

Relative Collection Efficiency of Rotorod and Burkard Spore Samplers for Airborne *Venturia inaequalis* Ascospores

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ABSTRACT

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Rotorod spore samplers and a Burkard volumetric spore sampler were placed next to each other in an orchard and used to simultaneously collect *Venturia inaequalis* ascospores naturally discharged during rain. In addition, these spore samplers were operated next to each other inside a closed room in which *V. inaequalis* ascospores were released via the exhaust of a spore-release tower. These data were used to determine the

relative collection efficiency of the two types of samplers for airborne *V. inaequalis* ascospores. In the relatively still air of the laboratory, E_{Rotorod} was $\sim 0.24 E_{\text{Burkard}}$. In the field, E_{Rotorod} was $\sim 0.37 E_{\text{Burkard}}$. A large part of this difference between the findings in the field and laboratory can be explained by the effects of wind on collection efficiency of the samplers.

Reliable, accurate samplers for monitoring airborne spores are indispensable for further understanding of the epidemiology of many plant diseases. For example, further development of epidemiological models of apple scab requires, among other things, a means of predicting and measuring the aerial concentration of ascospores in the orchard canopy. The Burkard volumetric spore sampler (11,21), by virtue of its long record for reliably and consistently recording airborne spores in all kinds of weather, has often been the sampler of choice in epidemiological studies (3,4,8,12,14) and over the years has been the sampler to which others have been compared (7,13,19,21). In an effort to expand the spatial extent of spore sampling, particularly at several heights above the ground, it is useful to operate a Burkard sampler in conjunction with a lighter weight, less-expensive sampler. The Rotorod is a field-proven sampler (6,19) that fits these requirements.

Both Burkard and Rotorod samplers have been used for monitoring airborne ascospores of *Venturia inaequalis* (Cooke) G. Wint. (19). *V. inaequalis* ascospores are released from ground-level, and their concentration in the air, C , varies considerably with height (3,19). Rotorods have a distinct advantage over Burkards because they are easy to deploy at several heights above the ground with minimum disturbance of the wind, and they are inexpensive enough to allow several to be deployed at a sampling site. Another advantage of Rotorods over suction-type

samplers is that their efficiency is relatively insensitive to changes in wind speed. This also makes them advantageous for measuring vertical spore gradients, because wind speed typically increases with height above the ground. A disadvantage of Rotorods is that they must be changed frequently to determine a temporal pattern and to avoid overloading the sampling surface with material. When used in combination with a Burkard sampler, their respective advantages complement each other in ways that are useful for monitoring spore dispersal near a ground-level source, as is the case for *V. inaequalis* ascospores.

To maximize the advantage of this complementary relationship, we need a basis of comparison in terms of the relative trapping efficiencies of Rotorod and Burkard samplers. This paper reports on side-by-side laboratory and field comparisons of these two types of samplers, which allow their relative collection efficiencies to be determined. The results are discussed in terms of some physical parameters that govern collection efficiencies of suction and rotary impaction samplers.

MATERIALS AND METHODS

Field study. During the 1991 and 1992 apple scab ascospore-release seasons, spore samplers were placed near the center of a 0.2-ha orchard in Mt. Carmel, CT, containing a 2-yr-old planting (during 1991) of dwarf resistant apple trees trained to a slender spindle at 1.8×3.6 m spacings. The ground in the orchard was uniformly "seeded" with apple leaves with scab lesions. The sampling site was essentially level. The number of airborne *V.*

inaequalis ascospores was monitored with a Burkard volumetric spore sampler (Burkard Scientific Sales, Ltd., Rickmansworth, Hertfordshire, England) located near the center of the orchard with its orifice at a height of about 0.45 m. The Burkard sampled air at approximately 10 L/min and yielded a continuous record of ascospores in the air. A needle was used through the trap orifice to make a time mark at both the start and end of the spore trap tape to precisely identify the beginning and ending of the sampling period. The tape was changed weekly. The clock of the spore trap kept time accurately to within 30 min or less during the 7-day sampling periods.

