

Fungicides and Soil Health: Challenges for the Industry

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Abstract

The challenge exists for the Organics industry to manage disease whilst protecting soil health. The use of copper (Cu) fungicides has been effective in the control of many pathogens, however, it has come at a cost. Copper residues in an avocado orchard caused significant reductions in biomass carbon (C_{mic}), even though the soils had similar or elevated levels of total organic carbon (C_{org}). The $C_{mic}:C_{org}$ ratio was significantly lower in all of the Cu contaminated soils. Microorganisms in the Cu contaminated soils were stressed as they had an increased metabolic quotient.

The composting worm *Eisenia fetida* was shown to avoid soils with even relatively minor elevation of copper (44mg/kg). Very significant earthworm avoidance was found when copper residues increased above 100mg/kg. In a related study, severely depleted populations of both native and exotic earthworms were found in copper contaminated orchards. Copper residues from fungicide application at an avocado orchard have impacted upon key soil health indicators. To protect soil health, alternatives to copper for disease control will need to be developed, along with remediation technologies for reducing the impact of Cu contamination in soils.

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Introduction

The use of chemical biocides to control microbial, fungal and insect plant pests has long been a feature of conventional agricultural practice and their use has made it possible to increase crop yields and food production (Lee 1985). However, many of these biocides have toxic effects that are not confined to their target species, and their application may have negative impacts on organisms that benefit the wider agroecosystem.

Copper-containing fungicidal sprays have been used in pome and stone fruit orchards, vineyards and vegetable crops for well over 100 years (Merry *et al.* 1983). In Australasia over 7500 t/year of Cu fungicides are used, representing 13% of the global total (Lepp and Dickinson 1994). Commonly used fungicides in avocado orchards on the north coast of NSW have included copper oxychloride, cuprous oxide, copper hydroxide and copper ammonium acetate. Fungal diseases controlled by copper include downy mildew (*Plasmopara viticola*), Anthracnose and Phytophthora root rot (*Phytophthora cinnamomi*). Recommended application rates for conventional orchards are 3-6kg/ha per application to foliage up to 15 times per year (QDPI). This can account for between 30-450mg/kg copper input into the top 1cm of the soil profile per year. However, use of copper-containing products is restricted in organic farming practices in Australia, in that Bordeaux mixes (copper sulphate and hydrated lime), copper hydroxide, copper sulphates and copper ammonium carbonate are permitted by certifying authorities but copper oxychloride is prohibited (BFA, 2000). Furthermore, from 2002, the International Federation of Organic Agriculture Movements (IFOAM) will be regulating total copper input on organic farms to a maximum of 8kg/Ha/year.

This restriction is in place to ensure that excessive copper does not build-up in orchard soils. Indeed, horticultural and viticultural operations with a long history of Cu fungicide application often have significant accumulations of Cu in surface horizons (Merry and Tiller 1986; Alva *et al.* 2000). The relatively long residence time of Cu in top soils, largely related to the high affinity of Cu for soil organic matter and hydrous oxides, means that long term accumulation of Cu is likely.

The accumulation of Cu in top soils also corresponds to the zone in the soil profile of greatest biological activity. Detrimental effects of elevated Cu concentrations upon mycorrhizal associations (Georgieva *et al.* 2002), microbial populations and function (Dumestre *et al.* 1999) and a range of mesofauna (Paoletti *et al.* 1998) have been documented.

It has been suggested (Potter *et al.* 1990) that to circumvent toxic effects, earthworms may avoid surface litter and soil layers contaminated by certain pesticides. Earthworms are highly mobile in soil and hence their ability to avoid areas of contamination would have significant ecological implications (Yearley *et al.* 1996).

This manuscript presents some work recently conducted by Merrington *et al.* (2002) on a Cu contaminated avocado orchard and describes some of the impacts of Cu contaminated soil on the composting worm *Eisenia fetida*.

