

Improved Management of Charcoal Rot of Strawberry in Australia with Soil Fumigation and Totally Impermeable Film

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ABSTRACT. Charcoal rot of strawberry [*Fragaria × ananassa* (Duchesne)] caused by the soil-borne fungus *Macrophomina phaseolina* (Tassi) Goid results in significant plant losses each year in Australia. Before the current research, industry applied the soil fumigants 1,3-dichloropropene and chloropicrin in combination to the bed (strip fumigation) under low-density polyethylene film (LDPE) as the standard practice to manage the disease. We conducted column and field experiments to evaluate soil fumigation with 1,3-dichloropropene/chloropicrin under totally impermeable film (TIF) and broadcast application (treatment of beds and inter-rows) for control of *M. phaseolina* and charcoal rot compared with the previous standard practice. The results showed that treatment under TIF increased the concentration × time (CT) values of fumigants in soil by up to 38% compared with those of LDPE. Strip fumigation under TIF reduced DNA concentrations of *M. phaseolina* in soil (0–10 cm) by 65%, charcoal rot by 93%, and increased fruit yields by 37% compared with application under LDPE. Furthermore, broadcast fumigation reduced DNA concentrations of *M. phaseolina* by an average of 55%, charcoal rot by 65%, and increased fruit yields by 40% compared with those of strip fumigation under LDPE. This work demonstrates the effectiveness of improved fumigation practices for minimizing the impact of charcoal rot in strawberry.

The cultivation of strawberries (*Fragaria × ananassa*) has considerable significance within the Australian horticultural landscape. In 2023, the industry produced 65,824 metric tons of fruit valued at AU\$429 million (Horticulture Innovation Australia 2023). Production occurs in every Australian state, but major regions in Queensland and Victoria collectively account for approximately 70% of the total output. The fruit sector in Victoria mostly relies on day-neutral cultivars of strawberry such as Albion and Cabrilla. These cultivars flower in response to moderate temperatures centered over the Victorian summer, with a fruiting season spanning from October to May. Conversely, the fruit sector in Queensland predominantly grows short-day cultivars of strawberry such as Red Rhapsody and Festival. These cultivars flower in response to short daylength and mild temperatures centered over the Queensland winter, with a fruiting season from May to October. This strategic distribution of production across various regions ensures a consistent year-round supply of strawberries to meet the

demands of the domestic market in Australia.

Charcoal rot is a disease of strawberry that affects the crown of the plant and is caused by the soil-borne fungus *Macrophomina phaseolina* (Tassi) Goid (Chamorro et al. 2016; Hutton et al. 2013). Charcoal rot causes wilting and plant death in strawberry and can devastate crop production with estimates of disease incidence as high as 20% in some states of Australia (McFarlane et al. 2019b). Charcoal rot of strawberry has been recorded in most continents, including Europe (Pastrana et al. 2017), Asia (Sharifi and Mahdavi 2012), North America and South America (Baino et al. 2011; Koike et al. 2016), and Australia (Fang et al. 2011b; Golzar et al. 2007; Hutton et al. 2013; Mattner et al. 2014). The incidence of charcoal rot in strawberry in Australia has increased since the soil fumigant methyl bromide (MeBr) was withdrawn from use in 2006 (Mattner et al. 2014; McFarlane et al. 2019b). MeBr was phased out for nonquarantine and pre-shipment uses under the Montreal Protocol because emissions are implicated

in degradation of stratospheric ozone (Butler 1995). Following the withdrawal of MeBr, strawberry industries across the globe adopted substitute practices for soil disinfestation to control soil-borne pathogens and weeds, such as alternative fumigants (Porter et al. 2004), steam (Fennimore and Goodhue 2016), biofumigation (Mattner et al. 2008), and anaerobic soil disinfestation (Roskopf et al. 2017). In Australia, most strawberry growers adopted strip fumigation (treatment of the strawberry beds but not the inter-rows) with mixtures of 1,3-dichloropropene (1,3-D) and chloropicrin (Pic) sealed with low-density polyethylene (LDPE) following the phase-out of MeBr (McFarlane et al. 2019a). However, this practice is less effective than MeBr/Pic at controlling *M. phaseolina* in plant debris, such as old strawberry crowns, found in the soil (Hutton et al. 2013).

Fumigants are chemicals that act in the gaseous phase and are toxic to many organisms, including soil-borne pathogens. To optimize the effect of soil disinfestation, fumigants should move evenly through soil and have adequate concentration × time (CT) values to kill all target pests. There are various fumigation practices that can increase the CT value of fumigants and their distribution through soil, including the type of plastic film used to seal the soil (Gao et al. 2012). Plastic films used for these purposes can be grouped into LDPE, high-density polyethylene, virtually impermeable film (VIF), and totally impermeable film (TIF) based on their composition and permeability to gaseous compounds, such as fumigants (Fennimore and Ajwa 2011). In addition to sealing fumigants in soil, strawberry growers use films as mulches on beds to retain soil moisture, manage soil temperatures for improved plant growth, suppress weed emergence, and protect fruit from soil contamination (Shiukhy et al. 2015).

Virtually impermeable films are plastic laminates that contain a layer of polyamide, whereas TIFs contain a layer of ethylene vinyl alcohol that is impermeable to gases. The degree that plastic films allow fumigants to diffuse through them can be attributed to their mass transfer coefficient (*h*) and is specific for each compound (Papiernik et al. 2001); a low *h* value indicates

lower permeability. Based on a study by Qian et al. (2011), the average h values for LDPE, VIF, and TIF were 4.4, 0.006, and 0.0003 cm/h for *cis*-1,3-D, respectively. Similar trends between these three film types have been shown for the fumigants *trans*-1,3-D and Pic (Qian et al. 2011).

Because of their low mass transfer coefficients, VIF and TIF are capable of sealing fumigants in the soil for a longer period than that of LDPE (McAvoy and Freeman, 2013a). Therefore, VIF and TIF films require lower rates of fumigants per unit area to achieve the same CT values as fumigation under LDPE (Gao et al. 2012; Samtani et al. 2010). Compared with LDPE, TIFs can increase the concentration of fumigants in soil (Fennimore and Ajwa 2011) and their distribution through the bed (Qin et al. 2011) when applied at the same rates. The low permeability of VIF and TIF also reduces emissions of fumigants to the atmosphere and may lower the exposure risks of operators and the community (Gao et al. 2012; Qian et al. 2011).

