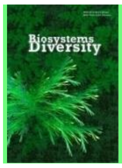

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Copper for crop nutrition

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Copper (Cu) is a transition redox-active metal, one of eight essential trace elements and one of 17 essential elements required by humans, animals, and plants in limited ranges of low concentrations. Copper exists in two oxidation states, Cu^{+} and Cu^{2+}. This property makes copper a key structural component and catalytic cofactor in many metalloproteins. These include enzymes involved in photosynthesis, respiration, stress protection, and lignin metabolism. Plant genomes contain an average of more than 70 copper enzyme genes, indicating its broad importance. Therefore, copper research is important for establishing the scientific basis for nutrition systems with high levels of resource efficiency. In classical plant physiology, redox homeostasis was considered primarily protective; however, recent results show that Cu pools are essential for growth and development, as well as for numerous interactions between plants and their environment. The components of redox homeostasis are also factors in the formation of high levels of nitrogen use efficiency and carbon accumulation during vegetation, as well as in the formation of increased levels of plant adaptation to extreme environmental conditions. Copper is an important regulator of nitrogen use efficiency (NUE). It improves nitrogen uptake and efficient consumption, which is key to reducing nitrogen losses in the environment and increasing crop profitability. This is of paramount importance for the development of crop cultivation technologies in resource-poor environments. An important component of copper's biological activity is its ability to increase plant resistance to disease pathogens. Copper deficiency systematically inhibits NUE, causing growth retardation, decreased enzyme activity, and chlorosis, leading to susceptibility to pests and diseases, impaired root system development, and reduced crop yield and quality. Thus, copper plays a very important role in achieving high levels of nitrogen use efficiency in cereal crops. Cu is also needed in legume crops (soybeans, peas, chickpeas). Unlike other micronutrients, copper is essential for the productivity of cultivated plants throughout the country. With the formation of optimal pools of available copper, the productivity of winter wheat reached 11.0–14.4 t/ha, and triticale – 16.6 t/ha in 2021–2024 in Kyiv region under moderate nitrogen nutrition.

Keywords: Cu; redox homeostasis; nitrogen use efficiency; NUE; plant productivity; saprotrophs.

Introduction

In recent years, crop production in Ukraine has become an important sector of the economy. The war in Ukraine has caused a global food crisis, which, according to the UN, could last for years. Therefore, the main trend in the development of cultivation technologies is resource conservation, ensuring high levels of nutrition efficiency, and crop protection (Schwartau, 2024). The creation and implementation of innovative varieties and hybrids is the basis of modern crop production in the country. Accordingly, the development and implementation of technologies for maximizing the genetic potential of new varieties is of paramount importance for ensuring the country's food security. Today, the number one problem in Ukrainian crop production is a total shortage of resources. Therefore, it is important to implement optimal cultivation systems with high levels of efficiency in the use of nitrogen, other nutrients, moisture, etc. Undoubtedly, the main driving force behind productivity is the provision of nitrogen to crops. Plants absorb less than 50% of the nitrogen applied, and the rest is lost to the environment, causing groundwater pollution, biodiversity loss, and greenhouse gas emissions. Increasing nitrogen use efficiency (NUE) is an urgent need for sustainable agriculture and reducing negative environmental impacts (Cassim et al., 2024; Schwartau, 2024). Improving nitrogen use efficiency is in line with the European Green Deal and the Paris Agreement, i.e., building a climate-neutral economy. Promising solutions for improving nitrogen use efficiency include positioning application zones, utilizing other factors such as disease, weed, and lodging control, applying digital technologies, and ensuring optimal copper pools for crops. The latter area is still one of the least researched components of crop fertilization systems.

Copper has been part of human culture for thousands of years, but it did not play a significant role in agriculture until the 1880s. In 1885, Alexis Millardet published a paper on the effective control of powdery mildew on grapes through the use of a combination of copper sulfate and lime, known as “Bordeaux mixture” (Borkow & Gabbay, 2005; La Torre et al., 2018). Since then, copper has been used to combat various fungal, bacterial, and oomycete diseases, mainly on grapes, fruit, and vegetable crops (Tamm et al., 2022; Pesce et al., 2025). In the 1930s, copper was first introduced as an important micronutrient for plant nutrition (Sommer, 1931; cited in Marschner, 1995). Research on the role of copper in nutrition is classic in plant physiology, from the works of Arnon & Stout (1939) to the current edition of Marschner's Mineral Nutrition of Plants (1995–2022). In Ukraine, the works of Prof. L. K. Ostrovska from the Institute of Plant Physiology and Genetics were pioneering in this field. Her achievements include the monograph “The Physiological Role of Copper and the Basics of Copper Fertilizer Application” (1961), the 1978 State Prize in Science and Technology for the creation, use, and application of complexons in the national economy, and more.

Copper (Cu) is a transition redox-active metal, one of eight essential trace elements (Zn, Cu, Mn, Fe, B, Mo, Co, Cl), and one of the 17 essential elements required for humans, animals, and plant nutrition in a limited range of very low concentrations (Xu et al., 2024). Copper, as a redox-active transition metal, exists in two oxidation states, Cu^{+} and Cu^{2+}, which allows it to participate in numerous biochemical processes associated with redox reactions (Yruela, 2009). This property makes copper a key structural component and catalytic cofactor in many metalloproteins, especially those involved in electron transport. This element participates in various morphological, physiological, and biochemical processes and plays an important role in antioxidant protection and signal transduction, among other things (Chen et al., 2022; Xu et al., 2024). Plant genomes contain an average of more than 70 copper enzyme genes, indicating its widespread importance (Mydy et al., 2021).

Mobility and localization of copper in plants

Copper is an important trace element that is essential for plant growth and development, and its absorption by plants is an active transport process (Ishka et al., 2022). The cell wall is the first barrier to the penetration of Cu ions into plant root cells, containing many macromolecular substances (including amino groups, phosphoric acid, carboxyl and hydroxyl derivatives) (Marschner, 2012).

When Cu passes through the cell wall, various complexes with different amino acids and proteins are formed inside the cell. Copper is then transported to various tissues and organs through the xylem and phloem. At the tissue level, Cu transport mainly involves root absorption, sequestration in vacuoles, loading into the xylem and phloem, and distribution and redistribution between compartments. At the cellular level, Cu transporters identified in plants are divided into two categories: 1) uptake transporters - responsible for transporting extracellular copper into the intracellular space, and 2) efflux transporters - responsible for transporting intracellular copper to the extracellular space or organelles (Chen et al., 2022). In plant cells, Cu is maintained in a relatively balanced state through the coordinated regulation of these two types of Cu transporters. This balance ensures normal plant growth and development and prevents symptoms of Cu poisoning (Ando et al., 2012; Andresen et al., 2018; Adhikari, 2022).

The copper content in many plants does not correlate with the concentration of this element in the soil (Wuana & Okieimen, 2011) and is usually between 5 and 20 mg/kg of dry matter of plant material, with an average value of about 6 mg/kg. Cu is absorbed in the form of Cu^{2+} or Cu chelate and, despite its low mobility in plants, can be transferred from old leaves to new ones. Its concentration in the dry mass of plants is low and usually ranges from 2 to 20 mg/kg. Deficiency symptoms begin when the copper level in tissues falls below 4–5 mg/kg in vegetative tissues, while toxicity for most plants is observed at levels above 20 to 100 mg/kg in dry plant matter (Marschner, 1995, 2011; Rengel et al., 2022).

Copper is limited in its mobility in plants. Experiments with cut roots of different plant species have established a close relationship between Cu^{2+} ions and the root apoplast, as well as the ability of Cu^{2+} ions to remove most other ions from its exchange sites. This suggests that the concentration of this element in the roots is higher than in other parts of the plant. Despite their relatively low mobility, copper ions can be transported through the phloem from the vegetative parts of the plant to the seeds. The uptake of this element is associated with nitrogen compounds due to the high affinity of copper to nitrogen amino groups, which act as copper transporters in the xylem and phloem. *Nicotiana tabacum* plants accumulate copper mainly in their roots, which indicates their potential for bioremediation (Havryliuk et al., 2021).

Xylem serves as the main pathway for copper transport from the root to the shoot. In plants, copper is usually transported in the form of copper complexes rather than free Cu(I) or Cu(II) ions, which helps prevent its toxicity during transport. Copper is transported in xylem sap as Cu(II), bound to specific metal chelants. The best ligands for copper transport in xylem are amino acids, particularly nicotianamine (NA) and histidine, due to their high binding constants for copper at xylem sap pH. Thus, 2′-deoxymugineic acid can serve as a specific copper chelator in the xylem sap of rice plants. In dicotyledonous plants, the re-oxidation of Cu(I) to Cu(II) can occur during transport from the root to the shoot to facilitate complex formation with long-distance transporters (Printz et al., 2016).

Although copper is considered relatively immobile in plants (http://nmsp.cals.cornell.edu/publications/factsheets/factsheet113.pdf) , it is actively remobilized and redistributed within the plant, especially from aging organs to metabolically active younger parts and developing seeds (Printz et al., 2016). This explains why copper deficiency symptoms often first appear on younger leaves. Members of the Yellow Stripe-Like (YSL) family, such as the phloem-localized transporter OsYSL16 in rice, function in the loading of metal complexes into the phloem, facilitating the transfer of copper from aging organs to actively functioning tissues. Metallothioneins (MTs), cysteine-rich proteins, coordinate Cu(I) and are involved in the mobilization of copper from senescent organs. *A. thaliana* mutants with four MT genes had lower copper concentrations in seeds and impaired copper remobilization from senescent leaves, indicating the involvement of MTs in copper redistribution to consumer/acceptor organs. Plant-specific copper chaperone (CCH) accumulates in the vascular veins of the midrib and petioles of senescent leaves, and its presence in phloem exudates indicates a role in the intercellular transport of copper from senescent to reproductive organs, which act as active consumers. COPT6, a protein localized in the plasma membrane, functions primarily for copper homeostasis in above-ground organs and is involved in the remobilization and redistribution of copper from green tissues to reproductive organs (Prinz et al., 2016).

Despite the general characterization of copper as a “low mobility” element in plants, the existence of specialized and highly regulated long-distance transport systems via both xylem (for movement from root to shoot) and phloem (for remobilization to consumer organs) reveals a complex strategy for the dynamic redistribution of copper. This ensures the efficient distribution of necessary copper to actively growing tissues and reproductive organs, directly supporting plant development and productivity formation processes. “Poor mobility” likely refers to the free, unregulated movement of copper. However, plants have developed highly specific and controlled transport systems to overcome this limitation. Chelation of copper into complexes (e.g., with nicotinamide) prevents its toxic reactivity during transport and allows it to be efficiently moved through the vascular system. Phloem-mediated remobilization influences resource allocation, ensuring that limited amounts of copper are prioritized for critical developmental processes such as seed filling/formation, even at the expense of older tissues. This dynamic copper redistribution system is an important adaptive feature.

Understanding and effectively utilizing these pathways (e.g., through breeding or biotechnology) may be a promising strategy for improving nutrient use efficiency and enhancing crop yields, especially in environments where copper availability is suboptimal, by optimizing the delivery of this vital micronutrient to the most developmentally important parts of the plant.