Copper influences on microbial indicators

This study examined the influences of copper contamination on soil health indicators in an avocado orchard located approximately 5 km north-east of the town of Alstonville, in northern-eastern NSW. The site is approximately 5 ha in size and is gently sloping (< 5°) with ferrosol soil. The orchard comprises 1100 trees of a range of commercial avocado varieties, ranging in age from 2 to 25 years. There is no history of Cu toxicity in the avocado trees in the orchard. Foliar application of Cu-based fungicides has been used extensively for the control of Anthracnose.

Soil sampling

Soil samples were collected from the orchards, as well as from a nearby control site comprising natural vegetation. The soil samples were then sieved (< 2 mm) soon after collection, sub sampled and half the soil sample was then stored at 4°C for the soil microbial tests. The second half was oven-dried at 60°C before chemical analysis including; 'total' Cu, 0.01M CaCl₂ extractable Cu, pH (1:5), organic C and total N. Free Cu²⁺ ion activity in soil solution extracts, microbial biomass and soil basal respiration were determined using methods described by Merrington *et al.* 2002.

Results and Discussion

Total soil Cu concentrations in both horizons (0-2 and 2-10 cm) from Block A and B in the avocado orchard were between 14 and 27 times greater than the reference soil. Soils from Block A had significantly higher concentrations of Cu in the 0-2 cm horizon, whilst there was no difference in Cu concentrations in the two horizons from Block B (Table 1). Both horizons in these orchard samples had significantly higher CaCl₂ extractable Cu than the respective horizons in the reference soil. In contrast to the total Cu concentrations, the 0-2 cm horizon in Blocks A and B had significantly higher concentrations of CaCl₂ extractable Cu than the lower horizon. In Block A CaCl₂ extractable Cu in the 0-2 cm horizon was 2.8 times the concentration measured in the lower horizon, with 1.8 times the concentration in Block B.

Sample site	'Total' Cu (mg/kg)	CaCl ₂ extractable Cu (mg/kg)	Organic C (%)	Total (%)	N	pH (1:5 CaCl ₂)
Block A, 0-2 cm	280 ± 33*	2.15 ± 0.39*	8.82 ± 0.48*	0.96 ± 0.06		6.01 ± 0.23*
Block A, 2-10 cm	176 ± 48*	0.76 ± 0.12*	4.88 ± 0.59	0.52 ± 0.05		4.50 ± 0.47
Block B, 0-2 cm	345 ± 78*	1.29 ± 0.21*	8.43 ± 1.00*	0.99 ± 0.11		5.80 ± 0.20*
Block B, 2-10 cm	301 ± 112*	0.71 ± 0.15*	5.37 ± 0.14	0.61 ± 0.02		5.34 ± 0.23
Reference, 0-2	13 ± 1.8	0.71 ± 0.19	6.74 ± 0.14	0.72 ± 0.02		4.43 ± 0.05
Reference, 2-10	14 ± 0.1	0.24 ± 0.05	4.93 ± 0.30	0.56 ± 0.03		4.51 ± 0.13

Table 1. Selected characteristics of the orchard and reference soils (derived from Merrington *et al.* 2002). Values are means ± standard error; n = 6. Significantly (**P* ≤ 0.05) different from respective soil layer in the reference soil.

Microbial basal respiration (results not shown), (CO₂ respired from incubated soil samples) in soils from the 0-2 cm horizon in both orchard blocks was almost double that observed in the equivalent horizon in the reference soil. Whilst there was no difference in basal respiration between the 0-2 and 2-10 cm horizon in the reference soil, significantly higher basal respiration was observed in the 0-2 cm horizons in the orchard soils (Block A, 5.5 times; Block B, 9 times) when compared to the 2-10 cm horizons. The 0-2 cm horizons of orchard soils also had higher *q*CO₂ values than reference soils (1.7 times) (Figure 1), and as with basal respiration observations, the

orchard 0-2 cm horizon samples had increased $q\text{CO}_2$ values when compared to the associated 2-10 cm horizons. Basal respiration was significantly higher in Cu contaminated orchard soils. Higher rates of respiration (with associated reductions in C_{mic} and lower $C_{\text{mic}}:C_{\text{org}}$ ratios [Figure 2]), are considered to be indicative of soil microbial population stress.

