

Proceedings of 8th International Conference on

Selenium in the Environment and Human Health

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Forward

Based upon the overwhelming success at previous seven International Conferences on Selenium in the Environment and Human Health (ICSEHH), including the 1st Conference held in Suzhou, China in 2009, the 2nd Conference held in Suzhou, China in 2011, the 3rd Conference held in Hefei, China in 2013, the 4th Conference held in Sao Paulo, Brazil in 2015, the 5th Conference held in Stockholm, Sweden in 2019, the 6th Conference held in Xi'an, China in 2019, and the 7th Conference held in Bangkok, Thailand in 2023, the 8th ICSEHH will be held at Sheraton Hotel in Chuzhou, China.

The conference series has created and provided an excellent platform for scientists, particularly environmental health and biological professionals working on all aspects of selenium across multitude disciplines to present their state-ofthe-art research findings, exchange research ideas, and initiate future collaborative research programs. We selected Chuzhou as the venue of the 8th Conference since it has become one of the major selenium research centres in China during recent years. While acknowledging the existence of both selenium-deficient and selenium-rich soils in China, the world community of selenium researchers will converge in Chuzhou and create a new vision on selenium research and application direction and accomplishment. It will be an inspiring event, thanks to a high number of young participants and experienced researchers, both nationally and internationally, who are eager to openly present and discuss their most recent results and concepts. The International Society for Selenium Research (ISSR) extends its gratitude and sincere appreciation to Anhui Science and Technology University, their faculty and student volunteers for their great work and enthusiasm. This conference is a result of group participation from all of us on a great subject -"selenium", which is taking place in a fantastic location in Chuzhou, China.

We hope that this 8th conference will become another great opportunity for worldwide participants who can discuss strategies for improving selenium's beneficial impacts on the environment and human health. We have all the components needed to have another successful conference in China, as well as selenium-enriching experience for all. Enjoy and learn from each other, get to know each other and celebrate together. Please remember our Selena's favourite slogan – "Long Live Selenium and Live Long with Selenium."

Lutz Schomburg

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A 17-year of Research and Practices of Se-Functional Agriculture in China (2008-2025): Framework, Progress and Future Cooperation

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Introduction: Academician Zhao Qiguo proposed a new concept of agricultural development- functional agriculture in 2008 to solve the global "hidden hunger" problems ^[1,2]. However, studies on the global development trend of functional agriculture reveal that selenium (Se)-functional agriculture is confronted with several prominent issues. These include a lack of unified standardization across soil systems, production technical regulations, Se product systems, and testing systems, as well as inadequate research and development of intelligent and precise regulation technologies. Consequently, these issues have severely impeded the development of the selenium-rich industry.

Methods: This study was based on two villages (Pandside in Chuzhou, Big Hill in Shitai county) with different soil Se levels. We analyzed Se transmission patterns in the soil-crop-food-human chain to modify precisionally the biofotification of several crops in Pandside village in Chuzhou and a Se-rich village in Shitai county. Centering on the links of "land - fertilizer - breeding - cultivation and breeding - harvesting and storage - processing - marketing - health", we will establish a comprehensive innovation chain for the of selenium - enriched functional agriculture, ensuring standardized process specifications and intelligent process control.

Results and Discussion: In response to the needs of the innovation chain in selenium - enriched functional agriculture, we have established the FAST multi-level standardized technology system. This system encompasses precision fertilization technologies, which can be applied to a variety of crops such as rice, wheat, corn, and tea. Additionally, we have carried out research on basic fertilizers and basic food materials. In terms of standard setting, we have established a series of national and industrial standards. For example, these include "Classification of Soil Selenium Content" (GB/T 44971 - 2024), "Determination of Five Selenium Species in Agricultural Products" (GH/T 1429 - 2023), "Selenium - Enriched Agricultural Products" (GH/T 1135 - 2024), and "Technical Specification for Traceability Operation of Quality and Safety of Functional Agricultural Products" (DB 13/T 5892 - 2024). Furthermore, we have actively expanded collaborative partnerships, jointly creating the FAST Shennong Model for intelligent functional agriculture with China Telecom. We have also joined forces with the First People's Hospital of Chuzhou to launch a medical and health evaluation project for functional agriculture. In terms of equipment development, we have introduced a range of FAST - specific equipment including the FAST 3D Food Printer and the FAST Mineral Rapid Tester.

