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Susceptibility of Basil Cultivars and Breeding Lines to Downy Mildew (*Peronospora belbahrii*)

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***Abstract*. Downy mildew, caused by *Peronospora belbahrii*, is a new disease of basil (*Ocimum* spp.) in the United States. In 2009, different basil species, cultivars, and advanced breeding lines of sweet basil (30 in total) were evaluated for susceptibility to basil downy mildew in field trials in southern and northern New Jersey. Popular commercial sweet basil cultivars such as Martina, Nufar, and Poppy Joe were among the most susceptible to downy mildew. Symptoms and sporulation of *P. belhahrii* on *Ocimum ×citriodorum* and *O. americanum* cultivars were present but far less than on most *O. basilicum* cultivars evaluated. The cultivars Spice, Blue Spice, and Blue Spice Fil were the least susceptible to basil downy mildew with no visible symptoms. Similar results were observed in both field trials. This is the first report of potential resistance in *Ocimum* spp. to basil downy mildew. Observations from this study show that the development of resistant cultivars may be possible. Selection criteria such as foliar morphology, plant architecture as well as the presence of secondary metabolites are being examined as potential traits for developing downy mildew resistant basil cultivars.**

Sweet basil (*Ocimum basilicum* L., Fam. Lamaiaceae) is the most commercially important annual culinary herb crop grown in the United States. Sweet basil is grown for culinary use for both fresh and dry consumption and as a source of essential oil and oleoresin for manufacturing perfumes, food flavors, and aromatherapy products (Simon et al., 1990). Although sweet basil is the most popular culinary herb and the most common type of basil grown in the United States, it is just one species of *Ocimum*, which includes more than 50 species of herbs and shrubs, many of which are also referred to as basil, and which originate from the tropical regions of Asia, Africa, and Central and South America (Paton et al., 1999). Phenotypes from other *Ocimum* spp. are also edible and available in the horticultural trade as ornamentals, which have a wide range of aromas and flavors (Simon et al., 1999; Vieira and Simon, 2006). Basils accumulate a wide variety of terpenes and phenylpropanoids, secondary plant products that are constituents of the plants' aromatic volatile oils, also known as essential oils. It is the presence and relative ratio of these major and minor volatile constituents such as linalool, methyl chavicol, citral, eugenol, methyl cinnamate, and others that impart particular aromas and flavors to basils (Charles and Simon, 1990; Deschamps and Simon, 2006; Simon et al., 1990, 1999). Sweet basils, for example, contain a high content of linalool followed by methyl chavicol, eugenol, and 1,8 cineole; anise or licorice basils are rich in methyl chavicol; lemon basils are rich in citral; and cinnamon basils are rich in methyl cinnamate (Simon et al., 1990; Vieira and Simon, 2006). Basils also vary significantly in their morphology relative to growth habit; size, shape, texture, and orientation of leaves; and color of leaf, stem, and flower, resulting in a wide variety of ornamental cultivars (Simon et al., 1999).

Basil downy mildew, caused by *Peronospora belbahrii*, is a new disease of basil (*Ocimum* spp.) in the United States (Roberts et al., 2009). Basil downy mildew has been previously reported as a destructive disease in several countries and continents (Belbahri et al., 2005; Garibaldi et al., 2004a, 2005; Hansford, 1933; Khateri et al., 2007; McLeod et al., 2006; Ronco et al., 2008). In the United States, the pathogen was first discovered in Florida in the fall of 2007 (Roberts et al., 2009). Since that time, basil downy mildew has been found throughout the eastern United States and in regions of commercial basil production in the Midwest and California (McGrath et al., 2010; Wick and Brazee, 2009). Basil downy mildew can develop and spread rapidly throughout plantings during periods of high humidity, mild temperatures, poor air circulation, and extended durations of leaf wetness (Garibaldi et al., 2005, 2007; Spencer, 1981). In northern areas (i.e., in mid-Atlantic and northeast regions of the United States), it is thought that the obligate pathogen cannot survive outdoors during winter months between field-grown basil crops. Importantly, the basil downy mildew pathogen can also be spread through contaminated seed (Garibaldi et al., 2004b). The symptoms of basil downy mildew on an infected plant resemble, and can be mistaken for, a nutrient deficiency such as nitrogen deficiency (Fig. 1). Like other downy mildew pathogens, basil downy mildew only sporulates on the abaxial surface of infected leaves. Numerous dark purplish brown sporangia are produced during weather conditions favorable for disease development (Fig. 2). Little is known about the epidemiology of basil downy mildew making predictions about its development and spread difficult. In 2009, a monitoring program developed by McGrath, Cornell University, was established with cooperation from vegetable plant pathologists throughout the eastern United States. The priorities of this monitoring program were to determine if basil downy mildew could move northward up the East Coast and whether such a monitoring program would be useful to basil producers in the eastern United States. In 2009, 49 reports of basil downy mildew were logged from 17 states and Canada (Fig. 3). These reports confirmed the first major basil downy mildew

