

Pesticidal Copper (I) Oxide: Environmental Fate and Aquatic Toxicity

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Contents

1	Introduction	1
1.1	Molecular Structure	2
1.2	Physical and Chemical Properties	3
1.3	Use in California	3
2	Environmental Fate	4
2.1	Copper Speciation in Surface Waters	4
2.2	Copper Speciation in Sediment	9
2.3	Copper Speciation in Soil	11
2.4	Copper Environmental Fate in Air	12
3	Effects on Aquatic Organisms	13
4	Nanocopper: Emerging Ecotoxicity Data	18
5	Monitoring and Ambient Water Quality Standards	19
6	Summary	21
	References	22

1 Introduction

Copper oxide is used in agriculture as a fungicide to protect coffee, cocoa, tea, banana, citrus, and other plants from major fungal leaf and fruit diseases such as blight, downy mildew, and rust (HSDB 2008). Copper oxide is used as an active ingredient in various pesticidal formulations. After the ban of tributyltin (TBT), in the late 1980s, the use of copper oxide in antifouling paint products increased. These products protect boat and ship hulls against biofouling by marine organisms. There are currently 209 pesticide products registered in California that use copper

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oxide as an active ingredient (CDPR 2009a). Examples of registered pesticide products include the following: 3M Copper Granules, Americoat 275E Antifouling Red, Copper Shield 45, Nordox, Super KL K90 Red, Ultra 3559 Green, and others.

Although copper is an effective biocide, it may also affect non-target organisms and pose environmental concerns. Copper may be washed into the aquatic environment from agricultural and urban application sites and may enter water when used as a biocide in antifouling paint formulations. The latter use may constitute a major copper pollution contributor to California marinas, because antifouling paints continually leach from and are regularly scrubbed off boat hulls, thus releasing copper-containing paint residues into the surrounding water and sediment. The resulting copper concentrations may potentially be high enough to threaten aquatic organisms.

Copper (Cu) is a naturally occurring element. Its average abundance in the earth's crust is about 50 parts per million (ppm) (U.S. DHHS 2004). Copper is a transitional metal and occurs in nature in four oxidation states: elemental copper Cu (0) (solid metal), Cu (I) cuprous ion, Cu (II) cupric ion, and rarely Cu (III) (Georgopoulos et al. 2001).

Copper is also a trace element that is needed for proper functioning of many enzymes in biological systems. At least 21 copper-containing enzymes are known, all of which function as redox catalysts (e.g., cytochrome oxidase, monoamine oxidase) or dioxygen carriers (e.g., hemocyanin) (Weser et al. 1979). Excess copper concentrations, on the other hand, retard organisms' vital processes by inactivating enzymes and by precipitating cytoplasmic proteins into metallic proteinates (Long 2006). Exposure to copper-containing compounds precedes the modern era; such compounds have been used as pesticides for centuries and are still being used today in various insecticide, fungicide, herbicide, algacide, and molluscicide formulations.

In this chapter, we review the environmental fate and effects of copper oxide, with special attention provided to surface waters: freshwater, saltwater, and brackish water. Since copper is a natural element, its speciation, environmental fate, and toxicity are complex and differ from that of organic pesticides. In water, Cu (II) (or Cu^{2+}) is the most prevalent form of copper (Georgopoulos et al. 2001). Therefore, in this review we will primarily focus on this ionic species. Additionally, because of rapid advances in nanotechnology and potential developments of nanopesticides, we will also address the current state of knowledge on the environmental fate and toxicity of nanocopper.

1.1 Molecular Structure

Copper (I) oxide is a mineral that has cubic structure. In the lattice structure, copper has two neighboring oxygen atoms, and oxygen has four neighboring copper atoms (Web Elements 2009). Copper (I) oxide is an IUPAC name; however, in this chapter synonymous names like copper oxide and cuprous oxide will be used interchangeably.

1.2 Physical and Chemical Properties

Copper oxide dissolves in strong acids, ammonium hydroxide, and aqueous ammonia and its salts (Goh 1987). Copper oxide is insoluble in water, organic solvents, and dilute acid unless an oxidizing agent is present (U.S. DHHS 2004) (Table 1).

1.3 Use in California

The majority of copper oxide is used in agriculture on nuts, citrus, apples, lettuce, olives, berries, spices, and other commodities (Fig. 1) (CDPR 2009b). The pounds of copper oxide used in 2007 are similar to those used in 1997, and amounts used have generally decreased since 2005. The amounts of copper (I) oxide used between 1993 and 1995 were unavailable and therefore represent data gaps in the chart depicted in Fig. 1.

Copper oxide is the most popular biocide used in antifouling paints today, appearing in about 90% of products registered in California (Singhasemanon et al. 2009). Antifouling uses of copper oxide include commercial and non-commercial applications for boat and ship hulls and miscellaneous applications such as underwater structures and piers. Efforts to estimate the total use in pounds of copper oxide for these purposes are challenging, because the use of copper as an antifouling agent is not required to be reported.

Table 1 Physical and chemical properties of copper (I) oxide

IUPAC name	Copper (I) oxide
Synonyms	Copper oxide; cuprous oxide; dicopper oxide
CAS number	1317-39-1
Molecular formula	Cu ₂ O
Molecular weight (g/mol)	143.09
Appearance	Yellow, red, or brown crystalline powder
Odor	None
Boiling point STP ^a (°C)	1800
Melting point STP (°C)	1235
Density (g/cm ³)	6
Refractive index	2.705
Vapor pressure	Negligible
Solubility	Soluble in dilute mineral acid to form copper (I) salt or copper (II) salt plus metallic copper; aqueous ammonia and its salts
Stability	Insoluble in water and organic solvents
Kow	Stable in dry air; in moist air oxidizes to cupric oxide
	Not applicable

Sources: CDPR (1991); Goh (1987); HSDB (2008); ILO (2008) (ICSC: 0421)

^aSTP: standard temperature and pressure

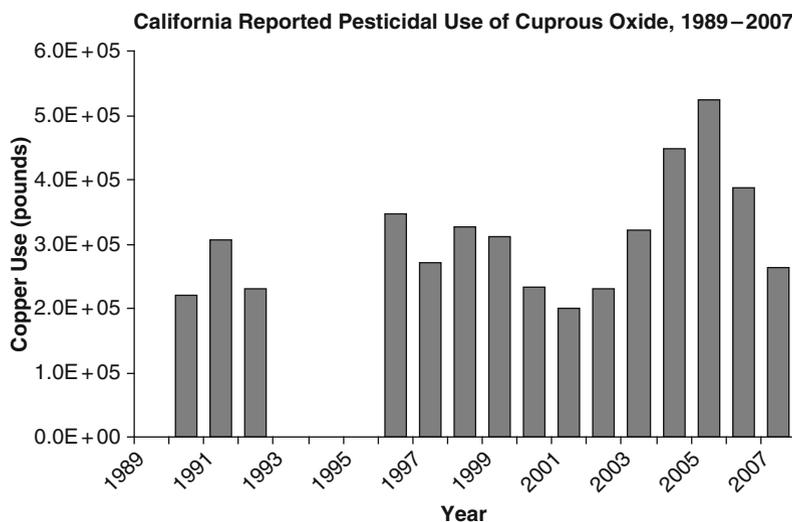


Fig. 1 Cuprous oxide total annual use in California between 1989 and 2007. *Source:* CDPR (2009b)