Conclusions: This study of natural and modified mineral transmission patterns in the soil-crop-food-body chain provides new perspectives and methods for the development of technology on functional agriculture. We are looking forward to collaborating with interested scientific groups worldwide in the field of functional agriculture innovation chains!

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Genetic Approaches to Biofortify Selenium in Rice

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Rice is a staple food for over half of the global population. However, approximately three quarters of the rice grains produced globally have insufficient levels of selenium (Se) to meet human's requirement. Two examples will be presented in this talk to demonstrate the potential of genetic modification for Se biofortification in rice. Using forward genetic approaches, we have identified two key genes regulating sulphur (S) and Se assimilation in rice. The serine hydroxymethyltransferase OsCADT1 is a negative regulator of S and Se uptake and assimilation. Its mutation leads to enhanced accumulation of S and Se in rice grain with little effect on plant growth and grain yield. OsASTOL1^{S189N} is a mutated gain-of-function cysteine synthase that markedly increases S uptake and biosynthesis of cysteine. Expression of OsASTOL1^{S189N} driven by a suitable promoter from rice enhanced the accumulation of S, Se, iron (Fe) and zinc (Zn), while simultaneously decreasing arsenic (As), in rice grain, with no adverse effect on grain yield.

New Insights into Selenium Biofortification to Scavenge ROS and Increase Yield of Crop Species

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Introduction: Selenium (Se) can be a benefit element for plants related to several physiological and biochemical characteristics that help to mitigate different types of abiotic stresses. Selenoproteins act as powerful antioxidants in plant metabolism through glutathione peroxidase (GSH) pathway, and provide an increased activity for enzymatic (SOD, CAT, and APX) and non-enzymatic (carotenoids, flavonoids, and tocopherols) compounds that act in reactive oxygen species (ROS) scavenging system and cell detoxification ^[1]. Foliar Se application at low concentration can increase antioxidant metabolism, photosynthesis, sugar translocation from source to sink. Better carbon allocation enhances flavonoid biosynthesis increasing nodulation of legume plants ^[2] and stimulates plant growth and yield of several crop species like grapes, coffee ^[3], sugarcane ^[4], and maize plants ^[5].

Methods: The experiments were carried out under greenhouse conditions for maize plants, and field trials for grapes, cowpea, coffee, sugarcane, soybean and peanuts. Foliar Se application was performed at 6 different rates (0, 10, 20, 40, 50 and 60 g/ha) using sodium selenate or Se-bis-glycinate as Se sources.

Results and Discussion: Selenium application reduced lipid peroxidation measured by lower malondialdehyde and hydrogen peroxide content in leaf tissues of all analyzed crop species. This reduction of lipid peroxidation was most pronounced at Se rate ranging from 20 to 50 g/ha mainly governed by increased carotenoids, chlorophylls and boosted enzymatic antioxidant metabolism triggered by superoxide dismutase, catalase, peroxidase and glutathione peroxidase activities. Scavenged ROS resulted in greener leaves of plants due to higher chlorophyll and photosynthesis rates resulting in better carbon translocation from leaves to fruits or seeds in these studied plant species. Foliar Se application at low rates promoted higher sugar biosynthesis, especially sucrose, glucose, mannitol and sorbitol in leaves and roots of plants. In addition, flavonoid daidzein was highly expressed and synthetized in roots of legume plants like soybean, cowpea and peanuts, resulting in better nodulation, increased? ureides concentration and biological nitrogen fixation, resulting in higher yield of soybean, cowpea and peanut plants. Foliar application rate greater than 50 g ha⁻¹ can cause visual symptoms of toxicity impairing leaf morphology and yield of plants.

Conclusions: Foliar Se application rates ranging from 20 to 50 g/ha enhanced antioxidant metabolism and increased enzymatic and non-enzymatic antioxidant metabolism, photosynthesis and sugar in leaf tissues of sugarcane, coffee, maize, soybean, cowpea and peanut plants. The flavonoid daidzein increased in response to Se fertilization showing enhanced nodulation, biological nitrogen fixation, and yield of legume plants.

