



Control Alt Delete: Enhancing Resiliency of Broccoli Production by Mitigating Alternaria Leaf Blight and Head Rot in the Eastern United States-Major Findings and Impact

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Alternaria leaf blight and head rot (ABHR) is caused by a complex of Alternaria species that include *Alternaria brassicicola*, *A. brassicae*, *A. raphani* (syn. *A. japonica*), and *A. alternata*. *A. brassicicola*, is an economically important necrotrophic pathogen of cultivated brassica crops such as broccoli, cabbage, kale, collards, and cauliflower that causes ABHR. Over the last several years, the occurrences of ABHR in broccoli exhibited an increasing trend in the eastern U.S. (Fig. 1). Some of the factors that can be attributed to this trend may include prevalence of conducive weather conditions for disease development, development of resistance against commonly used fungicides, and introduction of pathogen through infested seeds or seedlings in broccoli production fields. A coordinated, multi-state effort was led by Dutta and team to determine pathogen biology, population structure, and fungicide resistance profile of *Alternaria* spp. responsible for ABHR in broccoli. This newsletter is meant to showcase our work and impact that were made in this project that are done in broccoli among different collaborating states. There also is no need to add a heading for your introduction, as people assume that your manuscript will begin with some kind of introduction.

A. Pathogen Survey, Characterization and Fungicide Sensitivity in Alternaria Species

Survey of Commercial Broccoli Fields in Georgia

PD Dutta, post-doctoral research associates Petkar and Kaur from Georgia surveyed commercial broccoli fields from 2020-2023. Over 450 isolates were collected and characterized during this period. Based on the findings, majority of the *Alternaria* isolates belonged to *A. brassicicola*, which could be the predominant species across eastern US responsible for ABHR outbreaks. However, in some cases *A. alternata* and *A. japonica* were also recovered. Also, considerable variation in aggressiveness was observed with isolates from *A. brassicicola* on broccoli, cabbage, kale and collard with majority of the isolates are either highly or moderately aggressive on broccoli and kale but less aggressive on cabbage.

Fungicide Sensitivity of Alternaria Species

The *A. brassicicola* isolates collected in 2020-2023 survey were sensitive to azoxystrobin at both concentrations (10 and 100 μg/ml). Conversely, *A. alternata* isolates found on broccoli from fields in Connecticut and Virginia had reduced sensitivity to azoxystrobin.

Additionally, Co-PI Everhart's team (University of Connecticut) sequenced cytochrome b gene (cytb) of isolates to screen for mutation corresponding to quinine outside inhibitor (QoI) fungicides, they identified that there was a G143A mutation in *A. alternata* isolates collected from Connecticut and Virginia, and on isolates from weed samples collected from New York; however, there was no mutation present in *A. brassicicola* corroborating our lab sensitivity assays.

Survey of Commercial Broccoli Fields in New York

A group led by Co-PI Smart and her post doc Betaw, conducted a survey in New York state from commercial broccoli fields during 2021 and 2023. *A. brassicicola* isolates were characterized for their aggressiveness on brassica crops and their sensitivity to boscalid fungicide. Variability in disease severity on detached leaves of broccoli, cabbage, cauliflower, collard, and kale was investigated on 121 isolates. Significant variation was noted among brassica crops in response to ABHR (Fig. 2). Broccoli was the most susceptible, collard was relatively less susceptible while cabbage, cauliflower and kale were the least susceptible for all isolates. Disease percentage ranged from 6 to 56%. The subset of isolates from 2021 (24 isolates), 51 isolates (2023) collection and isolate '18062' were also examined for their sensitivity to boscalid, a commonly used fungicide for ABHR control, at two concentration levels and control (no boscalid) (Fig. 3). Out of the tested 72 isolates 58 isolates were less sensitive to boscalid with mycelial growth ratio > 0.5.

Survey of Commercial Broccoli Fields in Virginia, Connecticut and Massachusetts

The predominant species isolated was *A. brassicicola* (88% in 2022 and 94% in 2023) and the second most common was *A. alternata* (12% in 2022 and 6% in 2023), which was obtained from fields in Connecticut and Massachusetts in 2022, and in Virginia in both years. Alternaria japonica was not found in either year. Symptoms of *A. alternata* were indistinguishable from *A. brassicicola*, as were colony morphologies. While *A. alternata* is considered a generalist and of little consequence for broccoli, it is considered a pathogen of significance on multiple crops (blueberry, citrus, pistachios), but there remains scant information on the disease etiology on broccoli. Further investigations indicate that *A. alternata* can be pathogenic on broccoli and is capable of initiating infection and causing lesions typical of ABHR.

