Control of mango blossom malformation in South Africa

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Abstract
Mango blossom malformation (MM) caused by Fusarium mangiferae severely affects the crop in most of the mango producing areas of South Africa. Current control measures are to break out the malformed panicles as well as three additional nodes of the branch when malformed panicles are clearly visible. An increase in the incidence of MM which caused concern was, however, reported from the Hoedspruit area of South Africa during the last three seasons. The aim of this study was 1) to investigate and evaluate the influence of various management practices by producers on the incidence of malformation in the Hoedspruit area, 2) to develop a strategy for optimum control, 3) to determine the annual inoculum availability in an infected orchard at Nelspruit and 4) to evaluate the effect of prochloraz alone and in combination with sanitation on malformation.

In the Hoedspruit area, removal of malformed panicles by the producers over the three year period led to a decrease in malformation at Producer A (‘Kent’) and at Producer D (‘Keitt’) respectively. At Producer C, malformation was approximately the same over the three seasons and at Producer D malformation increased. A decrease in malformation was evident where removal of malformed panicles commenced early in the season, whereas an increase was observed where removal was only done late in the flowering period. In a ‘Sensation’ orchard at Nelspruit, removal of malformed panicles over the entire orchard in the 2012 flowering season led to an average decrease in malformation of 67% the following year. Further removal after harvest and a benomyl spray gave no further advantage. Low levels of conidia were detected in the months July to September. Detection of conidia increased from October and highest numbers were detected in November. The time when an increase in sporulation occurred, coincided with the time when the individual flowers on the panicles started to dry out. Spraying of prochloraz alone three times during the season had no effect on malformation incidence in the 2013 flowering season. Sanitation alone and sanitation combined with prochloraz reduced malformation. Any advantage of a prochloraz spray in addition to sanitation was not evident in the first season of application.

Introduction
Mango blossom malformation (MM) is one of the most serious diseases of mango (Mangifera indica L.) in South Africa, causing severe economic losses annually. Mango blossom malformation affects both flowers and vegetative tissue (Ploetz, 2001). To a greater or lesser extent, affected flowers take on the appearance of a cauliflower head. The flowers are mostly sterile and no fruit is produced (Manicom, 2008). Fusarium mangiferae has been proven in many studies to be the causal agent of MM disease worldwide, after conflicting reports of the causal agent existed for many years (Summanwar et al., 1966; Varma et al., 1974; Manicom, 1989; Freeman et al., 1999; Noriega-Cantu et al., 1999; Britz et al., 2002; Ploetz, 2003). Recently at least four other Fusarium species or sub-species, F. sterilhyphosum, F. mexicanum, F. proliferatum and F. tuiensi have also been associated with the disease (Britz et al., 2002; Marassas et al., 2006; Otero-Colina et al., 2010). Of these, only F. sterilhyphosum Britz, Wingfield & Marassas has been associated with MM in South Africa.

Recommendations in South Africa are removal of malformed panicles when these can be easily distinguished, but before the flowers have dried out and sporulation occurs (Manicom, 2008). Recommendations further require that malformed panicles should be broken out three nodes (growth flushes) back or at least 200 to 300 mm behind the malformed panicle, whichever is greatest. During the past three seasons an alarming increase in malformation was reported in South Africa from especially the Hoedspruit production area in the Limpopo Province (J du Preez, personal communication). This caused concern that current control measures were no longer effective.

Studies on the epidemiological aspects of mango blossom malformation disease by Youssef et al. (2007) have shown that infection is not systemic and
that infection of apical meristems most likely originates from conidia disseminated from malformed panicles. Recent studies on dispersal patterns of conidia of *F. mangiferae* also suggest that spores are spread by wind and that the primary infection sites are the apical buds (Gamiel-Atinsky et al., 2009a; 2009c; Noriega-Cantu et al., 1999). Based on these results and further studies, removal of infected panicles combined with timely sprays of fungicides to protect apical buds was recently recommended by Israeli researchers as a more effective management strategy (Dr Stanley Freeman, personal communication).

The aim of this study was 1) to investigate and evaluate the influence of various management practices by producers in the Hoedspruit area on the incidence of mango blossom malformation, 2) to develop a strategy for optimum control of MM, 3) to determine the inoculum availability through the year of *F. mangiferae* and 4) to determine the effect of prochloraz alone or in combination with sanitation on malformation.