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Effect of Organic Cropping on the Accumulation of Selenium and Other Mineral Elements in Tea

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Introduction: Selenium (Se) is widely recognized as an important trace element for the maintenance of human life and health. Others have shown that the type of fertilizer has a significant effect on the content of mineral elements in plants ^[1,2]. In organic cropping systems, the organic fertilizers employed are distinctly different from the inorganic fertilizers used in conventional agriculture. The unique nutrient release mechanisms of organic fertilizers may alter the concentration and bioavailability of mineral nutrients in the soil. Consequently, these changes may potentially influence the uptake and accumulation of mineral nutrients by plants. In this study, we evaluated the effect of organic cropping on the accumulation of Se and other mineral elements in tea.

Methods: Tea and soil samples in this study were collected in July 2022 from five organic tea gardens (O-1, O-2, O-3, O-4, and O-5) certified for organic production and five adjacent conventional tea gardens (C-1, C-2, C-3, C-4, and C-5) in the Pu'er region of Yunnan Province, China. The tea cultivar in all tea gardens was *Camellia sinensis* var. assamica cv. Yunkang 10, i.e. a major cultivar grown in local tea gardens. To determine the elemental composition (Se, Cu, and Zn) in tea and soil samples, ground tea leaf samples were digested using nitric acid (HNO₃), while soil samples were digested using aqua regia (HCl:HNO₃ = 3:1, v/v). The microelement content in the digest was determined using ICP-MS (Agilent ICP-MS 7700).

Results and Discussion: On an average, there was a significant difference in Se content in tea from organic and conventional tea gardens, with organic being 191.6% higher than conventional. However, there was no significant difference in the average Se content in the soils of the organic and conventional tea gardens. Furthermore, the bioconcentration factor (BCF) of Se for organic tea was 158.3% higher than conventional. The average Cu content of tea leaves in the five organic tea gardens was highly significantly lower than that in the conventional tea gardens, by 10.7%. As for Zn, no significant differences were observed in the mean Zn content in both tea and soil between conventional and organic farming systems.

Conclusions: Based on the research findings above, the content of Se and other trace elements in tea is influenced by the cultivation patterns, and tea grown under organic cropping systems are more likely to accumulate Se.

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Integrative Analysis of Selenium Accumulation, Physiology, and Rhizosphere Metabolism in Wheat Intercropped with *Cardamine hupingshanensis*

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Introduction: Selenium (Se) is essential for human and animal health. However, Se deficiency remains a global concern. Wheat is a major staple food crop and a target for Se biofortification. However, conventional fertilization approaches face limitations in improving Se content and bioavailability. This study explores the effects of intercropping wheat with the Se-hyperaccumulating plant *Cardamine hupingshanensis* and exames its effect on Se accumulation, plant physiological performance, and rhizosphere metabolism.

Methods: Pot experiments were conducted under three sodium selenite treatments (0, 5, and 50 mg/kg). Three planting patterns were established: wheat monoculture, *C. hupingshanensis* monoculture, and wheat - *C. hupingshanensis* intercropping. Physiological parameters (photosynthesis rate, chlorophyll content, biomass, and root morphology) were measured after 30 days ^[1]. Selenium concentration and speciation were determined using ICP-MS ^[2] and LC-AFS. Rhizosphere metabolomics was performed using UHPLC-MS.

Results and Discussion: Intercropping significantly increased Se accumulation in wheat tissues, particularly under high Se treatment, with aboveground Se content reaching 200 mg/kg. Selenocysteine levels increased while selenomethionine decreased. Intercropping improved photosynthetic rate, relative chlorophyll content, and root traits under Se treatment. Rhizosphere metabolomic profiling revealed significant shifts in amino acid, sugar, and phenolic metabolism under low and medium Se levels, suggesting reprogramming of metabolic pathways involved in stress adaptation and nutrient signaling by intercropping.

Conclusions: Intercropping wheat with *C. hupingshanensis* enhances Se accumulation and alters Se speciation, while improving physiological traits. This strategy provides a promising agroecological approach to Se biofortification and sustainable farming.