2 Environmental Fate

2.1 Copper Speciation in Surface Waters

In the water column, copper displays a complex biogeochemical and speciation cycle (Fig. 2). Important factors that determine copper's environmental fate relate to interactions between the metal and the physical/chemical properties of the water column. Seawater, water of increasing salinity in estuaries, and freshwater in rivers and lakes may have different copper speciation outcomes. These outcomes influence metal bioavailability and, thus, the toxicity to the aquatic organisms.

Copper oxide dissociates in water, and the most prevalent copper oxidation state is Cu^{2+} (cupric ion) (Georgopoulos et al. 2001). Cu^{2+} is also a form primarily responsible for coppers' biocidal effects. Thus, the following discussion will largely refer to water solubilized copper.

Copper can exist adsorbed to dissolved molecules or to particulate matter and is referred to collectively as the total copper (TCu) pool (Fig. 3). Even though copper adsorbs to particulate matter, it interacts most strongly with dissolved components in the water (Muller 1996). Hence, in speciation studies, total dissolved copper (TDCu) concentrations, sometimes referred to as dissolved copper (DCu), is the entity that is conventionally measured. TDCu is functionally determined by the filter pore size. Copper passing through a $0.45 \mu\text{m}$ or smaller filter pore size is considered to be dissolved.

TDCu can be further separated into labile copper (LCu) and organically complexed fractions. In this chapter, the term LCu means bioavailable copper

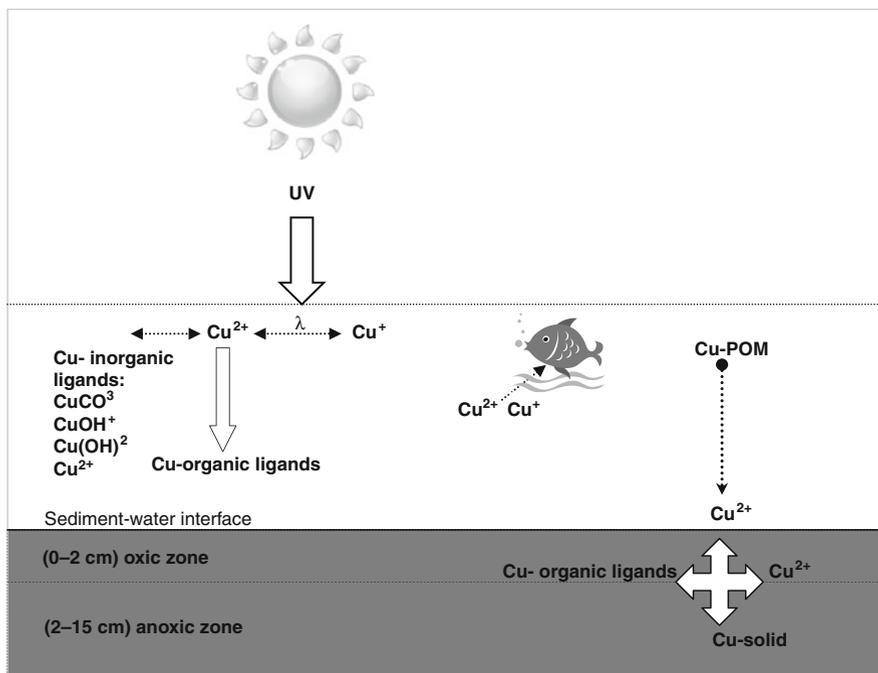
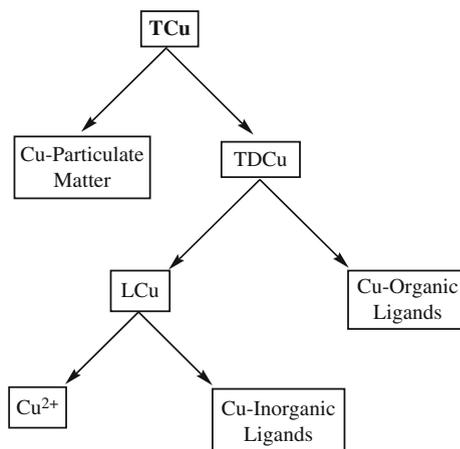


Fig. 2 Aquatic fate diagram for copper oxide (POM is particulate organic matter)

Fig. 3 Copper speciation pathways in surface water. TCu represents total copper, TDCu is total dissolved copper, and LCu is labile copper



and includes both free hydrated copper ions (Cu^{2+}) and inorganically complexed species. Organically complexed copper, on the other hand, is considered inert or non-bioavailable to biological organisms.

Organically complexed copper is bound to organic ligands. “Organic ligands” is a generic term describing heterogeneous molecules that are ubiquitous in water.

Their binding may or may not be metal specific. In some studies, organic ligands are interchangeably referred to as dissolved organic carbon (DOC) or dissolved organic matter (DOM). The sources of organic ligands are both natural and anthropogenic. Natural sources include humic and fulvic substances, as well as microorganism-produced copper-binding ligands (Buck et al. 2007; Shank et al. 2004). Anthropogenic sources include urban, industrial, and agricultural discharges and runoffs that carry organic molecules such as ethylenediaminetetraacetic acid (EDTA) and nitrilotriacetic acid (NTA) (Buck et al. 2007).

Organic ligands are characterized as weak, strong, and, sometimes, intermediate strength. The strength of ligands is experimentally determined by measuring their conditional stability constants, which reflect copper-ligand binding affinity. For example, conditional stability constants for weak ligands are reported to be $\sim 10^9$ and for strong ligands $\sim 10^{13}$.

2.1.1 Saltwater

In early copper speciation studies, results were often in conflict, because different analysis techniques and detection capabilities of analytical instrumentation were used. With advanced instrumentation, a more uniform picture emerged, showing that the majority of TDCu in seawater is organically complexed. Most authors of copper speciation studies have concluded that about 89–99% of TDCu is bound to organic ligands (Buckley and Van Den Berg 1986; Hirose et al. 1982; Lucia et al. 1994; Suda and Hanson 1987). Therefore, only a small fraction of TDCu constitutes LCu that is inorganically bound or exists as free Cu^{2+} ions. Among different studies, the incidence of LCu ranges between 0.03 and 6% of TDCu (Hirose et al. 1982; Suda and Hanson 1987; Van Den Berg 1984). Inorganically bound copper forms complexes primarily with carbonate (60% CuCO_3), hydroxide (32% CuOH^+ and $\text{Cu}(\text{OH})_2$), and 4% free Cu^{2+} ion (Van Den Berg 1984).

