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A Rapid Screening Approach to Identify Resistance to Basil Bacterial Leaf Spot (*Pseudomonas cichorii*)

Alexander J. Barrett
New Use Agricultural and Natural Plant Products (NUANPP), Rutgers University, New Brunswick, NJ, USA; and Department of Plant Biology, Rutgers University, 59 Dudley Rd., New Brunswick, NJ 08901, USA

Nrupali Patel and Don Kobayashi
Department of Plant Biology, Rutgers University, 59 Dudley Rd., New Brunswick, NJ 08901, USA

Christian A. Wyenandt
Department of Plant Biology, Rutgers University, 59 Dudley Rd., New Brunswick, NJ 08901, USA; and Rutgers Agricultural Research and Extension Center, Rutgers University, Bridgeton, NJ 08302, USA

James E. Simon
New Use Agricultural and Natural Plant Products (NUANPP), Rutgers University, New Brunswick, NJ, USA; Department of Plant Biology, Rutgers University, 59 Dudley Rd., New Brunswick, NJ 08901, USA; and Center for Agricultural Food Ecosystems (RUCAFE), Rutgers University, New Brunswick, NJ 08901, USA

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Abstract. **Basil (*Ocimum* spp.) is among the most economically important culinary herbs grown worldwide. *Ocimum basilicum* includes both the European style sweet basil and the distinct Thai basil. Field, greenhouse, and indoor production practices involve growing dense compact stands that facilitate ideal conditions for the development of significant foliar diseases. One such disease, which has increased in prevalence and severity over the past decade throughout basil production regions in the United States and Europe, is bacterial leaf spot (BLS) caused by the pathogen *Pseudomonas cichorii*, a common bacterial pathogen with a broad host range including both wild plant species and commercial crops. Under favorable conditions, BLS is capable of causing significant losses before harvest, with growers currently having few economically viable chemical controls available for the disease. At present, no known resistance to BLS exists in commercially available cultivars. This study establishes a rapid pathogenicity screening assay for identification of potential BLS resistant germplasm within the O. *basilicum* and broader *Ocimum* genus for use in breeding genetically resistant lines. Three basil accessions, 'Lettuce Leaf' (*O. basilicum*), 'Mrs. Burns' Lemon' (*O. africanum*), and the coded Rutgers' 'RUGBX 88' (*Ocimum* spp.), were inoculated with *P. cichorii* at 10^8 CFUs and screened for disease severity at the cotyledon, first true leaf, second true leaf, and mature growth stage (four to five nodes). Disease severity scores (0 to 5) were assessed daily from 3 to 6 days post-inoculation (DPI) to determine the best growth stage for rapid screening purposes. Disease severity scores at the cotyledon and first true leaf stage displayed significant variation from mature plants while there were no significant differences (Dunn's test, $\alpha = 0.05$) in ratings between the second true leaf stage and maturity for all three accessions; the second true leaf growth stage was then selected to screen a collection of commercial basil cultivars for BLS resistance. Pathogenicity assays at the second true leaf stage revealed substantial variation in disease severity scores with the lowest being 2.28 ('Queen of Sheba') and the highest being 4.65 ('Siam Queen') across all lines tested. This methodology demonstrates the screening of basils at the second true leaf growth stage can readily be applied to *Ocimum* spp. Germplasm collections for the identification of potential BLS-resistant lines for incorporation into breeding programs.**

Basil (*Ocimum basilicum* L.) is one of the most widely grown and economically important herbs in the world. According to the Food and Agriculture Organization of the United Nations (FAO), there was an estimated $3,086,654 (USD thousands) global annual import market and $2,829,966 (USD thousands) export market in 2013, with a year-over-year increase in the export market of 25% (FAO and World Health Organization 2017). The basil essential oil market prioritizes high yield and specific aromatic compositions in their cultivars, both of which can be severely affected by pathogen infection; the fresh culinary sweet basil herb market demands even greater stringency, requiring product that is undamaged by pathogens, visually attractive, aromatic, and high yielding (Simon et al. 1990, 1999). Thus, marketable basil crops require protection from abiotic and biotic stresses throughout the growing period and must maintain excellent aesthetic characteristics throughout shipping, shelf life, and after sale to consumers.