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Genetic Biofortification of *Brassica napus*: A Promising Strategy for MeSeCys Enrichment

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Introduction: Selenium (Se), as an essential trace element, has been demonstrated to be significant for human health, while Se deficiencies can cause serious health issues ^[1,2]. Food crops can uptake Se from a culture substrate and synthesize safer organic Se such as selenocysteine (SeCys2), methyl-selenocysteine (MeSeCys), selenomethionie (SeMet), etc. ^[3,4]. Thus, biofortification of food crops enriched with organic Se is the most effective strategy for providing nutritional supplements. Rapeseed (*Brassica napus*. L), belonging to the *Brassicaceae* family, possess extremely high capacity to uptake, assimilate and accumulate Se. MeSeCys is dominant in rapeseed, which demonstrates high bioavailability and superior pharmacokinetic characteristics, compared to other organic Se compounds. However, the molecular mechanism of Se accumulation in rapeseed has not yet clear investigated. In this study, we analyzed the forms and contents of Se in various tissues across different fertility stages in determining its distributional characteristics. Additionally, 349 accessions were selected for the purpose of performing genome-wide association analysis (GWAS), identifying four stable QTLs associated with MeSeCys levels in rapeseed.

Methods: The detection of Se species in rapeseeds was accomplished by high performance liquid chromatography-inductively coupled plasma mass spectrometry (HPLC-ICP-MS). The 50 K SNP array for *Brassica napus* developed by Huazhong Agricultural University, were screened for GWAS analysis. We conducted a genome-wide association study (GWAS) using three models: general linear model (GLM), mixed linear model (MLM), and Farm CPU.

Results and Discussion: We analyzed the Se species in different tissues fractions of selenite-treated rapeseed and found that the above-ground organic Se abundance was consistently higher than that in the roots, with MeSeCys and SeMet being the primary organic Se forms. It is particularly interesting that SeMet content decreased with continued selenite treatment, while the MeSeCys content showed the opposite trend. At the bolting stage, MeSeCys was the main organic Se component in the shoots, accounting for up to 63.25%. By conducting GWAS of phenotypes across four replicated experiments, we successfully pinpointed four stable QTLs linked to MeSeCys content. This breakthrough has greatly advanced the screening and cultivation of rapeseed varieties with superior Se absorption, transport, and accumulation traits. Our work is essential for further studies aimed at understanding and enhancing the ability of rapeseed to accumulate Se to levels that can significantly contribute to nutritional Se biofortification.

Conclusions: By detecting Se content in different tissues of rapeseed, we found that Se accumulation is dynamically correlated with the duration of Se treatment. Notably, rapeseed exhibited a robust ability to accumulate MeSeCys at the bolting stage. The QTLs obtained by GWAS can help us to selected extreme materials and construct F_2 populations to validate the QTL by bulk segregation analysis (BSA). In the future, we intend to perform RNA-seq analysis on the extreme materials and integrate our results to obtain the key candidate genes for Se metabolism.

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Selenium for Plants and Food: Pros, Cons, and Economic Value

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Introduction: Selenium (Se) is essential for human health, supporting antioxidant defense, immunity, and cancer prevention. Over 1 billion people lack adequate Se due to poor soil, with 72% of China's land Se-deficient. To address this deficiency, Se biofortification strategies using organic and nano-Se is a growing solution. This review summarizes global progress, benefits, challenges, and economic aspects of Se and nano-Se use in crops and food.

Methods: Figure 1 illustrates the process adopted for data extraction.



Figure 1. Methodology for information extraction and synthesis

Results and Discussion: Organic Se (e.g. plant-derived peptides) shows high bioavailability (94.6% in *Lucilia sericata* maggots) while inorganic Se shows low toxicity. Nano-Se further reduces phytotoxicity; for example, lignin-containing cellulose nanofiber (LCNF)/SeNP hybrids reduced Na+/K+ ratios by 40% in salt-stressed rice, enhancing yield. Selenium biofortification increased rice prices by 50–150% in China (\$1.2-2.0/kg vs. \$0.8/kg), supporting a Se-enriched food market growing at 7.2% CAGR (2023–2030). Nano-Se (20 mg/L) lowered H₂O₂ and MDA by 60%, boosting superoxide dismutase/peroxidase in arsenic-stressed cotton, while Se priming enhanced lead tolerance in faba beans via amino acid regulation. However, Se >150 mg/L reduced amino acids in tea by 25% and inhibited *L. sericata* growth at all nano-Se levels tested. Hybrid materials like LCNF/SeNPs show dual functionality in stress mitigation and environmental remediation (e.g. degrading 86% methylene blue) However, Nano-Se synthesis costs (e.g. microwave-assisted methods) are 20-30% higher than conventional fertilizers, limiting scalability. Standardizing Se thresholds in food (e.g. EU's 50–300 µg/day limits) and funding field trials (e.g. *Limosilactobacillus* sp. LF-17strains in rape seedlings) are critical for safe adoption. Figure 2. Maps Se's inter-industry roles and regulatory effects on environmental and food systems.



