# Relationship between concentrations of *Botrytis cinerea* conidia in air, environmental conditions, and the incidence of grey mould in strawberry flowers and fruits

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#### Abstract

Atmospheric concentrations of *Botrytis cinerea* conidia were monitored for two seasons in a strawberry crop in Moguer (Huelva, southwestern Spain). Concentrations of conidia were estimated using a Burkard volumetric spore sampler. A diurnal pattern of conidial release was observed. Airborne conidial concentration was significantly and positively correlated with the average solar radiation and mean temperature, and negatively with rainfall and relative humidity. Among the weather variables considered, solar radiation showed the most consistent results in the regression analysis, explaining over 40% of airborne conidial concentration variability. Correlation between Botrytis fruit rot incidence and accumulated number of conidia over seven days was significant and positive. Two regression models containing three variables explained over 62 and 52% of the fruit rot incidence variability. A positive but non-significant correlation was established between *B. cinerea* incidence in flowers and airborne conidial concentration or weather variables.

Abbreviations: ANOVA - analysis of variance; RH - relative humidity; SR - solar radiation

# Introduction

Strawberry (*Fragaria* × *ananassa*) is one of the most important crops in Huelva, southwestern Spain, with more than 7000 ha cultivated in 2001 (Anon., 2001). Strawberry grey mould is caused by *Botrytis cinerea* (teleomorph: *Botryotinia fuckeliana* (Faretra and Grindle, 1992). It is one of the most common diseases of the aerial parts of the plant. Under favourable conditions during flowering and harvest, yield loss can exceed 50%. The disease is often found in the field on fruits, flowers, leaves and petioles when high humidity conditions occur, and is widely found in harvested fruits (Sutton, 1998).

Botrytis cinerea can infect leaves, fruits, flowers, petioles and even crowns. Infected tis-

sues usually remain symptomless until they ripen, senesce or die (Sutton, 1990). Flowers show a typical brownish colouration in any flower part, which could eventually lead to flower death (Sutton, 1998). On ripe fruits, there are brown firm areas, spreading from the calyx to other parts of the fruit, especially in those parts in contact with already infected tissue. Over those affected areas a velvet-like mycelium grows with conidiophores formed in clusters (Maas, 1998). Under high humidity conditions, mycelia can be cottony white and conidiophores do not develop (Paulus, 1990). Finally, the affected fruit appears mummified and is covered by a powdery-like mass of mycelia and conidiophores (Hancock, 1999).

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Aerobiological studies could lead to a better understanding of the biology and epidemiology of B. cinerea, and to the development of a disease management programme to avoid unnecessary fungicide applications without increasing the risk of disease. Diseases caused by B. cinerea have been widely studied in perennial crops with June bearing cultivars (Wilcox and Seem, 1994; Xu et al., 2000). Boff et al. (2001) studied the epidemiology of B. cinerea in a spring-transplanted annual cropping system using cold-stored dormant transplants. The aim of this work was to study the relationship between environmental conditions and airborne conidial concentrations, and the relationship between these two factors and disease incidence in an autumn-transplanted annual strawberry crop with long harvest seasons (3-4 months) in south-western Europe.

#### Materials and methods

# Experimental plot

Sampling was carried out in an experimental strawberry farm located near Moguer (Huelva, southwestern Spain) for two growing seasons from October to May in 2001/02 and 2002/03, in the same plot. The soil (90% sand, 2.8% lime and 7.2% clay) had never been previously fumigated. The plot consisted of fungicide-treated subplots and untreated subplots of two cultivars (Andana and Camarosa) in a randomised complete block with four replicates. Each subplot was 1.5 m×12.5 m and had three raised beds. Treatments were foliar applications of chlorothalonil (Bravo 75PM) and fungal proteins (NBT Co. Ltd., Sevilla, Spain). Each year, during the last week of October, strawberry plants were planted. Plants were grown in an intensive annual system on dripirrigated raised beds (30 cm high and 50 cm wide) with black plastic mulch. Beds were covered with small polyethylene tunnels (microtunnels) from late November to early March for both seasons (López-Aranda and Bartual, 1999).