While basil downy mildew, caused by *Peronospora belbahrii*, is one of sweet basil's most economically important diseases, an increasing number of reports have identified bacterial leaf spot (BLS) caused by *Pseudomonas cichorii* as an increasingly impactful disease that causes severe leaf damage and decreased marketability (Patel et al. 2019; Webb et al. 2016; Wyenandt et al. 2015). *P. cichorii* has a large host range and infects a wide array of commercial and noncommercial plants worldwide (Hikichi et al. 2013). This includes lettuce, eggplant, and sweet pepper, which are commonly grown together with basil crops in field and greenhouse operations. First identification of *P. cichorii* causing BLS on basil in the United States occurred in a Florida nursery in 1986 (Burgess et al. 1986) and has since been reported in Louisiana in 1998 (Holcomb and Cox 1998), Indiana in 2016 (Webb et al. 2016), Hawaii in 2017 on Thai basil (Luiz et al. 2018), and 1 year later, in New Jersey on sweet basil (Patel et al. 2019). Additionally, BLS of basil has been reported in Japan (Kanehashi et al. 2006), Taiwan (Hseu et al. 2013), and Brazil (Moreira et al. 2015). The BLS pathogen is quickly becoming common throughout basil growing regions in the United States and worldwide, presumably spreading via infested seed, infected plant material, or through its unidentified presence on its diverse host range (Hikichi et al. 2013; Wick 2021). The characteristic symptom of BLS infection is an irregular blackened necrosis on the stems and leaves; however, the pathogen may also reside on asymptomatic host plants, making its identification, control, and containment difficult (McGovern 2023; Wick 2021).

Currently, there are no effective methods to control BLS on basil. Little is known about the etiology of the disease, and it can develop sporadically, making prophylactic bactericidal controls costly and unfeasible. Additionally, pathogen dissemination and disease severity are highly dependent on favorable environmental conditions. For this reason, and also its negative impact on the success of field trials, a rapid method to test for genetic resistance under controlled conditions is needed. Differential resistance to the disease has been previously reported among *O. basilicum* cultivars (Holcomb and Cox 1998), but identification of a completely resistant cultivar has not been reported, and none are available on the market.

Differential resistance to BLS-causing *Xanthomonas campestris* pv. *vitians* and inheritance of that resistance has been clearly

demonstrated in lettuce allowing for advancements in understanding of the molecular mechanisms and breeding efforts (Bull et al. 2007; Hayes et al. 2014b; Kandel et al. 2022; Sandoya et al. 2019). These advancements in lettuce and other crops may provide a model for BLS resistance breeding in sweet basil.

The *Ocimum* genus is richly genetically diverse and displays high variation in ploidy levels and species compatibility (Gupta et al. 2018; Matthew et al. 2022; Pyne et al. 2018; Vieira et al. 2003). This diversity has been successfully harnessed to develop basil downy mildew–resistant sweet basil (*O. basilicum*) lines using both intra- and interspecies crosses (Ben-Naim et al. 2015, 2018; Pyne et al. 2015). There is little understanding of susceptibility or resistance to BLS caused by *P. cichorii* in sweet basil and the broader *Ocimum* genus. A rapid BLS screening approach will allow for the potential identification of resistant material from large germplasm collections for incorporation into active basil breeding programs. Rapid BLS resistance screens have been established in lettuce, and differential temporal severity has been demonstrated (Lu and Raid 2013; Pauwelyn et al. 2011). Additionally, rapid screening approaches have been developed and used for downy mildew resistance within the *Ocimum* genus (Pyne et al. 2014). The objective of this study was to develop a methodology for rapidly screening BLS resistance in the *Ocimum* genus using three species including the *O. africanum* (formerly *O. citriodorum*) 'Mrs. Burns' Lemon' (MBL), *O. basilicum* 'Lettuce Leaf' (LL), and the coded Rutgers' line 'RUGBX 88' (BX; *Ocimum* spp.), which is yet to be taxonomically identified. MBL and LL were selected for evaluation as a moderately resistant and highly susceptible cultivar based on the results reported by Holcomb and Cox (1998), and BX was selected as a highly resistant accession based on preliminary disease evaluations (Barrett, unpublished). This study evaluated each species at multiple growth stages finding repeatable differences in BLS susceptibility.

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J.E.S. is the corresponding author. E-mail: jimsimon@rutgers.edu.

