

Review

The Legacy of Copper-Based Fungicide Use in Vineyard Soils: Accumulation, Ecotoxicity, Human Health Risks, and Sustainable Remediation

Marija Poljak¹, Ivica Kisić², Monika Zovko², Milan Mesić² and Aleksandra Perčin^{2,*}¹ Institute of Public and Environmental Health, Zaloška cesta 155, 1000 Ljubljana, Slovenia² Division of Agroecology, University of Zagreb Faculty of Agriculture, Svetošimunska cesta 25, 10000 Zagreb, Croatia* Correspondence: apercin@agr.hr**How To Cite:** Poljak, M.; Kisić, I.; Zovko, M.; et al. The Legacy of Copper-Based Fungicide Use in Vineyard Soils: Accumulation, Ecotoxicity, Human Health Risks, and Sustainable Remediation. *Earth: Environmental Sustainability* **2026**, *2*(2), 154–168. <https://doi.org/10.53941/eesus.2026.100011>

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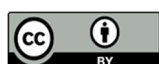
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Abstract: The long-term use of copper (Cu)-based fungicides in viticulture has led to substantial Cu accumulation in vineyard soils, raising concerns about soil health, ecological integrity, plant physiology, and potential human exposure. Since their introduction in the late 19th century, Cu-based fungicides have remained essential for controlling fungal diseases, particularly in organic production systems where viable alternatives are limited. In contrast to organic systems, conventional viticulture can rely on modern synthetic fungicides (such as fluxapyroxad, pyraclostrobin, tebuconazole, and fludioxonil), which provide less harmful alternatives to copper because they are applied at much lower rates and do not accumulate in soils, making them generally more environmentally benign than copper based fungicides. As a result, copper concentrations in vineyard soils frequently exceed European regulatory thresholds, often surpassing 200 mg/kg soil and occasionally exceeding 1000 mg/kg soil. Copper accumulation is influenced by soil physicochemical properties, climate, terrain, and vineyard management practices, while its bioavailability dictates its ecological and toxicological impacts. Elevated copper levels adversely affect soil biota, reducing microbial diversity, impairing enzymatic activity, and disrupting nutrient cycling and key soil functions that underpin fertility and ecosystem stability. Although mature grapevines demonstrate relatively high copper tolerance through deep rooting and physiological detoxification mechanisms, young vines and crops planted following vineyard conversion are more susceptible to copper toxicity. In humans, copper is an essential micronutrient with a U-shaped dose-response curve and toxicity is uncommon at typical dietary exposure levels. Nonetheless, copper residues in grapes and wine can occasionally exceed the maximum limits established by the International Organisation of Vine and Wine (OIV), underscoring the need for continued monitoring. Mitigating copper contamination requires preventive approaches, such as reducing copper inputs, employing forecasting tools, adopting precision viticulture technologies, and cultivating resistant varieties, alongside sustainable remediation strategies. Among these, phytoremediation, particularly phytoextraction, offers a promising, non-invasive, and cost-effective method for managing low to moderately copper contaminated vineyard soils.

Keywords: regulatory standards; soil enzymatic activity; soil functions; soil microorganisms; human health risk; phytoremediation; sustainable viticulture



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1. Introduction

Copper-based fungicides have been a central component of plant protection in European viticulture for more than a century [1–5], primarily because of their reliable efficacy against downy mildew (*Plasmopara viticola*) [6,7]. Despite substantial progress in the development of modern fungicides [8,9], copper (Cu) remains essential in organic production systems [10–12], where no fully effective alternative is currently available. Consequently, copper-based products continue to play a major role in organic vineyards in Europe [5]. The persistent use of copper in viticulture is accompanied by a steadily growing global market for copper-based fungicides. In 2024, the global copper fungicides market size was estimated at USD 478.38 million by Grand View Research [13] and is projected to reach USD 648.19 million by 2030. In the European Union, copper use is subject to strict regulatory limits due to its persistence in the environment and well-documented ecological risks. Current legislation restricts annual applications to 4 kg Cu/ha, averaged over several years, yet vineyards frequently operate at or near these limits [14]. Because copper is poorly mobile and strongly retained in the upper soil layers, repeated applications lead to progressive accumulation, particularly in long-established viticultural regions [4,15–24]. The accumulation of copper in vineyard soils has raised increasing concern regarding its effects on biocenosis [25], soil biochemical properties, soil functions [26], phytotoxicity [27,28] and the potential transfer of copper through the food chain [29,30]. These issues have become especially relevant in the context of sustainable viticulture, where producers must balance effective disease control with the need to reduce environmental pressures and comply with tightening regulatory frameworks.

