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Anaerobic soil disinfestation and endophytic bacterial treatments for disease and yield evaluation in annual hill strawberry plasticulture production system

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The south-Atlantic region of the United States, including the Commonwealth of Virginia, ranks third in the production of fresh market strawberries after California and Florida. Most growers in the region use the annual hill production system. In addition, the south-Atlantic region of the United States produces 2,342 acres of strawberries, with an average yield of 13,979 lb/acre and a total farm gate value of \$47,158,000. The strawberry plant is highly susceptible to a large variety of soil-borne pathogens, including Colletotrichum spp., Verticillium spp., and Phytophthora spp., which are considered the most damaging pathogens on this crop in the United States. Anaerobic soil disinfestation (ASD) is an alternative method for chemical fumigation that shows great promise to control diseases in strawberry production systems. Endophytic bacteria, such as Bacillus spp., are important biocontrol agents due to their capacity to reduce the growth of pathogens such as fungi, bacteria, and nematodes while also promoting plant growth. We proposed that using ASD and endophytic bacteria will reduce diseases such as anthracnose fruit rot and increase the yield of strawberries in the annual hill plasticulture production system.

In the 2022–23 strawberry growing season, a trial was conducted at the Hampton Roads AREC, Virginia Beach, VA, and strawberry plug plants from 'Chandler' were planted in various treatments as stated below: non-disinfested beds (Control); Pic-Clor 60 beds at 175 lbs/A (preplant soil fumigant) (Pic); ASD treated beds. Additionally, straw-

berry plants in each of these main treatments were treated with one of the following subtreatments: *Bacillus velezensis* only; TerraGrow (a mixture of five *Bacillus* spp.) at 1.5 lbs/A only; and TerraGrow at 1.5lbs/A plus Oxidate 5.0 (27% hydrogen peroxide and 5% peroxyacetic acid) at 2500gal/A. TerraGrow and Oxidate 5.0 were applied three times in the fall season followed by three times in the spring season and the *B. velezensis* treatment was applied two times in the fall followed by two times in the spring. Four plot replicates of 14 plants each were used for each treatment. The berries were harvested twice per week from March 31 through June 16, 2023 (Photo 1).

At each harvest, fruits from each plot were graded as marketable and nonmarketable. The nonmarketable category included small (<10 g), deformed, damaged fruits from insects and birds, and diseased fruits. Fruits that were infected with AFR were weighed separately. Fruits were weighed after grading, and yield is presented as lb/ acre. The total yields were calculated by adding the marketable yield to the non-marketable yield.

For anthracnose fruit rot (AFR), in non-disinfested soil treatment, the weight of diseased fruits decreased signifi-

cantly in plots treated with *B. velezensis*, TerraGrow, and TerraGrow plus Oxidate by about 93%, 80%, and 93%, respectively, compared with the untreated control. In general, the ASD treatment reduced the weight of the diseased fruits with AFR by about 93% compared to the non-disinfested control and 89% compared to the Pic-Clor 60 fumigation treatment (Fig. 1).

AFR on strawberries was controlled without using any chemical fungicide during the field production cycle by using *Bacillus* spp. as biocontrol agents. This bacterial species has the ability to reduce mycelial growth, conidial germination, fruit decay development, and disease severity. Further, the application of ASD may involve the production of organic acids and other biologically active volatiles that have antifungal properties. However, the total yield in the ASD treatments was 15,496 lb/acre, which was significantly lower than the Pic-Clor 60 fumigation of about 21,902 lb/acre and the untreated control treatments of about 19,229 lb/acre (Fig. 2).

The application of ASD with endophytic bacteria may be a helpful plant disease control strategy for organic growers, small farms, growers with limited resources, or growers



experiencing pest issues in buffer regions prior to strawberry transplanting. We are repeating this work for another growing season.

Photo 1. Set up of the experimental trial at the Hampton Roads AREC, Virginia Beach. Each plot was a section of the bed comprised of 14 plants.



