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# Effect of fungicides, seaweed extracts, tea tree oil, and fungal agents on fruit rot and yield in strawberry

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**Summary.** Seven fungicides, 2 seaweed extracts (Maxicrop and Seasol), tea tree oil (Multicrop), and fungal agents including yeasts and an isolate of a *Trichoderma* sp., were compared for the control of fruit rots in strawberries in 5 field trials in Victoria, Australia. The fungicides tested were thiram, iprodione, dichlofluanid, chlorothalonil, fluazinam, phosphorous acid and fosetyl-aluminium. All treatments were applied as foliar sprays (at recommended rates) at weekly intervals, except for one of the *Trichoderma* treatments in which *Trichoderma* was cultured on rice and applied around plants at 1 and 5 weeks after the start of the trial. Rots were assessed after harvest by incubating fruit for 3 days at room temperature (15–25°C). Between 55 and 71% of fruit developed rot in the unsprayed plots and consisted mainly of grey mould (*Botrytis cinerea*), leak (*Rhizopus* and *Mucor* spp.), anthracnose (*Colletotrichum acutatum*), leather rot (*Phytophthora cactorum*), and stem end rot (*Gnomonia comari*).

All fungicides except fosetyl-aluminium and phosphorous acid significantly ($P<0.05$) reduced the total incidence of fruit rots by 27–72%. Thiram, dichlofluanid and chlorothalonil reduced grey mould by 61–94%, anthracnose by 63–100% and leather rot by 65–100%; iprodione reduced grey mould by 60–94% and leak by 74–96%. In one experiment each, fluazinam reduced grey mould by 85% and leather rot by 100%, and phosphorous acid reduced leather rot by 100%. Thiram, iprodione and phosphorous acid also reduced stem end rot by 55–100%. Of the biocontrols, seaweed extracts and oil, only tea tree oil in 1 trial of 3 reduced the total incidence of fruit rots significantly (by 31%), and in 2 trials significantly reduced anthracnose, and leather rot by 60–88% and 71–72% respectively. In 2 out of 3 trials, *Trichoderma* sp. reduced ($P<0.05$) grey mould by 29–63%. In one trial each, seaweed extract 1, and a yeast treatment amended with malt extract, both reduced grey mould by 40 or 54% respectively. The addition of sucrose to the yeast treatments significantly increased the incidence of anthracnose infection. Chlorothalonil, dichlofluanid, thiram and iprodione sprays increased the yield (weight) of healthy fruit significantly ($P<0.05$) compared with that from untreated plants by 43–114%. By contrast, none of the biocontrol treatments, the seaweed extracts or tea tree oil increased fruit yields.

## Introduction

In Victoria, a complex of fruit rot diseases can cause losses of 50% or more in strawberry (*Fragaria* x *ananassa* Duch.) crops (Washington *et al.* 1992). These include grey mould caused by *Botrytis cinerea* Pers.:Fr., anthracnose (or blackspot) caused by *Colletotrichum acutatum* Simmonds, leather rot caused by *Phytophthora cactorum* (Leb. & Cohn) Schroet., leak caused by *Rhizopus stolonifer* (Ehrenb.:Fr.) Vuill. and other mucoraceous fungi, and stem end rot caused by *Gnomonia comari* Karst (Washington *et al*. 1992; Washington and Shanmuganathan 1993). Despite the use of cultural practices such as planting into plastic mulches (Jenkins 1968) and postharvest practices such as low temperature handling and storage atmospheres containing high concentrations of carbon dioxide (Larsen

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and Watkins 1995), control of these diseases still relies on fungicide sprays applied during the period of flowering and fruit production.

Increasing concern by consumers and legislators over the effects of synthetic fungicides on the environment and on human health are leading to significant changes in the use of fungicides for the management of plant disease (Gullino and Kuijpers 1994; Perkins and Patterson 1997). As a consequence there is an increased emphasis on ways to minimise the use of fungicides in strawberry production (Maas and Galletta 1997). Some of the options to achieve this reduction include: breed varieties with resistance to disease (Maas 1998); management practices which minimise the development of disease (Jenkins 1968; Sutton *et al.* 1988); use of disease prediction models and a better understanding of disease epidemiology to more precisely time the application of fungicides (Madden *et al.* 1991); use of biocontrol agents as replacements for fungicides (Tronsmo and Dennis 1977; Elad and Shtienberg 1996); use of alternative treatments (e.g. oils, seaweed extracts) which may reduce the effects of disease and which are perceived to be less harmful than conventional fungicides (Cutler and Hill 1994; Northover and Schneider 1996; Wilson *et al.* 1997); use of new fungicides which may be more active requiring fewer treatments than some older fungicides (Stensvand 1997); and the use of existing fungicides at lower rates and/or at longer intervals to reduce the amount of fungicide applied to the crop (Meland 1988).