The use of TIF or VIF can increase the effectiveness of fumigants for weed control (Fennimore and Ajwa, 2011; Gao et al. 2012; McAvoy and Freeman, 2013b; Samtani et al. 2010; Stevens et al. 2016; Thalavaisundaram

et al. 2015) and pathogen and disease control (Song et al. 2020; Washington 2019) and increase yield (Fennimore and Ajwa 2011) in strawberry crops compared with LDPE or high-density polyethylene. For example, Washington (2019) showed that strip fumigation with a mixture of 1,3-D and Pic (40:60) reduced the incidence of charcoal rot by more than 20% compared with application under LDPE. However, no study has determined the efficacy of fumigants applied under TIF for control of *M. phaseolina* in soil or charcoal rot in strawberry plants in Australia.

Fumigants are injected into soil using two main methods: broadcast (broadcast) and strip (bed) fumigation. Broadcast fumigation involves treatment of the entire field before bed formation, including soils that will form the inter-rows. With this method, overlapping strips of film are held in place with adhesive glue or hot-melted together. When the film is removed, the strawberry beds are formed and covered again with a plastic mulch. With strip fumigation, only the soil in the raised beds, and not the inter-rows, is treated. The fumigant is injected into the beds and immediately sealed with film, which remains in place for the entire season. Broadcast fumigation is typically more expensive than strip fumigation because of the extra resources required (i.e., films, fumigants, labor).

Studies conducted by Wilhelm (1965), a pioneer of research in soil disinfection, and Wilhelm et al. (1974) found that broadcast application was more effective than strip fumigation at controlling soil-borne pathogens, particularly *Verticillium dahliae*. This was reportedly caused by the inferior distribution of fumigants through the bed when using the strip method. However, no published study has directly compared the effectiveness of broadcast and strip fumigation for control of *M. phaseolina* in soil and management of charcoal rot of strawberry.

We conducted a series of field and soil column experiments to evaluate the effectiveness of 1,3-D/Pic using TIF compared with LDPE and broadcast compared strip fumigation for control of *M. phaseolina*, charcoal rot and weeds, and strawberry production. We also investigated the ability of TIF to increase the CT values and distribution of 1,3-D/Pic in soil compared with LDPE.

Materials and methods

Soil column experiment

COLUMN CONSTRUCTION AND DESIGN. Cylindrical columns were constructed from 3-mm-thick high-density polyethylene with a height of 40 cm and a diameter of 28 cm. Two bulk-head fittings were attached to the columns at depths of 10 cm and 25 cm from the top to provide gas-tight sampling ports that were accessible on the exterior surface of the columns.

The soil (silty clay: 26% silt, 41% clay, 33% sand) used to fill each column was obtained from a commercial strawberry farm in Silvan, Victoria, Australia (lat. 37°49'0.282"S, long. 145°24'37.335"E) with a concentration of *M. phaseolina* DNA of 40,000 DNA copies/g soil, a field capacity of 60%, a pH of 6.5, and an organic matter content of 2.4% (same site as the 2018–20 field experiment). A total of 26 ± 1 kg of soil was added to fill each column. Soil was added to the columns at 10-cm depths at a time and packed to mimic the predetermined bulk density of soil in prepared strawberry beds in the field. A cylindrical brass probe (length, 26 cm; diameter, 4 mm) (Nicholls et al. 1999) was inserted into each of the sampling ports and tightened with the bulk-head fittings to prevent gas leaks. The exterior side of the probes were temporarily closed with Teflon tubes that could be removed for sampling.

The soil surface of 10 of the columns were sealed with LDPE film (thickness, 30 µm), and five were sealed with TIF (thickness, 30 µm). Both films were manufactured by Richdale Plastics Pty Ltd. (Mentone, Victoria, Australia). Both films were sealed to the chambers with an adhesive fumigation tape (Venhart, Melbourne, Victoria, Australia). Fumigation of soil in the columns simulated shank injection in the field (see the Fumigation, crop agronomy, and design section of this article). In this procedure, a mixture of 1,3-D/Pic (20:80) (Tri-Form® 80; R&R Fumigation, Bayswater, Victoria, Australia) was manually injected into the center of the column to a soil depth of 5 cm using a hypodermic needle and syringe (Terumo Australia; Macquarie Park, New South Wales, Australia) at a rate equivalent to 400 kg/ha. Half of the chambers sealed with LDPE film and all chambers sealed with TIF film were fumigated. Soil temperature at a

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Table 1. Concentration \times time values (mg/L/h) of chloropicrin in soil at different depths when sealed with totally impermeable film (TIF) or low-density polyethylene (LDPE) in a column experiment (2–168 h after fumigation). Values assigned different letters in each column are significantly different, where $P \leq 0.05$. The 1,3-dichloropropene data were \log_{10} -transformed for analysis and back-transformed in the table.

Barrier film	Sampling depth	Chloropicrin	1,3-dichloropropene
LDPE	5 cm	5987 a	1314 a
	25 cm	5422 a	1315 a
TIF	5 cm	9617 b	2032 b
	25 cm	8894 b	1945 b

depth of 5 cm at fumigation was 10 °C and columns were placed outside under cover for the duration of the experiment. The columns were arranged as a randomized complete block design with five replicates.

VIABILITY OF *M. PHASEOLINA* IN BURIED CROWNS. Strawberry crowns were collected from plants showing symptoms of charcoal rot at field sites that had a history of the disease, including Silvan (lat. 37°49'07.0"S, long. 145°24'34.2"E) and Coldstream (lat. 37°43'25.2"S, long. 145°23'07.6"E), Victoria, Australia. Sub-samples of the collected crowns from each site ($n = 10$) were destructively tested in the laboratory to confirm that they were infected with *M. phaseolina*. Before fumigation, muslin bags containing five crowns from plants showing symptoms of charcoal rot were placed into each column at soil depths of

10 cm and 25 cm. Two weeks postfumigation, the muslin bags containing the crowns were recovered.

To test for the viability of *M. phaseolina*, crowns were broken apart under aseptic conditions to expose their xylem tissue. Four pieces of xylem tissue were removed from each crown using secateurs, surface-sterilized (1% NaOCl, 10 s), and placed on a petri dish containing potato dextrose agar (half strength) and 250 µg/L chloramphenicol. The plates were incubated at 22 °C and 30% humidity for 7 d, and fungi growing from the crowns onto the potato dextrose agar were morphologically assessed under a compound microscope. The presence of a *M. phaseolina* isolate or isolates confirmed the viability of each sample.