#### Inoculum data collection

Concentrations of airborne conidia were studied with a 7-day volumetric spore sampler (Burkard Manufacturing. Co. Ltd., Ricksmanworth, Hertfordshire, UK) located at the centre of the field, in between two microtunnels, and operated at a constant flow rate of 10 litres of air per minute. The sampling orifice was 40 cm above the beds. Melinex tape coated with petroleum jelly was used in the spore sampler. The tape was replaced weekly and cut into 48 mm long segments, each representing a single day. Each tape segment was mounted on a glass slide, with glycerine-jelly (50 ml of glycerine, 7 g of jelly, 1 g of phenol, and 42 ml of distilled water), and stained with acid-fuchsin (5 ml of glycerine-jelly solution plus 0.5 ml of acidfuchsin) (Domínguez-Vilches et al., 1992). Each tape segment was examined at 2 mm intervals in two longitudinal transepts to the direction of tape movement with a light microscope ( $\times 400$ ). Counts were corrected to compensate for the area sampled and recorded as the number of conidia per cubic meter of air sampled per day (Mediavilla-Molina et al., 1997). Daily and seven days accumulated airborne conidial concentration was calculated and compared with the disease incidence data. Botrytis cinerea conidia were identified by their morphological characteristics: size  $(8-14\times6-9 \ \mu m)$ , ellipsoid to rounded without any internal structure and with a scar on the point of union to the conidiophore (Jarvis, 1977; Maas, 1998). For this purpose we used an airborne spore and pollen atlas (Grant-Smith, 1986).

#### Collection of weather data

Meteorological data (Table 1) were obtained from an automatic weather station (Data Logger CR10X, Campbell Scientific Ltd. Leicestershire, UK; Humidity (RH) and Temperature Sensor HMP 45C, Vaisala Oyj, Helsinki, Finland; Tipping Bucket Rain gauge ARG100, Campbell Scientific Ltd.; solar radiation (SR) measured with a pyranometer SKS1110, Skye Instruments, UK). The weather station, part of the Meteorological Data Service Network (Junta de Andalucía), was located 100 m from the experimental plot, within the same farm.

# Disease data collection

All ripe fruits, considered as marketable fruits labelled from J-2 to J-3 on the Roudeillac fruit phenology scale (Roudeillac and Veschambre, 1987), were collected once a week in untreated sub-plots. Incidence of grey mould was expressed as the mean weight percentage of symptomatic fruit (Hancock, 1999). The incidence of flower infection was determined in the first six weeks every year. From each untreated plot, 10 white-bud stage flowers were randomly collected at each sampling time. The flowers were surface-sterilised with sodium hypochlorite (0.025% available chlorine, wt/vol.) for 15 min and rinsed with sterile water. Flowers were placed separately on filter paper thoroughly wetted with distilled water in plastic bags and incubated at 20 °C in the dark for 7 days. Flowers were then observed under magnification lenses to determine the presence of the pathogen. Flowers were considered colonised by B. cinerea when at least one conidiophore was observed (Xu et al., 2000).

### Statistical analysis

Whenever necessary, the transformation  $\log_{10}$  (conidial concentration + 1) was applied to data in order to reduce heterogeneity in variance (Everts and Lacy, 1990). The correlation between meteorological variables and conidial concentration was examined with Spearman's Correlation Test (Statistix 7, Analytical Software Ltd., La Jolla, CA, USA), which is a general test used for micro-organism infection data (Díaz et al., 1997). Simple regression analysis was used to quantify the relationship between airborne conidial concentration [log<sub>10</sub> (conidia concentration + 1)] and weather variables (Table 1).