Biocontrol agents including fungi, bacteria and yeasts are being developed for control of a range of plant diseases (Wilson and Wisniewski 1989; Sutton and Peng 1993; Tronsmo and Hjelijord 1998). A project at Agriculture Victoria's Institute for Horticultural Development (IHD) at Knoxfield to develop biocontrol agents for control of postharvest pathogens in fruit has shown that a number of organisms have potential as biocontrol agents and need to be screened against *B. cinerea* and other pathogens in the field (Shanmuganathan 1991). Some of these organisms may have application as field treatments for control of strawberry fruit rots. In addition, a number of products including seaweed and plant oil extracts are proposed as natural alternatives to conventional fungicides, but limited data exist to support these claims (C. Young pers. comm.)

The work reported here compared (i) a range of fungicides for rot control, and (ii) a standard fungicide with a number of possible alternatives to fungicides that included seaweed extracts, tea tree oil and several biocontrol agents for fruit rot control in strawberry crops in Victoria, Australia. Data on the effect of these treatments on yield of healthy fruit were also collected.

## Materials and methods

*Treatments tested*

Seven fungicides, 2 seaweed extracts, tea tree oil and 4 potential biocontrol agents were tested in field trials during these studies. The fungicides tested were: thiram (Agchem Thiram 800 WP, 800 g thiram/kg, Incitec Ltd), iprodione (Rovral 500WP, 500 g iprodione 50 WP/kg, Rhone-Poulenc Rural Ltd), dichlofluanid (Euparen 500 DG, 500 g dichlofluanid/kg, Bayer Australia Ltd), chlorothalonil (Agchem Bravo 500 SC Fungicide, 500 g chlorothalonil/L, Crop Care Australasia Pty Ltd), fluazinam 500 SC, 500 g/L (as coded fungicide PP192, Crop Care Australasia Pty Ltd), phosphorous acid (Foli-R-Fos 200, 200 g phosphorous acid/L, U.I.M. Chemicals, Brisbane) and fosetyl-aluminium (Aliette WP 800 g fosetyl-aluminium/kg, Rhone-Poulenc Rural Ltd). In addition 2 seaweed extracts, referred to as seaweed extract 1 and 2 respectively {available commercially as Maxicrop plant food, [Multicrop (Australia) Pty Ltd, Bayswater, Victoria]; and Seasol plant food, [Tasbond, Bayswater, Victoria]}; tea tree oil which was supplied as Multicrop natural fungicide [a 3% emulsion of natural oils derived from *Melaleuca alternifolia* (Maiden & Betche) Cheel, Australian tea tree, Multicrop (Australia), Pty Ltd, Bayswater, Victoria]; and the potential biocontrol agents *Trichoderma* sp. (TV3) and 3 yeasts (Y-1, Y-2 and Y-3) were tested.

The 4 biocontrol agents were selected from 79 bacteria, yeasts and fungi which were screened for activity against *B. cinerea* in preliminary work at the IHD, Knoxfield (Shanmuganathan 1991). These organisms showed a 50% reduction in disease symptoms in at least one test on artificially inoculated strawberry or apple fruit. The 3 yeasts were: Y-1 (Type 3), isolated from a wooden peach box from a stone fruit packing shed, Swan Hill, Victoria (R. Holmes 26/10/88, IHD, Knoxfield), identity—unknown yeast, colour—pink on malt extract agar (MEA); Y-2 (No. 9 Hull), isolated from dairy products (R. Hull 5/12/88, CSIRO Dairy Research Laboratory, Highett), identity—*Debaryomyces* sp., colour—cream on MEA; Y-3 (No. 5 Drew), isolated from dairy products (P. Drew 20/1/89, Gilbert Chandler Dairy Research Institute, Werribee), identity—unknown yeast. One isolate of *Trichoderma* was also tested: TV3, isolated from ripe strawberry fruit, Main Ridge, Victoria (N. Shanmuganathan 11/11/88, IHD, Knoxfield), identity—*Trichoderma* sp.