**Materials and methods**

**Hoedspruit** (24°24′12.7″S, 30°47′59.82″E) (Limpopo Province)

In the Hoedspruit area, the influence of various management practices on the incidence of malformation was evaluated for three consecutive seasons (2011 to 2013). An orchard with a history of MM was selected at each of four producers, A, B, C and D. Thirty branches were marked randomly at each site in the 2011 season, except at producer B where only 20 branches were marked. The total number of healthy and malformed panicles was counted before each removal of malformed panicles by the respective producers during the 2011, 2012 and 2013 seasons and incidence of mango blossom malformation determined at each producer. Records were kept of all husbandry practices followed.

**Nelspruit** (25°27′96″S, 30°58′14.43″E) (Mpumalanga Province)

**Trial A: Integrated strategy for optimum control**

The following methodology was followed in six rows of a 36 year old ’Sensation’ orchard at the Agricultural Research Council’s Institute for Tropical and Subtropical Crops (ARC-ITSC), Nelspruit. The trial consisted of three treatments set up in a randomized complete block design with four replicates (10 tree plots) per treatment. Data was only taken on three trees per plot. Pre-treatment data was recorded on 24 randomly selected branches per treatment by counting total, healthy and diseased panicles on each branch in October 2012. Data was used to calculate average healthy and diseased panicles per tree and disease incidence. Directly after monitoring, malformed panicles and branches across the entire trial were removed 300 to 500 mm behind malformed panicles by hand or securers connected to long poles. Fruit were harvested for atchar in January 2013. In the 2013 flowering season, malformed panicles were removed only in treatments 2 and 3. Pre-treatment data was recorded before removal of malformed panicles in 2013.

The three treatments applied after harvest in 2013 and treatments applied in the 2013 flowering season are listed in Table 1.

<table>
<thead>
<tr>
<th>Treatment no</th>
<th>Treatments applied in 2013</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Control - no further removal of malformed branches/panicles.</td>
</tr>
<tr>
<td>3</td>
<td>Removal of malformed branches/panicles after 2013 harvest and spray with benomyl 50% a.i. @ 75 g/100 l. Removal in September and October 2013. Spray with prochloraz 0.1% @ 100 ml/l after 2014 harvest.</td>
</tr>
</tbody>
</table>

**Trial B: Sporulation from malformed panicles and seasonal dynamics of airborne conidia on leaves**

Sporulation from malformed panicles was evaluated from July to December of the 2013 flowering season. Malformed panicles were randomly sampled once a week from a ’Sensation’ orchard at the ARC-ITSC research farm at Nelspruit. Both early and late panicles were collected weekly. In July some panicles were already fully expanded while the later panicles which represented most of the orchard were only 2 to 6 cm in length. Three panicles representing each of these stages were collected. Photos were taken of the development stage of the collected panicles. The panicles were grouped into early and late panicles. The small individual flowers on the malformed panicles from each group were removed separately, 40 g weighed off, and suspended in 250 ml of water to which Abamectin (18 g/l a.i.), a miticide, was added at a concentration of 1 ml/l l. The suspension was stirred every 15 minutes. After an hour the suspension was diluted 10 and 100 fold and 100 μl from each dilution from each group were transferred to 90 mm petri dishes containing Nash & Snyder medium semi-selective for *Fusarium* spp. Ten replicate plates
per panicle group were assessed for *F. mangiferae* after seven days, incubation at 25°C. Identification of the *Fusarium* spp. was carried out by microscopic observation. Each week a representative colony was stored on slants and later identified by specific polymerase chain reaction (PCR) amplification. The average number of conidia per gram of malformed panicle for each of the two flower groups was calculated for each sampling date.

Table 3. Treatments followed in the prochloraz trial, 2013 season.

