Effect of Crop Debris Management on Severity of Stemphylium Purple Spot of Asparagus

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ABSTRACT

Johnson, D. A. 1990. Effect of crop debris management on severity of Stemphylium purple spot of asparagus. Plant Dis. 74: 413-415.

Asparagus debris from the previous season's fern was either incorporated into the soil or left on the soil surface in large plots in a commercial field. Severity of Stemphylium purple spot on spears was significantly reduced when debris was incorporated into the soil. Incorporation of debris into the soil was equally effective in late fall and in late winter. Ascospores and conidia of *Pleospora herbarum* on debris from the previous year's fern growth served as primary inoculum. Volunteer asparagus seedlings became infected during the harvest season and may be important as a substrate for inoculum increase. They also may act as a bridge to carry inoculum through the harvest season to the time when the fern is allowed to regrow.

Stemphylium vesicarium (Wallr.) Simmons causes a disease of asparagus (*Asparagus officinalis* L.) spears and fern in many growing regions of the world (1,3,5,8). Symptoms on spears consist of elliptical, slightly sunken, purplish spots (8), which may result in rejection when the asparagus is graded for the fresh market. The disease appears on the fern as tan to brown lesions with dark purple margins, and cladophylls (small needle-like branches) may drop when infections are severe. Pseudothecia of the teleomorph of *S. vesicarium, Pleospora herbarum* (Pers. ex Fr.) Rabenh., form on senescing ferns in the fall and winter in Washington state. Ascospores mature by early spring, and conidia are also produced on the previous year's fern growth (5). Infections occur through open stomata and wounds on current-season green asparagus tissue when the temperature is favorable and moisture is present from rainfall or sprinkler irrigation (5,6).

An association between debris from the previous season's fern growth left on the soil surface and disease incidence of purple spot has been observed (4). Removal of this source of overwintering inoculum from the soil surface may be a practical method of managing Stemphylium purple spot on spears. The purposes of this study were to quantify the effect on disease severity of incorporating asparagus crop debris into the soil before the next harvest season, to investigate the role of conidia of *S. vesicarium* as primary inoculum, and to determine the effect of soil burial on development of ascospores of *P. herbarum*.

Plant Pathology New Series No. 0050. Project 0678. College of Agriculture and Home Economics Research Center, Washington State University, Pullman 99164.

Accepted for publication 29 December 1989 (submitted for electronic processing).

MATERIALS AND METHODS

The experiments were done in a commercial asparagus field planted in 1981 with Green Giant hybrid on a sandy loam soil east of Burbank, WA. Row spacing was 1.5 m, and the planting was watered by a center-pivot irrigation system. Plot size was 11 rows (16.5 m) by 91 m in 1987 and 15 rows (22.5 m) by 91 m in 1988 and 1989. The following three treatments were applied after the fern was killed by frost in 1986: 1) in November (fall) 1986, the season's fern growth was broken up and incorporated about 4–9 cm deep in the soil with a New Holland Ace Dynadrive Bombford tiller; 2) the fern was incorporated into the soil as before but in February (winter) 1987; and 3) the fern was chopped and left on the soil surface in February 1987. Only two treatments were applied in February before each of the 1988 and 1989 harvest seasons: 1) fern was incorporated as before, and 2) the fern was chopped and left on the soil surface. Treatments were arranged in a randomized complete block each year with six replicates in 1987 and 1988 and four replicates in 1989.

Yield data were collected from the two center rows of each plot by cutting and weighing the spears for 55 days from 15 April to 17 June 1987. Yields were not determined in 1988 and 1989 because of financial constraints and because yields did not differ in 1987.

Disease severity on spears was determined in the field two to eight times each year, usually 1–3 days after rainfall, by visually inspecting 100 spears greater than 10 cm in length in the center rows of each plot. Disease severity on volunteer seedlings also was determined once or twice each year by collecting eight seedling shoots, with cladophylls that had just expanded fully, from five locations 10 m apart along the center row. Spears and volunteer seedlings were graded into one of the following disease classes, based on the number of lesions per spear or seedling: 0 = no lesions; 1 = 1–10 lesions; 2 = 11–30 lesions; 3 = 31–50 lesions; and 4 = greater than 50 lesions. A disease severity index (DSI) ranging from 0 (no disease on any spear or seedling) to 100 (all spears or seedlings in class 4) was calculated for each sampling date, using the formula of Sherwood and Hagedorn (9): DSI = Σ (disease class $\times$ number of spears or seedlings in that class) 100/total number of spears or seedlings $\times$ 4.

The amount of debris in plots was estimated by raking and weighing debris from a 3- $\times$ 3-m section in the center of each plot on 9 June in 1987 and 26 May in 1989.

