Industry wide risk assessment: case study of Cu in vineyard soils

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Abstract

There are concerns over the environmental risks posed by Cu-based fungicide use, and there is community and regulatory pressure on viticultural industries to restrict the use of Cu-based fungicides. This study assesses the relative environmental risks posed by Cu-based and alternative synthetic organic fungicide compounds used in Australian vineyards, giving particular consideration to adverse effects on soil microbial activity and how risks vary across different viticultural regions. The study was guided by key steps in the ecological risk assessment (ERA) framework to analyse the risks of Cu-based fungicides towards soil organisms and involved four key steps: (1) problem formulation; (2) analysis (characterise exposure and effects); (3) risk characterisation; (4) risk assessment. There is evidence of a build up of Cu-based fungicide residues in Australian vineyard soils, although this has occurred over many years, thus allowing the availability of the Cu in the soil to be attenuated over time due to aging processes. On the whole it appears that Cu-based fungicide residues are currently unlikely to pose a significant risk to soil organisms in Australian vineyard soils. However, there are indicators that continued applications of Cu-based fungicides may well have implications on the use of impacted land for sustainable agricultural production. Further detailed studies are required to enable a more definitive characterisation of the risks posed by C-based fungicide residues, such as establishing a clearer link between the laboratory and agricultural settings, investigating effects on other indicators of microbial activity and biodiversity, and understanding the resilience of soil microbes to additional stressors. The challenge for agricultural industries and governments, both in Australia and globally, is to formulate appropriate plans to reduce the risks associated with Cu-based fungicide use. Further research is required to consider the relative risks of a wide range of alternative fungicide compounds to ensure that they pose a lower environmental risk than the Cu-based fungicides they may replace.

Keywords: Copper fungicides; synthetic organic fungicide alternatives; vineyard soils; relative environmental risk;

Introduction

Fungicides are widely used in horticultural production systems, in particular vine and orchard crops, to control fungal diseases such as downy mildew (*Plasmopara viticola*), grey mould (*Botrytis cinerea*) and black spot (*Diplocarpon rosae*). Agricultural best practices include adoption of preventative management strategies to control the spread of diseases and this typically involves the regular application of fungicides throughout the growing season whether or not fungal infection is present (McConnell et al. 2003). Regular use of Cu-based fungicides in Australian vineyards has led to an increase in the concentrations of Cu in the surface soils (Wightwick et al. 2010a,b). Although the proportion of available Cu is relatively low, Cu residues may adversely affect soil organisms (e.g. microbes, earthworms) which could have implications for the sustainable use of this land for agriculture.

There are concerns over the environmental risks posed by Cu-based fungicide use, and there is community and regulatory pressure on viticultural industries to restrict the use of Cu-based fungicides. However, in Australia the environmental risks of Cu-based fungicides to organisms in vineyards soils has been little studied, so there is little scientific basis to inform policies on the restriction to the use of these fungicides. Moreover, it is difficult to make generalisations about likely risks because the availability and toxicity of Cu to soil organisms varies widely across different soils (Wightwick et al. 2010a). Alternative synthetic organic fungicide compounds are registered for use, however little is known about their environmental fate and toxicity, so it is not known if they are likely to pose a lower environmental risk than Cu-based fungicides. This study assessed the relative environmental risks posed by Cu-based and alternative synthetic organic fungicide compounds used in Australian vineyards, giving particular consideration to adverse effects on soil microbial activity and how risks vary across different viticultural regions. This new information will help inform government and industry in formulating evidence-based policies for the future management of Cu-based fungicides in Australian vineyards.

Methodology

The study presented herein was guided by key steps in the ecological risk assessment (ERA) framework to analyse the risks of Cu-based fungicides towards soil organisms (US EPA 1998; NEPC 1999). In short, the ERA framework involves four key steps: (1) problem formulation; (2) analysis (characterise exposure and effects); (3) risk characterisation; (4) risk management. This

desk-top study itself broadly followed the same framework by conducting a literature review to formulate the problem to be investigated and to characterise exposure and possible effects, before assessing the environmental risks of fungicide use in Australian viticulture.