Figure 1: Effect of anaerobic soil disinfestation (ASD) and endophytic bacteria on anthracnose fruits rot (AFR). C (non-disinfested control), ASD (Anaerobic soil disinfestation), and PIC (Pic-Clor 60 fumigation). N (untreated control), BV (*Bacillus velezensis* IALR619 strain), TG (TerraGrow), and TG-O (TerraGrow + Oxidate 5.0). Yields are estimated based on 12,432 plants/acre.



Figure 2: Effect Anaerobic soil disinfestation (ASD) and endophytic bacteria on the total yield of strawberry. C (non-disinfested control), PIC (Pic-Clor 60 fumigation), and ASD (Anaerobic soil disinfestation). N (untreated control), BV (*Bacillus velezensis* IALR619 strain), TG (TerraGrow), and TG-O (TerraGrow + Oxidate 5.0). Yields are estimated based on 12,432 plants/acre.

Acknowledgments: We would like to thank the Northeast Sustainable Agriculture Research and Education (SARE) program for funding this project.





SAVE THE DATE

2023 SE Strawberry Expo Location: Charlotte, NC Dates: November 8 - 10, 2023



You're invited to our upcoming 2023 Southeast Strawberry Expo. The 2023 Conference & Farm Tour has an exciting agenda planned for our attendees. Our annual meeting with educational breakout sessions, networking opportunities, industry trade show & expo

will be held at the **Hilton Charlotte University Place** on **Thursday, November 9**. Don't miss out on the Farm Tour which will take place on **Friday, November 10**. We will be visiting four amazing farms! The tour will begin with our 1st stop at **Springs Farm** (Fort Mill, SC) - 2nd stop at **Bush-N-Vine** (York, SC) - 3rd stop at **Patterson Farm Markets** (Mount Ulla, NC) and end with an incrediable Farm-to-Table Dinner at **Carrigan Farms at The Quarry** (Mooresville, NC). The tour will begin promptly at 9:00AM and end around 6:30PM. Registration is required to attend the Farm Tour.

The **Hilton Charlotte University Place** lakeside hotel is located just minutes off I-85, next to UNC Charlotte and the Shoppes at University Place. Top Golf, PNC Music Pavilion, Charlotte Motor Speedway and Concord Mills (home of Bass Pro Shop) are all less than 10 minutes away so there's lots to do while you're in town. The Charlotte Light Rail is located 1/4 mile from the hotel which provides round-trip transportation to the upscale South Park area for an unforgetable shopping and dining experience.



Register here: https://ncstrawberry.com/southeast-strawberry-expo-2023/





2024 NASGA Annual Meeting and Conference Hershey Lodge, Hershey, PA January 29-31, 2024

Join the 2024 NASGA Annual Meeting and Conference in Hershey, PA in conjunction with the Mid Atlantic Fruit and Vegetable Convention.

The convention will begin with a full day workshop on strawberry production. The workshop will include presentations and discussion on soil health, site selection, varieties, fertility and nutrient management. Matted row production will be discussed but there will also be discussion on substrate production as well as other alternative production methods. Time will also be spent discussing insect, disease and weed management.

Day 2 and 3 will be filled with informative presentations as well as a number of panel discussions. There will also be the popular grower and nursery profiles. **Presentation Topics include:** Cyclamen mite management, new diseases, Anthracnose updates, variations between high tunnel and greenhouse production, weed management, runner management, plastic options, new strawberry varieties and more.

Be sure to book your accommodations early to ensure a room at Hershy Lodge. A special discounted Mid-Atlantic Convention room rate of \$169.00 plus 11% taxes is available to attendees.

<u>https://reservations.hersheypa.com/HRSApp/xhtml/home/groupBookingHome.xhtml</u> <u>?groupCode=MAFVC2024L&venue=hersheyLodge</u>

If you wish to make your reservation by phone, call the Hershey Lodge Reservation office at 855-729-3108, and ask for the room block for the Mid Atlantic Fruit & Vegetable Conference, January 28 – February 02, 2024.

