Cranberry Cottonball: Dispersal Periods of Primary and Secondary Inocula of *Monilinia oxycocci*, Host Susceptibility, and Disease Development

P. G. Sanderson and S. N. Jeffers

Department of Plant Pathology, University of Wisconsin, Madison 53706. Present address of first author: Mid-Columbia Agricultural Research and Extension Center, Oregon State University, Hood River, OR 97031. This research was supported by Hatch Project 3046, the Wisconsin Cranberry Board, Inc., and Ocean Spray Cranberries, Inc. We thank Wisconsin Moss Co. for providing the experimental site, M. K. Clayton for statistical advice, and S. A. Vicen for figure preparation.
Accepted for publication 21 October 1991 (submitted for electronic processing).

ABSTRACT

Sanderson, P. G., and Jeffers, S. N. 1992. Cranberry cottonball: Dispersal periods of primary and secondary inocula of *Monilinia oxycocci*, host susceptibility, and disease development. Phytopathology 82:384-392.

In 1987–1989, airborne ascospores and conidia of *Monilinia oxycocci*, the causal agent of cranberry cottonball, were trapped, and selected environmental parameters were monitored in a heavily infested commercial cranberry bed. In each year, each type of spore occurred in a single dispersal period that lasted 25–35 days. Single, distinct spore showers that lasted 10–14 days occurred in each dispersal period. Ascospore dispersal periods began just before or at budbreak and continued until 0–7 days before bloom. Conidium showers began 32–33 days after initiation of ascospore showers. The presence of ascospores and conidia in the air exhibited a diurnal periodicity. Ascospores were trapped predominantly between 1000 and 2100 h, with a peak occurring between 1600 and 1800 h. Conidium concentrations peaked between 1000 and 1800 h. Temperature of the duff and upper canopy and relative humidity correlated best with the daily presence of airborne ascospores during the peak period of dispersal. These three variables and wind speed correlated best with the daily presence of airborne conidia during the peak period of dispersal. Percentage of bloom and of shoots with symptoms of primary infection were highly correlated with conidium shower intensity in both 1988 and 1989. Trap plants were placed in the field during the periods of ascospore dispersal in 1989 and 1990 and conidium dispersal in 1989. Shoots that were beginning to expand and had several nodes visible were most susceptible to infection by ascospores. Shoots inoculated with conidia did not develop tip blight symptoms. Ascospores were shown for the first time to infect flowers and cause typical cottonball fruit rot. In the field, fruit rot incidence did not change over time as berries developed, and airborne inocula were not present after bloom. This evidence supports previous observations that fruit rot results only from infection of flowers during bloom.

Additional keywords: epidemiology, *Vaccinium macrocarpon*.

Cranberry cottonball, which is caused by *Monilinia oxycocci* (Woronin) Honey, is the most important disease affecting cranberries (*Vaccinium macrocarpon* Aiton) during the growing season in Wisconsin. In consequence, cottonball is one of the main reasons for fungicide use in cranberry production. Over the past 10 yr, disease management has relied on applications of triforine, which is the only fungicide currently registered for cottonball control (15). However, disease control has not been consistent or reliable, and some growers have incurred substantial economic losses.

Pathogenesis by *M. oxycocci* on cranberry is similar to that by other species of *Monilinia* that attack ericaceous hosts (5,10,12,16,17) and is characterized by two distinct phases (4,18,20,22,23,25). Primary infection by ascospores results in a blight of shoots and flowers known as tip blight, and secondary infection of flowers by conidia results in fruit rot. Apothecia of *M. oxycocci* develop from overwintered sclerotia around budbreak and discharge ascospores (4,18), which are reported to infect young developing shoots with little or no cuticle (25). Around bloom, conidia are produced on ectostromata that develop on infected shoots and are dispersed to blossoms where they infect developing fruit (4,7,18,20,22,23,25).

Few studies have been done on cottonball, and most of the available epidemiological information is based on qualitative field observations (4,23,25). Consequently, little is known about dispersal of inoculum of *M. oxycocci* or the conditions conducive to infection. Bain (4) described cottonball in the field and concluded that fruit infection was initiated by conidia during bloom. Furthermore, he concluded that disease incidence was greatest when there were prolonged wet periods during the time apothecia were mature, when rainy periods occurred during bloom, and when a plentiful supply of mummified berries was present (4). In his original description of *M. oxycocci*, Woronin (25) described infection of stigmas of the blossoms of *V. oxycoccus* L. after inoculation with conidia but, unfortunately, did not report the conditions under which infection took place.

The objective of this research was to identify periods when infection of cranberry by *M. oxycocci* is most likely to occur (i.e., when the host is susceptible, the pathogen inoculum is present, and the environmental conditions are conducive for infection). Appropriate disease management practices based on this information can then be developed. Preliminary reports have been published (18–20).

MATERIALS AND METHODS

Research was conducted during the 1987, 1988, and 1989 growing seasons in the southern portion of a 1.2-ha, 20-yr-old commercial cranberry bed (cultivar Bain McFarlin) located in Wood Co., WI. Cottonball incidence had been consistently high in this field during the 5 yr preceding this study. Fungicides were not applied to the portion of the bed (80 × 42.5 m) in which the research was conducted; other standard cultural practices were conducted routinely on the entire bed by the grower.

Quantifying airborne inocula. The presence of airborne inocula was monitored with a Burkard 7-day recording volumetric spore trap (Burkard Scientific Sales Ltd., Rickmansworth, Hertfordshire, England) that was placed in the center of the study area. Spores were trapped on Melinex tape (Burkard Scientific Sales Ltd.) prepared according to the manufacturer's specifications (8). The spore trap was fixed to a stationary wooden platform in the bed so that the trapping orifice was about 45 cm above the ground and about 30 cm above the canopy. In 1987 and 1988, the trap was placed in the field immediately after the last spring flood waters were removed (17 and 29 April,

respectively) and operated continuously until 29 and 28 July, respectively (10–12 days after bloom had ended). To determine if any ascospores were coming from outside the study area before removal of the spring flood, the spore trap was placed in the field on 12 April in 1989 and mounted on a platform just above the water surface (i.e., raised about 1 m above the ground). After the bed was drained (16 May), the spore trap was mounted on the low-lying platform as described above and operated continuously until 2 August (approximately 12 days after bloom ended).

Spore trap tapes that had been exposed for 7 days were cut into 48-mm strips, each corresponding to a 24-h period, and mounted on glass microscope slides in a mixture of Gelvatol and lactic acid (8). Cover glasses were mounted on the tapes with the same mixture supplemented with 0.05% cotton blue in lactophenol (~15:1, v/v). Tapes were examined at hourly intervals of deposition (every 2 mm) in traverses perpendicular to the direction of movement. All ascospores and conidia in 400 m wide areas (1.2-h periods of deposition [8]) were counted with a bright field microscope (×320). Ascospores discharged from apothecia onto membrane filters (21) were mounted and stained in the same manner and served as a reference to confirm the identification of ascospores on spore trap tapes. Conidia, on the other hand, were identified easily by their characteristic lemon-shaped appearance (25).

Spore trap data from each year were compared to determine whether consistent seasonal or diurnal patterns of dispersal occurred. Seasonal patterns of dispersal were examined by plotting daily totals of spores caught. Diurnal periodicity was evaluated by plotting the mean proportion of spores caught each hour for all 24-h periods (2400 to 2400 h) during the ascospore or conidium showers in each year.

