05) as determined by Fisher's LSD test.

2. Cooperate with other sweet corn projects within and outside of OSU.

Ocamb evaluated sweet corn plants for rot of the primary and adventitious roots, mesocotyl, stalk nodes, and crown as well as crown grayscale for J. Myers (OSU Horticulture, 920 corn plants), Rogers Brand Vegetable Seed (1630 plants), and TerraMax (175 plants for rot, 700 for yield).