Materials and Methods

Bacterial isolation and identification. Symptomatic samples of sweet basil 'Prospera' from a commercial basil farm located in Delray Beach, FL, USA, were collected in Jan 2022. Infected leaf samples were surface sterilized with deionized water. A 4- to 5-mm piece of leaf tissue was excised along the margins of the leaf lesion, macerated in sterile water, and then the extract was streaked onto King's B medium agar (KBM) (Schaad et al. 2001). Colonies developed at 30 °C within 24 h. Characteristic fluorescent colonies of *P. cichorii* were selected under 365-nm ultraviolet light and further cultured to pure colonies on KBM. A single colony in 3 mL of Luria broth (LB) was cultured overnight (12 h) at 30 °C and 250 rpm. A 20% glycerol stock solution was made and placed in a −80 °C freezer for long-term storage. One millilitre of the bacterial suspension was used for DNA extraction using the Wizard® Genomic DNA Purification Kit. Extracted DNA was amplified by polymerase chain reaction (30 cycles at 95 °C 2 min | 95 °C 30 s, 55 °C 30 s, 68 °C 2 min | 68 °C 8 min, 4 °C hold) for the 16S rRNA gene using the 27F and 1525R primers (∼1500 bp) (Lane 1991) and for a 520-bp region of the pathogenicity gene cluster *hrcRST* (Forward 5′ TTC TCC TGT TCG TGC TGG TC 3′, reverse 5′ CGA ATA CTC GGC ATC GGG AA 3′) commonly found in *Pseudomonas* species (Cottyn et al. 2011). Amplification was confirmed via gel electrophoresis and DNA bands were gel extracted for sequencing analysis (Genwiz by Azenta, South Plainfield, NJ, USA). The sequences were analyzed using the Basic Local Alignment Search Tool (BLAST) utility in the National Center for Biotechnology Information (NCBI) Database (https://www.ncbi.nlm.nih.gov/) to determine the isolate's sequence homology to *P. cichorii* (Altschul et al. 1990; Camacho et al. 2009). The consensus 16-s sequence was submitted to GenBank (PQ067361) and identified as strain RUPC1.

Identification of plant growth stage for rapid screening. Seeds of *O. africanum* MBL from Richters Herbs (Goodwood, Canada), *O. basilicum* LL from Urban Farmer (Westfield, IN, USA), and the Rutgers germplasm accession *Ocimum* spp. BX were each sown into two rows (24 seed total of each) of a 24-cell tray with a peat based soilless media (PRO-MIX BX) and grown to the cotyledon, first true leaf, second true leaf stage, or preflowering maturity (four to five nodes) under greenhouse conditions (23 to 27 °C, 16-/8-h day/night cycle, ∼400 µmol/m^2/s supplemental lighting) at the Rutgers New Jersey Agricultural Experiment Station Research Greenhouse located on Cook Douglas Campus, New Brunswick, NJ, USA. Plants designated for the mature growth stage were transplanted to 9-cm pots at the second true leaf stage to facilitate faster growth. Inoculum was prepared by adding a single *P. cichorii* colony to 100 mL of LB broth and incubated at 30 °C overnight on a rotary shaker (250 rpm). The liquid culture was centrifuged at 7500 g_n and rinsed twice with sterile water. The final bacterial pellet was resuspended in sterile deionized water and adjusted to 1×10^8 CFUs. Two to four hours before spray inoculation, 12 plants per accession per growth stage were randomly assigned to a tray and transferred to a constant mist chamber (100% humidity, 23 to 27 °C, 16-/8-h day/night cycle, ∼100 µmol/m^2/s supplemental lighting) for conditioning. After the conditioning period, plants at the cotyledon, first, and second true leaf stage were sprayed with a handheld sprayer five times each until leaf runoff, delivering ∼4 mL of inoculum suspension per individual plant. Mature plants were sprayed 10 times delivering about 8 mL of suspension. The day of conditioning and inoculation is considered day 0 in this screen. Post-inoculation, plants were randomly arranged and incubated in the mist chamber for 72 h before initial disease ratings to allow for symptom development. Inoculated plants remained in the mist chamber for the duration of the screen. Disease ratings were assessed four times from 3 d post-inoculation until day 6 using a 0 to 5 scale developed by Hayes et al. (2014a) for use in lettuce germplasm. Scores were evaluated as follows: 0 = no disease; 1 ≤ 10 lesions of <3 mm in size; 2 = individual disease lesions ≥ 3 mm or more than 10 lesions; 3 = large, coalesced lesions covering less than 20% of leaf area; 4 = lesions covering 20% to 50% of leaf area; 5 = lesions covering >50% of leaf area. Trials occurred from Apr to Sep 2023 and were fully replicated in triplicate.