The review aims to deliver a comprehensive synthesis of current knowledge regarding the long-term implications of copper-based fungicide application in viticultural systems. Particular emphasis is placed on copper accumulation in vineyard soils and its influence on key biological and chemical processes (soil health), including alterations in microbial community structure and enzymatic activities. The review further addresses copper phytotoxicity, its potential translocation through the food chain, and associated risks to human health. In addition, it examines the prevailing European regulatory framework and evaluates sustainable management and remediation strategies designed to mitigate ecological impacts and safeguard the long-term resilience of viticulture.

2. EU Regulatory Framework and Safety Standards

Cu-based pesticides are classified as inorganic pesticides and, together with sulfur-based pesticides, are the only inorganic pesticides approved for use in both conventional and organic farming [31]. Conventional and organic farmers in the EU can use five Cu-based active ingredients approved by the European Commission (cuprous oxide, copper hydroxide, Bordeaux mixture, copper oxychloride, and tribasic copper sulfate) [32]. Current approval of this Cu-based active substance will expire on June 30, 2029. Individual EU Member States may choose whether or not to register specific Cu compounds as plant-protection products; for example, five of the 27 Member States, Denmark, Estonia, Finland, the Netherlands, and Sweden, have not approved any copper-based products for this purpose [33–35].

EU member states set the threshold for Cu in agricultural soil through national legislation. However, due to the accumulation of Cu in soil and its potentially harmful effects, several EU legal acts bind member states. The Council directive on the protection of the environment, and in particular of the soil, when sewage sludge is used in agriculture (86/278/EEC) [36] defined limit values for Cu concentration in soil from 50 to 140 mg/kg. To initiate mechanisms to reduce the use of Cu in agriculture, the EU adopted Commission Implementing Regulation (EU) 2018/1981 [14], which limits the use of Cu-based plant protection products (PPPs) to a maximum application amount of 28 kg/ha over seven years. In the same way (28 kg/ha/7 year), the maximum permitted rate of Cu-based PPPs in organic agriculture is regulated by Commission Implementing Regulation (EU) 2021/1165 [37]. Demeter International standards have even stricter limits [38]. Cu may be used at an average of 3 kg/ha/year over 7 years, preferably with a maximum of 500 g/ha/spray, with possible exemptions in wine- and hop-growing regions with high fungal pressure. In these cases, the respective certifying organization may grant an exemption for the use of an average amount of up to 4 kg/ha/y over 7 years for grapes and hops. In addition, Cu is listed as a candidate for substitution (CFS) in part E of the Annex to Commission Implementing Regulation (EU) 540/2011 [39], indicating that the goal is to phase out Cu use in EU agriculture. Copper compounds are considered persistent and toxic and thus meet at least two of the three (persistent, bioaccumulative, toxic) criteria for CFS labelling [33].

This regulatory restriction reflects growing concern about the environmental footprint of copper use, particularly its tendency to accumulate in vineyard soils. Despite these strict limits, long-term application has resulted in substantial copper accumulation in surface soil layers. Understanding the extent and drivers of this accumulation is essential for assessing its ecological consequences.

3. Copper Accumulation in Vineyard Soils

Copper accumulation in vineyard soils, in both conventional and organic viticulture, results from the combination of intensive and long-term fungicide applications and the intrinsic chemical properties of copper [16–21]. Before the introduction of current regulatory limits of 4 kg Cu/ha per year, application rates in the 20th century commonly reached 20 to 30 kg Cu/ha annually, and in some cases exceeded 80 kg Cu/ha [33]. Although currently inputs are substantially lower, copper use still varies considerably between production systems; for example, in 2013 German organic winegrowers applied nearly three times more copper (2.29 kg Cu/ha) than conventional winegrowers (0.8 kg Cu/ha) [29,40]. In addition to these differences between production systems, the actual annual copper accumulation in soil is strongly shaped by weather conditions, site climate, latitude, exposition as well as grapevine cultivar [6,41,42]. After fungicide applications, 90% of the applied Cu is deposited in the soil [15]. On the other hand, the effectiveness of copper in foliar application is high due to its relatively low water solubility, which prevents it from being washed away by rainfall and allows it to remain on leaves longer than other fungicides, but still can contribute copper accumulation in soil [7].

Copper accumulation and distribution in vineyard soils are governed by a combination of soil properties, environmental conditions, and long-term management practices. Key physicochemical factors such as pH, organic matter content, clay fraction, and the presence of Fe, Al, and Mn oxides strongly influence copper retention [4,43,44], as copper binds readily to organic matter and clay minerals and therefore remains largely immobilized in the upper soil horizons. Although most copper is associated with stable solid-phase fractions, a smaller portion occurs as Cu^{2+} or as organic complexes in the soil solution, with mobility increasing under acidic conditions or in the presence of dissolved organic carbon [7]. Environmental factors such as rainfall, slope, erosion, and tillage can enhance copper redistribution [2,42,45], while the duration and intensity of fungicide use determine the overall copper load. In vineyard soils, copper is typically partitioned into bioavailable, potentially bioavailable, and immobile fractions (Figure 1), with shifts between these pools driven by changes in soil chemistry and environmental conditions [46–49].