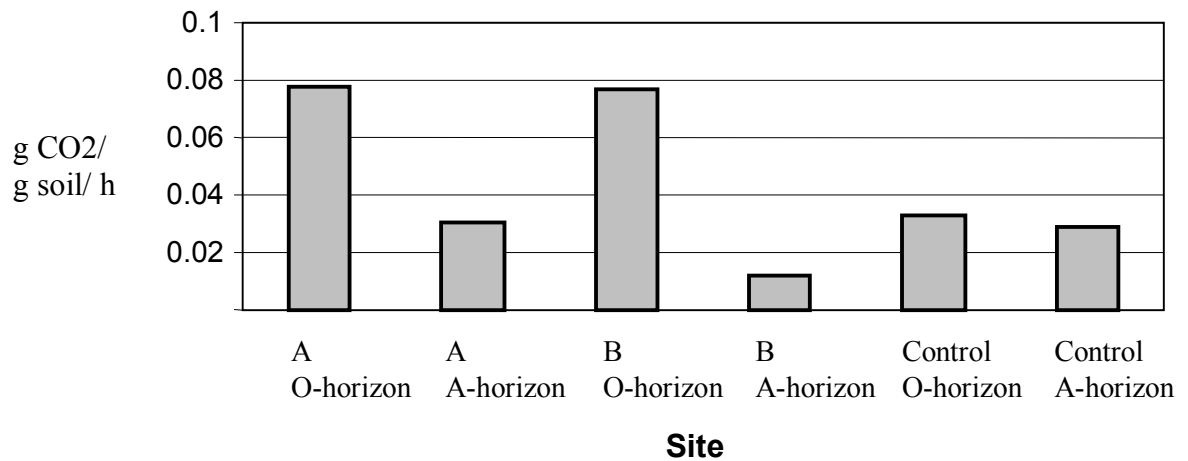


Figure 1. Metabolic quotient ($q\text{CO}_2$) of the orchard and control sites (grams of carbon dioxide produced per gram on biomass C per hour).

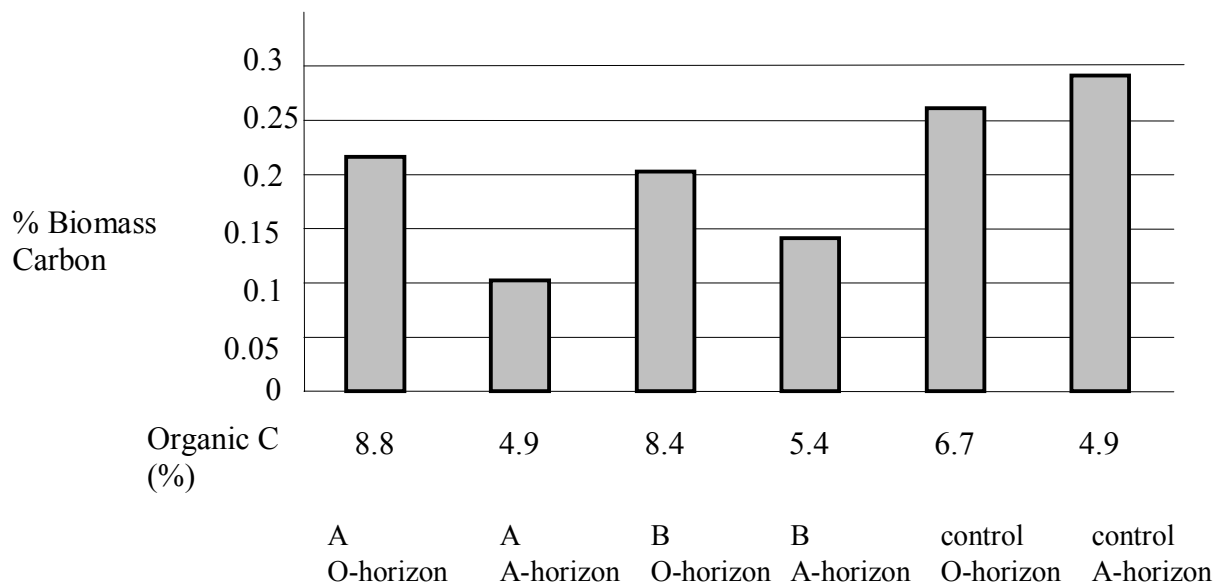


Figure 2. Biomass C (%) and total organic C (%) in orchard and control sites.