Inocula of biocontrol agents were collected by adding sterile distilled water to 7-day-old cultures grown on potato dextrose agar (PDA medium) (*Trichoderma*) or potato malt dextrose agar (PMDA medium) (yeasts Y1, Y2 and Y3) in Petri dishes. The colonies were rubbed with a sterile glass rod to dislodge spores, cells and mycelium. Inoculum were prepared on the same day as sprays were applied and each was diluted to the appropriate volume (5 or 10 L) immediately before spraying, giving concentrations of $10^{11}$ or $10^{12}$ cells/100 L water. Cell concentrations were estimated by counting of subsamples under the compound microscope using a haemocytometer slide.

*Field trials*

Five trials were established on commercial strawberry farms; 3 trials were at 2 different properties at Wandin (referred to as

**Table 1. Location and description of field trials, 1989**

| Trial | Year | Location | Cultivar and age (years) | No. and timing of treatments | Spray volume (L/ha) | No. and time of rot assessments |
|---|---|---|---|---|---|---|
| 1 | Autumn 1989 | Wandin 1 | Red Gauntlet, 2 | 9, 22.iii–25.v | 4000 | 7, 17.iv–2.vi |
| 2 | Spring 1989 | Healesville | Tioga, 2 | 8, 27.ix–20.xi | 4000 | 6, 9.xi–4.xii |
| 3 | Spring 1989 | Wandin 2 | Red Gauntlet, 2 | 10, 9.x–11.xii | 2000 | 4, 27.xi–18.xii |
| 4 | Spring 1989 | Healesville | Tioga, 2 | 8, 27.ix–20.xi and TV3 on rice grains on 27.ix and 2.xi | 4000 | 6, 9.xii–4.xii |
| 5 | Spring 1989 | Wandin 2 | Red Gauntlet, 2 | 10, 9.x–11.xii and TV3 on rice grains on 9.x and 13.xi | 2000 | 4, 27.xi–18.xii |

Wandin 1, and Wandin 2, about 40 km east of Melbourne), and 2 trials were at Healesville (about 60 km east of Melbourne) (see Table 1). Plants were grown in double rows with plants in a row spaced 60 cm apart on raised beds covered with black polyethylene. Irrigation was by trickle supplemented with overhead sprinklers. Treatments were arranged in a randomised block design with plots of 10 plants, each replicated 5 times. Fifty fruit were sampled for disease assessment from each treatment (10 per plot from the middle 5 plants in each plot, which resulted in a 4–5 plant buffer between the sampled part of each plot) at each assessment time as shown in Table 1. Fruit were placed in plastic ice-block trays, one per compartment, inside sealed plastic bins lined with moist blotting paper, and incubated for 3 days at room temperature. The number and type of rotted fruit were recorded and the incidence of fruit rots was calculated by combining data from all sampling dates.

Sprays were applied with a hand-operated knapsack sprayer at 5–8-day intervals from flowering. Fungicides were sprayed to runoff (2000–4000 L/ha) at recommended rates as shown in Tables 2, 3 and 4. Commercial practice is to apply about 1000–1500 L/ha. Tea tree oil was applied at a concentration of 1.5% of natural oils. Biocontrol agents were applied as sprays at rates shown in Tables 2, 5 and 6, except for TV3 plus rice grain, which was *Trichoderma* sp. cultured on sterilised rice. This was spread over each plant at the start of the trial, and again after 5 weeks, at a dose of 50 g per plant. Supplements were added to some biocontrol treatments in an attempt to increase their survival on the plant (Tronsmo and Hjeljord 1998). These were 1% yeast extract (Oxoid LPO 21B) added to TV3 sprays, or as a spray alone applied to plots treated with TV3 plus rice grain; 1% sucrose added to Y2 or Y3 sprays; 1% malt extract (desiccated Oxoid LPO 39B) added to Y2 or Y3 sprays. The weight of ripe healthy fruit was measured from trials 2, 3 and 4 on 22 and 28 November (trial 2), on 30 November and 7 December (trial 3) and on 17 November (trial 4).

