Control of powdery mildew (Sphaerotheca pannosa var. roae) on rose (Rosa L. sp.) using anhydrous milk fat and soybean oil emulsions

A. Ah Chee, K.V. Wurms and M. George

The New Zealand Institute for Plant & Food Research Limited, Private Bag 3230, Hamilton 3240, New Zealand
Corresponding author: Annette.AhChee@plantandfood.co.nz

Abstract Powdery mildew (PM) is a serious fungal disease of a wide range of horticultural crops and can adversely affect crop yield and quality. Anhydrous milk fat (AMF) and soybean oil (SBO) emulsions were evaluated for control of PM (Sphaerotheca pannosa var. roae) on potted rose plants (Rosa L. sp. 'Splendid Surprise' and 'Sahara') maintained in a controlled environment. Foliage was sprayed weekly with AMF (0.7% w/v), SBO (2% w/v), fungicide (Supershield™, 1% v/v), water, or left unsprayed. PM infection increased to 100% leaf area in water and unsprayed control plants over 6 weeks. The fungicide reduced infection to a severity rating of 5 (>40% leaf area infection). The AMF and SBO treatments gave significantly better disease control (P <0.0001) with a severity rating of 2 (2-5% infection). Some yellowing and necrosis of leaves was observed on plants in the SBO treatment, but overall leaf health was superior to that of control plants.

Keywords anhydrous milk fat, soybean oil, powdery mildew, greenhouse roses.

INTRODUCTION
Roses are one of the most important ornamental plants globally (Debener et al. 2004). Powdery mildew (PM) caused by the fungus Sphaerotheca pannosa var. roae is the most widespread and economically important disease in commercial cut rose production (Pasini et al. 1997). The first signs of PM appear on young leaves, which hold their colour but begin to crinkle. Subsequently, the disease appears as a whitish powder covering foliage, stems and buds (Rose Magazine Inc. 2011). Severe PM outbreaks can make entire crops unmarketable through damage such as leaf chlorosis and necrosis, bud distortion, defoliation, leaf rolling, stunted growth and twisted new stems (Morgan 2010). Although the home grower has many soft fungicide control options, such as sodium bicarbonate (Rose Magazine Inc. 2011), PM in commercial greenhouse roses is usually controlled by synthetic chemical products (Scarito et al. 2007). Moreover, the efficacy of sodium bicarbonate is very dependent on plant cultivar (Dik et al. 2003), can be associated with phytotoxicity (Pasini et al. 1997; Scarito et al. 2007) and may be less durable than traditional fungicides (Reuveni et al. 1994).

Given an increasing international demand to reduce the use of toxic pesticides, because of human health and environmental concerns (Tjosvold & Koike 2001), there is a need to seek more benign disease control alternatives. This is particularly relevant to the PM/rose interaction, since PM control can account for up to 40% of the...
pesticide volume applied to rose crops (Tjosvold & Koike 2001).

Milk has been reported to be effective for the control of PM in squash and wine grape crops (Bettiol 1999; Crisp & Bruer 2001), but the use of raw milk has had problems including difficulties in handling/storage and unwanted growth of non-target organisms. Different fractions of milk were tested and AMF was found to be the most toxic to PM on glasshouse-grown squash (K.V. Wurms, unpublished data). The anti-fungal activity of plant oils against PM in tomato has also been reported (Ko et al. 2003).

The purpose of this study was to evaluate emulsified formulations of anhydrous milk fat (AMF) and soybean oil (SBO) for their potential to control PM outbreaks in glasshouse-grown roses and to establish if there were any plant health issues associated with regular applications. This represents a novel use for these products.

MATERIALS AND METHODS

Trial 1

Rooted cuttings of the PM-susceptible rose cultivar ‘Splendid Surprise’, infected naturally with PM, were potted into 20-cm diameter pots with potting mix (General Purpose Mix, Daltons Ltd, Hamilton, New Zealand) and maintained in the glasshouse during late summer. Pots were watered daily with tap water through drip feed irrigation. Over the course of 8 weeks, five treatments (Table 1) were applied weekly, using mist bottles, to the adaxial and abaxial leaf surfaces until runoff to ensure full coverage. The experiment was a randomised block design with five plants per treatment.

