

# Validating spray timing for *Cercospora* spot

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## ABSTRACT

*Cercospora* spot of avocados, caused by *Pseudocercospora purpurea*, is one of the most severe pre-harvest fungal diseases in South Africa. A forecasting model, developed in the early 1980s, is currently used in the South African avocado industry to predict when spore release occurs as well as the optimum time for the first spray. Due to climate change, it is however crucial to re-evaluate this model. The aim of this study was to determine if the current prediction model is still valid for forecasting the first spray for effective control. The second aim was to refine the existing model by considering the effect of humidity and/or leaf wetness as determining factors for spore release. In the 2017/18 season, spore traps were positioned in an unsprayed 'Fuerte' orchard at both Halls and Agricultural Research Council – Tropical and Subtropical Crops (ARC-TSC) (Nelspruit area) and in the 2018/19 season only in the ARC-TSC orchard. Spore traps consisted of petroleum jelly-coated slides held both horizontally and vertically. Slides were changed and examined hourly, daily and weekly. To determine the critical infection periods, a bagging trial was carried out at the Halls site (2017/18 season) and at the ARC-TSC site (2018/19 season). Fruit were covered with paper bags at the beginning of the season before fruit became susceptible. Thirty to fifty bags were removed and replaced every fortnight in both seasons to allow infection. At harvest, fruit were evaluated for the presence of *Cercospora* spot. Weather data were correlated with spore trapping and disease index data in order to develop a forecasting model. A very good correlation was found between weekly conidia trapped and weekly rainfall. Disease index values showed highest correlation with maximum humidity. No significant correlations were found with any other weather parameter. Several new models were developed. Models need to be used in combination with monitoring of fruit size to determine correct timing of first sprays. Models will be further refined with the inclusion of the 2018/19 season's data. The best model developed will be used to determine high-risk infection periods, enabling accurate and more cost-effective timing of fungicide sprays.

## INTRODUCTION

From a quality point of view *Cercospora* spot or black spot is the most important avocado fruit disease. Although not particularly affecting the flesh, it can cause losses of up to 70% on unsprayed trees (Darvas and Kotze, 1987). According to the SAAGA Loss Factor Benchmark Report of 2017, the total loss across the industry for fruit delivered at the pack house due to *Cercospora* spot was 3% and for the 'Fuerte' cultivar specifically, 9%.

The first spray of the season during a normal rainfall year is the most important spray. The accurate timing of this spray and good coverage of fruit will determine how successful control will be for that particular season. Various factors, such as temperature, rainfall and fruit size, play a role in determining when the first spray should commence. Control is therefore most effective when weather conditions which affect spore release are monitored. A fix spray programme is not recommended.

In the early 1980s, Dr Darvas developed a predictive model to forecast the number of conidia in a

given area (Z value) as well as the optimum timing of the first spray (Darvas, 1982). The current model is  $Z = -58.99 + 3.22x(\text{Mean weekly temp.}) + 0.18y(\text{weekly rainfall in mm})$ . If Z is  $\geq 0$ , spore release occurs and if  $Z \geq 20$ , the potential for infection is high. According to the Subtrop orchard management schedule for 'Fuerte', first copper sprays should commence when fruit are  $\leq 2.5$  cm in diameter or  $Z \leq 15$  (Campbell, 2016).

Due to climate change it is necessary to re-investigate the existing models. New models need to be developed and possibly include more weather parameters such as humidity/leaf wetness. Lately, extremely high temperatures resulted in a Z value  $\geq 15$  without any rain, resulting in incorrect prediction of spore release and inaccurate timing of sprays.

## OBJECTIVE

The objective of the study was to determine if the equation developed by Darvas (1982) is still a valid model for forecasting spore release and the timing of the first spray for effective control of *Pseudocercospora purpurea*. The second objective was to con-



sider including humidity and/or leaf wetness in the formula as a determining factor.

## MATERIALS AND METHODS

### Spore trapping: 2017/18 season

The trial was carried out in an unsprayed 'Fuerte' orchard at both ARC-TSC and Halls in the Nelspruit area. Four spore traps were placed at each of the two sites. The 'Fuerte' orchard at the ARC-TSC had a very high disease incidence as it had not received any copper sprays for several seasons. Two of the four spore traps had four vaseline-coated slides held horizontally and two had four slides held vertically facing the four different wind directions. Slides were held in position with clothes pegs attached to the end of two metal strips fixed at right angles to one another, on top of a 1.5 m pole.