B. Varietal Screening

Varietal Screening in New York

Commercial broccoli cultivars were compared for ABHR disease susceptibility for three growing seasons (2021-2023) in New York. The plants were inoculated with *A. brassicicola* and rated for leaf disease percentage. There was a difference in susceptibility among cultivars (Fig. 4 and 5). Cultivars 'Marathon', 'Belstar', 'Burney' and 'Asteroid' were identified as less susceptible based on leaf disease severity and the latter three confirmed to be less susceptible to head disease as well. Very early maturing cultivars appear to have more head disease severity compared to the rest of the maturity groups (Fig. 5).

In the 2023 growing season, yield of six cultivars adapted to the eastern US environment was compared in uninoculated and inoculated plots with *A. brassicicola* (Table 1). Two field sites, to be either left uninoculated or inoculated with *A. brassicicola*, were set separately a quarter of a mile apart. No ABHR symptoms appeared in the uninoculated site. No significant differences were observed in head weight (total yield) both in uninoculated and inoculated sites. In the inoculated site, significant differences were observed among cultivars for leaf and head disease severity, marketable head number and yield. Cultivar Burney showed the lowest disease severity both on the head and leaf, highest marketable yield and number and largest percent of marketable head followed by cultivar Imperial (Table 1).

Varietal Screening in Georgia

A group led by Co-PIs Coolong, Duffey and Mcavoy evaluated 26 commercial broccoli cultivars in multiple years of trials in Watkinsville and Tifton, GA for resistance to ABHR. We inoculated plants at approximately 3 weeks after transplanting with a local isolate of Alternaria and foliar disease was evaluated throughout growth. At harvest we also evaluated every individual head for the presence/severity of disease as well. The evaluation was done by scraping the beads from the heads using a harvest knife and evaluating the presence of lesions. At times, Alternaria is present, but not readily apparent until some of the beads are removed. This allowed us to detect disease when it was present at low levels in the heads. While not readily apparent, it certainly would have been an issue for growers shipping heads as it would likely have shown up during transit.

Some consistently cultivars with notable resistance included 'Marathon', 'Vallejo', 'Emerald Pride', and 'Burney' (Fig. 6). Although some of these cultivars are not well suited to the fall climate in Georgia, our results should provide documentation for breeders looking to incorporate enhanced ABHR resistance into other cultivars suitable for the southeastern US.

C. Fungicide Programs

Fungicide Programs in Georgia

PD Dutta evaluated 12 conventional fungicides for the for the management of ABHR in broccoli in Tift County, Georgia, 2021-23. Alternaria leaf blight severity was significantly lower for Priaxor, Luna Sensation, and Inspire Super compared to other treatments and non-treated check. Head rot (%) was significantly lower for treatments (Miravis Prime, Topguard EQ, Luna Sensation, Priaxor, Inspire Super, Quadris Top, Quadris and Switch compared to other treatments and non-treated check. Phytotoxicity was not observed with any of the treatments used.

PD Dutta also evaluated 9 OMRI-listed fungicides for the management of ABHR in broccoli in Tift County, Georgia, 2021-23. Plots treated with OSO (6.5 fl oz), Bravo WeatherStik (conventional check), and Microthiol Dispress sulfur had significantly lower disease severity compared to Theia and non-treated check. Head rot (%) ratings were taken at harvest where plots treated with OSO at both rates (13 fl oz and 6.5 fl oz), Microthiol Dispress sulfur, and Bravo had significantly lower incidence compared to other treatments and non-treated check.

Fungicide Program in New York (Geneva)

Efficacy of 12 conventional and 9 OMRI listed fungicides was evaluated in 2021 and 2022 for control of ABHR disease on broccoli. All conventional fungicides (Inspire Super, Priaxor, Miravis Prime, Quadris Top, Luna Sensation, Fontelis, Quadris, Topguard EQ, Endura, Switch and Bravo), with the exception of a new copper zinc (CuZn) formulation evaluated only in 2022, significantly reduced the disease severity compared with the nontreated controls (Fig. 7). However, no fungicide provided complete control of the disease. We also evaluated Oso at both the full rate and half rate, and both rates were found to have good efficacy against leaf spot and head blight. In fact, Oso at either rate was not significantly different from Bravo, which was included as a conventional control (Fig. 8).

In the 2023 growing season, we evaluated three fungicide spray programs on two resistant broccoli cultivars 'Abrams', 'Burney' and a susceptible check 'Emerald Crown'. The fungicide programs include Program 1 – Bravo WS, Miravis Prime, Quadris Top; Program 2 – Bravo WS, Priaxor, Inspire Super; and Program 3 – Bravo WS, Quadris Top, Bravo WS. Significant differences among fungicide programs were observed only on susceptible cultivar 'Emerald crown' both in leaf and head disease reduction. Fungicide program 2 (Bravo WS, Priaxor, Inspire Super) was efficacious in reducing leaf and head disease severity, and yield loss in Emerald Crown.

