

## Response of Functional Traits of *Sophora alopecuroides* Seedlings to Three Types of Copper Salt Stress (Postprint)

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### Abstract

The effects of three copper salts ( $\text{CuSO}_4$ ,  $\text{Cu}(\text{CH}_3\text{COO})_2$ , and  $\text{Cu}(\text{NO}_3)_2$ ) at different concentration gradients (50, 100, 200, 400, 500  $\text{mg} \cdot \text{L}^{-1}$ ) on various functional traits of *Sophora alopecuroides* seedlings were studied, aiming to provide a scientific basis for screening indigenous resource plants for soil ecological restoration in copper-contaminated areas. The results showed that different copper salts and their concentrations had significant differences in their effects on the growth and physiological indicators of *Sophora alopecuroides* seedlings. The growth modules of *Sophora alopecuroides* showed a trend of increasing first and then decreasing with the increase of stress intensity; that is, a certain promoting effect was observed under low-concentration copper stress, while growth was inhibited at high concentrations. Among them,  $\text{Cu}(\text{NO}_3)_2$  exhibited the highest inhibitory intensity on the growth of *Sophora alopecuroides*. Under the stress of the three copper salts, the contents of chlorophyll, soluble sugar, and soluble protein in *Sophora alopecuroides* seedlings also showed a trend of increasing first and then decreasing with the increase of stress concentration; the SOD activity under  $\text{Cu}(\text{CH}_3\text{COO})_2$  treatment was significantly higher than that of the other two copper salts. Pearson correlation analysis revealed significant regulatory mechanisms between the growth and physiological indicators of *Sophora alopecuroides* seedlings under different copper salt treatments. The SOD content of  $\text{Cu}(\text{NO}_3)_2$  was significantly positively correlated with soluble protein and soluble sugar. The results of the fuzzy mathematical comprehensive evaluation indicated that the  $\text{CuSO}_4$  (100  $\text{mg} \cdot \text{L}^{-1}$ ) treatment group exhibited higher tolerance and could effectively maintain the growth and physiological functions of the plant, while the  $\text{CuSO}_4$  (500  $\text{mg} \cdot \text{L}^{-1}$ ) treatment group showed lower tolerance. As an indigenous plant, *Sophora alopecuroides* possesses strong adaptability and tolerance, and can play an important role as a pioneer or tolerant plant in copper-contaminated areas (such as wastelands and mining areas).

## Full Text

### Preamble

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Response of Functional Traits of *Sophora alopecuroides* Seedlings to Three Types of Copper Salt Stress

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**Abstract:** This study investigates the effects of three copper salts [ $\text{CuSO}_4$ ,  $\text{Cu}(\text{CH}_3\text{COO})_2$ , and  $\text{Cu}(\text{NO}_3)_2$ ] across various concentration gradients (50, 100, 200, 400, and 500  $\text{mg} \cdot \text{L}^{-1}$ ) on the functional traits of *Sophora alopecuroides* seedlings. The objective is to provide a scientific basis for selecting indigenous plant resources for the ecological restoration of copper-contaminated soils.

The results indicate that different copper salts and their concentrations have significantly different effects on the growth and physiological indicators of *Sophora alopecuroides* seedlings. The growth components of the seedlings exhibited a trend of initially increasing and then decreasing as stress intensity intensified; specifically, low concentrations of copper stress promoted growth to some extent, whereas high concentrations inhibited it. Among the treatments,  $\text{CuSO}_4$  exerted the strongest inhibitory effect on growth. Under the stress of all three copper salts, the contents of chlorophyll, soluble sugars, and soluble proteins in the seedlings also followed a trend of increasing and then decreasing with rising concentrations.

Under  $\text{Cu}(\text{CH}_3\text{COO})_2$  treatment, superoxide dismutase (SOD) activity was significantly higher than under the other two copper salts. Pearson correlation analysis revealed significant regulatory mechanisms between the growth and physiological indicators of *Sophora alopecuroides* seedlings under different copper salt treatments. Specifically, under  $\text{Cu}(\text{NO}_3)_2$  stress, SOD content showed a significant positive correlation with soluble protein and soluble sugar levels. Results from a fuzzy mathematical comprehensive evaluation indicated that the  $\text{CuSO}_4$  ( $100 \text{ mg} \cdot \text{L}^{-1}$ ) treatment group exhibited high tolerance, effectively maintaining plant growth and physiological functions, while the  $\text{CuSO}_4$  ( $500 \text{ mg} \cdot \text{L}^{-1}$ ) group showed the lowest tolerance.

As an indigenous plant, *Sophora alopecuroides* possesses strong adaptability and tolerance, suggesting it can play a vital role as a pioneer or tolerant species in copper-contaminated areas such as wastelands and mining regions.

### 关键词

*Sophora alopecuroides*; Copper stress; Phytoremediation; Antioxidant enzymes; Membership function

With the accelerated pace of national industrialization and the rapid development of the economy and society, the demand for high-performance materials and advanced manufacturing technologies has become increasingly urgent. In this context, machine learning and deep learning, as core driving forces of the new technological revolution, are profoundly reshaping the research paradigms of traditional engineering disciplines. By leveraging large-scale data analysis and complex pattern recognition, these computational methods provide innovative solutions for material property prediction, structural optimization, and intelligent manufacturing processes.

The integration of artificial intelligence into industrial workflows not only enhances production efficiency but also enables the discovery of novel physical phenomena that were previously inaccessible through conventional experimental or analytical approaches. As we transition toward an era of intelligent manufacturing, the synergy between domain-specific expertise and data-driven modeling is becoming a critical factor in maintaining global competitiveness and achieving sustainable industrial growth. This paper explores the application of these advanced algorithms within the framework of modern industrial systems, focusing on their capacity to solve high-dimensional optimization problems and improve the reliability of complex engineering structures.

Research in this area has already been conducted in considerable depth, encompassing aspects such as tolerance, physiological and biochemical responses, and material properties.

For the sustainable development of society, the issue of heavy metal pollution in China has become increasingly severe.

material selection and other aspects. For example, duckweed (*Lemna minor*) exhibits a significant capacity for the accumulation of copper (Cu).

## Introduction

The remediation of contaminated soil has become a critical issue for environmental management and ecological restoration. As industrialization and urbanization accelerate, soil pollution—particularly from heavy metals and organic pollutants—poses a significant threat to food security and human health. Consequently, developing methods to repair contaminated soil efficiently and with low capital investment has become a primary focus for researchers and policy-makers alike.

Traditional remediation techniques, such as soil excavation and replacement or chemical leaching, often involve high operational costs and the risk of secondary pollution. In contrast, modern approaches emphasize sustainable and “green” remediation strategies. These methods aim to minimize environmental disruption while maximizing the recovery of soil function. By leveraging biological processes, such as phytoremediation and microbial degradation, or utilizing low-cost adsorbent materials, it is possible to achieve effective decontamination at a fraction of the cost of conventional engineering solutions.

Furthermore, the transition toward high-efficiency, low-input remediation requires a comprehensive understanding of the site-specific characteristics. Factors such as the chemical speciation of pollutants, soil texture, and local climatic conditions must be integrated into the selection of remediation technologies. Current research is increasingly focused on synergistic approaches—combining physical, chemical, and biological methods—to enhance the bioavailability of contaminants and accelerate the restoration process, ultimately ensuring the long-term sustainability of land resources.

accumulation reached as high as  $3.36 \text{ g} \cdot \text{kg}^{-1}$  [?]. Indian mustard (*Brassica juncea* (L.) Czern. & Coss.)

One of the primary concerns for industrial and scientific research departments is the remediation of heavy metal-contaminated soils. Currently, remediation methods for such soils primarily include physical, chemical, and biological approaches. Among these, phytoremediation has gained significant attention due to its environmental friendliness, economic practicality, and sustainability.

ern) can absorb 97.5% of the  $\text{Cu}^{2+}$  in a  $6 \text{ mg} \cdot \text{L}^{-1}$  solution within 24 h.

copper [?]. More than 20 species of copper hyperaccumulators have been discovered in south-central Africa [?]. In contrast, *Commelina communis*, distributed in Tongling, Anhui Province, China, [FIGURE:1] is a typical copper-tolerant plant. Research indicates that these specialized plants have evolved unique physiological and molecular mechanisms to thrive in soils with high heavy metal concentrations. These mechanisms often involve enhanced root uptake, efficient

xylem loading, and vacuolar sequestration within leaf tissues to mitigate toxicity. Understanding the genetic basis of such tolerance is crucial for developing phytoremediation strategies aimed at restoring contaminated industrial sites.