During the 2017/18 season, spore release in the ARC-TSC orchard was monitored from 6 September 2017. Slides were changed weekly on the four traps and additionally, daily, on the two horizontal traps from 27 November to 22 December and from 9 January to end of February. For the daily spore trapping, 4 additional clothes pegs were added onto the horizontal traps to hold these slides. In November 2017, for a 10-day period (12 November to 21 November), slides were changed on an hourly basis from 23:00 in the evening until 03:00 in the morning.

Reading of the slides was carried out using methyl blue dissolved in water at 0.05% [m/v] as a stain for *P. purpurea* spores.

In the 2017/18 season, weather data were obtained from the ARC-TSC weather station for the ARC-TSC site and Hobo sensors were placed at the Halls site to record temperature and humidity. Additional weather data were obtained from the nearest weather stations. At the ARC-TSC site an EM50 data logger was placed in the orchard to record leaf wetness. Weather data were correlated with spore trapping data.

### Spore trapping 2018/19 season

During the 2018/19 season, two spore traps were placed in the 'Fuerte' orchard at ARC-TSC (Fig. 1) from September 2018. Only spore traps that had four horizontally-held vaseline-coated slides (Fig. 2) were used in the 2018/19 season, as results from the previous season showed that these slides trapped spores more consistently.

Slides were changed weekly on both traps from 10 October 2018. In addition, slides were also changed daily from September to December 2018. Reading of slides was carried out as described for the previous season. Weather data were obtained from the nearest weather station and correlated with spore trapping data.

## Infection periods

### 2017/18 Season

In the 2017/18 season, a bagging trial was carried out at Halls to determine the critical infection periods of avocado fruit by *Cercospora* spot. One thousand

five hundred fruit were covered with brown paper bags before they reached the susceptible stage of 40 mm in diameter. Every fortnight, fifty bags were removed to allow infection and then replaced at the end of the fortnight. In the 2017/18 season, fruit were covered when they were between 20 and 30 mm in diameter. Opening of fruit for fortnight periods started from 11 October. Brown paper bags were also covered with duct tape at the top end to prevent water from running onto fruit, in addition to the use of non-absorbent cotton wool.



Figure 1. A spore trap in the 'Fuerte' orchard at ARC-TSC.



Figure 2. A spore trap with horizontally-held vaseline slides.



Fruit were harvested on 10 April 2018. Immediately after harvest, fruit were evaluated for the presence of *Cercospora* spot on a scale of 0-5 (Table 1).

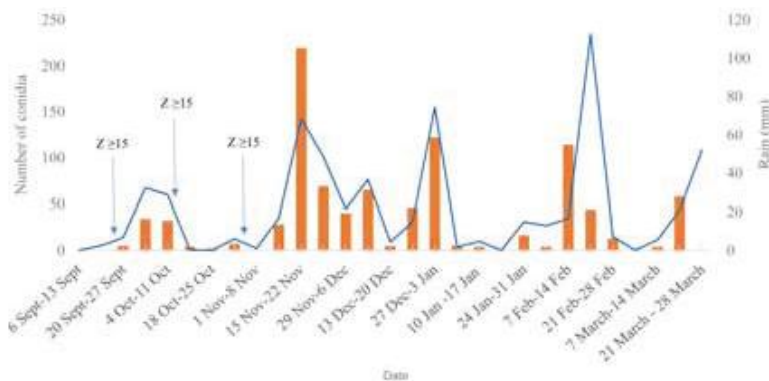
- 0 = clean fruit
- 1 = 1-5 spots with diameter of combined lesion area 1-5 mm
- 2 = 1-5 spots with diameter of combined lesion area 6-10 mm
- 3 = 1-5 spots with diameter of combined lesion area >10 mm
- 4 = 6-10 spots
- 5 = >11 spots.