Results and Discussion

Several surveys have generated information on the concentrations of total Cu in the surface soils of vineyards in different regions of Australia ([Pietrzak and McPhail 2004](#); [Wightwick et al. 2008](#)), and these showed evidence of a build up of Cu-based fungicide residues. This has occurred over many years, thus allowing the availability of the Cu in the soil to be attenuated over time due to aging processes. This latter is an important consideration, as it is generally considered that soil organisms are not adversely affected by strongly bound Cu (i.e. 'aged' Cu) in soil. However, until recently there was no information on the availability of the Cu-based fungicide residues in Australian vineyard soils, information required to evaluate the likely exposure concentrations for soil organisms. However, recently [Wightwick et al. \(2010b\)](#) characterised the environmental availability of Cu-based fungicide residues in vineyard soils from 10 different viticultural regions of Australia. The availability of the Cu (CaCl_2 extractable) in the Australian vineyard soils was found to be relatively low, typically being $< 0.5\%$ of the total Cu in the majority of vineyard soils. The availability of Cu in the vineyard soils was primarily influenced by the total Cu concentration of the soil, but also related to soil properties influencing cation exchange processes and capacity for soil to adsorb Cu (i.e. pH, clay, exchangeable K, silt, CaCO_3). In particular the availability of Cu increased with increasing soil acidity, and there were also some indications that availability increased with increasing soil alkalinity (above pH 8). This information may prove useful in devising strategies to reduce the availability of Cu in agricultural soils, where this is deemed to be required. It is also valuable information for understanding which soils are likely to have the greatest capacity to buffer bioavailable Cu with further increases in Cu-based fungicide residues.

In assessing the risks of Cu-based fungicide residues, toxicity assessments with field contaminated soils are preferred over the use of 'artificially' contaminated soils, as this accounts for Cu-aging in the soil and the adaptation of soil organisms to the gradual increases in soil Cu concentration. The ecotoxicological effects of long-term exposure of Cu-based fungicide residues on enzyme activities (as sensitive indicators of microbial activity) in Australia vineyards soils was reported by [Wightwick et al. \(2012a\)](#) using soils collected from vineyards in three different

viticultural regions of Australia representing soils with contrasting Cu availabilities. [Wightwick et al. \(2012a\)](#) used a soil microcosm approach to assess the effects of Cu accumulation on microbial function in vineyard soils. Surface soil samples were collected from 10 vineyards and a number of un-impacted reference sites in each of three different viticultural regions of Australia. The field collected soils were transferred to microcosms and maintained for up to 93 days in the laboratory at 20-22°C and 60 % of their maximum water holding capacity. The microbial function of the soils was indicated by measuring phosphomonoesterase, arylsulfatase, urease, and phenol oxidase activity. In general, the vineyard soils had greater concentrations of Cu and lower enzyme activities than in the reference soils. Assessment of the data from that experiment highlights the importance of taking a broader more regional and industry wide approach. Indeed, the results suggested that enzyme activities were being driven predominately by physical-chemical soil properties (i.e. organic C, pH) at the concentrations of Cu present in the studied soils (where 90 % of the vineyard soils contained total Cu concentrations less than 115 mg/kg). This suggests that increased Cu concentrations from the regular use of Cu-based fungicides are currently unlikely to pose a risk to the microbial activity (as indicated by enzyme activities) of Australia vineyard soils.

A limitation of the reductionist approach to the incubation experiment used by [Wightwick et al. \(2012a\)](#) is that whilst ‘real’ field-collected soils were used, the experiment was conducted outside of the agricultural setting. Consequently, it is challenging to relate those findings back to an agricultural context *per se*. So, whilst the results showed that, even within the discrete regions, physical-chemical soil properties were significant factors influencing enzyme activities, it is not known how different viticulture practices may have influenced these varied physical-chemical properties. Moreover, that study could have been improved by including a general measure of soil microbial biomass, as this would have enabled an assessment as to whether the influence of soil factors (e.g. Cu, physical-chemical properties) on enzyme activities was due to interactions with the enzymes themselves and/or due to influences on the overall microbial biomass/community.