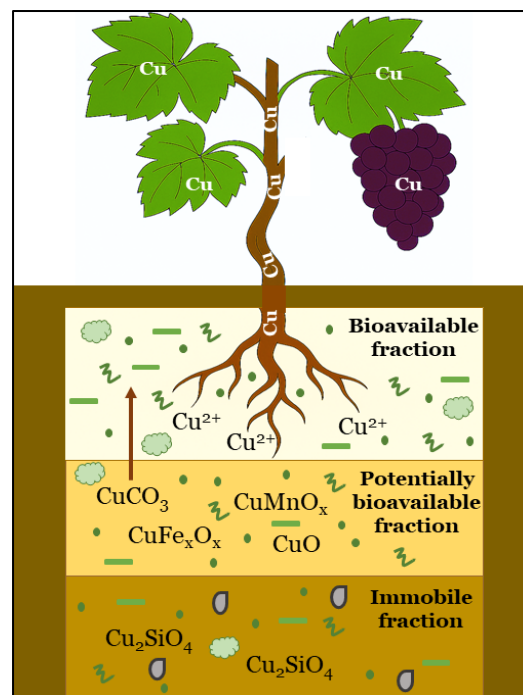


Figure 1. Conceptual Diagram of Copper Speciation and Bioavailability in Vineyard Soils. Source: Authors' own illustration based on the literature referenced in this section.

Long-term application of copper-based fungicides (≥ 30 years) in intensive horticultural systems leads to substantial copper accumulation in surface soil layers, often exceeding 150 mg Cu/kg, whereas vineyard soils commonly exhibit even higher copper concentrations [43]. Total copper concentrations in vineyard soils often exceed 200 mg/kg and can occasionally reach concentrations higher than 1000 mg/kg [22–24], while the maximum copper value registered in vineyard soils was as high as 3200 mg/kg [41]. Copper threshold values are often based on total soil metal content, but for assessing the potential toxicity of metals, information on the content of mobile and bioavailable forms of metals in the soil is much more important [2,50,51].

The intensive and long-term application of Cu-based fungicides can potentially harm human health through crop residues, reduce biodiversity by harming non-target organisms, increase pest resistance, and contribute to freshwater eutrophication, and marine ecotoxicity [5,33,52]. These ecological implications form the basis for the following sections, which examine in detail the effects of copper on soil biota, soil functions, and enzyme activity.

4. Effects of Copper on Soil Biota, Soil Functions and Enzyme Activity

Soil biota, which includes microorganisms, mesofauna, macrofauna and plant roots, represents one of the most sensitive components of vineyard ecosystems [53]. Approximately one quarter of global biodiversity resides in soil [54]. The response of soil organisms to copper depends primarily on its bioavailability, which is governed by soil pH, organic matter, clay content, Fe/Mn oxides, phosphate content, cation exchange capacity (CEC) and copper speciation [25,55]. Agrotechnical practices that alter soil structure, pH or organic matter content can indirectly influence copper mobility and toxicity [25]. Anthropogenic copper is generally more mobile and bioavailable than pedogenic copper, and therefore more toxic to soil organisms [10,50,56]. Copper bioavailability typically decreases with increasing pH and organic matter content [57] and rarely correlated with total soil copper [25].

The sensitivity of different soil organism groups to copper varies widely. Bacteria, algae and fungi are the most sensitive, while nematodes and earthworms show greater tolerance. Overall sensitivity follows the order: algae < bacteria < fungi < actinomycetes < protozoa < nematodes < collembola < earthworms [58]. Experimental studies show substantial reductions from 50% to 86% in bacterial growth and activity due to copper contamination in soil (508–8112 kg Cu/ha) [12]. Field study conducted in Australia on agricultural soil treated with copper sulphate at concentrations ranging from 0 to 3310 mg/kg monitored the effect of the treatment on soil microorganisms 12 years after application. The results show that treatments with copper doses higher than 200 mg/kg caused a selection pressure on the microbial communities and that the microbial communities in the soils treated with medium copper doses (200–700 mg/kg) differ significantly from the microbial communities in the soil treated with low (<200 mg/kg) and high doses of copper (700–>1000 mg/kg) [59]. Numerous authors report differences in metal solubility and potential toxicity between metal-spiked soils and field-contaminated soils [7,60]. Copper potential toxicity is higher in metal-spiked soils, which can be related to soil aging processes [60] because, with time, the proportion of the bioavailable Cu fractions in soil decreases while the proportion of the bound Cu fractions in soil increases [61]. Further to the above, authors from Belgium found no phytotoxic or adverse effect of copper on microbiological processes and invertebrates in the soil sampled from six European vineyards in which the copper content ranged from 142 to 689 mg/kg, while in spiked soil with the same amount of copper, a toxic effect on the mentioned organisms is present [1]. In addition, available studies indicate that different copper formulations (copper sulfate, copper (II) hydroxide, copper (II) oxide, copper oxychloride, and copper chloride) exert similar effects on soil organisms [12].

