Factors Affecting Airborne Concentrations of Podosphaera xanthii Conidia and Severity of Gerbera Powdery Mildew

Leah L. Granke¹, Layla E. Crawford², and Mary K. Hausbeck³,⁴
Department of Plant Pathology, Michigan State University, 107 CIPS, East Lansing, MI 48824-1311

Abstract. To determine the factors affecting airborne conidial concentrations of Podosphaera xanthii Braun and Shishkoff and powdery mildew severity in greenhouse-grown potted gerbera (Gerbera jamesonii H. Bolus), airborne conidial concentrations of conidia were monitored in a glass and polyethylene greenhouse. Temperature, relative humidity, and leaf wetness were recorded onsite, and the percentage of foliage with visible disease was assessed weekly at the glasshouse and every 2 weeks at the polyethylene greenhouse (1 to 10 visual rating scale). Peak airborne conidial concentrations occurred at 0800/1600 and 0900/1400 UTC at the glasshouse and polyethylene greenhouses, respectively. Few conidia were sampled between 2200 and 0500 UTC at either greenhouse. Worker activity was associated with conidial release in the glasshouse, but not in the larger polyethylene greenhouse, and worker activity may have influenced the daily periodicity of conidial concentrations. Airborne conidial concentrations were not related to environmental conditions in the same hour as conidial detection. An increase in disease severity was positively related to relative humidity and negatively related to leaf wetness at both greenhouses; in addition, temperature was negatively related to an increase in disease severity in the glasshouse. In light of the results of this study, frequent scouting and fungicide applications for powdery mildew are advised. Wide plant spacing and adequate ventilation are also recommended to reduce relative humidity in the microclimate.
weeks (three treatments). Drenches were not applied at the glasshouse, because all of the leaves displayed P. xanthii signs before 6 weeks following exposure to inoculum.

**Monitoring airborne conidia.** Concentrations of airborne conidia were monitored in each greenhouse using a 7-d volumetric spore sampler (Burkard Mfg. Co. Ltd., Rickmansworth, Hertfordshire, U.K.) from 2 Mar. to 12 Aug. 2004 in the glasshouse and from 13 July to 8 Nov. 2004 in the polyethylene greenhouse. The spore sampler was placed in the center of the center greenhouse bench with the orifice ≈0.5 m above the bench surface and operated at a flow rate of 10 L·min⁻¹. Conidia were impacted onto Melanex tape (Burkard Scientific, U.K.), which had been coated with a polyvinyl alcohol (Airvol; Burkard Scientific) and phenol mixture (35 g polyvinyl alcohol, 25 mL glycerol, 50 mL distilled water, 2 g phenol), dried overnight, and subsequently coated with an adhesive mixture of petroleum jelly and paraffin (9:1, wt/wt) dissolved in sufficient toluene (≤3 mL per 50 g of mixture) to result in a viscous consistency. Tapes were removed weekly, cut into 48-mm lengths, scored at hourly intervals, lightly stained with aniline blue in lactic acid (28 mg aniline blue, 20 mL distilled water, 10 mg glycerol, and 10 mL 85% lactic acid, diluted with 5 drops to 25 mL of distilled water), and mounted on glass slides beneath 22 × 50-mm coverslips. Each hourly interval on each slide was scanned vertically at 100× magnification. P. xanthii conidia were identified based on morphological characteristics including the presence of inclusion bodies (Braun et al., 2002) and enumerated. Conidial counts were converted to conidia per cubic meter of air sampled per hour.

**Collection of environmental and disease data.** Temperature (°C), relative humidity (%), and leaf wetness (0 to 15 scale) were recorded in each greenhouse every 15 min by a WatchDog 450 data logger (Spectrum Technologies Inc., Plainfield, IL) and an external leaf wetness sensor set at a 45° angle facing north placed within the plant canopy. Hourly averages were calculated for temperature and relative humidity. Before entering greenhouses, personnel documented date, time of day, and activity performed (applying pesticides, watering, pruning, assessing plant status).