Tissue samples from volunteer seedlings with lesions were collected during the spring harvest seasons in 1988 and 1989, washed, surface disinfested with 1% sodium hypochlorite for 30–60 sec, and plated on either 3% water agar or Difco potato dextrose agar (PDA). Conidia from eight resultant colonies in 1988 were each increased on PDA in petri dishes placed under continuous fluorescent light at 20–23 C for 14 days. Conidia were scraped and washed from the agar, filtered through cheesecloth, and inoculated at a concentration of 100,000 conidia per milliliter onto potted asparagus plants of the cultivar WSU-1. Plants were wounded as previously described (6) before innoculation. Inoculations were done by wetting the foliage of shoots in inoculum: shoots were laid horizontally and moved around on a 60- $\times$ 68-cm plastic sheet that contained 30 ml of a water suspension of conidia with one drop of Tween 20. After inoculation, plants were placed in a plastic mist cham-

ber for 48 hr and then placed in a greenhouse as previously described (6).

Asparagus debris from the previous season was collected from the test field in April 1988, cut into 7-cm pieces, and incubated in petri dishes containing moistened filter paper on a lab bench at 20–23 C for 4–7 days. Conidia of *S. vesicarium* formed on the debris, and four isolates from separate pieces of debris were increased and inoculated at a concentration of 10,000–300,000 conidia per milliliter onto healthy, potted asparagus shoots of the cultivar WSU-1, as described above.

Asparagus debris with pseudothecia from four different ferns were buried separately about 7–10 cm deep in a fine sandy loam soil in an asparagus field near Prosser, WA, on 12 December 1988. Four ferns with pseudothecia were left on the soil surface and secured to a stake with string. On 31 January 1989, debris from another fern with pseudothecia was buried. Before burial, samples of at least 200 pseudothecia per fern were examined with a compound microscope for asci and ascospores. On 17 April 1989, the debris samples were recovered and thoroughly washed in running water. At least 200 pseudothecia per sample were examined for asci and ascospores with a compound microscope. Pseudothecia containing mature ascospores from each sample were scraped with a sterile scalpel from the surface of the washed debris into a 50-ml beaker containing 25 ml of distilled water. Pseudothecia were macerated with a metal rod, and the suspension was filtered through four layers of cheesecloth. About 0.07 ml of each ascospore suspension was streaked on PDA in petri dishes. Potted plants of the cultivar WSU-1 that were wounded as previously described (6) were inoculated with the ascospore suspension. Plants were inoculated by laying shoots horizontally on a plastic sheet, as above, and wetting the foliage in 20 ml of water inoculum (containing 3,000 ascospores per milliliter) and one drop of Tween 20. Inoculated plants were placed in a mist chamber for 24 hr and then moved to a greenhouse with a temperature range of 20–24 C during the day and 17–20 C at night. The number of lesions were recorded 14 days after inoculation. Tissue samples from lesions on inoculated plants from all inoculations were washed, surface disinfested in 1% sodium hypochlorite for 60 sec, and plated on PDA.

Analysis of variance and single degree of freedom contrasts were used to analyze the data from 1987, and Student's *t* test was used for data from 1988 and 1989.

Table 1. Disease severity index of Stemphylium purple spot on asparagus spears in plots during three years when asparagus debris from the previous season's fern was incorporated into the soil or chopped and left on the soil surface[a]

	Fall incorporation	Winter incorporation	Chopped
1987[b]			
5 May	0.2**	0.4**	2.5
12 May	0.1*	0.1*	0.4
14 May	0.3**	0.3**	1.6
19 May	0.1**	0.1**	1.6
2 June	0.2**	0.4**	2.3
12 June	0.2*	0.2*	0.8
1988[c]			
18 April	...	1**	7
21 April	...	7**	27
22 April	...	5**	23
25 April	...	8**	41
2 May	...	1**	7
18 May	...	1**	4
1 June	...	3**	19
3 June	...	4**	16
1989[c]			
24 April	...	3**	17
3 May	...	2**	11

[a] Mean of six replicates in 1987 and 1988 and four replicates in 1989.
[b] Single degree of freedom contrast; * and ** are significantly different from chopped at $P = 0.05$ and 0.01, respectively.
[c] Student's *t* test; ** is significantly different from chopped at $P = 0.01$ level.