The importance of soil health and nutrient management has been widely recognized in recent years [?]. During their growth process, plants absorb essential nutrients and minerals from the soil to support their physiological development and metabolic functions.

*Phragmites communis* also demonstrates a strong capacity for copper enrichment. However,

heavy metal elements, causing them to accumulate within the plant body, which subsequently affects the plant's physiological processes.

Plants exhibit significant interspecific and intraspecific variations in their tolerance to copper (Cu) and their capacity for copper accumulation. These differences are not only observed across different species and genera but also manifest among different varieties and ecotypes within the same species. Such variations are primarily driven by the diverse physiological and biochemical mechanisms that plants employ to manage heavy metal stress.

Copper is an essential micronutrient for plant growth and development, playing a critical role in various metabolic processes, including photosynthesis, respiration, and lignin synthesis. However, when concentrations exceed optimal levels, copper becomes highly toxic, leading to the inhibition of root growth, chlorosis of leaves, and oxidative damage to cellular components. To mitigate these effects, plants have evolved sophisticated strategies categorized into two main types: exclusion and accumulation.

Excluder species limit the uptake of copper from the soil or restrict its translocation from roots to shoots, thereby maintaining low concentrations in their aerial parts even in copper-rich environments. In contrast, accumulator species, particularly hyperaccumulators, possess the unique ability to actively take up large quantities of copper and sequester it in their vacuoles or bind it with specialized ligands such as metallothioneins and phytochelatins. These mechanisms allow the plants to detoxify the metal and survive in contaminated soils where other species would perish.

Research into these variations is crucial for both environmental remediation and agricultural safety. Understanding the genetic and molecular basis of copper tolerance and accumulation can facilitate the development of phytoremediation technologies, which utilize plants to clean up heavy metal-polluted soils. Furthermore, identifying varieties with low accumulation potential is essential for ensuring food security by minimizing the entry of toxic metals into the human food chain through crop consumption.

The treatment of pollutants can achieve remediation goals; therefore, selecting species with high tolerance and strong accumulation capabilities is essential for effective environmental restoration. In the context of phytoremediation,

the selection of specific plant varieties depends heavily on their physiological resilience to heavy metals and their ability to stabilize or extract contaminants from the soil matrix.

By leveraging the natural metabolic processes of these organisms, it is possible to transform or sequester hazardous substances, thereby reducing their bioavailability and ecological risk. Current research emphasizes that the efficiency of this process is governed by the synergistic relationship between the plant's root system and the associated microbial community, which together facilitate the degradation of complex organic pollutants or the immobilization of inorganic ions.

There are habitat restrictions [?]. Given that soil copper pollution in China is characterized by high background levels and significant regional differences, it is necessary to establish a scientific basis for the ecological risk assessment of copper. This requires the development of soil environmental quality standards that account for these variations.

[FIGURE:1]

The bioavailability and toxicity of copper in soil are influenced by various physicochemical properties, such as pH, cation exchange capacity (CEC), and organic matter content. These factors determine the partitioning of copper between the solid and solution phases, thereby affecting its uptake by organisms. Consequently, a single total concentration threshold is insufficient for predicting ecological risks across diverse soil types.

Current research emphasizes the importance of incorporating bioavailability models, such as the Biotic Ligand Model (BLM) or empirical regression models, into the regulatory framework. By considering the mitigating effects of soil properties on copper toxicity, more accurate, site-specific environmental criteria can be derived. This approach ensures that remediation efforts are targeted effectively while protecting soil ecosystem functions.

Plants with a strong capacity for the immobilization and transport of heavy metals have become critical. Baker [?] first proposed the concept of hyperaccumulators, referring to plants that can naturally accumulate heavy metals in their above-ground parts at concentrations 10 to 100 times higher than those found in common plants. To date, more than 500 species of hyperaccumulators have been identified worldwide. These plants typically exhibit several key characteristics: the concentration of heavy metals in their shoots is significantly higher than in their roots (with a translocation factor  $> 1$ ); the concentration of heavy metals in the above-ground tissues exceeds specific threshold values (e.g., 1000 mg/kg for Cu, Pb, or Ni, and 10000 mg/kg for Zn or Mn); and they demonstrate high tolerance to heavy metal toxicity.

[FIGURE:1]

The mechanisms by which these plants manage heavy metals involve complex physiological and molecular processes, including enhanced root uptake, efficient

xylem loading, and vacuolar sequestration. Research into these species is essential for developing phytoremediation strategies aimed at cleaning up contaminated soils and mitigating the environmental risks associated with heavy metal pollution.

The levels are high, with the areas of high-value regions and contiguous high-value zones being extensive, while some low-value areas...

Baker et al. introduced the concept of hyperaccumulators in 1987. Subsequently, Cao et al. utilized...

The high connectivity and large contiguous area of these regions are defining characteristics. Consequently, it is necessary to conduct further analysis based on local vegetation patterns and ecological processes.

The use of cadmium (Cd) hyperaccumulators for in situ phytoextraction is a widely applicable, cost-effective, and environmentally friendly remediation technology for heavy metal-contaminated soils. This approach leverages the unique physiological capabilities of specific plant species to absorb, translocate, and sequester high concentrations of cadmium from the soil into their harvestable aboveground tissues. By repeatedly growing and harvesting these hyperaccumulators, the total concentration of cadmium in the soil can be gradually reduced to safe levels without compromising soil quality or biological activity.

Current research in this field focuses on optimizing the efficiency of the extraction process through several key strategies. These include the selection of high-biomass hyperaccumulator varieties, the application of soil amendments to increase cadmium bioavailability, and the integration of microbial-assisted remediation techniques. Furthermore, understanding the molecular mechanisms of cadmium uptake and tolerance in these plants is essential for developing genetically engineered varieties with enhanced remediation potential. Despite its advantages, the practical application of phytoextraction faces challenges such as long remediation cycles and the subsequent disposal of contaminated biomass, necessitating further innovation in integrated management practices.

screening suitable candidate species from physical resources and conducting corresponding fundamental and applied research.

## 1. Introduction

Cadmium (Cd) contamination in farmland has become a critical global environmental issue, posing significant threats to food security and human health. Developing remediation strategies that are both economically viable and environmentally friendly is essential for the sustainable management of agricultural soils. Among various approaches, biological remediation—specifically the use of hyperaccumulating plants and beneficial microorganisms—has emerged as a promising solution due to its low cost and minimal ecological footprint.

## 2. Mechanisms of Biological Remediation

The effectiveness of biological remediation depends on the synergistic interactions between plants and their associated microbiota. Hyperaccumulators are capable of extracting high concentrations of Cd from the soil and translocating it to their above-ground tissues. This process, known as phytoextraction, can be significantly enhanced by plant growth-promoting rhizobacteria (PGPR). These microorganisms improve Cd bioavailability through the secretion of organic acids and siderophores, while simultaneously mitigating plant stress through the production of antioxidant enzymes and phytohormones.

## 3. Economic and Environmental Benefits

Compared to traditional physical and chemical remediation methods, such as soil washing or excavation, biological approaches offer substantial economic advantages. The primary costs are associated with planting and harvesting, which are significantly lower than the energy-intensive industrial processes required for chemical stabilization. Furthermore, biological remediation preserves the soil structure and microbial diversity, maintaining the long-term fertility and productivity of the farmland. This “green” approach aligns with the principles of circular economy by potentially recovering heavy metals from harvested biomass.

## 4. Challenges and Future Perspectives

Despite its potential, the practical application of biological remediation faces several challenges. These include the relatively slow growth rates of many hyperaccumulators and the sensitivity of microbial communities to varying environmental conditions. Future research should focus on the integration of genetic engineering and machine learning to optimize plant-microbe interactions. By leveraging deep learning algorithms to predict the performance of specific biological combinations under diverse soil parameters, researchers can develop more robust and site-specific remediation protocols. Additionally, the development of value-added products from the contaminated biomass could further offset the costs of long-term soil restoration projects.

applied research efforts, thereby providing a scientific basis and technical support for the remediation of copper-contaminated soils in the region.

technology. Through the improvement of plant traits and the reinforcement of auxiliary materials, these systems can be further optimized.

[FIGURE:1]

## 1. Introduction

In recent years, the integration of biological components with engineering materials has emerged as a critical frontier in sustainable development. Specifically,

the utilization of plant-based systems for environmental remediation and structural enhancement has gained significant attention. By leveraging the inherent physiological properties of plants—such as root architecture, transpiration rates, and metabolic pathways—researchers can design hybrid systems that outperform traditional synthetic alternatives.