Copper speciation in seawater is affected by factors such as location, depth, and the state of equilibrium. TDCu concentrations were found to be between 0.92 and 3.2 nM (0.06 and 0.2 ppb) in the Sargasso and North seas and 16–39 nM (1.0–2.5 ppb) in the Irish Sea (Van Den Berg 1984; Van Den Berg and Donat 1992). Generally, TDCu concentrations in seawater have been observed to be much lower than ligand concentrations. Van Den Berg (1984) found TDCu concentrations of 16–39 nM and ligand concentrations of 58–156 nM.

Water depth plays a role in determining metal and ligand concentrations. In one organic speciation profile study (Buckley and Van Den Berg 1986), it was observed that an inverse relationship existed between free Cu^{2+} and ligand concentrations that was depth dependent. At the surface, free Cu^{2+} concentrations were low (2×10^{-13} M) and ligand concentrations were high (maximum 1800 nM). With increased depth, ligand concentrations dropped to 6–20 nM and free Cu^{2+} concentration increased to 7×10^{-13} M.

In the majority of copper speciation studies, it is assumed that free copper in the water is in a state of equilibrium. The state of equilibrium predicts that free copper in water exists primarily in the Cu^{2+} oxidation state. Moffett and Zilka (1983),

however, challenged this assumption. According to these authors, biochemical, photochemical, or thermodynamic processes create a non-equilibrium environment, in which copper redox chemistry may become an important part of copper speciation. For example, in the photic zone of the ocean, sunlight-generated free radicals like superoxide and hydrogen peroxide can reduce Cu^{2+} to Cu^+ . Thus, the thermodynamic model used by the authors predicted that 20–50% of total copper would be Cu^+ . Other factors that affect reduction to Cu^+ are pH, dissolved oxygen concentration, ligands, and reducing species (Moffett and Zilka 1983).

2.1.2 Brackish Water

The salinity of brackish water is less than that of seawater. Brackish water most commonly occurs in estuaries, where fresh river water meets seawater. Complex copper speciation takes place in this mixing zone.

Upon reaching water of increasing salinity, copper is largely sequestered by forming complexes with organic ligands (Apte et al. 1990; Buck and Bruland 2005; Hurst and Bruland 2005; Kozelka and Bruland 1998; Muller 1996). Apte et al. (1990) found TDCu concentrations to be higher at the freshwater end. The TDCu concentrations displayed a linear decrease with an increase in water salinity, changing from 76.4 nM in the freshwater samples to 6.8 nM in saline samples.

Seasonal and temperature fluctuations also affect TDCu and LCu levels. Beck and Sanudo-Wilhelmy (2007) studied seasonal TDCu cycling in the Long Island Sound, New York. They observed that TDCu levels did not vary greatly between spring and summer. Surface LCu concentrations, on the other hand, showed seasonal cycling to be higher in the summer. In another study, Jones and Bolam (2007) observed increased TDCu levels from winter to late summer and decreased levels during the autumn and winter, in UK marinas. Unlike Beck and Sanudo-Wilhelmy (2007), this group observed that the LCu fraction remained fairly constant throughout the year.

Although Jones and Bolam (2007) indicated that the natural environment has sufficient buffering capacity to keep LCu concentrations low, Beck and Sanudo-Wilhelmy (2007) linked the LCu fluctuation to water temperature and dissolved oxygen levels. At water temperatures above 21°C, LCu concentrations in the bottom waters increased exponentially. This indicates that copper remobilization was occurring and could explain the increased surface LCu levels during the summer. High levels of LCu were also observed under low oxygen conditions. The authors believe that there is a potential for copper remobilization if water temperatures rise and dissolved oxygen concentrations decrease as a result of global warming.

As occurred in seawater, the conclusion reached from the majority of the brackish water copper speciation studies was that 97–99.99% of TDCu in estuaries is organically complexed to ligands (Apte et al. 1990; Buck and Bruland 2005; Hurst and Bruland 2005; Kozelka and Bruland 1998; Muller 1996). The conditional stability constants of the ligands in brackish water are comparable to those of seawater. Organic ligand concentrations generally exceed TDCu concentrations, as well (Buck and Bruland 2005; Hurst and Bruland 2005; Kozelka and Bruland

1998). For example, TDCu levels in San Francisco Bay ranged between 17.9 and 49.6 nM and strong ligand concentrations were between 22 and 265 nM (Buck and Bruland 2005). As a result, free Cu^{2+} concentrations were low.

Since organic ligand levels consistently exceed TDCu concentrations, only a small percentage of copper constitutes the LCu fraction. Hence, there is a risk of overestimating the levels of copper that are available to cause toxicity. Buck and Bruland (2005) derived a saturation curve-shaped relationship between TDCu and Cu^{2+} . They estimated that for free copper to reach toxic levels of 10^{-11} M, the TDCu must be at least 100 nM. A similar finding that TDCu measurements tend to overestimate copper toxicity was also noted by Jones and Bolam (2007). Their calculated LCu/TDCu ratio predicted that TDCu overestimates the toxicity risk by a factor of 4.

2.1.3 Freshwater

Among saltwater, brackish water, or freshwater types, copper speciation in the latter appears to be the least studied and generates the most debated results. Water characteristics in rivers and lakes differ in ways that depend largely on the geochemistry of their particular location. Therefore, water properties (e.g., pH, hardness, and alkalinity) affect speciation and may produce different results among studies performed at different locations. The use of different techniques and inconsistent terminology can also produce different measurements or interpretations of results.

Copper speciation in freshwater is predominantly controlled by the TDCu that binds to organic ligands (Hoffman et al. 2007). According to the authors, almost all dissolved copper (>99.99%) is bound to strong ligands in river water, which produces free Cu^{2+} concentrations that are in the picomolar range ($0.9\text{--}6.5 \times 10^{-15}$ M). Most authors agree that, in freshwater, ligand concentrations consistently exceed those of TDCu.

The conditional stability constants of the freshwater ligands differ from those found in saltwater and estuaries. Hoffman et al. (2007) reported ligand conditional stability constants above 10^{13} . Additionally, the ligands had a higher affinity for copper than for other metals. This may indicate the existence of copper-specific ligands in freshwater systems. In contrast, Wang and Chakrabarti (2008) and Pesavento et al. (2003) reported the conditional stability constants for very strong ligands to be $\sim 10^{20}$ and 10^{17} , respectively. These numbers are many folds higher than the estimated strong ligand conditional stability constants found in saltwater and estuaries ($\sim 10^{13}$). Because of such inconsistent results, more studies in freshwater are needed to determine the nature and origin of such ligands.