Features of copper mobility and localization in tissues

Copper catalyzes the oxidation of terminal oxidases in living cells. The microelement has a high affinity for peptide, sulfhydryl (including cysteine-enriched proteins), carboxyl, and phenolic groups. The dominant fraction of Cu in plants is present in bound forms; concentrations of free copper in the cytoplasm are usually very low (Cakmak et al., 2023).

Nitrogen-fixing legumes such as beans, peas, and chickpeas usually have higher levels of copper accumulation. Plants absorb the divalent form (Cu^{2+}); cells can reduce it to the monovalent form (Cu^{+}) (Ogunkunle, 2019). The reduced form Cu^{+} binds mainly to sulfur-containing compounds with mercaptan or thioether groups, while the oxidized form Cu^{2+} coordinates mainly with oxygen or imidazole nitrogen groups (Murphy et al., 2020). Thus, Cu acts as a structural or catalytic cofactor in key cellular enzymes that exploit its redox potential (Xu et al., 2024).

In the cell, copper, like other metal ions, is required for the activation and stabilization of enzymes and transcription factors. However, high concentrations of the element disrupt intracellular processes and can lead to cell death. Copper homeostasis in plant cells and subcellular compartments requires coordination between copper-binding proteins, copper chaperones, and copper transporters. The latter also play an important role in the levels of copper mobility to intracellular compartments, such as thylakoid lumens, where it is incorporated into proteins or isolated, preventing the harmful effects of high concentrations of the element.

Specific copper transporters: COPT, ZIP, and YSL families

Copper transporters play a major role in its uptake and distribution in plants, with different protein families performing specific functions. It has been reported that the copper transporter (COPT), heavy metal ATPase (HMA), Yellow Stripe Like protein (YSL), and Cu chaperones are the main transporters and proteins involved in the uptake, root-to-shoot translocation, distribution, and redistribution of Cu in plants (Cui et al., 2022).

COPT copper transporters

COPT transporters are found exclusively in eukaryotes, including plants (known as COPT) and animals/fungi (Ctr). These proteins function as homo- or heterotrimers, forming a channel specifically responsible for transporting Cu^+ (monovalent copper ion) (González-Guerrero et al., 2016). The COPT family consists of five known members: COPT1, COPT2, COPT3, COPT4, and COPT5. In *Arabidopsis*, COPT1, a high-affinity Cu(I) transporter localized in the plasma membrane, especially at root tips, is crucial for copper uptake from the soil. Its expression is activated by SPL7 under copper deficiency conditions. COPT2 also shows increased expression in response to copper deficiency in an SPL7-dependent manner, although its role in copper uptake from the soil may be limited. COPT5 is located in the tonoplast (vacuolar membrane) and is responsible for the release of vacuolar copper for transport to reproductive organs, indicating its role in internal remobilization (Printz et al., 2016).

The sequential regulation of key components of copper uptake, such as COPT1 expression, by the transcription factor SPL7 highlights SPL7 as a central node in the plant copper homeostasis network (Printz et al., 2016). This indicates a complex hierarchical control mechanism that integrates the copper availability sensor with the transcriptional apparatus that controls uptake and distribution. SPL7, as a transcription factor, directly controls gene expression. Due to its sensitivity to copper content, it acts as a sensor and master regulator. Under conditions of copper deficiency, SPL7 activates genes necessary for increasing uptake (e.g., COPT1 for more copper intake) while simultaneously downregulating less important copper-containing proteins (Shahbaz et al., 2015) to prioritize/redistribute limited copper for critical plant functions (to plastocyanin in photosynthesis). This indicates a highly adaptive and efficient regulatory system that allows the plant to rapidly adapt its physiological response to fluctuations in copper availability, optimizing resource allocation and minimizing the harmful effects of both deficiency and excess. Such a regulatory system is important for improving the efficiency of copper use in agricultural crops and has potential for use in biotechnological developments.

ZIP transporters (proteins similar to transporters regulated by zinc and iron)

ZIP transporters are a widespread family of divalent metal transporters that predominantly transport Fe^{2+}, Zn^{2+}, Ni^{2+}, and Mn^{2+} (González-Guerrero et al., 2016). Although not their primary substrate, members of the ZIP family are involved in the uptake of copper ions. Depending on the copper concentration in the plant growth medium, ZIP1 or ZIP5 can act as copper transporters. Their involvement in copper uptake is often associated with competition with other metals, as they can also bind iron (Adamczyk-Szabela & Wolf, 2024).

YSL transporters

YSL transporters belong to the broader family of OPT (oligopeptide transporters) (González-Guerrero et al., 2016). Unlike COPT and ZIP transporters, YSLs do not transport free metal ions. Instead, they transport metal complexes with NA or its derivatives (Printz et al., 2016). NA is a non-proteinogenic amino acid synthesized from S-adenosylmethionine. Transport by YSL proteins occurs via H^+ symport. YSL transporters are important for the uptake of metals from the soil in monocotyledonous plants (strategy II plants) and for the distribution of metals in both monocotyledonous and dicotyledonous plants (González-Guerrero et al., 2016). Specific examples include AtYSL1, AtYSL2, and AtYSL3, which are plasma membrane proteins expressed in the xylem parenchyma of leaves. AtYSL2 can transport Cu-NA complexes, and AtYSL3 may play a role in copper transport in rice plants. The OsYSL16 transporter, localized in the phloem of plants, functions in loading metal complexes into the phloem and transporting copper from aging organs to younger parts and seeds (Printz et al., 2016). The AhYSL3.1 gene in peanuts is also involved in copper transport and the formation of resistance to its excess (Dai et al., 2018).

BdYSL3, the closest homologue of OsYSL16, found in *Brachypodium distachyon*. Wheat, like *B. distachyon*, has reduced fertility and altered flower development when copper is deficient. BdYSL3 is localized on the plasma membrane, transports copper in *Xenopus laevis* oocytes, and is expressed in the phloem. Loss of BdYSL3 function leads to reduced copper delivery to reproductive organs, resulting in delayed flowering, changes in inflorescence architecture, reduced seed set, and reduced grain weight and size. The importance of sufficient copper for plant reproductive development has been demonstrated, as low copper levels can affect crop productivity (Sheng et al., 2021).

The different roles and specificities of the COPT, ZIP, and YSL transporter families, combined with their different energy mechanisms, illustrate a very complex and multi-level strategy for copper uptake, distribution, and remobilization in plants. This coordinated system ensures optimal copper delivery to different tissues and organelles based on their specific needs and the form of copper available. This diversity indicates that plants have optimized their copper processing for different scenarios: COPTs are key for the highly selective, direct uptake of the reduced form Cu(I), which is crucial for initial uptake from the soil. ZIPs provide a broader mechanism for the uptake of divalent cations, potentially acting as a backup or general pathway for copper when concentrations are higher or in specific physiological conditions. YSLs specialize in the efficient transport of chelated copper (Cu-NA complexes), which is vital for the movement of copper from roots to shoots, as well as for remobilization to developing organs and seeds, where free ions can be toxic. This multifaceted approach allows plants to fine-tune copper homeostasis, ensuring that the right form of copper is delivered to the right place at the right time, minimizing toxicity and optimizing its usefulness. This level of control is important for plant survival and productivity in a variety of environments.

The NRAMP transporter family is involved in the transport of several divalent metal ions (Fe, Zn, Mn, Ni, and Cu). Its homologues are involved in iron transport and homeostasis in nodules, supporting symbiotic N_2 fixation. Natural resistance macrophage protein (NRAMP) transporters help transport harmful metal ions across cell membranes and vacuolar membranes in plants (Kumar et al., 2021).

Cu metal chaperones

Cu-chaperones belong to the family of cytosolic low-molecular-weight receptor proteins called metallochaperones. They participate in intracellular metal ion trafficking and the transport/transfer of copper to the active sites of Cu-dependent enzymes, playing a leading role in cellular Cu homeostasis (Llases et al., 2020).

Major cytosolic chaperones include CCH and ATX1 (antioxidant protein 1), which play a crucial role in maintaining copper homeostasis by chelating copper through conserved MxCxxC domains. ATX1 is involved in supporting plant growth under both copper excess and moderate copper deficiency (Printz et al., 2016).

Copper transport/delivery to chloroplasts is important, especially for the maturation of plastocyanin, a key protein in photosynthesis (Aguirre & Pilon, 2016). Transport to chloroplasts involves two P1B-type ATPases: AtHMA6 (PAA1), which imports cytosolic copper into the stroma, and AtHMA8 (PAA2), which imports/transfers copper from the stroma to the thylakoid lumen. Chaperones such as plastid chaperone 1 (PCH1) and stromal CCS may mediate copper delivery to these ATPases (Printz et al., 2016).

Three main types of Cu chaperones have been identified: CCS, CCH, and COX17. CCH regulates the movement of Cu to ATPases that contain Cu. CCH binds to Cu^+ and interacts with P-type heavy metal transporters, facilitating Cu transport. The expression of plant CCH depends on plant aging and oxidative stress. Stromal CCS promotes Cu transport to PAA, which is mainly involved in the transfer of cytosolic Cu to stromal Cu. COX17 mediates Cu transport across mitochondrial membranes. Copper is transported to mitochondria via AtCOX17, where Cu is used as a structural component of the cytochrome c oxidase complex (Pradeep & Aishwarya, 2023).

Vacuoles serve as important compartments for copper storage and detoxification, especially under conditions of copper excess. Excess copper can be sequestered in vacuoles to reduce the concentration of free, toxic ions in the cytosol. This sequestration may be mediated by transporters such as COPT5 (located in the tonoplast) and potentially HMA5 homologs. COPT5 is also involved in the release of accumulated vacuolar copper when it is needed for transport to other parts of the plant, such as reproductive organs (Printz et al., 2016).

The distribution of copper between cellular compartments is dynamically determined by cellular needs, including differentiation, mitochondrial biogenesis, changes in cellular redox status, hormonal signaling, and hypoxia (Yruela, 2009).

Intracellular compartmentalization of copper, which is facilitated by specific transporters and chaperones for targeted delivery to organelles (e.g., chloroplasts for photosynthesis) and sequestration in the vacuole, is a complex process. Copper delivery to chloroplasts is important for photosynthesis due to the function of plastocyanin (Shahbaz et al., 2015). Sequestration in the vacuole acts as a detoxification mechanism, effectively removing excess copper from metabolic activity. Such cellular precision in copper pool management is a fundamental aspect of plant survival, allowing plants to optimize the benefits of copper as an essential cofactor while minimizing its inherent toxicity. Disruption of this internal transport system, even with sufficient external copper, can lead to functional deficits or toxicity, affecting the overall health and productivity of plants.

The physiological role of Cu in photosynthesis and respiration

Copper plays a leading role in photosynthesis, the most important process in plant life. It is known (Xu et al., 2024) that up to 30% of all copper is found in the chloroplasts of green plants, of which 60–80% is localized in thylakoids. Such a high concentration indicates its exceptional importance for photosynthetic proteins and complexes.

Plastocyanin (PC) is the most common copper-containing protein that ensures the functioning of the photosynthetic electron transport chain in plants. It is located in the thylakoid lumen of chloroplasts and functions as an electron carrier/transporter. PC transfers electrons from cytochrome f (Cyt f) in the cytochrome b6f complex to P700 in photosystem I (PSI) (Xu et al., 2024). PC also provides efficient electron transport over long distances during photosynthesis, from the stacked grana of photosystem II (PSII) to distant unstacked areas of thylakoids containing PSI (Höhner et al., 2020). A sufficient pool of PC increases the linear electron flow while reducing the redox state of plastoquinone (PQ) and ferredoxin (Fd), thereby reducing the formation of reactive oxygen species (ROS), which is critical for protecting cells from damage. PC acts as a redox capacitor that influences photosynthetic electron transport (PET), CO_2 fixation, and adaptation to high light by modulating the redox states of the electron transport chain (Wu et al., 2025). During the transition from low to high light, PC accumulates to a significant level, effectively reducing the reduction state of PQ and restoring Fv/Fm (García-Cañas et al., 2021).