Although it appears that Cu-based fungicide residues are not directly affecting soil microbial activity, is that persistent Cu stress affecting the susceptibility of microbial communities to additional stressors/disturbances? To begin to address this question, [Wightwick et al. \(2012b\)](#) used ‘artificially’ contaminated soils to determine the Cu concentration at which an additional heat disturbance affected the soil’s phosphomonoesterase and urease activity. [Wightwick et al. \(2012b\)](#) suggest that at the concentrations of Cu typically reported in Australian vineyard soils,

Cu stress is unlikely to cause lasting alterations to the susceptibility of phosphomonoesterase and urease activity following a single heat disturbance, although, Cu-based fungicide residues in Australian vineyard soils may be harming microbial mediated P processes. This represents a loss of ecosystem service which in the long-term may have cost implications for viticulturists; if Cu-based fungicide residues continue to accumulate, disruption to P cycling is likely to result in the need to apply more P based fertilisers to meet plant/crop requirements.

Ecological investigation levels (EILs), or soil-specific guideline values for Cu have recently been included in the proposed revision of the National Environmental Protection Measure (NEPM) for site contamination in Australia (NEPC 2011). Normalisation relationships have also been included to account for known influences of physical-chemical soil properties on the toxicity of Cu to different species/processes/endpoints. Unlike the outgoing version of the NEPM, these EILs have been derived based on ecotoxicological data for plants, invertebrates and microbial processes in soil, with each of these groups being represented amongst the most sensitive endpoints. Using the guidance material in the revised NEPM, soil-specific EIL's were derived for vineyards in 10 different regions of Australia using data from [Wightwick et al. \(2008; Table 1\)](#). In all but three of the regions, the observed increased Cu concentrations in vineyards exceeded the soil-specific high ecological value EIL (Table 1), with 10-50 % of vineyards exceeding this EIL in each region except Margaret River and Tasmania. However, only one vineyard exceeded the less sensitive urban residential EIL which is based on 30% effect concentrations (EC₃₀), although many of the vineyard sites were nearing this EIL. The urban residential EIL is arguably of greater relevance to highly disturbed agricultural systems than the high ecological value EIL which is based on experimental lowest observed effect concentrations and is primarily for national parks and areas of high conservation value. Consequently, Cu may increasingly pose a risk to soil ecosystems if Cu-based fungicide residues continue to accumulate in Australian vineyards soils, with the risks likely to be greatest in regions with acidic soils (e.g. the Southern Highlands, Riverina). It is challenging to predict the time-frame in which Cu concentrations are likely to reach more critical concentrations as the accumulation of Cu-based fungicide residues is dependent on many factors (e.g. rate of Cu-based fungicide application, soil physical-chemical properties, disease pressure). However, based on data in [Wightwick et al. \(2008\)](#), Cu residues may reach concentrations exceeding the urban residential EIL in around 2030 – 2040 (based on an estimated annual accumulation of 2 – 5 mg/kg total Cu). Further studies are needed to model such predictions more precisely.

Table 1 Comparison of Cu concentrations in vineyard soils from 10 regions of Australia to soil-specific ecological investigation levels