In freshwater, as in saltwater and brackish water, organic complexation generally dominates copper speciation and, thus, toxicity. However, in freshwater, parameters like pH, alkalinity, and hardness have significant effects on copper speciation, as well. Gundersen and Steinnes (2003) studied eight rivers in Norway and determined that pH had the most significant influence on metal speciation. At low pH levels, most of the copper was dissolved, and at high pH levels, Cu occurred predominantly in colloidal or particulate form. Consequently, in river water of pH

3.1, almost all copper was in the dissolved fraction, and at a pH range of 6.9–7.2 (pH neutral rivers), all three fractions (dissolved, colloidal, and particulate) occurred in significant amounts. From these results, it can be inferred that LCu concentrations may be higher in acidic water.

Alkalinity is related to the capacity of water to neutralize strong acids (Snoeyink and Jenkins 1980a). Alkalinity (alkalinity ions HCO_3^{3-} and CO_3^{2-}) and water hardness (Ca^{2+} and Mg^{2+} ion concentrations) are related and usually increase or decrease together. According to Snoeyink and Jenkins (1980b), in a carbonate-buffered water system with pH below 6.5, the predominant copper species is Cu^{2+} . In the pH range of 6.5–9.5 (the pH range of most waters), CuCO_3 is the predominant copper species. Hence, in most waters, copper forms copper carbonate complexes. Moreover, at pH 7 an increase in alkalinity from 50 to 250 mg/L (as CaCO_3) decreases the Cu^{2+} levels from 25 to 9% of the total copper present. These results indicate that copper is more bioavailable and more toxic in soft, less alkaline water than in hard, more alkaline water.

2.2 Copper Speciation in Sediment

Although sediments tend to accumulate heavy metals, mass-balance estimates suggest that their remobilization could be a major source of some toxic metals in the water column (Beck and Sanudo-Wilhelmy 2007). Therefore, it is important to understand copper speciation in sediment pore water. Sediment pore water fills the spaces between grains of sediment. At the sediment–water boundary, physical, chemical, and biological changes take place (Fig. 2). Processes (physical, chemical, and biological) that bring about changes in the sediment (following its deposition) are referred to as diagenesis (Berner 1980). Copper speciation in pore water is influenced by diagenetic processes and depends on factors such as oxygen levels, temperature, and sediment type. Before addressing the specifics of copper speciation in sediment, it is important to explain how copper cycles globally to end up at the bottom of the water column.

The geochemical cycling of copper in the water column is linked to the cycling of organic carbon (Widerlund 1996). In surface water, copper adsorbs to scavenging particulate organic matter (POM) and, with the downward flux, eventually settles out. The settling action results in the formation of a thin, carbon-rich layer at the sediment–water interface (Klinkhammer et al. 1982). How much TCu settles along with POM depends on the location and season. Chester et al. (1988) reported that detritus-associated Cu comprised about 60% of TCu along the Atlantic coast, but only 18% in the open ocean. These differences reflect differences in oceanic biological activity. Landing and Feely (1981) observed increased copper flux during the summer algae bloom in the Icy Bay, Gulf of Alaska. Similarly, Helland and Bakke (2002) reported higher Cu–POM fluxes near the river mouth during a spring flood in the Gloma Estuary, Norway. Because many factors interplay in the global carbon cycling, the study authors reported that 10–50% of TCu is bound to suspended POM and settle out (Chester et al. 1988; Helland and Bakke 2002).

Upon settling, diagenetic processes break the Cu–POM association and copper is liberated into the sediment pore water; herein, copper may partition back into the water column or into the solid sediment phase. The residence time for copper in the pore water is approximately 2.1–10 days (Widerlund 1996).

The availability of oxygen determines whether copper is recycled back into the water column or is removed by precipitation. Experimental results from Widerlund (1996) suggest that oxic conditions (0–2 cm depth) play a major role in copper recycling, whereas anoxic (2–15 cm depth) conditions result in copper removal (Fig. 2). Other study results have also shown that, during early diagenesis, aerobic decomposition controls copper release back to the water column (Gerriga 1990; Skrabal et al. 2000). Gerriga et al. (1990) reported two kinds of aerobic degradation: fast and slow. Fast degradation was characterized by the rapid decline in POM concentration and high oxygen consumption. During fast degradation, copper in the sediment, which was relatively strongly bound, became relatively weakly bound. Slow degradation of POM was reflected by the transformation of ammonia into nitrite and nitrite into nitrate. It is through such transformations that copper, derived from degrading organic constituents of sediment, is continuously released into the sediment pore water. Copper concentrations were reported to be 10 times higher in the top 2 cm of the sediment pore water than in the overlying bottom waters (Klinkhammer et al. 1980). At the deeper levels of sediment pore water, where anoxic conditions prevail, copper is captured into the solid-phase sediment by precipitating metal sulfides (Skrabal et al. 2000; Widerlund 1996).

Temperature plays an important role in copper cycling. Seasonal temperature fluctuations can affect sediment conditions by changing biological activity and oxygen levels, thus influencing copper benthic flux. Widerlund (1996) noted that removal of dissolved copper from the pore water into the solid-phase sediment was temperature dependent. In September (core temperature 8°C), copper flux into the sediment was twice as high as in April (1°C). Hence, at higher water temperatures more copper is removed from the sediment pore water by precipitation. The authors also noted that since decomposition of the organic matter is temperature dependent, cold water promotes higher sediment accumulation and results in a more rapid burial of reactive (non-decomposed) organic matter.

In addition to oxygen availability and water temperature, sediment type and micro-flora and microfauna may play a role in the speciation of copper in sediment pore water. Goh and Chou (1997) observed that finer sediment has higher surface area onto which copper can adsorb. Skrabal et al. (2000) investigated the distribution of TDCu at two distinct locations: one with sulfidic muddy sediment, dominated by seasonal anoxia and poor biodiversity, and the other with sandy silt, dominated by extensive bioturbation and richness in benthic organisms. The authors determined that copper precipitates out as metal sulfides in anoxic, muddy sediment conditions. The study also allowed the authors to predict that, under these conditions, copper exists predominantly in the Cu⁺ oxidation state and is bound to sulfur-containing organic and inorganic ligands. In contrast, in oxic and biodiverse sandy sediment, the Cu²⁺ oxidation state dominates TDCu speciation. Under oxic conditions, the release of copper is controlled by the aerobic decomposition rate of organic matter.