The intention was to carry out PM and leaf health assessments on a fortnightly basis but the plants became severely infested with two-spotted spider mite (*Tetranychus urticae*), resulting in extremely poor plant health and leaf drop. Repeated attempts to control the mites were unsuccessful and even exacerbated plant phytotoxicity – two applications of Yates Mavrik® insecticide/miticide (10 ml/litre) were ineffectual, and a single application of 20 ml/litre Yates Mite Killer (active ingredient 250 g/litre fatty acid potassium salts) caused severe phytotoxicity (chlorosis and necrosis). To try to minimise further mite damage throughout the course of the experiment, the predaceous mite, *Phytoseiulus persimilis* (supplied by BioForce Ltd, Auckland, New Zealand), was introduced by draping bean leaves infested with *P. persimilis* onto the roses. Collection of quantitative data was abandoned but the spray programme was continued to see if any treatment effects on PM could be observed.

Trial 2

Mature rose plants of the PM-susceptible rose cultivar ‘Sahara’ were pruned to compact bushes of height ~40 cm, root-pruned and potted into 20-cm diameter pots with potting mix (General Purpose Mix, Daltons Ltd, Hamilton, New Zealand). Plants were maintained in a controlled environment (CE) room at 20°C with a 16 h photoperiod. Pots were hand-watered with tap water every 2–3 days. The plants were left for

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Treatment code</th>
<th>Application rate</th>
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</thead>
<tbody>
<tr>
<td>Unsprayed control</td>
<td>Unsprayed</td>
<td></td>
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<tr>
<td>Distilled water control</td>
<td>Water</td>
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<tr>
<td>Fungicide: 4.4 g/litre mycobutanil and 9.6 g/litre tau-fluvalinate (Supershield™; Yates, New Zealand)</td>
<td>Supershield</td>
<td>10 ml/litre</td>
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<tr>
<td>Emulsified anhydrous milk fat (New Zealand Milk Products Ltd, New Zealand)</td>
<td>AMF</td>
<td>7 g/litre</td>
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<tr>
<td>Emulsified soybean oil (Goodman Fielder, New Zealand)</td>
<td>SBO</td>
<td>20 g/litre</td>
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3 weeks to establish new leaf growth and to infect naturally with PM before commencement of the trial. Rose buds and flowers were removed throughout the course of the trial.

There were five treatment applications at weekly intervals. Treatments were the same as in Trial 1, and were applied using mister bottles to the adaxial and abaxial leaf surfaces until runoff to ensure full coverage. There were six plants per treatment. The plants were randomised within blocks on a single bench within the CE room, with one replicate plant from each treatment in each block.

Disease assessments were carried out immediately before each spray application and 1 week after the last spray. Assessments of PM severity were based on the rating scale in Table 2. Disease ratings were given for the mature portion of each plant as a whole, excluding the fresh unexpanded juvenile leaves. Individual leaves were not assessed because roses have relatively rapid growth and turnover of leaves.

Pest problems included an initial plague of slugs (which were removed by hand). Following the fifth treatment application, there was an invasion of two-spotted spider mite. It was decided that, rather than apply a miticide and continue with further treatment applications, the trial would be concluded 6 weeks after commencement.

PM severity data at each assessment date were analysed separately. Mean treatment differences were analysed using analysis of variance (ANOVA) with means separation by Fisher’s Least Significant Difference (LSD) (P<0.05), using Genstat, version 8.1. The ANOVA was adjusted using the initial disease assessment as a covariate.

RESULTS

Trial 1

After 8 weeks of spray applications, the roses treated with AMF and SBO were the healthiest looking, with the least PM and mite infestation (Figure 1). The fungicide treatment was also relatively free of PM and mites, but appeared to be slightly phytotoxic, causing leaf chlorosis, necrosis and rolling (Figure 1). There was heavy PM infection and leaf chlorosis in the unsprayed and water control plants (Figure 1).

Overall, the SBO treatment provided the best balance between leaf health and disease control (Figure 2).

Trial 2

With natural PM infection, the rose plants commenced the trial with a high mean PM severity rating of 4 (21-40% leaf area infected) (Figure 3). Variation in severity rankings between plants at this stage was also quite high, so plants were arranged such that same levels of variation were found across all treatments. Disease severity increased to the maximum ranking of 6 (100% leaf area infected) in the control plants 35 days after the first treatment application, but remained approximately the same as the initial severity in the Supershield™ treatment. However, in the AMF and SBO treatments, PM severity decreased to ratings of 2 (2-5% leaf area infected) and 1 (≤1% leaf area infection), respectively, 15 days after the first treatment application. These low PM severity ratings were maintained in the natural product treatments until the conclusion of the trial.