<table>
<thead>
<tr>
<th>Treatment no</th>
<th>Treatments</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.1% Prochloraz (45% a.i.) applied three weekly during the flowering period (end July - Oct).</td>
</tr>
<tr>
<td>2</td>
<td>Removal of malformed panicles three times during the season (Aug, Sept, Oct).</td>
</tr>
<tr>
<td>3</td>
<td>0.1% Prochloraz applied three weekly during the flowering period + removal of malformed panicles three times during the season (Aug, Sept, Oct).</td>
</tr>
<tr>
<td>4</td>
<td>Untreated control.</td>
</tr>
</tbody>
</table>

In order to determine the presence of airborne conidia of *F. mangiferae* on leaves during the season, leaves were collected from the orchard described above from July 2013 to March 2014. Ten leaves were collected weekly. The leaves were cut into small pieces and 20 g suspended in 250 ml water. The rest of the procedure was similar to that followed for malformed panicles, except that no dilutions were made and only five replicate plates were assessed. The average number of conidia detected per gram of leaves was calculated for each sampling date.

**Trial C: Prochloraz trial**

In the same orchard adjacent to the above trials, a trial to evaluate prochloraz sprays at regular intervals was carried out in the 2013 flowering season. Treatments are listed in Table 3.

The trial consisted of four treatments in a randomized complete block design with three replicates (6 tree plots) per treatment. Data was only collected from three trees. On each data tree, a branch was marked in each quadrant of the tree. Data was recorded by counting diseased, healthy and total number of panicles on each branch three times during the season before removal of malformed panicles. Average panicles per tree on marked branches were recorded and percentage infection calculated. Yield data was recorded at harvest in 2014. Treatments will be applied for three consecutive years.

**Results and discussion**

**Hoedspruit**

The incidence of malformation at each of the four orchards after monitoring over three seasons (2011 to 2013) in the Hoedspruit area is shown in Figures 1 to 4. Producer A (cultivar Kent) removed malformed panicles three times during the 2011 and 2013 seasons and twice in the 2012 season, starting in early August (Fig. 1). At this early stage, malformation could be detected on the green, half expanded panicles, but was not always definite. Malformation decreased from 17% to 10.4% from 2011 to 2012 and from 17% to 11% from 2011 to 2013. The 2011 and 2013 seasons were ‘on’-seasons. Although removal was done regularly and started early, removal was not always done completely and malformed panicles were missed. A relatively good reduction in malformation incidence was observed. With more complete removal in several consecutive seasons and by protecting the apical buds with an effective fungicide, it should be possible to lower the incidence of malformation to insignificant levels.

Producer B (cultivar Keilt) removed malformed panicles only once per season (Fig. 2). Removal was done early to late October. At this stage it was very easy to identify the malformed panicles, which were large and bear no fruit, but at this stage sporulation has occurred. Blossom malformation decreased from 12.9% to 5% in the first year after removal. From the 2011 to 2013 seasons which were both ‘on’-years, malformation increased from 12.9% to 21.9%. This
increase is probably due to the fact that malformed panicles were not removed early enough during the 2011 season. In the 2012 season flowering was poor and many branches only produced a flush and no panicles. Vegetative malformation was observed on many branches. In the 2013 season which was again an ‘on’-year, blossom malformation was severe due to infections from the 2011 season, manifested as vegetative malformation in 2012.

Producer C (cultivar Kent) removed malformed panicles once in the 2011 season (early September) when panicles were fully developed (Fig. 3). Malformation was about the same the following season. In 2012 malformation was removed once late in the season at the end of October. Malformation in August 2013 was the same as the previous two seasons (between 5 and 6%) and malformation was only removed thereafter. Malformation was low in this orchard from the initiation of the trial and inconclusive results were obtained.

Producer D (cultivar Keitt) removed malformed panicles once in the flowering season (end September) in 2011 and 2012. Panicles were fully developed and fruit were set on the healthy panicles. In the 2013 season, malformation was removed twice during the season in early August and again early September: Malformation decreased from 13.7% to 4.6% the first year after removal and to 2.2% in the second year. Removal in this orchard was done completely, resulting in a 83.9% decrease in malformation from 2011 to 2013.