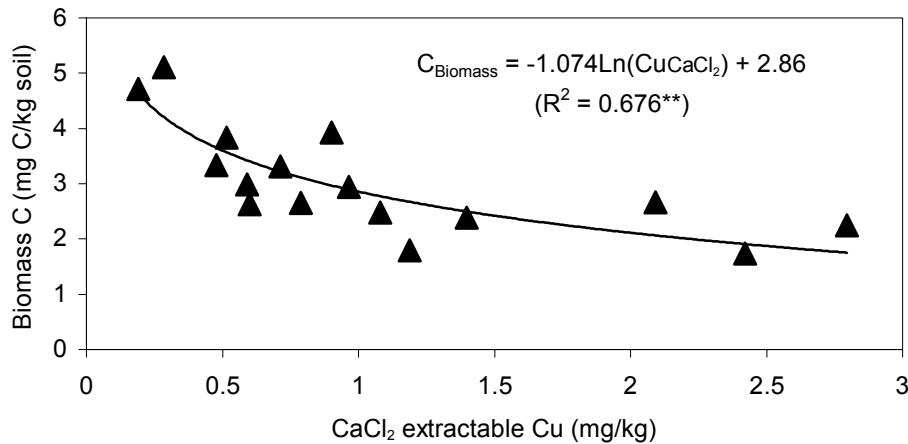


Figure 3. Relationship between microbial biomass-C and the concentration of CaCl₂ extractable Cu in the 0-2 cm horizons of the orchard and reference soils.

Figure 3 demonstrates the correlation between bioavailable (CaCl₂ extractable) Cu and microbial biomass in orchard and control soils. As the available Cu increased, microbial biomass decreased.

These data strongly suggest that microbial populations in the orchard soils are lower, not responding to additional carbon substrate (such as mulch), and show levels of metabolic stress in soils with Cu accumulation. These findings are in line with previous studies reporting the response of these indicators to soil heavy metal contamination. Whilst intensive horticultural management would have been expected to modify microbial properties within the orchard soils (Saggers *et al.* 2001), it is generally accepted that any effects of management would be minimal compared to impacts resulting from the long-term use of Cu fungicides (Graham *et al.* 1986; Filser *et al.* 1995).

Earthworm avoidance study

This study was designed to demonstrate the impacts of Cu contamination on earthworms. The composting worm *Eisenia fetida* was chosen for this study as it is prescribed as a test organism by the Organisation for Economic Co-operation and Development (OECD) for toxicity testing (OECD 1984) and is easily reared in a

commercially available worm farm. Although not a typical soil species for the local area it occurs in soil rich in organic matter and its susceptibility to chemicals resembles or exceeds that of true soil-inhabiting species (OECD 1984; Kula and Larink 1997). *E. foetida* is very prolific, has a short life cycle, and colonies are readily maintained in a laboratory.

Two reference soils were used - an artificial soil prepared according to OECD guidelines (OECD 1984), and a soil sourced from a site adjacent to the test soil but with no known history of Cu application. These were tested against a known Cu-contaminated soil sourced from beneath the drip zone of established trees at two local avocado orchards. Soil samples were taken from 0-10cm, below the litter layer. The Cu-contaminated soil was diluted by mixing it with the relevant reference soil to give 0, 5, 10, 25, 50 and 100 percent Cu-contaminated soil. Each dilution regime was tested in replicates of ten. Dilution mixtures were prepared based on their dry weights. Soil pH and moisture content were adjusted.