V. inaequalis ascospores also were monitored during fixed 2-h time intervals with Rotorod spore samplers with retracting-type sampling heads (model 82, Sampling Technologies, Inc., Los Altos Hills, CA), which were deployed under rain shields (19). The collecting surface was one side of a 32 mm long by 1.59 mm square cross section of plastic rods. Two Rotorod samplers were placed next to each other near the center of the orchard with the center of the sampling rod (when swung down during operation) at a height of about 0.40 m above the ground. The Rotorods were triggered to operate during rain events occurring during daylight hours. The first Rotorod operated for 2 h after the start of rain and then switched off. The second Rotorod then was turned on and operated for an additional 2 h. The Rotorods and the Burkard sampler were located about 3 m apart.

The spore collecting surfaces for both kinds of samplers were made sticky with a thin layer of high-vacuum grease (Dow Corning, Midland, MI) thinned with hexane before application to the surfaces. The number of *V. inaequalis* ascospores deposited on sampling surfaces were counted at 200 \times with a microscope. Tapes from the Burkard spore trap were cut into daily segments (48 mm) and mounted on glass slides with double-stick tape and then Gelvatol (Monsanto Chemical Co., St. Louis, MO) under a coverslip. Ascospores were counted on eight consecutive 0.5-mm traverses (4 mm) centered on each 2-h exposure. Ascospores collected on the plastic rods of the Rotorods were counted by making traverses across the rod over a 20 mm length of rod, beginning 2 mm from the free end. Spore counts were converted to counts per unit volume of air sampled, G (spores per liter), by dividing by the sampling rate (~ 10 L/min for the Burkard and ~ 38 L/min for the Rotorods), and by the duration of the sampling period (usually 120 min).

Wind speed and direction, air temperature, and relative humidity were monitored continuously at the center of the orchard with a data logger (model 21X, Campbell Scientific, Inc., Logan, UT). Wind speed was measured with a cup anemometer (model 014A, Met-One, Inc., Grants Pass, OR) located at a height of 0.6 m above the ground; wind direction was measured with a vane (Met-One model 024A) placed 3.2 m above the ground. A temperature/relative humidity probe (Campbell, model 207) was shielded from the sun and located at a height of 1.5 m. A leaf wetness-resistance grid (Campbell, model 731) was used to trigger Rotorod spore traps to turn on at the beginning of rainfall. Rainfall amounts were obtained with a tipping bucket rain gage (model RG2501, Sierra-Misco, Inc., Berkeley, CA).

Laboratory study. The source of ascospores was scabbed leaves collected from the ground in the test orchard, described above, during April–May each year and kept dry at 4 C until used. The ascospores were released into the air in the laboratory by placement on a screen at the top of a spore-release tower through which air was drawn by a fan and exhausted into the lab. The leaves were kept wet by misting periodically with a spray bottle. The air in the lab was stirred by an oscillating fan directed toward the ceiling. The speed of the air flow around the spore samplers was ~ 0.10 – 0.15 m/s.

Rotorod samplers were placed on either side (total of two) of the Burkard sampler (0.28 m from) and at the same height as the Burkard sampler's orifice (about 0.45 m above the floor). Spore samplers were turned on, and spore release was initiated and maintained for a 2-h period. The sample collection tape on the Burkard was marked at the beginning and end of the sampling period as described above.

Relative sampler efficiency. As noted above, both kinds of samplers tested yielded G , i.e., the number of spores collected per unit volume of air sampled. To obtain the aerial concentration, C (spores per cubic meter), which is the quantity of epidemiological significance, G must be divided by the particle collection efficiency of the sampler. The relative collection efficiency of the two kinds of samplers, E_{rel} , is

$$E_{rel} = G_{Rotorod} / G_{Burkard} \quad (1)$$

The efficiency of the Rotorod, $E_{Rotorod}$, can be estimated from equation 1, if the efficiency of the Burkard sampler, $E_{Burkard}$, is known. The relationship between $G_{Rotorod}$ and $G_{Burkard}$ was examined by regression with a linear model with no constant (SYSTAT, Inc., Evanston, IL).

