Evaluation of Four Spore Traps for Monitoring Discharge of Ascospores of Venturia inaequalis

T. B. Sutton and A. L. Jones

Research Associate and Associate Professor, respectively, Department of Botany and Plant Pathology, Michigan State University, East Lansing 48824. Present address of senior author: Department of Plant Pathology, North Carolina State University, Raleigh 27607.

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ABSTRACT

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Four spore traps were compared for detecting airborne ascospores of *Venturia inaequalis*. Burkard traps and Rotorod samplers trapped ascospores more frequently and in larger numbers than wind vanes with vertically mounted microslides and rods. Rotorod samplers were judged more practical than Burkard traps. Ascospore catch by Rotorod samplers was significantly greater at 0.45 m above the orchard floor than at 1.60 or 2.74 m. Sampler maintenance was reduced by activating them electronically only during rain periods and by changing spore impaction surfaces only after wetting periods sufficient to initiate spore dischage.

Additional key words: epidemiology, pest management, apple scab.

Contemporary apple scab warning services identify primary infection periods on the basis of host plant development, weather data and, in some cases, estimates of maturity and discharge of ascospores of *Venturia inaequalis* (Cke.) Wint. from freshly collected leaves (4, 6, 7, 8, 9, 10). Because of the nature of a particular wetting period or because of changes in ascospore maturation after sampling, spore discharge in the orchard does not always reflect laboratory estimates of spore release. We evaluated four spore traps for detection of ascospore discharge in Michigan orchards. Our objective was to develop an economical and simple system for monitoring ascospore discharge that could be used within scab warning systems or for estimating ascospore densities in fungicide testing programs.

MATERIALS AND METHODS

Trapping sites.—Spores were trapped in 12 commercial orchards in southwest, west central, and northwest Michigan. The sites were: Berrien Springs, Watervilet, Hartford, and Paw Paw in the southwest area; Grand Rapids, Belding, Conklin (two sites), and Kent City in the west central area; and Hart, Summit, and Beulah in the northwest area. All orchards had a history of severe incidence of apple scab the previous season. Two sites were used in 1973; all were used in 1974.

Spore traps.—In 1973, four wind vanes with rod and microslide samplers, four Rotorod® samplers (Metronics Assoc., Inc., 3201 Porter Drive, Palo Alto, Calif.), and one Burkard trap [Burkard Scientific (Sales) Limited, Rickmansworth, Hertfordshire, England] were placed at Paw Paw and at Grand Rapids. In 1974, four Rotorod samplers and one Burkard trap were compared at Grand Rapids. Two Rotorod samplers were placed at each of the other locations. Trap efficiencies have not been determined for ascospores of *V. inaequalis* and have been shown to vary with trap type and wind speed in studies with other fungi (1). Spore counts are reported as total catch per trap per sampling period, or as a logit transformation of the proportion of the cumulative total for the season caught during each sampling period. Differences in trap efficiency were not considered. Sampling periods corresponded to wetting periods and, because we utilized spore counts from the Burkard tape only during the Rotorod or slide sampling periods, the periods were of equivalent length for each trap.

Each Burkard trap was set 2 m from the trunk of an apple tree and with the intake orifice about 44.5 cm above ground. Traps were adjusted to sample 10 liters of air per minute. Counts were made by mounting the Melinex tapes from the traps in lactophenol and traversing their width at 2-mm intervals under a microscope. Counts were corrected for the area examined.

The wind vanes and Rotorod samplers were placed, one of each per quadrant, 2-3 m from the trunk of the same tree as the Burkard trap and with the mid-points of their trapping surfaces about 44.5 cm above the ground. A cylindrical glass rod (7 × 75 mm) and a microscope slide (25 × 75 mm) were mounted vertically on the front of each wind vane. The rod was wrapped with a 20 × 55 mm strip of cellophane tape and positioned so that the center of the tape faced the wind. Both rods and slides were thinly coated with silicone grease (General Electric G-697). The tape was removed after exposure and placed on a microscope slide for examination. Counts were taken from 20 passes across each tape and slide sampler, and were corrected for the area examined.

