SURVIVAL AND VIABILITY OF SCLEROTIA OF *SCLEROTINIA SCLEROTIORUM* IN TWO KIWIFRUIT ORCHARDS IN THE BAY OF PLENTY

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SUMMARY

Low numbers of sclerotia were present in soil in two kiwifruit orchards during pre-blossom (22 September 91) and post-blossom (9 January 92) with about 0 to 0.13 sclerotia/kg soil. Sclerotia number significantly increased during preharvest (15 April 92) to 0.4 sclerotia/kg soil in Orchard A and 0.8 sclerotia/kg soil for Orchard B. *S. sclerotiorum* was isolated from 41% of sclerotia from Orchard A and from 31% in Orchard B during preharvest. Viability of sclerotia buried in orchard soil decreased by 30% after 111 days and 48% after 208 days while decomposition of the sclerotia was 1% and 13% respectively. Species of *Trichoderma* and *Gliocladium* were isolated from non-viable sclerotia. Their potential in reducing sclerotia viability in kiwifruit orchards warrants further investigation.

Keywords: sclerotia survival, kiwifruit, *Sclerotinia sclerotiorum*, *Trichoderma*, *Gliocladium*.

INTRODUCTION

In species of *Sclerotinia* (Fuckel) about 90% of the life cycle is spent in the soil as sclerotia (Adams and Ayers 1979). These are the primary means of survival and source of inoculum for *S. sclerotiorum* (Lib) de Bary. Under dry conditions, sclerotia of *S. sclerotiorum* remain viable for at least 10 years (Brown and Butler 1936), in contrast to the estimated 4 to 5 years survival for sclerotia in field soil (Adams and Ayers 1979). Antagonism and parasitism by species of fungi and bacteria appear to be the most significant components of soils affecting survival of sclerotia but proof of parasitism for most of these micro-organisms is lacking and information about their importance or behaviour in natural soil is scant (Ayers and Adams 1979). However, mycoparasitism of sclerotia of *S. sclerotiorum* by *Sporidesmium sclerotivorum* (Ayers and Adams 1981), *Gliocladium* spp (Tu 1980), *Trichoderma* spp. (Knudsen *et al* 1991) and *Coniothyrium minitans* (Huang 1977) have consistently been reported.

Quantitative information on the overwintering and survival of sclerotia under kiwifruit is lacking. Almost all research on *S. sclerotiorum* has been on annual crops (Purdy 1979) where cultural practices, cropping conditions and disease management (eg. crop rotation, deep ploughing) are different from those used in a perennial crop such as kiwifruit.

This paper reports on the survival and viability of sclerotia of *S. sclerotiorum* in the soil of two adjacent kiwifruit orchards in the Bay of Plenty.

MATERIALS AND METHODS

Sclerotia populations

The number of sclerotia in the soil under female kiwifruit vines at two Bay of Plenty orchards was determined at pre-blossom (22 September 1991), post-blossom (9 January 1992) and preharvest (15 April 1992). On the first sampling, 10 soil cores (5 cm diameter and 5 cm deep) per plot were taken and bulked. On subsequent sampling occasions soil quadrat samples (30 x 30 x 2 cm depth) were taken.

Sclerotia were extracted from soil samples by a method adapted from Hoes and Huang (1975), Utkhede and Rahe (1979) and Thomas *et al* (1983). The soil samples

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were weighed, washed under running water through two stacked sieves of 4.0 mm and 0.8 mm mesh openings respectively. Roots and other large organic particles were discarded. The fractions containing the sclerotia on the 0.8 mm sieve were firstly placed in water to remove plant debris by flotation and then in magnesium sulphate (MgSO₄) solution (density 1.18 to 1.20 g/cm). Sclerotia floating in the MgSO₄ solution were removed manually under binocular microscope.

The sclerotia recovered from soil samples were surface sterilised in 1.0% sodium hypochlorite (NaOCl) solution for 2 min and then rinsed twice in sterile distilled water. The sclerotia were placed on potato dextrose agar (PDA) and incubated at 20°C under 12:12 L:D. Growth of *S. sclerotiorum* and other fungi from these sclerotia was observed and recorded after 4 to 10 days.

Sclerotia survival

Sclerotinia sclerotiorum was cultured on autoclaved kiwifruit slices for 2 to 3 weeks at ambient temperatures (20–25°C). Sclerotia that formed were removed, air-dried for 4 to 6 weeks in Petri dishes and kept in a refrigerator at 5°C for 2 to 3 months (Hoyte *et al* 1992). Twenty sclerotia produced by these cultures were placed inside a 10 x 10 cm nylon bag (1 mm mesh). On 27 September 1991 eight bags were buried to 2 cm depth in each of five plots under kiwifruit vines in a Tauranga orchard. One bag from each plot was retrieved 111 and 208 days after burial, the sclerotia surface sterilised in 1% NaOCl for 2 min, rinsed twice in sterile distilled water, placed on PDA and incubated at 20°C under 12:12 L:D. Growth of *S. sclerotiorum* and other fungi was recorded after 4 to 10 days.