Commercial cultivar screening. BLS susceptibility was determined for 35 commercial cultivars by screening plants at the second true leaf stage (Table 2). The same procedures established for the growth stage screening were followed for plant growth and inoculation. Twelve plants per cultivar were screened in a single randomized unreplicated trial. A single trial was conducted due to the consistency of response detected in the growth stage screen. Additionally, all individuals screened were moderately to highly susceptible to BLS. Replicated trials are recommended for cultivars displaying mean disease severity scores ≤2. Trials occurred from Jan to Feb 2024.

Data analysis. Data analysis was done in Rstudio using the kruskal.test function and the dunn.test function from the dunn.test package (Dinno 2017; R Core Team 2022). Thirty individual plants from three replicated screens of 10 plants each of LL, MBL, and BX were analyzed per growth stage to determine the inter- and intra-accession differences in means. Ten individual plants per cultivar were used in the commercial screening study.

Results and Discussion

Variation in disease severity resulting from *P. cichorii* inoculation was observed among the three basil species evaluated within the growth stage comparison screen.

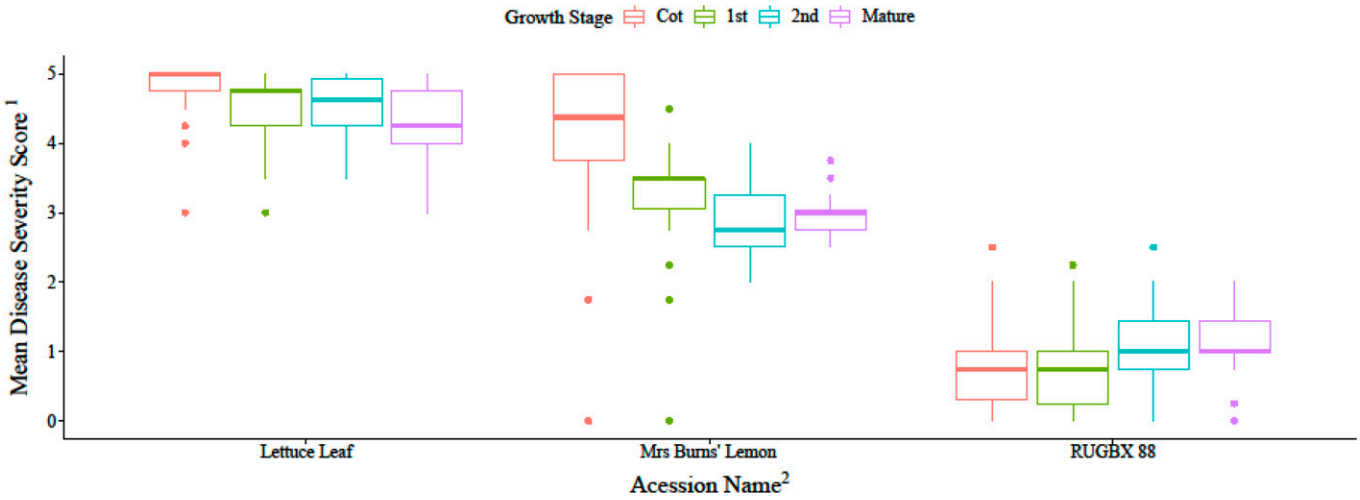

Fig. 1. Mean bacterial leaf spot severity score of three basil accessions at different growth stages. [1]Severity Score calculated as mean of 30 plants from 3 to 6 d post-inoculation. [2]Lettuce Leaf = *Ocimum basilicum*, Mrs. Burns' Lemon = *Ocimum africanum*, RUGBX 88 = *Ocimum* spp.

Basil LL displayed high susceptibility across all growth stages to *P. cichorii* inoculation with 3 to 6 d post-inoculation (DPI) mean scores ranging from 4.23 to 4.77; the lemon basil cv. MBL displayed intermediate susceptibility (2.92 to 3.33) at the first true leaf to maturity growth stages, to high (4.09) susceptibility at the cotyledon stage; BX displayed consistently low susceptibility across growth stages with a mean severity from 0.72 to 1.08. (Fig. 1). Results observed for MBL and LL are similar to those described previously by Holcomb and Cox (1998) wherein MBL was identified as a moderately resistant cultivar and LL a highly susceptible one.