Figure 2. Process of applications, advantages, and disadvantages in plants and food development - integrating statistical and economic perspectives.

Conclusions: Selenium and nano-Se have transformative potential in agriculture and functional foods. Despite toxicity and cost challenges, innovations like nano-formulations (e.g. LCNF/SeNPs) and precision farming could enable scalable solutions. Government and industrial collaboration on standardized protocols and eco-toxicity studies is key for safe, viable adoption.

Healthy China and Functional Foods

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With the social economic development in China, health has become a focus on the theme of people's life. People's demands for agricultural products and foods are no longer merely at the stage of solving the problem of food and clothing and ensuring safety, but they also have higher requirements, hoping that they can integrate functionality, nutrition and health. Definition of health: WHO makes a new definition for people's health, namely "health is not only no disease, but also physical health, mental health, social adjustment and moral good health". The assessment results from WHO on the factors affecting human health show that:

Health = 15% genetic factors + 10% social factors + 8% medical conditions + 7% climatic conditions + 60% self-care

The "Proactive Health" model advocates the concept that everyone is responsible for their own health. It establishes and practices the health management concept of being responsible for one's own health, and everyone actively learns health knowledge, improves health literacy, and strengthens health management. The latest statistics from the health census of Chinese residents show that the number of patients with various chronic diseases in China has exceeded 300 million. In 2019, the number of deaths from chronic diseases in China each year accounted for 88.5% of the total deaths. Chronic diseases have become the greatest threat to the health of the nation. Over the past decade, China's health care expenditure has continued to rise. In the "Healthy China 2030 Plan Outline" issued by The State Council in 2016, the goals and tasks for building a healthy China were put forward, emphasizing the implementation of prevention first and the promotion of a healthy lifestyle. Vigorously developing the cause of national nutrition and health is the need of "the times." Chronic diseases are closely related to dietary structure. Choosing a reasonable dietary structure is crucial for the prevention of chronic diseases. The Global Burden of Disease Study shows that in 2017, 11 million people worldwide died due to unhealthy diet, accounting for 22% of total deaths (ranking first). The WHO report states: Dietary factors (13%) have the second-largest impact on health, second only to genetic factors (15%), and greater than medical factors (8%). Unhealthy diet is the most significant factor contributing to disease occurrence and death among Chinese people. Dietary factors can explain 30.6% of deaths and 16.3% of disease burden in China, ranking first among all related risk factors. In recent years, the food consumption trend of Chinese residents shows that functional foods with nutritional and health benefits functional foods - have become the preferred choice of consumers. From ordinary foods to functional foods there is an inevitable trend in market development. Due to the relatively lagging market of functional foods in China, it is necessary to deeply analyze the existing problems and combine the development trends of functional foods at home and abroad to apply functional factor resources to the development of functional foods, creating new types of functional foods with clear information on functional factor structure, content, and mechanism of action, based on scientific evidence, to lay the foundation for promoting the development of functional foods and implementing the "Healthy China" national strategy.