Trial Set-up

Avoidance trials were based on methodologies previously described by Yeardley *et al.* (1996). Transparent plastic containers (170x120x70mm) were divided in half by drawing a line on the outside and labelling the sides according to soil type. Test soil (200 g wet weight) was added to one side of the container and control/reference soil (200g wet weight) added to the other side of the container. A plastic divider was used between the soil types to avoid mixing, and help with even distribution. After addition of the soil, the divider was removed and the container lightly tapped to settle the soil. Ten clitellate worms were placed on the soil surface in each container along the centre line and the containers placed in a controlled-environment chamber at 22 °C (+/- 2 °C) under 24h/day illumination. Holes were punched in the lids of the containers and the lids put in place once the worms were observed to enter the soil.

After 48 hours, the containers were gently removed one at a time from the chamber for tallying, and the lids removed. The control and test soils were separated along the mid-line with a stainless steel spatula and the divider replaced. Soils from each side of the container could then be removed separately and the numbers of worms counted.

Worms found on the line between the two soils were excluded from the statistical analysis of avoidance.

Results and Discussion

A considerable increase in the number of earthworms avoiding the copper contaminated orchard soil was observed with increasing copper concentration (Fig. 4). The level of avoidance was significant ($p < 0.05$) at a soil copper concentration of 48 mg kg^{-1} , and remained significant across all concentrations above that threshold. At 715 mg kg^{-1} copper (ie undiluted orchard soil), only 7.5% of earthworms remained in the orchard soil, with 91.5% being found in the OECD control portion of the tub. Some worms were mid-line and did not account for avoidance statistics.

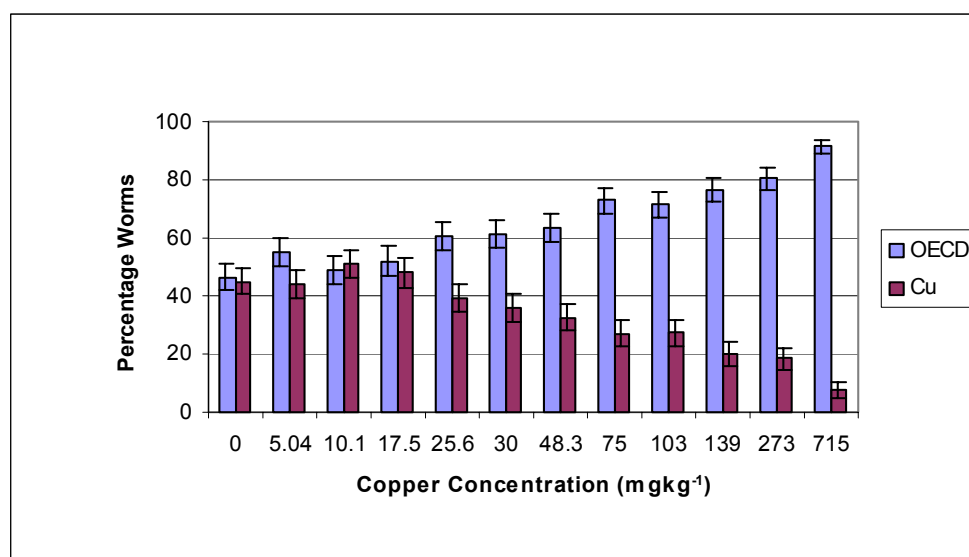


Figure 4. Percentage of earthworms ($n=200$) found in copper-contaminated (Cu) soil and in control (OECD) soil over dilution treatments (ie original soil had 715 mg kg^{-1} copper contamination).

When given the option of moving into an area of field-sourced control soil (soil near the orchard with no Cu contamination), the level of avoidance response with increasing copper concentration was more pronounced than that observed with OECD soil as a control (Fig. 5).