When copper disassociates from POM and is liberated into the sediment pore water, it enters the TDCu phase. As occurs in the water column, TDCu speciation and bioavailability in the sediment pore water are controlled by strong and weak organic ligands (Gerriga et al. 1991; Skrabal et al. 2000). Since sediments are rich in organic matter, they provide a large pool of ligands to the pore water that is available for metal complexation. In addition, Skrabal et al. (2000) suggest that sediment pore water also supplies as much as 10–50% of copper complexing ligands to the overlying water column. Skrabal et al. (2000) found ligands to always be in large excess relative to TDCu concentrations in the sediment pore water. As a result, 87–99.9% of copper exists as organic complexes, and free inorganic copper concentrations are low.

Organic ligands in the sediment pore water, in contrast to their water column analogs, are subject to much faster biological degradation. Gerriga et al. (1991) reported differences in ligand degradation rates. Strong ligands were subject to oxidation, and their concentrations decreased faster than did weak ligand concentrations. Weak ligands were more resistant to degradation. After strong ligand concentrations became depleted, weaker ligands began to dominate TDCu speciation. This resulted in a sharp increase of free copper concentration (from 10^{-12} to 10^{-9} M) (Gerriga et al. 1991). Although sediments are a rich source of organic ligands, when TDCu speciation is dominated by weak ligands, copper may be much more bioavailable and, thus, more toxic to the aquatic organisms.

2.3 Copper Speciation in Soil

Copper oxide is used in agriculture to protect various crops from fungal diseases. Soil is also a major repository of copper. Thus, copper pollution can affect soil-dwelling organisms and plants and make its way to the food chain. Understanding copper speciation in soil is important to a better understanding of its effects on the soil ecosystem. The factors that control the environmental fate of copper in soil include the organic and inorganic content and the pH.

Copper persists in the topsoil and generally accumulates in the upper 15 cm (Rodríguez-Rubio et al. 2003). Today, elevated copper concentrations are found in the vicinity of former smelters and chemical spill areas that did occur, or may have occurred, decades previously (Kabala and Singh 2001; McBride and Bouldin 1984). The results from several studies show that copper preferentially associates with soil organic matter (Boudescque et al. 2007; Jacobson et al. 2007; Liu and Wang 2004; Strawn and Baker 2008). In the Liu and Wang (2004) speciation study, 50% of copper in the contaminated soil was associated with organic matter, 28% formed CuCO_3 , 11% Cu_2O , and 11% CuO . Boudescque et al. (2007) determined that the copper associated with soil organic matter is formed via inner sphere complexes, which occur when copper ions adsorb directly to the organic particle in the soil. Because of the strength of such complexes, organic matter plays an important role in determining the degree of mobility and bioavailability of copper (Boudescque et al. 2007). Additionally, copper distribution in the soil may not be uniform. As

reported by Jacobson et al. (2007), entire regions of vineyard soils were devoid of the metal. Localized hotspots of copper were associated with minimally degraded organic matter, which may have been the result of reduced microbial degradation in those places.

Although the majority of study authors agree that soil organic matter is a very important component in copper speciation, there are some controversies among them on calcareous soils. A few authors have concluded that the main mechanism of copper retention is its precipitation as CuCO_3 (Ponizovsky et al. 2007; Rodriguez-Rubio et al. 2003). In contrast, Strawn and Baker (2008) concluded that copper in calcareous soil was predominantly associated with soil organic matter and not with metal oxides, silicates, phosphates, or carbonates.

Soil pH plays an important role in copper retention and mobility. The concentration of uncomplexed copper increases at low pH, thus increasing its mobility (McBride and Bouldin 1984; Temminghoff et al. 1997). At pH 3.9, only 30% of copper was associated with soil organic matter. In comparison, at pH 6.6 the copper–soil organic matter association was 99% (Rodriguez-Rubio et al. 2003; Temminghoff et al. 1997). Thus, uncomplexed, free copper may be more toxic to plants, especially in higher acidity soil.

2.4 Copper Environmental Fate in Air

Copper is found as a trace element in atmospheric water (e.g., fog, clouds, and rain) as a result of its global cycling (Kieber et al. 2004). Atmospheric copper derives from both natural and anthropogenic sources. Natural sources include windblown dust, plant exudates, and sea salt spray. Anthropogenic sources include iron, steel, and non-ferrous metal manufacturing; the burning of fossil fuels; waste incinerators; and terrestrial pesticide use (CDPR 2009b; Hsiao et al. 2002; Kieber et al. 2004). Research results have indicated that the majority of the copper that is released into the atmosphere originates from continental anthropogenic sources.

The primary route by which copper is removed from the atmosphere is wet deposition (Church et al. 1984; Kieber et al. 2004). Kieber et al. (2004) estimated the amounts of Cu per year removed by rain to be 150×10^6 kg. This number also represents the total estimated copper input (continental and marine) into the global atmosphere. Few studies have been performed on the contribution of atmospheric copper to water bodies. Williams et al. (1998) estimated that about 6% of the total copper input into the Irish Sea comes from the atmospheric compartment. Giusti et al. (1993) estimated the atmospheric copper input to the oceans to be $14 - 45 \times 10^6$ kg/year for the dissolved form (e.g., rainwater) and $2 - 7 \times 10^6$ kg Cu/year as particulates.

In air, copper may exist as several chemical species. In fly ash, generated from municipal solid waste incinerators, copper was found to exist as CuCO_3 , Cu(OH)_2 , and CuO (Hsiao et al. 2002). Atmospheric copper is subject to redox reactions and tends to be hydrated. Researchers who investigated the speciation of copper in rainwater reported the volume-weighted average concentrations of TDCu to be

5.3 nM, Cu^+ 1.4 nM, and Cu^{2+} 3.2 nM (Kieber et al. 2004). About half of the TDCu was bound to strong organic ligands and the remainder was bound to weaker organic ligands, inorganic ligands, or existed as free hydrated ions.

3 Effects on Aquatic Organisms

Copper is an essential trace element needed at miniscule levels for the proper functioning of all organisms. However, excessive amounts of copper interfere with vital biological functions. Different species, and even organisms within the same species, can exhibit different sensitivities to elevated copper levels in the water column. Organisms have different mechanisms by which they cope with and process copper. Some organisms bioaccumulate and store copper, whereas others actively regulate its levels. In general, copper is actively regulated in fish, decapod crustaceans, and algae and stored in bivalves, barnacles, and aquatic insects (Brix and Deforest 2000). Therefore, to properly evaluate the copper-related effects on aquatic life, one must understand the factors that affect the biological fate of copper and the mechanisms by which it acts to produce its toxicity.

Copper undergoes complex speciation in natural waters; some species are bioavailable (free Cu^{2+} and Cu^+ ions (Fig. 2)), while others are not. Only bioavailable forms of copper are considered to be toxic to exposed organisms. The reference to “copper” and “free copper” in the following discussion refers to its bioavailable form. The bioavailability, biodistribution to various parts of the organism, and bioaccumulation of copper are dramatically influenced by water chemistry. Therefore, water pH, hardness, organic content, and salinity play important roles in copper-induced toxicity.