Plants were exposed to inoculum on 27 Feb. 2004 (glasshouse) and 12 July 2004 (polyethylene greenhouse) by placing one severely infected gerbera plant (=100% of foliage showed pathogen signs) with actively sporulating P. xanthii colonies in the center of each greenhouse bench adjacent to healthy gerberas. Inoculum-bearing plants remained on the greenhouse bench for the duration of the experiment. Powdery mildew disease severity was assessed every week in the glasshouse (six ratings, 375 plants) and every 2 weeks (seven ratings, 209 plants) in the polyethylene greenhouse based on a visual estimation and rated on a scale of 1 to 10, where 1 = no disease, 2 = trace to 10%, 3 = 11% to 20%, 4 = 21% to 30%, 5 = 31% to 40%, 6 = 41% to 50%, 7 = 51% to 60%, 8 = 61% to 70%, 9 = 71% to 80%, and 10 = 81% to 100% of foliage infected. Ratings were conducted more frequently in the glasshouse because disease progressed more quickly in this greenhouse than in the polyethylene greenhouse.

**Statistical analysis.** Statistical analyses were performed using the SAS statistical package Version 9.1 (SAS Institute, Inc., Cary, NC). The GAM procedure of SAS used β-spline and local regression methods to model smoothed seasonal trends for daily conidial concentrations and daily temporal trends for non-zero hourly conidial concentrations in each greenhouse. The smoothing parameter was chosen to minimize the generalized cross-validation criterion.

To determine if conidial release was more likely during periods with worker activity, the FREQ procedure was used to conduct a one-sided Fisher’s exact test to determine if the probability of conidial release and detection was greater in the activity group vs. the time periods without worker activity, and an odds ratio was used to estimate relative risk of conidial dispersal during periods of worker activity.

Hourly conidial concentrations and corresponding environmental conditions were temporally autocorrelated time series (7). Hence, time series analyses were used to study changes in these variables through time and uncover patterns and relationships in the data. Using the ARIMA procedure, regression analyses were completed, regressing conidial concentrations on covariates (environmental conditions) assuming an autoregressive (AR) moving average (MA) error structure (6, 42). The AR portion of the error model accounts for the dependency of the current value of airborne conidial concentrations on past values, and the persistence of error terms beyond one observation for conidial concentrations is the MA portion of the error model (Box and Jenkins, 1976). The mean was subtracted from each time series before analysis and first-order differencing was completed for all of the variables (including covariates) to induce stationarity [ARIMA (p,1,q)]. Differencing removes trends from the time series by taking the difference of the series from one period to another (1 = the number of lags) (8). Stationarity was evaluated using plots of the expected values of the series and its autocorrelation function to ensure temporal trends were removed. Lags, time periods before the current time period, of up to 5 h were permitted for each environmental factor to allow for the influence of environment on pathogen sporulation in the hours before conidial dispersal. Series of environmental variables were prwhitened using an autoregressive moving average (ARMA) filter to achieve a white noise (random) residual series before crosscorrelation (Box and Jenkins, 1976). The goodness of fit of models was measured using the Akaike information criterion (Akaike, 1974). Variables with $P \leq 0.05$ were considered statistically significant.

The GAM procedure was used to develop models relating the increase in disease severity

Fig. 1. Colonies of powdery mildew (<em>Podosphaera xanthii</em>) on (A) leaves of gerbera and (B) a gerbera flower. (C) Chains of <em>Podosphaera xanthii</em> conidia.

a hose and water breaker as needed; care was taken to avoid wetting the foliage. Plants were fertilized during watering with 200 ppm of Peter’s 20N–20P–20K general purpose fertilizer (Scotts-Sierra Horticultural Products Co., Marysville, OH) at 2- to 3-d intervals. Substrate pH was maintained between 5.8 and 6.2 and electrical conductivity between 1.2 and 1.5 mmhos/cm by treating water as necessary with phosphoric acid.

Temperatures in both greenhouses were set at 20 to 22 °C and venting occurred when temperature exceeded 22 °C. To manage root rot at the polyethylene greenhouse, drenches of etridiazole/thiophanate-methyl 0.8 g product/liter (Banrot 40WP; Scotts-Sierra Crop Protection Co.) and mefenoxam 0.2 mL product/L (Subdue MAXX 21.3EC; Syngenta Crop Protection, Inc., Greensboro, NC) were applied in alternation every 6
from one disease rating to the next (calculated by subtracting the previous severity rating from the current severity rating for each plant) with conidial concentrations and environmental conditions in that same period. For both greenhouses, GAM yielded a parametric model. The increase in disease severity was square root transformed before analysis to satisfy the assumption of normality.