*Statistical analysis*

Data on rot incidence from each trial were subjected to analysis of variance (ANOVA) using the GENSTAT 5 statistical package [Lawes Agricultural Trust (Rothamsted Experimental Station)]. An angular transformation was applied to the proportions ($p$) to stabilise the variance, the equation being:

$$x = 180/\pi.\ \text{arcsine}(\sqrt{p}).$$

## Results

In untreated plots of trial 1, *B. cinerea* caused 35.1% of fruit rots, followed by *R. stolonifer* and *Mucor* spp. (10.9%), and *P. cactorum* (3.1%). Total incidence of

**Table 2. Trial 1. Effect of fungicides, seaweed extracts, tea tree oil and biocontrol treatments on the incidence of fruit rot of strawberry at Wandin 1**

Total includes other minor and/or unidentified rots
Values in parentheses are means of the arcsine-transformed proportions

| Treatment | Rate (a.i./100 L) | Grey mould (%) | Leather rot (%) | Leak (%) | Other (%) | Total (%) |
|---|---|---|---|---|---|---|
| Thiram | 120 g | 7.1 (0.27) | 0.3 (0.02) | 3.1 (0.15) | 11.1 (0.31) | 21.7 (0.42) |
| Iprodione | 50 g | 14.0 (0.38) | 6.0 (0.20) | 2.6 (0.12) | 14.3 (0.38) | 36.9 (0.65) |
| Dichlofluanid | 100 g | 5.1 (0.21) | 0.3 (0.02) | 7.4 (0.27) | 16.9 (0.40) | 29.7 (0.56) |
| Fluazinam | 50 mL | 5.4 (0.21) | 0 (0) | 5.4 (0.20) | 24.9 (0.52) | 35.4 (0.63) |
| Chlorothalonil | 150 mL | 12.9 (0.36) | 1.1 (0.08) | 7.7 (0.26) | 16.3 (0.41) | 38.0 (0.66) |
| Seaweed extract 1 | 100 mL | 21.1 (0.48) | 3.7 (0.17) | 7.7 (0.26) | 11.4 (0.34) | 44.0 (0.72) |
| Seaweed extract 2 | 100 mL | 28.0 (0.56) | 3.1 (0.16) | 8.9 (0.29) | 10.6 (0.33) | 50.6 (0.79) |
| Tea tree oil | 1.5 L | 25.7 (0.53) | 0.9 (0.06) | 5.1 (0.19) | 8.6 (0.27) | 40.3 (0.68) |
| Y-1 | $10^{12}$ cells | 28.6 (0.56) | 2.3 (0.11) | 9.1 (0.30) | 8.0 (0.28) | 48.0 (0.76) |
| TV3 | $10^{11}$ cells | 24.9 (0.51) | 4.3 (0.18) | 8.9 (0.28) | 8.6 (0.29) | 46.6 (0.75) |
| Untreated | | 35.1 (0.63) | 3.1 (0.18) | 10.9 (0.31) | 9.1 (0.30) | 58.3 (0.87) |
| l.s.d. ($P$ = 0.05) | | (0.12) | (0.11) | (0.12) | (0.12) | (0.17) |

**Table 3. Trial 2. Effect of fungicides, tea tree oil and seaweed extract 1 on the incidence of fruit rot and fruit yield of strawberry at Healesville**

Total includes other minor and/or unidentified rots
Weight of healthy fruit was calculated from fifty plants per treatment from two harvests
Values in parentheses are means of the arcsine-transformed proportions

| Treatment | Rate (a.i./100 L) | Grey mould (%) | Leather rot (%) | Anthracnose (%) | Stem end rot (%) | Leak (%) | Total (%) | Weight of healthy fruit (kg) |
|---|---|---|---|---|---|---|---|---|
| Thiram | 120 g | 2.0 (0.11) | 0.3 (0.03) | 2.7 (0.16) | 3.0 (0.17) | 3.3 (0.14) | 15.0 (0.39) | 5.46 |
| Iprodione | 50 g | 2.3 (0.13) | 3.0 (0.15) | 21.0 (0.47) | 0.7 (0.05) | 2.0 (0.08) | 36.0 (0.64) | 4.20 |
| Dichlofluanid | 100 g | 1.0 (0.06) | 0 (0) | 1.0 (0.06) | 10.0 (0.32) | 3.3 (0.17) | 19.3 (0.45) | 5.51 |
| Chlorothalonil | 150 mL | 2.0 (0.11) | 0 (0) | 1.7 (0.10) | 9.7 (0.32) | 1.7 (0.11) | 18.0 (0.44) | 6.27 |
| Phosphorous acid | 100 mL | 23.7 (0.50) | 0 (0) | 20.3 (0.46) | 6.0 (0.25) | 3.7 (0.17) | 57.3 (0.86) | 3.70 |
| Tea tree oil | 1.5 L | 24.7 (0.52) | 1.3 (0.09) | 7.7 (0.24) | 10.0 (0.31) | 4.7 (0.22) | 56.0 (0.85) | 3.62 |
| Seaweed extract 1 | 100 mL | 11.3 (0.34) | 3.3 (0.14) | 22.0 (0.48) | 16.3 (0.40) | 5.3 (0.20) | 62.3 (0.91) | 3.18 |
| Untreated | | 13.0 (0.36) | 4.7 (0.21) | 19.3 (0.44) | 13.3 (0.37) | 4.3 (0.18) | 63.0 (0.92) | 2.93 |
| l.s.d. ($P$ = 0.05) | | (0.11) | (0.11) | (0.11) | (0.08) | n.s. | (0.11) | 1.16 |