A comparative overview of threshold values and observed effects is listed in Table 1, which summarizes the main organism groups affected in vineyard soils.

Table 1. Sensitivity of soil organisms to copper contamination.

Soil Organism Group	Copper Input and Copper Soil Levels	Observed Effects	Notes	Source
Bacteria	150–200 mg/kg *	Reduced growth, community shifts	Highly sensitive	[62]
Bacteria	>200 mg/kg *	Community restructuring	At >800 mg/kg Cu, communities become dominated by biofilm-forming species	[59]
Fungi	2028–4056 kg/ha **	Growth rate decreased by 20%	Not affected (508–1016 kg/ha Cu) Growth rate decreased 45% at 8112 kg/ha Cu	[12]
Actinomycetes	200 mg/kg *	Inhibited growth	Less tolerant than gram negative heterotrophic and nitrogen fixing bacterial population	[63]
Nematodes	>3200 kg/ha **	Minimal effect unless pH < 5	Sensitive to acidity	[12]
Collembola	400–4000 kg/ha ** (acute)	Reduced diversity	Strong soil-type dependence	[12]
Earthworms	>33–80 mg/kg *	Reduced abundance, impaired activity	Reliable bioindicators	[64,65]

* The values given in mg/ha represent copper amounts in vineyards soils. **The values given in kg/ha represent copper application rates in vineyards.

Soil functions, including nutrient cycling, organic matter decomposition, carbon sequestration, water storage and purification, are closely linked to the activity of soil biota [66]. Excessive copper (>200 mg/kg) disrupts these functions primarily through its effects on microorganisms and soil fauna [2,26,66,67], which ultimately negatively affects the nitrification rates, organic matter decomposition, mineralization of essential nutrients, carbon cycling, soil structure and nutrient availability (Table 2). Along with that, consequent reduction in earthworms in the soil due to excessive copper accumulation negatively affect the aeration and water infiltration [68]. Field studies show that higher earthworm densities are associated with greater crop yields, suggesting that copper related declines in earthworm abundance may reduce vineyard productivity [65]. Excessive copper (>200 mg/kg) can also reduce soil respiration [58] and negatively affect soil aggregation and stability [26,67].

Table 2. Effects of copper on key soil functions.

Soil Function	Change	Mechanism/Cause	Source
Nitrification	Decrease	Inhibition of nitrifying microorganisms	[10]
Mineralization (N, P)	Decrease	Reduced microbial activity	[17]
Organic matter decomposition	Slower	Inhibited enzymes and microbial biomass	[69]
Carbon cycling	Disturbed	Reduced respiration and altered C storage	[69]
Soil structure	Degradation	Reduced earthworm activity and aggregation	[68]
Nutrient availability	Decrease	Changes in microbial turnover and sorption	[70]

Soil enzymes are essential components of soil biochemical functioning, particularly in the decomposition of organic matter and the cycling of C, N, P and S [71–73]. Heavy metal contamination, including copper, can inhibit key enzymes involved in these processes [26,74,75]. The extent of inhibition depends on several soil properties, especially pH: lower pH increases copper bioavailability and enhances its negative effects, whereas higher pH reduces toxicity and limits enzymatic inhibition [75]. Copper can reduce enzyme activity through multiple mechanisms: toxicity to microorganisms that produce enzymes, direct binding to enzyme active sites, and formation of enzyme–substrate complexes that reduce catalytic efficiency [76].

Enzymes associated with carbon and sulphur cycling tend to be more sensitive to copper than those linked to nitrogen and phosphorus cycling, and endoenzymes are generally more affected than exoenzymes [12,77]. Differences in soil pH, organic matter content, soil temperature and clay content further influence the degree of enzymatic inhibition [76,77]. Organic amendments (cattle and chicken manure) can mitigate copper toxicity at lower contamination levels by improving soil structure and binding copper, although this effect diminishes as copper concentrations increase [76]. Copper formulation itself appears to have little influence on enzymatic responses, as different copper compounds show broadly similar effects [26]. Because some enzymes, particularly dehydrogenase and urease, consistently show strong inhibition under copper stress, they are considered reliable indicators of copper contamination and useful tools for assessing soil biochemical health [75].

Table 3 synthesizes reports effects of copper on soil enzyme activity across different studies, which indicates that monitoring soil enzyme activity can serve as an effective screening tool for assessing soil contamination; for instance, a reduction in dehydrogenase (DHA) activity, an intracellular enzyme, reflects the inhibitory effects of Cu on soil microbial communities [26].

Table 3. Effects of copper on soil enzyme activity.

