SPINACH SEED CROP (*Spinacia oleracea*)
Cladosporium leaf spot; *Cladosporium variabile*
Stemphylium leaf spot; *Stemphylium botryosum*

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Evaluation of fungicides for control of leaf spot in spinach seed crops, 2004.

Fungicides were evaluated for control of leaf spot in a hybrid spinach seed crop trial established on a Puget silt loam soil at the Washington State University NWREC. The field site was fumigated on 2 Oct 03 with Tri-Con 57/43 (methyl bromide/chloropicrin) at 350 lb/A to a depth of 8 in. and sealed with plastic tarp, for control of Fusarium wilt caused by *Fusarium oxysporum* f. sp. *spinaciae*. The tarp was removed on 6 Nov 03. Male and female stock seed of a proprietary spinach hybrid were planted on 23 Apr 04 at a ratio of two male rows for every six female rows, with 22 in.-spacing between rows and 2 in.-spacing within rows. Fertilizer (18-46-0) was applied in-furrow (375 lb/A) at planting. The trial was tilled between rows on 1 Jun and side-dressed with 34-0-0 (150 lb/A) on 2 Jun. Plots, 20 ft-long and consisting of six female rows and a male row on each side, were established in the field on 17 Jun with 15 ft alleys separating plots at each end. The experiment was arranged in a randomized complete block design with four replications. The first fungicide applications were initiated at the start of anthesis. Due to windy and rainy conditions, the first application was completed over four days, with the control and Dithane applications made on 9 Jul; the Bravo WeatherStik, Cabrio EG, Manex, and Rovral 4F applications on 11 Jul, and the remaining fungicides applied on 12 Jul. Fungicides were applied again on 22 Jul, and 8 and 19 Aug. All fungicides were applied in 44.5 gal/A at 40 psi using a CO_2-pressurized tractor-mounted sprayer fitted with a 7 nozzle boom (8003 flat fan tips at 20-in. spacing). R-11 surfactant was used with all fungicide applications at 1.0 pt/100 gal. Control plots were sprayed with water and R-11. Each plot was inoculated with approx. 0.18 gal of a mixed spore suspension of *C. variabile* and *S. botryosum* in 0.01% Tween 80 on 13 Jul (1.7×10^5 and 1.7×10^4 spores/ml, respectively), 23 Jul (2.1×10^4 and 1.1×10^4 spores/ml, respectively), 9 Aug (2.1×10^4 and 1.5×10^4 spores/ml, respectively), and 20 Aug (1.9×10^3 and 9.0×10^3 spores/ml, respectively). All inoculations were made at 15 psi using the sprayer described above. Male rows were tilled under on 30 Jul. Severity of leaf spot was assessed on 17 and 31 Aug. The middle 10 ft of the center four female rows were cut by hand from each plot on 7 Sep. Because of persistent rain, harvested plants were placed onto 10 ft x 12 ft sections of Remay and transported to covered storage to dry. Plants from the driest plots were threshed on 16 Sep and the remaining plots were threshed on 23 Sep. The seed was cleaned and sized according to commercial specifications for the proprietary hybrid. In Oct, a freeze-blotter assay was used to determine the incidence of *Stemphylium*, *Cladosporium*, and other fungi in 100 seed/plot. The seed was surface-sterilized for 1 min in 1.2% NaOCl, triple-rinsed in sterile deionized water, and placed onto damp blotters in plastic Petri plates (20 seed/plate). The seed imbibed on the blotters in the dark for 24 h, and was then incubated at –4ºF for 24 h followed by 12 d at 75ºF under a 12h/12h day/night cycle with near-UV and cool white fluorescent light by day. The seed was examined using a dissecting microscope (8 to 100X magnification) 5, 8, and 11 days after plating. Germination of 100 seed/plot was assessed using the AOSA (Association of Official Seed Analysts) protocol. Mean temperature and total rainfall for Apr, May, Jun, Jul, Aug, and Sep were 52.3°F and 0.28 in., 56.2°F and 3.68 in., 61.2°F and 1.8 in., 64.5°F and 0.51 in., 65.3°F and 2.6 in., 57.6°F and 3.4 in., respectively.

All fungicides except Topsin M 70WP reduced severity of leaf spot relative to the control plots. Greatest control was provided by Pristine EG and Cabrio EG, followed by Rovral 4F, Amistar, and Sovran. By 31 Aug, only Cabrio EG and Pristine EG applications kept severity of leaf spot below 40%, compared to 94% for control plots and 46 to 91% for the other fungicide treatments. Bravo WS and Dithane DF, the primary fungicides currently used by spinach seed growers in the Pacific Northwest for control of leaf spot, displayed moderate efficacy against leaf spot. Seed yield was significantly greater for plots treated with Dithane DF, Rovral 4F, Cabrio EG, Amistar, and Pristine EG compared to control plots and plots treated with Topsin M 70WP. No significant differences in seed germination or incidence of seedborne *C. variabile* were observed among fungicide treatments. However, the incidence of seedborne *S. botryosum* differed significantly among treatments. All fungicides except Topsin M 70WP and Bravo WeatherStik significantly reduced the incidence of seedborne *S. botryosum* compared to control plots (73%). Pristine EG and Rovral 4F were the most effective at reducing infection of seed by *S. botryosum* (<14%), followed by Cabrio EG and Amistar (<31%). Severity of leaf spot on 17 Aug was significantly correlated with seed yield ($r = -0.37$, $P = 0.0187$) and incidence of seedborne *S. botryosum* ($r = 0.84$, $P < 0.0001$). The predominant leaf spot symptoms were of Stemphylium leaf spot, reflecting the higher incidence of seedborne *S. botryosum* compared to *C. variabile*.

Treatment and rate/A	Leaf spot severity (%)[z] Aug 17	Leaf spot severity (%)[z] Aug 31	Seed yield (lb/40 ft)[y]	% Seed infected[x] *Stemphylium botryosum*	% Seed infected[x] *Cladosporium variabile*
Control (water) …….....	84 a[w]	94 a	0.66 b	73.0 a	2.0 a
Topsin M 70WP 1.5 lb …….....	84 a	91 ab	0.62 b	66.5 abc	0.0 a
Bravo WeatherStik 3.0 pt …….	69 b	83 ab	0.92 ab	69.5 ab	0.8 a
Manex 1.6 qt ……	69 b	80 bc	0.84 ab	57.0 bcd	1.3 a
Dithane DF Rainshield 2.0 lb …..	59 b	70 c	1.21 a	53.8 cd	2.0 a
Amistar 5.0 oz ……..	21 c	48 d	1.09 a	30.8 e	0.3 a
Rovral 4F 2.0 pt ……..…	21 c	46 de	1.19 a	13.0 f	1.8 a
Sovran 4.8 oz ……	19 c	51 d	0.93 ab	51.3 d	0.3 a
Pristine EG 0.7 lb …….	13 c	33 f	1.08 a	12.5 f	0.0 a
Cabrio EG 1.0 lb ……..	11 c	35 ef	1.16 a	22.5 ef	0.3 a

[z] Severity of leaf spot rated as % foliage with symptoms of Cladosporium leaf spot or Stemphylium leaf spot per plot.
[y] Seed yield = lb of marketable seed (between screen sizes 6 and 11) per 40 ft of row (10 ft from center four rows per plot).
[x] Mean % of 100 seed infected/plot. Seed assayed using a freeze-blotter assay described above.
[w] Means followed by the same letter within a column are not significantly different based on Fisher's Protected LSD ($P \leq 0.05$).