Analysis of variance (ANOVA) was performed for Botrytis fruit rot and flower infection, using Statistix 7. Regression analysis was used to quantify the relationship between Botrytis fruit rot, weather variables and concentration of airborne conidia. The arcsin-transformed percentage of infected fruits was the dependent variable and various weather variables (Table 1), calculated as the

Table 1. Weather variables\* used in regression analysis

Р	=	daily	prec	cipita	ation	(m	ım)	

AP = accumulated precipitation (mm)

- RD = number of rainy days
- $T_{max}$  = average maximum daily temperature (°C)  $T_{min}$  = average minimum daily temperature (°C)
- $T_{min}$  = average mean daily temperature (°C)
- $RH_{mn}$  = average mean daily relative humidity (%)
- $SR = average solar radiation MJ m day^{-2}$

weekly mean were the independent variables in the analysis. The correlation between disease incidence and the accumulated number of airborne conidia captured during the previous 7 days (accumulated number of conidia) was examined using Spearmańs Correlation Test. Simple regression analysis was used to determine the relationship between the arcsin-transformed percentage of infected fruits and the accumulated number of conidia.

Regression analysis was used to quantify the relationship between incidence of flower infection, weather variables (Table 1) and concentration of airborne conidia. The arcsin-transformed percentage of infected flowers was the dependent variable and various weather variables, measured 24 h prior to flower collection, were the independent variables in the analysis. A simple regression analysis was used to determine the relationship between the arcsin-transformed percentage of infected flower and airborne conidial concentration  $[\log_{10} (\text{conidia concentration } + 1)].$ 

#### Results

# Seasonal periodicity and influence of meteorological factors

In both years, conidia were first detected early in October and their number increased as the crop matured (Figures 1 and 2). During the 2001–2002 season, the concentration of conidia in sampled air ranged from 0 to  $2.0 \times 10^3$  conidia m<sup>-3</sup> until the first day of harvest (February 13th, 2002). Conidial concentration over  $5.0 \times 10^2$  conidia m<sup>-3</sup> was registered on 34 non-consecutive days (Figure 1a). During these days, the average temperature was 12.6 °C (Figure 1d), average RH<sub>mn</sub> was 76.3% (Figure 1c), and no rainfall over 5 mm occurred (Figure 1b). During the harvest period (from February 13th to May 20th), the concentration of conidia ranged from 0 to  $2.6 \times 10^4$  conidia m<sup>-3</sup> (Figure 1a). Conidial concentration over  $3.0 \times 10^3$ conidia m<sup>-3</sup> was registered on 23 non-consecutive days (Figure 1a). During these days, the average temperature was 18.8 °C (Figure 1d), average  $RH_{mn}$  was 69.41% (Figure 1c) and rainfall >5 mm occurred on one day (Figure 1b). Minimum concentration of conidia (zero) was recorded on three non-consecutive days during the season. For these days, the average temperature was 11.8 °C



*Figure 1.* Daily total airborne conidia of *Botrytis cinerea* (a), rainfall (b), mean daily relative humidity (c) and mean air temperature (d) at the Moguer experimental strawberry plot during 2001–2002 season. Concentrations of airborne conidia were determined with a Burkard volumetric spore sampler operated continuously. Arrows indicate dates of planting and beginning of harvest.



*Figure 2.* Daily total airborne conidia of *Botrytis cinerea* (a), rainfall (b), mean daily relative humidity (c) and mean air temperature (d) at the Moguer experimental strawberry plot during 2002–2003 season. Concentrations of airborne conidia were determined with a Burkard volumetric spore sampler operated continuously. Arrows indicate dates of planting and beginning of harvest.

(Figures 1a and 1d), average  $RH_{mn}$  was 70.3% (Figure 1a, c) and rainfall was > 5 mm on only one out of the three days (Figure 1a, b). The highest concentration of conidia (2.6 × 10<sup>4</sup> conidia m<sup>-3</sup>) occurred on a day with an average temperature of 21.5 °C, 55% RH<sub>mn</sub> and no rainfall (Figure 1).