Table 2. Disease severity of Stemphylium purple spot on volunteer asparagus seedlings in plots during three years when debris from the previous season's fern was incorporated into the soil or chopped and left on the soil surface[a]

	Fall incorporation	Winter incorporation	Chopped
1987[b]			
21 May	23**	26**	37
2 June	25*	27**	36
1988[c]			
26 May	...	30**	66
3 June	...	7**	23
1989[c]			
26 May	...	19**	47

[a] Mean of six replicates in 1987 and 1988 and four replicates in 1989.
[b] Single degree of freedom contrast; * and ** are significantly different from chopped at $P = 0.05$ and 0.01, respectively.
[c] Student's *t* test; ** is significantly different from chopped at $P = 0.01$ level.

RESULTS

Disease severity on spears (Table 1) and volunteer seedlings (Table 2) was less on all sampling dates ($P = 0.01$ and 0.05) when asparagus debris from the previous season was incorporated into the soil either in late fall or winter than when debris was left on the soil surface. No difference was found in disease severity ($P = 0.05$) between the fall and winter incorporation dates.

The yield of harvested spears did not vary ($P = 0.05$) among the three treatments in 1987. Mean yields of the treatments per harvest day were 2.327, 2.341, and 2.318 kg/182-m row for the fall incorporation, winter incorporation, and chopped treatments, respectively.

The mean weights of debris were 0.26, 0.31, and 1.91 kg/9 m^2 for the fall incorporation, winter incorporation, and chopped treatments, respectively, in 1987. They were 0.34 and 2.04 kg/9 m^2 for the winter incorporation and chopped treatments, respectively, in 1989. Differences in the amount of debris per unit area between the soil incorporation and chopped treatments were statistically significant each year ($P = 0.01$).

S. vesicarium was isolated from volunteer seedlings collected in 1988 and 1989 and produced lesions typical of those on plants in the field when inoculated onto healthy plants. Conidia of *S. vesicarium* from overwintered asparagus debris also produced lesions typical of purple spot. *S. vesicarium* was reisolated from lesions on inoculated plants.

Before burial, pseudothecia on asparagus debris contained cytoplasm but not asci and ascospores. On 17 April 1989, nearly 100% of the pseudothecia examined from the four buried samples contained asci and mature ascospores. From the sample on the soil surface, 20–95% of the pseudothecia contained asci, ranging from immature asci without ascospores to asci with either immature or mature ascospores. This variation was associated with variation in moisture content within the sample from the soil surface. Ascospores were more mature and abundant on portions of the fern debris that were directly on the soil sur-

face, where the moisture content of the debris was higher, than on debris material held above the soil surface by underlying debris. Ascospores from all samples, either buried or left on the surface, germinated on PDA and produced lesions on inoculated plants typical of the disease in the field. *S. vesicarium* was reisolated from all inoculated plants.

DISCUSSION

Several foliar pathogens of asparagus overwinter on asparagus debris. Among these are *Cercospora asparagi* Sacc. (2), *Puccinia asparagi* DC. (7), and *Pleospora herbarum* (1,3,5). Burial of crop debris frequently favors decomposition of the substrata and activity by organisms antagonistic to the pathogen (2). Sanitation, such as crop burial, is particularly important for disease management when pathogens overwinter in the debris and when crops are grown in the same field for several years (10). Asparagus is a perennial plant and, in Washington State, asparagus shoots are generally harvested from early April to mid-June, after which shoots are allowed to develop fern growth.

In this study, burial of the previous year's fern growth reduced severity of Stemphylium purple spot on spears during the harvest season. In south-central Washington's semiarid environment, burial of asparagus debris would be an effective management practice for purple spot on spears. Falloon had previously observed less disease on spears in fields where the previous year's fern was removed than in fields where it was left on the soil surface (4). Burial in late fall or late winter was equally effective in reducing disease. Less soil erosion from wind would be expected with the late-winter than with the late-fall incorporation of debris.

Pseudothecia of *P. herbarum* did not decompose during the 10–14 wk that they were buried in soil. Asci and ascospores developed on fern debris beneath the soil surface, and ascospores from buried debris were capable of infecting asparagus. Burying asparagus debris physically prevented ascospores from becoming airborne and reaching an infection court. Asparagus substrata and pseudothecia would have decomposed if buried longer, and perhaps ascospores would not have been viable after burial for 14 wk if they had been in a different soil environment.

Pseudothecia of *P. herbarum* on fern debris are an important source of inoculum for purple spot of asparagus spears (5). Conidia of *S. vesicarium* formed in the spring on the previous year's asparagus fern and caused infection. Thus conidia, as well as ascospores, served as a source of primary inoculum.

Volunteer asparagus seedlings that become infected during the harvest season may be important as a substrate for the increase of inoculum and as a bridge to carry inoculum from the harvest period, when spears are consistently removed, to the period when they are allowed to fern.

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