Effects of Growth Conditions on Postharvest *Botrytis* Infection in Gerbera - a Nursery Comparison

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Abstract

***Botrytis cinerea* is one of the main postharvest problems in gerbera cut flowers. There are clear differences among growers in the percentage of flowers showing symptoms of *Botrytis* infection after harvest. Because the factors causing these differences are uncertain, cultivation parameters of twelve different growers were followed and a nursery comparison was carried out. Gerbera flowers 'Dino' were sampled twelve times; six times in autumn 2006 and six times in spring of 2007. At the nurseries climate (air temperature, relative humidity, light intensity and CO_2 concentration) was logged, and plant density, plant age and other characteristics were monitored. Before each harvest *Botrytis* spores were trapped in the greenhouses and counted. After sampling, flowers were treated according to a standard transport simulation. Thereafter *Botrytis* infection was monitored, and statistical analyses were performed on the effect of the pre-harvest conditions on *Botrytis* infection in the postharvest stage. Clear influence of the spore level in the greenhouse was observed. When a sufficient number of spores were present in the greenhouse environment, the humidity level and the intensity and duration of irradiation had the strongest influence on infection. In general, factors those lead to a dry microclimate such as the use of ventilators, the use of supplemental lighting and a low plant density were related with a lower number of lesions on gerbera petals in the postharvest phase.**

INTRODUCTION

Infection with *Botrytis cinerea* results in severe losses during distribution of gerbera flowers. Economical loss due to visible infection of gerbera cut flowers at Dutch auctions was estimated at € 207.000,- in 2002 (Vrind, 2005), and total economical loss in the postharvest chain was estimated € 2.5 million in 2007 (Marcelis, unpublished). One of the major reasons for such a high loss is that gerbera cut flowers are commonly used in mixed flower arrangements. In those mixes gerbera is often the first flower that shows damages and this can easily lead to a turn down of the complete flower arrangement.

Besides large differences in susceptibility of varieties, clear differences in infection and damaged flowers from different nurseries were found (Vrind, 2005). This may indicate that growth conditions play an important role in the occurrence of postharvest damage by *Botrytis*. Since it is not certain which factors during cultivation are causing these differences of infection among different growers, a nursery comparison was performed. This procedure proved to be a good tool to determine the correlation between postharvest quality and growth conditions (Marissen and Benninga, 2001; Slootweg, 2005).

MATERIALS AND METHODS

Twelve growers of the cut flower *Gerbera jamesonii* H. Bolus 'Dino' located in the western part of the Netherlands were randomly selected for nursery comparison. During twelve weeks (six weeks in autumn 2006 and 6 weeks in spring 2007) 20 flowers were sampled weekly from a plot of ca. 40 m^2 randomly assigned in each greenhouse. After harvesting the flower stems from the plant, the stems were directly placed in tap

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water and transported to the laboratory. All stems were stored for 24 hours at 8°C, 80% RH in water with Chrysal CVB (Chrysal, Naarden, the Netherlands) to prevent bacterial growth. After storage, flowers were put in closed cardboard boxes for transport simulation at 8°C, 80% RH, for 4 days. After transport simulation, the flowers were placed in vases with tap water (1 stem/vase of 1 L) in a climate room at 20°C, 60% RH, 12 h light/day of fluorescent tubes (TL 84) with a light intensity of 14 $\mu mol.m^{-2}.s^{-1}$. The number of flowers with *Botrytis* lesions was counted next day.

In the greenhouse, within the sampling plot climate (temperature, relative air humidity, photosynthetic active radiation (PAR), and CO_2) at the position of the flowers was continuously measured with calibrated sensors and logged in 5 minute averages in a data logger (CaTec, Delft, the Netherlands). Next to climate other cultivation parameters such as planting density, plant age, intensity and duration of the use of supplemental lighting, use of mechanical ventilators, type of substrate, substrate water content and nutrient content of drain water were registered.