Monitoring environmental parameters. Environmental parameters were recorded with a 21× micrologger (Campbell Scientific, Inc., Logan, UT). In 1987, temperatures of the soil (at a depth of about 4 cm), duff or leaf litter layer, upper plant canopy, and ambient air were measured with thermistors; wind speed at 0.5 m above the soil surface was measured with a three-cup anemometer; rain and irrigation, which were not distinguished, were measured with a tipping-bucket rain-gauge; leaf wetness was measured with resistance grid sensors that were coated with a green-tinted latex paint (1). Temperature and leaf wetness sensors were replicated. In 1988, some of the thermistors were replaced or supplemented with Cu-Cn thermocouples, and relative humidity was sensed with a sulfonated polystyrene resistance grid sensor. A hygrothermograph also was operated to verify and backup the micrologger. The same variables were measured in 1989, but all temperature measurements were made with thermocouples.

Temperature, relative humidity, and wind speed were sensed every 10 s; data were summarized as hourly and daily averages and as daily maxima and minima. Rain-irrigation measurements were summarized as hourly and daily totals. Leaf wetness sensors were sampled every 15 min, and readings were scaled from 0 to 100 such that 0 was completely dry and 100 was completely wet. A value of 70 was selected as the lower limit of wet periods based on field observations of actual leaf wetness. Therefore, leaves were recorded as wet if readings of 70 or above occurred in at least one of the sensors. Each year, all sensors were calibrated before placement in the field and again after removal at harvest. The hygrothermograph and relative humidity sensor were calibrated weekly in the field with a sling psychrometer.

Cumulative degree-days (0001 to 2400 h) were calculated for duff and canopy temperatures (C) as follows: Σ[maximum daily temperature + minimum daily temperature]/2. Values were used to compare seasonal temperature trends during the spore dispersal periods each year.

Host phenology and disease progress. Observations of cranberry phenology (budbreak, shoot development, and bloom) and disease incidence (numbers of symptomatic shoots and those with secondary sporulation) were recorded each year. In 1987, the site was visited weekly. The initiations of budbreak and bloom were recorded, and the percentage of shoots with tip blight in 10 randomly selected areas (0.25 m^2) was determined 17 June (day of the year 168). In 1988 and 1989, 10 plots (0.5 × 0.5 m) were established in randomly selected sites in the bed before budbreak. Host phenology and primary disease incidence observations were made every 3–4 days through day 197 in 1988 and every 2–3 days through day 195 in 1989. Corners of plots were marked with wooden stakes to facilitate repeated and consistent placement of a wooden frame with a fixed grid, which was used for all enumerations. Percentage of bloom was the percentage of flowering upright shoots with open blossoms in two opposite corner sections of the grid (313 cm^2 total). The percentage of shoots that became symptomatic or on which ectostromata were produced was calculated based on the total number of actively growing shoots in each plot.

The proportion of shoots in different stages of development in the center section of the grid (625 cm^2) was recorded. Vegetative budbreak occurred when greater than 50% of shoots had begun to elongate. Stages of early season shoot development were divided into three classes based on shoot length and corresponding morphological changes. Shoots in class 1 had just begun to elongate, were 1–10 mm long, and had leaves that were in a tight cluster at the shoot tip. Shoots in class 2 were 11–20 mm long, still had leaves tightly clustered at the shoot tip, but had several nodes visible at the proximal end of the stem. Shoots in class 3 were greater than 21 mm long, and leaves in the apical cluster had begun to open and expand.

Isolations from diseased shoots were made periodically in 1987 and 1988 to determine whether *M. oxycocci* was able to persist after the conidium dispersal period. Shoots were surface-disinfested by first wetting in 95% ethanol and then soaking for 30 s in 0.5% NaOCl. Shoots were blotted dry, and stem and leaf sections were placed on potato-dextrose agar (PDA; Difco Laboratories, Detroit, MI) in 9-cm-diameter petri dishes. Dishes were placed at room temperature (22–25 C) and examined daily for up to 10 days.

In 1988 and 1989, samples of fruit were collected weekly from the end of bloom until harvest to determine if the incidence of diseased fruit increased after bloom. Berries were collected 12 times beginning 12 July (day 194) in 1988 and 10 times beginning 18 July (day 199) in 1989. All of the fruit in 10 areas (625 cm^2) were collected. The areas sampled were randomly selected; those for the final collection each year were from the small plots used to monitor host phenology and primary disease incidence. Berries less than 4 mm in diameter were discarded because they could not be assessed accurately. Fruit were cut in half transversely, and locules were examined visually for characteristic mycelia or stromata of *M. oxycocci* (4,18,23,25).

Host susceptibility. *Infection by ascospores.* Trap plants with shoots in each of the three morphology classes described above were used to determine if cranberry plants with shoots in different stages of development varied in susceptibility to infection by either ascospores or conidia of *M. oxycocci.* Individual plants were grown from hardwood cuttings taken from dormant, 1-yr-old upright shoots (cv. Searles) with terminal buds. Cuttings were grown individually in a peat/sand mixture (3:1, v/v) in 2.5 × 16 cm planting cones (Ray Leach "Cone-tainer" Nursery, Canby, OR) in a greenhouse at 20–25 C with a 16-h photoperiod. Plants for use as trap plants were first hardened outside for at least 3 days just before placing in the field.

In 1989, 100 plants in each morphology class were placed in the infested field during the period of ascospore dispersal and exposed to natural inoculum for 3 days. Plants were then moved to another commercial cranberry field 5–10 km away, which had no history of cottonball, for symptom development. At the same time, 100 uninculated plants in each class also were placed in the uninfested field to serve as controls. Disease development was assessed as soon as symptoms became apparent. Symptomatic plants were removed from the bed before ectostromata and conidia developed.

Five single-plant replicates of each class were placed in each of 20 randomly located blocks in the infested field. Uninoculated

control plants were placed in a solid block in the uninfested field. In all instances, planting cones with trap plants were inserted into the cranberry sod so that the trap plant shoots were at the same level as the surrounding canopy. When inoculated plants were brought into the uninfested field, they were placed in a contiguous block with control plants. The experiment was conducted six times beginning on days 143, 145, 149, 152, 155, and 161. Numbers of diseased plants from each treatment were compared with chi-square analysis.

A similar experiment was conducted four times in 1990 on days 138, 144, 147, and 151. Ten-plant replicates of each morphology class were placed in each of 10 randomly located blocks and exposed to inoculum for 3 days in the infested field. Equal numbers of plants were blocked and kept outside in Madison, WI, as controls. Exposed plants were brought back to Madison, placed in blocks with their respective controls, and grown until symptoms developed. Disease incidence in each replicate was calculated, and data were analyzed by one-way analysis of variance.

In addition, plants with open blossoms were placed in the infested field during the ascospore shower to determine whether ascospores could cause cottonball fruit rot. Equal numbers of flowering plants were placed outside in Madison as unexposed controls. Blossoms were hand-pollinated to ensure fruit set. Plants were kept in the infested field for 14 days and then returned to Madison for incubation with control plants while fruit developed. The experiment was conducted twice, the first time with 26 plants and the second time with 30 plants.

Open blossoms of plants growing in a greenhouse also were inoculated artificially with ascospores. Plants were propagated and grown in planting cones as described above. Ascospores, which had been collected previously and stored dry on membrane filters (21), were transferred with a fine-bristled brush directly to the stigmas of blossoms on 25 plants immediately after the blossoms had been hand-pollinated. Pollinated but uninoculated blossoms on 25 other plants served as controls. Senescent blossoms were removed at the time of inoculation. Locules of berries were examined visually for mycelia or stromata of *M. oxycocci* 25 days after inoculation, and the proportion of berries that were diseased was calculated.