Region	Typical Cu concentrations and physical-chemical soil properties of vineyards ^a				Soil-specific ecological investigation level (EIL) ^b			
	pH ^c	TOC	CEC	Added Cu	EIL ^b	Sites exceeding EIL (%)	Added Cu ^c	
		(%) ^c	(cmol _c /kg) ^d	(mg/kg) ^{d,e}			(%) ^{d,e}	
Murray Valley	8.7	1.1	16.5	55 (26 – 114)	80	10	69	(33 – 142)
Southern Highlands	5.6	3.6	14.3	79 (18 – 217)	55	44	144	(33 – 395)
Riverland	7.7	2.8	23	67 (42 - 108)	95	10	71	(44 – 114)
Barossa	7.2	2.8	14.2	38 (11 – 80)	85	0	45	(13 – 94)
South-east, SA	7.8	2.9	26.7	63 (4 – 133)	75	30	8	4 (5 – 178)
Riverina	6.6	1.7	21.1	83 (40 – 152)	80	50	103	(61 – 190)
Mudgee	6.0	1.6	8.5	47 (26 – 78)	60	22	78	(44 – 129)
Swan Valley	5.9	1.4	1.6	17 (3 – 34)	15	30	113	(20 – 224)
Margaret River	5.9	5.2	9.0	14 (0 – 29)	60	0	24	(0 – 48)
Tasmania	6.4	4.1	19.5	35 (8 – 69)	70	0	49	(12 – 98)

^a Data from [Wightwick et al. \(2008\)](#); Mudgee, n = 9; Margaret River, n = 13; Tasmania, n = 7; southern highlands, n = 9; all other regions, n = 10;

^b Derived using guidance in the NEPM (site contamination) (NEPC 2011); EIL for national parks and areas of high conservation value

^c Added Cu concentration relative to EIL

^d, data mean, as calculated from [Wightwick et al. \(2008\)](#)

^e, data range in [Wightwick et al. \(2008\)](#)

Table 2 Summary of the relative behaviour and toxicity of five key classes of fungicides ([IUPAC 2010](#))

Risk factor	Copper-based	Phthlamide	Dithiocarbamate	Triazole	Strobilurin
Mobility in soil	Low	Low	Low to medium	Low to medium	Low
Persistence in soils	High	Low	Low	Moderate to high	Low to moderate
Toxicity to earthworms	Low to moderate	Moderate	Low to moderate	Low to moderate	Low to moderate
Toxicity to aquatic invertebrates	High	Moderate	Low	Low	Low to moderate
Toxicity to fish	Moderate	Moderate	Low to moderate	Low	Low to moderate
Toxicity to aquatic primary producers	Moderate	Low to moderate	Low	Low	Low

In addition to the more widely documented risks to terrestrial soil organisms the use of Cu-based fungicide can pose a risk to nearby surface water bodies due to the run-off of accumulated Cu from agricultural soils. For instance, [Bereswell et al. \(2012\)](#) reported that surface water sediment Cu concentrations can be in the same range as those actually measured in cultivated vineyard soils in the Palatinate wine-growing region of south-west Germany. So, does the use of Cu-based fungicides pose a risk to organisms in streams or lakes in popular viticultural areas? To begin to address this question [Wightwick et al. \(2012c\)](#) conducted a reconnaissance survey of the presence and concentration of copper in sediments at 18 sites within the Yarra River catchment. Observed Cu concentrations in sediment samples from sites in important horticultural areas of the region (mean (95% confidence interval) 12.0 (10.6-13.6) mg/kg dry weight) were similar to the concentrations present in the samples from reference sites in the catchment (mean (95% confidence interval) 12.0 (6.7-16.8) mg/kg dry weight). The data on Cu in the sediments suggests that there is unlikely to have been wide spread, diffuse, off-site transport of Cu from the soils of horticultural properties to nearby surface waterways in the Yarra River catchment, and that that observed sediment metal concentrations are unlikely to pose an ecological risk to sediment dwelling organisms at the study sites.