The majority of studies in which the toxicity of copper has been addressed were performed on freshwater species. Copper is generally more toxic to organisms in freshwater than in saltwater. One of the reasons for this difference is that freshwater lacks cations, which compete with Cu^{2+} at the biological action sites, thus reducing copper toxicity (Brooks et al. 2007). Consequently, pH and water hardness play more important roles in freshwater than in saltwater environments. Increased pH accentuates copper toxicity because of the reduced competition between copper and hydrogen ions at the cell surface (Wilde et al. 2006). Cations that are involved in water hardness (i.e., Ca^{2+} and Mg^{2+}) also compete with Cu^{2+} for biological binding sites (Boulanger and Nikolaidis 2003). Therefore, Cu^{2+} is less bioavailable in hard water than in soft water.

Although water pH and hardness protect organisms against Cu toxicity to some degree, the DOC content is among the most important factors in reducing copper toxicity to both fresh- and salt-water species. DOC forms organic complexes with copper and thereby reduces copper’s bioavailability. According to McIntyre et al. (2008), water hardness and pH are unlikely to protect fish from copper-induced sensory neurotoxicity. However, water that contains high DOC concentrations does diminish the toxic effects of copper on the peripheral olfactory nervous system in

Coho salmon (*Oncorhynchus kisutch*) (McIntyre et al. 2008). High DOC levels also significantly decrease acute copper toxicity to the freshwater flea, *Daphnia magna*, and the estuarine copepod, *Eurytemora affinis* (Hall et al. 2008; Kramer et al. 2004).

Study results show that the water salinity gradient can also significantly affect the biological fate of copper. Water salinity influences the biodistribution and bioaccumulation of copper and can affect its toxicity as well (Amiard-Triquet et al. 1991; Blanchard and Grosell 2005; Grosell et al. 2007; Hall et al. 2008). The biodistribution of copper throughout the gill, intestine, and liver of the common killifish, *Fundulus heteroclitus*, is salinity dependent (Blanchard and Grosell 2005). According to these authors, the gill and the liver are important target organs for copper accumulation at low salinities, whereas the intestine is a target organ at high salinities. In addition, the liver is a major organ involved in copper homeostasis and accumulates the highest amounts of copper. For this reason, the liver may be a potential target organ during chronic copper exposure. Water salinity influences the biodistribution and the toxicity of copper. Grosell et al. (2007) found killifishes to be most tolerant to copper exposure at intermediate salinities, and the acute toxicity was significantly higher in the lowest and highest salinity water. Increased fish sensitivity at both salinity extremes can be attributed to two factors: changes in copper speciation and changes in fish physiology in changing aquatic environments.

In general, water salinity may be more important to species that actively regulate internal osmotic pressure. The majority of invertebrates, however, are osmoconformers. Hence, to them the salinity gradient may be less important. Although in bivalves, the biological fate of copper was salinity dependent, in copepods (*Eurytemora affinis*) the toxicity of copper correlated better to DOC content than water salinity (Hall et al. 2008). In oysters, copper accumulation was inversely related to salinity (Amiard-Triquet et al. 1991). Some species can adapt to tolerate higher pollutant levels. Damiens et al. (2006) described adult oysters that lived in polluted water, wherein their larvae become less sensitive to pollution over time. Phytoplankton species have different sensitivities to copper toxicity: cyanobacteria appear to be most sensitive, coccolithophores and dinoflagellates show intermediate sensitivity, and diatoms are resistant to copper (Brand et al. 1986; Beck et al. 2002).

In many aquatic animals, copper causes toxicity by impairing osmoregulation and ion regulation in the gill (Blanchard and Grosell 2005; McIntyre et al. 2008). When bioavailable Cu^{2+} enters the cell, it is reduced to Cu^+ . This copper oxidation state has a high affinity to sulfhydryl groups that are abundant within ATPase enzymes (Katranitsas et al. 2003; Viarengo et al. 1996). The best studied copper toxicity pathways involve the inhibition of ATP-driven pumps and ion channels. Katranitsas et al. (2003) discovered that, in brine shrimp, copper inhibited Na/K ATPase and Mg^{2+} ATPase enzyme activity. Similarly, in the mussel, *Mytilus galloprovincialis*, copper interfered with Ca^{2+} homeostasis in the gill, causing disruptions in Na/K ATPase and Ca^{2+} ATPase (Viarengo et al. 1996). In an in vitro study, Corami et al. (2007) investigated lysosomal activity and found that copper acted at two different sites: the proton pump and Cl^- selective channels. Therefore, copper acts by inhibiting enzymes, ATP-driven pumps, and ion channels, resulting in cell

toxicity from disruption of cell homeostasis and leading to changes in internal pH balance, membrane potential, and osmosis.

In addition to inhibiting ATPase enzymes and disrupting ion flow, copper toxicity can be induced by generating reactive oxygen species (ROS) (Bopp et al. 2008; Viarengo et al. 1996). ROS can lead to different outcomes: genotoxicity via DNA strand break and impaired cell membrane permeability via lipid peroxidation. Both pathways compromise normal cell functions.

A less understood effect of copper is neurotoxicity to fish olfaction. There is evidence that exposure to sublethal copper levels results in the loss of chemosensory function, which affects predator-avoidance behavior (McIntyre et al. 2008). The exact mechanisms are not yet completely understood and are still under investigation. Tilton et al. (2008) revealed that copper depresses the transcription of key genes within the olfactory signal transduction pathway.

The environmentally relevant copper levels that interfere with fish chemosensory mechanisms are very low. TDCu concentrations in the range of 0–20 ppb affected sensory capacity and behavior in salmon (Sandahl et al. 2007). At higher levels, copper caused a degeneration of the sensory epithelium (Bettini et al. 2006; Hansen et al. 1999). These effects were observed within hours of exposure. Hence, fish olfaction is a vulnerable endpoint that should be considered in environmental risk assessment.

The developmental stage of fish during their exposure to elevated copper levels may be a critical factor in their sensitivity. Carreau and Pyle (2005) showed that exposure to copper during embryonic development can lead to permanently impaired chemosensory functions. In contrast, fish that are exposed to elevated copper later in life can recover their chemosensory ability after the toxicant is removed.

Copper is stored and transported inside an organism as inorganic and organic complexes. In killifishes, copper bioaccumulates in target organs primarily as copper carbonate (CuCO_3) and, to a lower extent, as copper hydroxide (CuOH^+ and $\text{Cu}(\text{OH})_2$) (Blanchard and Grosell 2005). Bivalves accumulate considerable amounts of copper that is associated with a cytosolic protein called metallothionein (Claisse and Alzieu 1993; Damiens et al. 2006). Although copper bioaccumulates and biodistributes to different organs, it is an internally regulated essential micronutrient. Therefore, according to Brix and Deforest (2000), there is an inverse relationship between metal concentrations in the water and in the organism. Hence, the bioconcentration factor (BCF) is not a suitable concept to describe the bioconcentration of copper.