FUMIGANT CONCENTRATIONS. Gastec 131La and 134 detector tubes

(Gastech Australia, Wangara, Western Australia) were attached to the sampling ports of the chambers using Teflon tubing. Then, soil air was sucked through the detector tubes using a 60-mL hypodermic syringe to determine the respective concentrations of 1,3-D and Pic. This method was adapted from the approach described by Chellemi and Mirusso (2006). Each sampling port was sealed between readings. Concentration measurements for 1,3-D and Pic were adjusted using correction factors recommended by the Gastec manufacturer and recorded at 2 h, 24 h, 72 h, and 168 h after treatment. The area under the curve (fumigant concentration \times time) was used to calculate the CT values (mg/L/h) for each replicate from 2 to 168 h.

***M. PHASEOLINA* CONCENTRATIONS IN SOIL.** Soil samples (500 g) were taken from each column at 14 d post-fumigation. Soil samples were taken from depths of 0 to 5 cm and 25 to 30 cm with the use of a disinfested hand trowel (composites of 10 sub-samples). Soil samples were submitted to a commercial laboratory [South Australian Research and Development Institute (SARDI), Waite Campus, Adelaide, South Australia], DNA were extracted, and a quantitative polymerase chain reaction was performed for *M. phaseolina* using the general procedures described by Ophel-Keller et al. (2008). The efficiency and consistency of the SARDI method in comparison with commercial extraction kits has been confirmed (Haling et al. 2011).

Silvan field experiment (2018–20)

FUMIGATION, CROP AGRONOMY, AND DESIGN. A field trial was conducted on a commercial strawberry farm at Silvan, Victoria, Australia (lat. 37°49'07.0"S long. 145°24'34.2"E) with a known history of charcoal rot. Before fumigation, soil was sampled (500 g sub-sample) at depth of 0 to 10 cm and 30 to 40 cm in a "W" pattern across the trial site (30 m \times 110 m). The concentration of *M. phaseolina* DNA in each sample was determined using quantitative polymerase chain reaction analyses conducted by SARDI (see *M. phaseolina* concentrations in soil section in this article).

The soil was prepared for fumigation by rotary-hoeing to a depth of 25 cm. Broadacre fumigation was conducted during summer (Jan 2018) at

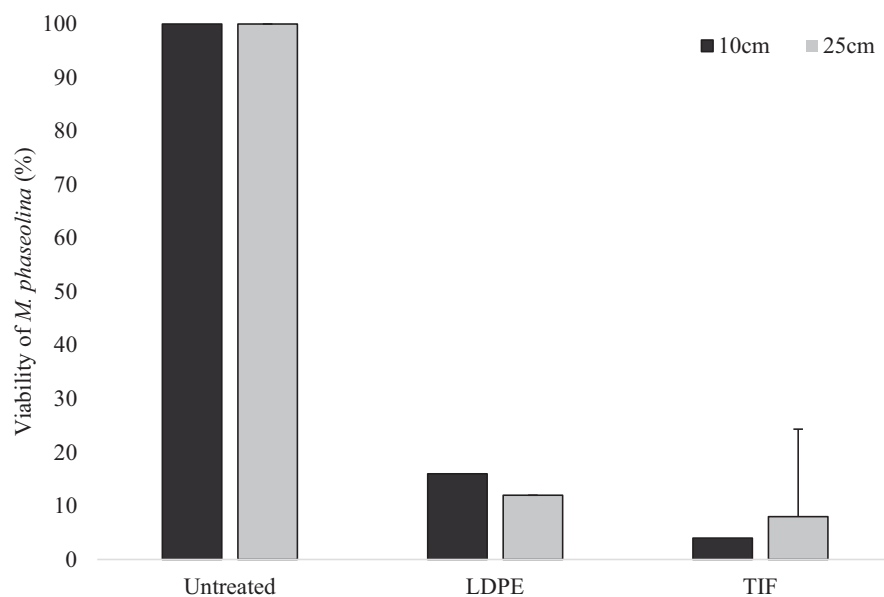


Fig. 1. The viability *M. phaseolina* in buried strawberry crowns after fumigation with 1,3-dichloropropene/chloropicrin in soil columns. The fumigated treatments were covered in either totally impermeable film (TIF) or low-density polyethylene (LDPE). The bar represents the least significant difference (interaction between treatments), where $P = 0.05$.

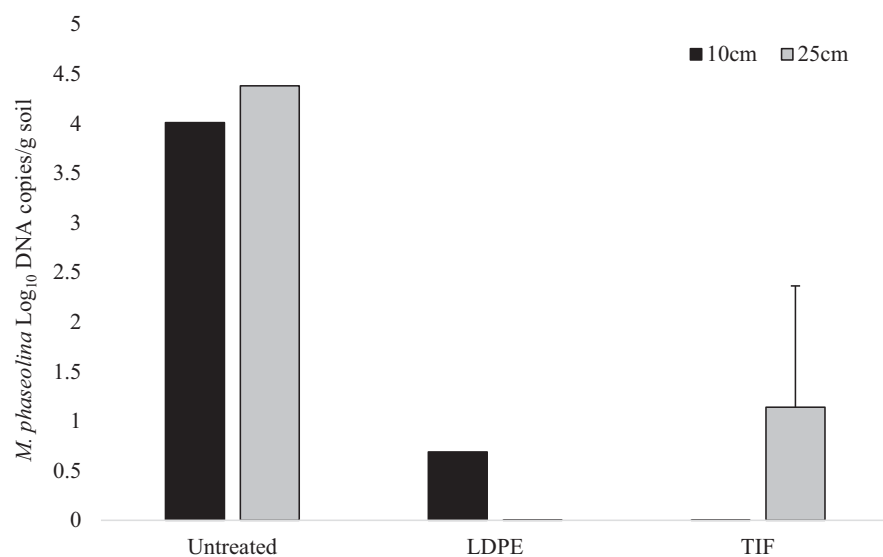


Fig. 2. Log₁₀ DNA copies of *M. phaseolina* in the soil after fumigation with 1,3-dichloropropene/chloropicrin in soil columns. The fumigated treatments were covered in either totally impermeable film (TIF) or low-density polyethylene (LDPE). The bar represents the least significant difference (interaction between treatments), where $P = 0.05$.

the site on a portion of the block. With this procedure, 1,3-D/Pic (20:80) was shank-injected at a depth of 15 cm at a rate of 400 kg/ha with a nine-tine rig (2.71 m width). A single 3-m-wide sheet of either TIF or LDPE film (see Column construction and design section of this article) was placed and buried on each edge as the fumigant was injected. The position of the film was centered over what would become the strawberry bed. This meant that both the bed and inter-row were fumigated in the broadcast treatment. The soil temperature at a depth of 5 cm at fumigation was 15 °C. The plastic was removed 1 week after fumigation and beds (width, 85 cm; 1.5-m centers) were raised across the site.