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Fig. 1. Chlorosis (yellowing) of sweet basil 'Nufar' infected with basil downy mildew on the left and healthy, uninfected *O. americanum* × *O. basilicum* 'Blue Spice' on the right.

Fig. 2. An underside of a sweet basil leaf infected by basil downy mildew.

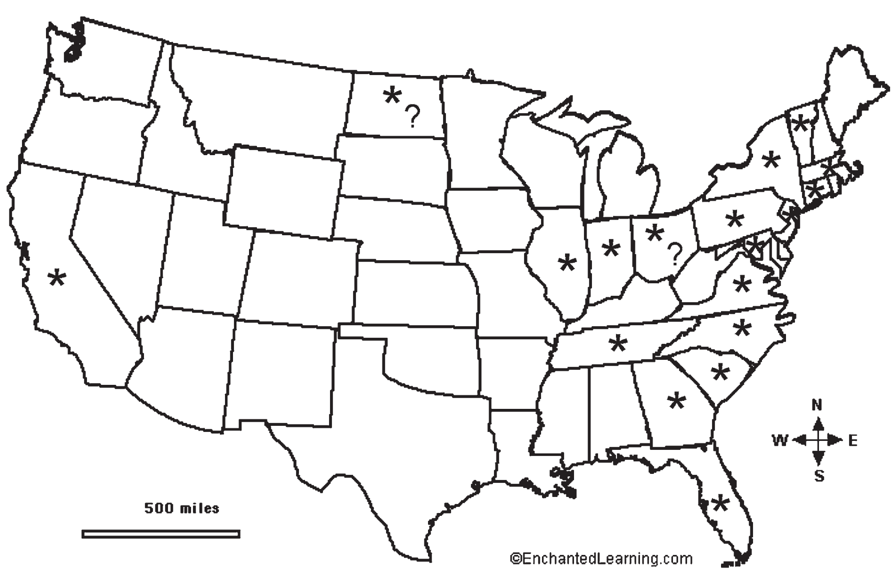

Fig. 3. States reporting basil downy mildew in 2009.

outbreak in the eastern United States and helped establish that under ideal weather conditions, basil downy mildew can disseminate over a wide geographic region in a single growing season in the United States (McGrath et al., 2010). In 2009, 100% losses occurred on some farms in New Jersey and elsewhere is the United States. Once basil plants become infected and develop symptoms, they are no longer marketable as a fresh product (Fig. 2). There are few registered chemical control options for commercial basil producers. Currently, there is no known resistance or tolerance to basil downy mildew leaving 100% of the sweet basil acreage in the eastern United States vulnerable to the pathogen. Without adequate chemical control options and genetic resistance, basil downy mildew has the potential to destroy basil production in the eastern United States and in all other areas where basil is being produced.

The objective of this study was to evaluate current commercial cultivars, sweet basil breeding lines, and other *Ocimum* spp. for their potential susceptibility to basil downy mildew.