**Results**

**Atmospheric conidial concentrations.** Temperature in the glasshouse ranged from 11.3 to 30.8 °C, and temperatures in the polyethylene greenhouse ranged from 11.3 to 37.9 °C during the course of this study. Conidial concentrations were monitored for 52 d in the glasshouse and 119 d in the polyethylene greenhouse until 100% of the foliage was infected. Conidia were detected on Day 21 of monitoring of the glasshouse and Day 25 of monitoring for the polyethylene greenhouse, and conidia were sampled until the cessation of the experiment. At the glasshouse, infected plants were observed before the first conidium was sampled suggesting that conidial concentrations below the detection limit of the spore sampler were sufficient for disease development. At the polyethylene greenhouse, the first conidium was detected before the first disease rating occurred so a comparison between conidium detection and the onset of disease symptoms could not be made.

Disease severity and conidial concentrations generally increased over time. When daily conidial concentrations were modeled using nonparametric regression, peak concentrations were detected at both sites when almost all of the plant foliage displayed pathogen signs (Fig. 2A–B). When hourly conidia counts were modeled using nonparametric regression, a daily periodicity was observed where more conidia were caught during the day than at night (Fig. 3A). Peak concentrations were observed at \( \approx 0800/1600 \) hr and 0900/1400 hr in the glasshouse and polyethylene greenhouses, respectively (Fig. 3A). Relatively few conidia (less than 6%/total) were trapped in the 7-h period from 2200 to 0500 hr in both greenhouses (Fig. 3A). The probability of dispersal was higher during periods of worker activity in the glasshouse (\( P = 2.344 \times 10^{-5} \)) (odds ratio, 2.55; 95% confidence interval, 1.83 to 3.59) but not in the polyethylene greenhouse (\( P = 0.2999 \)). At the glasshouse, the time of day when worker activity was most frequently conducted coincided well with the daily periodicity observed for conidial concentrations (Figs. 3A and 3E).

When ARIMA models were developed for hourly conidial concentrations, none of the environmental parameters (covariates) measured were significant for the glasshouse, and these were not included in the model for that site. The hourly conidial concentrations for the glasshouse could be modeled as follows: conidia = 0.53\( z_{t-1} + 0.26 z_{t-2} \), where conidia is the number of conidia sampled at time \( t \) and \( z_{t-k} \) are the autoregressive error terms for \( k \) hours before conidial sampling. The hourly conidial concentrations for the polyethylene greenhouse could be modeled as follows: conidia = 1.12RH \( t-1 - 2.18 \) RH \( t-2 + 28.83 LW \( t-1 + 0.99 z_{t-1} + 0.67 e_{t-1} - 0.23 e_{t-2} + 0.09 e_{t-3} \), where conidia is the number of conidia sampled at time \( t \), RH \( t-k \) is the average relative humidity \( k \) hours before conidial sampling, and \( z_{t-k} \) and \( e_{t-k} \) are the autoregressive and moving average error terms for \( k \) hours before conidial sampling, respectively.

**Disease development.** Powdery mildew colonies were observed on gerbera by 20 d after exposure to inoculum (13 Mar.) in the glasshouse and 25 d after exposure to inoculum (16 Aug.) in the polyethylene greenhouse at the first disease rating. By 52 and 119 d after exposure to inoculum in the glasshouse and every 2 weeks at the polyethylene greenhouse (at least 100% of the foliage was covered with signs of the disease), the disease severity of powdery mildew was sampled suggesting that conidial concentrations in both greenhouses were greater than 5000 conidia/m\(^3\)/d were cropped from the figure but included in analyses (0 of 52 d at the glasshouse, 2 of 119 d at the polyethylene greenhouse). Curves were fitted by nonparametric regression (PROC GAM) and relate season to predicted conidial concentrations (conidia/m\(^3\)/d) to illustrate seasonal trends.