For Classes 1-3, all fruit had 1-5 spots but were differentiated on the basis of combined lesion diameter as most of the fruit that was not clean fell into these 3 categories and it was necessary to differentiate between fruit with 1-5 large lesions and fruit with 1-5 very small lesions. Results are expressed in terms of a disease index according to Wheeler (1969) where infection index = (Sum of all numerical ratings/total number of fruit) x (100/Maximum disease category (5)).

Disease index data were correlated with weather data.



**Figure 3.** 'Fuerte' avocado fruit covered with brown paper bags at ARC-TSC.



**Figure 4.** The mean weekly number of conidia trapped at the ARC-TSC site and weekly rainfall in the period September 2017 to March 2018.

### 2018/19 Season

During the 2018/19 season, the bagging trial was carried out at the ARC-TSC site as this site had a much higher disease incidence in comparison with the Halls site used in the 2017/18 season. One thousand fruit were covered with brown paper bags from beginning of October when fruit were between 20 and 25 mm in diameter (Fig. 3). Every fortnight, 30 bags were removed to allow infection and then replaced at the end of the fortnight. Towards the end of the season, only 20 bags were removed and replaced as many bags were lost due to November drop and adverse weather storms (hail) experienced during the season. In the 2018/9 season, this was carried out from 10 October 2018 until end February 2019.

In addition to the two-weekly exposure periods, 25 fruit were also opened for a monthly time exposure period from 10 October until end February 2019.

Fruit will be harvested in April 2019 and evaluated as described for the 2017/18 season.

## RESULTS

### 2017/18 Season

#### Weekly spore trapping

In the 2017/18 season, more than 1000 slides were placed in the orchards at the two trial sites from September 2017. At the Halls trial site, only weekly spore trapping data were collected, while at the ARC-TSC site both weekly, daily and hourly data were collected. Due to the low disease incidence at the Halls site, no spores were trapped in the 2017/18 season. Weekly conidia trapped at the ARC-TSC site from September 2017 – March 2018 are presented in Figure 4.

Conidia were first detected in the period 20 to 27 September. Conidia were also detected in the following two periods with the commencement of the rain season. The Z value for the first period in the season was  $\geq 15$  from 12 to 17 September, predicting spore release, but no spores were trapped in this period. The reason for the Z value being  $\geq 15$  at this stage is that very high temperatures





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were recorded in September. The next period that the Z value was  $\geq 15$  was on 5 October for a one day period only. Average fruit size at this stage was still below 25 mm in diameter and spraying using the Z value model not yet recommended. By using the Z value model, the recommendation will be to spray soon after this period i.e. as soon as fruit reached 25 mm in diameter. Conidia were also detected in the period 25 October to 1 November, as some rain was recorded in this period. In this period a Z value of  $\geq 15$  was calculated using Darvas' equation and fruit size was above 25 mm, indicating that spraying should have commenced before this date. Conidia numbers also started to increase rapidly after this period, so spraying before this period is important. Conidia numbers reached a peak in the period 15 to 22 November when more than 68 mm rain was recorded. Several other peaks were observed throughout the season following the trend of rainfall, however, this was not observed towards the end of the season. Spores were trapped as late as the third week in March as rain still occurred.

When the weekly conidia trapped was compared with weekly rainfall, a very good correlation ( $r = 0.898$ ) was found between conidia numbers and rainfall for the period September to December 2017. For the entire season (September to March) the correlation was  $r = 0.609$ , somewhat lower due to lower numbers of spores trapped despite high rainfall in mid-February. No significant correlations were found with any other weather parameters (temperature, humidity or leaf wetness).

#### Daily and hourly spore trapping

Daily conidia trapped at the ARC-TSC site for a 26-day period in November/December 2017 and 51-day period in January/February 2018, are presented in Figure 5. The daily data is presented as weekly counts.

The general observation is that spores were trapped predominantly in periods when rainfall occurred,

as was found with the weekly spore trapping. When the daily spore trapping data was correlated with all the weather parameters, a poor correlation was found between the number of conidia trapped and all of the weather parameters analysed. The best correlation was obtained for rain with a correlation coefficient of  $r = 0.224$ . When daily data are however presented as weekly counts and correlated with weekly weather data, weather parameters influencing spore release are smoothed out and good correlations were found between weekly conidia counts and rainfall, as was found with weekly spore trapping data.