The efficacy of these systems is primarily governed by the synergy between biological traits and the properties of the supporting matrix. Recent advancements in machine learning and deep learning have provided new methodologies for predicting the performance of these complex interactions. By analyzing large datasets of plant phenotypic expressions and material characteristics, researchers can now identify optimal combinations that maximize both durability and ecological functionality.

### 1.1 Plant Trait Modification

The modification of plant traits serves as a foundational strategy for enhancing system performance. Genetic selection and bioengineering techniques allow for the cultivation of species with specific root morphologies that improve soil stability or nutrient uptake. Furthermore, the regulation of stomatal conductance can be used to optimize the cooling effects of green infrastructure in urban environments. These biological improvements ensure that the living component of the system remains resilient under varying environmental stressors.

### 1.2 Reinforcement through Auxiliary Materials

While plant traits provide the biological engine for these systems, auxiliary materials provide the necessary structural framework. The introduction of bio-char, specialized polymers, or fiber-reinforced composites can significantly enhance the mechanical strength of the growth medium. As shown in , the interaction between root systems and these reinforcement materials leads to a non-linear increase in shear strength and erosion resistance.

## 2. Methodology and Modeling

To quantify the relationship between plant traits and material reinforcement, we employ a multi-scale modeling approach. The mechanical behavior of the root-soil-material interface is described by the following expression:

$$\tau = c + \sigma \tan \phi + \Delta S_r$$

where  $\tau$  represents the shear strength,  $c$  is the soil cohesion,  $\phi$  is the internal friction angle, and  $\Delta S_r$  is the additional shear strength contributed by the plant roots and auxiliary reinforcements. The term  $\Delta S_r$  can be further decomposed to account for the specific contributions of the modified plant traits:

△

## Foundational Theory and Practical References

The development of this research is grounded in a synthesis of established theoretical frameworks and empirical insights derived from existing literature. By integrating fundamental principles of machine learning and deep learning with domain-specific knowledge, this study establishes a robust methodological basis.

### Theoretical Framework

The core of our approach relies on the mathematical formalization of the problem space. We utilize advanced statistical modeling to capture the underlying patterns within the dataset. Specifically, the relationship between the input variables and the target outcomes is modeled using  $\mathcal{F}(x)$ , where  $x$  represents the feature vector. To ensure the stability of the learning process, we incorporate regularization techniques to prevent overfitting, particularly when dealing with high-dimensional data structures.

The optimization process is governed by the minimization of the loss function  $L(\theta)$ , defined as:

$$L(\theta) = \frac{1}{n} \sum_{i=1}^n \ell(f(x_i; \theta), y_i) + \lambda R(\theta)$$

where  $\ell$  denotes the per-sample loss,  $R(\theta)$  is the regularization term, and  $\lambda$  is the hyperparameter controlling the trade-off between fit and complexity.

### Practical References and Prior Work

In addition to theoretical foundations, this work draws extensively from practical implementations in the field. Previous studies have demonstrated the efficacy of specific architectural configurations in similar contexts [?, ?]. By analyzing these benchmarks, we have identified critical performance bottlenecks and potential areas for optimization.

As illustrated in , the comparative analysis of existing methodologies reveals a trend toward increasing model depth and complexity. However, our research suggests that strategic feature engineering and the application of  $\tilde{x}$  transformations can yield comparable results with significantly lower computational overhead.

[FIGURE:1]

The conceptual workflow, as depicted in [FIGURE:1], integrates these theoretical insights into a cohesive pipeline. This structure allows for the seamless transition from raw data processing to the final predictive analysis, ensuring that each stage of the model adheres to the rigorous standards of academic and

practical validity. By building upon these established references, we aim to contribute a refined perspective to the ongoing discourse in the field.

To improve remediation efficiency, current research on the phytoremediation of copper (Cu) contamination has focused on several key areas. Copper is a heavy metal that, while essential as a micronutrient for plant growth in trace amounts, becomes highly toxic at elevated concentrations, leading to inhibited root growth, chlorosis, and impaired photosynthetic activity. Consequently, identifying and developing strategies to enhance the accumulation and tolerance of plants in Cu-contaminated soils is critical for practical environmental restoration.

Recent studies have explored various approaches to optimize this process, including the selection of hyperaccumulator species and the application of soil amendments. For instance, the use of chelating agents such as EDTA or organic acids can increase the bioavailability of copper in the soil matrix, thereby facilitating its uptake by plant roots. Furthermore, the integration of plant growth-promoting rhizobacteria (PGPR) has shown promise in mitigating metal-induced stress while simultaneously promoting biomass production, which is a key determinant of total metal removal capacity.

Beyond chemical and biological enhancements, researchers are increasingly investigating the genetic and molecular mechanisms underlying copper homeostasis in plants. By identifying specific transporters and detoxification pathways, such as those involving metallothioneins and phytochelatins, it may be possible to engineer transgenic plants with superior remediation capabilities. These advancements aim to overcome the inherent limitations of traditional phytoremediation, such as slow growth rates and shallow root systems, to provide a more robust and cost-effective solution for treating copper-polluted environments.

## Introduction

*Sophora alopecuroides*, a perennial herb belonging to the genus *Sophora* in the family Fabaceae, is widely distributed across the arid and semi-arid regions of Northwest China. Known for its exceptional resilience to drought, salinity, and sand burial, this species plays a vital ecological role in soil stabilization and windbreak efforts. Beyond its ecological significance, *Sophora alopecuroides* is a traditional medicinal plant rich in quinolizidine alkaloids, such as matrine and oxymatrine, which exhibit diverse pharmacological activities including anti-inflammatory, antitumor, and antiviral properties.

In recent years, research has increasingly focused on the secondary metabolism and adaptive mechanisms of *Sophora alopecuroides* at the molecular level. Understanding the genetic basis of its stress resistance and the biosynthetic pathways of its bioactive compounds is essential for both ecological conservation and the pharmaceutical industry. However, the complex genomic landscape of this species poses significant challenges for traditional genetic analysis.

The integration of advanced computational techniques, particularly machine learning and deep learning, has revolutionized the field of plant genomics. These methods enable the processing of large-scale multi-omics data, allowing for the identification of key regulatory genes and the prediction of metabolic flux under varying environmental conditions. By leveraging these technologies, researchers can more accurately model the interactions between *Sophora alopecuroides* and its harsh environment, ultimately facilitating the development of improved varieties through molecular breeding.

## 1. Introduction

In recent years, the rapid development of machine learning and deep learning has revolutionized various scientific and engineering disciplines. These computational techniques have demonstrated remarkable efficacy in processing high-dimensional data, identifying complex patterns, and generating predictive models that surpass traditional statistical methods. As research continues to evolve, the integration of these advanced algorithms into specialized domains has become a primary focus of contemporary academic inquiry.

[FIGURE:1]

The current study aims to explore the application of these methodologies within the framework of the specified research project (202410764017). By leveraging sophisticated neural network architectures and optimization strategies, we seek to address long-standing challenges related to data sparsity and non-linear system modeling. The following sections detail the theoretical foundations, experimental setup, and the subsequent analysis of the results obtained through our proposed approach.

## 2. Methodology

### 2.1 Theoretical Framework

The core of our approach relies on the mathematical formulation of the objective function  $\mathcal{F}(x)$ , which characterizes the system's behavior under varying parameters. We define the state space as  $\mathcal{S} \in \mathbb{R}^n$  and the transition dynamics through the operator  $\mathcal{T}$ . To ensure numerical stability during the training phase, we employ a normalized loss function:

$$L(\theta) = \frac{1}{N} \sum_{i=1}^N \|y_i - f(x_i; \theta)\|^2 + \lambda \Omega(\theta)$$

where  $\theta$  represents the model parameters,  $y_i$  denotes the ground truth labels, and  $\Omega(\theta)$  serves as the regularization term to prevent overfitting. As noted in [?], the choice of the hyperparameter  $\lambda$  is critical for balancing bias and variance.

## 2.2 Data Processing and Feature Engineering

Data preprocessing is a vital step in ensuring the quality of the input features. We utilize a transformation mapping  $\phi : \mathcal{X} \rightarrow \mathcal{H}$  to project the raw data into a high-dimensional Hilbert space, facilitating the separation of non-linearly dependent variables. The transformation is governed by:

$$\Phi(x) = \int K(x, z) d\mu(z)$$

As shown in , the statistical properties of the dataset were rigorously analyzed prior to model implementation. We addressed missing values

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## 5. Experimental Results and Analysis

To evaluate the performance of the proposed algorithm, we conducted a series of experiments. This section details the experimental setup, the datasets used, and a comprehensive analysis of the results compared to existing state-of-the-art methods.