Enzyme	Sensitivity Threshold	Direction of Effect	Notes	Source
Dehydrogenase (DHA)	200–250 mg/kg	Strong inhibition	Highly sensitive; good bioindicator	[77]
β-glucosidase	200–250 mg/kg	Inhibition	Linked to C cycling	[77]
Phosphatase (acid/alkaline)	150–200 mg/kg	Inhibition	First enzyme affected	[77]
Urease (UA)	≥100 mg/kg	Variable	Sometimes inconsistent response	[76]
Arylsulfatase (ARY)	No single threshold	Strong inhibition	Sensitive; linked to S cycling	[75]
Catalase (CAT)	Elevated Cu	Inhibition	Less sensitive than DHA	[75]
Acid phosphatase (Pacid)	100 mg/kg	Stimulation	Observed in some soils	[76]
Enzymes of C & S cycling	Lower thresholds	Strong inhibition	More sensitive than N & P enzymes	[12]

5. Copper Toxicity in Grapevines and Transfer through the Food Chain

Grapevines take up copper from the soil through their roots [78] and like most plant species, depends on copper as a constituent of several enzymes which play important roles in physiological processes of grapevine (e.g., photosynthesis, respiration, water permeability, reproduction and disease resistance) [79,80]. But excessive amounts of copper are toxic to grapevine, especially young and cultivated varieties [11,81–83]. For example, copper exposure reduces plant growth, leading to shorter plants, smaller leaves, a reduced root system, and decreased total shoot biomass in young grapevines [62]. Young grapevines whose roots are shallow in the soil are

often affected by excessive copper [79,84,85], showing leaf chlorosis, copper accumulation in the roots [11], changed root morphology [86] and anatomy [87], including darkening of the roots, reduced root density, modifications in cell walls, and alterations in the arrangement of root apex tissue, resulting in thickening of the roots [87]. In addition, one pot experiment on young grapevines determined changes in root morphology at a dose of 40 mg Cu/kg, but at the same dose in treatments where grapevines are grown in intercropping with native grass, root length and surface area were not depressed. However, a dose of 80 mg Cu/kg affected root morphology regardless of the presence of native grass [85]. Also, grapevine seedlings are sensitive to elevated copper levels, and if they are treated with CuSO₄ solution (10 mM/L) they induce the overproduction of reactive oxygen species (ROS), leading to oxidative damage in cells, reduced chlorophyll content and disrupts normal metabolic pathways, especially those related to energy production, nitrogen metabolism, and stress-response signalling [88]. On the other hand, wild grapevine varieties tend to be more tolerant to elevated copper levels in the soil. Authors from Spain determined that exposure to elevated copper levels reduced biomass in *Vitis vinifera* ssp. *sylvestris* by approximately 35%, primarily due to a decline in net photosynthesis linked to disruptions in the photosynthetic electron transport chain. Despite this reduction, the subspecies showed high tolerance, with growth remaining unaffected at foliar copper concentrations up to 35 mg/kg and survival maintained even at much higher copper levels, confirming that *V. vinifera* ssp. *sylvestris* is more copper tolerant than cultivated grapevine varieties [89]. This trend is further supported by other authors, who compared the effects of excessive copper on the grapevine rootstock “41B” and two wild grapevine populations. While all plants exhibited nutrient imbalances and reduced photosynthetic performance under high copper exposure (23 mM), the wild grapevines showed significantly higher tolerance [81]. Furthermore, research shows that the timing of foliar copper application in vineyards plays a critical role in determining potential negative effects, while elevated foliar doses themselves usually have only a limited impact on grapevine health. Authors from Romania concluded that copper applications enhanced several physiological processes in grapevine, as higher doses of copper improved must sugar concentration, increase photosynthetic efficiency, and promote greater carbohydrate accumulation in canes, particularly when copper was applied from the lag phase. Conversely, late or very early applications were less effective, while the highest copper dose (2.42 kg/ha) consistently enhanced wood maturation and bud viability, demonstrating that both the amount and timing of copper treatments are critical for optimizing vine performance [90]. Additionally, many authors report that, despite the high concentrations of copper in the vineyard soil, adult grapevines usually do not exhibit symptoms of the toxic effects of copper [1,11,16,84], which confirms that old grapevines are one of the most copper tolerant crop species [91]. All because most of the copper is accumulated in the soil surface layer, while grapevines are deeply rooted, where copper concentrations are lower and therefore less copper is available for absorption [1,10].

Grapevines in general accumulate most of copper in the root and so prevent and/or reduce excess copper translocation to aerial parts [79] (Table 4). Following root uptake, copper must be efficiently redistributed within the plant, a process mediated by specific copper transporters. Recent studies have provided detailed insights into one such transporter, VvCTR1, which is predominantly expressed in the root system, consistent with the role of roots as the primary site of copper uptake from the soil. Its localization to the trans-Golgi network, the pre-vacuolar compartment and the tonoplast indicates that this transporter participates in intracellular copper trafficking, including vacuolar sequestration and mobilization. Functional complementation assays confirm that VvCTR1 mediates copper transport across endomembrane systems, highlighting its role in maintaining copper balance under both deficiency and excess conditions [92]. Copper accumulation in roots is considered a tolerance mechanism that prevents or reduces the excess translocation of copper to shoots, where it could cause greater damage to the plant’s important physiological processes [85]. The application of copper-based fungicides can lead to accumulation in grapes and by-products, such as must and wine [29,30,93], but transport of copper from the roots seems to play only a minor role in determining its levels in the above-ground and edible parts of the plant [30,93].