Infection by conidia. To determine whether shoots were susceptible to infection by conidia, 20 plants with shoots in each morphology class and 20 plants with open blossoms were placed in the infested field, during the period of conidium dispersal but after that of ascospore dispersal, in 1988. The plants were randomly assigned to blocks in the field with one plant from each class per block. Plants were similarly placed outside in Madison as uninoculated controls. After 2 days of exposure to conidia, plants were brought back to Madison and placed with the control plants. Plants were observed daily for tip blight symptoms. After 45 days, berries that developed were cut open and examined for mycelia or stromata. The experiment was conducted three times, beginning days 180, 182, and 184.

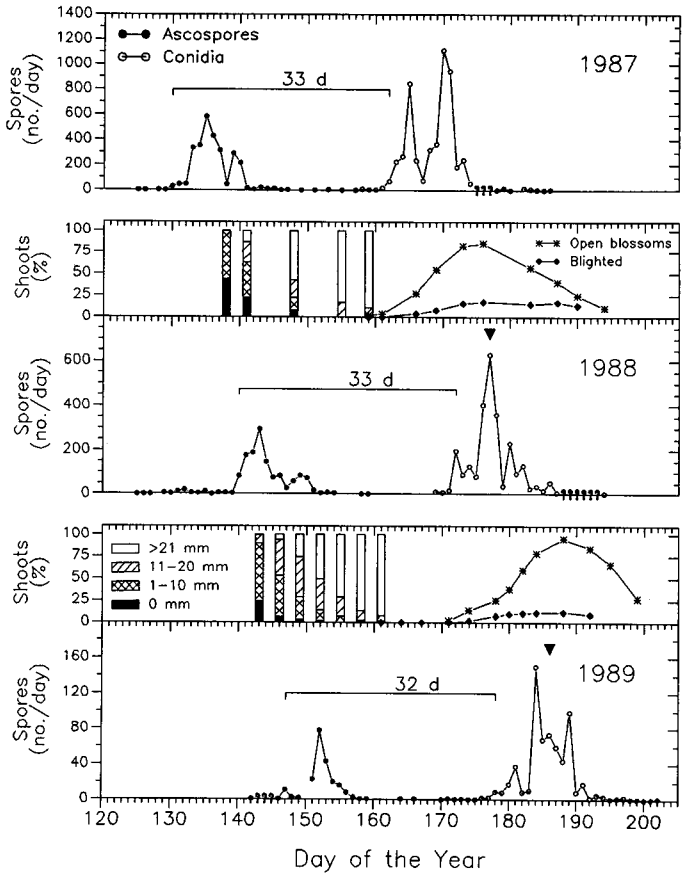

Fig. 1. Total numbers of ascospores and conidia of *Monilinia oxycocci* detected per day with a Burkard 7-day recording volumetric spore trap in a commercial cranberry field (cultivar Bain McFarlin) in 1987–1989, and the relationship of spore numbers to host phenology and disease development in 1988–1989. Host phenology is shown as the percentage of shoots in 0.25-m^2 areas that were in four length categories when ascospores were present and that had open blossoms when conidia were present. Disease development is the percentage of shoots that became blighted in 0.25-m^2 areas. Only spore numbers greater than zero are plotted; question marks in spore showers are when the spore trap malfunctioned. Bracketed intervals are the number of days (d) between initiations of ascospore and conidium showers. Triangles above conidium showers in 1988 and 1989 mark the day on which half of the cumulative total number of spores had been caught. In 1989, actual numbers of conidia detected per day were 10 times the numbers shown.

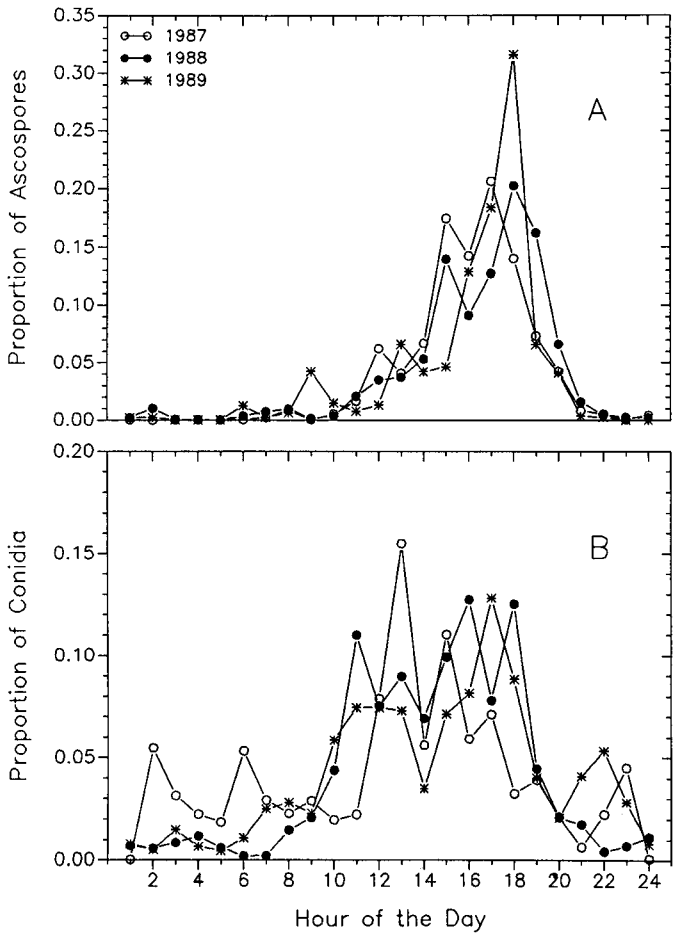

Fig. 2. Diurnal periodicities of **A**, ascospores and **B**, conidia of *Monilinia oxycocci* in a commercial cranberry field (cultivar Bain McFarlin) in each of 3 yr. Data are mean hourly proportions of spores caught each day with a volumetric spore trap during the period of peak spore abundance.

Data analysis. All analyses were conducted with MINITAB statistical software, release 7.2 (Minitab, Inc., State College, PA). Relationships among hourly summaries of environmental variables and spore numbers were examined by regression and correlation analyses. Numbers of spores and proportional data were transformed to natural logarithms and arcsine-square root values, respectively, for analyses in which residual plots showed it necessary to stabilize variances (24).

The apparent relationship between peak bloom and conidium showers that was observed in 1988 and 1989 was examined by correlation. The number of conidia that accumulated between each field observation was used as an expression of conidium shower intensity. Correlations were determined for the natural logarithms of these values and either percentage of bloom or percentage of tip blight at each field observation in each year. The relationship between bloom and symptom development also was examined by correlating percentage of bloom with percentage of shoots blighted at each field observation.