The Rotorod samplers, with fixed sampling heads with plastic "I" rods (60 × 1.59 mm) were mounted on vertical pipes 20 mm in diameter and were operated at 2,400 rpm. A 25 × 25 cm piece of sheet metal was positioned about 10 cm above each sampler as a rain shield. After exposure, "I" rods were removed and examined microscopically. If ascospore numbers were high, 10 passes were made at random across the short axis. If numbers were low, 20 passes, or roughly one-third of the total sampling area of the rod, were made. Counts were adjusted to a rod total.

To reduce maintenance of the samplers and the accumulation of trash on "I" rods during dry periods, a transistorized control unit and a moisture sensor were developed for us by Mr. Cyril G. Small, 7184 Slayton Settlement Road, Lockport, New York 14094, for activating the Rotorod samplers only during wet weather. Certain of the control units were designed to operate from the pen arm of a 7-day recording hygrograph. Thus, the time and the duration of operation of the Rotorod samplers were known.

Traps were maintained from 28 March to 6 June in 1973 and from 6 April to 22 June in 1974. Trapping surfaces in the Burkard traps and the Rotorod, rod, and slide samplers were usually changed at 0800 hour after any rain.

Trap height and ascospore catch.— The best height for detecting ascospore discharge with the Rotorod sampler was determined by positioning three samplers on a tower with the midpoint of their trapping surfaces 0.45, 1.60, and 2.74 m above ground level. One tower was placed in each quadrant of a tree in an orchard with moderate scab the previous year.

Ascospore production in overwintered leaves.—A random sample of overwintering leaves was gathered from each trapping site before discharge occurred in 1974. Ascospore productivity was determined using a modification of the method of Hutton and Burchill (3). Disks were cut from the leaves with a No. 9 cork borer to give five, 50-disk replicates per site. The disks were placed in a 250-ml bottle containing 75 ml of distilled water and agitated for 2 hours by bubbling air through the water. Disks were removed from the bottles and the liquid, along with water from rinsing, was passed through a 47-mm cellulose-acetate grid filter of 0.8-μm pore size (Millipore Corporation, Bedford, Mass.). The filter was cut in half, and each half was mounted in lactophenol on a microscope slide. Ascospores were counted in five grid squares chosen at random and counts were adjusted to give the number of ascospores per square centimeter of leaf tissue. Disks were sampled until no spores were recovered on successive wettings. Between wettings, disks were held at 20 C for 3 days in closed plastic boxes, followed by 7 days in boxes with the lids of the boxes cracked open.

RESULTS

Spore trap comparisons.—On wind vanes the catch of ascospores was 1.5 to 4.0 times as great on rods as on slides, but both samplers caught significantly fewer ascospores during all spore discharges than the Burkard trap and Rotorod samplers. Only heavy spore discharges were detected by the rod and slide samplers. In 1973