Materials and Methods

In 2009, at the Rutgers Agricultural Research and Extension Center (RAREC) near Bridgeton, NJ, different basil species, cultivars, and advanced breeding lines (30 in total) were evaluated for susceptibility to basil downy mildew in a randomized complete block design with four replications (Table 1). On 27 July, 6-week-old basil seedlings were transplanted into raised beds spaced 1.5 m apart with black plastic mulch and drip irrigation. All seedlings were transplanted at 23-cm spacing between plants in two rows spaced on a 41-cm center per bed. The total number of plants per plot varied, but in general, each plot consisted of no less than 20 plants. On 31 July, the field was artificially infested with *P. belbahrii* by transplanting 10 infected sweet basil plants in a staggered form throughout the entire field. All plots were irrigated two to three times per week for ≈2 h and fertilized as needed using 0.91 kg 20N–20P–20K (Peters Professional 20-20-20; Scotts-Sierra Horticultural Products Company, Marysville, OH). On 21 Aug., all plots were manually pruned back to approximately a 15- to 30-cm height by hand depending on growth habit. On 20 Aug. and 21 Sept., basil plants were rated for downy mildew infection using an ordered categorical scale where 0.0 equaled no visible symptoms (i.e., chlorosis) or downy mildew sporulation, 1.0 equaled symptoms of downy mildew (i.e., chlorosis) and light sporulation, 2.0 equaled chlorosis and heavy downy mildew sporulation, or 3.0 equaled chlorosis and heavy, dense downy sporulation on the abaxial surface of leaves. Rainfall accumulation at RAREC for the months of July, August, September, and October were 6.15, 20.4, 11.8, and 11.3 cm, respectively. A separate observational trial with only two replications using some of the same cultivars and advanced breeding lines was established in a similar fashion at the Clifford E. and Melda C. Snyder Research

Table 1. Basil species, cultivar, or advanced breeding line and rating scores to basil downy mildew infection and general leaf architecture on 20 Aug. and 21 Sept. 2009 at the Rutgers Agricultural Research and Extension Center, Bridgeton, NJ.

Species	Cultivar/breeding line	Rating date[z] 20 Aug.	Rating date[z] 21 Sept.	Leaf type[y]
O. basilicum	Genovese	2.00 ab	3.00 a[x]	D
O. basilicum	Magical Michael	0.75 defg	3.00 a	FL, U
O. basilicum	Mariden	1.50 bcd	3.00 a	FL, D
O. basilicum	Superbo	2.00 ab	3.00 a	FL, D
O. basilicum	Queentette	2.00 ab	3.00 a	U
O. basilicum	Aroma 2	0.75 defg	3.00 a	FL, D
O. basilicum	Italian large Leaf	1.75 bc	3.00 a	D
O. basilicum	Genoveser Martina	1.25 bcde	3.00 a	D
O. basilicum	E93.7962	1.75 bc	3.00 a	D
O. basilicum	Poppy Joe's	2.75 a	3.00 a	FL, D
O. basilicum	Opal Purple Variegated	2.00 ab	3.00 a	FL, U, D
O. basilicum	Gecofure	1.25 bcde	2.75 ab	FL, D
O. basilicum	Nufar	2.00 ab	2.75 ab	D
O. basilicum	Rubin	0.50 efg	2.75 ab	FL, D
O. basilicum	Cinnamon	1.00 cdef	2.50 b	U
O. basilicum	E93.8109	1.00 cdef	2.00 c	FL, U
O. basilicum	Amethyst Imp	1.75 bc	1.75 c	FL, D
O. basilicum	Mrs. Burn's Lemon	0.50 efg	1.00 d	FL, U
O. basilicum	Red Leaf	0.25 fg	1.00 d	U
O. citriodorum	Lemon Std	1.00 cdef	1.00 d	FL, U
O. citriodorum	Lemon Mrs. Burns	1.00 cdef	1.00 d	FL, U
O. citriodorum	Lemona	1.00 cdef	1.00 d	U
O. sp.	Lime	0.00 g	1.00 d	U
O. basilicum	Red Rubin	0.75 defg	1.00 d	FL, U, D
O. basilicum	Sweet Aden	0.00 g	0.75 d	FL, U
O. citriodorum	Lemon	0.00 g	0.75 d	U
O. americanum × *O. basilicum*	Blue Spice	0.00 g	0.00 e	FL, U
O. sp.	Spice	0.00 g	0.00 e	FL, U
O. basilicum	Blue Spice F1	0.00 g	0.00 e	U