Table 4. Grapevine organs and their copper concentrations.

Grapevine Organs	Copper Concentrations	Note	Source
Roots	peak values 11 to 164 mg/kg	Primary site of accumulation; mechanism of tolerance.	[94]
Canes	low-moderate values 4 to 21 mg/kg	It depends on the rootstock and on the age of the grapevine	[94]
Leaves	low values 7.1–10.5 mg/kg; although higher concentrations may occur in young, stress-affected plants.	Translocation is limited; symptoms are visible in young plants.	[11]
Shoots	low-moderate levels, 8.0–11.7 mg/kg;	It depends on the rootstock and the phenological stage.	[11]
Bunches (berries)	low levels approximately 7.0 mg/kg	Most of the copper on the surface originates from fungicides, not from the roots. Translocation from the roots is minimal.	[11]

Other factors that may influence copper concentration in grapes and their by-products include the amount of copper applied, the number of applications, the amount of precipitation during the period between the last application and harvest, and the number of days between the last application and harvest [30,95,96]. Furthermore, the use of copper-based winemaking equipment and copper-containing processing aids, such as copper sulphate or copper citrate, in winemaking to remove H₂S can also increase copper levels in grapes, must, and wine [93,96]. Red wine generally contains more copper than white wine, which can be attributed to the fact that during the initial winemaking process for red wine, the grape skins are not removed [30,96–98]. However, there are also exceptions, where white wines contain significantly more copper than red wines [97], or where there is no significant difference in copper content between the two types of wine [30]. The most frequently cited maximum residue limits (MRLs) for Cu in grapes and wine, at 20 mg/kg and 1 mg/L, respectively, in the scientific literature, are those established by the International Organization of Vine and Wine [30,96,98]. Copper in wines is usually not high and is below the MRL due to its elimination in the lees during the fermentation process in the form of insoluble salts [30,42]. However, MRL limits are sometimes exceeded in both grapes and wines [30,99,100]. For example, from 2014 to 2017, Chinese customs detained numerous batches of wines imported from Argentina, Spain, Chile, Cyprus, Ukraine, France, and other countries due to excessive Cu content [98].

6. Copper Toxicity in Humans

Copper is an essential trace metal (micronutrient) for humans with health effects described by a “U-shaped” dose-response curve, where both insufficient and excessive intakes may produce adverse health consequences [95,101–103]. The metal participates in multiple biochemical and physiological processes within the human body. It is a cofactor for a variety of enzymes, mainly oxidases involved in antioxidant defences, immune regulation, iron homeostasis, and the synthesis of hormones, neuropeptides, and neurotransmitters [102–106]. The essential nature of copper is evidenced by its involvement in numerous physiological processes, including fetal and infant growth, neurodevelopment, bone integrity, glucose and cholesterol metabolism, pigment synthesis, and immune function [105]. Disruption of copper homeostasis in the human body can result in either deficiency or overload, both of which are associated with toxic effects [103,107]. For example, Wilson’s disease is characterized by copper accumulation, exemplify the pathological outcomes of impaired copper regulation caused by mutations in Cu-ATPase transport genes [102,103,106,107]. The human body cannot synthesize copper, so it must be obtained through diet [97]. However, high levels of copper exposure (acute or chronic) to humans can result in toxicity. Acute copper poisoning is characterized by symptoms that can include nausea, vomiting, gastrointestinal disturbances, abdominal and muscle pain, hemolysis, and tissue damage in the intestines, kidneys, and liver [27,97,102,103]. Fatal cases of acute copper poisoning, primarily reported in developing countries, are typically linked to intentional ingestion of substantial quantities of copper compounds [106]. Exposure to high levels of copper can cause brain and kidney damage, liver cirrhosis, intestinal irritation, immunotoxicity, and may cause nervous breakdown [27,105,108]. There is evidence that excessive copper contributes to the pathogenesis of Alzheimer’s disease (AD), Parkinson’s disease (PD), amyotrophic lateral sclerosis (ALS), and multiple sclerosis (MS) [109]. In the above-mentioned neurodegenerative disorders, including Huntington’s disease (HD) and amyotrophic lateral sclerosis (ALS), copper homeostasis is disrupted [107,109,110]. The total copper content in an adult human body generally ranges from 50 to 120 mg and is predominantly localized in the brain, liver, kidneys, and bones [107]. In the general population, copper exposure occurs predominantly through the diet [100,106], whereas specific groups such as agricultural and industrial workers may additionally encounter Cu via inhalation or dermal contact [102].