RESULTS

Periodicity of inocula. *Seasonal periodicity.* Ascospores were collected in a single dispersal period each year (Fig. 1). In 1987 and 1988 (a leap year), ascospores first were detected in low numbers on 5 and 4 May, respectively (both day 125). They were trapped on 27 days of a 31-day dispersal period in 1987, and on 31 days of a 35-day dispersal period in 1988. In 1989, ascospores were trapped on 16 days of a 25-day dispersal period that began day 142, 6 days after the spring flood was removed; spores were not collected on 3 days due to a spore trap malfunction. Single, distinct showers within ascospore dispersal periods occurred in each year that lasted 11, 12, and 10 days and began on days 130, 140, and 147 in 1987, 1988, and 1989, respectively. While the numbers of ascospores caught per day decreased (a maximum daily catch of 585, 295, and 78 occurred in 1987, 1988, and 1989, respectively) and spore showers occurred later in each consecutive year, similar seasonal patterns of spore

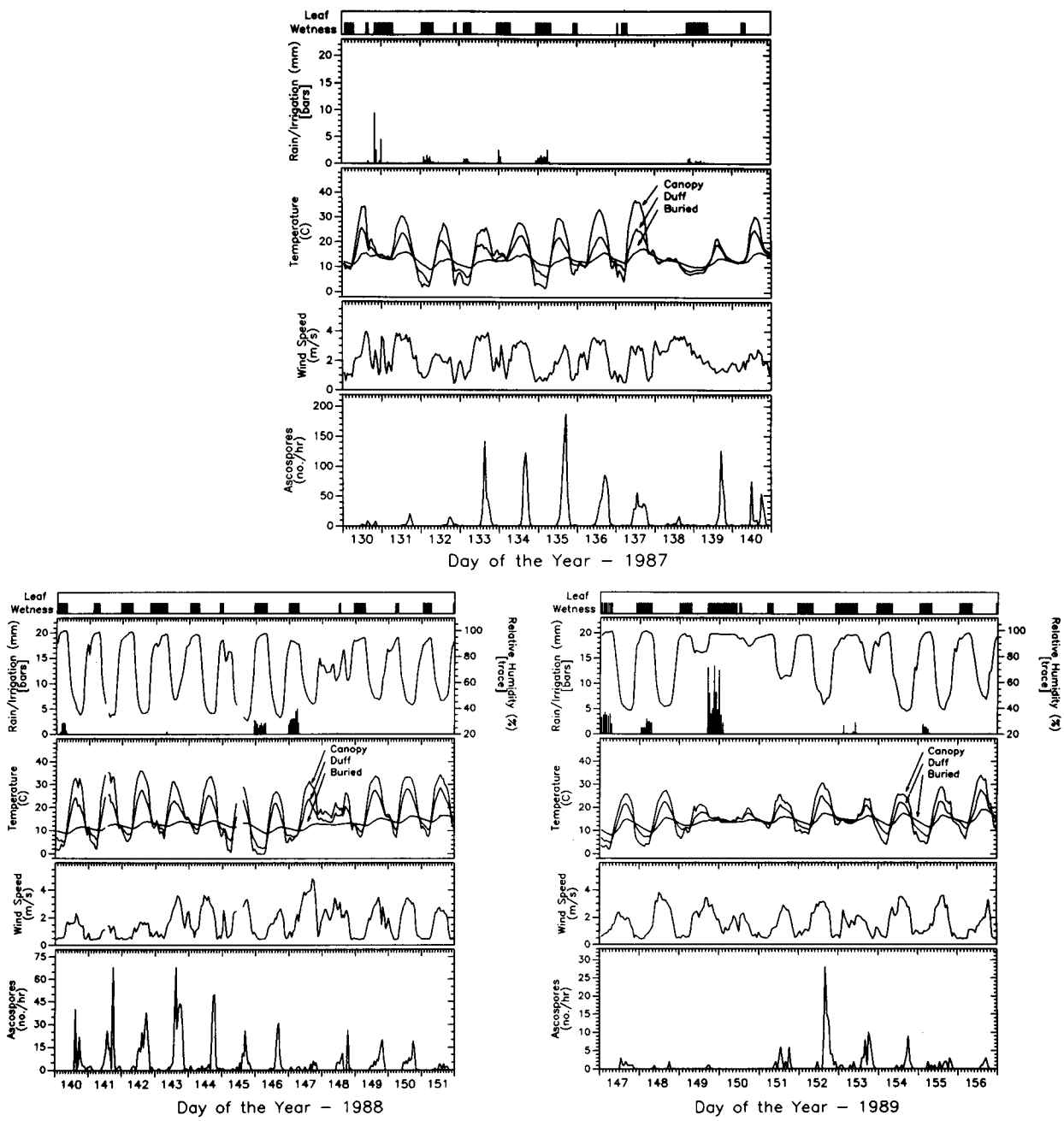

Fig. 3. Numbers of ascospores of *Monilinia oxycocci* collected per hour and hourly summaries of environmental variables during the peak periods of inoculum dispersal in a commercial cranberry field (cultivar Bain McFarlin) in 1987–1989. Relative humidity was not recorded in 1987. Days of the year are divided into 2-h increments.

dispersal were observed (Fig. 1).

Conidia also were collected in a single dispersal period each year (Fig. 1). In 1987, conidia were collected on 26 days of a 30-day dispersal period that began day 157; the spore trap malfunctioned on 3 of the 4 days (days 175–177) that conidia were not caught. In 1988, conidia were collected on 19 consecutive days of what appeared to be a 26-day dispersal period that began day 169; again, the spore trap malfunctioned during the 6-day period (days 188–193) immediately preceding the last day spores were caught. In 1989, conidia were caught for 33 consecutive days beginning day 170. Single showers of conidia began on days 162, 172, and 178 in 1987, 1988, and 1989, respectively. Showers were 13, 15, and 14 days in duration during the respective years. The maximum number of conidia caught per day in each year was 1,112, 630, and 1,498 in 1987, 1988, and 1989, respectively.

The time period from the start of the ascospore shower to the start of the conidium shower was consistent from year to year; 33 days in 1987 and 1988, and 32 days in 1989. Little difference in the local climate of the cranberry field was observed among years. Accumulated degree-days for canopy temperatures from days 137–196 were 1,252.8, 1,253.8, and 1,218.5 in 1987, 1988, and 1989, respectively. Degree-days for duff temperatures during the respective time periods were 1,123.3, 1,085.0, and 1,099.1.

Diurnal periodicity. The pattern of mean hourly proportions of ascospores caught over a 24-h period showed a distinct diurnal periodicity (Fig. 2A). Most spores were collected each year between 1100 and 2100 h. In 1987, the maximum spore catch occurred at 1700 h, whereas in 1988 and 1989, it occurred at 1800 h. Patterns of mean hourly proportions of conidia caught over a 24-h period also showed a diurnal periodicity (Fig. 2B), although it was not as pronounced as that for ascospores. Most conidia were caught during daylight hours, and the greatest numbers of spores were caught between 1100 and 1800 h.

A comparison of hourly summaries of environmental data during the periods when ascospore or conidium showers occurred (Figs. 3,4) also showed no striking difference among years in the local climate of the cranberry field. In all instances, both