Fungicide Programs in New York (Western New York)

FRAC 7 fungicides such as Priaxor, Luna Sensation and Fontelis were the most effective products, which were closely followed by FRAC 3 fungicides such as Quadris Top and Inspire Super. FRAC 11 fungicide Quadris resulted in significantly less control than FRAC 3 + 11 Quadris Top, and FRAC M5 Bravo was significantly better than the nontreated, but generally not as effective as the other conventional fungicides (Fig. 9 and Fig. 10). In 2023, fungicide trials were artificially inoculated with local Alternaria isolate and disease severity observations were recorded. In summary:

- FRAC 11 was the most effective for controlling ABHR and head rot in broccoli, followed by FRAC 9 and 12, while FRAC M5 had mediocre and poor-to-no efficacy on controlling foliar ABHR and head rot, respectively.
- ABHR appeared to have developed fungicide resistance to FRAC 7 a.i. boscalid (in Endura) and should be tested along with the other FRAC 7 active ingredients, fluopyram (in Luna Sensation) and pydiflumetofen (in Miravis Prime) for fungicide resistance. It was later confirmed that the majority of Alternaria isolates collected in NY as part of this project had developed fungicide resistance to boscalid.
- Similarly, ABHR may be developing fungicide resistance to FRAC 3 (as in Quadris Top and Inspire Super), which should also be further investigated.

Organic Fungicide Summary:

- The best organic treatment for ABHR in broccoli was FRAC 19 Oso, which performed similarly to conventional fungicide Bravo, but both failed to adequately control head rot under moderate to high disease pressure.
- Adding adjuvant Nufilm-P, increasing rate to 13 fl oz/A and tank mixing with Cueva did not significantly improve ABHR control over Oso 6.5 fl oz/A alone.
- FRAC M1 Kocide 3000-O also showed some activity on ABHR, which was slightly better than other FRAC M1 Cueva, while Carb-O-Nator (a.i. potassium bicarbonate) had no activity.
- Phytotoxicity in the form of leaf necrosis was observed when Kocide 3000 was used in the fall, especially when it was tank mixed with an adjuvant, that resulted in more foliar ABHR than the nontreated.
- Fungicides alone, especially when a susceptible broccoli variety is grown, will not suffice when disease pressure is high making organic growers vulnerable to significant economic losses from ABHR in broccoli.

D. Irrigation Timing

To determine the role of timing of irrigation in disease severity, four overhead irrigation treatments were compared in 2021 and 2022. Treatments were 1) 15 minutes of overhead irrigation at 7 am; 2) 15 minutes of overhead irrigation at noon; 3) 15 minutes of overhead irrigation at 6:00 PM; 4) no additional overhead irrigation (control). No significant difference was observed among the treatments. However, plots overhead irrigated in the morning showed lower disease severity both in head and leaf in both years in New York (Fig. 11). For the simulated irrigation trial, significant differences were determined in head disease, with irrigation later in the day causing more disease and less marketable heads in Georgia. When possible, drip irrigation or early morning overhead irrigation should be used to decrease disease and increase marketable yield.

E. Nitrogen Management

Five treatments were compared to determine the effect of nitrogen fertility for severity of the disease in 2021 and 2022. The treatments were 50, 75, 100, 125 and 150% of locally recommended nitrogen (N) treatment. Half of the recommended fertilizer was applied at planting for the entire plot of the trial. Six weeks after transplanting, the five nitrogen rate treatments were side dressed. No significant difference was observed among the treatments in all parameters except in head disease severity in 2021, where 50% N fertilizer from recommended showed significantly higher disease severity compared to the rest of the treatments (Table 2). Similarly, no significant differences between fertility treatments were detected in 2021 and head disease severity was higher in the Tifton location compared to the Watkinsville location (Georgia). In 2022, less disease was determined in the 50%, 75% and 150% fertility treatments in Watkinsville, but no differences were determined in the percent of marketable heads at either location. Lower fertility rates may impact disease progression but the trade off for lower quality heads and yield is not beneficial.

F. Leaf Epicuticular Wax-Mediated Resistance to A. brassicicola in Broccoli and Modification of Fungicide Programs

PD Dutta and post-doctoral research associates Gangurde and Kaur demonstrated that leaf epicuticular wax is associated with resistance against *A. brassicicola* in broccoli. During the pathogenicity and whole plant inoculation assays, we observed significantly higher disease severity in the older leaves at the bottom, as compared to the younger leaves at the top. The symptoms seemed to appear first in the lower and older leaves and gradually progressed upwards to the younger leaves, ultimately reaching the broccoli head. Epicuticular wax analysis showed a significantly higher amount of wax deposition on the younger leaves at the top as compared to the older leaves at the bottom. Further regression analysis showed a negative correlation between wax per unit area and disease severity.