Most current national regulations do not define thresholds for copper content in food. Therefore, the health risk to humans can be assessed by calculating the hazard quotient (HQ), which is equal to the ratio of the average daily dose (ADD) to the reference dose (EFD (0.04 mg/kg/day) [2,111,112]. If the HQ value >1, there is a possible health risk [2]. It should also be highlighted that EFSA revised the acceptable daily intake (ADI) in 2023 from 0.15 to 0.07 mg Cu/kg bw/day, reflecting growing concern over copper-induced liver toxicity [102].

Data from studies conducted in Italy [2], Iran [113,114], the Czech Republic [115], and Pakistan [116] suggest that consuming grapes or wine poses no potential health risk for consumers due to copper content. Similar to previously reported findings, a nationwide survey in China showed that copper concentrations were higher in grape skins ($5.02 \pm 3.18 \mu\text{g/g}$) than in pulps ($3.74 \pm 1.48 \mu\text{g/g}$) and based on the estimated daily intake the study concluded that consumption of grapes does not pose a health risk to consumers, even in areas with elevated copper levels [95]. Nevertheless, a degree of caution is always advisable, because relatively elevated concentrations of potentially hazardous metal ions, including copper have been reported in both red and white wines from various countries. When consumed daily at a volume of 250 mL, such wines can yield very high HQ values, indicating that long-term intake may pose health concerns [117]. Reported HQ values typically ranged from 50 to 200, with

some Hungarian and Slovakian wines reaching up to 300. Among the analysed elements, vanadium, copper and manganese exerted the strongest influence on those HQ values, indicating that copper is consistently among the key contributors to potential non-carcinogenic risk associated with wine consumption [117].

7. Sustainable Viticulture and Remediation Strategies

Grapevine (*Vitis* spp.) is one of the most extensively cultivated fruit crops worldwide [118], with recent estimates indicating 7.07 million hectares and 78 million tons of global grape production in 2024 [119], and European vineyards accounting for more than 60% of the total cultivated area [120]. At the same time, viticulture relies on the long-term application of inorganic pesticides, leading to the accumulation of these contaminants in soils and other environmental compartments, with potential implications for ecosystem integrity and human health. Copper is the most frequently occurring pollutant in vineyard soils, as its concentrations very often exceed European legal limits [64]. Under current regulations, it is evident that the EU has identified a problem with copper contamination of agricultural soils. However, to systematically address the problem and prevent potential negative consequences for the environment and human health, it is necessary to invest additional effort in reducing copper inputs into vineyard soils and in green remediation of already contaminated vineyard soils.

Efforts to reduce copper use in viticulture have been ongoing for decades. However, many proposed alternatives in organic vineyards have shown limited success under real field conditions [121,122], particularly in years with high disease pressure. Measures such as reduced application rates, improved forecasting systems, precision spraying, adoption of resistant cultivars, and the use of cover crops can contribute to lowering copper inputs [123–125], but their overall impact remains modest. For example, in organic vineyards studies show that copper-peptidate formulations can reduce the required copper dose while maintaining disease control. However, field trials in both northern and southern Italy demonstrated that these formulations also caused substantial phytotoxicity under humid conditions, indicating the persistent difficulty of identifying safe and effective copper-reduction strategies in organic systems [122]. Similarly, results from the VITIFIT project in Germany show that reducing copper inputs in organic vineyards remains highly challenging under real field conditions. Even when copper doses were lowered from 3 to 2 kg Cu/ha, satisfactory control of *Plasmopara viticola* was only achievable in years with low infection pressure, while difficult seasons such as 2021 required the full permitted dose to avoid severe crop losses. Although microencapsulated copper formulations (CuCaps) demonstrated promising efficacy, the study confirms that organic viticulture still lacks reliable, scalable solutions for substantial copper reduction [121].

In contrast, in conventional viticulture, a wide range of modern synthetic fungicides with favourable ecotoxicological and toxicological profiles, such as fluxapyroxad (SDHI), pyraclostrobin (QoI), tebuconazole (DMI), and fludioxonil (phenylpyrrole), provide less harmful alternatives to copper. But findings indicate that fungicide efficacy is highly compromised by the development of resistance, particularly in *Aspergillus uvarum* populations exposed to in conventional vineyards. In contrast, *A. tubingensis* isolates remained fully sensitive to QoI and showed only low-level resistance to DMI, fludioxonil, SDHI, suggesting that these fungicides are still largely effective against this species [126]. A similar pattern is observed in *Plasmopara viticola*, where the long-term use of QoI fungicides has led to the spread of the G143A mutation in the *cytb* gene, causing a marked decline in QoI efficacy across European vineyards. Resistant isolates reached frequencies of up to 23% and showed no detectable fitness cost, remaining fully competitive even in the absence of fungicide pressure. These findings indicate that resistance to key single-site fungicides can become stable and persistent in conventional vineyards, underscoring the need for diversified resistance-management strategies despite their favourable toxicological profiles [8]. A comparable trend is recorded in Brazil, in *Plasmopara viticola*, where intensive QoI and carboxylic acid amide (CAA) fungicide use has selected for the G143A and G1105S mutations, resulting in widespread resistance across multiple regions. Populations carrying these mutations were completely insensitive to azoxystrobin and mandipropamid, with G143A frequencies often exceeding 90% and frequently occurring together with CAA resistance. The emergence of dual resistance shows how quickly even highly effective single-site fungicides can lose efficacy under strong selection pressure [9].