During the 2002–2003 season, the concentration of conidia ranged from 0 to  $2.8 \times 10^3$  conidia m<sup>-3</sup> until the first day of harvest (February 6th, 2003). Conidial concentration over  $5.0 \times 10^3$  conidia m<sup>-3</sup> was registered on 47 non-consecutive days (Figure 2a). During this period, the average temperature was 12.9 °C (Figure 2d), average RH<sub>mn</sub> was 81.0% (Figure 2c) and rainfall, <5 mm, was registered on six of these days (Figure 2b). During the harvest period (from February 6th to May 19th) concentration of conidia varied between 0 and  $1.3 \times 10^5$  conidia m<sup>-3</sup> (Figure 2a). Conidial concentration over  $3.0 \times 10^3$  conidia m<sup>-3</sup> was registered on 51 non-consecutive days (Figure 2a). During these days, the average temperature was 15.9 °C (Figure 2d), average RH<sub>mn</sub> was 74.95% (Figure 2c) and rainfall > 5 mm occurred on 5 days (Figure 2b). Minimum concentration of conidia (zero) was recorded on four non-consecutive days during the season. For these days, the average temperature was 14.2 °C (Figure 2a, d), average RH<sub>mn</sub> was 86.5% (Figure 2a, c) and rainfall, > 5 mm, occurred on only one out of the four days (Figure 2a, b). The highest concentration of conidia  $(1.3 \times 10^5 \text{ conidia m}^{-3})$  was on a day with mean temperature of 16.2 °C, 75% RH<sub>mn</sub> and no rainfall (Figure 2).

There were significant positive correlations between  $\log_{10}$  (daily conidial concentration +1) and SR, and the average temperature on the previous day (t-1) and on the same day in both years. There were significant negative correlations between  $\log_{10}$  (daily conidial concentration + 1) and RH<sub>mn</sub> in both years, and with rainfall, only in the first year. Weather variables explained 42% and 54% of the variability in the number of B cinerea conidia trapped during the first and second season respectively. In both years,  $\log_{10}$  (daily conidial concentration + 1) was positively correlated with SR, and the average temperature recorded 3, 7 and 10 days previously (Table 2), but it was not consistently correlated with rainfall and RH<sub>mn</sub> for the same delayed periods (data not shown).

The equations derived from multiple stepwise regressions relating  $\log_{10}$  (daily conidial concen-

tration + 1) to weather variables varied for both years (data not shown). The simple regression analysis between the transformed concentration of conidia and weather variables was consistent over the two years (Table 3). Average SR on the same day explained 39.6% and 42.5% of the variability for the first and second season respectively. Correlation coefficients indicated a moderately strong relationship between these two variables.

#### Diurnal periodicity

The incidence of airborne conidia trapped generally followed a diurnal pattern (Figure 3). Conidial concentration was greatest between 8:00 h and 15:00 h during the 2001–2002 season, with a peak between 9:00 and 11:00 h. During the second season, most conidia were trapped between 6:00 and 17:00 h, with a peak between 12:00 and 14:00 h.

#### Disease incidence on fruit

Results of ANOVA on Botrytis fruit rot incidence showed that the effect of cultivar was not significant. Therefore, the mean was calculated for the

*Table 2.* Spearman's rank correlation between number of daily conidia trapped<sup>a</sup> in the air and meteorological variables on untreated subplots during the 2001–2002 and 2002–2003 season in an experimental plot in Moguer (Huelva, southwestern Spain)

Weather variables <sup>b</sup>	Season <sup>c</sup>			
	2001–2002	2002-2003		
T <sub>mn</sub>	0.49 ***	0.43 ***		
SR	0.63 ***	0.58 ***		
RH <sub>mn</sub>	-0.29 ***	-0.28 ***		
Rainfall	-0.16 *	-0.10 NS		
$T_{mn t} = 1$	0.50 ***	0.41 ***		
$SR_{t-1}$	0.57 ***	0.59 ***		
$T_{mn, \ell} = 3$	0.41 ***	0.36 ***		
$SR_{t-3}$	0.51 ***	0.53 ***		
$T_{mn}$ / $-7$	0.23 **	0.34 ***		
$SR_{t-7}$	0.43 ***	0.56 ***		
$T_{mn t} - 10$	0.23 **	0.27 ***		
$SR_{t-10}$	0.43 ***	0.56***		

<sup>a</sup>Transformation:  $\log_{10}$  (daily concentration + 1).