Effect of air speed on sampler efficiency. Suction samplers, such as the Burkard, are subject to nonisokinetic sampling errors (5,16). The collection efficiency, E_C , of a thin-walled suction head that is aligned with and facing into the flow is a function of ambient air speed, sampler inlet flow speed, and particle size and can be expressed as (5,16)

$$E_C = C_s / C_a = 1 + [(U_a / U_s) - 1] \times f(S) \quad (2)$$

and

$$f(S) = 2 \times S / (1 + 2 \times S) \quad (3)$$

in which C is the aerial concentration of particles, U is air-flow speed, and the subscripts s and a refer, respectively, to the regions just inside the suction head and in the undisturbed air flow upwind of the sampler. S is the Stokes' number (1,5,9,10) based on the sampler inlet size and the free-stream flow speed, i.e.,

$$S = v_s U_a / gL \quad (4a)$$

in which v_s is the settling speed of the particle in still air (for *V. inaequalis* ascospores $v_s = 2$ mm/s [9]), g is the acceleration of gravity, and L is the width of the sampler entrance port. The sampling orifice of the Burkard sampler is 2 mm wide by 14 mm long. At a flow rate of 10 L/min, the average value of U_s is ~ 6 m/s. Thus, S for the Burkard orifice is a function of wind speed and is calculated as $0.00102 U_a$.

In laboratory tests, U_a was ~ 0.1 m/s. Using equation 2, we expect $E_{Burkard}$ for *V. inaequalis* ascospores to be ~ 0.98 of its maximum efficiency, which is $\sim 90\%$ for particles the size of *V. inaequalis* ascospores (G. M. Wili, Burkard Manufacturing, *personal communication*).

The collection efficiency of a Rotorod sampler also depends on a Stokes' number, in which U_a is replaced by the tangential speed of the rotating rod, U_r , so

$$S = v_s U_r / gL \quad (4b)$$

Here, v_s and g have the same meaning as above, and L is the width of the rod. For the I-rod retracting heads, U_r is ~ 10.8 m/s, yielding $S = 1.4$. Equation 4b does not depend on wind speed, and the collection efficiency of Rotorods is expected to be relatively independent of wind speed.

The impaction efficiency for particles (droplets) on a stationary cylinder or ribbon in an air stream with speed U_r can be approximated by (2,15,17)

$$E_i = 0.86 / (1 + 0.442 / S^{1.967}) \quad (5)$$

which for $S = 1.4$ gives a theoretical value of $E_i = 0.70$. Generally, the overall efficiency of a Rotorod sampler is less than the theoretical value because the sampler disturbs the air flow and because the retention of the sticky surface is less than 100% (18). For example, Ogden and Raynor (18) found that the efficiency of rotoslides for collecting ragweed (*Ambrosia artemisiifolia*) pollen was 64% compared to a theoretical value near 100%.

Likewise, the actual efficiency of Rotorods for collecting *V. inaequalis* ascospores is expected to be less than the theoretical value given above.

RESULTS

The number of *V. inaequalis* ascospores per unit volume of air collected by the two kinds of samplers in the laboratory tests (Table 1) was highly correlated ($r = 0.98$, $P < 0.000001$). The relative efficiency of collection of ascospores in the laboratory by the Rotorod samplers (compared to the Burkard sampler) was 0.24 ± 0.01 (slope \pm SE [standard error]).

TABLE 1. Relative sampling efficiency, $E_{rel.}$, of Rotorods compared to Burkard spore samplers determined in laboratory tests

Test	$G_{Burkard}^a$	$G_{Rotorod}^a$	$E_{rel.}^b$
1	158.30	31.64	0.20
		33.39	0.21
2 ^c	163.75	34.27	0.21
		40.86	0.25
3	50.83	15.82	0.31
		14.06	0.28
4	83.33	17.58	0.21
		20.65	0.25
5	365.80	110.29	0.30
		94.91	0.26
6 ^c	791.43	171.24	0.22
	
7	180.00	45.70	0.25
		58.88	0.33
8	121.70	25.05	0.21
		21.09	0.17

^a Number of spores caught on trapping surface per cubic meter of air sampled.