Copper homeostasis disruption (deficiency/excess) causes photosynthesis inhibition, chlorophyll content reduction, and chlorosis. Copper deficiency reduces photosynthetic activity, leading to decreased energy production and overall plant growth retardation.

In contrast, high/excessive Cu concentrations can cause oxidative stress and replacement of Mg^{2+} ions in chlorophyll molecules with Cu^{2+} ions, rendering the pigment unstable and inactive (Yang et al., 2022; Xu et al., 2024). Cu competes with the binding of -SH residues, replacing cofactors (such as Mg^{2+}, Zn^{2+}, or Fe^{2+}) in photosynthetic pigments or proteins in chloroplasts, affecting photosynthetic activity (Shabbir et al., 2020). Excess copper can damage thylakoid membranes and inhibit the activity of photosynthetic enzymes due to its ability to catalyze the formation of ROS, which damage lipids, proteins, and DNA. Thus, copper is an element that is essential for the activity of the photosystem, but in excess it destroys it, which indicates the dose dependence of the biological activity of the metal.

In addition to photosynthesis, copper is a redox-active metal that is essential for cellular respiration processes (Xu et al., 2024). It plays a key role in the mitochondrial electron transport chain, acting as a coordinating ion in the cytochrome c oxidase (Cox) enzyme (Complex IV) (Yruela, 2009). Cox catalyzes the transfer of electrons from cytochrome *c* to oxygen, reducing it to water and forming a proton gradient that is used to synthesize ATP, which provides cells with energy, while superoxide dismutase 1 (SOD1) participates in the detoxification of superoxide to hydrogen peroxide (Shim & Han, 2023).

Traditionally, it was believed that respiration in photosynthetic plant cells plays a secondary role compared to photosynthesis, serving as a source of energy mainly at night or in non-photosynthetic tissues. However, recent studies conducted on *Physcomitrium patens* mosses have completely refuted this concept (Vera-Vives et al., 2025). It was found that mutations that prevent cytochrome c oxidase activity are lethal. This confirms that respiration is an indispensable process even in photosynthetically active cells (Vera-Vives et al., 2025).

The study showed that disruption of the cytochrome pathway caused by inactivation of complex IV had significant consequences for the entire carbon and nitrogen metabolism, not just for ATP production. A significant disruption in nitrogen assimilation was observed, which led to an overall decrease in the amount of amino acids. Partial restoration of mutant growth occurred only with the external supply of amino acids, but not sugars. This indicates that respiration in photosynthetic cells plays a key role at the interface between carbon and nitrogen metabolism and provides carbon skeletons for amino acid biosynthesis (Vera-Vives et al., 2025). Thus, copper-dependent complex IV (Cox) acts as the main mechanism for coordinating cellular metabolism, emphasizing the fundamental, structural-metabolic role of copper in respiration.

Copper deficiency reduces Cox activity and respiratory efficiency, leading to decreased ATP production and negatively affecting plant growth and development. Copper-dependent transporter proteins in mitochondria (chaperones), such as Cox 17 and Sco1/2, are necessary for the proper assembly and functioning of Cox. Copper deficiency causes disruption of the photosynthetic transport chain. In other words, copper is a key element in plant energy efficiency. It directly supports the plant's ability to generate energy and organic matter by ensuring efficient electron transport and chlorophyll stability. Thus, copper deficiency directly affects energy metabolism, leading to reduced growth and yield, even when other nutrients are available.

The role of copper in antioxidant protection of plants

The role of copper in antioxidant protection of plants is dual / contradictory and can be described as an “antioxidant paradox.” On the one hand, copper is a key element in systems that prevent oxidative stress (Xu et al., 2024). In particular, it acts as a cofactor for copper/zinc superoxide dismutase (Cu/Zn-SOD), which is responsible for converting superoxide radicals ($O_2^{\bullet-}$) into less dangerous compounds, thus protecting cells from damage (Yruela, 2009). This function is especially important in chloroplasts, where large amounts of reactive oxygen species (ROS) can be produced during photosynthesis.

On the other hand, the same redox-active nature of copper that makes it indispensable can be a source of toxicity at excessive concentrations. An excess of free copper ions (Cu^{+} and Cu^{2+}) can participate in Fenton reactions, leading to the formation of highly reactive hydroxyl radicals ($^{\bullet}OH$). These radicals damage cell membranes, proteins, and DNA, inducing severe oxidative stress that leads to chlorosis, necrosis, and stunted plant growth (Yruela, 2009).

Copper ions at low pH cause oxidative stress and disrupt intracellular calcium homeostasis in winter wheat roots (Riazanova et al., 2015).

Plants have several mechanisms to protect themselves from copper toxicity. The cell wall is the first barrier that can adsorb excess copper ions, preventing them from entering the protoplast. This process occurs due to the presence of macromolecular groups on the surface of the cell wall that bind copper ions (Xu et al., 2024). Thus, copper is a regulator of ROS homeostasis, but when this homeostasis is disturbed, it itself becomes a toxic factor.

The role of Cu in other enzymatic processes in plants

Copper is an important cofactor for numerous enzymes involved in various physiological processes.

Laccases (EC 1.10.3.2): multi-copper oxidases that catalyze the one-electron oxidation of phenolic substrates, playing a crucial role in the biosynthesis of lignin, which provides mechanical support and protection to terrestrial plants (Chen et al., 2022). They also influence plant development by cross-linking flavonoids and contribute to defense (Mydy et al., 2021).

Ascorbate oxidases (EC 1.10.3.3): Enzymes found exclusively in plants and fungi mediate the oxidation of ascorbate, which is vital for redox regulation in the extracellular space, reducing the formation of reactive oxygen species (ROS). They are also involved in the regulation of stress responses and plant growth (Mydy et al., 2021).

Type III polyphenol oxidase (PPO): This class of enzymes includes catechol oxidases and tyrosinases. They catalyze the oxidation of o-diphenols and monophenols (tyrosinase), contributing to the metabolic defense of plants, such as fruit browning (Chen et al., 2022). Polyphenol oxidase, a Cu-rich enzyme found in plant thylakoids, catalyzes the conversion of monophenol to o-dihydroxyphenyl and o-quinone in poplar and spinach plants, causing their black or brown pigmentation (Zhang & Sun, 2021; Zhang et al., 2023). The enzymes are distributed in cell walls but are also localized in thylakoid membranes (Lulai et al., 2020). Polyphenol oxidases are involved in the biosynthesis of lignin and alkaloids. In tissues that are deficient in copper, polyphenol oxidase activity is strongly inhibited and phenol accumulation is observed. It converts highly reactive superoxide radicals into less dangerous compounds, protecting cells from oxidative stress (Mydy et al., 2021).

Diamine oxidases are localized in the apoplasm, including the epidermis and xylem of mature tissues (Lulai et al., 2020; Prabhjot et al., 2020). Diamine oxidase activity decreases in plants with copper deficiency and in young leaves, and can be restored when proper copper nutrition is resumed.

Copper-containing amino oxidases (CuAO): These enzymes catalyze the deamination of primary amines using molecular oxygen and water, releasing ammonia and hydrogen peroxide (H_2O_2) (Mydy et al., 2021). Hydrogen peroxide (H_2O_2), produced by copper-containing amino oxidases (CuAO), is essential for lignification, cross-linking of cell wall proteins, and programmed cell death, and also acts as a signaling molecule in stress resistance and growth regulation (Chen et al., 2022).

Cu,Zn-Superoxide dismutase (Cu,Zn-SOD, E.C. 1.15.1.1): an enzyme that is a critical component of the plant's antioxidant defense system, neutralizes molecular oxygen radicals by converting them into hydrogen peroxide (H_2O_2) and molecular oxygen (Chen et al., 2022). It is found in various cell compartments and promotes lignification and prevents DNA mutations (Mydy et al., 2021).

Ethylene receptors: copper also acts as a cofactor for ethylene receptors (Binder, 2020), such as ETR1, which are crucial for plant growth and development (including seed germination, stem thickening, fruit ripening, stomatal opening), as well as for responses to stress (biotic and abiotic) (Binder, 2020; Husain et al., 2020; Chen et al., 2022; Huang et al., 2023). Ethylene is sensed by a series of membrane-bound receptors that are negative regulatory molecules in the ethylene response. When receptors bind to ethylene, their activation response signaling is turned off (Azhar et al., 2023).

Nitrate reductase (NR) and glutamine synthetase (GS): copper indirectly affects NR activity through its important role in the biosynthesis of molybdenum cofactor (Moco), which is the catalytic center for NR activation and nitrate reduction. The application of copper can significantly increase the activity of NR and GS enzymes, nitrogen content, and biomass, promoting ammonium assimilation and the transformation of organic nitrogen compounds (Wen et al., 2024).

Disruption in one copper-dependent enzymatic system can cause a cascade effect, leading to complex symptoms. For example, copper deficiency as a universal cofactor can simultaneously disrupt lignin synthesis, pollen viability, and antioxidant protection. This interrelationship explains why the same deficiency manifests itself in the form of lodging (due to the absence of lignin), increased susceptibility to disease (due to reduced activity of protective enzymes), and reduced yield (due to pollination problems). Copper acts as a central regulator, ensuring the functioning of several vital processes simultaneously.

The role of copper in redox homeostasis and its effect on nitrogen metabolism

The unique nature of copper, which allows it to be both essential for life and potentially toxic, explains the evolution of complex homeostasis mechanisms in plants. Cells do not simply passively absorb copper, but actively and selectively regulate its movement. This process involves the activity of specific protein systems, such as Cu-chaperones, which are involved in intracellular ion trafficking, and transporters responsible for its absorption and redistribution. This complex network of metal trafficking/transport enables the biological benefits of copper to be exploited while minimizing its toxic effects. Understanding the role of copper in plants must begin with this fundamental principle – the principle of maintaining homeostasis (Xu et al., 2024).

Plants have developed complex mechanisms for the proper regulation of copper uptake, transport, and cellular homeostasis. This is due to the dual role of copper: on the one hand, Cu is an essential cofactor, and on the other, it is a potentially toxic element at high concentrations (Xu et al., 2024). Copper uptake at the root surface in dicotyledonous plants mainly occurs through the reduction of Cu(II) to Cu(I) by ferric reductase enzymes (FRO4/5), after which Cu(I) is transported through the roots by COPT/Ctr-like proteins, such as COPT1. Other transporters, including P-type ATPases, the ZIP family, and the NRAMP family, are also involved in copper uptake and redistribution in plants (Xu et al., 2024).

To prevent the formation of reactive oxygen species (ROS), intracellular copper is chelated and delivered to partner proteins by specific chaperones, such as CCH and ATX1. Excess copper can be sequestered in vacuoles (e.g., by HMA5, COPT5) or removed from the cell. Long-distance copper transport occurs via the xylem, often in the form of Cu complexes with amino acids such as nicotinamide (NA) or histidine, facilitated by YSL transporters (Xu et al., 2024). Remobilization of copper from aging organs to younger ones involves metallothioneins (MT) and COPT6 (Printz et al., 2016).