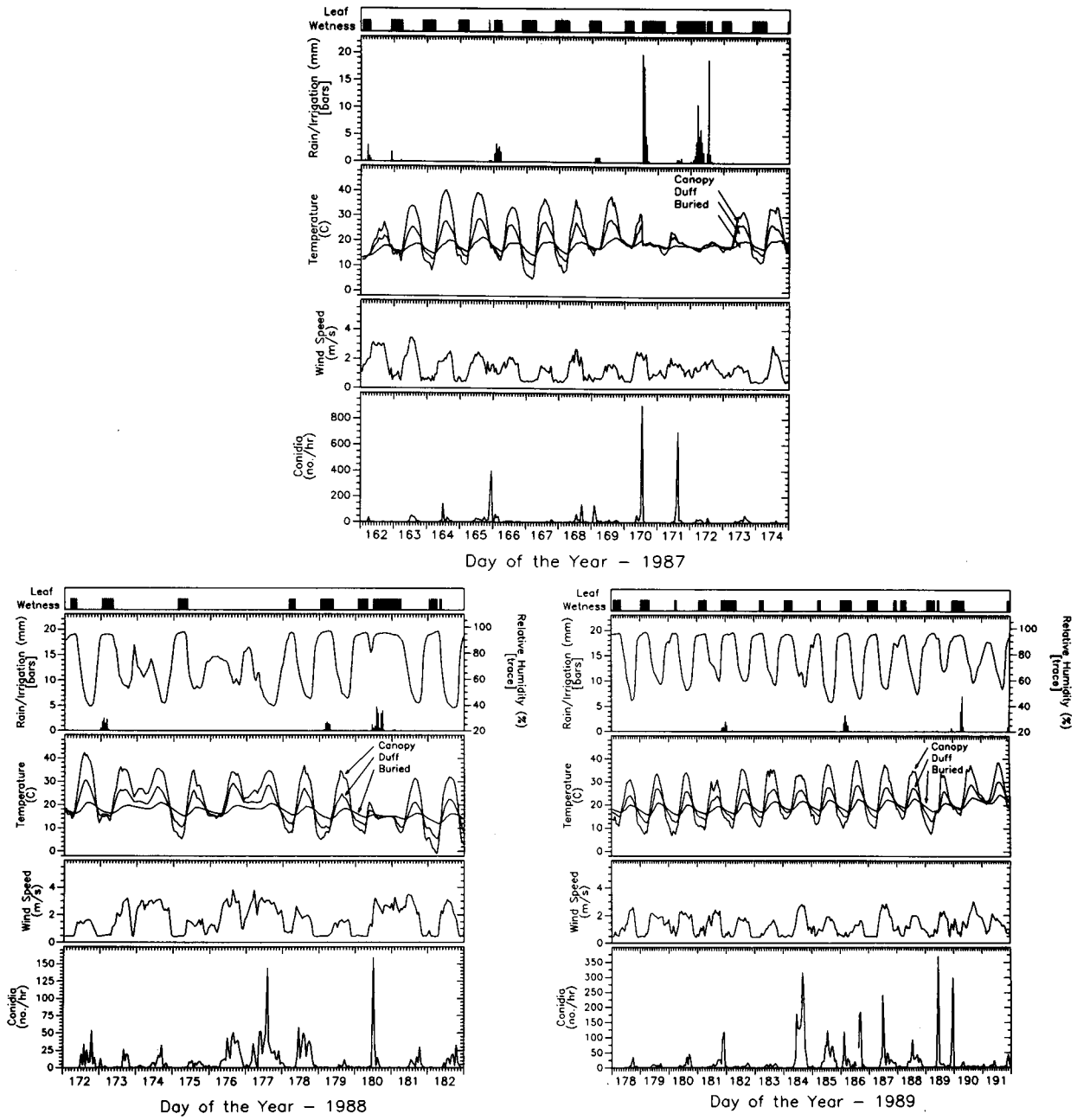

Fig. 4. Numbers of conidia of *Monilinia oxycocci* collected per hour and hourly summaries of environmental variables during the peak periods of inoculum dispersal in a commercial cranberry field (cultivar Bain McFarlin) in 1987–1989. Relative humidity was not recorded in 1987. Days of the year are divided into 2-h increments.

TABLE 1. Correlation coefficients (r) between hourly summaries of environmental variables and the natural logarithm of numbers of ascospores or conidia of *Monilinia oxycocci* that were caught each hour during periods of spore showers in a commercial cranberry field (cultivar Bain McFarlin) over a 3-yr period

Year	Number of hourly samples[a]	Temperature		Wind speed	Rain and irrigation	Leaf wetness	Relative humidity
		Duff	Canopy				
Ascospores							
1987	264	0.54	0.56	0.34	−0.08	−0.35	...[b]
1988	288	0.55	0.57	0.41	−0.15	−0.37	−0.63
1989	240	0.36	0.36	0.32	−0.09	−0.29	−0.36
Conidia							
1987	312	0.50	0.48	0.43	0.29	−0.31	...[b]
1988	360	0.60	0.62	0.58	−0.02	−0.37	−0.58
1989	336	0.50	0.56	0.49	0.12	−0.25	−0.51

[a]Spores were caught with a Burkard 7-day recording volumetric spore trap. Periods of ascospore dispersal, which were not always continuous, were from day of the year 130–140 in 1987, 140–151 in 1988, and 147–157 in 1989; periods of conidium dispersal were from day of the year 162–174 in 1987, 172–186 in 1988, and 178–191 in 1989.
[b]Relative humidity was not recorded in 1987.

TABLE 2. Difference between natural logarithms of mean numbers of ascospores and conidia of *Monilinia oxycocci* caught in a commercial cranberry field (cultivar Bain McFarlin) during hours of leaf wetness or rain and irrigation and during dry hours in each of 3 yr

Year	Leaf wetness[a]			Rain and irrigation		
	Wet (n^b)	Dry (n^b)	t^c (P)	Wet (n)	Dry (n)	t (P)
Ascospores						
1987	0.21	1.32	8.51	0.13	1.13	7.94
	(79)	(185)	(<0.001)	(37)	(227)	(<0.001)
1988	0.19	1.11	9.11	0.2	0.9	5.84
	(85)	(202)	(<0.001)	(24)	(264)	(<0.001)
1989	0.11	0.47	5.16	0.06	0.36	5.46
	(101)	(139)	(<0.001)	(36)	(204)	(<0.001)
Conidia						
1987	0.84	1.74	5.74	2.55	1.16	−4.70
	(140)	(172)	(<0.001)	(40)	(272)	(<0.001)
1988	0.41	1.43	9.06	1.00	1.16	0.69
	(99)	(261)	(<0.001)	(29)	(331)	(0.50)
1989	1.14	1.92	4.73	2.68	1.62	−2.73
	(101)	(234)	(<0.001)	(19)	(316)	(0.013)

[a]Hours were considered wet if either of two resistance grid sensors connected to a micrologger registered values ≥ 70 within an hour (see text).
[b]n = Number of hourly samples.
[c]Data were analyzed by two-sample Student's t tests; P = significance of t.

types of spores were trapped each day when percentage of relative humidity decreased, temperature increased, and wind speed increased (Figs. 3,4). The effects of heavy and light rains on conidium dispersal were readily apparent on days 170 and 171, respectively, in 1987. On day 170, daily spore dispersal was stopped abruptly at the onset of a heavy rain, whereas on day 171, the greatest number of spores was caught during a light rain shower (Fig. 4).

Of the environmental variables measured, duff and canopy temperatures and percentage of relative humidity correlated best with hourly ascospore catches during the peak period of dispersal in each year (Table 1). Leaf wetness and wind speed had lower correlation values, and rain-irrigation correlated poorly with numbers of ascospores caught per hour over the 3-yr period. Similar relationships were observed between hourly summaries of environmental variables and numbers of conidia caught per hour (Table 1), except that correlations with wind speed were somewhat higher for conidia than for ascospores. The significance of correlation coefficients (Table 1) and regression analyses of hourly summaries of environmental variables and spore numbers could not be determined accurately because of autocorrelation among hourly summaries of the same variable and intercorrelation among different variables.

Numbers of ascospores and conidia caught during periods of leaf wetness and rain-irrigation events were compared to those caught during dry periods with Student's t tests. Significantly more ascospores were caught during dry periods than during wet periods in each year (Table 2). A similar relationship was found between numbers of conidia and periods of leaf wetness. In contrast, significantly more conidia were trapped during periods of rain-irrigation than during dry periods in 1987 and 1989 (Table 2).