Registration details can be found on the NASGA website at https://www.nasga.org/n-american-strawberry-growers-conference.htm



Laboratories That Offer Testing for Fungicide Re- strawberries. Because of the difference in the severity of sistance in the Strawberry Grey Mold (Botrytis cinerea) and Anthracnose (Colletotrichum spp.) Pathogens, and for Identifying Aggressive Neopestalotiopsis

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Fungicide resistance poses a challenge to growers of many crops. It is vital to know if fungicides are likely to be effective so that diseases are managed and money is not wasted. Populations of Botrytis cinerea, which causes grey mold in strawberries, have developed resistance to a number of fungicides formerly effective against it. Furthermore, resistance to the quinone outside inhibitor (QoI, FRAC 11) group of fungicides has been identified in the pathogen that causes anthracnose fruit rot of strawberries (Colletotrichum acutatum species complex; Figure 1). Forcelini and Peres (2018) found that 49 percent of 137 isolates collected from fields in the southeastern US during the 2015-2016 season were resistant to azoxystrobin.



Figure 1. Fruit infested with anthracnose fruit rot.

Another challenge to southeastern US strawberry growers in recent years has been Pestalotia leaf spot and fruit rot (Figures 2a and 2b). A *Neopestalotiopsis* sp. genetically distinct from Neopestalotiopsis rosae has been identified by Baggio et al. (2021) as the cause of severe disease on

disease associated with the newly identified Neopestalotiopsis sp. and other Neopestalotiopsis species, Kaur et al. (2023) have developed a molecular technique for detecting the aggressive genotype.





Figures 2 a and 2 b. Pestalotia leaf spot (left) and fruit spot (right) on strawberry.

This article provides information about labs in the southeastern US that offer tests for fungicide resistance in the strawberry grey mold (Botrytis cinerea) and anthracnose (Colletotrichum spp.) pathogens. It also identifies labs that detect aggressive Neopestalotiopsis using the Kaur et al. (2023) protocol.

Clemson's Molecular Plant Pathogen Detection (MPPD) Lab (Phone no. 864-646-2133) offers tests for fungicide resistance in *Botrytis* and *Colletotrichum*. They test *Botrytis* tion of a new Neopestalotiopsis sp. associated with disisolates for resistance to the following active ingredients: thiophanate methyl (FRAC 1); iprodione (FRAC 2); boscalid, fluopyram, isofetamid, and penthiopyrad (FRAC 7); cyprodinil (FRAC 9); pyraclostrobin (FRAC 11); fludioxonil (FRAC 12); and fenhexamid (FRAC 17). In Colletotrichum, they test for resistance to pyraclostrobin, as a representative of the QoI fungicides. In-state (South Carolina) and out-ofstate samples cost \$80 and \$100, respectively. For *Botrytis* samples, up to ten isolates are each tested for resistance to each active ingredient. Clemson's MPPD Lab also tests for aggressive Neopestalotiopsis. Contact the lab for pricing information.

The University of Georgia (UGA) Plant Molecular Diagnostics Lab (MDL) (Phone no. 229-386-3372) offers resistance testing in both Botrytis and Colletotrichum for the following active ingredients: difenoconazole and prothioconazole (FRAC 3); benzovindiflupyr, fluopyram, penthiopyrad, and pydiflumetofen (FRAC 7); and azoxystrobin and pyraclostrobin (FRAC 11). They test five isolates per sample. The cost for both in- and out-of-state samples is \$200.

UGA's MDL also tests for aggressive Neopestalotiopsis. They charge \$60 for in- and out-of-state samples. If the lab determines that DNA sequencing needs to be done to confirm the identification, there is an additional \$20 charge.

The University of Florida Gulf Coast Research and Education Center's Plant Clinic (Phone no. 813-419-6599) tests strawberry samples for aggressive *Neopestalotiopsis*. They charge \$40 for in- and out-of-state samples.

References

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Postharvest Physiology of Small Fruits at Auburn University

Dr. Marlee Trandel-Hayse

Postharvest physiology is key to maintaining the shelf-life of small fruits and reducing food loss. Typically, research deals with the plant and fruit's response to different technologies and applications that increase quality and delay senescence (decomposition). Fruit quality is a major component of postharvest physiology as it relates to consumer acceptance. Appearance, aroma, color, flavor, texture, and nutritional composition typically define fruit quality. Due to the healthful benefits, consumption of small fruits like strawberry, blueberry (Figure 1), blackberry and raspberry continues to steadily increase, placing pressure to deliver high-quality fruit with a longer shelf-life. As the Assistant Professor in Postharvest Physiology at Auburn University, it is my job to understand the quality issues of small fruits grown in Alabama and deliver high-quality fruits to our consumers.