Toxicity data for aquatic species for copper oxide, selected from the U.S. EPA ECOTOX database, are summarized in Table 2 (U.S. EPA 2009a). The table is divided into sections for freshwater and saltwater organisms. Data are presented for fish, invertebrates, and plants. The toxicity endpoints are also presented in the table, as is the chemical concentration that was lethal (LC_{50}) or produced an effect (EC_{50}). There is a large range in copper toxicity values for different freshwater algae.

Table 2 Copper (I) oxide toxicity to aquatic organisms

Toxicity to freshwater aquatic organisms		Toxicity to aquatic organisms		
Species name: scientific/common	Endpoint	Duration/effects	Concentration $\mu\text{g/L}$ (ppb)	Purity (%)
Fish: <i>Danio rerio</i> /Zebra danio	LC ₅₀	96 h/mortality	75	100
Invertebrates: <i>Daphnia similis</i> /Water flea <i>Biomphalaria glabrata</i> /Snail	EC ₅₀ LC ₅₀	48 h/mortality 48 h/mortality	42 179	100 100
Plants: <i>Pseudokirchneriella subcapitata</i> /Green algae	EC ₅₀	30 min, 35 min physiology/photosynthesis	90, 1900, >4500	Not reported
	EC ₅₀	96 h/physiology/ photosynthesis	1300, 1600	Not reported
	EC ₅₀	96 h/population/ Abundance	30, 60, 230	Not reported

Table 2 (continued)

Toxicity to saltwater aquatic organisms					
Species name: scientific/common	Endpoint	Duration/effects	Concentration $\mu\text{g/L}$ (ppb)	Purity (%)	
Fish:					
<i>Cyprinodon variegatus</i> /Sheepshead minnow	LC ₅₀	96 h/mortality	>173	93	
<i>Melanogrammus aeglefinus</i> /Haddock	LT ₅₀	4.5 h, 5.7 h Mortality	1800	100	
Invertebrates:					
<i>Americamysis bahia</i> /Opossum shrimp	LC ₅₀	96 h/ mortality	69.7	97	
<i>Balanus improvisus</i> /Barnacle	LC ₅₀	12 h/mortality	700	Not reported	
	LC ₅₀	24 h/mortality	500	Not reported	
	LC ₅₀	48 h/mortality	350	Not reported	
	LC ₅₀	72 h/mortality	140	Not reported	
	LC ₅₀	96 h/mortality	20	Not reported	

Source: U.S. EPA (2009a) ECOTOX database (accessed: 11/05/09)

4 Nanocopper: Emerging Ecotoxicity Data

A definition of nanotechnology, according to the U.S. EPA White Paper produced in 2007, is “research and technology development at the atomic, molecular, or macromolecular levels using a length scale of approximately 1–100 nanometers in any dimension; the creation and use of structures, devices and systems that have novel properties and functions because of their small size; and the ability to control or manipulate matter on an atomic scale” (U.S. EPA 2007). A nanometer is one billionth of a meter (10^{-9}); this is equal to the diameter of a single-strand DNA molecule. Indeed, manipulating materials at the molecular and atomic scale produces novel materials that have new physical and chemical properties that may vary from their bulk forms. Rapid growth of the nanotechnology industry and increasing mass production of engineered nanomaterials will inevitably result in environmental exposure to these types of chemicals.

Today, metal nanoparticles are among the most popular types of nanomaterials. Metal nanoparticles like CuO, ZnO, TiO₂, nanosilver, and nanogold have a wide variety of applications, including use in industry, consumer products, medicine, and pesticide products. Copper oxide nanoparticles are used as additives in inks, plastics, lubricants; as coatings for integrated circuits and batteries; and as bactericides for air and liquid filtration (Griffitt et al. 2007; Midander et al. 2009). Thus, metal nanoparticles from various sources, including a growing number of pesticide products, could make their way to the surface waters.

Unfortunately, little published information exists on the environmental fate of nanometals, including nanocopper. Metal nanoparticles, when added to the water, can aggregate, sediment out of the water column, adsorb to nutrients, and disassociate to release soluble metal ions (Griffitt et al. 2009; Kahru et al. 2008). Gao et al. (2009) indicated that both water chemistry and the reactivity of the nanoparticle itself should be considered in environmental speciation studies. Hence, laboratory experiments that use deionized water and artificial methods to suspend nanoparticles may not realistically reflect what occurs in natural environments.

The effects of nanocopper on aquatic organisms have not been well studied. Existing studies indicate that copper toxicity strongly depends on particle size. As particle size decreases, toxicity increases. Among the studies that have been performed, there is a 15- to 65-fold increase in toxicity when nano-sized copper particles are used (Table 3). In most studies, the increase in nanocopper toxicity is attributed to an increase in solubility and, consequently, bioavailability (Aruoja et al. 2009; Heinlaan et al. 2008; Mortimer et al. 2010).

However, increased solubility does not always explain increased nanocopper toxicity. Copper nanoparticles can induce toxicity by mechanisms that are different from those of soluble ions (Griffitt et al. 2007, 2008, 2009). When exposed to equivalent bioavailable amounts of nano- and soluble metal-forms, gill copper uptake was identical in zebrafish. However, nanocopper caused greater damage to the gill. Nanocopper produced different morphological effects and global gene expression patterns in the gill than did soluble copper ions alone. Similarly, Kasemets et al. (2009) reported that soluble copper ions explained 50% of nanocopper toxicity in

Table 3 Bulk vs. nanocopper toxicity to different species of aquatic organisms

Species	Duration/ effect	Bulk-CuO (mg/L)	n-CuO (mg/L)	Cause of toxicity	Reference
<i>Pseudokirchneriella subcapitata</i> (algae)	72 h EC ₅₀	11.55	0.71	Soluble Cu ²⁺	Aruoja et al. (2009)
<i>Vibrio fischeri</i> (bacteria)	1/2 h EC ₅₀	3899	79	Soluble Cu ²⁺	Heinlaan et al. (2008)
<i>Daphnia magna</i> (crustacean)	48 h EC ₅₀	164.8	3.2	Soluble Cu ²⁺	Heinlaan et al. (2008)
<i>Thamnocephalus platyurus</i> (crustacean)	24 h EC ₅₀	94.5	2.1	Soluble Cu ²⁺	Heinlaan et al. (2008)
<i>Saccharomyces cerevisiae</i> (yeast)	8 h EC ₅₀	1297	20.7	50% soluble Cu ²⁺	Kasemets et al. (2009)
<i>Tetrahymena thermophila</i> (protozoa)	24 h EC ₅₀	873	13.4	Soluble Cu ²⁺	Mortimer et al. (2010)
	4 h EC ₅₀	1705	128		
	24 h EC ₅₀	1966	97.9		

n = nano

yeast. In vitro studies provided evidence to show that copper nanoparticles have the ability to cause mitochondrial (Karlsson et al. 2009) and DNA damage (Midander et al. 2009). Although the mechanisms of nanoparticle toxicity are not well understood, the findings to date suggest that both ionic copper and nanoparticulate copper are responsible for the toxicity that is produced.