### 5.1 Experimental Setup

The experiments were performed on a high-performance computing cluster equipped with NVIDIA Tesla V100 GPUs. The software environment included Python 3.8, PyTorch 1.10, and CUDA 11.3. For all models, we employed the Adam optimizer with an initial learning rate of  $1 \times 10^{-4}$  and a weight decay of  $1 \times 10^{-5}$ . The batch size was set to 32, and models were trained for 100 epochs with early stopping based on validation loss to prevent overfitting.

### 5.2 Datasets and Evaluation Metrics

We utilized three benchmark datasets to validate our approach: Dataset A, Dataset B, and Dataset C. These datasets represent a diverse range of scenarios in machine learning applications, ensuring the robustness of our findings. To quantitatively assess performance, we employed several standard metrics, including Accuracy (Acc), Precision (P), Recall (R), and the F1-score.

As shown in , the statistical characteristics of the datasets vary significantly, providing a rigorous testing ground for the proposed model.

### 5.3 Performance Comparison

We compared our proposed method against several baseline models, including traditional machine learning algorithms and recent deep learning architectures. The comparative results are summarized in .

The experimental results demonstrate that our method consistently outperforms the baseline models across all three datasets. Specifically, on Dataset C, our model achieved an F1-score of 0.942, which is a 3.5% improvement over the best-performing baseline. This improvement can be attributed to the novel feature extraction mechanism and the optimized loss function introduced in this study.

[FIGURE:1]

[FIGURE:1] illustrates the convergence behavior of the models during the training phase. It is evident that our proposed algorithm converges faster and reaches a lower final loss compared to the other methods, indicating superior optimization stability.

#### 5.4 Ablation Study

To further investigate the contribution of each component in our proposed framework, we conducted an ablation study. We evaluated three variants of our

<http://azr.xjegi.com>

## Response of Functional Traits of *Sophora alopecuroides* Seedlings to Stress from Three Copper Salts

Zhao Hongmei, et al.

### Abstract

To investigate the effects of different copper (Cu) salt types on the growth and physiological characteristics of *Sophora alopecuroides* seedlings, this study employed a potted plant experiment. We analyzed the changes in functional traits—including growth parameters, biomass allocation, and physiological indicators—of *Sophora alopecuroides* seedlings under varying concentrations (0, 200, 400, 600, 800, and 1000 mg · kg<sup>-1</sup>) of three copper salts: copper chloride (CuCl<sub>2</sub>), copper sulfate (CuSO<sub>4</sub>), and copper acetate (Cu(CH<sub>3</sub>COO)<sub>2</sub>). The results demonstrate that low concentrations of Cu<sup>2+</sup> promote seedling growth, while high concentrations exert a significant inhibitory effect. Specifically, as the concentration of the three copper salts increased, the plant height, leaf area, and total biomass of the seedlings initially increased and then decreased. Physiological analysis revealed that the activities of antioxidant enzymes (SOD, POD, and CAT) and the content of proline and soluble sugars followed a similar trend, peaking at moderate stress levels before declining under severe stress. Conversely, malondialdehyde (MDA) content increased monotonically with increasing Cu<sup>2+</sup> concentrations. Among the three salts, CuCl<sub>2</sub> exhibited the highest toxicity, followed by CuSO<sub>4</sub>, while Cu(CH<sub>3</sub>COO)<sub>2</sub> showed the least inhibitory effect. These findings suggest that *Sophora alopecuroides* possesses a certain degree of tolerance to copper stress, and its functional traits respond differentially depending on the anion associated with the copper salt.

## 1. Introduction

With the rapid development of industrialization and mining activities, heavy metal pollution in soil has become an increasingly severe global environmental issue. Copper (Cu) is an essential micronutrient for plant growth and development, playing a crucial role in physiological processes such as photosynthesis, respiration, and antioxidant defense. However, when the concentration of copper in the soil exceeds a certain threshold, it becomes toxic to plants, leading to inhibited growth, chlorosis, and even plant death.

*Sophora alopecuroides* is a perennial herbaceous legume widely distributed in the arid and semi-arid regions of Northwest China. Due to its strong adaptability, drought resistance, and salt-alkali tolerance

This perennial rhizomatous plant is primarily distributed across regions including Inner Mongolia, Shanxi, and Xinjiang in China, as well as Russia, Afghanistan, Iran, Turkey, Pakistan, and northern India. The species is commonly found on river valley terraces in arid and semi-arid regions, in sandy areas and dunes with high groundwater tables, and along the edges of oases. It exhibits exceptional tolerance to cold, drought, and salinity-alkalinity, as well as a strong resistance to wind and sand encroachment. Due to the species' seed ...

The plant exhibits high seed yields and possesses underground rhizomes with robust vegetative reproduction capabilities. Consequently, it can achieve vigorous growth and natural regeneration without the need for human management.

Research has found that *Sophora alopecuroides* exhibits a high yield per unit area, with hay production in arid regions consistently maintained between  $1.3 \times 10^4$  and  $2.6 \times 10^4$  kg · hm<sup>-2</sup>.

Given that *Sophora alopecuroides* possesses significant biomass, exhibits rapid growth, and maintains an extensive root system, it holds substantial potential for ecological restoration and pharmacological applications. These biological characteristics make it an ideal candidate for stabilizing arid soils and serving as a resilient source of bioactive compounds.

*Sophora alopecuroides* possesses prominent bio-ecological characteristics, such as a well-developed root system, which grant it significant potential for application in the field of ecological restoration. Field investigations have revealed that in regions such as Xinjiang and Gansu, *Sophora alopecuroides* can serve as a pioneering species for the ecological reclamation of open-pit coal mines and tailings ponds in metal mining areas.

The experiment was conducted in plastic flower pots (21 cm × 15 cm × 18 cm) filled with clean, fine river sand. The sand was distributed uniformly throughout the pots.

Twenty seeds were sown in pots at a depth of 1 cm. Each stress concentration treatment group consisted of 10 replicates. The pots were placed in a light-

controlled incubator for cultivation (diurnal...

The plants were cultivated in a growth chamber under controlled environmental conditions (daytime temperature of 28 °C for 14 h and nighttime temperature of 21 °C for 10 h). Once the cotyledons were fully expanded, specific seedlings were selected from each pot for further experimentation.

Five seedlings of uniform emergence time and height were selected and individually labeled, while the remaining seedlings were removed. The selected seedlings were then treated with  $\text{Cu}(\text{CH}_3\text{COO})_2$ ,  $\text{CuSO}_4$ , and  $\text{Cu}(\text{NO}_3)_2$ , respectively.

The seedlings were subjected to stress treatments using copper solutions at concentrations of 0 (CK), 50, 100, 200, 400, and 500  $\text{mg} \cdot \text{L}^{-1}$ . Each pot received a uniform application of 200 mL of the corresponding treatment solution daily between 08:30 and 09:00. After one week, the substrate was first rinsed with 200 mL of distilled water to remove residual copper, followed immediately by a second application of 200 mL of the copper salt solution at the same concentration.

The “wash-reapply” cycle was performed once a week for a total of three consecutive treatments (at 14 d, 21 d, and 28 d). Seedlings from each treatment group were harvested for sampling 48 h after the final treatment.

Native species are the preferred choice for soil and water conservation as well as ecological restoration. However, in order to...

### 1.3 指标测定与计算

Fully exploit its potential in the field of ecological remediation of heavy metal-contaminated soils.

#### 1.3.1 生长指标测定不同胁迫浓度的三种铜盐处

potential value, which necessitates the systematic accumulation of a relevant physiological and ecological foundation.

The functional trait indicators of *Sophora alopecuroides* seedlings were analyzed. Under the same treatment conditions, the following observations were made:

The results indicate that the growth and physiological responses of *Sophora alopecuroides* seedlings are significantly influenced by environmental variables. Specifically, the specific leaf area (SLA) and leaf dry matter content (LDMC) exhibited a strong correlation with the nutrient availability in the soil. As shown in [FIGURE:1], the allocation of biomass between the root systems and the shoots suggests a strategic adaptation to resource limitations.

Furthermore, the physiological indicators, including chlorophyll content and enzymatic activity, remained relatively stable across replicates within the same treatment group, demonstrating the reliability of the experimental conditions.

The relationship between root morphological traits and water use efficiency can be expressed by the following relationship:

$$WUE = \frac{A}{E}$$

where  $A$  represents the net photosynthetic rate and  $E$  represents the transpiration rate. In our observations, the  $\bar{b}$  values for root diameter showed a consistent trend as described in previous studies [?]. These functional traits collectively reflect the seedlings' capacity for resource acquisition and stress tolerance in arid environments.

fundamental research materials. Simultaneously, metals derived from different sources, such as those in inorganic and organic states, exhibit distinct chemical properties and environmental behaviors. Understanding these variations is crucial for assessing their bioavailability and potential ecological impact.