Figure 1. Blueberry bushes cultivated for fruit production and harvested blueberries for postharvest quality research.



Postharvest physiology is all-encompassing as there are many preharvest factors that can impact postharvest quality, including plant genetics, cultivar selection, production techniques, maturity, harvest conditions, and packaging. Strategies for fruit after harvest include removal of field heat, storage temperatures and atmosphere, edible films/ coating, and other techniques to reduce respiration or ethylene (e.g., the plant ripening hormone). A key issue with many of the small fruits grown in Alabama is their short shelf-life, exacerbated by generally warm to hot conditions during the harvest season. Blackberry and raspberry shelflife can range from 2 to 10 days. If mishandled during postharvest, the fruit can have a 100% loss of salability within 48 hours of harvest. Part of my program will focus on these pertinent issues by teaming with growers and other university professors to preserve shelf-life.

The overarching goal of my program is to understand the impact of preharvest production, breeding and cultivation techniques on the postharvest quality of Alabama small fruits. My research will focus on shelf-life and texture to understand variability among cultivars and guide grower techniques to extend shelf-life. Fruit texture/firmness and variability of firmness can greatly affect fruit shelf-life. Typically, fruit with lower firmness have a higher rate of decomposition and shorter shelf-life. My program will focus on these firmness differences by addressing inner pulp quality. Microscopy work (example of micro-scan in Figure 2) can deliver important information regarding the type and size of cells laid within the fruit. Other aspects of inner quality include middle lamella separation and fiber, lignin and cell wall polysaccharide composition through shelf-life and storage. All of these can elucidate when and how fruit degradation is occurring.



Figure 2. Example of confocal microscopy work on internal flesh quality. Microscopy on small fruits can further elucidate cell structure, size and density which can relate to fruit texture and ripening events.

General quality assessments of color (L*, a*, b*, C* and hue angle), soluble solids content (Brix), pH, titratable acidity, weight loss and tissue firmness will be conducted on small fruits. Postharvest instruments such as handheld brix meters and automatic titrimeters will be used to assess general quality changes in fruit (Figure 3). Performing these assessments in large variety trials will provide useful information regarding cultivar selections with increased postharvest quality and shelf-life. Other research includes determining the optimal storage conditions and application of postharvest treatments, such as ethylene oxidizers or blockers.





Figure 3. Postharvest equipment such as the hand-held Brix meter (A) or automatic titrator (B) which measures titratable acidity is important for quality data collection on small fruits.

Another aspect of my program will address nutritional and bioactive content of small fruits. High nutritional quality and flavor are critically important for the acceptance of our consumers. Nutritional quality encompasses vitamins, minerals, fiber, as well as bioactive phytonutrients such as carotenoids, anthocyanin, flavonoids, non-flavonoids, and other secondary metabolites. Flavor is another critical component of postharvest quality and volatile analysis offers the ability to characterize flavor compounds.

Currently, two important postharvest projects focusing on small fruits are in the pipeline. I am eager to tackle the rabbiteye blueberry quality issues our Alabama growers are facing and I will be working with Dr. Sushan Ru, the rabbiteye blueberry breeder. I am also teaming with Drs. Jay Spiers and Edgar Vinson to address blackberry shelf-life and delay crop loss. My program will report data on these projects in a timely fashion to address grower needs on postharvest guality. In the interim, please feel free to reach out to me via email (mat0141@auburn.edu) or cell phone (334-734-6838) to ask any questions about my program.



Biostimulants did not affect crop yield and postharvest strawberry fruit quality

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In the past few years, there's been an increased interest in the use of biostimulants for improved crop production. Biostimulants are materials that can promote plant growth

much nutrition. These are composed of different organisms, compounds or plant extracts; they include beneficial fungi and bacteria, humic and fulvic acid, seaweed extracts, and protein hydrolysates.