Based on these observations, we modified our fungicide spray program that included initial fungicide sprays with systemic fungicides (Luna Sensation, Bayer; Miravis Prime, Syngenta; Inspire Super, Syngenta) at a 10-day interval followed by Bravo (Adama) at a 7-day interval until head-set. After the head set, the modified program relied on systemic fungicides stated above at a weekly interval until harvest maturity. The modified fungicide program was compared with grower's standard that relied on solely Bravo (at a weekly interval) until head set followed by a weekly spray of systemic fungicides until harvest maturity. Based on disease assessments over a period of 8-weeks indicated significant reduction in foliar severity and head rot (%) with our modified fungicide program compared with the growers' standard. The results from the field trials suggest that our modified fungicide spray program could significantly reduce foliar disease severity and head rot (%) compared with the grower's standard. The improved fungicide program can potentially reduce economic losses due to ABHR by 20% (data obtained from field trials), which can account for \$160 per acre in savings. If the improved management strategy is utilized over the entire broccoli acreage in U.S. (130,000 acres), a total savings of \$20.8 million can potentially be achieved.

G. Seed as a Source of Inoculum

Seed can serve as a potential source of inoculum for the transmission of *A. brassicicola* in broccoli as demonstrated earlier; however, seed-to-seedling transmission of pathogen was never characterized empirically. Our results indicate that artificial seed inoculation with *A. brassicicola* can result in consistent seed-to-seedling transmission with significant impact on seed germination. Further seed-to-seedling transmission is not dependent on the aggressiveness of *A. brassicicola* isolates and, seed treatment with Miravis can significantly reduce pathogen transmission in broccoli seedings. Overall, our study provides detailed characterization of seed-to-seedling transmission of *A. brassicicola* in broccoli that can be further used to determine inoculum threshold, which has potential applications in seed-health testing and sample size determination. Further we also provide options for effective seed treatments that can significantly reduce *A. brassicicola* seed-to-seedling transmission and may potentially aid in managing seedborne fungal infection.

We also characterized natural seed infestation and seed-to-seedling transmission of A. brassicicola in broccoli. Two hundred commercial seedlots from two broccoli cultivars; Cultivar 1 (n=100 seedlots) and Cultivar 2 (n=100 seedlots) were evaluated for the presence of A. brassicicola under in-vitro conditions using a seedling grow-out assay. Alternaria spp. was detected in 31 and 28% of the commercial seedlots of Cultivar 1 and Cultivar 2, respectively (Fig. 12). The seed-to-seedling transmission (%) varied considerably within each positive infested seedlot, which ranged from 1.3 to 17.3%. Subsequent molecular identification showed that ninety-six percent of the isolates formed a cluster with a known A. brassicicola. One hundred percent of the A. brassicicola seed isolates were either highly- or moderately- aggressive on broccoli (cv. Emerald Crown) based on a detached leaf assay (Fig. 13). Sensitivity of representative A. brassicicola isolates to azoxystrobin was evaluated for a representative set of isolates. A. brassicicola isolates from naturally infested commercial broccoli seeds were sensitive to azoxystrobin with considerably low EC50 values in the range of <0.0001 ppm to 0.33 ppm; however, there were a few isolates (14%), which showed 100-fold reduced sensitivity from the most sensitive isolate (EC50 =0.0001 ppm). Our results confirm that commercial broccoli seedlots can be naturally contaminated with pathogenic and aggressive A. brassicicola. We also provide evidence for the potential presence of A. brassicicola isolates with reduced azoxystrobin-sensitivity in naturally infested commercial broccoli seedlots. Together, these findings may have critical implications in considerations for seed-health testing, seed treatments and greenhouse scouting to limit the introduction of infested seedlots in commercial broccoli fields.

Cross-Resistance to SDHI Fungicides in Seed Isolates

Interestingly, we also observed that 15% (n=9/58) that had 100-fold reduced sensitivity to a QoI fungicide (azoxystrobin) as determined above also had reduced sensitivity to boscalid (60-fold) and penthiopyrad (45-fold). The majority of isolates (93%) showed reduced sensitivity to boscalid and penthiopyrad, with high EC₅₀ values (>50 ppm for boscalid and >10 ppm for penthiopyrad), confirming resistance to two SDHI fungicides in *A. brassicicola* isolates from seeds (Fig. 14). However, isolates were sensitive to fluopyram suggesting that fluopyram is still effective in management of ABHR. Our findings provide the first molecular and phenotypic evidence that *A. brassicicola* from naturally infested commercial broccoli seeds in the United States exhibited multiple resistance to two SDHI fungicides (boscalid and penthiopyrad) and a reduced sensitivity to azoxystrobin.

H. Weeds as a Source of Inoculum

Since *Alternaria* species can survive on weeds, weed samples infected with Alternaria were collected from New York (2021), and from Connecticut and Massachusetts (2022- 2023) from within fields and from field margins (Fig. 15). From these weeds, only *A. alternata* has been isolated using the Alt al markers. Currently, there has been no positive identification of *A. brassicicola* infecting weeds in broccoli fields in these states. The G143A mutation that confers resistance to azoxystrobin has been identified in two New York weed isolates. This suggests that *A. alternata* can survive on weeds and play an important role in initiating outbreaks.