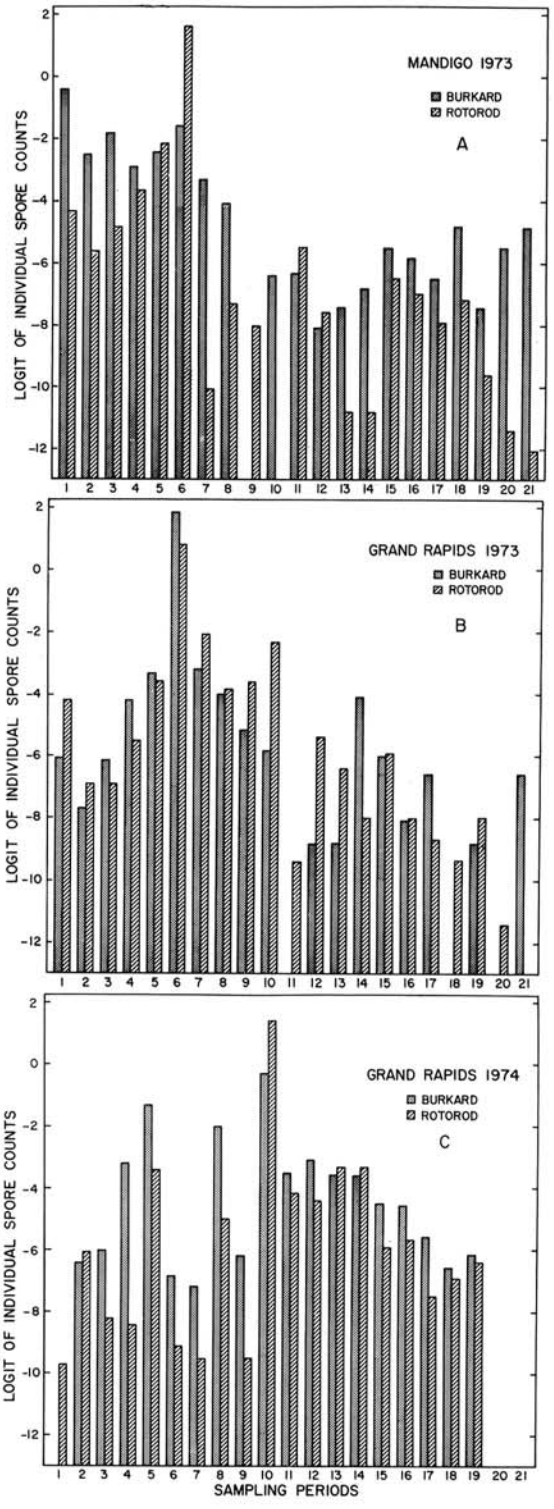

Fig. 1-(A to C). Comparison of Burkard trap and Rotorod sampler catches of ascospores of *Venturia inaequalis* during 21 sampling periods at **A)** Paw Paw and **B)** Grand Rapids in 1973, and **C)** during 19 sampling periods at Grand Rapids in 1974. Individual ascospore catches are expressed as a proportion of final cumulative spore counts and subjected to logit transformation.

during 21 wetting periods at Paw Paw, the Burkard, Rotorod, rod, and slide traps caught ascospores on 20, 20, 5, and 8 occasions, respectively. During 21 wetting periods at Grand Rapids, catches were recorded 18, 20, 5, and 5 times, respectively.

Comparisons of Rotorod and Burkard catches over 21 sampling periods at two locations in 1973 and 19 sampling periods at one location in 1974 indicated that both traps usually detected periods of heavy spore discharge and gave individual catches of similar magnitude in relation to the total catch for the season (Fig. 1). In 1973 at Grand Rapids (Fig. 1-B), the Burkard trap caught about four times more spores than the Rotorod sampler, but at Paw Paw (Fig. 1-A) the Rotorod sampler caught about seven times more ascospores than the Burkard trap. The reduced catch with the Burkard trap at this location may be related to the difficulty of maintaining a constant sampling rate with battery operated traps. If counts from period 6 are removed from the total catch, Rotorod catches were only 1.3 times as great as the Burkard catches. In 1974, the Burkard trap caught about 1.4 times as many spores as the Rotorod sampler (Fig. 1-C). Total numbers of ascospores caught per trap for the season were 6,437 and 46,729 at Paw Paw in 1973; 12,924 and 3,048 at Grand Rapids in 1973; and 6,466 and 4,663 at Grand Rapids in 1974 for the Burkard trap and Rotorod sampler, respectively.

Relationship between trap height and ascospore catch.—The number of ascospores trapped with the Rotorod samplers during 19 wetting periods was negatively correlated with placement height. Catches at 1.60 and 2.75 m were 17 and 8%, respectively, of those at 0.45 m.

Rotorod catches in relation to spore release from overwintered leaves.—Numbers of ascospores per square centimeter of overwintered leaf tissue obtained by discharging perithecia in leaf disks were positively correlated with numbers of ascospores trapped in the orchards ($r = 0.76$; $P = 0.05$). Estimated total catches per Rotorod sampler ranged from 44 at Kent City to 171,272 at Berrien Springs. Spores were trapped only five and six times at Kent City and Conklin, respectively, compared to 19 to 23 times at the other monitoring sites. Orchard locations where disk samples produced 224 or more ascospores per cm^2 of overwintering leaf tissue gave a minimum of 6,172 ascospores per Rotorod sampler per season and were the most suitable trapping sites. Rotorod samplers detected ascospores only during heavy discharge periods at locations where disk samples produced less than 224 ascospores per cm^2 of overwintering leaf tissue.