Leaf health results were the same as in Experiment 1, so photographs are not presented. AMF- and SBO-treated leaves were the healthiest, with little PM infection and no chlorosis. The Supershield™ treatment resulted in leaves with moderate infection and chlorosis, and the control plants were completely infected with PM and exhibited severe chlorosis and necrosis.

Table 2: Powdery mildew severity rating scale for leaves, from Spencer (1977).

<table>
<thead>
<tr>
<th>Powdery mildew rating</th>
<th>Percent leaf area infection</th>
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<tbody>
<tr>
<td>0</td>
<td>No infection</td>
</tr>
<tr>
<td>1</td>
<td>≤1% infection</td>
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<tr>
<td>2</td>
<td>2-5% infection</td>
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<td>3</td>
<td>6-20% infection</td>
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<td>4</td>
<td>21-40% infection</td>
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<tr>
<td>5</td>
<td>&gt;40% infection</td>
</tr>
<tr>
<td>6</td>
<td>100% infection</td>
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</table>

Figure 1 Powdery mildew infection and leaf health of ‘Splendid Surprise’ roses after eight weekly spray applications. (a) Unsprayed, (b) Water, (c) 10 ml/litre Supershield™, (d) 7 g/litre anhydrous milk fat and (e) 20 g/litre soybean oil.

Figure 2 ‘Splendid Surprise’ roses after eight weekly spray applications with 20 g/litre soybean oil (left) or unsprayed (right).
DISCUSSION

In the absence of disease control, severe PM infection and phytotoxicity were observed on 'Splendid Surprise' and 'Sahara' rose cultivars. Supershield™ fungicide was able to control the disease, but caused leaf chlorosis, while applications of AMF and SBO resulted in glossy, disease-free leaves in both cultivars. Within 2 weeks of the first treatment application on 'Sahara' roses, AMF and SBO treatments significantly reduced PM severity rating from 4 (21-40% leaf area infection) to a rating of 2 (2-5% infection) in the AMF treatment and 1 (≤1% infection) in the SBO treatment. These levels of infection were maintained until the conclusion of the trial. The natural product treatments significantly outperformed the fungicide Supershield™.

It could be argued that some decrease in PM severity in the whole plant data was attributable to the emergence of new leaves and the abortion of heavily infected leaves. However, if this were the case, the same trends would be expected in all the treatments, whereas PM severity increased in the control plants over time to 100%, whilst plants treated with Supershield™ maintained the initial infection severity for the duration of the trial. The Supershield™ result would be expected of a pesticide with protectant activity only but Supershield™ has been reported to have both protectant and eradicant activity (Roberts 2010). This indicates that Supershield™ was generally ineffective under the CE conditions, which favour the pathogen and therefore represent a stringent test on any potential product intended to control PM. The whole-plant PM severity data obtained for AMF and SBO treatments are indicative of eradicant activity, which has been observed previously where crops have a very open canopy, for example, in individually trained and spaced squash plants in glasshouse trials (K.V. Wurms, unpublished data).

Rose growers tend to avoid oil-based products for PM control because of leaf burning (L. Arkesteijn, Moffatt Flower Co. Ltd, New Zealand, pers. comm.), but no phytotoxicity was observed with AMF and SBO treatments on two different rose cultivars under glasshouse or CE conditions. In contrast, chlorosis was associated with plants that had been treated with Supershield™, which was probably associated with damage caused by the PM.

Control of fungal diseases is a major constraint of cut-rose cultivation in greenhouses and transportation around the world (Koning-Boucoiran et al. 2009). The search for alternatives to synthetic fungicides is particularly important given customer preferences for some cultivars with low PM resistance, increasing pressure on growers to reduce the use of toxic pesticides (Tjosvold & Koike 2001) and the variable efficacy of natural products that are currently available on the market (Reuveni et al. 1994; Pasini et al. 1997; Dik et al.)
This study indicates that AMF and SBO give an effective, durable and non-toxic alternative to the use of pesticides. Further research will be required to test the effect of these products on mature flowering roses and outdoor roses.

ACKNOWLEDGEMENTS
Special thanks to Plant & Food Research for supporting this research programme, which has led to a provisional patent (PCT/NZ2005/000167; WO 2006/006878 A1); Leonard Arkesteijn (Moffatt Flower Co. Ltd, Christchurch, New Zealand) for provision of rose plants and valuable advice, and to John Waller (AgResearch, Hamilton, New Zealand) for statistical analyses.

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