Combining host resistance and organic amendments as a management strategy for the control of Macrophomina crown rot in strawberry

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Introduction

Macrophomina crown rot, caused by the soilborne fungus *Macrophomina phaseolina*, is a problematic pathogen in California strawberry production (Koike et al., 2016). *M. phaseolina* can be difficult to manage due to its persistence in soil and crop residues as microsclerotia. Differential host resistance of strawberry genotypes to *M. phaseolina* has been observed (Winslow et al. 2017). Utilizing host resistance of specific genotypes will be a critical tool for managing this disease in the post-methyl bromide era. Additional studies have demonstrated some control of Macrophomina crown rot with the incorporation of mustard seed meal (MSM) as an organic amendment (Mazzola et al. 2017), or with anaerobic soil disinfestation (Muramoto et al. 2016) but the control has been inconsistent and incomplete. The objective of this research was to evaluate the combination of six strawberry cultivars and four organic amendment soil treatments for their effects on the development of Macrophomina crown rot.

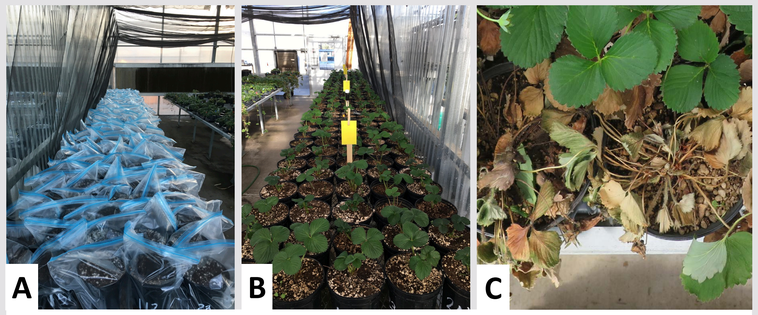

Figure 1. **A**. Anaerobic disinfestation of artificially inoculated potting substrate amended with rice bran (9 ton acre^{-1}) and mustard seed meal (2 ton acre^{-1}). **B**. Early transplants approximately three weeks after inoculation. **C**. Symptoms of Macrophomina crown rot include wilting and plant discoloration, and potentially plant collapse.

Materials and Methods

- The experimental design was a two factor, completely randomized design (CRD), consisting of 24 total treatments.
- The factor cultivar had six levels: 'Albion', 'Del Rey', 'Fronteras', 'Festival', 'Monterey' and 'Petaluma.'
- Cultivars 'Albion,' 'Festival,' and 'Monterey' had been previously characterized as susceptible to *M. phaseolina* and 'Fronteras,' 'Petaluma,' and 'Del Rey' had been characterized as resistant to *M. phaseolina* (Winslow et al. 2017).
- The factor soil treatment had four levels: Inoculated-unamended control, ASD with rice bran (9 ton acre^{-1}), *Brassica juncea* mustard seed meal (2 ton acre^{-1}), and a steam control.
- Plants were grown in a mixture of soil:coconut-coir:perlite 1:1:1 (v:v:v) referred to as potting substrate, with 10 plant replicates for each treatment.
- All soil treatments were artificially infested with a cornmeal sand *M. phaseolina* inoculum (Mihail 1992) to obtain 100 CFU/g soil potting substrate before soil treatment.
- The inoculated potting substrate was irrigated and sealed in plastic bags for three weeks to simulate ASD (Fig. 1A).
- Quantification of *M. phaseolina* in potting substrate was completed with a Andersen sampler onto Sorsensen's NP-10 media.
- Plant health assessments were completed every two weeks with a 0 to 5 scoring, with 0 being a healthy plant and 5 being a dead plant.
- Eight assessments were used to derive area under disease progress curve (AUDPC).
- An additional qPCR assay was conducted on the strawberry crowns to quantify the level of colonization by *M. phaseolina*.