A Kruskal–Wallis rank sum test was performed to determine significant differences in mean disease severity scores within each accession across the four growth stages. A significant difference was found within all three accessions: LL, $\chi^2 = 21.684$, $df = 3$, $P = 7.591e^{-05}$; MBL, $\chi^2 = 40.808$, $df = 3$, $P = 7.183e^{-09}$; and BX, $\chi^2 = 12.545$, $df = 3$, $P = 0.005731$, at $\alpha = 0.05$. Dunn's test with a Bonferroni correction was then performed for pairwise comparisons of the mean disease severity scores at each growth stage within each accession (Table 1). In LL, the cotyledon stage was the only significantly different (z-score: 4.641, $P = 0.00$, $\alpha = 0.05$) growth stage compared with maturity, with a mean severity score of 4.77 and 4.23, respectively. These scores, although significantly different, do both accurately capture the high susceptibility of the LL accession. The variation in these scores appears to be due to the early onset of severe intense symptoms at the cotyledon stage compared with the mature score as the 3 DPI mean severity score was 4.33 and 3.60, respectively. In MBL, both the cotyledon (z-score: 5.46, $P = 0.00$, $\alpha = 0.05$) and first true leaf stage (z-score: 2.839, $P = 0.0136$) mean severity scores were significantly different from maturity. The cotyledon stage rapidly developed severe disease symptoms and was unable to replicate the intermediate resistance phenotype displayed in mature plants, whereas the first true leaf stage initially displayed similar resistance to mature plants but quickly declined by 6 DPI (Table 1). The second true leaf stage's mean score did not significantly differ but did display a slightly higher disease severity by 6 DPI. In BX, only the first true leaf stage (z score: −2.845, $P = 0.0133$, $\alpha = 0.05$) significantly differed to maturity with a score of 0.72 and 1.08 respectively. The cotyledon stage, although scoring similarly to the first true leaf stage (0.78), did not pass the $\alpha = 0.05$ threshold (z-score: −2.331, $P = 0.0592$). These results demonstrate that the disease severity scores across the three basil species screened at the second true leaf stage were not significantly different from those recorded at plant maturity. This indicates that the second true leaf stage can accurately replicate the mature plant response to *P. cichorii* infection. Therefore, the second true leaf stage was used to pilot the rapid pathogenicity screening test for commercial basil cultivars.

The comparative screen included 35 commercial basil cultivars, and the three accessions used to determine pathogenicity parameters for BLS resistance at the second true leaf stage. The basil cultivars screened included Genovese, Italian, Thai, Lemon, Globe, Lettuce, and Purple Basil types (Table 2). The cultivars

Table 1. Summary of bacterial leaf spot disease severity across growth stages in three accessions of basil.

		Disease severity[i,ii]				
Accession	Growth stage[iii]	3 DPI	4 DPI	5 DPI	6 DPI	Avg
'Lettuce Leaf'	Cotyledon	4.33 ± 0.19	4.80 ± 0.09	5.00 ± 0.00	4.93 ± 0.07	4.77 ± 0.08[iv]
	First leaf	3.73 ± 0.16	4.50 ± 0.14	4.87 ± 0.08	4.93 ± 0.05	4.51 ± 0.09
	Second leaf	4.00 ± 0.14	4.50 ± 0.14	4.63 ± 0.09	4.93 ± 0.05	4.52 ± 0.08
	Mature	3.60 ± 0.11	4.10 ± 0.15	4.53 ± 0.12	4.70 ± 0.11	4.23 ± 0.10
'Mrs. Burns' Lemon'	Cotyledon	3.63 ± 0.26	4.00 ± 0.23	4.40 ± 0.22	4.33 ± 0.23	4.09 ± 0.21[iv]
	First leaf	2.33 ± 0.14	3.23 ± 0.20	3.67 ± 0.20	4.10 ± 0.20	3.33 ± 0.16[iv]
	Second leaf	2.40 ± 0.10	2.67 ± 0.10	3.17 ± 0.13	3.43 ± 0.15	2.92 ± 0.10
	Mature	2.60 ± 0.09	2.90 ± 0.10	3.10 ± 0.07	3.10 ± 0.06	2.92 ± 0.06
'RUGBX 88'	Cotyledon	0.80 ± 0.18	0.80 ± 0.14	0.80 ± 0.10	0.73 ± 0.15	0.78 ± 0.11[iv]
	First leaf	0.50 ± 0.12	0.73 ± 0.14	0.87 ± 0.10	0.77 ± 0.10	0.72 ± 0.10
	Second leaf	0.77 ± 0.11	1.03 ± 0.14	1.17 ± 0.14	1.37 ± 0.14	1.08 ± 0.11
	Mature	1.10 ± 0.12	1.07 ± 0.14	1.10 ± 0.12	1.07 ± 0.10	1.08 ± 0.11