5 Monitoring and Ambient Water Quality Standards

Because of the heavy use of copper oxide-based boat antifouling paints in poorly flushed marine environments, copper monitoring data in marinas are particularly useful for assessing potential water quality impacts. Several copper monitoring studies have been conducted in California marinas (Table 4). Singhasemanon et al. (2009) reported median DCu concentrations from 23 marinas that reflected a range of water salinities. Different water types were determined by measuring water electrical conductivity (EC) in micro-Siemens/centimeter ($\mu\text{S}/\text{cm}$). The values are 0–1,500 for freshwater, 1,500–15,000 for brackish water, and >15,000 for saltwater. “There were significant differences in median DCu concentrations among the three water types (one-way ANOVA, $F_{2,64} = 8.90$, $p < 0.0005$), with freshwater marina median DCu concentrations being significantly less than those in salt and brackish

Table 4 Dissolved copper concentrations ($\mu\text{g/L}$) measured in the water column of California marinas at different water salinities and the CTR standard values ($\mu\text{g/L}$)

Source	Saltwater	Brackish water	Freshwater
	DCu	DCu	DCu
Singhasemanon et al. (2009)	3.7 (median)	2.6	1.4
RWQCB (2007)	4.27 (mean)	–	–
Schiff et al. (2007)	7.0 (mean)	–	–
CTR ^a	3.1/4.8	3.1/4.8	9.0/13 ^b

Source: U.S. EPA (2000)

(CTR- California Toxics Rule; CCC- criterion continuous concentration; CMC- criterion maximum concentration; DCu- dissolved copper)

^aCCC and CMC, respectively

^bThe CTR values for freshwater are based on total water hardness (100 mg/L) and will change depending on the individual fresh water body

water marinas (Tukey's Test, family error rate = 0.05). In contrast, there was no significant difference between median DCu concentrations in salt and brackish water marinas" (Singhasemanon et al. 2009). This suggests that there are similarities in the sources of dissolved copper or in the physical and chemical processes that are driving the cycling of dissolved copper in saltwater and brackish water marinas.

Median DCu concentrations of marina samples were greater than median concentrations found at their associated local reference sites (LRSs) (Singhasemanon et al. 2009). Among the three water types, median DCu concentrations were 3.7, 2.6, and 1.4 $\mu\text{g/L}$ for saltwater, brackish water, and freshwater marinas, respectively. For comparison, the associated LRS median concentrations were 0.6, 1.6, and 0.5 $\mu\text{g/L}$. Through source analysis, Singhasemanon et al. (2009) concluded that during the dry weather season (July through October), antifouling paints are probably a major source of copper pollution in California saltwater and brackish water marinas. Similar data from studies performed in Southern California also indicated elevated DCu concentrations in saltwater marinas during the dry season (Table 4) (RWQCB 2007; Schiff et al. 2007). The authors of a study conducted by the Santa Ana RWQCB (2007) further concluded that DCu from copper-containing boat coatings may be settling in marina sediments of Lower Newport Bay.

California Toxics Rule (CTR) standards pertain to the chemical concentrations in inland surface waters and enclosed bays and estuaries and are intended to protect human health and the environment (U.S. EPA 2009b). CTR standards establish freshwater and saltwater thresholds for chemicals, based on criterion continuous concentrations (CCC) for chronic toxicity endpoints and criterion maximum concentrations (CMC) for acute toxicity endpoints (Table 4). In their study, Singhasemanon et al. (2009) found that 51 and 30% of their brackish water and saltwater marina samples exceeded the CTR's CCC and CMC standards, respectively. In contrast, none of their freshwater marina samples exceeded the CTR standards. This suggests that elevated copper concentrations in some saltwater and brackish water marinas may pose a risk to aquatic life.

6 Summary

Besides being a naturally occurring element and an essential micronutrient, copper is used as a pesticide, but at generally higher concentrations. Copper, unlike organic pesticides, does not degrade, but rather enters a complex biogeochemical cycle. In the water column, copper can exist bound to both organic and inorganic species and as free or hydrated copper ions. Water column chemistry affects copper speciation and bioavailability. In all water types (saltwater, brackish water, and freshwater), organic ligands in the water column can sequester the majority of dissolved copper, and therefore, organic ligands play the largest role in copper bioavailability. In freshwater, however, the geochemistry of a particular location, including water column characteristics such as water hardness and pH, is a significant factor that can increase copper bioavailability and toxicity. In most cases, organic ligand concentrations greatly exceed copper ion concentrations in the water column and therefore provide a large buffering capacity. Hence, copper bioavailability can be grossly overestimated if it is based on total dissolved copper (TDCu) concentrations alone. Other factors that influence copper concentrations include location in the water column, season, temperature, depth, and level of dissolved oxygen. For example, concentrations of bioavailable copper may be significantly higher in the bottom waters and sediment pore waters, where organic ligands degrade much faster and dissolved copper is constantly resuspended and recycled into the aquatic system.

Aquatic species differ greatly in their sensitivity to copper. Some animals, like mollusks, can tolerate high concentrations of the metal, while others are adversely affected by very low concentrations of copper. Emerging evidence shows that very low, sublethal copper levels can adversely affect the sense of smell and behavior of fish. The developmental stage of the fish at the time of copper exposure is critical to the reversibility of sensory function effects. The fish olfactory system may be the most sensitive structure to copper pollution.

The major factors that influence copper-induced toxicity are dissolved organic carbon and water salinity. Dissolved organic carbon reduces copper toxicity by sequestering bioavailable copper and forming organic complexes with it. Salinity, on the other hand, influences copper bioavailability at the biological action site and also affects metal biodistribution and bioaccumulation in the organism. Therefore, the salinity gradient can increase or decrease copper toxicity in different aquatic species. In some killifish, copper may affect different organs at different times, depending on the water salinity.

The most studied and best explained copper toxicity mechanisms involve inhibition of key enzymes and disruption of osmoregulation in the gill. Other toxicity mechanisms may involve reactive oxygen species generation and changes of gene transcription in the fish olfactory signaling pathway.

More studies are needed to evaluate the potential magnitude of copper remobilization from the sediment that may result from climate change and its effects on surface waters. Moreover, the environmental exposure, fate, and ecotoxicity of emerging metal nanoparticles, including nanocopper, will require additional studies as new forms of copper appear from application of nanotechnology to copper compounds.

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