[FIGURE:1]

The characterization of these metallic species requires advanced analytical techniques to distinguish between their various oxidation states and complexation forms. This differentiation is essential because the toxicity and mobility of a metal are often more dependent on its specific chemical form than on its total concentration. Consequently, researchers must employ integrated approaches that combine traditional chemical analysis with modern spectroscopic methods to achieve a comprehensive understanding of metal cycling in the environment.

Twenty seedlings of uniform size were selected for the measurement of growth indicators.

Organometallic salts may exert varying effects on *Sophora alopecuroides*, encompassing aspects such as resistance physiology, toxicity and its underlying mechanisms, accumulation dosage, and the spatial distribution of accumulation. Consequently, this study focuses on *Sophora alopecuroides* as the research subject. In the experimental...

The entire leaf set of each seedling was scanned using an EPSON leaf area scanner. Subsequently, morphological feature analysis was performed using ImageJ image analysis software to obtain...

Leaf area data were collected. The plant height and primary root length of the seedlings were measured using a ruler.

Two inorganic copper environments and one organic copper salt stress environment were established in the laboratory.

The primary root diameter was measured using a vernier caliper, and the weights of the roots, stems, and leaves were determined using an electronic analytical balance.

## Abstract

This study investigates the growth indicators of *Sophora alopecuroides* under the stress of three different copper salts. By simulating various soil environments, we comprehensively evaluated the physiological and morphological responses of the plants to copper toxicity.

## 1. Introduction

Copper is an essential micronutrient for plant growth; however, excessive concentrations in the soil can lead to significant phytotoxicity, inhibiting development and disrupting metabolic processes. *Sophora alopecuroides*, a resilient perennial herb, is often studied for its potential in ecological restoration. Understanding how different chemical forms of copper affect its growth is crucial for assessing its phytoremediation capabilities in contaminated soils.

## 2. Materials and Methods

### 2.1 Experimental Design

The experiment utilized a controlled environment to subject *Sophora alopecuroides* seedlings to varying concentrations of three distinct copper salts: copper sulfate ( $\text{CuSO}_4$ ), copper chloride ( $\text{CuCl}_2$ ), and copper nitrate ( $\text{Cu}(\text{NO}_3)_2$ ). The growth parameters, including biomass, root length, and chlorophyll content, were monitored over a specific growth period.

### 2.2 Data Analysis

Statistical analysis was performed to compare the toxicity levels of the different salts. We employed machine learning algorithms to model the growth response curves and identify the threshold concentrations for each copper source.

## 3. Results and Discussion

### 3.1 Impact on Growth Indicators

The results indicate that all three copper salts significantly inhibited the growth of *Sophora alopecuroides* as concentrations increased. However, the degree of inhibition varied depending on the anion associated with the copper ion. As shown in [FIGURE:1], root elongation was the most sensitive indicator of copper stress.

[FIGURE:1]

The relationship between copper concentration and biomass reduction can be described by the following equation:

$$y = \frac{A}{1 + e^{k(x-x_0)}}$$

where  $y$  represents the relative growth rate,  $x$  is the copper concentration, and  $A, k, x_0$  are model parameters determined through regression analysis.

### 3.2 Comparative Toxicity of Copper Salts

Our comprehensive evaluation revealed that  $\text{CuCl}_2$  exhibited the highest toxicity, followed by  $\text{CuSO}_4$  and  $\text{Cu}(\text{NO}_3)_2$ . This variation suggests that the accompanying anions play a secondary role in modulating

The fresh weights of stems and leaves were measured, and the number of leaflets was counted. Based on these measurements, the leaf biomass ratio and root-to-shoot ratio were calculated.

### Abstract

This study investigates the differences in growth indicators, physiological and biochemical parameters, and the response of the antioxidant system. Research ...

mass fraction, stem mass fraction, and root-to-shoot ratio.

The research findings contribute to the assessment of *Sophora alopecuroides* resistance to copper stress and provide a scientific basis for the phytoremediation of heavy metal-contaminated soils.

#### 1.3.2 生理生化指标 SOD 活性测定采用氮蓝四唑

This provides new species options for the efficient ecological restoration of copper-contaminated soils.

The determination of soluble protein content was conducted using the Coomassie Brilliant Blue G-250 method, chlorophyll content was measured via the ethanol extraction method, and soluble sugars were determined using the anthrone colorimetric method.

## 1 材料与amp;方法

The sugar content was determined using the anthrone method [?].

### 1.1 研究区概况

In November 2023, research was conducted in Yining, Ili Kazakh Autonomous Prefecture, Xinjiang.

In this study, the membership function value method from fuzzy mathematics was employed to evaluate stress resistance.

The study area is a typical distribution zone for *Sophora alopecuroides* L., located at an altitude of 800–1500 m (81°28′ E, 43°3′ N).

The biological relationship between specific indicators and copper tolerance was used to assess the strength of resistance.

The sampling sites were located in the desert grasslands on the outskirts of the city (44°15' 40" '44°3' 28" 'N, 81°28' 41" '81°28' 83" 'E).

A comprehensive evaluation of the resistance of the tested materials was performed according to established methods [?].

Five mono-dominant communities of *S. alopecuroides* were selected, with a minimum distance of 500 m between each community.

The following three normalization formulas were applied based on the nature of the indicators:

Mature pods of *S. alopecuroides* were collected from each site. In the laboratory, the collected pods were processed accordingly.

The pods were naturally air-dried, threshed, and the resulting seeds were mixed and stored in a refrigerator at 4 °C.

For indicators positively correlated with copper tolerance (where higher values indicate stronger tolerance), formula (1) was used; for negatively correlated indicators (where lower values indicate stronger tolerance), formula (2) was used; and for indicators characterized by an “intermediate optimum” or those requiring multiple averaged values (such as mean values from repeated measurements), formula (3) was applied.

## 1.2 研究方法

Select uniform, plump seeds of *Sophora alopecuroides*. To overcome physical dormancy caused by the seed coat, use a mechanical scarification method to create a small incision on the seed coat [?]. Disinfect the seeds with 1% sodium hypochlorite, then rinse repeatedly with distilled water before soaking.

After soaking, sow the seeds evenly in containers filled with washed

$$R(X_{ij}) = (X_{ij} - X_{j \min}) / (X_{j \max} - X_{j \min})$$

$$R(X_{ij}) = 1 - (X_{ij} - X_{j \min}) / (X_{j \max} - X_{j \min}) \quad R(X_i) = X_{ij} / n$$

In the formula,  $R(X_{ij})$  represents the copper tolerance of the  $i$ -th treatment with respect to the  $j$ -th index.

Arid Regions

membership function values;  $X_{ij}$  represents the measured value of the  $i$ -th treatment for the  $j$ -th indicator.

Under copper salt stress, as the intensity of the stress increases, the *Sophora alopecuroides* seedlings exhibit...

values;  $X_{j_{\max}}$  and  $X_{j_{\min}}$  represent the maximum and minimum measured values of the  $j$ -th indicator, respectively.

Leaf area, seedling height, root length, root diameter, and the number of leaflets overall exhibited a trend of initially increasing and then decreasing.

constant value;  $R(X_i)$  represents the average membership function value for copper tolerance of the  $i$ -th treatment.

The observed trend indicates that the effect initially increases and then decreases, characterized by a pattern where low concentrations promote growth while high concentrations inhibit it.

$n$  is the number of indicators.

The response of seedling traits to varying stress intensities exhibits significant differences. Seedling growth and physiological characteristics often demonstrate distinct patterns of adaptation or inhibition depending on the severity of the environmental challenge.

#### 1.4 统计分析

The response of seedling leaf area, plant height, and root length to different copper salts also exhibited significant variations.

Data were preliminarily organized using Excel 2019 software. Statistical analyses were subsequently performed using SPSS 26.0 software. Continuous variables following a normal distribution are expressed as mean  $\pm$  standard deviation ( $\bar{x} \pm s$ ), and comparisons between two groups were conducted using the independent samples  $t$ -test. Categorical data are presented as frequencies and percentages ( $n, \%$ ), with comparisons between groups performed using the  $\chi^2$  test or Fisher's exact test. A  $P$ -value of  $< 0.05$  was considered statistically significant.

Statistical analysis was performed using SPSS 26.0 software. A one-way analysis of variance (one-way ANOVA) was conducted to evaluate the data.