Then, strip fumigation was conducted, whereby 1,3-D/Pic was shank-injected at a depth of 15 cm into the raised beds at a rate of 400 kg/ha with a two-tine rig. The rig simultaneously

formed the bed, laid black film (TIF or LDPE) and a trickle irrigation tape, and applied fertilizer (Mattner et al. 2023). Soil temperature at a depth of 5 cm at fumigation was 14 °C. The inter-rows between the beds were not fumigated in the strip fumigation procedure. The controls were not strip-fumigated or broadcast-fumigated, but they were covered in black TIF or LDPE film. The rows that were previously broadcast-fumigated were also not strip fumigated but were covered in black TIF or LDPE film.

Two weeks after strip fumigation (Jan 2018), two rows of planting holes were cut into the film over each bed (30 cm spacings). Four weeks after strip fumigation (Feb 2018), certified strawberry transplants ('Albion'; Toolangi Certified Strawberry Runner Growers Cooperative, Toolangi, Victoria, Australia) were planted into the planting holes. Crops were initially watered by overhead

sprinklers during establishment (approximately 1 month) and then with drip irrigation following the grower's schedule. The crop remained in the ground until Jan 2020. During this cycle, the crop was trimmed and ratooned over winter (July–August). All agronomic practices followed the grower's standard practices (Stirling et al. 2019), including fertigation based on soil and plant nutrient tests, the use of a rotation of fungicides to manage foliar diseases such as gray mold (caused by *Botrytis cinerea*) and powdery mildew (caused by *Podosphaera aphanis*) (Mattner et al. 2023), and the use of red spider mite predators (*Phytoseiulus persimilis*) to manage two-spotted mites (*Tetranychus urticae*).

The experiment was conducted as a randomized complete block design with a total of six treatments. The treatments were composed of two factors: application method (three levels: untreated, strip-fumigated, and broadcast-fumigated) and barrier film (two levels: TIF and LDPE). Individual plots had a length of 5 m, and measurements of 20 plants were taken in the middle of the plots. Each treatment was replicated three times across the site.

VIABILITY OF *M. PHASEOLINA* IN BURIED CROWNS. On the day of fumigation, muslin bags containing five crowns from plants showing symptoms of charcoal rot (see Viability of *M. phaseolina* in buried crowns section of this article) were buried into each plot at a depth of 25 cm on the shoulder of the bed. This position was chosen because it is the most difficult point to kill *M. phaseolina* in the strawberry bed (Chamorro et al. 2016). The muslin bags were recovered from 1 week after fumigation, and the viability of *M. phaseolina* in the crowns was assessed in the laboratory (see Viability of *M. phaseolina* in buried crowns).

FUMIGANT CONCENTRATIONS. The concentration of 1,3-D and Pic in the soil air was measured as described previously at a depth of 15 cm in the center of the bed. Fumigant concentrations were recorded from each main plot at 2, 24, 72, and 144 h after treatment.

***M. PHASEOLINA* CONCENTRATIONS IN SOIL.** Shallow soil samples (0–10 cm) were taken with a sanitized hand trowel from each plot at 4, 49, 188, 279, 293, 348, 385, 452, and 651 d after planting. Samples were 500-g composites of 10 sub-samples. Soil samples were also

Table 2. Concentration × time values (mg/L/h) of chloropicrin and 1,3-dichloropropene in soil from 2 to 144 h after fumigation with 1,3-dichloropropene/chloropicrin in a field experiment at Silvan, Victoria, Australia. The fumigated treatments were covered in either totally impermeable film (TIF) or low-density polyethylene (LDPE). Values assigned different letters in each column are significantly different, where $P \leq 0.05$.

Application method	Barrier film	Chloropicrin	1,3-dichloropropene
Broadcast	LDPE	15,267 a	6271 ab
Broadcast	TIF	19,478 a	7950 b
Strip	LDPE	32,191 b	5038 a
Strip	TIF	39,528 b	8417 b

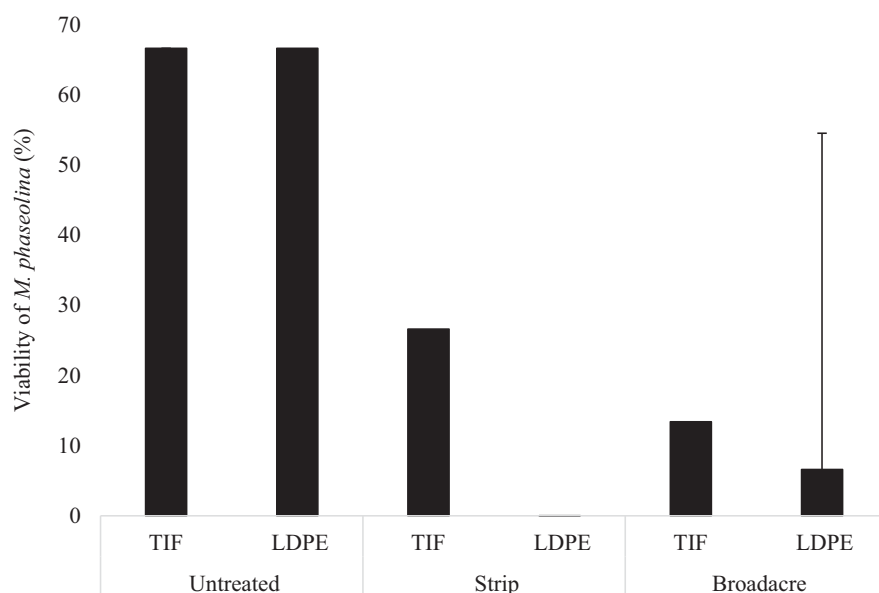


Fig. 3. The viability *M. phaseolina* in buried strawberry crowns after fumigation with 1,3-dichloropene/chloropicrin in a field trial at Silvan, Victoria, Australia. The application method was strip or broadacre fumigation, and treatments were covered in either totally impermeable film (TIF) or low-density polyethylene (LDPE) barrier film. The bar represents the least significant difference (interaction between treatments), where $P = 0.05$.

taken from depths of 30 to 40 cm with the use of an electric drill-bit (Power Planter™) on 26 Feb 2018. All soil samples were submitted to a commercial laboratory (SARDI), DNA were extracted, and a quantitative polymerase chain reaction was performed for *M. phaseolina* (see *M. phaseolina* concentrations in soil section of this article). The average \log_{10} copies of *M. phaseolina* DNA/g of soil were 3.2 at a depth of 0 to 10 cm and 3.6 at a depth of 30 to 40 cm before fumigation.