Pathogenicity of Weed Isolates in Selected Brassicas

Since weeds can serve as a source of inoculum, scientists from Virginia Tech led by Drs. Langston and Batista,

also collected 30 *Alternaria* isolates (2021-2023) from weeds located adjacent to broccoli fields. The weeds included *Oenothera laciniata* (curleaf evening primrose), *Brassica kaber* (D.C.) L.C. Wheeler (wild mustard), *Chenopodium album* (lambsquarter), and *Cyperus rotundus* (nutsedge). Using PCR-based marker analysis, four were identified as *A. japonica*, sixteen tested positive for *A brassicicola*, and ten were neither *A. brassicicola* nor *A. japonica*. Pathogenicity tests demonstrated varying degrees of virulence in broccoli, kale, cabbage, and collard (Fig. 16). Specifically, 30% of the *A. brassicicola* isolates were virulent to broccoli, 75% to kale, 56% to cabbage, and 69% to collard. All *A. japonica* isolates were virulent to broccoli, kale, cabbage, and collard, except for one isolate which was not virulent on broccoli. Additionally, a few isolates obtained from cutleaf evening primrose and lambsquarter were re-inoculated into the same weeds to confirm pathogenicity (Fig. 17). This work will lead to a better understanding of inoculum sources of *Alternaria* spp. pathogenic on brassica crops.

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- 2. Higgins, D. S., Adams, C. M., and Fletcher, A. L. 2023. Evaluation of broccoli cultivar response to Alternaria leaf blight and head rot, 2023. 18:V072
- 3. Higgins, D. S., Adams, C. M., and Fletcher, A. L. 2023. Evaluation of DMI fungicides for control of Alternaria leaf blight and head rot in broccoli, 2023. 18:V071
- 4. Dutta, B. and Foster, M. J. 2022. Evaluation of organic fungicides for the management of Alternaria leaf blight and head rot in broccoli in Tift County, Georgia, 2022. 17:V134.
- 5. Dutta, B. and Foster, M. J. 2022. Evaluation of conventional fungicides for the management of Alternaria leaf blight and head rot in broccoli in Tift County, Georgia, 2022. 17:V135.
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Table 1. Disease severity and yield components of uninoculated and inoculated broccoli varieties at Geneva, NY in 2023.

		Leaf Head AUDPC ^a %disease		head	Total nead Marketable eight head weight Kg) ^b (kg)		%Marketable head weight ^c		Number of total head	Marketable head number			
Inoculated trial													
Burney	26.3	b	4.3	b	1.50	a	1.10	a	69.8	a	10	7.3	a
Imperial	27.0	b	1.7	b	1.28	a	0.77	ab	61.4	ab	10	7.3	a
Exp3622	74.0	ab	25.0	a	1.38	a	0.38	ab	24.7	bc	10	2.3	ab
Eastern Crown	77.7	ab	37.5	a	1.78	a	0.03	b	2.4	c	10	1.0	b
Emerald Pride	120.4	a	26.7	a	1.58	a	0.18	ab	12.8	c	10	1.7	ab
Emerald Crown	123.4	a	30.0	a	1.54	a	0.05	b	4.8	c	10	0.7	b
Uninoculated trial													
Burney	-		-		1.12	a	1.12	a	100		10	10	
Imperial	-		_		1.57	a	1.57	a	100		10	10	
Exp3622	-		-		1.19	a	1.19	a	100		10	10	
Eastern Crown	-		-		1.43	a	1.43	a	100		10	10	
Emerald Pride	-		-		1.28	a	1.28	a	100		10	10	
Emerald Crown	-		-		1.47	a	1.47	a	100		10	10	

^aMeans in a column followed by the same letter(s) are not significantly different at $P \le 0.05$ according to Tukey's honestly significant difference test.

^bTotal and marketable head weight were determined by measuring the weight of broccoli head from 10 plants per plot (Plot size was 0.9 x 2.4 m² double row, 20.3 cm within 91.44 cm between row spacing).

^c% Marketable head weight was calculated as (Marketable head weight)/(Total head weight) x 100.

Table 2. Alternaria leaf blight and head rot severity (2021-2022) and unmarketable head percentage (2022) in five levels of nitrogen fertilizer in broccoli plants

N % from recommended	Leaf	AUDPC	Head A	Unmarketable head (%)		
	2021	2022	2021	2022	2022	
50%	99.0 a	288.4 a	37.4 b	167.7 a	67.2 a	
75%	85.0 a	266.4 a	20.7 a	139.6 a	58.2 a	
100%	66.0 a	275.6 a	19.5 a	136.4 a	57.3 a	
125%	71.0 a	203.9 a	15.6 a	74.9 a	55.8 a	
150%	76.0 a	272.1 a	11.4 a	146.5 a	77.3 a	



Figure 1. The characteristic symptoms of *Alternaria brassicicola* on broccoli at different growth stages. Panel A to C depicts the initial symptoms of ABHR symptoms on broccoli that include small dark brown to black color necrotic lesions, followed by dark brown to black color necrotic lesions with concentric rings and a yellow halo (B and C). In heavily infected plants necrotic lesions on head can be seen (D-F).