Removal of malformed panicles at producer A (‘Kent’) and D (‘Keitt’) led to a decrease in MM over the three seasons. At both these producers malformation was removed earlier in the season than at producer B. At Producer D removal was done more completely than at Producer A, resulting in a higher decrease. At Producer C malformation was low and stayed about the same over the three year period. Based on these results, recommendations are to remove malformed panicles early in the season when the first malformed panicles are present to remove the inoculum source before sporulation occurs. Removal should be done at regular intervals during the season. Malformed panicles should be removed 30 to 50 cm behind malformed panicles and care should be taken to remove all malformed panicles at each removal time. If early malformed panicles are missed, these panicles produce spores before the second round of removal. In Israel only removal of malformed panicles are recommended and according to Dr Freeman (personal communication) it is not necessary to remove part of the branch behind the malformed panicle, as only the source of inoculum should be removed. In orchards with a high incidence of malformation, we still recommend removal of malformed panicles and branches as far back as possible, as the fungus is present in the lateral buds of the wood several growth flushes back.

Nelspruit

**Trial A: Integrated strategy for optimum control**

**2012 flowering season:** Percentage infection in September 2013 after treatments were applied after harvest in January 2013 is presented in Table 4.

In each treatment, 40 trees were included – 10 per replicate in four replicates. Data were taken from three data trees in each replicate. Average healthy and malformed panicles per tree was recorded and % infection calculated.

Percentage infection before removal of malformed panicles across the entire trial in 2012 was between 19.2% and 21.1%. The removal of malformed panicles after harvest alone and removal + benomyl spray did not result in improved control of malformation in comparison with the control treatment in 2013. All treatments had about 67% less malformation than in 2012. This decrease in malformation is most probably due to the removal of malformed panicles across the entire trial in October 2012. The treatment effects will

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**Figure 3:** The incidence of malformed panicles monitored on marked branches at Producer C in the 2011 to 2013 seasons. (Time of removal of malformed panicles is indicated by red arrows.)

**Figure 4:** The incidence of malformed panicles monitored on marked branches at Producer D in the 2011 to 2013 seasons. (Time of removal of malformed panicles is indicated by red arrows.)
be more evident after another season’s data has been obtained.

The removal of malformed panicles in September and October 2013 has led to a decrease in malformation in the removal and removal + spray treatments (data not shown). Where malformed panicles were not removed (control) in the 2013 season, malformation increased to 13.5% in the 2013 flowering season. The influence of removal of malformed panicles in the 2013 season only in treatments 2 and 3 would only be evident in the 2014 flowering season. Prochloraz was applied after harvest in the 2013/14 season and the influence will also only be seen in 2014.

Yield data recorded in January 2013 and 2014 are presented in Table 5.

There were no treatment differences after two season’s harvest, but we hope to see differences at the following harvest in Jan 2015.

**Trial B: Sporulation of malformed panicles and seasonal occurrence of airborne conidia on leaves**

The sporulation of malformed panicles during the 2013 flowering season is presented in Figure 5 (early panicles) and Figure 6 (late panicles).

**Early panicles:** Low levels of conidia were detected in the months July to September ranging from 0.1 to 25.5 conidia/g of malformed panicles. Sporulation increased slowly through October, then jumped dramatically in November, decreasing thereafter to below 1000 conidia/g of malformed panicles in mid-December. At this stage there were few malformed panicles

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**Table 4.** Effect of removal of malformed panicles after harvest in 2013 alone and in combination with a benomyl spray on the incidence of malformation.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Sept 2012 Pre-treatment data % infection</th>
<th>All removed Oct 2012</th>
<th>Apply treatments</th>
<th>Post-treatment data Sept 2013</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Control – no further removal of malformed branches/panicles.</td>
<td></td>
<td></td>
<td></td>
<td>6.1</td>
</tr>
<tr>
<td>2. Removal of malformed branches/panicles after 2013 harvest.</td>
<td></td>
<td></td>
<td></td>
<td>6.7</td>
</tr>
<tr>
<td>3. Removal of malformed branches/panicles after 2013 harvest and spray with benomyl 50% a.i. @ 75 g/100 l.</td>
<td></td>
<td></td>
<td></td>
<td>6.8</td>
</tr>
<tr>
<td>Average</td>
<td>19.9</td>
<td></td>
<td></td>
<td>6.5</td>
</tr>
</tbody>
</table>