There are currently 47 different fungicides (active ingredients) registered for use in Australian vineyards representing 16 different chemical classes ([Wightwick et al. 2012d](#)). Like Cu, the majority of these fungicides are designed as preventative sprays to protect against disease infection before it occurs. Many current synthetic fungicides were developed in the 1940 – 50's and are based on 'old fungicide chemistry'; these have broad-spectrum multi-site modes of action and are applied at high application rates (1.5 – 3.5 kg/ha), e.g. the dithiocarbamates and phthlamides ([Russell 2005](#)). Other fungicides developed since 1970 are based on 'modern fungicide chemistry'; these have specific modes of action, greater activity and are applied at relatively low application rates (0.13 – 0.25 kg/ha; [Russell 2005](#)). In conventional viticulture some of these synthetic organic fungicide compounds may be used more frequently in the future. But are the alternatives to Cu-based fungicides in fact less risk for the environment? To begin to address this question, [Wightwick et al. \(2012e\)](#) conducted laboratory experiments comparing the effects of captan and trifloxystrobin and copper hydroxide on soil enzyme activity. Compared to an untreated control, Cu did not inhibit either enzyme activity, even at the highest dose used (156 mg/kg). At their respective highest doses, captan (96 mg/kg) and trifloxystrobin (144 mg/kg) did not cause inhibition of phosphomonoesterase activity, but did inhibit urease activity. Consequently, [Wightwick et al. \(2012e\)](#) suggest that the copper hydroxide alternatives, captan

and trifloxystrobin, do not pose a short term risk to P cycling processes in soil, although the results do suggest that they are more toxic than copper hydroxide to N cycling processes in soil. Overall, captan and trifloxystrobin are likely to pose a lower long-term risk to soil enzyme activity than Cu-based fungicides.

The presence of synthetic organic fungicide residues in surface waters of Australian horticultural production catchments was studied by [Wightwick et al. \(2012d\)](#). Although fungicide residues were frequently detected (i.e. in 63 % of water samples), [Wightwick et al. \(2012d\)](#) suggest that the risks to aquatic ecosystems and organisms at the concentrations observed are unclear as there is relatively little ecotoxicological data available with which to compare the field data, particularly for aquatic fungi and micro-organisms. Moreover, often multiple fungicides were detected in any given sample. The significance of these mixtures on toxicity to aquatic organisms is also largely unknown. Nonetheless it appears that, synthetic organic fungicide compounds are likely to pose long-term (chronic) rather than short-term (acute) risk to aquatic ecosystems. Some information on the expected behaviour and toxicity of synthetic organic fungicide compounds was reported in [Wightwick et al. \(2010a\)](#), and can also be used to indicate their relative risks (Table 2). This comparative analysis suggests that increased use of phthalamides (e.g. captan), dithiocarbamates (e.g. mancozeb) and strobilurins (e.g. trifloxystrobin) over Cu-based fungicides is likely to result in decreased risks to soil organisms due to their much lower persistence in soil. Furthermore, a move away from Cu-based fungicides is likely to result in decreased risks to aquatic organisms, particularly if dithiocarbamates and triazoles are used. Finally, triazoles need to be used with caution as they may pose a risk to soil organisms as they may persist in surface soils for several months.

Cu-based fungicide residues can present other issues in vineyard soils beyond risks to soil organisms. For instance, elevated concentrations of Cu in soil can adversely affect plants. The accumulated Cu in vineyard soils is not expected to harm mature grapevines because they tend to have deep root systems that extend below the surface soils ([Chopin et al. 2008](#)). However, Cu may be toxic to shallow rooted plants used for cover crops within the vineyard, or other crops in the case of changed land use ([Brun et al. 2001](#); [Michaud et al. 2007](#)). For instance, since 2000 many old vineyards in France have gone out of production due to economic pressures, with much of this land being converted to wheat production ([Michaud et al. 2007](#)). Unfortunately, the elevated soil Cu concentrations from the long-term use of Cu-based fungicides in France are causing issues with phytotoxicity and reduced yields in wheat crops ([Michaud et al. 2007](#)).

Significant areas of old grapevine plantings have also recently gone out of production. It is not yet clear what the new land uses will be, but based on published soil and ecotoxicological data (Wightwick et al. 2008; NEPC 2011), the Cu concentrations currently in the soils of old Australian vineyards may well pose a risk to a number of agricultural plant species, including wheat, barley, ryegrass, and tomato.