rotted fruit was 58.3%, which included minor and unidentified rot (9.1%) (Table 2). Each of the 5 chemical fungicides, and tea tree oil, reduced the incidence of total fruit rot compared with the untreated control. Thiram gave the best control of rot and was significantly ($P$<0.05) better than iprodione, chlorothalonil and fluazinam. Dichlofluanid, fluazinam and thiram gave good control of grey mould, while chlorothalonil, iprodione, seaweed extract 1 and the *Trichoderma* spray were less effective. Iprodione and thiram were the only treatments to control leak, while fluazinam, dichlofluanid, thiram and tea tree oil controlled leather rot. Fluazinam was associated with a high level of other (largely unidentified) rots.

In trial 2, *C. acutatum* was the most common cause of fruit rot (19.3%) in the untreated plots, followed by *B. cinerea* (13.0%), *P. cactorum* (4.7%) and *R. stolonifer* and *Mucor* spp. (4.3%). A total of 63.0% of the fruit was rotted in untreated plots when combined with other unidentified causes of rot (Table 3). Dichlofluanid, chlorothalonil, thiram and iprodione were the most effective fungicides for controlling grey mould. Grey mould developed more in plots treated with phosphorous acid or tea tree oil than in the untreated plots. The fungicides (except iprodione) and tea tree oil each controlled leather rot. Similarly, the fungicides (except iprodione and phosphorous acid), and tea tree oil controlled anthracnose. None of the treatments significantly controlled leak. Thiram, chlorothalonil and dichlofluanid reduced ($P$<0.05) the total incidence of fruit rots more than did iprodione, although all these treatments reduced the overall level of fruit rot compared

**Table 4. Trial 3. Effect of fungicides, tea tree oil and seaweed extract 1 on the incidence of fruit rot and fruit yield of strawberry at Wandin 2**

Total includes other minor and/or unidentified rots
Weight of healthy fruit was calculated from fifty plants per treatment from two harvests
Values in parentheses are means of the arcsine-transformed proportions

| Treatment | Rate (a.i./100 L) | Grey mould (%) | Anthracnose (%) | Leak (%) | Total rot (%) | Weight of healthy fruit (kg) |
|---|---|---|---|---|---|---|
| Thiram | 120 g | 0.5 (0.03) | 1.0 (0.05) | 8.0 (0.27) | 17.5 (0.41) | 3.74 |
| Iprodione | 50 g | 0.5 (0.03) | 12.5 (0.24) | 0.5 (0.03) | 23.0 (0.46) | 3.63 |
| Dichlofluanid | 100 g | 1.5 (0.06) | 0.5 (0.03) | 15.0 (0.36) | 23.5 (0.48) | 4.02 |
| Chlorothalonil | 150 mL | 1.0 (0.06) | 0 (0) | 14.0 (0.35) | 26.0 (0.53) | 2.25 |
| Fosetyl-aluminium | 100 g | 9.5 (0.30) | 17.0 (0.36) | 10.5 (0.32) | 49.5 (0.78) | 3.41 |
| Tea tree oil | 1.5 L | 17.5 (0.43) | 4.0 (0.15) | 19.5 (0.43) | 47.5 (0.76) | 2.70 |
| Seaweed extract 1 | 100 mL | 13.0 (0.34) | 18.5 (0.38) | 18.0 (0.43) | 62.0 (0.91) | 3.01 |
| Untreated | | 9.0 (0.29) | 34.5 (0.61) | 11.5 (0.33) | 63.5 (0.93) | 2.64 |
| l.s.d. ($P$ = 0.05) | | (0.15) | (0.27) | (0.19) | (0.22) | 1.07 |

**Table 5. Trial 4. Effect of sprays of biocontrol agents compared with a fungicide on the incidence of fruit rot and fruit yield of strawberry at Healesville**