Host phenology and disease progress. Ascospore dispersal coincided with early cranberry shoot development. In 1987, buds began to break about day 132, 7 days after the start of ascospore dispersal but only 2 days after the start of the ascospore shower. In 1988, budbreak had occurred by day 138, 14 days after the start of ascospore dispersal but only 2 days before the spore shower began (Fig. 1). The median number of ascospores (i.e., half of the cumulative total of spores caught during the shower) occurred on day 143, at which time (by interpolation) the majority of shoots probably were 1–3 cm long (Fig. 1). In 1989, budbreak occurred on day 141, 1 day before the start of ascospore dispersal (Fig. 1). The median number of ascospores occurred on day 152, at which time about 90% of the shoots were in morphology classes 2 and 3 (>11 mm long).

Although the onset of conidium dispersal differed relative to the time of first bloom, the median number of conidia caught coincided with peak bloom in both 1988 and 1989. In 1988, bloom began about day 158, and peak bloom was about day 176 (Fig. 1). Conidium dispersal began approximately 11 days after the start of bloom with the onset of the conidium shower occurring at approximately 75% bloom. However, the median number of conidia caught occurred on day 177, 1 day after peak bloom. The conidium shower began abruptly, and the median number of spores had accumulated after only 5 days. In contrast, in 1989, the conidium shower began more gradually, and the median number of spores had accumulated 10 days later (Fig. 1). In 1989, bloom and the conidium shower began at approximately the same time (about day 170), with peak bloom occurring during day 188. The conidium shower began about 25% bloom, and the median number of conidia was reached on day 186. Initiations of bloom and the conidium shower coincided in 1987.

In 1987, tip blight symptoms were first observed on day 154; conidia were observed on 5–10% of these shoots by day 161. On day 168, 21 ± 8% (mean ± standard deviation) of shoots were symptomatic. Conidia produced on these shoots resulted in 51 ± 11% diseased fruit. Tip blight symptoms in small plots first were observed on day 159 in 1988 and on day 161 in 1989 (Fig. 1). In general, conidia first were observed on blighted shoots about 1 wk after initial symptoms. In 1988, numbers of symptomatic shoots increased for 17 days, at which time (day 176) 18 ± 11% of shoots were blighted; whereas, numbers increased for 28 days in 1989, at which time (day 188) 11 ± 10% of shoots were blighted.

In 1988, conidia eventually were produced on 76 ± 8% of all blighted shoots, and 37 ± 10% of fruit (mean from all sampling dates combined) were diseased (Fig. 5). In 1989, conidia eventually were produced on 88 ± 7% of blighted shoots, and 33 ± 13% of fruit were diseased (Fig. 5). There were no significant differences in disease incidence among sample dates in either year ($P = 0.556$ in 1988 and $P = 0.389$ in 1989). In addition, diseased shoots desiccated after conidia on ectostromata had been shed and bloom was over; *M. oxycocci* was not recovered from those shoots.

In both 1988 and 1989, conidium shower intensity and percentage of bloom were highly correlated ($r = 0.868$ [$n = 9$], and $r = 0.919$ [$n = 12$], respectively). Similarly, conidium shower intensity and the proportion of shoots that were symptomatic also were highly correlated in each year ($r = 0.952$ [$n = 9$] in 1988, and $r = 0.960$ [$n = 12$] in 1989). Percentage of bloom and of blight also were highly correlated ($r = 0.849$ [$n = 11$], and $r = 0.919$ [$n = 12$] in 1988 and 1989, respectively).

Host susceptibility. In 1989, low numbers of trap plants that had been exposed to inoculum in the infested field at each of the planting dates became diseased (Table 3). However, some control plants also became diseased because the supposedly uninfested field was infested with *M. oxycocci*. Because disease was present in the control plants and because they were not blocked, direct comparisons of the numbers of plants in each morphology class that became diseased could not be made. Consequently, only comparisons of numbers of plants that became diseased between locations within each class could be made. Significant differences within classes occurred on days 149, 152, and 155 (Table 3). Although differences between morphology classes could not be determined, on day 152, considerably more plants with shoots in classes 2 and 3 became diseased than those with shoots in class 1. The 3-day period in which these plants were exposed to inoculum in 1989 coincided with the apex of the ascospore shower (Fig. 1).

Disease incidence was greater on shoots of plants kept in the infested field in 1990 (Table 4) than in 1989 (Table 3). Tip blight occurred in three of the four trials. The relative proportions of diseased shoots in each morphology class differed among trials; however, no significant differences were observed among different morphology classes (Table 4).

None of the shoots in any morphology class that were exposed to conidia in 1989 became symptomatic. However, averaged over the three trials, 36 ± 17% of berries became diseased. None of the 16 fruits produced by plants with open blossoms that initially were exposed to ascospores on day 147 in 1990 became diseased; however, about 5% of berries produced on plants initially exposed on day 153 developed typical signs of cottonball fruit infection. In addition, 24% of berries that developed from blossoms inoculated with dry ascospores in the greenhouse became diseased.

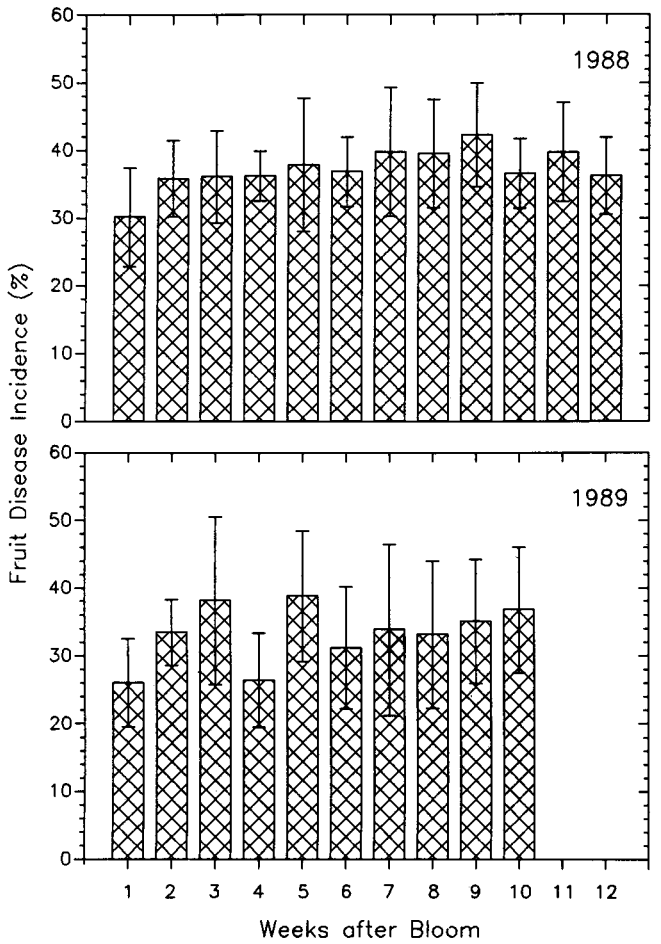

Fig. 5. Percentage of cranberry fruit (cultivar Bain McFarlin) >4 mm in diameter that contained mycelia or stromata of *Monilinia oxycocci*. Percentages are means from 10 randomly selected 625-cm² areas in a commercial cranberry field. Fruit were collected weekly from the end of bloom until harvest in 1988 and 1989. Bars are 95% confidence intervals of the means.