Disease severity at each rating (every week at the glasshouse and every 2 weeks at the polyethylene greenhouse) at (A) the glasshouse and (B) the polyethylene greenhouse over the course of monitoring. Conidial concentrations (dots) greater than 5000 conidia/m\(^3\)/d were cropped from the figure but included in analyses (0 of 52 d at the glasshouse, 2 of 119 d at the polyethylene greenhouse). Curves were fitted by nonparametric regression (PROC GAM) and relate season to predicted conidial concentrations (conidia/m\(^3\)/d) to illustrate seasonal trends.

Conidia were not detected for the first \( \approx 20 \) d after the addition of inoculum source plants until pathogen signs were present on non-source plants. Conidia were detected from that point forward until cessation of the experiment. Thus, it appears that the detection limit of a Burkard spore sampler is greater than the number of conidia necessary to initiate an epidemic, and visual scouting for signs of the disease is more effective than spore sampling as a trigger to initiate fungicide applications.

**Discussion**

Conidia were detected for the first

- **Fig. 2.** Daily concentrations of airborne *Podosphaera xanthii* conidia and average powdery mildew severity ratings (1 = no disease, 2 = trace to 10%, 3 = 11% to 20%, 4 = 21% to 30%, 5 = 31% to 40%, 6 = 41% to 50%, 7 = 51% to 60%, 8 = 61% to 70%, 9 = 71% to 80%, and 10 = 81% to 100%, rated every week at the glasshouse and every 2 weeks at the polyethylene greenhouse) at (A) the glasshouse and (B) the polyethylene greenhouse over the course of monitoring. Conidial concentrations (dots) greater than 5000 conidia/m\(^3\)/d were cropped from the figure but included in analyses (0 of 52 d at the glasshouse, 2 of 119 d at the polyethylene greenhouse). Curves were fitted by nonparametric regression (PROC GAM) and relate season to predicted conidial concentrations (conidia/m\(^3\)/d) to illustrate seasonal trends.

Conidia of *Podosphaera xanthii* were detected by 20 d after exposure to inoculum in the glasshouse and 25 d after exposure to inoculum in the polyethylene greenhouse. For the polyethylene greenhouse at the first disease rating. By 52 and 119 d after exposure to inoculum in the glasshouse and every 2 weeks at the polyethylene greenhouse, respectively, 100% of the foliage was covered with *P. xanthii* mycelia and conidia.

For both greenhouses, a parametric linear model was sufficient to describe the increase in disease severity at each rating (every week at the glasshouse and every 2 weeks at the polyethylene greenhouse). The increase in disease severity at the glasshouse could be modeled as follows: increase = -0.91 + 0.01RH - 0.01LW, where increase is the increase in disease severity rating from one 2-week period to the next (1 to 10 visual scale), Temp is the average temperature (°C), RH is the average relative humidity (%), and LW is the number of hours of leaf wetness in the 2-week period preceding a disease rating.

**Discussion**

Conidia were not detected for the first 20 d after the addition of inoculum source plants until pathogen signs were present on non-source plants. Conidia were detected from that point forward until cessation of the experiment. Thus, it appears that the detection limit of a Burkard spore sampler is greater than the number of conidia necessary to initiate an epidemic, and visual scouting for signs of the disease is more effective than spore sampling as a trigger to initiate fungicide applications.

Hourly airborne conidial concentrations were not related with environmental conditions in the same hour as conidial sampling at either greenhouse monitored in our study. A diurnal pattern was observed for airborne conidial concentrations in both greenhouses with a peak in the morning (\( \approx 0900 \) hr) and a peak in the early afternoon (\( \approx 1500 \) hr) (Fig. 3A). Very few conidia were sampled from 2200 to 0500 hr in either greenhouse. Other studies have also found peak airborne conidial concentrations in the middle of the day for other powdery mildew pathogens including powdery mildew of hawthorn (Khairi and Preece, 1978), grapes (Willocquet and Clerjeau, 1998), cherry (Grove, 1991), apples (Sutton 1998), cherry (Grove, 1991), apples (Sutton 1998).
and previous studies (Deng and Harbaugh, 2000) have suggested that correlations between airborne conidial numbers and weather variables were weak when the circadian pattern was removed (Xu et al., 1995), in agreement with the results of our study.