<sup>&</sup>lt;sup>b</sup>Weather variables determined on the current day of the capture, and recorded 1 (t-1), 3 (t-3), 7 (t-7), and 10 (t-10) previous days.

<sup>&</sup>lt;sup>c</sup>Significance levels: Non-significant at P > 0.05 (NS), significant at P = 0.05 (\*), significant at P = 0.01 (\*\*), significant at P = 0.001 (\*\*\*).

*Table 3.* Simple regression analysis of the airborne conidia of *B. cinerea* as a function of average solar radiation<sup>a</sup>

Term	Parameter	2001-2002	2002–2003
Constant	b	$2.09 \pm 0.07$	$2.19 \pm 0.08$
SR Variation accounted (%)	а	$0.051 \pm 0.004$ 39.6	$0.062 \pm 0.005$ 42.5

<sup>a</sup>The log of daily conidia concentration was a function average solar radiation, where *a* and *b* are parameters, therefore  $\log_{10}$  (daily conidia concentration + 1) =  $a \cdot SR + b$ .

two cultivars. During the first season, disease incidence was low from March 20th to March 26th, although the concentration of conidia was high (Figure 4a). For these days, the average temperature was between 13 and 20.8 °C, RH<sub>mn</sub> was between 56 and 79% and there was no rainfall. During the second season, incidence of Botrytis fruit rot was high (36%) from April 17th to April 24th (Figure 4b). During this period, the mean temperature was >15 °C, RH<sub>mn</sub> was >80% and for 43% of these days rainfall was >0.5 mm. Maximum concentration of conidia was reached one week later, with a mean temperature 16.4 °C, mean RH 77%, and one day of rainfall >0.5 mm.

The correlation between disease incidence on fruit and the accumulated number of conidia was positive and significant (P = 0.01 for both seasons).



*Figure 3*. Diurnal periodicity of *Botrytis cinerea* airborne conidia trapped from the air over a strawberry experimental plot in Moguer, Spain. Values given are the average of conidia trapped at the same hour of the day during the season 2001– 2002 and 2002–2003. Concentrations of airborne conidia were determined with a Burkard volumetric spore sampler operated continuously.

Simple regression analysis between the arcsintransformed percentage of infected fruits and accumulated number of conidia indicated a significant relationship between them (P < 0.01). Equations derived from the analysis are shown in Table 4.

Multiple stepwise regression conducted between arcsin-transformed percentage of infected fruits and weather variables resulted in two models containing three variables: average SR, average RH<sub>mn</sub> and average maximum temperature (Table 5). Model 1 included data from the first season (2001/02) and Model 2 was for both seasons (2001/02 and 2002/03).

# Incidence of B. cinerea on flowers

ANOVA results showed that the effect of cultivar on B. cinerea incidence was not significant. Therefore, the mean was calculated for the two cultivars. During the first year, the highest incidence of B. cinerea on flowers (10.9%) was reached when conidia also reached a high level  $(2 \times 10^3 \text{ conidia m}^{-3})$  (Figure 5a). For this day average temperature was 10.4 °C, RH<sub>mn</sub> was >80% and no rainfall occurred. There was a positive but non-significant correlation between B. cinerea incidence on flowers and the airborne conidial concentration (P > 0.10). Weather variables accounted for about 80% of the total variation in the incidence of B. cinerea on flowers. Multiple stepwise regression resulted in the best model that contained two variables (T<sub>mn</sub> and RH<sub>mn</sub>), but this model was not statistically significant (P > 0.10).