DISCUSSION

Each year, single periods of ascospore and conidium dispersal occurred. Within each dispersal period, a discrete spore shower was observed. No clear pattern of environmental events was observed to account for the initiation of these peak periods in

TABLE 3. Numbers of cranberry trap plants (cultivar Searles) with shoots in three different morphology classes that developed cottonball tip blight after exposure to ascospores of *Monilinia oxycocci* in two fields in 1989

	Initial day of exposure[a]											
	143		145		149		152		155		161	
Class[b]	+	−	+	−	+	−	+	−	+	−	+	−
1	1	0	3	2	3	0*[c]	2	1	1	0	0	0
2	0	0	0	0	2	0	13	2**	5	1*	1	0
3	0	0	0	1	0	1	11	0**	1	0	0	0

[a] Day of the year. One hundred plants of each class were kept for 3 days in a field infested with *M. oxycocci* (+); matching control plants were kept in a field with no history of cottonball (−). After exposure to inoculum, plants were moved from the infested field to the other field for tip blight symptoms to develop.

[b] Morphology classes: class 1 = shoots 1–10 mm long, leaves tightly clustered at the shoot tip; class 2 = shoots 11–20 mm long, leaves tightly clustered at the shoot tip and several nodes visible on the proximal end of the shoot; class 3 = shoots >21 mm long and apical leaf cluster beginning to open.

[c] Numbers of plants that developed symptoms at the two locations were significantly different at $P = 0.1$ (*) or $P < 0.005$ (**) based on a chi-square analysis.

TABLE 4. Percentage of 100 cranberry trap plants (cultivar Searles) with shoots in three different morphology classes that developed cottonball tip blight after 3 days of exposure to ascospores of *Monilinia oxycocci* in an infested cranberry field in 1990

	Initial day of exposure[a]			
Class[b]	138	144	147	151
1	0	4	26	18
2	0	11	33	7
3	0	9	10	5
P[c]	...	0.405	0.072	0.247

[a] Day of the year that plants were first placed in the field.

[b] Morphology classes: class 1 = shoots 1–10 mm long, leaves tightly clustered at the shoot tip; class 2 = shoots 11–20 mm long, leaves tightly clustered at the shoot tip and several nodes visible on the proximal end of the shoot; class 3 = shoots >21 mm long and apical leaf cluster beginning to open.

[c] Significance of F statistics from one-way analyses of variance; percentages were transformed to arcsine-square root values before analyses.

any year; however, once dispersal periods had begun, they were essentially continuous until they ceased. Although there were days within dispersal periods when no spores were detected, these occurred at the tails of the periods when the relative abundance of spores was low. The concentration of spores at these times was probably below the trapping threshold of the spore trap. Ascospores and conidia of *M. vaccinii-corymbosi* (Reade) Honey were dispersed in a similar pattern and for similar durations in a highbush blueberry field in Michigan (17).

A consistent 32- to 33-day lag between the initiation of ascospore showers and that of conidium showers occurred each year. In 1975, an identical lag period was observed between ascospore and conidium showers of *M. oxycocci* in a cranberry field in British Columbia (H. S. Pepin, *personal communication*). These results suggest that shoots infected at the beginning of the ascospore shower would be those shedding conidia 32–33 days later and that the onset of conidium dispersal was a function of the latent period, the duration of which was not affected by the range of environmental conditions observed in these studies.

A diurnal periodicity was observed in the patterns of ascospore catches, which correlated most strongly with temperature and percentage of relative humidity in each year. Greatest numbers of ascospores were caught between 1700 and 1800 h, at or just after the period of maximum temperature and minimum percentage of relative humidity each day. A similar relationship was reported for ascospores of *M. vaccinii-corymbosi* (16).

Ascospores of *M. oxycocci* are actively released from apothecia. It is likely that they are released in response to changes in temperature, relative humidity, and other stimuli (13), which could explain this diurnal periodicity. In contrast, inocula of many other fungus pathogens that actively release spores tend to be most abundant at night or in early to mid-morning (3).

Dispersal patterns of conidia of *M. oxycocci* also exhibited a diurnal periodicity, although less pronounced than that of ascospores. These findings differ from those of Ramsdell et al (16), who reported no diurnal periodicity for dispersal of conidia of *M. vaccinii-corymbosi.* As was observed for ascospores, temperature and percentage of relative humidity correlated most strongly with conidium catches in each year. Although levels of significance could not be determined for correlations between hourly totals of spores caught and hourly summaries of environmental variables due to autocorrelation, correlations were similar each year.

Significantly fewer ascospores and conidia were detected during periods of leaf wetness than during dry periods. Most leaf wetness periods occurred at night, as a result of dew, when numbers of ascospores and conidia in the air were less than during the day. Daily spore dispersal appeared to stop abruptly during periods of heavy rain. At these times, spores were probably quickly scrubbed from the air (9). However, significantly greater numbers of conidia were detected during rain-irrigation periods than during dry periods. Softer rains and irrigation events probably served to increase conidium dispersal by effecting the release of spores by water droplets tapping shoots (11).

As reported for *M. vaccinii-corymbosi* (16), wind speed was relatively well correlated with the presence of conidia of *M. oxycocci.* Disjunctors between catenulate conidia are characteristic of the section (Disjunctoriae) within *Monilinia* to which these two species belong and have been speculated to be adaptations to aid in wind dissemination (12). Turbulence may be the most important component of wind for the removal of spores from plant surfaces (2,3). Average wind speed may be an indication of the speed of those wind gusts that would serve to remove spores most effectively.

Although the correlation between the presence of ascospores of *M. oxycocci* and wind speed was relatively low, our findings contrast with those for *M. vaccinii-corymbosi* in which concentrations of ascospores were negatively correlated to wind speed (16). This contrast may be due to differences in the environments in which apothecia of *M. oxycocci* and *M. vaccinii-corymbosi* develop. Apothecia of *M. oxycocci* are found under the dense cranberry canopy among the stems of cranberry vines in which wind speeds would be reduced as compared to the relatively open environment at the base of a blueberry bush. High average wind speeds probably indicate faster gusts that would allow more ascospores to escape the plant canopy compared to lower wind speeds, but also would dilute the concentration of spores in the air.

The seasonal occurrence of inoculum of *M. oxycocci* coincided with host phenological development. Ascospore showers began at different times relative to budbreak each year, but the median numbers of ascospores were caught at times when the majority of shoots were between approximately 11 and 30 mm in length. Cranberry shoots appeared to be most susceptible to infection by ascospores when elongating, and several nodes were exposed. Although the response of shoots in different morphological classes to infection by ascospores could not be compared directly in the trap plant experiments in 1989, disease incidence was higher in classes 2 and 3. The results of trap plant trials in 1990 were somewhat equivocal because disease incidence among classes was not significantly different and because relative disease incidence among classes differed in each trial.

A complicating factor in interpreting the results of the trap plant experiments is that each morphology class was defined as a range of shoot lengths, and shoots may have advanced into the next class during the period that they were exposed to inoculum. In the 1989 trials and the trial planted on day 144 in 1990, shoots in classes 1 and 2 were distributed throughout the size ranges. In the trials planted on days 147 and 151 in 1990, the majority of the shoots in these classes were at the longer end of the range, and many probably grew into the next class during the period that they were in the infested field. In each trial in 1990, plants with shoots in class 3 were represented by many shoots that were 40 mm in length or longer and were probably no longer susceptible to infection. Therefore, although significant differences among classes were not observed in 1990, a trend was evident that supports Woronin's (25) claim that primary infection only occurs on very young, elongating leaves and stems that had no or only a very thin cuticle.

Conidium spore shower intensity was correlated with bloom. Dispersal periods and showers of conidia began at different times relative to bloom in both 1988 and 1989, but the days on which median numbers of conidia were collected each year were within 2 days of full bloom. The correlation between numbers of conidia accumulated between field visits and percentage of shoots with open blossoms demonstrated that there was a close relationship between spore shower intensity and the proportion of shoots with open blossoms. Therefore, either shoots continued to release conidia up to peak bloom, after which diseased shoots began to desiccate and spore dispersal ceased, or shoots that became symptomatic just before peak bloom bore more conidia than those that became diseased earlier in the epidemic.