During the 2022-23 growing season we evaluated three different biostimulant products in annual hill strawberry plasticulture production. 'Ruby June' plants were transplanted on 24 Oct, 2022 on non-fumigated beds and were maintained as per grower standard practices. Treatments were as follows (i) no biostimulant (ii) preplant AminoSalmon (220 lb/A) applied during bed making (3) plugs dipped for 20 seconds in TerraGrow Liquid (TGL, 3ml/10 gal) prior to transplanting followed by a foliar spray and a drip application (20 fl. oz/A) at one (10/25/2022), fourteen (11/7) and thirty (11/21) days after transplanting, resuming monthly during spring (4/13/2023 and 5/11); and (iv) EZ-GRO 16-0-0 (3.5lb/A) drip application 14 days after transplanting (11/7) and every 14 days during fall (11/21 and 12/9), resuming during spring (4/13, 4/27, 5/11 and 5/25). All non-treatment irrigation valves were closed during treatment injection through the drip lines. After treatments were injected, the lines were flushed then these valves were closed, and the others opened to irrigate for the length of time used to inject treatments.

Preventative fungicide was applied as a foliar spray three times in the fall, beginning with Captan then rotating with Elevate and Thiram at the recommended rates. During the spring, Captan was applied twice, then rotated with Luna Sensation and Abound/Thiram due to high incidence of anthracnose fruit rot. Row covers (1.2 oz) were utilized once in the winter on 23 December to 3 January, then again in early spring from 14-22 March. Acramite miticide was applied twice on 21 December and 4 April. Weekly spring fertigation began on 27 March at the rate of 7lbN/ acre. Tissue samples were collected on 4 April to determine fertilizer needs, prompting the addition of Epsom salts at 1.5lb S/acre to the fertigation regime on 20 April and 4 May. Ripe fruit was harvested beginning 31 March and continued twice a week until 16 June (Photo 1).

Fruit from each harvest was graded marketable and nonmarketable (less than 10g, deformed, damaged or diseased) and weighed by category. Fruit size was estimated when applied in amounts so small that they do not provide as g/fruit by weighing 10 marketable fruits weekly. Five

marketable fruits were measured weekly for firmness using a penetrometer then stored at -20 °C for later analysis of pH and total soluble solids (°Brix) using a digital refractometer.



Photo 1. Study of the different biostimulant beds during the harvest season.

compared to the other treatments and the untreated control (Fig. 1). The TGL product contains five different Bacil*lus* species that typically protect the plant from harmful microbes and make nutrients available to plants, which may help to improve the total yield and markable yield. Although there were several diseases diagnosed during the season, such as anthracnose fruit rot (AFR), botrytis rot, and other diseases, the treatments did not affect the weight of diseased fruits per bed (data not shown). For strawberry fruits, quality, firmness, total soluble solids (TSS), and pH are important factors affecting fruit quality and customer acceptance, with sugars being the primary soluble metabolites impacting taste and ripeness. While fruit firmness and pH were not influenced by treatments, the application of both the amino salmon and EZ-GRO biostimulants significantly improved TSS values compared to the untreated control (Fig. 2). In conclusion, during this first growing season, none of the biostimulant treatments stood out. We will be repeating this study with same treatments and adding two more biostimulant products, namely iQForte and PVent Microbial WP.

Result and discussion

For total yield and marketable yield, no treatment differences were found. However, the plots treated with TGL showed slight increases in the total and marketable yields



Figure 1. Effect of Amino Salmon, EZ-Gro, and TerraGrow Liquid on total yield. NTC (untreated control), Amino Salmon (preplant), EZ-Gro 16-0 -0 at 3.5lb/A drip application, and TGL (TerraGrow Liquid at 20 fl. oz/acre foliar and drip). The total yield was calculated based on 12,432 plants/acre.



Figure 2. Effect of application of Amino Salmon, EZ-G, and TerraGrow Liquid on the total soluble solids (TSS). NTC (untreated control), Amino Salmon (preplant), EZ-Gro at 3.5lb/A drip application, and TGL (TerraGrow Liquid at 20 fl. oz/acre foliar and drip). The total yield was calculated based on 12,432 plants/acre.

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