During the second season, low incidence (1.7%)was reached when conidial concentration reached the highest level  $(3.6 \times 10^3 \text{ conidia m}^{-3})$  (Figure 5b). On this day the average temperature was 12.8 °C,  $RH_{mn}$  was >78% and no rainfall occurred. The correlation between the B. cinerea incidence on flowers and the airborne conidial concentration was negative and non-significant. Simple regression analysis showed that there was no significant relationship between daily concentration of conidia and incidence of B. cinerea on flowers (P > 0.10). Weather variables accounted for about 80% of the total variation in the incidence of B. cinerea on flowers. Multiple stepwise regression indicated that the best model, containing two variables (SR and RH<sub>mn</sub>), was not



*Figure 4.* Weekly incidence of Botrytis fruit rot in an experimental plot of strawberry plants in Moguer during 2001–2002 (a) and 2002–2003 (b) seasons. Disease incidence was estimated as the average percentage of rot fruit produced in untreated subplots. Conidia of *Botrytis cinerea* in the air are given as accumulated number of conidia over seven days trapped in the plot by a Burkard volumetric spore sampler operated continuously. Vertical bars represent standard errors.

significant (P > 0.10). It was not possible to fit a consistent regression model to relate flower incidence to conidial concentration or weather variables.

#### Discussion

Conidia were trapped over the crop during all of the season but appeared to increase after the initiation of harvest in both years. Conidial concentration in the air over the crop was closely related to the environmental conditions on the same day and also on previous days. However, negative but not consistent correlations were found between rainfall and conidial concentration. Raindrops apparently washed conidia from the air (Paulus, 1990), so when rainfall occurred, the concentration of conidia in the air was reduced.

Mean temperature and solar radiation were positively correlated with conidial concentration up to ten days previous to the capture. Sosa-Alvarez et al. (1995) reported that a large number of spores are produced under a temperature near 15–22 °C. In our study, the conidial peak period corresponded with periods of temperature

*Table 4.* Simple regression analysis of the incidence of Botrytis fruit rot as a function of inoculum (accumulated number of spores captured for the 7 previous days to the corresponding dates)<sup>a</sup>

Term	Parameter	2001–2002 + 2002–2003	2002–2003
Constant Inoculum	b a	$\begin{array}{r} 0.067 \ \pm \ 0.03 \\ 2.37 \cdot 10^{-6} \\ \pm 4.95 \cdot 10^{-7} \end{array}$	$\begin{array}{c} 0.085 \pm 0.04 \\ 2.2 \cdot 10^{-6} \\ \pm 5.4 \cdot 10^{-7} \end{array}$
Variation accounted (%)		48.0	53.9

<sup>a</sup>The arcsin-transformed percentage of Botrytis fruit rot was a function of inoculum, where *a* and *b* are parameters, therefore arcsin [SQRT (percentage of infected fruits /100)] =  $a \cdot \text{accumulated number of conidia} + b$ 

*Table 5.* Multiple regression analysis of the incidence of Botrytis fruit rot as a function of weather variables<sup>a</sup>

Term	Parameter	Model 1 (2001–2002)	Model 2 (2001–2002 + 2002–2003)
Constant	d	$-0.67 \pm 0.55$	$-1.35 \pm 0.44$
SR	а	$0.05\pm0.01$	$0.05\pm0.01$
RH <sub>mn</sub>	b	$0.01\pm0.005$	$0.02\pm0.004$
T <sub>max</sub>	с	$-0.04\pm0.01$	$-0.03\pm0.01$
Variation accounted (%)		61.5	52.3

<sup>a</sup>The arcsin-transformed percentage of Botrytis fruit rot was a function of weather variables determined as the average of seven days previous to the sampling data, where *a*, *b*, *c* and *d* are parameters, therefore arcsin [SQRT (percentage of infected fruits /100)] =  $a \cdot \text{SR} + b \cdot \text{RH} + c \cdot \text{T}_{\text{max}} + d$ .

>15 °C. Solar radiation consistently accounted for a high percentage of the conidial concentration variability. The importance of light on the formation and maturation of *B. cinerea* conidia, mainly through cellular compounds such as mycosporine, has been described by Cooley-Smith et al. (1980) and Tan and Epton (1974).

*Botrytis cinerea* conidia have an aerial dispersion. Two phases can be distinguished: firstly there is a liberation of conidia from the conidiophore mediated by a hygroscopic mechanism that controls the turgidity of the conidiophore, and secondly, there is a transport episode mediated by physical agents such as wind and rain drops (Jarvis, 1980). Therefore, as observed, conidial liberation often happens early in the morning when RH changes, and as the temperature rises. Minimal liberation during the night permits recovery for the next day.