To measure the number of spores that depose on the open flowers in the sampling plot, four open Petri dishes filled with a *Botrytis* selective medium (Kerssies, 1990) were horizontally placed at flower height 24 h before each harvest. After harvest, the dishes were closed and stored at 21°C in the dark. *Botrytis* colonies were counted after one week and spore counts were classified (1= less than 10, 2= 10-50, 3= 51-100 and 4= more than 100 colonies/Petri dish).

To determine which factors affected *Botrytis* infection of the flowers after transport simulation, data were analysed with multivariate and univariate statistical procedures mainly using the PROC FACTOR and the PROC GLM procedures of the statistical software package SAS (ver 9.1.4; Cary, NC, USA).

RESULTS

Large differences in percentage flowers with lesions between the 12 nurseries were found (Fig. 1) and those differences were the same in both autumn 2006 and spring 2007 (R^2 of 11 nurseries 0.89). This suggests that there is a clear nursery effect on *Botrytis* damage in postharvest. Mean spore pressure per nursery is shown in Figure 2. Spore pressure has a direct influence on lesions when the conditions for lesion formation (i.e. high humidity) is optimal. Such that there is lesion formation on nurseries with high spore pressure and high humidity levels, whereas nurseries with high humidity but with a low spore pressure had only few lesions (Fig. 3). However, only at very low spore pressure levels ($\leq$2; i.e. less than 51 spores trapped in 24 h/Petri dish), the percentage of *Botrytis* infection after transport simulation was also low. Our measurements showed that low spore pressure could be attained when air humidity around the plants was kept low. This could either be achieved by keeping the average vapour pressure deficit (VPD) of the air high, e.g. above 0.5 kPa one week before harvest during at least 20% of the time, decreasing the spore pressure by 50% (Fig. 4). A low atmospheric vapour deficit of the greenhouse air (VPD) from time of flower opening to harvest lead to a high risk on *Botrytis* infection as shown in Figure 5. However, the humidity conditions during night time had a stronger effect than at day time; the incidence of *Botrytis* lesions in postharvest doubled when the total duration of the nighttime VPD <0.3 kPa increased from 50 to 75% during the week before harvest (Fig. 5).

A light sum of less than 50 MJ/m^2 in the last 3 days before harvest, doubled the *Botrytis* incidence compared to a light sum of more than 90 MJ/m^2 in the same period. Besides the effect of the total light sum, there was also an effect of the use of supplemental lighting. The use of supplemental light with an intensity of more than 75 $\mu mol.m^2s^{-1}$ lead to three times less flowers with *Botrytis* damage than the use of light with less than 40 $\mu mol.m^2.s^{-1}$.

A higher plant density in the greenhouse showed more postharvest *Botrytis* damage, than a lower plant density. The correlation of *Botrytis* versus plant density was 43%. There was no correlation between plant age and postharvest *Botrytis*. The use of ventilators in the greenhouse reduced postharvest *Botrytis* infection by 50%.

DISCUSSION

Our results show that the incidence of *Botrytis* in the postharvest stage of gerbera flowers is strongly connected to the cultivation procedures, such that high or low incidences of *Botrytis* damages are special to certain growers. We showed that cultivation conditions affect both the production of the spores by the mycelium (located in the older leaves of the crop) as well as on the start of the secondary disease cycle: the deposition, adhesion, germination of the spores and the penetration of the germ tube into the tissue and thus the start of lesion growth. Although the effect of the number of spores in the greenhouse on the later postharvest *Botrytis* damage of flowers was limited, we found that a very low spore pressure strongly prevented damage, i.e. in greenhouses with very few spores present, a low VPD did not lead to lesions (i.e. no spores means no problems when air humidity is high). However, on the other hand low VPD conditions in the greenhouse can lead to more spores. An other experiment of similar set up (Wageningen UR Greenhouse Horticulture, unpublished) with specific measurements of the microclimate suggested that using a more directed climate control regime having a dry microclimate in the leaf canopy, results in a low spore pressure. In these conditions no damage can occur, unrespectable the humidity or light conditions around the flowers (that are some 30-40 cm above the leaves). The observed higher levels of infection at high planting densities and not using ventilators were probably indirect effects and might also have been an effect of high humidity in the microclimate. However, a high air velocity may have a negative effect when spores are present, i.e. when the ventilator effect on leaf drying is not successful and thus spores are being produced, the additional wind speed by the ventilators may support spreading the spores. This is probably the reason that Hammer and Evensen (1996) found a negative effect of the use of ventilators in the greenhouse.