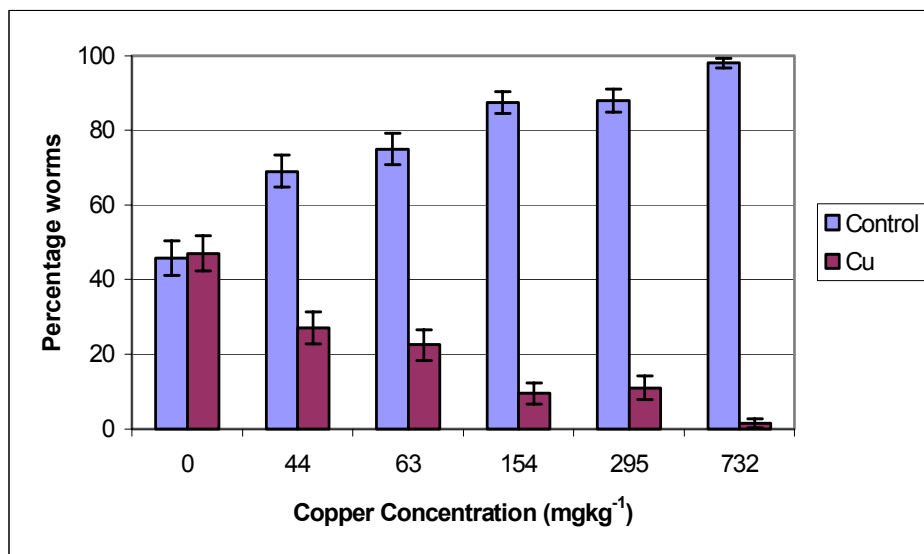


Figure 5. Percentage of earthworms (n=200) found in Cu-contaminated soil and in control soil over dilution treatments.

Earthworms aid decomposition and incorporation of organic matter, increase water soluble aggregates, improve water infiltration, aeration, drainage, root penetration, and increase microbial activity in soil (Edwards and Lofty 1977; Baker *et al.* 1994; Yeardeley 1996). Earthworm casts and burrow walls exhibit higher concentrations of total and plant-available nutrients than surrounding soil and it has been recognised that surface feeding species horizontally and vertically transport microorganisms, spores, pollen and seeds (Makeschin 1997) and can reduce plant pathogens through digestion of fungal spores (Hirst *et al.* 1955). Hence, as Cu has been shown to impart detrimental effects upon a range of beneficial soil organisms, including earthworms at concentrations as low as 12-34 mg/kg (Graham *et al.* 1986; Helling *et al.* 2000) consequences for orchard soils in Australia are expected.

Copper in the Organics Industry

This Natural Heritage Trust funded project jointly conducted by NSW Agriculture, Tuckombil Landcare and the University of Adelaide has demonstrated the impacts of even relatively low concentrations of copper in soil. Copper depleted biomass carbon

and increased respiration of microorganisms- indicating the microorganisms are stressed. Another more obvious impact of Cu in soil is its effect on earthworm populations. Our studies with *Eisenia fetida* in the laboratory have shown that this earthworm avoids soils with even relatively low concentrations of copper. Consistent with this, severely depleted populations of field earthworms were found in an orchard survey where Cu residues were detected.

Soil health is fundamental for the sustainability of agriculture, and in particular Organic industries. It has been discussed that if soil is improved, the whole agricultural system's health is improved (Pretty *et al.* 2002). The benefits of having healthy soils are numerous. For example, soil microorganisms produce polysaccharides that act as gums to bind and stabilise soil aggregates, hence influence soil structure (Stirling, 2001a). Microorganisms in healthy soils play a major role in nutrient cycling, making many nutrients available to plants. Healthy soils are also able to buffer against soil-borne diseases and nematodes (Stirling, 2001b), thus improving the health of crops. Agricultural sustainability starts with the soil by seeking to reduce erosion, and to make improvements to soil physical structure, organic matter content, water holding capacity and nutrient balance (Pretty *et al.* 2002). Contamination of organic orchards by Cu compromises soil health, and opposes the ideals of organic production.

The Organics Industry still relies on copper for disease control in numerous horticultural crops, however, with the restrictions placed on the use of this chemical, alternatives acceptable to the industry will need to be developed. NSW Agriculture will be conducting a review and producing an inventory of alternatives to copper for disease control (funded by RIRDC) in 2003. Future scientific evaluation of these alternatives is needed, and technologies will need to be developed to reduce the toxicity of Cu in contaminated organic orchards.

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