RESULTS

Soil collected from both orchards at pre-blossom (20 September 91) and post-blossom (9 January 92) had 0 to 0.13 sclerotia/kg soil (Table 1). The number of sclerotia was higher at preharvest sampling (0.4 - 0.8 sclerotia/kg soil).

TABLE 1: Sclerotial inoculum density in soil of two kiwifruit orchards in the Bay of Plenty.

Growth stage	Pre-blossom Estimated number of sclerotia	Post-blossom Estimated number of sclerotia	Pre-harvest Estimated number of sclerotia per kg soil
Orchard A	0.13(0.07) ^a	0.05(0.03)	0.38(0.07)
Orchard B	0(0)	0.06(0.06)	0.80(0.26)

^a Numbers in brackets are standard error of mean.

The low number of sclerotia recovered from soil in September 1991 and January 1992 yielded mainly *S. sclerotiorum*. Of the sclerotia retrieved from soil in April 1992, only 31 to 41% yielded *S. sclerotiorum*. The non-viable sclerotia yielded species of *Trichoderma*, *Gliocladium*, *Botrytis* and *Fusarium* while few viable sclerotia harboured these fungi (Table 2).

TABLE 2: Incidence of *Sclerotinia sclerotiorum* and other fungi isolated from sclerotia recovered from soil samples during pre-harvest.

	Total sclerotia recovered	Incidence of fungus isolated from sclerotia (%)				
		<i>Sclerotinia sclerotiorum</i>	<i>Trichoderma spp</i>	<i>Gliocladium spp</i>	<i>Botrytis spp</i>	<i>Fusarium spp</i>
Orchard A	22	41(12) ^a	18(11)	27(9)	4(4)	23(9)
Orchard B	35	31(13)	43(16)	31(13)	0	6(6)

^a Numbers in brackets are standard error of mean.

TABLE 3: Survival and viability of laboratory cultured sclerotia of *Sclerotinia sclerotiorum* buried in Orchard A in the Bay of Plenty.

Days buried in orchard soil	Decomposed sclerotia (%)	Total sclerotia recovered from soil (%)	Incidence of fungi isolated from sclerotia (%)			
			<i>Sclerotinia sclerotiorum</i>	<i>Trichoderma</i> spp	<i>Gliocladium</i> spp	<i>Fusarium</i> spp
0	0	-	92(5)	0	0	0
111	1(1) ^a	99(1)	62(6)	23(6)	20(6)	14(4)
208	13(6)	88(6)	44(8)	36(8)	11(4)	17(5)

^a Numbers in brackets are standard error of mean.

Burial in orchard soil reduced the survival and viability of the sclerotia (Table 3). Decomposition accounted for 1% loss of sclerotia after 111 days and 13% after 208 days. A high proportion of the remaining non-viable sclerotia yielded species of *Trichoderma*, *Gliocladium* and *Fusarium* when plated on PDA (Table 3).

DISCUSSION

The low numbers of sclerotia in soil under kiwifruit vines in both orchards during pre- and post-blossom period may have been influenced by two factors. Firstly, low levels *S. sclerotiorum* infection on the buds, flowers and fruitlets resulted in little shedding of diseased plant parts and hence minimal sclerotia production on the orchard floor (Goh and Lyons 1992). Secondly, few sclerotia may have survived over the winter. The numbers of sclerotia in the orchards increased during the pre-harvest period but their viability was low. Infection by species of *Trichoderma* and *Gliocladium*, fungi known to parasitise sclerotia (Huang 1977; Knudsen *et al* 1991), was common in the non-viable sclerotia. In the sclerotia burial experiment, colonisation of the sclerotia by species of *Trichoderma*, *Gliocladium* and *Fusarium* was associated with loss of sclerotia viability. *Gliocladium virens*, *Trichoderma harzianum* and *T. viride* have been reported to parasitise both mycelia and sclerotia of *S. sclerotiorum* reducing germination and apothecia production (Jones and Watson 1969; Tu 1980; Knudsen *et al* 1991). The presence of these fungi on non-viable sclerotia from the kiwifruit orchards suggests mycoparasitism. Previous reports of mycoparasitism on sclerotia of *S. sclerotiorum* by species of *Trichoderma* and *Gliocladium* have not previously been reported from kiwifruit orchards. *Fusarium* spp isolated from sclerotia are typically inhabitants of soil (Domsch *et al* 1980) and have not been reported to parasitise sclerotia.

As sclerotia are overwintering structures of *S. sclerotiorum* and the primary source of inoculum of the disease, any significant reduction in overwintering sclerotia number may greatly influence the development of disease epidemics in the following spring. The effectiveness of these potential biological control fungi on the ecology and epidemiology of *S. sclerotiorum* warrants further studies.

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