Our data also show that light has an effect on postharvest *Botrytis* damage, such that lower light levels during cultivation lead to a higher incidence of *Botrytis*. This could be explained by the decreased spore vitality at high light levels as described by Kerssies (1994) or through the increased susceptibility of tissue grown under low light conditions due to a softer tissue structure. Usually *Botrytis* does not invade healthy green tissue unless an injured or dead area is present, where leaves in the lower canopy are often infected and then the fungus can spread (Körner and Challa, 2003). However, the effect of high air humidity can be stronger or weaker depending on the specific susceptibility of the tissue as the fungus can easily invade soft flower petals (Marois et al., 1988). During daytime there are many more spores present in the greenhouse (Kerssies, 1994; Hausbeck and Pennypacker, 1991) and a dry microclimate around the flower petals can then prevent adhesion and germination (van Kan, 2005). During night humidity in greenhouses is generally higher than during day time and with very high humidity conditions or even free water on the leaves, the fungus can attack the leaf tissue through tiny wounds.

The presented study can be used to create an early warning system for postharvest *Botrytis* damage in gerbera. The grower can actively control the cultivation process and decrease the risk for *Botrytis* damage in postharvest and therefore decrease the economical losses in the gerbera chain.

ACKNOWLEDGEMENTS

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Figures

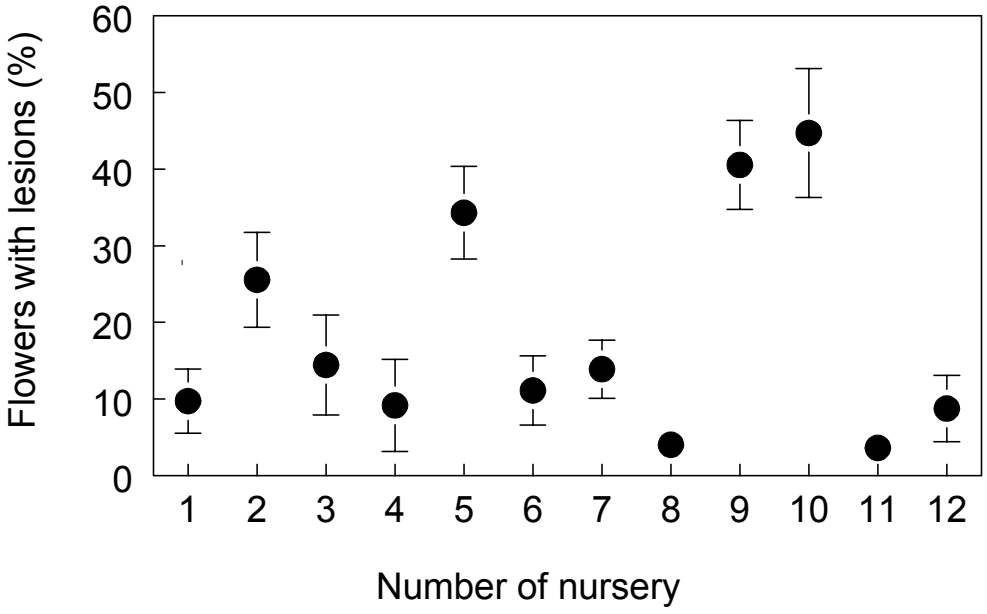

Fig. 1. Percentage of flowers with one or more lesions after transport simulation (including one day of vase life). Mean values of 12 tests per nursery; vertical bars indicate the standard error of the mean (SEM).