Maintaining optimal copper levels is challenging because there is a very narrow range of concentrations that are optimal for plants (typically 2–20 mg Cu/kg dry weight for higher plants). This limited concentration range, which ensures optimal plant metabolism and development (Yruela, 2009), means that both copper deficiency and excess can have detrimental effects on plant growth and development. This balance requires precise control, as copper has low mobility in the soil (Kaiser & Rosen, 2023: Copper for crop production. University of Minnesota Extension. https://extension.umn.edu/micro-and-secondary-macronutrients/copper-crop-production). That is, the accumulation of copper from previous applications, such as fungicides and copper-containing fertilizers, can persist in the soil for many years (Chen et al., 2022). Therefore, managing copper in agriculture requires not just increasing its amount, but precisely maintaining optimal concentrations, which emphasizes the need for sophisticated diagnostic tools and precision farming methods.

The relationship between copper and nitrogen metabolism

Plants absorb the main forms of nitrogen – nitrates (NO_3^-) and ammonium (NH_4^+) – from the soil solution through specific transport proteins known as nitrate transporters (NRTs) and ammonium transporters (AMTs), respectively. NO_3^-, after being absorbed by plant roots, can be reduced to NH_4^+ by nitrate reductase (NR) and nitrite reductase (NiR). NH_4^+ is then assimilated into amino acids for protein synthesis via the glutamine synthetase (GS)/glutamate synthase (GOGAT) cycle (Cui et al., 2022). The efficiency of these transport systems directly affects the overall nitrogen uptake by the plant. Studies Cui et al. (2022) have shown that providing rice plants with copper significantly improved nitrogen uptake, increasing it by 9.52–30.64%. In turn, nitrogen supply promoted copper translocation from roots to shoots by 27.28–38.45%. This two-way interaction was a key aspect of the synergistic effect between copper and nitrogen observed in plants.

At the molecular level, copper actively regulates the initial stages of nitrogen uptake by the plant. It significantly enhances the expression of genes encoding nitrate and ammonium transporters, such as OsNRT1.1B, OsNRT2.1, OsNRT2.3a, OsNRT2.4, OsAMT1.2, OsAMT1.3, and OsAMT3.1 (Cui et al., 2022). Thus, copper does not simply indirectly affect nitrogen metabolism, but actively regulates the molecular mechanisms responsible for nitrogen uptake. Improved nitrogen uptake in the presence of copper indicates that plants can use available nitrogen more efficiently, potentially reducing the need for excessive nitrogen fertilizer application and lowering the environmental impact.

The efficiency of these transport systems directly affects the overall nitrogen uptake by the plant. Copper application increases NR and GS enzyme activity and nitrogen content in both leaves and roots of Chinese cabbage (Wen et al., 2024).

Copper also directly influences nitrate reductase (NR) activity through its role in the biosynthesis of molybdenum cofactor. NR activity is important for the reduction of nitrates to ammonium (Wen et al., 2024).

High copper levels reduce nitrogen uptake and accumulation by downregulating nitrate reductase (NR1) and low-affinity nitrate transporters (NRT1 family). Although high-affinity NRT2 transporters can increase regulation under moderate copper stress, this is often insufficient to restore nitrogen uptake (Feil et al., 2023). Excess copper can also inhibit nitrogenase activity in symbiotic bacteria, which is crucial for nitrogen fixation (Costa et al., 2025).

In a study by Cui et al. (2022), no significant effect of Cu supply on nitrogen concentration in rice plants was observed, while Zhou et al. (2020) reported that N application increased Cu content in castor bean (*Ricinus communis* L.) roots. In plants with different Cu or N application rates, the interaction between Cu and N may vary. For example, a negative effect of N supply (280 kg/ha) on Cu uptake was observed in *Boehmeria nivea* grown on Cu-contaminated soils (Rehman et al., 2021). In addition, high Cu levels affected nitrogen metabolism in rice plants and inhibited nitrogen uptake and upward translocation in Chinese cabbage (Xiong et al., 2006; Huo et al., 2020).

Excessive copper concentrations can also lead to stress responses, such as increased total ascorbate levels and decreased glutamine synthetase (GS) activity (Llorens et al., 2000). This dual nature of copper – from an essential activator to a toxic inhibitor – indicates that exceeding the optimal threshold can activate stress defense mechanisms and degradation of key enzymes, negating all positive effects on NUE.

Therefore, to achieve maximum fertilizer efficiency, copper and nitrogen must be considered together, rather than as separate elements. Optimizing one without considering the other can lead to suboptimal results and resource losses.

Copper interacts synergistically and antagonistically with other nutrients, affecting their absorption and overall plant metabolism. Copper application cannot be considered in isolation; it is part of a complex network of nutrients. If Ukrainian soils are already deficient in copper, zinc, and phosphorus then applying only copper may exacerbate the deficiency of other essential nutrients if not carefully balanced. For example, increasing copper content may further reduce already low iron and zinc levels. Conversely, eliminating nitrogen deficiency (Teixeira da Silva et al., 2023) with copper can be very synergistic. Various transporters (COPT, ZIP, NRAMP, YSL) and chelators (NA, histidine, MTs) (Xu et al., 2024) highlight the complex molecular mechanisms that regulate these interactions. Disruption of one pathway (e.g., by excess copper) has cascading effects on others, requiring a precise approach to nutrient application in agriculture, particularly in Ukraine. Soil and tissue analysis should not only determine copper status but also the balance of other key micronutrients. The goal should be to achieve optimal fertilizer ratios, not just individual nutrient levels, in order to optimize synergistic benefits and mitigate antagonistic effects, ultimately leading to more efficient nutrient use and higher yields.

Thus, copper does not simply indirectly influence nitrogen metabolism, but actively regulates the molecular mechanisms responsible for nitrogen uptake. Improved nitrogen uptake in the presence of copper indicates that plants can use available nitrogen more efficiently, potentially reducing the need for excessive nitrogen fertilizer application and lowering the environmental burden.

Interaction of copper with Fe and Zn

Copper uptake is not an isolated process; it is significantly influenced by the presence and concentration of other essential micronutrients. Significant antagonistic interactions between copper and zinc are often observed, with increased copper content in roots inhibiting zinc uptake and low zinc levels stimulating copper uptake. This suggests that similar carrier sites may be involved in the uptake and transport mechanisms of both metals (Adamczyk-Szabela & Wolf, 2024). In addition, it has been shown that excess copper in the soil reduces the uptake of iron (Fe), zinc (Zn), and manganese (Mn) (When More Is Less: How Excess Nutrients Can Cause Deficiencies, 2025. https://extension.missouri.edu/publications/g9069). Conversely, iron or zinc deficiency can enhance copper uptake (Shi et al., 2011). High copper concentrations can also impair the ability of plants to absorb nitrogen (N) and phosphorus (P) by affecting their absorption mechanisms at the molecular and physiological levels (Feil et al., 2023). Such antagonistic interactions reveal the complex interrelationship of nutrient uptake. This means that an imbalance in copper availability can cause secondary deficiencies or toxicity of other essential elements, even if these other nutrients are present in sufficient quantities in the soil. For example, excess copper from fungicide application in vineyards (Feil et al., 2023) can lead to induced deficiencies of iron, zinc, or manganese in the plant. This highlights a complex problem in nutrient management, as fertilizers must take into account not only the absolute levels of individual nutrients, but also their ratios and potential synergistic or antagonistic interactions. Ignoring these interactions can lead to suboptimal/negative effects on plant health and reduced yields, impacting agricultural sustainability.

It is known that high Cu concentrations reduce the uptake of macronutrients such as calcium (Ca), potassium (K), magnesium (Mg), and phosphorus (P), as well as micronutrients such as Fe, Mn, or Zn, or alter the distribution of these elements in roots and shoots (Xu et al., 2024).

High copper levels in plants reduce iron content. The optimal copper-iron ratio varies for different plant species. In particular, the absorption and transport of iron are significantly influenced by the concentrations and ratios of other heavy metals (Adamczyk-Szabela & Wolf, 2024). Deficiency of Fe or Zn can enhance Cu uptake, while a concentration of 100 µM Cu significantly inhibited the accumulation of Fe, Zn, and Mn in the roots of *Commelina communis* (Shi et al., 2011). Copper toxicity can also disrupt the functioning of the electron transport chain by replacing iron in Fe-S clusters.

The interaction between Cu and Zn can often be antagonistic due to their competition for adsorption sites in the soil and uptake mechanisms by plant root systems (Alloway, 2008). Increased copper content in roots can inhibit zinc uptake, suggesting similar carrier sites (Adamczyk-Szabel & Wolf, 2024). High zinc levels can also reduce copper availability (Parvej et al., 2025), while excess copper can reduce soil zinc availability (Akila & Masu, 2023). Thus, high Cu concentrations (2.0–3.0 kg/ha) sharply reduced the nutrient content in rice. The use of lower copper concentrations (0.5–1.5 kg Cu/ha) significantly increased the content of macronutrients (total N, P, K) and micronutrients (Fe, Mn, Zn) in rice leaves, grains, and straw. Grain yield at 1.5 mg/kg Cu increased by 62.9% compared to the control. The concentration of Cu in leaves, grains, and straw increased with increasing Cu application rates. Excess copper reduced the availability of zinc in the soil (Akila & Masu, 2023). It has been shown that copper fertilization changes the dynamics of Zn in the soil and vice versa (Luo & Rimmer, 1995; Tani & Barrington, 2005) reported that copper fertilization had an antagonistic effect on the translocation and absorption of Zn by buckwheat plants. In other studies, copper fertilization had no significant effect on Zn uptake by rice, but had a synergistic effect on Zn uptake by bean plants (Fageria, 2002).

It is known that sandy soils are more likely to be deficient in micronutrients, while soils with high pH, carbonate, and organic matter content can also adsorb and limit the availability of Cu and Zn (Rahman et al., 2020, 2022; Rahman & Schoenau, 2022). In field experiments, Karamanos et al. (2003) demonstrated that copper fertilization resulted in a significant increase in wheat grain yield in cases where the copper content extracted with diethylenetriaminepentaacetic acid (DTPA) was less than 0.4 mg/kg.

Rahman & Schoenau (2022) showed that combined copper and zinc fertilization at a rate of 5 kg/ha on phosphorus-deficient soils had an antagonistic effect on spring wheat plants in western Canada, reducing grain yield on two of the three soils used in a controlled environment. Zinc concentrations in soil and plant tissues increased to toxic levels after fertilizer application, especially under phosphorus-deficient conditions, potentially due to an imbalance in the P:Zn ratio. Zinc concentrations in plant tissues increased steadily with fertilizer application, especially in zinc-deficient soils. Most of the added copper and zinc fertilizers remained in the soil in a form available to plants after harvest. It has been shown that copper and zinc bound to carbonates together with phyllosilicate species are the dominant products of the reaction of copper and zinc sulfate fertilizers, which regulate the exchangeability and bioavailability of trace metal elements in agricultural soils. It has been found that Cu forms a complex with oxyhydroxide minerals and organic matter, while Zn is adsorbed on oxyhydroxide minerals (Rahman et al., 2020).