There are a number of low cost in-situ techniques that can be used to ameliorate the effects of Cu toxicity should land holders be required to remediate vineyard soils on changed land-use, including stabilisation / immobilisation (e.g. the addition of organic or inorganic soil amendments to reduce the availability of Cu), phytoremediation (e.g. use of plants to extract metals from soils) and soil mixing (e.g. decreasing Cu concentrations in surface soils by mixing them with less contaminated horizons; Bolan and Duraisamy 2003; Mench et al. 2007; Pietrzak and Uren 2011). Given the strong influence of pH, CEC and TOC on the availability of Cu in Australian vineyard soils (Wightwick et al. 2010b), the use of inorganic (e.g. lime) and organic (e.g. composts) amendments may be a cost effective approach with the added benefit of improving the overall soil health.

Globally there are societal and regulatory pressures to move towards restrictions on Cu-based fungicide use. In particular, the European Commission via its Plant Protection Products Directive is requiring countries to develop and set pesticide risk reduction targets. Specifically, steps need to be taken to ensure the rate and number of Cu-based fungicides applications is minimal to achieve desired effects (EC 2009). Promotion of Australian viticultural products (particularly wine) is to a large extent reliant on the industry being 'environmental responsible' (Russell and Battaglene 2007), and these European Commission policy decisions are likely to place viticultural industries and governments in Australia under pressure to follow suit, to maintain consumer confidence and access to export markets. As such, viticultural industries should be considering appropriate risk management options; these can range from longer term passive approaches (e.g. self-regulation, improved farm management practices) to high level active approaches utilising regulatory instruments (e.g. restrictions on use, phase out).

The risks currently posed by Cu in Australian vineyards are relatively low, and it would seem appropriate for industry to adopt passive risk management approaches. The industry is already taking a lead on this, for instance by promoting best management practices which encourage adoption of integrated pest management (IPM) to reduce reliance on fungicide use (McConnell et

al. 2003). Indeed, many viticulturists are aware of concerns around Cu-based fungicide use and are self-regulating restrictions on their use (pers comm. Geoff McCorkelle, Vineyard Technical Manager, McWilliams Wine Group, April 2011). However, perhaps further steps could be taken to encourage reduced Cu-based fungicide use before resorting to ‘heavy handed’ regulatory enforcement instruments. For instance, the Australian wine industry has an environmental stewardship program which is providing an economic incentive for participating viticulturists to improve their ‘environmental performance’ (WFA 2011). As part of this environmental stewardship program the industry could more explicitly state agreed restrictions on Cu-based fungicide use in order to facilitate improved risk minimisation and to demonstrate appropriate risk management actions to consumers and markets.

Conclusions

Several recent publications have generated new information on the chemistry and ecotoxicological effects of Cu-based fungicide residues in Australian vineyard soils. This has provided scientific evidence that viticultural industries in Australia can use for environmental reporting, benchmarking against viticultural/wine industries in other countries, and formulating evidence-based policy decisions on the future management of Cu-based fungicides. On the whole it appears that Cu-based fungicide residues are currently unlikely to pose a significant risk to soil organisms in Australian vineyard soils. However, there are indicators that continued applications of Cu-based fungicides may well have implications on the use of impacted land for sustainable agricultural production. Further detailed studies are required to enable a more definitive characterisation of the risks posed by C-based fungicide residues, such as establishing a clearer link between the laboratory and agricultural settings, investigating effects on other indicators of microbial activity and biodiversity, and understanding the resilience of soil microbes to additional stressors. However, regardless of the actual risks posed, there are still societal and regulatory pressures to introduce restrictions on the use of Cu-based fungicides. The challenge is now for agricultural industries and governments, both in Australia and globally, to formulate appropriate plans to reduce the risks associated with Cu-based fungicide use, and this should be the focus of future research and development. This needs to consider the relative risks of alternative fungicide compounds to ensure that, on the whole, they are likely to pose a lower environmental risk than the Cu-based fungicides they replace.

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