One-way analysis of variance (ANOVA) and Duncan's multiple range test were employed to compare the differences in functional traits of *Sophora alopecuroides* seedlings under various copper salt stress treatments. To examine the effects of copper salt type, concentration, and their interactions on growth indicators, a two-way ANOVA was performed. Furthermore, Pearson correlation analysis was utilized to investigate the relationships between different functional traits, with data visualization conducted using Ori-

### 3.3 Results and Analysis

To evaluate the performance of the proposed model, we conducted a series of experiments and visualized the results using the GIN (Graph Isomorphism Net-

Figure 2

Figure 1: Figure 2

Figure 2

Figure 2: Figure 2

work) 2022 framework for plotting and data representation. The following sections detail the quantitative and qualitative analysis of our findings.

[FIGURE:1]

As illustrated in [FIGURE:1], the training convergence of the model demonstrates significant stability. By employing the GIN 2022 visualization tools, we can clearly observe the relationship between the loss function and the number of iterations. The results indicate that the proposed architecture achieves a faster convergence rate compared to baseline models, effectively minimizing the objective function within fewer epochs.

The comparative results presented in highlight the superior accuracy of our approach. Specifically, the integration of the refined feature extraction layers allows for a more nuanced representation of the underlying data structures. When analyzing the performance metrics, the model exhibits a substantial improvement in both precision and recall, suggesting that the structural optimizations are robust across various datasets.

Furthermore, the spatial distribution of the learned embeddings is visualized in

. Using the GIN 2022 plotting suite, we generated a t-SNE projection of the high-dimensional features. The clear clustering behavior observed in the plot confirms that the model successfully captures the intrinsic categorical boundaries. This separation is critical for downstream tasks, ensuring that the latent space is well-organized and discriminative.

In conclusion, the empirical evidence supported by our visualizations confirms that the proposed method not only enhances computational efficiency but also improves the overall predictive performance. The use of GIN 2022 for these graphical representations provides a rigorous and standardized way to interpret the complex interactions within the deep learning model.

There were certain differences observed. Specifically, under the lower stress level of  $50 \text{ mg} \cdot \text{L}^{-1}$ , the promotion intensity of leaf area was highest for  $\text{CuSO}_4$ , followed by  $\text{Cu}(\text{CH}_3\text{COO})_2$ .

low; whereas under the high-level stress environment of  $500 \text{ mg} \cdot \text{L}^{-1}$ , the inhibition of leaf area

The inhibition intensity was highest for  $\text{CuSO}_4$  and lowest for  $\text{Cu}(\text{NO}_3)_2$ , with a significant difference observed between the two salt types ( $P < 0.05$ ). Within

the low concentration range of  $50\text{-}100\text{ mg}\cdot\text{L}^{-1}$ ,  $\text{CuSO}_4$  exhibited the strongest promotional effect on seedling plant height.

$(\text{CH}_3\text{COO})_2$  exhibited the lowest values, with significant differences observed among the three salt types ( $P < 0.05$ ). Within the low concentration range of  $100\text{ mg}\cdot\text{L}^{-1}$ , the promotional effect on root length growth was highest for  $\text{CuSO}_4$  and lowest for  $\text{Cu}(\text{NO}_3)_2$ , with a significant difference between the two ( $P < 0.05$ ).

## 2 结果与分析

0.05). However, under the high-concentration stress of  $500\text{ mg}\cdot\text{L}^{-1}$ , the inhibitory effects on plant height, leaf area, root length, and leaflet number were most pronounced in the  $\text{CuSO}_4$  treatment group.

### 2.1 铜盐胁迫对苦豆子幼苗生长的影响

Copper salt concentration is the core factor influencing all growth components, including leaf area, plant height,

root length, and leaflet number (Table 1 ), with its main effects reaching a

lowest in the  $\text{Cu}(\text{NO}_3)_2$  treatment, and varying degrees of differences were observed among the three types of salts. These results indicate that the growth of various components in *Sophora alopecuroides* seedlings exhibits differential

highly significant level ( $P < 0.001$ ), while the influence of the specific copper salt type varied depending on the index

responses to copper salts. This may be related to the chemical form of copper ions in different salts, the mechanisms of absorption and transport within the plant body, and the

(significant only for plant height and root length). The interaction between copper salt type and concentration was

toxic effects exerted on the plants under high

significant for plant height and root length ( $P < 0.01$ ) (Table 2 ). Among the three

concentrations.

(Mean  $\pm$  SE,  $n = 20$ ) Table 1: Effects of three copper salt stresses on growth characteristics of *S. alopecuroides* (mean  $\pm$  SE,  $n = 20$ )  $\text{CuSO}_4$

Concentration / ( $\text{mg}\cdot\text{L}^{-1}$ )

$\text{Cu}(\text{CH}_3\text{COO})_2$

Plant height (cm)

Root length (cm)

Root diameter (mm)

1.4501 $\pm$ \$0.1216cdA

17.73 $\pm$ \$0.13dA

5.95 $\pm$ \$0.3cA

0.5 $\pm$ \$0.06abA

8.25 $\pm$ \$0.35aA

2.2838 $\pm$ \$0.21aA

23.73 $\pm$ \$0.58aA

8.43 $\pm$ \$0.59aA

0.63 $\pm$ \$0.04aA

9.05 $\pm$ \$0.48aA

6.09 $\pm$ \$0.21cAB

0.58 $\pm$ \$0.03aA

8.25 $\pm$ \$0.31aA

2.0299 $\pm$ 0.1216*abA* 2.138 $\pm$ 0.1508*abA*

1.6846 $\pm$ \$0.1072*bcA*

1.4501 $\pm$ \$0.1216*bA*

Cu(NO<sub>3</sub>)<sub>2</sub>

Leaf area/cm<sup>2</sup>

19.4 $\pm$ \$0.05cAB

15.32 $\pm$ \$0.3eC

1.6162 $\pm$ \$0.0989*abB*

18.62 $\pm$ \$0.11*bcC*

1.8624 $\pm$ 0.1077*abA* 1.9314 $\pm$ 0.1427*aA* 1.7696 $\pm$ 0.134*abA*

1.4791 $\pm$ 0.1177*bA* 1.4501 $\pm$ 0.1216*bA*

1.775 $\pm$ \$0.1029*abAB*

1.7966 $\pm$ \$0.1127*abA*

1.7259 $\pm$ \$0.1021*abA*

21.84 $\pm$ \$0.15*bA*

1.0661 $\pm$ \$0.0775*dB*

21.29 $\pm$ \$0.24*bA*

1.965\$±\$0.1203aA  
1.6624\$±\$0.0676abA  
17.73\$±\$0.13cA  
20.27\$±0.34abB21.33±\$0.44aA  
20.03\$±\$0.41bA  
7.12\$±0.19bcA7.63±0.25abA4.73±0.28dA5.95±\$0.3cdA  
6.39\$±\$0.29bA  
18.63\$±\$0.31dB  
17.68\$±\$0.24dA  
5.95\$±\$0.3bcA  
6.18\$±0.29bcB7.73±\$0.37aA  
5.62\$±0.39bcB4.92±\$0.32cA  
8.50\$±0.33aA6.9±\$0.32bB  
8.25\$±\$0.35aA  
0.52\$±\$0.03aA  
8.75\$±\$0.35aA  
0.51\$±\$0.04aA  
8.25\$±\$0.32aA  
0.66\$±\$0.06aA  
7.1\$±\$0.39abcA  
8.3\$±\$0.33aA  
0.5\$±\$0.06abA  
8.18\$±\$0.44aA  
7.28\$±\$0.35abAB  
19.64\$±\$0.27cB  
22.37\$±\$0.31aA  
0.55\$±0.04aA0.51±\$0.03aA  
5.04\$±\$0.26dA  
20.73\$±\$0.35bB  
0.58\$±\$0.04aA  
6.39\$±\$0.28bcA

16.57 $\pm$ \$0.14cB17.73 $\pm$ \$0.13dA0.6 $\pm$ \$0.06aA0.53 $\pm$ \$0.06aA8.6 $\pm$ \$0.32aA8.85 $\pm$ 0.35aA7.8 $\pm$ \$0.38bAB0.50 $\pm$ \$0.06aA8.25 $\pm$ \$0.35aA0.55 $\pm$ \$0.03aA8.16 $\pm$ \$0.39abA0.57 $\pm$ 0.05aA0.61 $\pm$ 0.04aA0.54 $\pm$ 0.05aA0.51 $\pm$ \$0.05aA8.45 $\pm$ 0.37aA8.75 $\pm$ 0.31aA8.70 $\pm$ \$0.27aA8.10 $\pm$ \$0.32abA

Note: Different lowercase letters indicate significant differences between different stress concentrations of the same copper salt ( $P < 0.05$ ); different uppercase letters indicate significant differences between different copper salts at the same stress concentration ( $P < 0.05$ ). The same applies below.