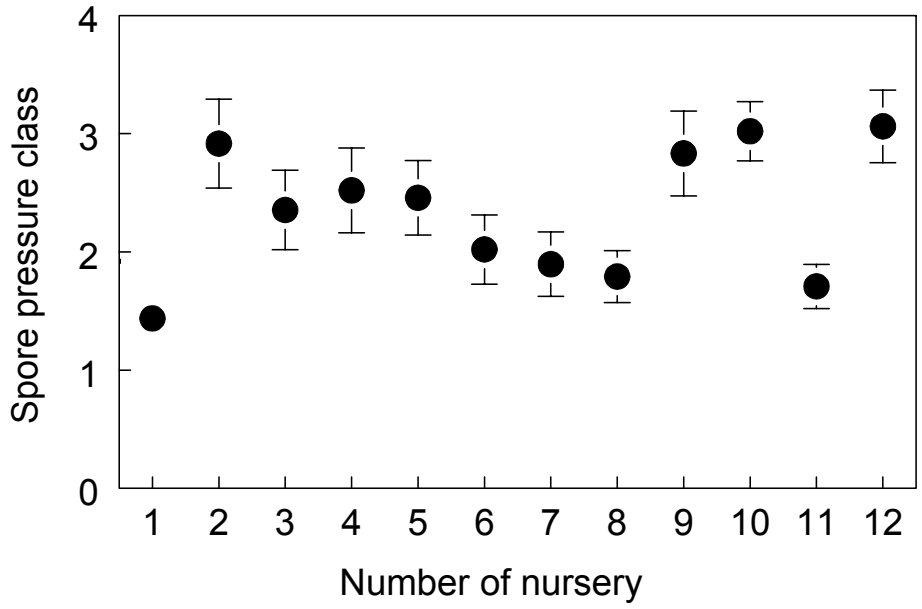

Fig. 2. Mean spore pressure per nursery. Spore pressure class 1= less than 10, 2= 10-50, 3= 50-100 and 4= more than 100 colonies per Petri dish; vertical bars indicate the standard error of the mean (SEM).

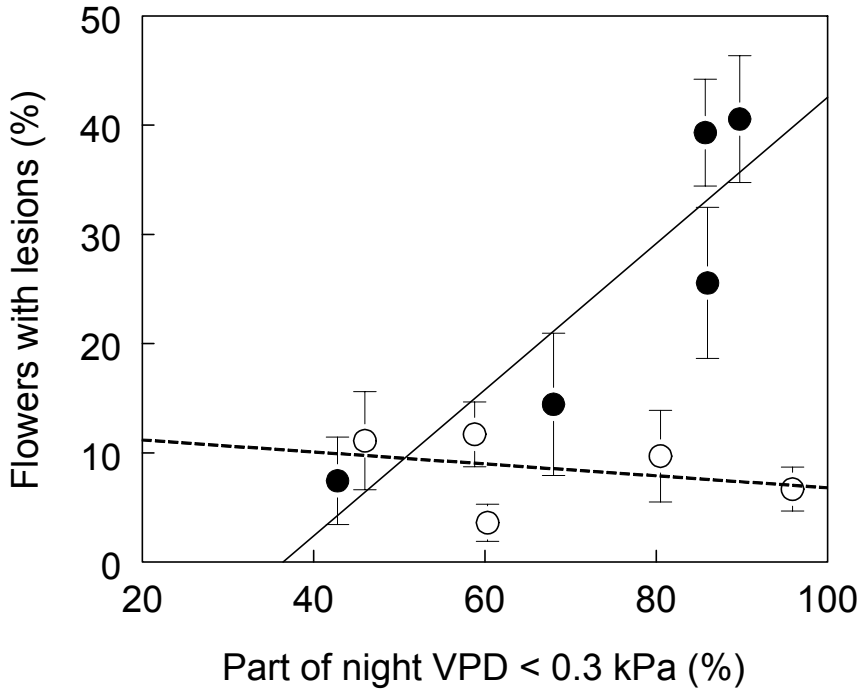

Fig. 3. Flowers with lesions for separate nurseries as a result of the mean night time vapour pressure deficit of the air (VPD, kPa) during the last week before harvest for the 5 nurseries with highest spore pressure (●) or the 5 nurseries with lowest spore pressure (○); lines are regressions, vertical bars indicate the standard error of the mean (SEM).