**Table 5.** Yield data recorded in January 2013 and 2014.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>All malformation removed</th>
<th>Apply treatments after harvest</th>
<th>Jan 2013</th>
<th>Sept 2013</th>
<th>Oct 2013</th>
<th>Jan 2014</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treatment 1 Control</td>
<td>1166 kg</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>7808 kg</td>
<td></td>
</tr>
<tr>
<td>Treatment 2 Removal</td>
<td>729.14 kg</td>
<td>Removal</td>
<td>7808 kg</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Treatment 3 Removal + spray after harvest on flush</td>
<td>1210 kg</td>
<td>Removal</td>
<td>7749 kg</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
present, as heavy rains had resulted in many of the malformed panicles disintegrating and falling to the ground. No further collections were made.

Late panicles: Very low levels of conidia were detected in the months July to September ranging from 0.1 to 6.4 conidia/g of malformed panicles. Sporulation increased from early October but was still relatively low when compared to the early panicles, in which individual flowers had already dried. Sporulation peaked at the end of November.

Taken together, the maximum sporulation in this year extended over the entire month of November. The time when an increase in sporulation occurred, coincided with the time when individual flowers on the panicles started to dry out and in this orchard it was on 9 October on the early panicles and after 23 October on the late panicles. As a time cannot be linked to an increase in sporulation in all the production areas, due to climatic differences, malformed panicles should be removed as soon as they can be identified and regularly thereafter to ensure that they are removed before individual flowers start to dry out and produce spores.

The seasonal occurrence of airborne conidia of F. mangiferae on leaves is presented in Figure 7.

Low levels of conidia were detected on the leaves in the months July to September 2013. In July the average number of conidia per gram of leaves was below 0.1, in August no spores were detected and in September it was below 0.05. Low numbers of conidia are produced in October from panicles and the only reason why higher numbers of conidia was detected on the leaves in October in comparison with November was because rain has not yet started. In November, when maximum spores are produced on the panicles and a peak in spores is expected on the leaves, spores are washed from leaves, resulting in the lower numbers detected on leaves. After November there is a decrease in sporulation on panicles resulting in a decline in spores on leaves. Rain also continues to wash off spores. As most of the malformed panicles has disintegrated and fallen to the ground, no new spores are released after the end of the main rain pe-

Figure 7. Seasonal pattern of conidia detected on leaves in a ‘Sensation’ orchard at Nelspruit during the 2013 flowering season.

Table 6. The effect of sanitation/removal and prochloraz spray on the incidence of mango blossom malformation.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Total panicles (avg/tree)</th>
<th>Malformed panicles (avg/tree)</th>
<th>Infection % 15/10/2013</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. 0.1% prochloraz (45% a.i.) three weekly during the flowering period (end July - Oct.)</td>
<td>70.9</td>
<td>13.3</td>
<td>18.8</td>
</tr>
<tr>
<td>2. Removal of malformed panicles three times during the season (Aug, Sept, Oct.)</td>
<td>77.0</td>
<td>7.2</td>
<td>9.4</td>
</tr>
<tr>
<td>3. 0.1% prochloraz three weekly during the flowering period + removal of malformed panicles three times during the season (Aug, Sept, Oct.)</td>
<td>67.1</td>
<td>5.9</td>
<td>8.8</td>
</tr>
<tr>
<td>4. Untreated control</td>
<td>78.1</td>
<td>14.8</td>
<td>18.9</td>
</tr>
</tbody>
</table>
period. In January through to March the average number of conidia was below 0.2.

The presence of spores on leaves coincides with the time that the sporulation of malformed panicles was high. This is also the time when infection of buds will take place. Spraying of fungicides to protect the terminal buds should be applied in the period before high numbers of spores are released to keep the inoculum available as low as possible and to protect the terminal buds from infection.

**Trial C: Prochloraz trial**
The effect of prochloraz alone or in combination with sanitation is presented in Table 6. Prochloraz applied three weekly during the flowering period, starting from end of July, did not result in improved control in comparison with the untreated control. Removal of malformed panicles in treatment 2 and 3 significantly reduced malformation. Prochloraz sprays three weekly during the flowering period combined with removal three times during the season was no different from removal alone. Any advantage of a prochloraz spray in addition to sanitation was not yet evident in the first season of application.

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**References**