Another study (Qane et al., 2024) found that the combined application of zinc and copper, both in conditions of manganese deficiency and sufficient manganese in the soil, contributed to a significant increase in wheat yield and quality. The highest wheat grain yield was achieved on soils with sufficient Mn when treated with Zn at a dose of 10.0 mg/kg and Cu at a dose of 10.0 mg/kg. A significant increase in protein content was observed only up to a Cu level of 7.5 compared to Cu 2.5, and a further increase in Cu from 7.5 to 10.0 in soil with sufficient Mn content did not significantly affect the protein content. The optimal treatment options were 5.0 mg/kg of zinc sulfate x 7.5 mg/kg of copper sulfate, in terms of using less zinc and copper fertilizers and having a beneficial effect on plants. The use of micronutrients such as Zn and Cu should be considered an integral part of a holistic approach to nutrition to support optimal wheat growth and quality.

Medicinal herbs grown in copper-treated soil had lower iron content in their roots, while manganese content was higher, except for borage, where manganese content in the roots was reduced. The addition of both copper and zinc increased the total phenolic content, especially in common nettle and basil. Peppermint and borage were less responsive to the supplements. Zinc and copper had no significant effect on photosynthesis (Adamczyk-Szabela, & Wolf, 2024).

Foliar application of zinc can significantly mitigate copper toxicity and, at the same time, increase nitrogen uptake. This confirms the well-known antagonistic interaction between these two elements. It has also been found that excessive N application can exacerbate copper toxicity under certain conditions (Tzortzakis et al., 2025). These results suggest that the synergism between copper and nitrogen is only part of a more complex system, where interactions with other elements (Zn, Fe, P) and environmental conditions play a key role.

Molybdenum is a cofactor for enzymes involved in nitrogen fixation and nitrate reduction (https://farmonaut.com/mining/copper-molybdenum-essential-sustainable-uses-in-2025), but its excess can also lead to a decrease in copper content in tomato fruits (Sabatino et al., 2019). In addition, the use of copper hydroxide as a fungicide increases the number of productive stems and increases the content of free nitrogen, phosphorus, and sulfur anions in winter wheat leaves (Riazanova & Schwartau, 2015).

Thus, the interaction of copper with other micronutrients can be either synergistic/additive or antagonistic for the growth and yield of oats. The nature of these interactions depends on the specific elements, their concentrations, and the overall nutrient status of the soil. Understanding these interactions is important for preventing nutrient imbalances when applying copper fertilizers.

Cu toxicity

Copper toxicity creates a systemic nutrient imbalance, not just a specific nitrogen problem. The plant suffers from a "cascading failure of several nutrients," where the toxicity of one element causes a deficiency of several others. This complex interaction means that simply addressing the nitrogen deficiency (e.g., by adding more nitrogen fertilizer) will be ineffective and potentially wasteful, as the plant's ability to absorb and utilize other essential nutrients is impaired. Therefore, these antagonistic and synergistic relationships, which are crucial for remediation measures, must be taken into account. Toxic levels of copper cause nutritional disturbances by altering the accumulation of macro- and micronutrients in both shoots and roots (Feil et al., 2023). Excess copper reduces the uptake of phosphorus (P) by affecting the permeability of root cell membranes, the expression of phosphorus membrane transporters, and root area; manganese (Mn) in the above-ground parts of some plants, increasing it in the roots; as well as potassium (K), calcium (Ca), and iron (Fe) (Chen et al., 2022).

Excessive amounts of Cu can negatively affect plant growth and metabolism; significantly hinder plant growth and development and nutrient uptake; inhibit photosynthesis, root development, and leaf expansion; and affect the functions of some key cellular components, such as proteins, lipids, DNA, and RNA (Gong et al., 2021). High concentrations of Cu reduce nitrogen uptake and accumulation in plants. The reduction in nitrogen uptake occurs through a decrease in the expression of nitrate reductase NR1, low-affinity nitrate transporters (NRT1 family), and bZIP transcription factors such as TGA1 and TGA4, which regulate nitrate transporters (Hippler et al., 2018). Excess copper led to a decrease in plant photosynthesis by inhibiting chlorophyll and PSII biosynthesis, negatively affecting plants. Therefore, it is important to strictly control the Cu balance in plants (Chen et al., 2022).

Thus, the formation of copper pools in the soil can increase nitrogen use efficiency, thereby improving overall plant growth and productivity. Note that both copper deficiency and excess can hinder nitrogen uptake. According to Epstein (Marshchner, 1995), there are 100 copper atoms per 1 million nitrogen atoms in the above-ground part of plants; or 0.1 μmol Cu/g dry matter per 1 mmol N/g dry matter.

Plants, depending on their sensitivity to copper deficiency, are classified as highly sensitive to copper fertilizers – beet (*Beta vulgaris* L. *crassa* group), lettuce (*Lactuca sativa* L.), onion (*Allium cera* L.), spinach (*Spinacia oleracea* L.), and wheat (*Triticum aestivum* L.). asparagus (*Asparagus officinalis* L.), beans (*Phaseolus vulgaris* L.), peas (*Pisum sativum* L.), and potatoes (*Solanum tuberosum* L.) show low sensitivity to copper (Marshchner, 1995).

The most sensitive to copper fertilizers are cereals (wheat, oats, barley), root crops (beets, carrots), sunflowers, vegetables, and fruit and berry crops. Among vegetables, lettuce, spinach, onions, cucumbers, beans, and peas are particularly in need of copper. A good effect from the use of copper-containing fertilizers is achieved in table beets and carrots, and the least effect is achieved in white cabbage, celery, and tomatoes.

Copper deficiency in plants

Micronutrient deficiencies in some crops occur due to increased nutritional requirements resulting from high yields, the application of high doses of NPK fertilizers with low micronutrient concentrations, loss of the topsoil layer due to soil erosion, which contains organic matter and copper; liming of acidic soils, which reduces the concentration of copper by increasing the pH of the soil and adsorbing this element. Copper deficiency can also be caused by certain herbicides applied to the soil or foliar.

Insufficient copper can lead to poor growth and reduced nitrogen use efficiency. Symptoms of Cu deficiency may include chlorosis, poor root development, and reduced enzyme activity, which negatively affects nitrogen uptake and assimilation.

Symptoms of copper deficiency are clearly species-specific and often depend on the stage of deficiency. In copper deficiency, the lower leaves and other organs contain more copper than the upper ones; plants do not remobilize copper from the lower organs to build the upper leaves. This is due to the relative immobility of copper in plants.

The low rate of Cu migration in the phloem causes typical symptoms of Cu deficiency, which first appear on young leaves in the form of chlorosis of the leaf tips, curling of the edges, leaf deformation, or even necrosis (Marschner, 2011, 2022; Farid et al., 2021; Rengel et al., 2022).

In most plants, copper deficiency manifests itself in the appearance of rosette leaves, necrotic spots, and changes in leaf morphology. Many plant species also experience loss/absence of turgor and discoloration of some tissues. Symptoms of copper deficiency in lentils, horse beans, chickpeas, and wheat include chlorosis, stunted growth, twisted young leaves, wilted leaf tips, and general wilting of the plant despite the availability of water.

In plants, copper deficiency usually manifests as a potential disease that can affect changes in the expression of genes that regulate various biological processes, as well as the morphology of roots and leaves (Yruela, 2005; Thomas et al., 2013, 2016). The average Cu content in the dry weight of a plant is approximately 10 μg g/dry matter, while a critical Cu deficiency is usually 1–5 μg g/dry matter (Yruela, 2005; Cohu & Pilon, 2007; Yruela, 2009). Cu deficiency leads to stunted plant growth, accelerated senescence, and reduced productivity (Marschner, 2011; Thomas et al., 2016) due to damage to the electron transport chain, resulting in impaired photosynthesis and respiration (Hajiboland, 2012).

Copper deficiency limits the activity of many plant enzymes, including ascorbate oxidase, phenolase, cytochrome oxidase, diamine oxidase, SOD, as well as plastocyanin, plastoquinone, the content of unsaturated C_{18} fatty acids, the synthesis of isopropyl lipids, a decrease in CO_2 fixation, and sensitivity to environmental stress (Ayala et al., 1992; Rengel et al., 2022). Thus, a number of authors observed a noticeable decrease in ascorbate oxidase activity in corn and wheat grown on soils with limited copper nutrition and a correlation with a decrease in wheat yield. With insufficient amounts of this element, carbon dioxide fixation, electron transport, and thylakoid lipid synthesis were inhibited compared to plants that received adequate nutrition. It is known that the main target of Cu deficiency is photosynthesis (Hajiboland, 2012). A study of the effect of copper deficiency on sugar beet plants found inhibition of PSII and PSI electron transport. Moreover, PSII electron transport activity was not restored by the addition of artificial electron donors. These results may indicate both a direct effect of copper on PSII electron transport, through its participation in electron transfer as a component of the transporter, and an indirect effect, through changes in the polypeptide composition of PSII complex membranes. Copper also increases the activity of NADPH-dependent oxidase, which is responsible for the production of superoxide radicals, and indirectly affects the photosynthetic apparatus of the plant by changing the degree of saturation of individual lipids in the thylakoid membrane, as well as reducing the content of pigments and the synthesis of plastoquinone. In general, copper deficiency-induced disturbances in photosynthesis and respiration affect plant energy metabolism, which can cause a cascade of secondary physiological effects (Droppa et al., 1984).

Copper deficiency reduces plastocyanin formation, which significantly affects PSI electron transport, thereby reducing the net rate of photosynthesis (Shikanai et al., 2003).

Severe Cu deficiency in plants can likely cause the breakdown of PSII complexes, directly altering the structure of thylakoids and promoting the degradation of pigments (chlorophyll and carotenoids) (Ayala et al., 1992; Peng et al., 2013; Thomas et al., 2016).

Cu deficiency leads to reduced grain/seed yield in agricultural crops (Marschner, 2011). The decrease in crop fertility with copper deficiency is the result of many factors, including stunted growth of the stigma, decreased pollen fertility, and pollination ability (Zhang et al., 2018).

It has been established that during the development of wheat plants, copper deficiency affects metabolism, leading to the accumulation of free amino acids and a decrease in the amount of reducing sugars. The accumulation of free amino acids and changes in their composition may indicate the effect of copper stress on nitrate reductase activity and nitrogen metabolism. It is likely that copper deficiency causes greater plant sensitivity to powdery mildew, which is due to insufficient lignification of plant cell walls, as well as a decrease in the activity of polyphenol oxidase and amino oxidases (Mir et al., 2021).

A characteristic feature of Cu^{2+} deficiency in cereals is strong tillering of the plant with white twisted leaf edges and reduced ear / panicle formation. Plants have a light green color, similar to that observed in nitrogen deficiency, although the nitrate content may be relatively high. With severe copper starvation, plants begin to tiller intensively, and the stem gradually dries out. With a weaker manifestation of the element deficiency, individual plants still produce ears, but with incompletely developed spikelets and partial emptiness. Inhibition of the lignification process in tissues due to copper deficiency leads to incomplete development of xylem vessels and increases susceptibility to lodging.

In fruit crops, copper deficiency causes young shoots to die off and delays flowering and fruiting. Symptoms are manifested by the appearance of unusually wide dark green leaves on long soft shoots. Trees take on a “drooping” shape, which develops as a result of a disruption in the lignification process, including a decrease in the activity of laccase and phenolase. With severe copper starvation, the leaves curl severely, and the leaf blade acquires a light green color with darker veins. Red-brown growths appear on the fruits, and a significant portion of them fall off.