Results

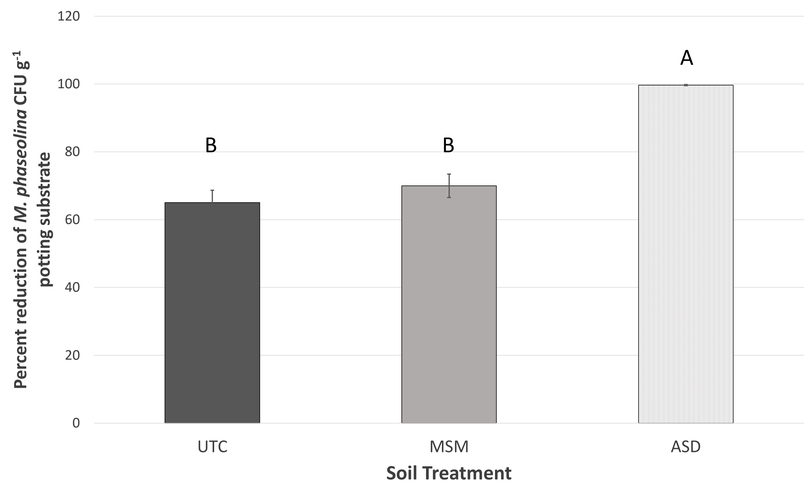

Figure 2. The percent reduction of *M. phaseolina* CFU g^{-1} soil potting mixture from before and after soil treatments. All potting substrate was inoculated with 100 CFU g^{-1} potting substrate mixture before soil treatment. UTC = inoculated but non-amended control, MSM = mustard seed meal applied at 2 tons acre^{-1}, ASD = 9 ton acre^{-1}. Error bars represent one standard error of the mean. Pairwise comparisons were completed with a Tukey's HSD (α = 0.05), means that share the same letter are not statistically different. Steam was not included in the analysis because of 100% reduction of CFU.

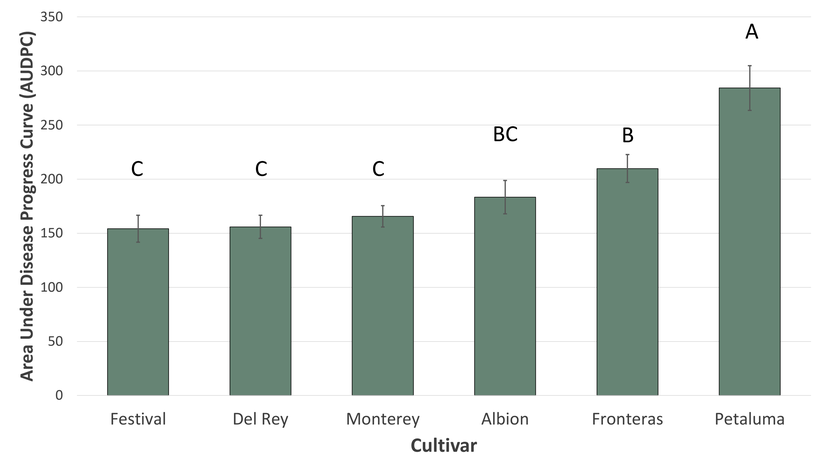

Figure 3. Disease severity expressed as the area under disease progress curve (AUDPC) of six strawberry cultivars. AUDPC is calculated from a plant disease severity rating with 0 = healthy plant, 1 = <10% necrotic foliage, 2 = <25% necrotic foliage, 3 = severe stunting and <50% necrotic foliage, 4 = <90% necrotic foliage, 5 = >90% necrotic foliage or plant death. Eight assessments were conducted. Error bars represent one standard error of the mean. Pairwise comparisons were completed with a Tukey's HSD (α = 0.05), means that share the same letter are not statistically different.