Once copper has accumulated in vineyard soils, remediation becomes technically challenging, costly, and often only partially effective [127]. Conventional remediation methods, such as immobilisation, pH modification, organic matter manipulation, or soil mixing, tend to provide temporary or superficial improvements and may even compromise soil structure [16,91]. Phytoremediation, particularly phytoextraction, is considered more suitable for soils with low to moderate copper contamination and is compatible with organic production systems [16,85,128]. Species such as *Brassica juncea*, *Plantago lanceolata*, and several cover crop legumes and grasses have shown higher copper accumulation capacities [129,130], although their practical implementation requires careful management to avoid competition with grapevines or the promotion of pests and diseases. Enhancing copper

bioavailability through chelating agents or copper mobilising microorganisms can improve phytoextraction efficiency, and harvested biomass can be processed through drying, ashing, composting, or emerging phytomining and ecocatalysis techniques to recover copper and increase economic feasibility [55,131].

8. Conclusions

Copper contamination in vineyard soils remains a persistent environmental and agronomic challenge resulting from more than a century of copper-based fungicide use. Although copper continues to play a crucial role in disease control, especially in organic viticulture where alternatives are limited, its accumulation in topsoil affects soil biota, ecosystem functions, and, under certain conditions, vine health. At the same time, copper levels in grapes and wine generally remain below regulatory limits, and current dietary exposure assessments indicate low risk for consumers, highlighting the need to balance environmental concerns with realistic food-safety evidence. Since copper residues in soils cannot be resolved through regulatory restrictions alone, sustainable viticulture requires a shift toward integrated strategies that reduce copper inputs, improve forecasting and application efficiency, and promote resistant cultivars and soil-protective practices. Among available remediation options, phytomanagement and phytoremediation offer promising, vineyard-compatible solutions for low to moderately contaminated soils, particularly when combined with cover crops that enhance soil function and copper stabilization or extraction. Ensuring long-term sustainability will depend on refining bioavailability-based soil thresholds, improving risk assessment frameworks, and optimizing preventive and phytomanagement approaches under changing climatic conditions.

9. Future Perspective

Despite decades of research, the legacy of copper fungicide use in vineyards remains a complex challenge that requires an integrated approach. Future research should focus on a more precise understanding of the long-term fate of copper in soil, including processes of binding to organic matter, transformation into different chemical fractions, and interactions with the microbiome. It is particularly important to develop standardized protocols for assessing copper bioavailability, as existing methods often conduce to contradictory results and make comparisons between studies difficult.

Given the growing concern for human health, future studies should focus on quantifying the transfer of copper through the food chain, including risk assessment for consumers and vineyard workers. The development of improved exposure models, which integrate data on environmental concentrations, dietary habits and toxicokinetics, will be crucial for making informed regulatory decisions.

In the field of remediation, sustainable strategies (e.g., phytoremediation, bioremediation, biochar application, and improved organic amendments) are expected to play an increasingly important role. However, their effectiveness in real viticultural conditions is still not sufficiently investigated. Future work should focus on long-term field trials, economic analyses, and assessment of the impact on grape and wine quality.

In the long term, the transition to integrated pest management systems and reduced reliance on copper will be essential, while in organic production the development of alternative, more environmentally friendly fungicides will be particularly important for ensuring sustainable viticulture. A combination of technological innovation, improved understanding of ecological processes and stricter regulatory frameworks could enable a balance to be struck between productivity and environmental protection. This would mitigate the legacy of historical copper use and ensure the sustainable development of viticultural ecosystems.

Author Contributions

M.P.: Conceptualization, Data Collection, Writing—original draft, Visualization; I.K.: Validation; M.Z.: Data Collection; M.M.: Validation; A.P.: Conceptualization, Visualization, Supervision, Writing—review & editing. All authors have read and agreed to the published version of the manuscript.

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The authors declare no conflict of interest.

Use of AI and AI-Assisted Technologies

During the preparation of this work, the authors used the Grammarly app to check spelling, grammar, punctuation, clarity, and writing engagement. After using this tool, the authors reviewed and edited the content as needed and take full responsibility for the content of the published article.

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