In addition, the correlation between the same conidium accumulation data and tip blight incidence suggested that the intensity of the spore shower was greatest at the time that most shoots had developed symptoms. It should be emphasized that percentage of shoots with tip blight is different from the percentage of diseased shoots bearing conidia; conidia were not found on diseased shoots until approximately 1 wk after tip blight symptoms first were observed and different proportions of diseased shoots eventually bore conidia in 1988 and 1989.

Although the initiation of the conidium shower may be a function of latent period, once the shower began there appeared to be a direct association between bloom and conidium dispersal. The correlations observed between both conidium shower intensity and bloom or tip blight development and between bloom and tip blight may be reflections of physiological interactions between host and pathogen. This interaction may influence the number of conidia disseminated per shoot, the length of time that conidia were disseminated from diseased shoots, or when infected shoots desiccated and ceased to disperse conidia.

None of the shoots exposed to conidia in the field in 1989 became diseased, thus supporting the tenet that conidia do not cause tip blight. Whereas this has been assumed for *M. oxycocci*

and related species (4,5,12,22,23), it had not previously been substantiated. On the other hand, fruits that developed from blossoms that had been inoculated with ascospores, either naturally in the field or artificially in the greenhouse, developed typical signs of cottonball fruit infection. This is the first report of cottonball fruit rot caused by ascospores. Previously, ascospores of *M. oxycocci* and other species in the section Disjunctoriae were thought only to cause tip or shoot blight (4,5,12,22,23). However, fruit rot caused by ascospore infection should not cause economic loss in Wisconsin because the peak period of ascospore dispersal and bloom rarely, if ever, would coincide.

Fruit rot incidence did not increase after bloom in any year. After diseased shoots had desiccated in the field, the fungus only occurred within fruit infected during bloom, substantiating previous observations that fruit are only infected at that time (4,22,23,25). Although conidia of *M. vaccinii-corymbosi* have been found on fruit of highbush blueberry on rare occasions (5), conidia of *M. oxycocci* were never observed on or in cranberry fruit during the course of this study.

This investigation has shown that inocula of *M. oxycocci* were present in the field at periods in which susceptible plant organs were most abundant. Cottonball infection periods were single continuous events that occurred during 10- to 14-day spore showers. By precisely timing fungicide applications to host phenology, cottonball probably could be managed effectively with triforine (15). Applications should be scheduled to adequately cover the narrow windows of time when susceptible organs are present. However, relatively few infected shoots can produce large numbers of conidia, and only 1% fruit rot can result in economic loss (14). Consequently, unless all primary infection could be eliminated, emphasis of future research should be placed on developing practices to manage secondary infection.

LITERATURE CITED

1. Alberga, A. H. 1987. Microclimate influences on *Pseudomonas syringae* bacteria on snap beans. Ph.D. thesis, University of Wisconsin, Madison. 248 pp.
2. Aylor, D. E. 1978. Dispersal in time and space: Aerial pathogens. Pages 159-180 in: Plant Disease: An Advanced Treatise, Vol. II. J. G. Horsfall and E. B. Cowling, eds. Academic Press, New York.
3. Aylor, D. E. 1990. The role of intermittent wind in the dispersal of fungal pathogens. Annu. Rev. Phytopathol. 28:73-92.
4. Bain, H. F. 1926. Cranberry Disease Investigations on the Pacific Coast. U.S. Dept. Agric. Bull. No. 1434. Washington, DC. 29 pp.
5. Batra, L. R. 1983. *Monilinia vaccinii-corymbosi* (Sclerotiniaceae): Its biology on blueberry and comparison with related species. Mycologia 75:131-152.
6. Batra, L. R. 1988. *Monilinia gaylussacia*, a new species pathogenic to huckleberries (*Gaylussacia*) in North America. Mycologia 80:653-659.
7. Batra, L. R. 1988. Disjunctors in *Monilinia* (Discomycetidae: Sclerotiniaceae) and their significance in classification. Mycologia 80:660-664.
8. Burkard Manufacturing Co. Ltd. Operating instructions. Seven day volumetric spore trap. Rickmansworth, Hertfordshire, England. 6 pp.
9. Gregory, P. H. 1973. The Microbiology of the Atmosphere, 2nd edition. Leonard Hill, London, U.K. 377 pp.
10. Harada, Y. 1977. Studies on the Japanese species of *Monilinia* (Sclerotiniaceae). Bull. Fac. Agric. Hirosaki Univ. 27:82-109.
11. Hirst, J. M., and Stedman, O. J. 1963. Dry liberation of fungus spores by raindrops. J. Gen. Microbiol. 33:335-344.
12. Honey, E. E. 1936. North American species of *Monilinia*. I. Occurrence, grouping, and life histories. Am. J. Bot. 23:100-106.
13. Ingold, C. T. 1971. Fungal Spores, Their Liberation and Dispersal. Clarendon Press, Oxford, U.K. 302 pp.
14. Jeffers, S. N. 1991. Managing cranberry cottonball caused by *Monilinia oxycocci* with fungicides. Plant Dis. 75:502-506.
15. Mahr, D. L., Jeffers, S. N., Stang, E. J., and Roper, T. R. 1990. Cranberry Pest Management in Wisconsin. Univ. Wis.-Ext. Publ. No. A3276. Madison, WI. 16 pp.
16. Ramsdell, D. C., Nelson, J. W., and Myers, R. 1974. An epidemiological study of mummy berry disease of highbush blueberry. Phytopathology 64:222-228.
17. Ramsdell, D. C., Nelson, J. W., and Myers, R. 1975. Mummy berry disease of highbush blueberry: Epidemiology and control. Phytopathology 65:229-232.
18. Sanderson, P. G., and Jeffers, S. N. 1989. The cottonball disease of cranberry in Wisconsin: Occurrence, symptoms, and disease progress. Acta Hortic. 241:312-317.
19. Sanderson, P. G., and Jeffers, S. N. 1989. Seasonal occurrence of spores of *Monilinia oxycocci* in a Wisconsin cranberry field. (Abstr.) Phytopathology 79:1163-1164.
20. Sanderson, P. G., and Jeffers, S. N. 1990. Cottonball disease progress in a Wisconsin cranberry field. (Abstr.) Phytopathology 80:970.
21. Sanderson, P. G., and Jeffers, S. N. 1992. Collection, viability, and storage of ascospores of *Monilinia oxycocci*. Phytopathology 82:160-163.
22. Shawa, A. Y., Shanks, C. H., Jr., Bristow, P. R., Shearer, M. N., and Poole, A. P. 1984. Cranberry Production in the Pacific Northwest. PNW 247. Pacific Northwest Cooperative Extension, Oregon, Washington, Idaho. 50 pp.
23. Shear, C. L., Stevens, N. E., and Bain, H. F. 1931. Fungous Diseases of the Cultivated Cranberry. U.S. Dept. Agric. Tech. Bull. No. 258. Washington, DC. 57 pp.
24. Snedecor, G. W., and Cochran, W. G. 1980. Statistical Methods, 7th edition. The Iowa State University Press, Ames. 507 pp.
25. Woronin, M. 1888. Uber die Sclerotienkrankheit der Vaccinieen-Beeren. Mem. Acad. Imp. Sci. St.-Petersbourg, Ser. VII 36:1-49.