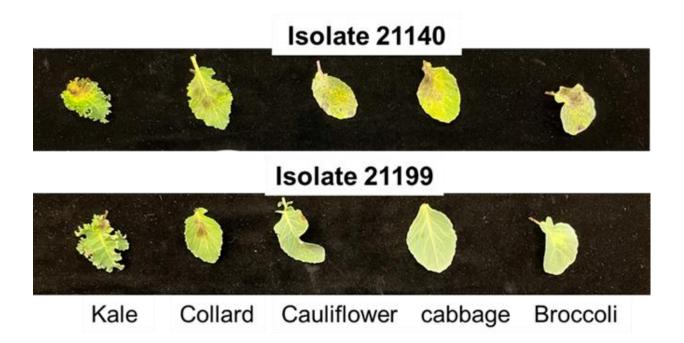


Figure 2. Disease symptoms in brassica crops inoculated with an aggressive ('21140') and a less aggressive ('21199') *A. brassicicola* isolates (5 days post inoculation).

Boscalid concentration 0 0.5 50 µg ml-1 µg ml-1 µg ml-1 Sensitive isolate '18062' Resistant Isolate '21154'

Figure 3. Mycelial growth of a sensitive isolate of *A. brassicicola* '18062' and a resistant isolate '21154' on day five after mycelial plug transfer on PDA (potato dextrose agar). Three concentrations were used: no boscalid, low $(0.5 \,\mu g \,ml-1)$, and high $(50 \,\mu g \,ml-1)$ boscalid.

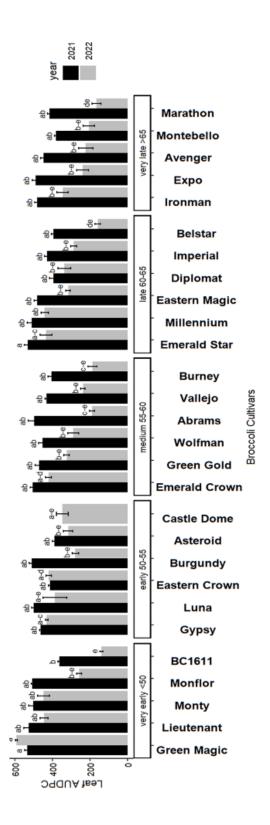


Figure 4. Leaf disease severity for broccoli cultivars and maturity groups in 2021 and 2022.

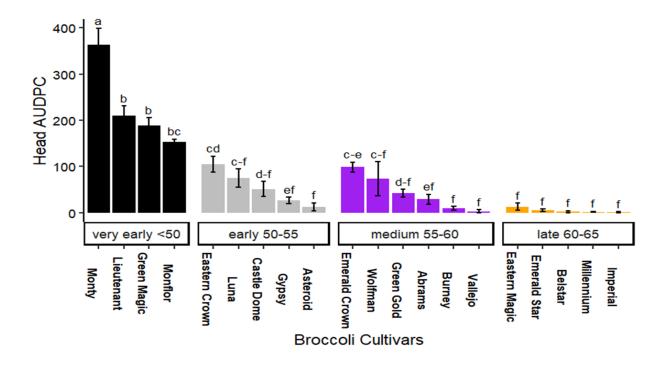


Figure 5. Head disease severity in broccoli cultivars and maturity groups in 2022.



Figure 6. Differences in Alternaria head rot severity in four cultivars in the trial conducted at UGA. Significant differences existed for commercial cultivars grown over several years in Tifton and Watkinsville, GA.

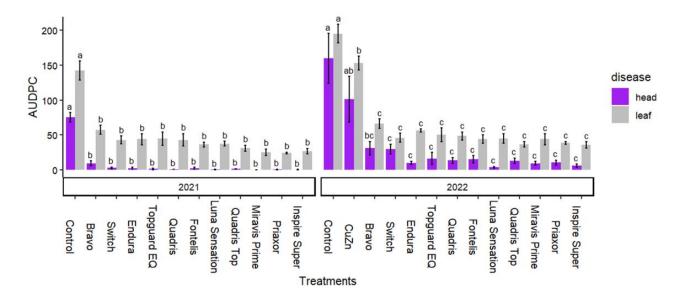


Figure. 7 Efficacy of conventional fungicides for Alternaria leaf blight and head rot in 2021 and 2022.

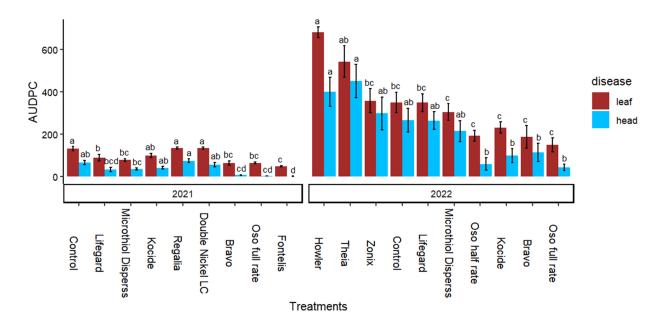


Figure 8. Efficacy of OMRI listed fungicides for Alternaria leaf blight and head rot in 2021 and 2022.