[z]Mean ratings for downy mildew development using an ordered categorical scale in which 0.0 equaled no visible symptoms (i.e., chlorosis) or downy mildew sporulation, 1.0 equaled symptoms of downy mildew (i.e., chlorosis) and light sporulation, 2.0 equaled chlorosis and heavy downy mildew sporulation, or 3.0 equaled chlorosis and heavy, dense downy sporulation on the abaxial surface of leaves. The *O. basilicum* advanced breeding lines of sweet basil each exhibited downy mildew injury ranging from 2.0 to 3.0.

[y]General architecture (i.e., structure) of basil leaves in which FL equaled mostly flat shaped leaves on plant, U equaled plants with leaves with upward flaring margins, and D equaled plants where margins of leaves were mostly downward flaring.

[x]Means followed by the same letter are not significantly different according to least significant difference test at $P \leq 0.05$.

and Extension Farm, Pittstown, in northern New Jersey. The evaluation trial in northern New Jersey was allowed to become naturally infested with basil downy mildew. This trial was evaluated once on 6 Oct. using the same rating scale. At the first rating date in each trial, general leaf-type architecture was described by visually examining leaves of each cultivar or breeding line. General leaf architecture was recorded as those plants whose leaves were mostly flat (FL), had mostly upward flaring leaf margins (U), or plants where margins of leaves were mostly downward flaring (D) (Tables 1 and 2).

Analysis of variance using the PROC GLM procedure with SAS Version 9.1.3 (Version 9.13; SAS Institute, Cary, NC) was done to determine any significant differences in basil downy mildew development between cultivars and/or breeding lines. Model assumptions were investigated using scatter plots of standardized residuals across predicted values and box plots of standardized residuals across all factors in the model.

Results and Discussion

In southern New Jersey in 2009, *Ocimum basilicum* was the most susceptible species among all *Ocimum* spp. evaluated (Table 1). Although sporulation ratings varied among the sweet basil cultivars by rating date, all sweet basil cultivars and breeding lines were susceptible to basil downy mildew and popular commercial cultivars such as Martina, Nufar, and Poppy Joe were among the most susceptible (Table 1). Symptoms and sporulation on *Ocimum ×citriodorum* and *O. americanum* cultivars were present but far less than on *O. basilicum* cultivars at both rating dates. In some instances, chlorosis resulting from downy mildew was visible on *O. citriodorum*, but no sporulation was present on the underside of leaves in cultivars such as Lemon Std. or only very light sporulation was visible on the margins of older, chlorotic leaves in cultivars such as Lemon or Lime. The cultivars Spice, Blue Spice, and Blue Spice Fil were the least susceptible to basil downy mildew with no visible symptoms of chlorosis or sporulation developing on the leaves at both rating dates and locations (Table 1). In ornamental cultivars such as Magical Michael, sporulation was present in small, irregular brown lesions on some leaves, which were quite different from the chlorosis of leaves that were present and preceded downy mildew sporulation in other *Ocimum* spp. and cultivars. Similar findings were observed in a smaller, non-inoculated (i.e., naturally infested) basil cultivar evaluation trial with two replications at the Snyder Agricultural Research Center, Pittstown, located in northern New Jersey (Table 2) and in trials conducted in Florida in 2009 (Raid et al., unpublished data).