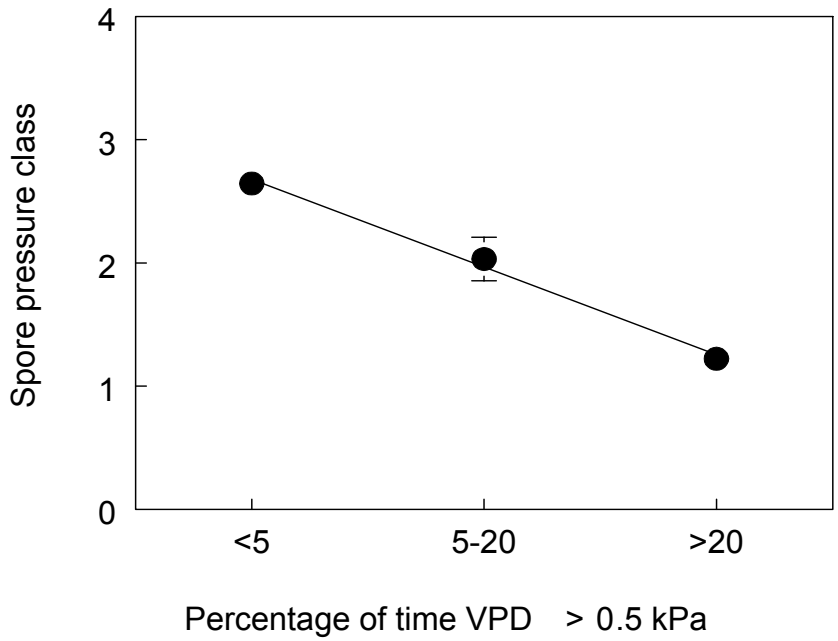

Fig. 4. Spore pressure in the greenhouse as a result of the atmospheric vapour pressure deficit (VPD) during the last week before harvest. Spore pressure class 1= less than 10, 2= 10-50, 3= 51-100 and 4= more than 100 colonies per Petri dish; vertical bars indicate the standard error of the mean (SEM).

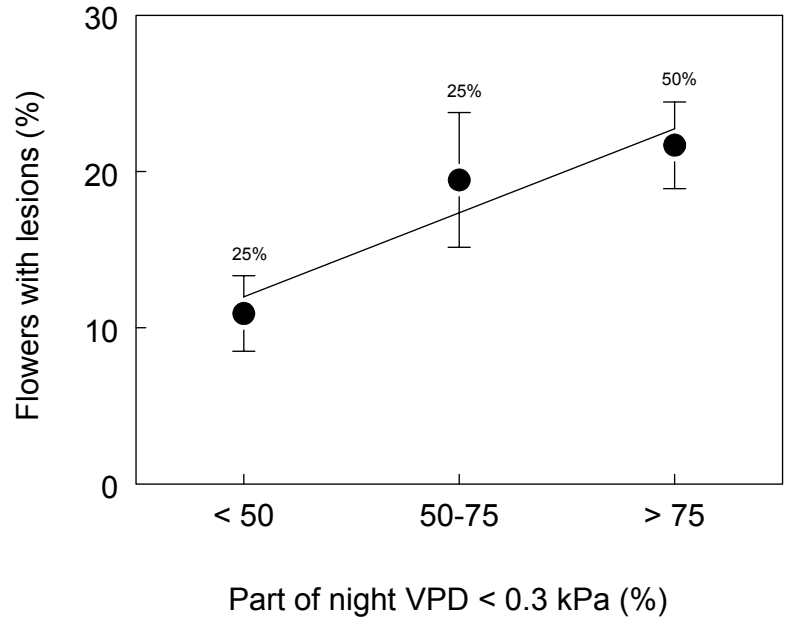

Fig. 5. Percentage of flowers with lesions as a result of the night time vapour pressure deficit of the air (VPD, kPa) during the last week before harvest. Percentages in the graph indicate the data points used (from the total of 144: 12 growers, 12 weeks); vertical bars indicate the standard error of the mean (SEM).