CHARCOAL ROT INCIDENCE AND SEVERITY. All plants were assessed for charcoal rot incidence and severity using the scale described by Mattner et al. (2018) (i.e., a 6-point categorical scale) at 49, 232, 279, 299, 343, 372, 390, 474, 615, 658, and 683 d postplanting. The charcoal rot severity and incidence scores were converted to decline/death indices based on the method described by Fang et al. (2011b) where:

$$\%DI = \left\{ [(a \times 0) + (b \times 1) + (c \times 2) + (d \times 3) + (e \times 4) + (f \times 5)] \times 100 \right\} / [(a + b + c + d + e + f) \times 5]$$

and where a, b, c, d, e, and f are the number of plants with a score of 0, 1, 2, 3, 4, and 5, respectively. A subsample of diseased plants (maximum of three plants) was collected from

each plot at the end of the experiment to confirm the presence of *M. phaseolina* in crowns (see Viability of *M. phaseolina* in buried crowns section of this article).

WEED ASSESSMENTS. The emergence of weeds was counted in each planting hole at 146, 490, and 700 d postplanting.

FRUIT YIELDS AND REVENUE. Strawberries were picked, counted, commercially graded, and weighed two to three times per week during the peak season (summer) and once per week when production was low (spring and autumn). A total of 75 picks were performed from Mar 2018 to Jan 2020. Revenue from fruit was calculated based on weekly wholesale prices at the Melbourne market (Freshlogic, Hawthorn, Victoria, Australia).

Partial budget analysis. Based on the revenue generated by the sale of fruit throughout the experiment and differential treatment costs, a partial budget analysis was performed to compare the economics of different fumigation treatments (see Fumigation, crop agronomy, and design section of this article). The untreated control with LDPE film was the baseline treatment. The additional costs associated with strip or broadacre fumigation and/or the use of TIF

were based on commercial quotes provided by R&R Fumigation (Bayswater, Australia) and VIC Plastics (Ravenhall, Australia).

Kinglake field experiment (2022)

FUMIGATION AND DESIGN. An experiment was conducted on a commercial strawberry farm at Kinglake, Victoria, Australia (lat. 37°49'48.3"S, long. 145°31'54.6"E). The soil at the site was a silty clay loam (30% silt, 33% clay, 37% sand) with a pH (water) of 5.7. Soil was prepared and strip-fumigated with 1,3-D/Pic (20:80) at a rate of 400 kg/ha in Mar 2022, as described previously. During fumigation, soil was sealed with black TIF or LDPE, as described previously (see Fumigation, crop agronomy, and design section of this article). A single (nonreplicated) 90-m-long bed of soil was assessed for each treatment (TIF and LDPE).

FUMIGANT CONCENTRATIONS. The concentrations of 1,3-D and Pic in soil air were measured as described previously at 4, 24, 96, and 168 h after treatment. Measurements were taken at three positions across the bed: from the center of the bed, on the tine line (12.5 cm from the center of the bed), and on the shoulder of the bed (30 cm from the tine line). The CT values were calculated and data were used to form contour maps through the cross-section of beds using the "contour plot" function in Genstat version 22 (VSN International).

Statistical analysis

All statistical analyses were performed with Genstat using an analysis of variance. Homogeneity of variance was determined by examining plots of fitted values and residuals, while histograms of residuals were examined for normality of distribution. When variance was heterogeneous across treatments, appropriate data transformations (e.g., \log_{10} transformations) were performed to restore homogeneity. Fisher's least significant difference test was used to identify significant differences between treatment means. The level of significance was $P \leq 0.05$.

Results

Soil column experiment

FUMIGANT CONCENTRATIONS. The untreated columns did not contain any detectable concentrations of Pic

Table 3. The average concentrations of *M. phaseolina* log₁₀ DNA copies/g of soil following treatment with the fumigant 1,3-dichloropene/chloropicrin in a field experiment at Silvan, Victoria, Australia. The application method was strip or broadacre fumigation and treatments were covered in either totally impermeable film (TIF) or low-density polyethylene (LDPE) barrier film. Soil samples were collected at depths of 0 to 10 cm from Feb 2018 to Dec 2019. Values assigned different letters in each column are significantly different, where $P \leq 0.05$. Note the seasonal conditions of Victoria, Australia during summer (December, January, February), fall (March, April, May), winter (June, July, August), and spring (September, October, November).

Application method	Barrier film	26 Feb 2018	12 Apr 2018	29 Aug 2018	28 Nov 2018	12 Dec 2018	5 Feb 2019	14 May 2019	20 May 2019	5 Dec 2019
Untreated	TIF	4.43	3.42	0.82	3.66	3.16	3.60 b	3.85 b	4.29 b	5.09
Untreated	LDPE	3.86	3.51	3.82	3.59	3.64	4.17 b	3.71 b	4.30 b	5.02
Strip	TIF	0	0.72	0	0	0	0 a	0 a	0 a	1.53
Strip	LDPE	1.15	3.17	1.99	0.96	2.41	2.90 b	3.23 b	4.27 b	4.26
Broadacre	TIF	0	0	0	0	0	0 a	0 a	0.96 a	2.10
Broadacre	LDPE	1.00	0.001	0	0	0	0 a	1.78 a	0 a	1.84
Application method × barrier film interaction (P)		0.051	0.389	0.051	0.394	0.083	<0.001	0.01	<0.001	0.208
Application method (P)		<0.001	<0.001	0.005	<0.001	<0.001	<0.001	<0.001	<0.001	0.015
		(untreated > strip > broadacre)	(untreated > strip, broadacre)	(untreated > strip, broadacre)	(untreated > strip > broadacre)	(untreated > strip > broadacre)	(untreated > strip, broadacre)	(untreated > strip, broadacre)	(untreated > strip > broadacre)	(untreated > strip, broadacre)
Barrier film (P)		0.009	0.348	0.003	0.388	0.041	<0.001	0.001	0.022	0.287
		(LDPE > TIF)	(LDPE > TIF)	(LDPE > TIF)	(LDPE > TIF)	(LDPE > TIF)	(LDPE > TIF)	(LDPE > TIF)	(LDPE > TIF)	(LDPE > TIF)

or 1,3-D. There was no significant interaction between sampling depth (10 cm and 25 cm) and barrier film regarding the CT values of Pic ($P = 0.341$) and 1,3-D ($P = 0.806$) (Table 1). The use of TIF significantly increased the CT values (mg/L/h) of Pic ($P < 0.001$) by 62% and 1,3-D ($P < 0.001$) by 51%. Sampling depth (10 cm and 25 cm) did not significantly affect the CT values of Pic ($P = 0.341$) and 1,3-D ($P = 0.806$).