This is the first report demonstrating potential resistance in *Ocimum* spp. to basil downy mildew. Observations from this study show that some basils, including a number of exotic *Ocimum basilicum* types as well as other *Ocimum* spp., may provide a potential source of resistance or tolerance to basil downy mildew. All sweet basils evaluated in this study were susceptible to basil downy mildew. Although sources of resistance in basil may be available, those which exhibited potential resistance in this study are not commercially acceptable as sweet basil as a result of their distinct morphological and aromatic characteristics. Importantly, the popular sweet basil cultivars (*O. basilicum*) that are susceptible to basil downy mildew can be hybridized with other more exotic members of this species, which showed tolerance as well as with basils originating from *O. citriodorum* and *O. americanum*. The challenge will be to introduce basil downy mildew resistance while maintaining the phenotypic and characteristic aroma and quality of the traditional sweet basil that is acceptable in the marketplace. Selection criteria such as foliar morphology, plant architecture as well as the presence of secondary metabolites and other factors that provide a less favorable microenvironment to the pathogen need to be examined as potential avenues for developing downy mildew-resistant sweet basil cultivars. Until this can be achieved, basil growers will have to rely

Table 2. Basil species, cultivar, or advanced breeding line and susceptibility scores to basil downy mildew infection and general leaf architecture on 6 Oct. 2009 at the Clifford E. and Melda C. Snyder Research and Extension Farm, Pittstown, NJ.

Species	Cultivar/breeding line	6 Oct.[z]	Leaf type[y]
O. basilicum	Magical Michael	3.00 a[x]	FL, U
O. basilicum	Gecofure	3.00 a	FL, D
O. basilicum	Mariden	3.00 a	FL, D
O. basilicum	Superbo	3.00 a	FL, D
O. basilicum	Queentette	3.00 a	U
O. basilicum	Nufar	3.00 a	D
O. basilicum	Aroma 2	3.00 a	FL, D
O. basilicum	Italian large Leaf	3.00 a	D
O. basilicum	Genoveser Martina	3.00 a	D
O. basilicum	E93.7962	3.00 a	D
O. basilicum	Amethyst Imp	3.00 a	FL, D
O. basilicum	Opal Purple Variegated	3.00 a	FL, U, D
O. basilicum	Red Rubin	3.00 a	FL, U, D
O. basilicum	Rubin	3.00 a	FL, D
O. basilicum	Cinnamon	2.00 b	U
O. basilicum	Red Leaf	1.50 c	U
O. basilicum	E93.8109	1.50 c	FL, U
O. basilicum	Mrs. Burn's Lemon	1.00 d	FL, U
O. citriodorum	Lemon Mrs. Burns	1.00 d	FL, U
O. citriodorum	Lemona	1.00 d	U
O. citriodorum	Lemon	1.00 d	U
O. americanum × *O. basilicum*	Blue Spice	0.00 e	FL, U
O. sp.	Spice	0.00 e	FL, U
O. sp.	Lime	0.00 e	U

[z]Mean ratings (two replications) for downy mildew development using an ordered categorical scale in which 0.0 equaled no visible symptoms (i.e., chlorosis) or downy mildew sporulation, 1.0 equaled symptoms of downy mildew (i.e., chlorosis) and light sporulation, 2.0 equaled chlorosis and heavy downy mildew sporulation, or 3.0 equaled chlorosis and heavy, dense downy sporulation on the abaxial surface of leaves. The *O. basilicum* advanced breeding lines of sweet basil each exhibited downy mildew injury ranging from 2.0 to 3.0.

[y]General architecture (i.e., structure) of basil leaves in which FL equaled mostly flat shaped leaves on plant, U equaled plants with leaves with upward flaring margins, and D equaled plants where margins of leaves were mostly downward flaring.

[x]Means followed by the same letter are not significantly different according to least significant difference test at $P \leq 0.05$.

on multiple applications of the few commercial fungicides currently registered to produce a marketable crop. Additionally, for organic basil growers, control of basil downy mildew will be even more challenging because there are fewer approved products labeled for organic use.

Literature Cited

Belbahri, L., G. Calmin, F. Lefort, and J. Pawlowski. 2005. Phylogenetic analysis and real time PCR detection of a new *Peronospora* species responsible for downy mildew disease of sweet basil and sage. Mycol. Res. 109:1276–1287.