[i]Disease severity scored on a 0 to 5 scale with 0 = no symptoms, 5 = more than 50% leaf area covered in necrotic lesions.
[ii]Standard error among three repeated experiments.
[iii]First leaf = first set of true leaves, second leaf = second set of true leaves, mature = four to five sets of true leaves.
[iv]Significant difference to Mature growth stage within each accession according to Dunn's test (alpha = 0.05).
DPI =days post-inoculation.

Table 2. Susceptibility to bacterial leaf spot for a selection of commercial basil cultivars at the second true leaf stage.

Accession information		
Name	Source	Avg disease severity[i]
RUGBX 88	Rutgers University	0.99 ± 0.07
Queen of Sheba	Hazzard's Seeds	2.28 ± 0.25
Boxwood	Hazzard's Seeds	2.42 ± 0.07
Profumo	Territorial Seed Co.	2.55 ± 0.11
Profumo di Genova	Renee's Garden	2.55 ± 0.19
Aurelia	Renee's Garden	2.58 ± 0.31
Amethyst Improved	Johnny's Selected Seeds	2.75 ± 0.23
Mrs. Burns' Lemon	Richters Herbs	2.84 ± 0.07
Italian Large Leaf	Pinetree Seeds	2.85 ± 0.08
Geno-Pro	Hazzard's Seeds	2.90 ± 0.10
Oriental Magic	Hazzard's Seeds	2.90 ± 0.06
Everleaf Emerald Towers	Harris Seeds	2.95 ± 0.07
Prospera 2	Johnny's Selected Seeds	3.01 ± 0.07
Red Rubin	Johnny's Selected Seeds	3.02 ± 0.19
Rutgers' Obsession	KBC	3.04 ± 0.09
Dwarf Greek	Hazzard's Seeds	3.08 ± 0.08
Persian Zing	Hazzard's Seeds	3.08 ± 0.13
Crimson King	Hazzard's Seeds	3.17 ± 0.12
Superbo	Dalponte Farms	3.21 ± 0.09
Italian Mountain Sweet	Adaptive Seeds	3.22 ± 0.09
Red Rosie	High Mowing Seeds	3.25 ± 0.11
Red Boza	Hazzard's Seeds	3.38 ± 0.11
Aroma 2	Johnny's Selected Seeds	3.42 ± 0.12
Rutgers' Devotion	KBC	3.42 ± 0.11
Columnar	Hazzard's Seeds	3.45 ± 0.25
Deep Purple	High Mowing Seeds	3.45 ± 0.20
Queenette Thai	Renee's Garden	3.45 ± 0.06
Siracusa	Adaptive Seeds	3.45 ± 0.10
Aromatto	Hazzard's Seeds	3.50 ± 0.16
Red Freddy Genovese	Hazzard's Seeds	3.52 ± 0.13
Amelia	NE Seed	3.55 ± 0.15
Golden Mountain Sweet Thai	Adaptive Seeds	3.72 ± 0.11
Licorice	Pinetree Seeds	3.88 ± 0.07
Red Crimson	Richters Herbs	3.98 ± 0.16
Lettuce Leaf	Urban Farmer	4.10 ± 0.09
Goddess	Hazzard's Seeds	4.45 ± 0.08
Oriental Breeze	Hazzard's Seeds	4.60 ± 0.08
Siam Queen	Renee's Garden	4.65 ± 0.11
Mean		3.23
Median		3.21
Range		3.66

[i]Mean score of 10 plants from 3 to 6 d post-inoculation with standard error between sample means. Scale: 0 = no symptoms, 5 = More than 50% leaf area covered in lesions.