Worker activity resulted in a higher probability of conidial release (1.83 to 3.59 more likely than without activity) at the glasshouse but not at the polyethylene greenhouse. Worker activity may have resulted in increased air turbulence, which has been shown to influence conidial dispersal for other powdery mildew pathogens (Hammett and Manners, 1971; Willocquet and Clerjeau, 1998). Previous work in a glasshouse also found that worker activity around diseased barley plants resulted in more new colonies of *Erysiphe graminis f. sp. hordei* (causal agent of barley powdery mildew) than when no worker activity had occurred (Frinking and Scholte, 1983). Frinking and Scholte (1983) suggested that worker activity may be more important than environmental conditions in smaller greenhouses such as the glasshouse in our study. In our study, worker activity may have influenced the daily periodicity observed for the glasshouse because the most frequent times for worker activity coincided well with peaks in airborne conidial concentrations. Future controlled studies specifically looking at the effects of worker activity on conidial dispersal and airborne conidial concentrations may be necessary to fully understand the dynamics of *P. xanthii* in a greenhouse environment. Future studies describing the dynamics of airborne spores in a greenhouse environment should carefully monitor and record worker activity, because this may be a contributing factor to spore release.

In both the glasshouse and the polyethylene greenhouse, 100% of the plants were completely covered with powdery mildew symptoms and/or pathogen signs when the disease was allowed to progress without fungicide sprays. Significant increases in disease severity were observed week to week when disease was rated weekly suggesting that more frequent scouting is necessary if fungicide sprays are to be applied before disease is established.

In the glasshouse, the increase in disease severity was positively related with relative humidity and negatively related with average temperature and the total hours of leaf wetness in the week before rating the plants for disease. A similar relationship was observed in the polyethylene greenhouse where the increase in disease severity was also positively related to average relative humidity and negatively related to leaf wetness. Disease severity was positively related to relative humidity for both greenhouses in this study; previous studies (Deng and Harbaugh, 2008; Quinn and Powell, 1982; Schnathorst, 1965; Tija, 1984) have also suggested that high relative humidity favors powdery mildew development.

**Fig. 3.** Daily periodicity of (A) airborne *Podosphaera xanthii* conidia, (B) mean temperature (°C), (C) relative humidity (RH, %), and (D) leaf wetness (0 to 15 scale) at the glasshouse and the polyethylene greenhouse. Models were fitted using nonparametric regression (PROC GAM) to relate the time of day (h) to conidial concentrations and weather variables to illustrate hourly patterns over the course of the entire experiment. (E) The number of times that a worker or workers entered the greenhouse for each time of day. Note differences in the y-axes.
development. Wide plant spacing and adequate ventilation may reduce relative humidity in the microclimate, creating a less favorable environment for disease progression.

In our study, average temperature was not significant in the model for disease increase in the polyethylene greenhouse, but it was included in the model for the glasshouse. This may have been the result of the longer interval between disease ratings (2 weeks) at the polyethylene greenhouse or because temperatures were generally warmer in the polyethylene greenhouse. Temperatures greater than 25 °C were found to negatively impact conidial release and disease development for powdery mildew of poinsettias in a greenhouse (Byrne et al., 2000). It has been previously suggested that moderate temperatures encourage development of powdery mildew on gerbera (Deng and Harbaugh, 2008).

The results of our study indicate that leaf wetness is unfavorable for the development of powdery mildew on gerbera. Management of gerbera powdery mildew should follow previous recommendations of wide plant spacing and adequate ventilation to reduce relative humidity in the microclimate (Tija, 1984). Because disease caused by Botrytis cinerea is also favored by high relative humidity and moderate temperatures (Salinas et al., 1989), these recommendations should be helpful to manage both fungal pathogens. Treatment with fungicides on a regular basis will encourage development of powdery mildew on gerbera (Deng and Harbaugh, 2008). The results of our study indicate that leaf wetness is unfavorable for the development of powdery mildew on gerbera. Management of gerbera powdery mildew should follow previous recommendations of wide plant spacing and adequate ventilation to reduce relative humidity in the microclimate (Tija, 1984). Because disease caused by Botrytis cinerea is also favored by high relative humidity and moderate temperatures (Salinas et al., 1989), these recommendations should be helpful to manage both fungal pathogens. Treatment with fungicides on a regular basis will encourage development of powdery mildew on gerbera (Deng and Harbaugh, 2008).

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**Literature Cited**