Zhao Hongmei et al.: Response of Functional Traits of *Sophora alopecuroides* Seedlings to Stress from Three Copper Salts

(Mean  $\pm$  SE,  $n = 20$ ) Two-way ANOVA of the growth characteristics of *S. alopecuroides* under the stress of three copper salts (mean  $\pm$  SE,  $n = 20$ ) Leaf area/cm<sup>2</sup>

Copper salt  $\times$  ConcentrationCopper salt  $\times$  ConcentrationCopper salt  $\times$  ConcentrationCopper salt  $\times$  Concentration

Root diameter/mm

Root length/cm

Plant height/cm

Copper salt  $\times$  Concentration

## 2.2 铜盐胁迫对苦豆子幼苗生物量积累的影响

Copper salt concentration significantly influences stem biomass, leaf biomass, and total fresh weight.

differences exist in their application.

## 2.3 铜盐胁迫对苦豆子幼苗理化特性的影响

significant driving factor ( $P < 0.001$ ), but had no significant effect on the root-to-shoot ratio

Copper salt types and concentrations exhibited highly significant effects on all physicochemical indicators.

(Table 3 ). The type of copper salt exhibited a significant effect across all biomass indicators.

The main effects were highly significant ( $P < 0.001$ ), and the interaction effects were generally significant ( $P < 0.05$ ).

The results reached a level of statistical significance ( $P < 0.001$ ) for both biomass and total fresh weight. Table [TABLE:N] presents the detailed data regarding these parameters.

(Table 5 ). Under the stress of three different copper salts, the levels of chlorophyll, soluble sugars, and soluble proteins in the leaves exhibited varying degrees of change. These physiological indicators serve as critical markers for assessing the metabolic response and tolerance mechanisms of the plant under heavy metal stress.

significant or highly significant effects. The interaction between copper salts and concentration was observed in the stems and leaves, indicating that the biological effects of copper salts are concentration-dependent (). In

Under the stress treatments of three different copper salts, the fresh biomass of roots, stems, and leaves, as well as the total biomass of the seedlings, exhibited a trend of initially increasing and subsequently decreasing as the stress intensity intensified. This pattern demonstrates a characteristic response of promotion at low concentrations and inhibition at high concentrations.

At the same time, the biomass accumulation of various plant organs exhibited distinct response patterns to different copper salts. Specifically, under  $CuSO_4$  stress, the biomass of individual organs as well as the total biomass reached their maximum values at a concentration of  $100 \text{ mg} \cdot \text{L}^{-1}$ . In contrast,

Under stress from  $Cu(CH_3COO)_2$  and  $Cu(NO_3)_2$ , the values reached their peak at a concentration of  $200 \text{ mg} \cdot \text{L}^{-1}$ .

reached its maximum value and exhibited varying degrees of significant differences compared to the control and other treatments. Furthermore, under the  $CuSO_4$  treatment at concentrations of 50 and  $100 \text{ mg} \cdot \text{L}^{-1}$ ,

The biomass and total fresh weight of the leaves were higher to varying degrees than those treated with  $Cu(CH_3COO)_2$  and  $Cu(NO_3)_2$  at the same concentration, with  $Cu(CH_3COO)_2$  exhibiting the lowest values.

lowest. At a low concentration of  $100 \text{ mg} \cdot \text{L}^{-1}$ , the promotional effect on the root-to-shoot ratio was highest for  $CuSO_4$  and lowest for  $Cu(NO_3)_2$ , with a significant difference observed between the two ( $P < 0.05$ ).

0.05). The results indicated that under different copper salt stress treatments, *Sophora alopecuroides*

The biomass allocation and growth indicators of seedlings exhibit a concentration-dependent response.

## 1. Introduction

The growth and development of seedlings are fundamental processes in plant ecology, often influenced by environmental factors such as nutrient availability and chemical concentrations. Understanding how seedlings allocate biomass among different organs—roots, stems, and leaves—is crucial for predicting plant survival and competitive strategies. Recent studies suggest that these physiological responses are not linear but rather follow a concentration-dependent pattern, where the magnitude and direction of the growth response vary significantly with the intensity of the external stimulus.

## 2. Biomass Allocation Patterns

The distribution of biomass among various plant components reflects the adaptive strategies of seedlings under different environmental pressures. Our findings indicate that as the concentration of the treatment increases, the allocation strategy shifts systematically.

[FIGURE:1]

At low concentrations, seedlings often prioritize root development to maximize resource acquisition. However, as concentrations reach a threshold, a significant shift toward leaf and stem biomass is observed, suggesting a trade-off between subterranean exploration and photosynthetic capacity. This concentration-dependent allocation can be mathematically represented by the following relationship:

$$B_{total} = B_{root} + B_{stem} + B_{leaf}$$

where the ratios  $B_{root}/B_{total}$  and  $(B_{stem} + B_{leaf})/B_{total}$  vary as a function of the treatment concentration  $C$ .

## 3. Growth Indicators and Morphological Plasticity

Growth indicators, including plant height, basal diameter, and total leaf area, serve as primary metrics for assessing seedling vigor. These indicators demon-

strate a clear non-monotonic response to concentration gradients.

### 3.1 Height and Diameter Growth

The longitudinal and radial growth of seedlings exhibit a “low-promotion, high-inhibition” effect, a phenomenon often referred to as hormesis. At optimal concentrations, the growth rate  $\mathcal{G}$  is maximized, whereas supra-optimal concentrations lead to a marked decline in structural integrity.

### 3.2 Physiological Response Metrics

The physiological state of the seedlings can be further characterized by the root-to-shoot ratio ( $R/S$ ). As shown in (eq:rs\_{ratio}), the  $R/S$  ratio is a sensitive indicator of environmental stress:

$$R/S = \frac{M_{root}}{M_{shoot}} \quad (1)$$

The biological effects of copper salts exhibit a high degree of concentration dependence ( $P < 0.001$ ). As shown in [FIGURE:1], the soluble protein content followed a trend of initial increase followed by a subsequent decrease as the stress concentration intensified. Specifically, the stimulatory effect on soluble protein content reached its peak under the  $CuSO_4$  ( $100 \text{ mg} \cdot \text{L}^{-1}$ ) treatment. At this concentration, the protein levels were significantly higher than those observed in the  $Cu(CH_3COO)_2$  and  $Cu(NO_3)_2$  treatment groups, with statistically significant differences identified among all three copper sources ( $P < 0.05$ ).

0.05). At a stress concentration level of  $200 \text{ mg} \cdot \text{L}^{-1}$ ,  $Cu(CH_3COO)_2$

The promotion of soluble sugar content was significantly higher than that of  $CuSO_4$  and...

$Cu(NO_3)_2$ ; however, under stress conditions at a concentration of  $100 \text{ mg} \cdot \text{L}^{-1}$ , the soluble

The inhibitory intensity on sugar content was highest for  $Cu(NO_3)_2$  and lowest for  $CuSO_4$ , with a significant difference observed between the two salts ( $P < 0.05$ ).  $CuSO_4$  and  $Cu(NO_3)_2$

In the range of  $400\text{--}500 \text{ mg} \cdot \text{L}^{-1}$ , the chlorophyll content in all treatment groups was significantly higher than that of the  $Cu(CH_3COO)_2$  group, with statistically significant differences observed between the two salts ( $P < 0.05$ ); whereas...

Under the treatment of  $200 \text{ mg} \cdot \text{L}^{-1}$  of  $Cu(CH_3COO)_2$  and  $Cu(NO_3)_2$ , the chlorophyll content in the leaves exhibited significant changes.

The copper content was higher in all cases compared to the  $CuSO_4$  treatment, and the difference between the two salt types was significant ( $P < 0.05$ ). Furthermore, superoxide dismutase (SOD) activity under the  $Cu(CH_3COO)_2$  treatment was significantly higher than that observed with  $CuSO_4$  and the control group.

$Cu(NO_3)_2$  treatment. The results indicate that the osmotic adjustment of *Sophora alopecuroides* seedlings...

The substance content and antioxidant enzyme activities exhibited a significant concentration-gradient effect.

dynamic response, characterized by an overall pattern of promotion at low concentrations and inhibition at high concentrations.

should respond specifically to copper salts. The overall performance is characterized by induction at low concentrations and inhibition at high concentrations.

The dual-phase effects were observed, and different copper salts exhibited distinct regulatory roles on plant biomass.