#### Hourly spore trapping

On the slides that were changed hourly between 23:00 at night and 03:00 in the morning for the 10-day period 12-21 November 2017, no spores were trapped.

#### Infection periods

The infection indices for each of the 12 periods as well as the index for fruit covered for the entire period and fruit exposed the entire period of the 2017/18 season's trial at the Halls site, are presented in Figure 6.

The highest disease indices for Cercospora spot were recorded for fruit exposed during the entire period. Of the two weekly exposure periods, highest disease indices were recorded for the period 8-22 November. This two-weekly period coincided with the two weekly periods when 27 and 218 spores were trapped, respectively. The 218 spores trapped in the period 15-22 November also represents the period when spores trapped reached a peak during the season.



**Figure 5.** The daily number of conidia trapped at the ARC-TSC site and rainfall in the period November 2017 to February 2018. Daily data are presented as weekly counts.



**Figure 6.** Cercospora disease severity during 2017/18 season in the unsprayed 'Fuerte' orchard at Halls.



## 2018/19 Season

### Weekly spore trapping

Weekly conidia trapped at the ARC-TSC site from September 2018 – December 2018 are presented in Figure 7.

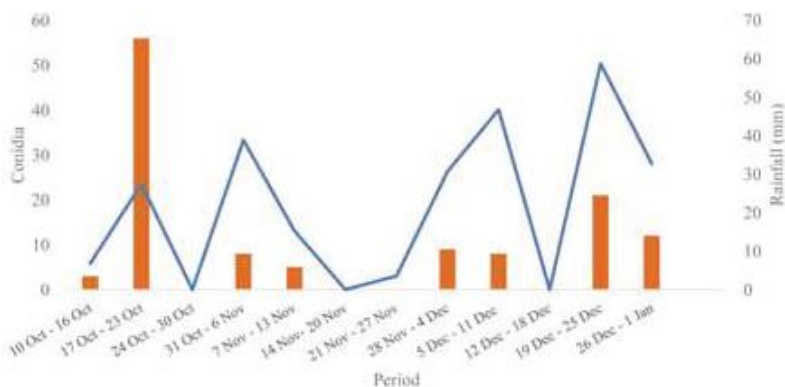
Very low numbers of conidia (3) were detected in the first period starting from 10 October 2018. In the second period from 17-23 October, a high number of conidia was trapped. During this period rainfall also occurred and spores were trapped, as was found in the 2017 season in periods where rain occurred. On 24 October, average fruit size was 29.7 mm in diameter and fruit not regarded as susceptible, although recommendations by Subtrop suggest that spraying should commence when the fruit is smaller or equal to 25 mm in diameter. Whether infection has already occurred in this period can only be confirmed when fruit has been harvested and disease index calculated for this period. Spores trapped in the periods 31 October to 6 November and the following period also coincided with periods when rainfall was recorded as well as in the periods from 28 November to 11 December. No spores were trapped in the periods where no rainfall was recorded.