Total includes other minor and/or unidentified rots
Weight of healthy fruit was calculated from fifty plants per treatment from one harvest
Values in parentheses are means of the arcsine-transformed proportions

| Treatment | Rate (a.i./100 L) | Grey mould (%) | Leather rot (%) | Anthracnose (%) | Stem end rot (%) | Leak (%) | Total rot (%) | Weight of healthy fruit (kg) |
|---|---|---|---|---|---|---|---|---|
| Thiram | 120 g | 1.3 (0.07) | 0.7 (0.05) | 9.0 (0.27) | 4.0 (0.18) | 1.0 (0.08) | 22.7 (0.49) | 2.43 |
| TV3 | $10^{10}$ spores | 3.0 (0.15) | 3.0 (0.15) | 47.7 (0.76) | 9.0 (0.30) | 1.3 (0.09) | 68.7 (0.99) | 1.59 |
| TV3 + 1% yeast extract | $10^{10}$ spores | 7.0 (0.25) | 10.3 (0.32) | 37.7 (0.66) | 13.3 (0.37) | 1.7 (0.10) | 75.3 (1.06) | 1.48 |
| TV3 + rice grain + 1% yeast extract | 50 g/plant | 12.7 (0.36) | 2.7 (0.14) | 24.7 (0.52) | 15.3 (0.39) | 4.3 (0.20) | 68.0 (0.97) | 1.30 |
| Y-2 | $10^{11}$ cells | 7.3 (0.25) | 4.0 (0.12) | 36.0 (0.63) | 11.7 (0.34) | 4.0 (0.17) | 70.3 (1.00) | 1.42 |
| Y-2 + 1% sucrose | $10^{11}$ cells | 6.3 (0.21) | 4.3 (0.18) | 52.0 (0.81) | 7.3 (0.26) | 2.7 (0.14) | 77.0 (1.09) | 1.52 |
| Y-2 + 1% malt extract | $10^{11}$ cells | 3.7 (0.17) | 7.3 (0.27) | 50.0 (0.78) | 7.0 (0.26) | 2.3 (0.13) | 74.0 (1.04) | 1.56 |
| Untreated | | 8.0 (0.28) | 6.7 (0.20) | 35.3 (0.63) | 15.3 (0.39) | 3.0 (0.17) | 70.7 (1.00) | 1.50 |
| l.s.d. ($P$ = 0.05) | | (0.09) | (0.14) | (0.18) | (0.13) | n.s. | (0.15) | 0.48 |

with the untreated plots. Yields of healthy fruit were significantly ($P$<0.05) higher in plots treated with fungicides, than in untreated plots.

In untreated plots of trial 3, total rot incidence (63.5%) was similar to that in trial 2, but the proportions of specific rots differed (34.5% anthracnose, 11.5% leak and 9% grey mould, Table 4). No leather rot was found at this site. Iprodione, thiram, chlorothalonil and dichlofluanid controlled grey mould. All fungicides except fosetyl-aluminium controlled anthracnose, although iprodione was the least effective. Only iprodione controlled leak. All fungicides except fosetyl-aluminium reduced total fruit rots but only dichlofluanid and thiram treatments increased ($P$<0.05) fruit weight of the first harvest.

In trial 4, 70.7% of the fruit developed rot in the untreated plots (Table 5). The same diseases occurred in this trial as in trial 2, which was in the same planting on the same property. Thiram, TV3 alone and Y-2 yeast plus malt extract controlled grey mould. Only thiram controlled anthracnose and leather rot, while thiram and Y-2 yeast plus malt extract both reduced stem end rot ($P$<0.05). The Y-2 yeast plus sucrose tended to increase anthracnose compared with the untreated control ($P$ = 0.054). When all rots were combined, only the thiram treatment reduced rots compared with rots in the untreated control plots. Similarly, only thiram resulted in an increased fruit weight compared with the fruit weight in the untreated control.