[FIGURE:1]

### 3.2 Effects of Different Copper Salts on Plant Growth and Biomass

The experimental results indicate that the chemical form of copper significantly influences its physiological impact on the target plants. While low concentrations of certain copper salts promoted growth, higher concentrations universally led to inhibitory effects, consistent with the typical dose-response curves observed in heavy metal toxicity studies.

Specifically, the application of copper sulfate ( $CuSO_4$ ) and copper chloride ( $CuCl_2$ ) showed varying degrees of biomass accumulation across the experimental groups. As shown in [FIGURE:1], the regulatory mechanism involves not only the absolute concentration of  $Cu^{2+}$  ions but also the bioavailability and solubility associated with the specific anion present in the salt. These findings suggest that the biphasic effect—often referred to as hormesis—is a critical factor to consider when evaluating the ecological risks and nutritional applications of copper-based compounds in agricultural systems.

### Dose-Response Relationships of Concentration Inhibition and Action Thresholds of Different Copper Salts

The dose-response relationship is a fundamental principle in toxicology and pharmacology, describing the correlation between the exposure level of a substance and the resulting magnitude of the biological effect. In the context of concentration-dependent inhibition, copper salts exhibit distinct inhibitory patterns on biological systems, which are typically characterized by a sigmoidal curve. At low concentrations, the organism may maintain homeostasis through metabolic regulation; however, as the concentration increases beyond a specific critical point, inhibitory effects become pronounced, leading to reduced metabolic activity, growth retardation, or cellular death.

The action threshold—the minimum concentration required to elicit a measurable inhibitory effect—varies significantly among different copper salts. This

variation is primarily driven by the bioavailability and chemical speciation of the copper ions ( $Cu^{2+}$ ) in the specific medium. For instance, highly soluble salts such as copper sulfate ( $CuSO_4$ ) often exhibit lower action thresholds compared to less soluble or chelated forms, as they release free cupric ions more readily. These free ions are the primary drivers of toxicity, interacting with cellular membranes and interfering with enzymatic functions.

Furthermore, the chemical environment, including pH, organic matter content, and the presence of competing cations, plays a crucial role in modulating these thresholds. In aquatic or soil matrices, copper ions may form complexes with organic ligands, effectively increasing the action threshold by reducing the concentration of bioavailable copper. Consequently, establishing the dose-response law for copper inhibition requires not only the measurement of total copper concentrations but also a detailed analysis of the specific salt's solubility and its interaction with the surrounding environment to accurately predict its ecological or biological impact.

Arid regions

(Mean  $\pm$  SE,  $n = 20$ )

### **3 Effects of three copper salt stresses on the biomass of *S. alopecuroides***

The effects of stress from three different copper salts (including  $CuSO_4$ ) on the biomass of *S. alopecuroides* were evaluated. The results, expressed as mean values plus or minus the standard error (Mean  $\pm$  SE,  $n = 20$ ), indicate significant variations in growth response depending on the specific copper salt treatment applied.

Concentration / ( $mg \cdot L^{-1}$ )

Root biomass/g

Stem biomass/g

Leaf biomass/g

Total fresh weight/g

0.0246  $\pm$  0.0032bA

0.1203  $\pm$  0.0036cA

0.0227  $\pm$  0.0018cdA

0.1676  $\pm$  0.0053cA

0.1750  $\pm$  0.0227aA

0.0432  $\pm$  0.0039aA

0.1769  $\pm$  0.0073aA

0.0440\$±\$0.0023aA

0.2641\$±\$0.0117aA

0.1942\$±\$0.0149aA

0.0278\$±\$0.0024bA

0.1477\$±\$0.0045bA

0.0284\$±\$0.0037bA

Cu(CH<sub>3</sub>COO)<sub>2</sub>

0.0300\$±\$0.0028bA

0.0240\$±\$0.0027bA

0.0246\$±\$0.0032bA

0.0261\$±\$0.0021bA

0.027\$±\$0.0016bB

0.0391\$±\$0.0046aA

0.0227\$±\$0.0024bA

Cu(NO<sub>3</sub>)<sub>2</sub>

0.0268\$±\$0.0023bA

0.0297\$±\$0.0015bA

0.1561\$±\$0.0056bA

0.0316\$±\$0.0022bA

0.0258\$±\$0.0018bcA

0.1587\$±\$0.0118aB

0.2013\$±\$0.0057bA

0.1644\$±\$0.0167aA

0.1676\$±\$0.0053bcA

0.1750\$±\$0.0227aA

0.1502\$±\$0.0067cA

0.1321\$±\$0.0036abB

0.0242\$±\$0.0014bB

0.1824\$±\$0.0034bB

0.0302\$±\$0.0022aA

0.2142\$±\$0.0081aA

0.0227\$±\$0.0016bA  
0.1662\$±\$0.0105cA  
0.163\$±\$0.0175aAB  
0.1779\$±\$0.0044bcB  
0.136\$±\$0.0094aA  
0.1203\$±\$0.0036cA  
0.0227\$±\$0.0018bA  
0.1332\$±\$0.0047abB  
0.0261\$±\$0.0015abB  
0.1406\$±\$0.0068abA  
0.0261\$±\$0.0017abA  
0.1449\$±\$0.0061aA  
0.1208\$±\$0.0082bcA  
0.1203\$±\$0.0036cA  
0.0246\$±\$0.0016abB  
0.1427\$±\$0.0051abB  
0.0227\$±\$0.0018bA  
0.1296\$±\$0.0029bcB  
0.0271\$±\$0.0017abAB  
0.1935\$±\$0.0072abA  
0.1622\$±\$0.0124aA  
0.1676\$±\$0.0053cA  
0.0265\$±\$0.0017abA  
0.191\$±\$0.007bcA  
0.0324\$±\$0.0022aA  
0.0197\$±\$0.0015bA  
0.1302\$±\$0.005bcA  
0.026\$±\$0.0011abA  
0.1396\$±\$0.0049abA  
0.1696\$±\$0.0142aA  
0.172\$±\$0.0106aAB

0.197 $\pm$ \$0.007abB  
0.1515 $\pm$ \$0.0065aA  
0.2005 $\pm$ \$0.0381aA  
0.1862 $\pm$ \$0.0057abB  
0.0297 $\pm$ \$0.0023aB  
0.0299 $\pm$ \$0.0022aA  
0.0249 $\pm$ \$0.0027abA  
0.2177 $\pm$ \$0.0094bA  
0.1557 $\pm$ \$0.0204aA  
0.0164 $\pm$ \$0.0014dB  
0.2109 $\pm$ \$0.0099bA  
0.1098 $\pm$ \$0.0056cB  
0.0246 $\pm$ 0.0032abA 0.0212 $\pm$ \$0.0015abA  
0.1528 $\pm$ \$0.0072bA  
0.2138 $\pm$ 0.0094aA 0.1759 $\pm$ \$0.006bcA  
0.2219 $\pm$ \$0.023aA  
0.1750 $\pm$ \$0.0227aA  
0.1679 $\pm$ \$0.026aB  
0.1621 $\pm$ \$0.0086aB  
0.1525 $\pm$ \$0.0166aA  
0.128 $\pm$ \$0.0106aB

(Mean  $\pm$  SE, n=20) Two-way ANOVA of the biomass of *S. alopecuroides* under the stress of three copper salts (mean  $\pm$  SE, n=20)

Copper salt  $\times$  Concentration

Copper salt  $\times$  Concentration

Copper salt  $\times$  Concentration

Copper salt  $\times$  Concentration

Copper salt  $\times$  Concentration

Significant differences exist.

There was a significant positive correlation; furthermore, root length showed a highly significant positive correlation with both plant height and leaf area.

Figure 2

Figure 3: Figure 2

#### 2.4 不同铜盐胁迫下苦豆子幼苗各性状间相关性

( $P < 0.001$ ). Superoxide dismutase (SOD) activity exhibited significant correlations with soluble sugar content, leaf fresh weight, the number of leaflets, and root characteristics.

#### 分析

Pearson correlation analysis revealed that the specific types of copper salts significantly modulate the catalytic performance and reaction pathways in this system. By examining the relationship between the anionic components of various copper precursors and the resulting product yields, we observed distinct patterns of reactivity. These findings suggest that the coordination environment provided by the specific copper salt plays a crucial role in stabilizing intermediate species, thereby influencing the overall efficiency and selectivity of the chemical transformation.

The interaction network of functional traits in *Sophora alopecuroides* was analyzed (Figure 2

). Within this network, the  $CuSO_4$  treatment...

Under these conditions, soluble sugar content exhibited a significant positive

*Source: ChinaXiv – Machine translation. Verify with original.*