DISCUSSION

Inconsistence in spore catches between Rotorod samplers and Burkard traps occurred primarily at low spore levels and is probably the result of our experimental design. Ascospores were trapped with Rotorod samplers but not the Burkard trap during sampling periods 11, 18, and 20 at Grand Rapids in 1973 and period 1 in 1974 (Fig. 1). Less than three ascospores per sampler were detected on these occasions. Because four Rotorod samplers but only one Burkard trap was used, detection of low numbers of ascospores would presumably be weighted in favor of the Rotorod samplers. The inconsistence in ascospore catch during periods 9 and 10 at Paw Paw (Fig. 1) occurred on adjacent days and may reflect an error in matching up catches with the two traps. If the sampling periods are combined, the relative catches for the traps are similar.

The Rotorod sampler seems better suited for a regional apple scab management program than the other traps evaluated. Rod and slide samplers are not suitable because of their high detection threshold. In addition because neither sampled isokinetically, evaluation of individual catch was difficult and the relationship between different catches could not be determined.

As reported in other studies (1, 2, 5), the Burkard volumetric trap (Hirst type) was reliable, and because it samples isokinetically over a wide range of wind speeds, the variation in relative discharge from one wetting period to another could be evaluated. However, since the Burkard traps were serviced after each rain rather than weekly, considerable time was required for removal and replacement of the Melinex tape on the drum assembly. A further disadvantage of the Burkard or Hirst traps is relative high cost.

The Rotorod sampler proved satisfactory for routine monitoring of apple scab ascospores. It detected small numbers of ascospores, was reliable, is inexpensive and simple to maintain and operate. Catches with the Rotorod sampler were generally of the same magnitude relative to the season totals as with the Burkard trap. Rotorod samplers, operated only during wet weather, ran 4-6 weeks per battery charge compared to 3-5 days for Burkard traps. The "I" rods were easy to remove and required less time to examine than the Melinex tape from the Burkard trap. This is important because rapid determination of spore catches is critical if counts are to be used in timing applications of fungicides for scab control. Experience gained during 1974 indicated that inexperienced personnel can quickly master the techniques of trap maintenance and spore counting. Because of the low relative cost per sampler, it was possible to use more than one sampler at a site and to place several samplers in each fruit-growing region of the state.

One disadvantage of the Rotorod sampler controlled by a moisture sensor was that fair weather discharges such as observed by Miller and Waggoner (5) would not be detected. However, discharges without wetting are exceptional, and of doubtful importance in most commercial orchards where the carryover of scab from season to season is low. Moreover, care in construction and placement of the moisture sensor is required to prevent premature inactivation of the Rotorod samplers under fast drying conditions and a subsequent underestimate of ascospore concentrations.

Because the detection of low concentrations of spores may be erratic, catch with the Rotorod sampler should be maximized through proper trap placement and orchard selection. In this study, samplers 0.45 m above ground and in orchards with a relatively large overwintering population of *V. inaequalis* had the highest ascospore catch. Orchards with high inoculum levels are most

suitable as monitoring sites because of the likelihood of detecting airborne spores during each discharge period. Assuming weather favorable for infection, growers following recommendations based on catches in orchards with low numbers of maturing perithecia might go unprotected early in the season when spore discharge is light and later might relax their spray program before the ascospore discharge period is over.

We believe Rotorod samplers are valuable to apple pest management programs and to fungicide testing programs as a tool for determining relative ascospore discharge during each infection period. Currently, we are using spore trapping in combination with temperature and duration of leaf wetness data to assess the likelihood of primary apple scab infection. As quantitative relationships among ascospore catch, infection, and the effectiveness of particular spray programs are established, spore trap results should be more useful and may help to reduce fungicide usage.

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