Charles, D.J. and J.E. Simon. 1990. Comparison of extraction methods for the rapid determination of essential oil content and composition of basil. J. Amer. Soc. Hort. Sci. 115:458–462.

Deschamps, C. and J.E. Simon. 2006. Terpenoid essential oil metabolism in basil (*Ocimum basilicum* L.) following elicitation. J. Essential Oil Res. 18:618–621.

Garibaldi, A., D. Bertetti, and M.L. Gullino. 2007. Effect of leaf wetness duration and temperature on infection of downy mildew (*Peronospora* sp.) basil. J. Plant Dis. Prot. 114:6–8.

Garibaldi, A., A. Minuto, and M.L. Gullino. 2005. First report of downy mildew caused by *Peronospora* sp. on basil (*Ocimum basilicum*) in France. Plant Dis. 89:683.

Garibaldi, A., A. Minuto, G. Minuto, and M.L. Gullino. 2004a. First report of downy mildew on basil (*Ocimum basilicum*) in Italy. Plant Dis. 84:1154.

Garibaldi, A., G. Minuto, D. Bertetti, and M.L. Gullino. 2004b. Seed transmission of *Peronospora* sp. of basil. J. Plant Dis. Prot. 111:465–469.

Hansford, C.G. 1933. Annual report of the mycologist. Rev. Appl. Mycol. 12:421–422.

Khateri, H., G. Calmin, N. Moarrefzadeh, L. Belbahri, and F. Lefort. 2007. First report of downy mildew caused by Peronospora sp. on basil in northern Iran. J. Plant Pathol. 89:S70.

McGrath, M.T., C.A. Wyenandt, R.N. Raid, M. Babadoost, and R.L. Wick. 2010. Occurrence of basil downy mildew in the eastern US in 2009. Phytopathology 100:S196.

McLeod, A., S. Coertze, and L. Mostert. 2006. First report of a *Peronospora* species on sweet basil in South Africa. Plant Dis. 90:1115.

Paton, A., M.R. Harley, and M.M. Harley. 1999. *Ocimum*: An overview of classification and relationships, p. 1–38. In: Hiltunen, R. and Y. Holm (eds.). *Basil the genus* Ocimum. Harwood Academic Publishers, Amsterdam, The Netherlands.

Roberts, P.D., R.N. Raid, P.F. Harmon, S.A. Jordan, and A.J. Palmateer. 2009. First report of downy mildew caused by a *Peronospora* sp. on basil in Florida and the United States. Plant Dis. 93:199.

Ronco, L., C. Rollán, Y.J. Choi, and H.D. Shin. 2008. Downy mildew of sweet basil (*Ocimum basilicum*) caused by *Peronospora* sp. in Argentina. BSPP New Disease Reports. 8 Aug. 2010. <http://www.bspp.org.uk/ndr/jan2009/2008-57.asp>.

Simon, J.E., M.R. Morales, W.B. Phippen, R.F. Vieira, and Z. Hao. 1999. Basil: A source or aroma compounds and a popular culinary and ornamental herb, p. 499–505. In: Janick, J. (ed.). Perspectives on new crops and new uses. ASHS Press, Alexandria, VA.

Simon, J.E., J. Quinn, and R.G. Murray. 1990. Basil: A source of essential oils, p. 484–489. In: Janick, J. and J.E. Simon (eds.). Advances in new crops. Timber Press, Portland, OR.

Spencer, D.M. 1981. The downy mildews. Academic Press, London. UK.

Vieira, R.F. and J.E. Simon. 2006. Chemical characterization of basil (*Ocimum* spp.) based on volatile oils. Flavour Frag. J. 21: 214–221.

Wick, R.L. and N.J. Brazee. 2009. First report of downy mildew caused by a *Peronospora* species on sweet basil (*Ocimum basilicum*) in Massachusetts. Plant Dis. 93:318.