^b Relative collection efficiency defined by equation 1 as the ratio $G_{Rotorod}/G_{Burkard}$.

^c Duration of tests 2 and 6 were 1.33 and 1.75 h, respectively; for all others, it was 2 h.

^d Sample was ruined during handling.

The number of ascospores per unit volume of air collected by the two kinds of samplers in the orchard (Table 2) also was highly correlated ($r = 0.77$, $P = 0.00005$). The collection efficiency of the Rotorod samplers when outdoors relative to the Burkard sampler was about $E_{rel.} = 0.37 \pm 0.05$ (slope \pm SE). After the Burkard data were adjusted for the effect of wind speed (Table 2) on collection efficiency (equations 2-4), the corrected overall relative efficiency $E_{rel.}'$ was equal to 0.32 ± 0.05 . There was no significant effect of wind speed, wind direction, air temperature, or amount of rain on the variation of $E_{rel.}'$.

DISCUSSION

Rotorods and Burkard samplers both collect *V. inaequalis* ascospores efficiently, and their advantages complement each other when used to study apple scab epidemiology (19). Sutton and Jones (19) compared catches of *V. inaequalis* ascospores by Burkard and Rotorod samplers and demonstrated the utility of Rotorods in studying apple scab epidemiology, but they did not consider differences in sampler efficiency. In their study, Rotorods sometimes caught more and sometimes caught fewer ascospores than did a Burkard sampler. The ratios (Rotorod/Burkard) of the total seasonal catches of ascospores for their three sampling sites were 7.26, 0.24, and 0.72. In the present study, Rotorods always collected fewer ascospores (on a per unit volume of air sampled basis) than did the Burkard sampler, and, thus, this ratio was always less than 1.0. The large value found in Sutton and Jones' study may have been due to differences in placement of the samplers relative to inoculum density in an unevenly distributed source region.

Assuming that $E_{Burkard}$ for collecting *V. inaequalis* ascospores in the slight air currents in the laboratory is $\sim 90\%$ implies that $E_{Rotorod}$ is $\sim 21\%$. A collection efficiency of 21% for *V. inaequalis* ascospores by Rotorods (for which $S = 1.4$) is in good agreement with Noll's (17) findings for a comparable target diameter and tangential speed of the rod on his sampler (points designated A3 in literature citation 17, Fig. 7). Outdoors the Rotorods appeared to be relatively more efficient than Rotorods indoors

TABLE 2. Comparison of *Venturia inaequalis* ascospores collected by Rotorod and Burkard samplers in an orchard during 2-h sampling periods

Year	DOY ^a	G_B^b	G_R^b	U^c	T_{air}^c	Rain ^c	θ^c	$G_b'^d$	$E_{rel.}^e$	$E_{rel.}'^e$	
1991	103.1	154.4	50.3	0.8	7.0	0	170	176.6	0.33	0.28	
	103.2	150.9	70.5	0.6	4.6	1	105	166.3	0.47	0.42	
	105.1	229.8	127.2	0.6	4.9	1	190	253.3	0.55	0.50	
	105.2	1,173.5	836.6	0.8	5.3	1	144	1,338.8	0.71	0.62	
	105.3	1,328.1	203.4	1.1	6.0	0	126	1,555.2	0.15	0.13	
	105.4	529.8	148.1	0.9	6.6	4	118	612.1	0.28	0.24	
	107.1	534.9	275.7	1.0	11.2	0	133	621.1	0.52	0.44	
	107.2	22.5	18.0	1.1	11.4	0	142	26.6	0.80	0.68	
	107.3	28.9	5.3	1.0	9.7	0	140	33.6	0.18	0.16	
	120.1	287.7	70.3	0.7	9.4	2	146	322.1	0.24	0.22	
	120.2	544.7	91.4	0.6	10.0	6	144	601.3	0.17	0.15	
	1992	113.1	50.8	23.7	0.7	14.9	1	142	57.4	0.47	0.41
		113.2	31.7	30.3	0.8	15.3	0	151	36.2	0.96	0.84
113.3		60.8	18.5	0.8	15.3	0	144	69.5	0.30	0.27	
113.4		72.5	30.3	0.8	16.1	1	143	82.6	0.42	0.37	
129.1 ^f		838.9	343.7	2.3	12.6	0	53	1,043.4	0.41	0.33	
129.2		516.7	65.0	2.0	12.5	4	46	640.2	0.13	0.10	
129.3		20.0	10.1	1.7	12.1	1	38	24.5	0.51	0.41	
137.1		111.7	43.1	0.8	11.9	3	42	127.6	0.39	0.34	
137.2		186.7	76.9	0.8	12.6	1	46	212.7	0.41	0.36	
147.1		72.5	15.8	0.6	11.4	0	171	79.8	0.22	0.20	