Copper deficiency also leads to insufficient water transport and delayed reproductive development of plants, damage to apical meristems (Epstein et al., 2005; Zhang et al., 2019; Rengel et al., 2022), a decrease in the number and size of grains, and incomplete spikelets, which directly affects yield and quality. Cu deficiency has a significant impact on pollen development and viability, seed embryonic development, and fruit formation (Burkhead et al., 2009; Yan et al., 2017).

The role of copper in improving nitrogen use efficiency (NUE)

NUE is an important indicator in modern agriculture. It refers to the ability of crops to absorb and utilize nutrients to produce optimal yields. NUE involves three main processes in plants: nutrient uptake, assimilation, and utilization (Hawkesford & Riche, 2020; Mishra et al., 2023). Low nitrogen fertilizer use efficiency (25–50%) leads to significant economic and environmental losses (Javed et al., 2022; Cassim et al., 2024).

Copper plays an important role in increasing NUE. Copper supply significantly improved nitrogen uptake by rice plants (Cui et al., 2022), which is a major component of overall nitrogen use efficiency. Copper, along with manganese and zinc, is important for the efficient use of nitrogen by wheat plants. The synergistic effect of copper and nitrogen observed in rice plants (Cui et al., 2022) emphasizes that copper can optimize the overall nitrogen use by plants.

The synergistic effect of copper on nitrogen metabolism also occurred under optimal element utilization. Copper at a concentration of 50 mg copper/kg of soil increased the biomass of fresh shoots of green gram (*Vigna radiata* L.), microbial respiration of the soil, and soil pH. A positive effect of copper on the length of the taproot, leaf area, and root volume was noted, while copper concentrations ranging from 100 to 250 mg/kg of soil significantly reduced total chlorophyll content, root length, plant biomass, and nodule formation in green gram (Begum et al., 2024). Thus, providing plants with sufficient / optimal amounts of copper is a direct route to increasing NUE.

Copper deficiency has a systemic inhibitory effect on NUE. It can lead to stunted growth, reduced enzyme activity, and chlorosis (Printz et al., 2016). In particular, a decrease in nitrate reductase activity in the roots and leaves of grapes was observed with copper deficiency

(Llorens et al., 2000). In addition, copper deficiency leads to a decrease in nitrogen concentration in leaves (Zambrosi, 2025) and a slowdown in overall plant growth (Printz et al., 2016). Symptoms of copper deficiency, such as twisting/curling or whitening of young leaves, damage to the apical meristem, reduced seed setting and grain yield, directly affect overall nitrogen use efficiency (Cui et al., 2022). These effects are not isolated; they form a cascade where reduced enzyme activity (e.g., nitrate reductase (NR)) directly impairs nitrogen assimilation, resulting in lower nitrogen concentrations in tissues. This, in turn, also affects chlorophyll synthesis (chlorosis) and overall plant growth, resulting in reduced yield. This systemic effect means that copper deficiency reduces NUE at many stages, from initial uptake to final utilization. Thus, eliminating copper deficiency is a prerequisite for achieving high NUE, as it removes fundamental physiological barriers to efficient nitrogen metabolism.

So, copper doesn't just provide one step in nitrogen metabolism; it improves the plant's ability to absorb and metabolize nitrogen, thereby reducing losses and increasing productivity. Thus, the application of copper can be a powerful tool for improving NUE, contributing to both environmental sustainability (reducing nitrogen losses) and economic profitability (more efficient use of fertilizers).

Interaction between copper deficiency and nitrogen excess / effect of Cu deficiency at high N doses

Copper deficiency can cause specific symptoms that significantly affect plant growth and development. These include distortion or whitening of young leaves, damage to the apical meristem, and a reduction in seed setting and overall grain yield (Cui et al., 2022). In cereals, copper deficiency manifests itself in the form of chlorosis and leaf curling due to the death of their tips. The top leaves, where copper deficiency first appears, are very large and pale in color, and the ovary is weakened. Even hidden “copper starvation” in cereals, which often goes unnoticed or is attributed to other factors, can lead to a 20% loss in yield. Copper deficiency is more common in sandy and peaty soils. Copper availability for plants is higher on acidic soils than on soils with neutral and alkaline reactions, so it is most effective to apply copper-containing fertilizers on calcareous soils.

The interaction between copper deficiency and high doses of nitrogen is critical to understanding optimal plant nutrition. Copper plays an important role in preventing crop lodging, which is a common problem, especially with high doses of nitrogen fertilizers. This suggests that high doses of nitrogen, which stimulate vegetative growth, without an equivalent amount of copper can significantly exacerbate the problem of lodging.

If a crop is deficient in nitrogen, this can also extend to copper due to its poor transfer from old tissues to young ones, indicating the interdependence of the transport of these elements. Plants grown without copper but with high nitrogen levels showed the lowest nitrate reductase activity. That is, high levels of available nitrogen cannot compensate for the lack of copper for effective nitrogen metabolism. Copper deficiency led to a decrease in nitrogen concentration in soybean leaves (Zambrosi, 2025) and had a direct impact on the overall nitrogen status of the plant.

These data indicate that copper deficiency is a “bottleneck” for effective nitrogen nutrition. High doses of nitrogen aimed at achieving maximum yield increase the plant's need for copper to maintain structural integrity (lignin biosynthesis) and effective nitrogen assimilation (NR activity). Without sufficient copper, plants are prone to lodging, and their ability to process and utilize large amounts of nitrogen is significantly impaired, resulting in reduced nitrate reductase (NR) activity and lower nitrogen concentration in the leaves. This means that simply increasing nitrogen fertilizers without addressing copper deficiency is ineffective.

Interaction between copper, nitrogen, and drought

Drought is one of the most serious environmental stresses affecting plant productivity, reducing biomass, quality, and energy, as well as disrupting morphological, physiological, biochemical, and molecular processes (Seleiman et al., 2021). Drought stress leads to a reduction in root absorption area, which causes a decrease in nitrogen uptake and reduces nitrogen use efficiency (NUE) (Cossani et al., 2012). During drought, copper availability to plants is often limited, especially in the upper soil layers, which are prone to drying out. Lack of soil moisture limits the uptake of copper into the roots by diffusion and reduces their activity in searching for this element. Given the critical role of copper in nitrogen uptake, assimilation, and overall energy supply to plants, drought-induced copper deficiency can significantly impair a plant's ability to utilize available nitrogen, even when it is present in the soil. This creates a double negative effect: drought directly causes stress in plants and, at the same time, indirectly causes copper deficiency, which further impairs nitrogen use efficiency and overall plant resilience. Thus, in arid regions, copper nutrition (e.g., foliar application) becomes even more critical to ensure optimal nitrogen use efficiency and mitigate combined stress effects.

Interaction between copper, nitrogen, and salinity

Soil salinity is a significant abiotic stress that can lead to substantial crop losses. Under salt stress conditions, copper and nitrogen-coded carbon dots (Cu, N-CDs) can effectively alleviate oxidative damage in cucumber seedlings by modulating antioxidant defense mechanisms and absorbing reactive oxygen species (ROS). Cu, N-CDs significantly enhance the activity of key antioxidant enzymes such as superoxide dismutase (+34.08%), catalase (+28.11%), peroxidase (+17.54%), and ascorbate peroxidase (+31.54%). In addition, they increase the levels of non-enzymatic antioxidants, including polyphenols (+23.60%), flavonoids (+15.43%), and carotenoids (+51.73%), which strengthens the overall ability of plants to absorb ROS. Cu, N-CDs also induce a significant increase in soluble sugars (+27.27%) and soluble proteins (+32.58%), which improves osmotic pressure and stress resistance in plants (Li et al., 2025).

Excess copper in nutrient solutions can disrupt nitrogen uptake and induce oxidative stress. However, moderate nitrogen levels may allow *Sideritis cypria* plants to tolerate excess copper without significant reduction in yield (Tzortzakis et al., 2025). This indicates that copper, especially in certain formulations, can be used to increase plant resistance to salinity by supporting internal defense mechanisms and osmotic balance, which directly affects nutrient uptake and metabolism.

Interaction of copper, nitrogen, and temperature stress

Temperature stress, both low and high temperatures, negatively affects growth, development, photosynthesis, and nitrogen metabolism in plants (Mishra et al., 2023). Low temperatures have a greater impact on photosynthesis and also limit nutrient uptake by slowing root growth and reducing transporter activity. A balanced nitrogen supply is crucial for increasing/improving both photosynthetic capacity and nitrogen use efficiency under temperature stress (Xiong et al., 2025). Although the direct relationship between copper and nitrogen metabolism under temperature stress is not described in detail in the literature, the general role of copper in supporting photosynthesis and energy metabolism (Swaminathan & Gohil, 2022) suggests its indirect influence. Temperature stress severely disrupts these processes. Therefore, by ensuring the optimal functioning of these copper-dependent pathways, copper indirectly contributes to the plant's ability to cope with temperature extremes and maintain nitrogen assimilation, which is an energy-intensive process. Maintaining optimal copper status may be an auxiliary strategy for improving plant resistance to temperature fluctuations by strengthening the basic energy-generating and metabolic apparatus, thereby indirectly increasing nitrogen use efficiency under stress conditions.

Carbohydrate, lipid, and nitrogen metabolism

Copper affects carbohydrate, protein, and lipid metabolism in plants. In experiments with potatoes, it was found that treating plants with copper sulfate led to an increase in soluble sugar content during the growing season, increased invertase and amylase activity, and increased yield and starch accumulation in tubers. In wheat plants, copper deficiency after flowering leads to the accumulation of soluble sugars in the leaves and roots, which is associated with the absence of carbohydrate outflow due to the destruction of flowers. Pollen sterility, which develops as a result of disruption of the lignification process of the cell walls of anthers, leads to a decrease in the amount of grain that remains green.

With a pronounced copper deficiency, the composition of membrane lipids also changes towards a decrease in double bonds in unsaturated fatty acids. The effect of copper deficiency on electron transport in PSII is manifested in a change in its lipid microenvironment. Insufficient copper nutrition leads to an increase in the amount of saturated fatty acids in thylakoid phospholipids, which play a regulatory role in the functioning of PSII (Yruela, 2013).

Low sugar concentrations in plants experiencing copper deficiency may explain reduced pollen formation and fertility and are the main cause of reduced nodulation and nitrogen fixation in legumes. Experiments with barley have established the feasibility of applying copper fertilizers to increase nitrogen uptake. The nature of phosphorus metabolism changes, as does the level of redox processes. The use of copper fertilizers accelerates the synthesis of phosphatides and complex proteins – nucleoproteins (Robson & Reuter, 1981). A trend towards an increase in organic phosphorus is already observed in the tillering phase.

Nitrogen affects the availability and mobility of copper in the plant. Thus, increased nitrogen nutrition leads to a decrease in the rate of copper translocation from old leaves to new growth areas. Thus, intensive nitrogen nutrition increases the copper requirements of plants and contributes to the exacerbation of copper deficiency symptoms (Marshchner, 2012).