VIABILITY OF *M. PHASEOLINA* IN BURIED CROWNS. There was no significant interaction between sampling depth (10 cm and 25 cm) and treatment (i.e., untreated, fumigation under LDPE or TIF) ($P = 0.266$) for the viability of *M. phaseolina* in buried crowns (Fig. 1). Fumigation with 1,3-D/Pic significantly ($P < 0.001$) reduced the viability of *M. phaseolina* in crowns by an average of 90% compared with the untreated control. However, there was no significant difference between the columns that were fumigated and covered by either LDPE or TIF. Sampling depth (10 cm and 25 cm) did not affect the viability of *M. phaseolina* within buried crowns ($P = 0.686$).

***M. PHASEOLINA* CONCENTRATIONS IN SOIL.** There was no significant interaction between sampling depth (10 cm and 25 cm) and treatment ($P = 0.111$) for the concentration of *M. phaseolina* in soil (Fig. 2). Fumigation with 1,3-D/Pic significantly ($P < 0.001$) reduced the concentrations in soil compared with the untreated controls by an average of 99.5%. However, there was no significant difference between the columns that were fumigated and covered by either LDPE or TIF. Sampling depth (10 cm and 25 cm) did not affect the concentration of *M. phaseolina* in soil ($P = 0.431$).

Silvan field experiment (2018–20)

FUMIGANT CONCENTRATIONS. The untreated plots did not contain any detectable concentrations of 1,3-D or Pic. There was no significant application method × barrier film interaction regarding the CT values of Pic ($P = 0.734$) or 1,3-D ($P = 0.297$) (Table 2).

The CT values of Pic were significantly higher ($P = 0.006$) for the two strip treatments compared with the two broadacre treatments by an average of 106%. There was no significant difference ($P = 0.625$) between the two strip treatments compared with

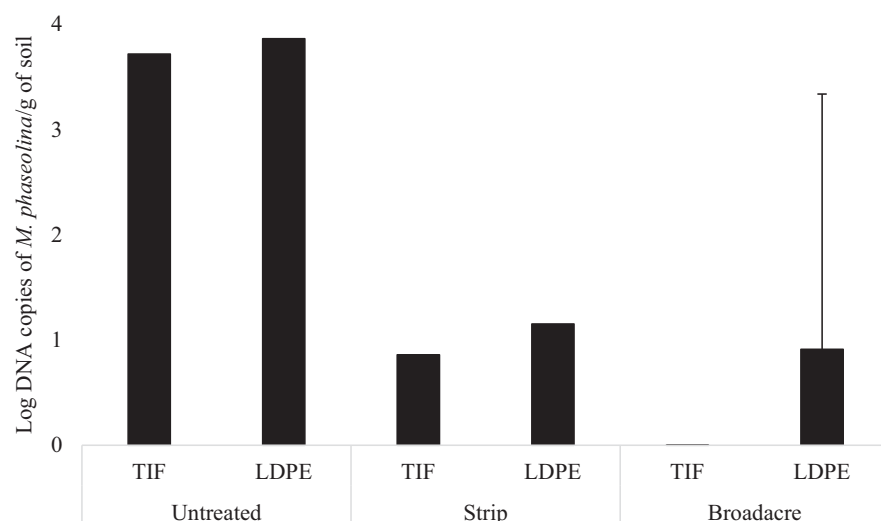


Fig. 4. The average concentrations of *M. phaseolina* log₁₀ DNA copies/g of soil following fumigation with 1,3-dichloropene/chloropicrin in a field experiment at Silvan, Victoria, Australia. The application method was strip or broadacre fumigation, and treatments were covered in either totally impermeable film (TIF) or low-density polyethylene (LDPE) barrier film. Soil samples were collected at depths of 30 to 40 cm on 26 Feb 2018. The bar represents the least significant difference (interaction between treatments), where $P = 0.05$.

the two broadacre treatments regarding the CT values of 1,3-D.

Sealing soil with TIF significantly increased ($P = 0.015$) the CT values of 1,3-D by 45% compared with the standard practice of LDPE film. Conversely, the plots fumigated under TIF did not differ significantly in CT values of Pic ($P = 0.237$) compared with those fumigated under LDPE.

VIABILITY OF *M. PHASEOLINA* IN BURIED CROWNS. There was no significant application method \times barrier

film interaction ($P = 0.67$) for the viability of *M. phaseolina* in buried crowns (Fig. 3). Fumigation significantly ($P = 0.006$) reduced the viability of *M. phaseolina* inside crowns compared with the untreated control by an average of 82%. However, there was no significant difference between the plots that were either strip-fumigated or broadacre-fumigated. There was no significant difference ($P = 0.392$) between plots covered by either LDPE or TIF.

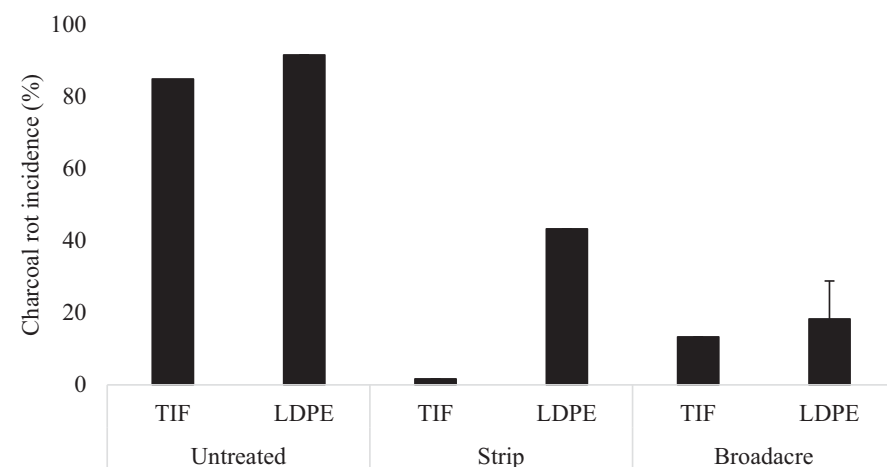


Fig. 5. The incidence of charcoal rot of strawberry at final harvest (Dec 2019) in a field experiment at Silvan, Victoria, Australia. The application method was strip or broadacre fumigation, and treatments were covered in either totally impermeable film (TIF) or low-density polyethylene (LDPE) barrier film. The bar represents the least significant difference (interaction between treatments), where $P = 0.05$.