### Infection periods

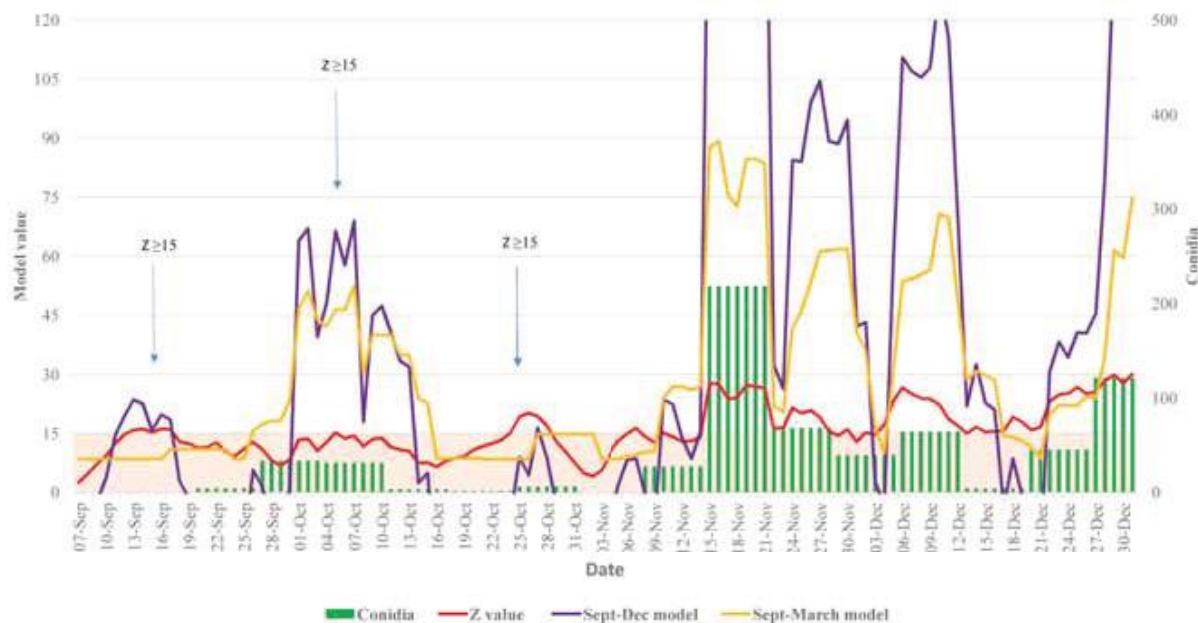
Fruit from the 2018/19 season will be harvested in April/May 2019 and evaluated for *Cercospora* spot and the results expressed in terms of an index as described for the 2017/18 season. The disease index data and spore trapping data will be correlated with weather data.

### New models developed

Using the spore trapping data of the 2017/18 season, various new models were developed. A preliminary model for the period September to December was developed using multiple stepwise linear regression analysis of the weekly number of conidia trapped and weekly weather parameters. The equation for the best fitted regression line is  $y = -234.60 + 2.82 * \text{Rain} + 7.44 * \text{MaxT}$  with  $R^2 = 0.92$  and Adjusted  $R^2 = 0.91$ . The model obtained for the entire period (Sept-March), is  $y = 8.4166 + 1.1 * \text{Rain}$  with  $R^2 = 0.371$  and Adjusted  $R^2 = 0.348$ . With the multiple stepwise linear regression analysis of two-weekly number of conidia trapped and two-weekly weather parameters for September to March, the model developed is represented by the equation  $y = 9.80233 + 2.24767 * \text{Rain}$  with  $R^2 = 0.8532$  and Adjusted  $R^2 = 0.816$ .



**Figure 7.** The mean weekly number of conidia trapped at the ARC-TSC site and weekly rainfall in the period October 2018 to December 2018.



**Figure 8.** A comparison of the Z value model with the new models (Sept-Dec and Sept-March).



The disease index values for the 2017/18 season for each period were also correlated with weather data and a disease index model was developed. The highest correlation was between disease index and max Humidity. The multiple linear regression model is  $y = -110.149 - 4.92 * \text{MinT} + 2.08 * \text{MaxH}$  with  $R^2 = 0.816$  and Adjusted  $R^2 = 0.693$ .

A comparison of the Z value model and the new models for the period September to December 2018 showed that the new model follows a very similar pattern compared with the Z value model in the 2017/18 season, but with a few discrepancies (Fig. 8).

The first time in the season that the Z value was  $\geq 15$  was in the period 12 to 17 September. Both the Z value model and Sept-Dec model use temperature as a criterion in the equation and due to the high temperatures observed in September, these models predicted spore release in mid-September. No conidia were however trapped in this period. The Sept-March model which only uses rainfall as criterion, did not predict any spore release in this period. The next prediction of spore release using the Z value was on 5 October for a one-day period only. The Sept-Dec and Sept-March models also predicted spore release during this period, but for a longer period starting earlier and continuing for a longer period (1-13 October and 26 September to 15 October, respectively). The next period the Z value was above 15, was 24 to

28 October and fruit size was  $>25$  mm. The Sept-Dec model and the Sept-March model also predict spore release in this period but starting and ending a few days later.

## CONCLUSION

Models will need to be further refined with the inclusion of the 2018/19 spore trapping data. Although the models followed the same trend, some differences were found as outlined under results. The best model developed will be used to determine high risk infection periods enabling accurate and more effective timing of fungicide sprays.

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