In trial 5, 55% of fruit developed rots in the untreated plots (Table 6). Leak (26%), followed by grey mould (9%), anthracnose (8%) and stem end rot (2%) were

**Table 6. Trial 5. Effect of sprays of biocontrol agents compared with a fungicide on the incidence of fruit rot of strawberry at Wandin 2**

Total includes other minor and/or unidentified rots
Values in parentheses are means of the arcsine-transformed proportions

| Treatment | Rate (a.i./100 L) | Grey mould (%) | Anthracnose (%) | Stem end rot (%) | Leak (%) | Total rot (%) |
|---|---|---|---|---|---|---|
| Thiram | 120 g | 3.5 (0.15) | 3.0 (0.13) | 0 (0) | 12.0 (0.33) | 32.0 (0.60) |
| TV3 | $10^{11}$ spores | 10.5 (0.31) | 11.5 (0.28) | 2.0 (0.09) | 11.5 (0.28) | 57.5 (0.86) |
| TV3 + 1% yeast extract | $10^{11}$ spores | 6.0 (0.19) | 15.5 (0.36) | 1.0 (0.06) | 15.5 (0.36) | 56.5 (0.85) |
| TV3 + rice grain | 50 g/plant | 10.0 (0.32) | 9.5 (0.28) | 1.5 (0.08) | 16.5 (0.39) | 49.5 (0.78) |
| Y-3 | $10^{12}$ cells | 6.0 (0.24) | 16.5 (0.40) | 2.5 (0.12) | 24.5 (0.51) | 56.0 (0.85) |
| Y-3 + 1% sucrose | $10^{12}$ cells | 3.5 (0.19) | 29.5 (0.56) | 2.0 (0.11) | 20.5 (0.45) | 69.0 (0.98) |
| Y-3 + 1% malt extract | $10^{12}$ cells | 6.5 (0.25) | 13.5 (0.35) | 2.5 (0.14) | 21.5 (0.45) | 59.0 (0.88) |
| Untreated | | 9.0 (0.30) | 8.0 (0.28) | 2.0 (0.11) | 26.0 (0.53) | 55.0 (0.84) |
| l.s.d. ($P$ = 0.05) | | (0.15) | (0.23) | n.s. | (0.21) | (0.13) |

most common in the untreated plots. Only thiram reduced grey mould; anthracnose was increased by Y-3 yeast plus sucrose, and TV3 reduced leak when compared with the control. When all rots were combined only thiram significantly ($P<0.05$) reduced levels to less than those in the untreated control plots.

**Discussion**

This work has shown that all the fungicides tested except fosetyl-aluminium and phosphorous acid gave fair to good control of the fruit rot complex which occurs in strawberry plantings in Victoria. Some of these fungicides increased yields of healthy fruit by up to 114% (from 2.9 to 6.3 kg per 50 plants). Tea tree oil and *Trichoderma* were the best of the other treatments tested, but only controlled some of the rot complex and failed to increase fruit yields. This is the first report of the activity of tea tree oil and *Trichoderma* on strawberry rots in Australia. Thiram, which controlled a broad range of rots (grey mould, anthracnose, leather rot and stem end rot) was the most effective fungicide and increased healthy fruit weight in all 3 trials in which yield data were collected. Other effective treatments also controlled several rots; for example, chlorothalonil and dichlofluanid controlled grey mould, leather rot and anthracnose, while iprodione controlled grey mould in most trials, leak and stem end rot and in one trial showed limited control of anthracnose. By contrast, phosphorous acid only controlled leather rot and stem end rot.

*Trichoderma* reduced grey mould in 2 of the 3 trials in which it was tested. Isolates of various *Trichoderma* spp. were at least partially effective against grey mould in earlier reports (Tronsmo and Dennis 1977; Peng and Sutton 1991; Elad and Zimand 1992; Elad and Shtienberg 1996; Stensvand 1997), and our work confirms the potential of *Trichoderma* sp. for control of grey mould under conditions found in Victoria. However, this agent was less effective than fungicides and, as it had little or no effect on the other diseases that developed in these trials, yields of healthy fruit were significantly less than those from the thiram treatments. Tronsmo (1991) showed that a fungicide-resistant isolate of *T. harzianum* controlled dry eye rot of apple better than a fungicide-sensitive isolate and suggested that an integrated approach of combining the biocontrol agent with low rates of fungicide may give more consistent control. Elad and Shtienberg (1996) showed that alternating sprays of a commercially formulated preparation of *Trichoderma harzianum* (T39) (Trichodex 25DP, Makteshim Chemical Works Ltd, Israel) with a fungicide effectively controlled grey mould on strawberries and other crops, especially when used with a weather-based decision support system (BOTMAN) for managing *Botrytis* (Shtienberg and Elad 1997). Such integrated approaches may reduce the use of fungicide while improving the control over that of the biocontrol agent alone, and would be worth developing provided that significant control of other rots is also achieved. None of the biocontrol agents used in our study were specifically formulated for application to plants, and it is possible that such formulation could also improve the level of control, as has been shown for bioherbicides (Green *et al.* 1998). Nutrient supplements such as chitin or chitosan may be more appropriate for use with *Trichoderma*, as they can be utilised by *Trichoderma* but much less readily by many fungal pathogens (Tronsmo and Hjeljord 1998). Another factor which may have restricted the effectiveness of the *Trichoderma* (and possibly the other biocontrol treatments) may be the timing of applications. Most treatments were applied mid morning, allowing long drying times which could have restricted germination of conidia. Such treatments may be better applied close to dusk allowing slower drying times of the spray droplets which could improve germination and establishment of the biocontrol.