The establishment of grey mould varies with the meteorological variables (Jarvis, 1980; Xu et al., 2000). For *B. cinerea* on strawberry, the optimal temperature range for germination is 15-25 °C, with RH requirements >90% for long periods of time (Wilcox and Seem, 1994; Xu et al., 2000). Although the infection process of *B. cinerea* needs a high RH, these conditions are often associated with rainy periods when conidia are washed out. Botrytis fruit rot is positively correlated with rainfall (Jarvis, 1964; Wilcox and Seem, 1994). However, we have not observed a consistent relationship between rainfall and fruit rot, not even when the number of rainy days was considered. SR, mean RH and average temperature measured up to 7 days before sampling date explained between 52 and 62% of fruit rot variability.

Boff et al. (2001), applied Bulger's models (Bulger et al., 1987) to predict fruit rots, and observed a lack of correlation between observed and predicted data. This could be due to the fluctuations of inoculum over the crop under field conditions. Although other authors had reported that infections of intact fruit from airborne conidia rarely occur under field conditions (Boff et al., 2003; Jarvis, 1962), our results suggested otherwise. The positive correlation between conidia concentration in the air and Botrytis fruit rot incidence indicates that the presence of higher conidia levels leads to a higher incidence of the disease.

The symptomatic infection of flowers could lead to the development of flower rot. Meanwhile, the symptomless infection of these tissues could lead to fruit rot, although the incidence of fruit infection was shown to have a relationship with the incidence of petal necrosis (Bulger et al., 1987). Studies conducted on annual crops of strawberry to assess the importance of petals as an inoculum source of grey mould proved that the retention of petals until harvest greatly enhanced the incidence of grey mould (Boff et al., 2003). Strawberry cultivars used in our study naturally dropped petals by the end of flowering, and petals were rarely found on green or ripe fruit. It is likely that in south-western Spain where cvs Camarosa and Andana are widely cultivated, infected petals could not be considered as an important source of inoculum for fruit infection by B. cinerea.

Xu et al. (2000) described relationships between *B. cinerea* flower infection incidence, weather conditions and inoculum concentration and



*Figure 5*. Weekly incidence of *Botrytis cinerea* on strawberry flowers, in an experimental plot in Moguer during 2001–2002 (a) and 2002–2003 (b) seasons. Disease incidence was estimated as the mean percentage of infected flowers among ten flowers collected from each untreated subplot. Conidia of *Botrytis cinerea* in the air are given as the daily average of conidia trapped in the plot by a Burkard volumetric spore sampler operated continuously. Vertical bars represent standard errors.

developed models that satisfactorily predicted infection incidence. In our study, there were no significant correlations between conidial concentration and incidence of *B. cinerea* on flowers, and between disease incidence and weather variables. This is probably due to the use of everbearing strawberry plants.

Many efforts have been made to develop disease prediction models for *B. cinerea*. Recently, BOT-CAST (BOTRYTIS FORECAST) and BOTMAN (BOTRYTIS MANAGEMENT) were developed based on meteorological data (Shtienberg and Elad, 1997). Miller and Wagoner (1957) and Jarvis (1962) studied the presence of *Botrytis cinerea* conidia on strawberry crops and its relation to grey mould incidence. Boff et al. (2001) reported that harvest fruit rot incidence was not correlated with the incidence of *B. cinerea* on flowers. Our observations suggest that in annual strawberry cropping systems, the incidence of Botrytis fruit rot is related to environmental conditions, temperature, RH, SR and the concentration of inoculum in the air.

Based on the results obtained, weather and inoculum data collected from several locations in

the growing area will be used to determine the critical threshold for each variable considered which may lead to high disease levels. It will provide strawberry growers with a valuable tool to achieve a better-integrated management of the crop (Montesinos et al., 1995; Llorente et al., 2000).

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