Figure 9. Nontreated broccoli in back row and broccoli treated with Quadris Top in the front.

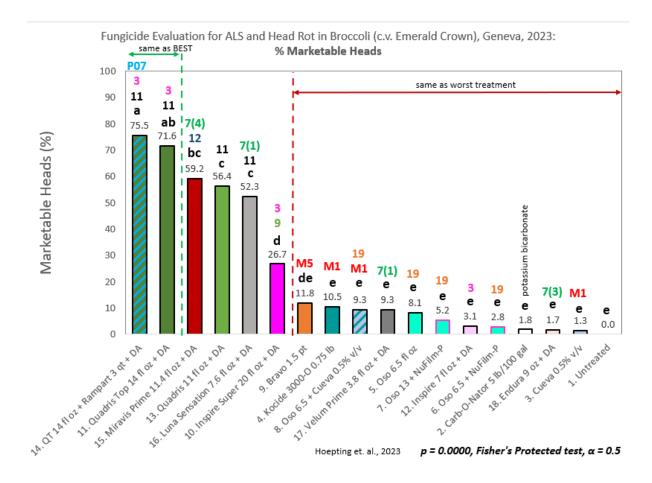


Figure 10. FRAC 3 + 11 fungicide Quadris Top was the only treatment that resulted in greater than 70% marketable heads under moderate to high ABHR disease pressure. Four of the top five best-performing treatments included FRAC 911.

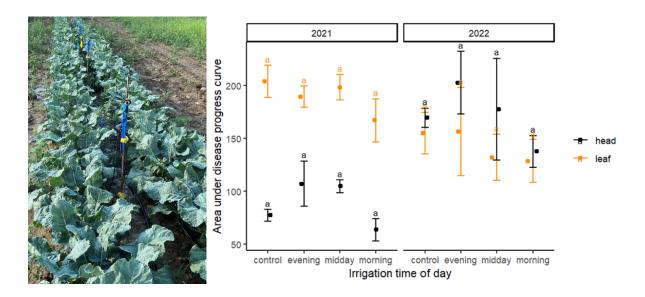


Figure 11. Broccoli leaf and head disease severity in different irrigation timing.

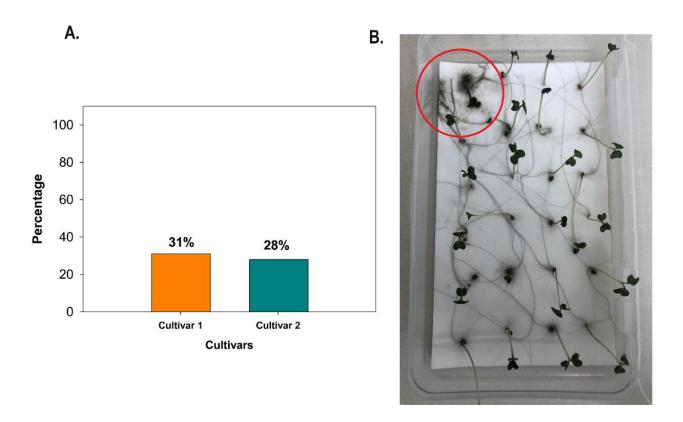


Figure 12. Percentage of naturally infested seedlots from two commercial broccoli cultivars with *Alternaria* spp. infestation. Naturally infested commercial broccoli a) percent infested seedlot (%) from each cultivar and b) seed-to-seedling transmission (%) of *Alternaria* spp. were determined. The red-circle in panel B indicates symptomatic transmission of *Alternaria* spp. with dark olivaceous green colored sporulation on infected seedlings.

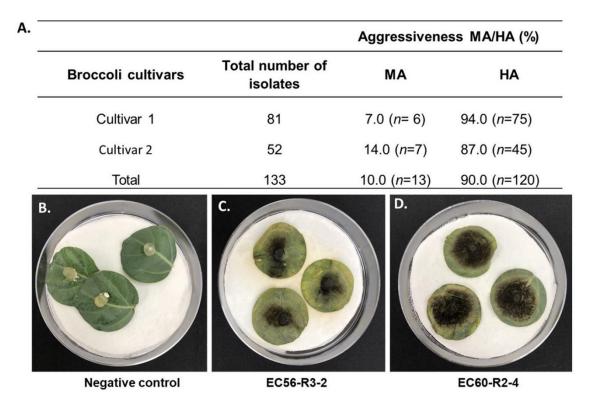


Figure 13. Percentage of *Alternaria brassicicola* isolates from naturally infested seedlots in broccoli that were moderately or highly aggressive (MA/HA) in a detached leaf assay. The panel A indicates the percent *A. brassicicola* isolates in each aggressiveness category isolated from Cultivar 1 and Cultivar 2. The panel B includes negative control while panels C to D shows the pictorial representation of a moderately aggressive (EC56-R3-2) and a highly aggressive (EC60-R2-4) *A. brassicicola* isolate in a detached leaf assay on broccoli.