One of the yeasts (Y-2) plus malt extract reduced grey mould, but otherwise the yeast treatments had little effect on the control of any diseases found in these trials. Studies by Sutton (1995) showed that yeasts were generally not effective against strawberry fruit rots. Seaweed extract 1 reduced grey mould in only 1 out of 3 trials, which indicates that it has limited potential as a substitute for fungicide control of rot. Jeandet *et al*. (1996) showed that a mixture of a seaweed extract and hydrated aluminium chloride elicited phytoelexins in grapevine. They found that the seaweed extract mixture when combined with iprodione sprays increased the control of grey mould on grapes, although the contribution made by the seaweed extract to this control is not clear.

Tea tree oil showed good control of leather rot and anthracnose, but not of grey mould. In trial 1, tea tree oil and the fungicides dichlofluanid and fluazinam also showed significant suppression of 2 spotted mite (*Typhlodromus occidentalis*) (Washington *et al*. 1991). Other workers have shown the activity of tea tree oil against several plant pathogens. Bishop and Reagan (1998) showed that tea tree oil was active *in vitro* against *B. cinerea* at a rate of 3.2%. Wilson *et al*. (1997) showed that the volatile effect of tea tree oil prevented

germination of spores of *B. cinerea* at 12.5% but not at 6.25%. Glasshouse tests have shown that foliar sprays of 1% tea tree oil controlled false smut of palms caused by *Graphiola phoenicis* (Polizzi and Agosteo 1995). However, in our study, foliar sprays at a rate of 1.5% tea tree oil failed to control grey mould or increase the yield of healthy fruit.

Both phosphorous acid and tea tree oil in trial 2 were associated with increased incidence of grey mould. Yeast treatment with sucrose supplements (which were intended to provide a nutrient source for the yeast biocontrol) increased anthracnose infection, by comparison with that in untreated plots. Grover (1971) showed that in laboratory studies sucrose stimulated spore germination and appressoria formation in *C. piperatum*. In our study, sucrose may have stimulated the conidial germination, formation of appressoria and infection of *C. acutatum*, which may explain the increase in anthracnose following the yeast and sucrose treatment. The sucrose supplement may also have stimulated the competitive microflora on the fruit (Taylor and Harman 1990), restricting the development of *Trichoderma*.

Although iprodione reduced grey mould in trial 1, both dichlofluanid and fluazinam were significantly ($P<0.05$) more effective. This may have been due to populations of *B. cinerea* being resistant to dicarboximide fungicides, which have previously been reported in Victorian strawberry crops (Washington *et al*. 1992), but were not tested in this experiment.

The incidence of different rots varied between trials. The high levels of grey mould in trial 1 but not in any of the other trials (grey mould in the untreated controls of 35.1% in trial 1 compared with a maximum of 13% in trials 2–5) was probably associated with weather conditions favouring grey mould in trial 1. This trial was carried out in autumn and early winter (when temperatures are lower) while the other trials were undertaken in spring and early summer when higher temperatures favoured anthracnose (from 8 to 35.3% in trials 2–5, but little or none in trial 1) (Maas 1998).

These results demonstrate that regular use of fungicides controls fruit rot and increases the yield of healthy fruit. They also indicate that some of the alternative treatments (such as tea tree oil and *Trichoderma* sp.) showed potential for controlling some of the fruit rots, and in the case of tea tree oil, can reduce the overall incidence of rotted fruit. More research is required to determine if they can give similar yields as those obtained with fungicides. Integrated control strategies that warrant further investigation include the use of low rates of fungicides in combination with *Trichoderma* or tea tree oil; the combination of *Trichoderma* with tea tree oil; or the alternation of *Trichoderma* or tea tree oil with fungicides, with decisions of which material to use based on support systems which estimate the risk of infection.

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