Copper content in soil and availability to plants

Under physiological conditions, the transition metal Cu exists in two common forms: reduced Cu(I) and oxidized Cu(II). Depending on its state, Cu can bind to different substrates. In its reduced form, Cu(I) predominantly binds sulfur-containing compounds with a thiol or thioether group, while the oxidized form Cu(II) coordinates predominantly with oxygen or imidazole nitrogen groups (Cohu & Pilon, 2007). This dual chemistry of Cu allows it to interact with a wide range of molecules, including proteins, to stimulate biochemical reactions or stabilize structural features (Festa & Thiele, 2011). However, the absence of Cu's redox activity can directly lead to the formation of reactive oxygen species (ROS) through Fenton reactions, thereby damaging proteins, DNA, and other biomolecules (Hänsch & Mendel, 2009).

Copper can exist in mineral soils in the form of simple or complex compounds in both Cu^{2+} and Cu^{+} forms. The two main isotopes of this element, Cu 63 (69%) and Cu 65 (31%), coexist (Jouvin et al., 2012). Plant roots mainly absorb copper in the form of Cu^{2+} ions (https://icl-growingsolutions.com/agriculture/categories/the-importance-of-copper-in-plant-nutrition/). In the soil, Cu exists in a complex set of chemical forms, each of which affects its mobility, bioavailability, and potential toxicity. These forms include dissolved copper (in the form of free Cu^{2+} ions or soluble organic/inorganic complexes), colloidal copper (bound to humic acids and clay particles), organically bound copper (Cu-Org), oxide-bound copper (Cu-Ox), carbonate-bound copper (Cu-Carb), and residual copper (Cu-Res) (Tu et al., 2024). The most available and phytotoxic form is soluble copper, which is most prevalent at lower soil pH values (Alva et al., 2000). Organically bound copper (Cu-Org) is often identified as the dominant fraction in soils and has been reported to constitute up to 40.7% of total copper (Kabirinejad et al., 2014). Plants predominantly absorb isotopically light copper (lower $^{\delta\ 65}$Cu values), indicating a predominant mechanism involving the reduction of Cu^{2+} to Cu^{+} on the root surface prior to absorption (Jouvin et al., 2012).

Copper cations easily interact chemically with organic and mineral substances, therefore they are precipitated by various anions (sulfide, carbonate, hydroxide) to form immobile forms. Water-soluble copper compounds make up a small part (up to 1%) of its total amount in the soil and are easily washed out of the root zone. This is especially true for sandy loam and sandy soils with low absorption capacity. In addition to water-soluble forms, exchange-sorbed forms of copper are readily available to crops when it is absorbed by organic or mineral colloids in the soil or clay minerals.

The influence of pH, organic matter content, and particle size distribution on copper availability

Soil pH is the main factor affecting copper availability. Copper solubility decreases as pH increases to 7 and above, as higher pH increases the strength of the bond between copper and soil clays and organic matter, making it less available. Conversely, copper availability is generally higher in acidic soils, which increases its potential toxicity (Xu et al., 2024). The optimal pH for copper availability is 6.5–7.0 (Alva et al., 2000).

It is known that on average 50% of Cu in the soil is insoluble and unavailable, 30% is bound by organic soils, 15% is in the form of oxides, and only 5% is available for absorption by plants (Barber, 1984). Thus, in Europe, the average background concentration of Cu in soils ranges from 11.4 to 17 mg Cu/kg (Alloway, 2013), while the concentration of copper in soils in the midwestern United States ranges from 1 to 40 mg/kg, with an average concentration of 9 mg/kg (Havlin et al., 1999), but only 2–21% of copper in sod is isotopically exchangeable (Barber, 1984). It is believed that 0.5 mg/kg is sufficient for normal growth of agricultural crops, particularly cereals. Factors such as pH, organic matter content, and plant species can affect the availability of micronutrients (Zaimenko et al., 2022) and copper in particular.

Copper binds more strongly to organic matter than any other micronutrient (Matijevic, 2014). High organic matter content in the soil can lead to copper deficiency. Also, sandy soils are more prone to copper deficiency than loams and clays. Sandy soils with very low organic matter content can cause copper to leach from the surface horizon (Sun et al., 2019). Copper-deficient, low-humus sandy, drained marsh, sod-podzolic soils with a light granulometric composition, and peaty soils, where copper is found in organic compounds that are difficult for plants to access, need to be enriched by applying copper-containing fertilizers. Clay soils tend to retain more copper in an exchangeable form (www.tfi.org/wp-content/uploads/2024/01/tfi-copper.pdf). Well-drained aerobic soils usually have low concentrations of free copper ions due to sorption processes and organic complexes. Under reduction conditions (e.g., waterlogging, peat soils), copper sulfides can form, affecting their solubility (Römkens et al., 2004). Waterlogging can negatively affect the uptake of nutrients (Neaman et al., 2024), copper in particular. Soil waterlogging can cause oxygen deficiency, leading to anaerobic respiration in the roots, disrupting nutrient uptake, and affecting plant morphology, growth, and metabolism by increasing ethylene production and disrupting vital plant physiological functions (Zhang et al., 2025).

Analysis of Ukrainian soils (Polissya, Forest-Steppe, Steppe) for copper content

Copper is found in abundance in yellow soils and red soils. There is slightly less copper in saline soils and chernozems. Sod-podzolic, gray soils, and chestnut soils contain lower concentrations of this microelement. Upland peat bogs and sod-carbonate soils are the poorest in terms of copper content.

The total copper content, for example, in the main seven soil types of Ukraine ranges from 10.4 to 38.5 mg/kg (Balyuk & Fateev, 2012). The soils of Polissya, with a gross copper content of 7.4 mg/kg, have rather low reserves and correspond to the lower threshold content. Other soil and climatic zones have an optimal (20–34 mg/kg) copper content. The distribution of mobile forms of Cu decreases from Polissya to the Forest-Steppe, Steppe, and Donbas, i.e., from soils with a light granulometric composition and high acidity to heavy loam and clay soils with a neutral soil solution reaction. Low copper content in cereal crops is characteristic of such soil zones of Ukraine as Polissya, Donbas, Crimea, Precarpathia, and Transcarpathia (Balyuk & Fateyev, 2012). There is a significant deficiency of micronutrients, including copper, in Ukrainian soils. According to data from the Institute for Soil Science and Agrochemistry Research named after O.N.Sokolovsky of the National Academy of Agrarian Sciences, out of 32 million hectares of arable land, 18 million hectares (56%) have low (about 0.20 mg/kg) content of mobile zinc, 2.5 million hectares (8%) have low (1.5–1.9 mg/kg) content of mobile copper, and 8 million hectares (25%) have low (0.3–0.5 mg/kg) content of mobile boron. It should be noted that to obtain average yields, for example, 6–7 t/ha of wheat, it is necessary to additionally apply copper fertilizers on all fields in the country. The main reasons for the low content of micronutrients in Ukrainian soils are the very low application of organic fertilizers, which significantly enrich the soil with both macro- and micronutrients, and failure to adhere to crop rotation patterns (https://uarostok.ua/statt/mkrodobriva-uarostok-zaporuka-otrimannya-visokih-vrozhav-slskogospodarskih-kultur). Copper deficiency can be observed in 90% of Ukrainian soils, often together with zinc and phosphorus deficiency, making them the most limiting elements (https://superagronom.com/blog/1086-problemi-ukrayinskih-gruntiv-poglyadom-nimetskogo-fahivtsya).

Copper deficiency is more pronounced in sandy and peaty soils, as well as in alkaline (calcareous) soils, where its availability is reduced (www.tfi.org/wp-content/uploads/2024/01/tfi-copper.pdf). It is known that liming can also reduce copper availability (Xu et al., 2024). Widespread copper deficiency in Ukraine leads to unrealized yield potential and systemic underperformance in agriculture. The presence of visual symptoms (chlorosis, stunted growth) may mask the true extent of the problem, leading to significant unrealized profits and economic losses.

Micronutrient deficiencies can also occur under unfavorable soil and climatic conditions. For example, boron and magnesium leaching can occur on light sandy soils. Copper becomes unavailable to plants on peatlands. In alkaline environments, the availability of most micronutrients (Zn, Cu, B, Mn, Fe) is limited. Early spring cold spells lead to a delay in root system development, which negatively affects the absorption of soil micronutrients. High doses of nitrogen and phosphorus fertilizers also reduce the availability of copper, zinc, iron, and boron compounds.

Environmental risks of copper use

Copper contamination of agricultural soils has accelerated due to its widespread and repeated use in agriculture and horticulture in fertilizers or fungicides to protect grapes, citrus fruits, and other fruit crops from fungal diseases. Cu applied from various agrochemical sources in the agricultural environment can be adsorbed and transported to the groundwater level and contaminate it, in addition to contaminating the soil. The use of copper-based fungicides in vineyard soils is known worldwide. It has been found that in many countries the concentration exceeds 100 mg/kg. The importance of studying Cu transport arises from the fact that Cu is absorbed by soils and also reaches the groundwater level, thus contaminating both soil and groundwater. It is important to be able to assess different fractions of copper: mobile, readily soluble, exchangeable, or plant-available fractions of Cu content in soil as a more direct indicator of the likelihood of harmful or toxic effects on soil and groundwater (Elhawat et al., 2013; Mir et al., 2021).

In viticulture, Cu-based fungicides are used in typical doses of 2–4 kg Cu/ha/year, resulting in soil Cu concentrations that can exceed 3000 mg Cu/kg soil, thereby exceeding the concentration range acceptable for most cultivated crops, and thus preventing their growth (Alloway, 2013). In grapevines, excessive Cu content due to prolonged use of fungicides can lead to root growth retardation, leaf chlorosis, and reduced fruit yield due to oxidative damage and inhibition of Fe and Zn uptake.

In grain production, the use of copper is limited to the vegetative growth period. The application of copper during the period from ear formation to ear filling can lead to increased accumulation of heavy metals in the grain.

A look at Ukrainian and international experiments on increasing crop yields at optimal Cu pools

Copper is an important micronutrient that plays an active role in the growth, development, reproduction, and formation of grain yield and crop quality (Marschner, 1995, 2011; Ishka et al., 2022). A sufficient supply of copper improves the level of nutrients in the plant and the activity of the mechanisms necessary for normal growth and better yield (Arif et al., 2016). Studies conducted on the valuable medicinal mushroom *Trametes versicolor* have shown that copper citrate significantly stimulates mycelium growth, increases the assimilation rate of nitrogen sources, and increases biomass synthesis by 79% compared to the control group. Copper citrate significantly increases the nitrogen assimilation rate and biomass synthesis of the fungus *Trametes versicolor* compared to copper sulfate (Al-Maali, 2017).