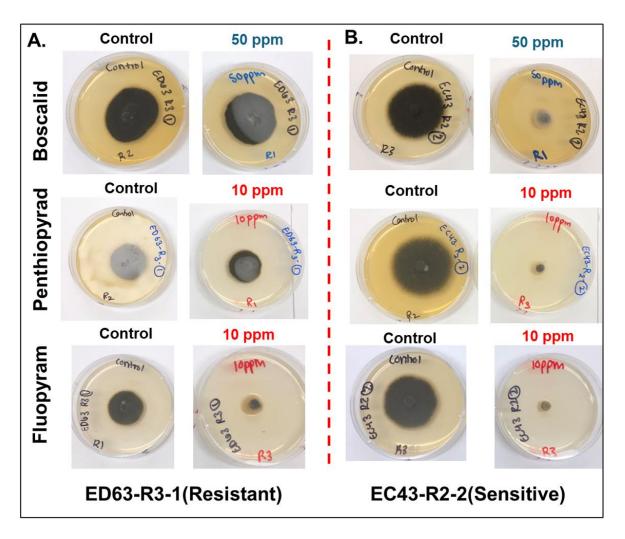


Figure 14. Radial growth assay of *Alternaria brassicicola* isolates (n = 58) from naturally infested commercial broccoli seedlots to three succinate dehydrogenase inhibitor fungicides (SDHI) fungicides, boscalid, penthiopyrad and fluopyram. Panels depict representations of radial growth assay for a resistant isolate (ED63-R3-1; panel A) and a sensitive isolate (ED23-R2-1; panel B).



Figure 15. Symptoms of Alternaria leaf blight on weeds found within and in the margins of fields in Connecticut in 2022. Single spore isolates collected from these weeds were sequenced and identified as *A. alternata*.

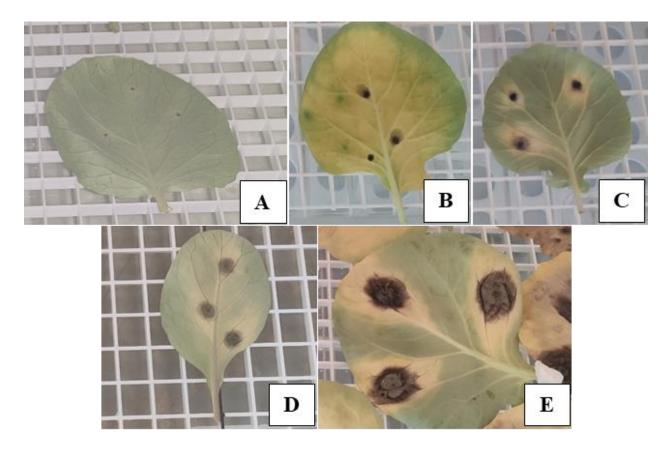


Figure 16. Levels of virulence of different isolates in cabbage five days after inoculation. Panels depicts the following: A= negative reaction; B= double size ca. 0.5cm^2 of the inoculation site plus chlorosis; C=>0.5 cm², but <1 cm² plus leaf chlorosis; D= increase in colonization with spores 'production, lesion >1 cm² and <4 cm² and leaf chlorosis; and E = rapid spore production with lesion size >4cm².

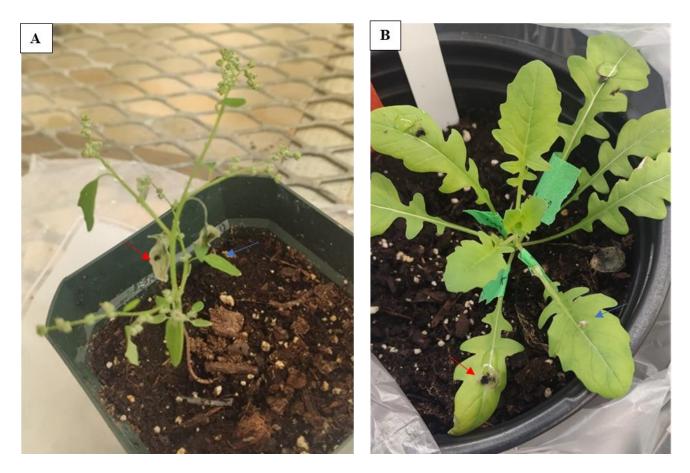


Figure 17. Lambsquarter (A) and cutleaf evening primrose (B) infected with *Alternaria brassicicola*. Red arrows indicate inoculation with the pathogen and blue arrows inoculated with water agar.