M. PHASEOLINA CONCENTRATIONS

IN SOIL. Shallow depths (0–10 cm). At most sampling dates, there was no significant application method \times barrier film interaction ($P > 0.05$) for the concentration of *M. phaseolina* in soil at 0 to 10 cm (Table 3). From 5 Feb 2019 to 20 May 2019, however, there was a significant interaction ($P < 0.05$) whereby TIF reduced *M. phaseolina* concentrations in soil more than LDPE in strip fumigated plots only. Overall, fumigated plots consistently contained significantly lower concentrations of *M. phaseolina* DNA compared with those of the untreated plots. On four of the nine sampling dates, the broadacre treatments contained significantly lower concentrations of *M. phaseolina* DNA compared with the strip treatments. Furthermore, the plots covered with TIF contained significantly lower concentrations of *M. phaseolina* DNA compared with those covered in LDPE on six of the nine sampling dates.

Deep depths (30–40 cm). There was no significant application method \times barrier film interaction ($P = 0.87$) for *M. phaseolina* concentration in soil at 30 to 40 cm (Fig. 4). Fumigation significantly ($P = 0.003$) reduced the viability of *M. phaseolina* in the soil compared with the untreated controls by an average of 80%. However, there was no significant difference between the plots that were either strip-fumigated or broadacre-fumigated. There was no significant difference ($P = 0.49$) between plots covered by either LDPE or TIF.

CHARCOAL ROT INCIDENCE AND SEVERITY. At the end of the experiment, all fumigant treatments significantly ($P < 0.05$) reduced the incidence of charcoal rot compared with the untreated controls. Furthermore, there was a significant interaction ($P = 0.037$) between application method and barrier film at final harvest regarding the incidence of charcoal rot. Strip fumigation under TIF and broadacre fumigation under TIF or LDPE controlled the incidence of charcoal rot significantly more than the industry standard practice of strip fumigation under LDPE by 96%, 69%, and 58%, respectively (Fig. 5).

The charcoal rot index in plants increased exponentially in nonfumigated plots from Dec 2018 to Mar 2019 (summer) (Fig. 6). The climatic conditions, including the temperature

Table 4. Cumulative strawberry fruit production and economics per plant (2018–20) in soils treated with 1,3-dichloropene/chloropicrin in a field experiment at Silvan, Victoria, Australia. The application method was strip or broadcast fumigation and treatments were covered in either totally impermeable film (TIF) or low-density polyethylene (LDPE) barrier film. Values assigned different letters in each column are significantly different, where $P \leq 0.05$.

Application method	Film	Total fruit yield (g)	Marketable fruit yield (g)	Total number of fruit	Marketable number of fruit	Marketable fruit size (g/berry)	Marketable fruit revenue (AU\$)	Partial profit from fruit (AU\$) ⁱ
Untreated	TIF	870.92 a	533.79 a	52.23 a	29.22 a	18.37 a	2.94 a	2.92 a
Untreated	LDPE	939.18 a	551.80 a	56.70 a	30.35 a	18.41 a	3.09 a	3.09 a
Strip	TIF	1686.93 c	986.97 c	103.27 c	55.95 c	17.60 a	5.55 c	5.48 c
Strip	LDPE	1213.20 b	709.32 b	73.63 b	38.17 b	18.59 a	4.04 b	3.99 b
Broadcast	TIF	1680.82 c	1005.32 c	98.37 c	53.93 c	18.73 a	5.73 c	5.57 c
Broadcast	LDPE	1729.99 c	972.69 c	103.03 c	52.18 c	18.65 a	5.65 c	5.53 c
LSD ($P = 0.05$)		214.33	126.20	13.50	6.85	ns	0.34	0.34

ⁱFruit revenue less the additional costs (film and labor) above the standard practice of strip fumigation under LDPE.

LSD = least significant difference.

95% and, subsequently, increased fruit yields by up to 39% compared with the standard practice of strip fumigation under LDPE. Furthermore, the data supported previous research showing that fumigation under TIF increased the CT values of fumigants in soil (McAvoy and Freeman 2013a; Stevens et al. 2016), reduced the concentration of pathogenic inoculum in soil, improved control of charcoal rot (Washington 2019), and, subsequently, increased fruit yields compared with the standard practice (i.e., strip fumigation with LDPE) (Fennimore and Ajwa 2011; Washington 2019). These results suggest that sites with a history of charcoal rot of strawberry and that use of strip fumigation with 1,3-D/Pic under LDPE could improve disease outcomes from the adoption of TIF and/or broadcast application.

Fumigation with 1,3-D/Pic using the strip method under TIF and broadcast treatments (averaged across TIF and LDPE because there was no significant difference between treatments) increased marketable fruit revenue of strawberry in a field infested with *M. phaseolina* by \$1.51/plant and \$1.65/plant, respectively, compared with the current standard practice of strip application under LDPE. Furthermore, these treatments were cost-effective because they increased partial profits by \$1.49/plant and \$1.56/plant, respectively, above the industry's standard practice. Therefore, even though TIF and broadcast fumigation with 1,3-D/Pic are more expensive than strip application under LDPE, they proved more profitable in a field infested with *M. phaseolina*. Other benefits from the

improved fumigation practices not accounted for in the partial budget analysis included reduced costs of hand weeding resulting from improved weed control and the potential environmental and operator exposure/human health benefits of TIF, such as the reduction in fumigant and N₂O emissions to the atmosphere (Li et al. 2022).

The results from the current study showed that the CT values of 1,3-D and Pic in soil were up to 67% and 60% higher, respectively, under TIF compared with LDPE in the soil column and field experiments. These results align with existing literature that showed that fumigants, including Pic and 1,3-D, applied under TIF have greater concentrations in soil than those applied under LDPE (Fennimore and Ajwa 2011; Gao et al. 2012). In the current study, the Kinglake experiment demonstrated that TIF improved the CT values and distribution of fumigant concentrations in soil across the bed compared with LDPE, particularly on the shoulders and at greater depths. We hypothesized that the improved distribution of fumigants in soil under TIF is key to the improved efficacy of the treatment against *M. phaseolina* compared with fumigation under LDPE.