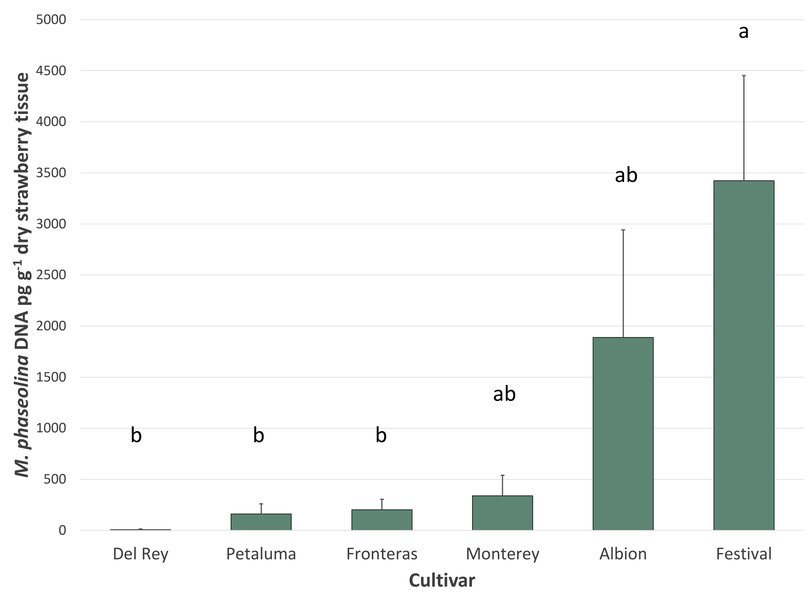

Figure 4. Comparison of the main effect of strawberry cultivar on the mean quantity of *M. phaseolina* DNA detected in strawberry crowns across all soil treatments grown in inoculated potting substrate. Mean values are derived from twelve plant samples per cultivar. Error bars represent one standard error of the mean. Pairwise comparisons were completed with a Tukey's HSD (α = 0.05), means that share the same letter are not statistically different.

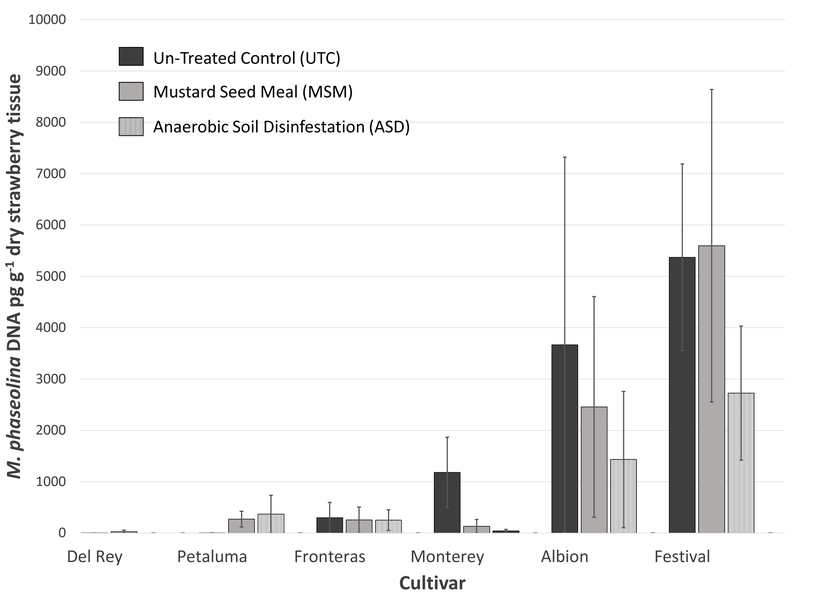

Figure 5. The mean quantity of *M. phaseolina* DNA detected in strawberry crowns of different cultivars by soil treatment. Strawberry plants were grown in infested potting substrate. Error bars represent one standard error of the mean. Pairwise comparisons were not completed due to lack of statistically significant differences. Steam treatment is not included due to lack of detection of *M. phaseolina* DNA.

Discussion

The results indicated that the ASD and steam treatments were able to reduce the CFU g^{-1} potting substrate of *M. phaseolina* by 99.7-100% (Fig. 2). However, disease severity and host colonization of multiple strawberry cultivars was not reduced when grown in the treated potting substrate (Fig. 3; Fig. 5). Organic amendment control of plant symptoms due to *M. phaseolina* was incomplete, which is consistent with literature (Mazzola et al. 2017). In addition, some of the cultivars we tested demonstrated reduced pathogen colonization in regard to *M. phaseolina* DNA using a qPCR assay (Fig. 4). Cultivar 'Petaluma' had increased disease severity in this experiment compared to a previous study, but had minimal pathogen colonization suggesting the symptoms observed were not due to *M. phaseolina* (Winslow et al. 2017). The data presented did not support our hypothesis that strawberry cultivars would have an interactive relationship with organic amendment treatments but demonstrated instead that host resistance were the main effect determining disease severity and host colonization. Future studies assessing the effects of soil treatments on *M. phaseolina* would benefit from a more sensitive assay then the Andersen sampler culture based method presented in this study.

Acknowledgements and References

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