displayed varying disease severity at 3 DPI when ratings were initiated that continued as the disease progressed (Fig. 2). The basil cultivars 'Queen of Sheba' (2.28) and 'Boxwood' (2.42) displayed the lowest susceptibility, although neither replicated the response of the highly resistant control BX (0.99). The most susceptible cultivars were the two Thai basils 'Siam Queen' (4.65) and 'Oriental Magic' (4.6), both with higher ratings than the susceptible control LL (4.1). A majority of the screened cultivars displayed a high level of susceptibility with severity scores ranging from 3.0 to 3.5. This pathogenicity screening data indicate that there is substantial variation in susceptibility to *P. cichorii* infection within current commercial cultivars, but complete resistance may not exist. For practicable breeding purposes, we consider accessions with severity scores ranging from 2.5 to 5.0 to be highly susceptible, and thus they should not be used in BLS-resistance breeding programs. It is recommended to focus on breeding individuals with mean severity scores at 2.0 or below, with the rare inclusion of individuals from 2.0 to 2.5. The 2.0 cutoff score ensures the exclusion of accessions that develop large necrotic lesions and maintains those that display limited disease severity while not forcing selection only to highly resistant individuals who likely reside outside of *O. basilicum*.

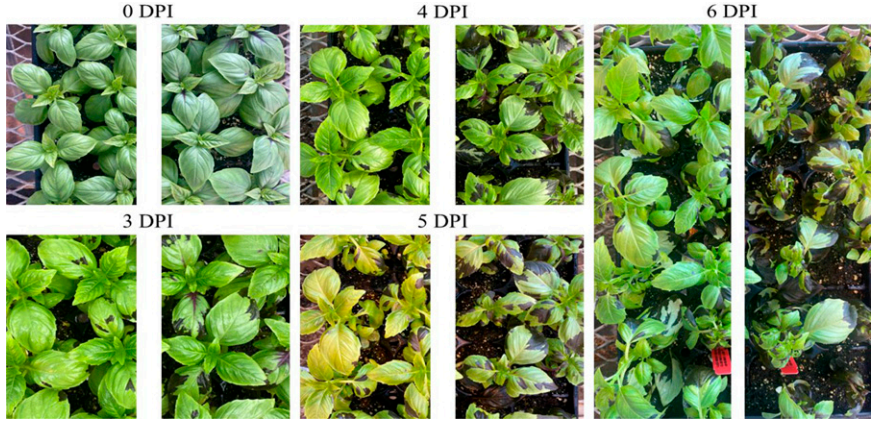

Fig. 2. Bacterial leaf spot progression from 0 to 6 d post-inoculation across two Rutgers coded *O. basilicum* Accessions RUGBX-127 (left) and RUGBX-128 (right).

The BX (*O.* spp.) highly resistant control used in this study does not readily intercross with *O. basilicum* species. Occasionally, these species barriers can be overcome with laborious tissue culture techniques such as embryo rescue (Ben-Naim et al. 2018). The *Ocimum* genus is known to have high amounts of diversity, admixture, and interspecies crossing events (Carović-Stanko et al. 2011; Gurav et al. 2020; Matthew et al. 2022; Pyne et al. 2018; Vieira et al. 2003). Thus, integrating resistance genes from the broader *Ocimum* genus is possible but challenging. The taxonomic identification of resistant germplasm within the *Ocimum* genus will be crucial for future successful breeding programs. It should be noted that 'RUGBX 88' phenotypically displays traits found in *O. gratissimum*, *O. teniflorum*, and *O. americanum*, although genetic identification for speciation of 'RUGBX 88' has not yet been completed. Nonetheless, this exotic basil line exhibits a high level of BLS resistance, emphasizing the utility and applicability of the disease screening methodology independent of host species.

The results of this study demonstrate that evaluation of BLS disease severity of basil germplasm at the second true leaf stage is replicable, streamlined, and comparable to mature plant responses. Given the ephemeral nature of BLS, field-level pathogenicity screenings are not reliable predictors of resistance because of the nonuniform dispersion and distribution in cultivated fields. Furthermore, the time, resources, and personnel required to screen accessions in the field is far greater than greenhouse evaluations. This rapid greenhouse approach uses a controlled high disease pressure environment, allowing for consistent inoculations and effective symptom development. This replicable and high throughput method allows for the screening of large germplasm collections for BLS resistance and is a crucial tool in the evaluation of breeding lines and their progeny.

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