Wilhelm (1965) and Wilhelm et al. (1974) stated that broadcast applications of fumigants are superior to strip applications because of the increased distribution of the fumigants in soil. Unfortunately, we did not test for the distribution of fumigants in soil (i.e., fumigant concentrations were sampled from the same position within the plots) in the Silvan field experiment. Nonetheless, we hypothesize that the

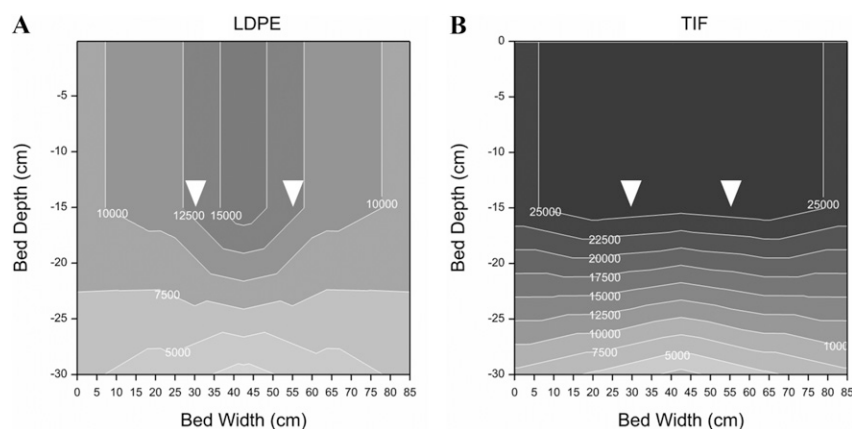


Fig. 8. Cross-section contour maps of the concentration \times time values (mg/L/h) of chloropicrin in strawberry beds sealed with (A) low-density polyethylene (LDPE) and (B) totally impermeable film (TIF) from 4 to 168 h after strip fumigation with 1,3-dichloropene/chloropicrin in a field experiment at Kinglake, Victoria, Australia. Bottom apices of white triangles indicate the points of fumigant injection in the bed.

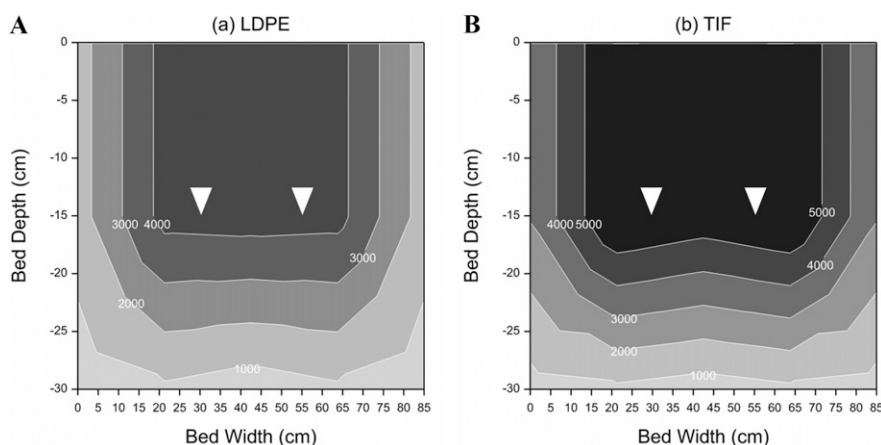


Fig. 9. Cross-section contour maps of the concentration \times time values (mg/L/h) of 1,3-dichloropene/chloropicrin in strawberry beds sealed with (A) low-density polyethylene (LDPE) or (B) totally impermeable film (TIF) from 4 to 168 h after strip fumigation with 1,3-D/ Pic in a field experiment at Kinglake, Victoria, Australia. Bottom apices of white triangles indicate the points of fumigant injection in the bed.

even application of fumigants across the soil profile may be an advantage of broadcast compared with the strip method. This is because all soil in the subsequent beds are treated with similar doses of fumigants. This is dissimilar to strip fumigation, whereby the semi-cylindrical shape of the raised soil beds during treatment may result in an uneven distribution of fumigants (i.e., soil in the edges of the beds were exposed to lower concentrations of fumigants, Figs. 8 and 9). In the Silvan field experiment, this factor may have been associated with the delayed recolonization of *M. phaseolina* in soil in the broadcast-treated plots compared with the standard practice of strip fumigation under LDPE.

The data from the Silvan experiment showed that the concentration of *M. phaseolina* DNA in the soil fluctuated over seasons in the cropping cycle. In general, the concentrations peaked in summer (particularly evident in untreated plots). Studies by Fang et al. (2011a) and Zveibil et al. (2012) also showed that the amount of *M. phaseolina* present in soil increased during the summer months when temperatures exceeded 30 °C. These results corroborate the thermophilic nature of *M. phaseolina* and support previous authors who have postulated that charcoal rot of strawberry will increase in importance under global warming scenarios (Cohen et al. 2022; Pennerman et al. 2024).

Previous studies have demonstrated that strawberry crowns infected with

M. phaseolina in the soil from previous crops are an important inoculum source for the infection of healthy plants (Baggio et al. 2019; Oag 2022). Additionally, research by Hutton et al. (2013) and Chamorro et al. (2016) confirmed that 1,3-D/Pic and other treatments are ineffective for eradicating *M. phaseolina* in infected crowns within soil. In the current study, we did not detect differences in the ability of different fumigation methods to control *M. phaseolina* in buried crowns. Furthermore, no fumigation method eradicated *M. phaseolina* in buried crowns. Nonetheless, fumigation with 1,3-D/Pic significantly reduced the viability of *M. phaseolina* in infected strawberry compared with that of the untreated controls (Fig. 1). Based on these results, it is important for the industry to consider alternative management strategies to enhance control of *M. phaseolina* in infested strawberry crowns in the soil. Recent research suggested that practices such as crop termination with metam sodium might directly reduce the survival of pathogenic inoculum in old strawberry crowns and improve the efficacy of subsequent fumigation (Khatri et al. 2019; Oag 2022). Furthermore, studies suggest that additional farm biosecurity practices and production of strawberry cultivars with increased resistance to *M. phaseolina* would also complement the industry's current management practices (Gomez et al. 2020; McFarlane et al. 2019a).

Conclusions

Treatment with 1,3-D/Pic under TIF increased the CT values of these fumigants in soil compared with application under LDPE by up to 38%. Moreover, strip fumigation under TIF reduced DNA concentrations of *M. phaseolina* in soil (0–10 cm) by 65% and charcoal rot index by 93% and increased fruit yields by 37% and partial profits from fruit by AU\$1.49/plant compared with LDPE. Additionally, broadcast fumigation reduced DNA concentrations of *M. phaseolina* by an average of 55% and charcoal rot index by 65% and increased fruit yields by 40% and partial profits from fruit by AU\$1.56/plant compared with strip fumigation under LDPE. The results of this study suggest that adoption of strip fumigation under TIF or broadcast applications of 1,3-D/Pic could improve management of charcoal rot of strawberry in Australia in a cost-effective manner compared with the standard practice of strip fumigation under LDPE.

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