^a DOY is day of the year from January 1, in which the added decimal indicates individual 2-h sampling periods on the same day.

^b G is the number of ascospores collected per cubic meter of air during the sampling period, in which the subscripts B and R represent Burkard and Rotorod samplers, respectively.

^c Average 2-h values of wind speed (U [meters per second]), air temperature (T_{air} [C]), wind direction ($\theta = 0$ degrees is north), and rainfall amount (R) during periods of ascospore collection by the samplers. $R = 0$ means no rain was recorded (or equivalently, $R <$ the least count of the rain gage, i.e., < 1 mm).

^d G_b' is equal to G_B divided by the reduced efficiency due to nonisokinetic sampling, E_C , calculated with equation 2 and measured values of U .

^e $E_{rel.}$ is calculated as G_R/G_B ; $E_{rel.}'$ is calculated as G_R/G_b' .

^f This sampling period was 54 min, all others were 2 h.

compared to a Burkard sampler. Part of this discrepancy could be explained in terms of the expected reduction of efficiency of the Burkard sampler operating in the wind speeds encountered in the field. After making this correction (equations 2-4; Table 2), the average efficiency in the field and laboratory differs by only about 11%, which is within the margin of error of these experiments.

Differences in vertical and horizontal placement of the two kinds of samplers could cause slight differences in the determination of relative efficiency. The height of the orifice of the Burkard sampler in the field was a nominal ~5 cm higher than the Rotorod samplers. In another study (3), the vertical variation of *V. inaequalis* ascospore concentration in the height range of 0.4 to 2.0 m decreased exponentially with height as $\exp(-b \times z)$, in which b averaged about 1.0 (range 0.6 to 1.6). Thus, it is reasonable to expect that the concentration at the Burkard orifice could have been about 5% smaller than at the sampling heads of the Rotorods. This correction is small and would change the average relative efficiency only from 0.32 (after correction for wind-speed effects) to 0.30.

Suction-type samplers also can undersample spores if they are misaligned with the direction of the wind (10,16). In very light winds (speeds <25-30 cm/s), the Burkard was sometimes misaligned with the wind by as much as 90 degrees. Alignment information was not available during the actual test runs, and this correction cannot be made. However, in view of the moderate average wind speeds observed during the tests (Table 2), misalignment of the Burkard with the wind direction should have been small but might help account for the greater variation in relative sampling efficiency found in the field compared to the laboratory. The differences remaining after correction for the effect of average wind speed on efficiency may be due to uneven distribution of source leaves on the ground relative to the positions of the samplers and due to atmospheric turbulence, which could have caused the Burkard to undersample ascospores by as much as 15-25% (20).

Rotorod samplers equipped with I-rod retracting heads collected airborne *V. inaequalis* ascospores in the laboratory with an efficiency of ~21%. Rotorod and Burkard samplers can be used to measure comparable concentrations of *V. inaequalis* ascospores in the field (over a range of wind speeds at the height of the sampler equal to 0.5-2.3 m/s) using a relative efficiency (Rotorod/Burkard) of 37%. If wind speed at the height of the Burkard orifice is known, then the correction for the effect of wind speed on collection efficiency described here can be made, and a corrected relative efficiency of 32% can be used. Both types of samplers have advantages for sampling ascospores and can be used in a complementary way to add to our understanding of apple scab epidemiology.

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