Over the past decade, numerous studies both globally and in Ukraine have confirmed the important role of copper in optimizing the growth, development, and productivity of agricultural crops, especially in the context of nitrogen nutrition. For example, studies on rice have demonstrated a pronounced synergistic effect between copper and nitrogen, leading to improved plant growth and yield. It was found that copper significantly improved nitrogen uptake, and nitrogen, in turn, promoted copper translocation in the plant (Cui et al., 2022). Foliar application of copper on corn showed that the optimal dose (up to 100 g/ha) significantly increased chlorophyll content, leaf area, cob diameter and length, thousand-kernel weight, and overall yield. However, higher doses (over 100 g/ha) were toxic and reduced plant productivity. Copper deficiency in soybeans led to a significant reduction in dry weight and the number of nitrogen-fixing nodules, as well as nitrogen concentration in the leaves. Foliar/leaf treatment of plants with copper restored the negative effects of copper deficiency on pod yield, although it did not always completely improve nodule formation, nitrogen nutrition, and vegetative growth (Zambrosi, 2025). The application of copper to Chinese cabbage (*Brassica pekinensis*) increased plant biomass, the activity of nitrate reductase (NR) and glutamine synthetase (GS) enzymes, expression of the corresponding genes (NIA and Gln2), and nitrogen content in both shoots and roots (Xiong et al., 2006). In Ukraine, copper fertilizers are used in liquid form because of their better availability. They are more readily absorbed by plants showing signs of deficiency, helping to reduce the risk of lodging and susceptibility to smut, which is important for cereal crops. Copper had a positive effect on the germination energy of cereals, as well as on the effective consumption of nitrogen by wheat plants. Copper deficiency is more common in sandy and peaty soils, as well as in soils with neutral and alkaline reactions, which requires specific approaches to fertilization. The use of microfertilizers containing copper prevents copper deficiency and lodging of crops (especially against the background of high doses of nitrogen), contributes to an increase in protein in cereals, sugar in root crops, and vitamin C in fruits. Retardants such as Terpal had a positive effect on the accumulation of copper, zinc, and other micronutrients in winter wheat grain (Miroshnichenko et al., 2017).

Copper deficiency can lead to serious crop losses and weak plants. Symptoms include distortion or whitening of young leaves, damage to the apical meristem, reduced seed setting, and grain yield. For example, in tomatoes, copper deficiency during growth leads to low copper content in fruits and changes in ripening processes (Ishka et al., 2022).

Copper deficiency also significantly reduces dry weight and the number of nitrogen-fixing nodules in legumes, which is directly reflected in a decrease in nitrogen concentration in the leaves. In soybeans, copper deficiency sharply reduces dry weight (–88%) and pod number (–86%), although the effect on shoot biomass is less pronounced (Zambrosi, 2025). Foliar copper treatment could reverse the negative effects of copper deficiency on pod yield, but did not improve nodule formation, nitrogen nutrition, and vegetative growth (Zambrosi, 2025). This indicates that foliar treatment can be effective in restoring yield in the later stages, but does not always completely eliminate all physiological effects of deficiency, especially in the early stages of root system development and nitrogen-fixing/nitrogen-fixing nodules. Excess copper is also toxic, leading to reduced yield, plant productivity, and biomass (Alves et al., 2023). Toxic levels of copper can cause leaf bronzing/necrosis, inhibit leaf expansion, reduce the absorption of iron (Fe), zinc (Zn), manganese (Mn), and cobalt (Co), and cause excessive production of hydrogen peroxide (H_2O_2) (Rather et al., 2020). The above approaches to improving resource use efficiency, nitrogen use efficiency, etc. under the influence of copper in the form of hydroxide have been implemented in the cultivation of wheat varieties. In recent years, the following results have been achieved in realizing the genetic potential of winter wheat varieties selected by the Institute of Plant Genetics and Soybean Breeding of the National Academy of Sciences of Ukraine at moderate levels of nitrogen nutrition ($N_{110–120}$) in the conditions of Kyiv region:

– 2021: innovative winter wheat varieties Sofia Kyivska, Horodnytsia, and Kyivska-19 yielded 11.0 to 13.6 t/ha, with an average yield in Ukraine of 4.53 t/ha;

– 2022: Kyivska 19, Sofia Kyivska, and Gorodnytsia yielded 10.7-11.7 t/ha, with an average yield in Ukraine of 3.93 t/ha;

– 2023: the Horodnytsia variety yielded 13.48, Sofia Kyivska – 14.41, the high-quality wheat variety Zdoba Kyivska –10.65 (protein content up to 15%), the Kyivska 19 variety yielded 13.23 t/ha, compared to the average yield in Ukraine of 4.75 t/ha;

– 2024: the growing season was marked by extremely high temperatures during the grain filling period of winter crops: in the last 2.5 weeks of wheat growth, the air temperature reached 40–42 °C. Under these conditions, the Kyivska 20 variety yielded 13.3 to 14.2 t/ha, and Zvenyhora yielded up to 13.5 t/ha, compared to an average yield in Ukraine of 4.5 t/ha.

With moderate costs for growing Okovyta triticale, yields of 14.6 to 16.6 t/ha were obtained in 2024. The crop is promising for the south and central parts of Ukraine, for regions with constant and periodic droughts.

The high productivity potential of modern winter wheat and triticale varieties is realized with reduced cultivation costs, increased nitrogen use efficiency, and, accordingly, a significant reduction in greenhouse gas emissions. The application of copper (twice in BBCH 31–32 and BBCH 37–39 at 200 g/ha in the form of hydroxide) is part of optimal crop nutrition and protection technologies.

Conclusion

Copper (Cu) is a transition redox-active metal that is one of eight essential trace elements and one of 17 essential elements required by humans, animals, and plants in limited ranges of low concentrations. Copper exists in two oxidation states, Cu^+ and Cu^{2+}. This property makes copper a key structural component and catalytic cofactor in many metalloproteins. These include enzymes involved in photosynthesis, respiration, stress protection, and lignin metabolism. Plant genomes contain an average of more than 70 copper enzyme genes, indicating its widespread importance. Therefore, copper research is important for establishing the scientific basis for nutrient systems with high levels of resource efficiency. In classical plant physiology, redox homeostasis was considered primarily protective; however, recent results show that Cu pools are essential for growth and development, as well as for the formation of numerous interactions between plants and their environment. The influence of copper extends to growth, yield, and product quality. It is essential for photosynthesis, chlorophyll formation, and lignin biosynthesis, which prevents lodging, especially at high nitrogen doses. Optimal copper supply also increases the protein content in cereals, sugar in root crops, and vitamin C in fruits, improving the nutritional value of the crop.

The components of redox homeostasis are also factors in the formation of high levels of nitrogen use efficiency and carbon accumulation during vegetation, forming increased levels of plant adaptation to extreme environmental conditions. Copper is not just a trace element, but a key regulator of many aspects of nitrogen metabolism. Copper is an important regulator of nitrogen use efficiency (NUE). It improves nitrogen uptake and efficient consumption, which is key to reducing nitrogen losses in the environment and contributes to increased crop profitability. This is of paramount importance for the future development of crop cultivation technologies in a context of resource scarcity. Copper deficiency systematically inhibits NUE, causing stunted growth, reduced enzyme activity, and chlorosis. Cu deficiency can also cause plant weakening, growth retardation, and susceptibility to pests and diseases; it can lead to poor root system development and reduced crop yield and quality. An important component of copper's biological activity is its ability to increase plant resistance to disease pathogens.

Copper has been found to interact synergistically with nitrogen, improving its uptake by plants by activating the expression of nitrate and ammonium transporter genes. It also plays a dual, dose-dependent role in regulating the activity of nitrate reductase, a key enzyme in nitrogen assimilation. Optimal copper levels increase the activity of this enzyme, while both deficiency and excess can lead to its reduction or dysfunction. Although copper is not a direct cofactor of nitrogenase, it is critical for biological nitrogen fixation, providing energy for bacteroids and protecting the nitrogen-fixing apparatus from oxidative stress. The fundamental importance of copper is also confirmed by its role in cytochrome oxidase, which provides cellular energy (ATP) for all energy-intensive processes, including nitrogen assimilation.

The consequences of copper deficiency at high nitrogen doses are particularly devastating, as high nitrogen levels cannot compensate for copper deficiency, leading to increased lodging, reduced activity of key nitrogen metabolism enzymes, and overall deterioration in growth and productivity. This emphasizes that copper deficiency is a critical "bottleneck" for efficient nitrogen utilization.

The above approaches to improving the efficiency of resource use, nitrogen, etc., under the influence of copper in the form of hydroxide have been implemented in the cultivation of wheat varieties. It is necessary to determine the varietal characteristics of wheat and their sensitivity to copper deficiency or excess when determining fertilizer application rates. In recent years, the following results have been achieved in realizing the genetic potential of winter wheat varieties selected by the Institute of Plant Genetics and Soybean Breeding of the National Academy of Sciences of Ukraine at moderate levels of nitrogen nutrition ($N_{110–120}$) in the conditions of the Kyiv region:

– 2021: innovative winter wheat varieties Sofia Kyivska, Horodnytsia, and Kyivska-19 yielded 11.0 to 13.6 t/ha, compared to the average yield in Ukraine of 4.53 t/ha;

– 2022: Kyivska 19, Sofia Kyivska, and Gorodnytsia yielded 10.7–11.7 t/ha, with an average yield in Ukraine of 3.93 t/ha;

– 2023: the Gorodnitsa variety yielded 13.48, Sofia Kyivska – 14.41, the high-quality wheat variety Zdoba Kyivska –10.65 (protein content up to 15%), the Kyivska 19 variety yielded 13.23 t/ha, compared to the average yield in Ukraine of 4.75 t/ha;

– 2024: the growing season was marked by extremely high temperatures during the grain filling period of winter crops: in the last 2.5 weeks of wheat growth, the air temperature reached 40–42 °C. Under these conditions, the Kyivska 20 variety yielded 13.3 to 14.2 t/ha, and Zvenyhora yielded up to 13.5 t/ha, compared to an average yield in Ukraine of 4.5 t/ha.

With moderate costs for growing Okovyta triticale, yields of 14.6 to 16.6 t/ha were obtained in 2024. The crop is promising for the south and central parts of Ukraine, for regions with constant and periodic droughts.

The high productivity potential of modern winter wheat and triticale varieties is realized through reduced cultivation costs, increased nitrogen use efficiency, and, accordingly, a significant reduction in greenhouse gas emissions. The application of copper is a component of optimal crop nutrition and protection technologies. Thus, the role of copper in achieving high levels of nitrogen use efficiency in grain crops is very important. Cu is also needed in legume crops (soybeans, peas, chickpeas) and in the southern regions of Ukraine. Unlike other micronutrients, copper is essential for the productivity of cultivated plants throughout the country.

In conditions of drought, salinization, and temperature stress, the role of copper becomes even more significant. Drought limits the availability of copper, exacerbating its deficiency and impairing nitrogen utilization. In saline soils, copper can enhance the antioxidant systems of plants, protecting them from oxidative damage and supporting metabolism. Copper also indirectly contributes to the formation of resistance to temperature stress by supporting energy and photosynthetic processes. Its role in antioxidant systems, such as superoxide dismutase, is fundamental to protecting the integrity of cells/plants under stressful conditions.

A summary of experimental data, including Ukrainian studies, confirms the need to use copper to increase the yield and quality of agricultural crops. Practical recommendations include accurate diagnosis of available copper pools, selection of optimal application methods (foliar for quick correction, soil-based taking into account soil conditions), accurate dosing, and a comprehensive approach to nutrition that takes into account the interaction of copper with nitrogen and other nutrients.

Future research should focus on deepening our understanding of the molecular mechanisms of copper and nitrogen interactions and studying their interactions under conditions of combined stress. This will improve copper pool management, which is key to increasing NUE and ensuring the sustainability of agricultural systems in the face of climate change and resource scarcity.

The research was carried out in accordance with the scientific work “Fundamentals of rational nutrition and protection to increase the productivity of winter wheat and other crops”, state registration number 0119U002235; and “Creating and transferring the first domestic varieties of new cereal crops with unique characteristics of grain for the production of healthy food products and ensuring food safety of Ukraine”, state registration number 0125U00707.

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