Effect of Plant Growth-promoting Rhizobacteria on Regulating Selenium Absorption in Non-heading Cabbage

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Introduction: Selenium (Se) is an essential micronutrient for humans and animals, primarily obtained through dietary intake. However, China faces significant challenges as an internationally recognized "Se-deficient country". This necessitates the implementation of Se biofortification strategies to enhance Se levels in vegetables and crops. Plant growth-promoting rhizobacteria (PGPR) play a pivotal role in mediating plant Se biofortification. These microorganisms can directly promote Se accumulation in plants and/or indirectly facilitate Se uptake by improving its mobility in soil. Building upon our previously screened 15 candidate PGPR strains, this study focuses on three popular non-heading Chinese cabbage cultivars ('Shanghaiqing', 'Suzhouqing', and 'Wuyueman') as experimental subjects. Through comprehensive field trials, pot experiments, and hydroponic cultivation systems, we systematically investigate the growth-promoting effects, nutritional quality enhancements, and Se absorption capabilities induced by these PGPR candidates. The ultimate goal is to identify superior PGPR strains with exceptional Se biofortification potential.

Methods: Field trials and pot experiments were conducted at the Agricultural Experiment Station of Yichun University, while hydroponic studies were performed under controlled laboratory conditions. Test cultivars were sourced from Nanjing Ideal Agriculture Technology Co., Ltd. The 15 candidate PGPR strains were isolated from rhizosphere soils of dominant plants in three natural Se-rich regions: Yutangba Se mining area, Ziyang County, and Yuanzhou District. These PGPR strains were inoculated onto non-heading Chinese cabbage plants through pot, field, and hydroponic systems to investigate their effects on biomass production, nutritional quality, and Se bioaccumulation. Aboveground biomass dry weight was precisely measured using an analytical balance. Vitamin C, total protein, and soluble sugar contents were quantified following protocols provided by Nanjing Jiancheng Bioengineering Institute's assay kits ^[1]. Selenium concentration determination adhered to the GB 5009.258-2016 National Food Safety Standard for multielement analysis. Statistical analyses were performed using SAS at $\alpha = 0.05$.

Results and Discussion: Analysis of PGPR effects on aboveground biomass in non-heading Chinese cabbage revealed 11 superior strains (1,3-8,10-13) meeting the biomass optimization criterion (occurrence frequency \geq 3 across experimental replicates). For the 'Shanghaiqing' cultivar, seven strains (1,3,4,6,8,11,13) demonstrated consistent biomass enhancement. Evaluation of nutritional quality impacts identified 10 PGPR strains (1-5,8,11,13-15), which significantly improved vitamin C, protein, and soluble sugar levels (occurrence \geq 3). Intersection analysis selected six dual-functional strains (1,3,4,8,11,13) that excelled in both biomass and nutritional enhancement. Selenium biofortification screening designated ten strains (3,5,7-13,15) with enhanced Se assimilation (occurrence \geq 2). Final integration identified four multifunctional PGPR (3,8,11,13) simultaneously optimizing growth, nutrition, and Se accumulation. As an essential micronutrient, Se exhibits critical biological functions including antioxidant activity, immune modulation, antitumor potential, heavy metal antagonism, and protein synthesis regulation. PGPR enhanced micronutrient uptake and Se biofortification through multiple mechanisms: growth stimulation, nutrient mobilization, and stress tolerance induction^[2]. Yasin et al. demonstrated that inoculating wheat with *Bacillus cereus* YAP6 and *Bacillus licheniformis* YAP7 increased Se accumulation in stems and grains by 375% and 154%, respectively, compared to controls.

Conclusions: In this study, 11 superior strains (1,3-8,10-13) that significantly enhanced biomass production were identified. Further evaluation focusing on 'Shanghaiqing' cultivar revealed 6 dual-functional strains (1,3,4,8,11,13) demonstrating concurrent improvements in both biomass and nutritional quality (vitamin C, total protein, and soluble sugar content). Ultimately, four multifunctional PGPR strains (3,8,11,13) were selected for their tripartite capabilities: promoting plant growth, elevating nutritional value, and enhancing Se assimilation.

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Effects of PGPR on Selenium Biofortification of Pak Choi (*Brassica campestris*) in a Native Se-rich Yellow Soil

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Introduction: Plant growth-promoting rhizobacteria (PGPRs) are recognized as potential candidates for Se biofortification in sustainable agriculture. It is reported the plant Se promotion depends on enhancing soil Se bioavailability ^[1], while others insist PGPR are the driving factors for the boosting of the root system development ^[2]. It is elusive; however, which mechanism plays a dominant role in crop Se biofortification, particularly in seleniferous acid soil?

Methods: The yellow soil, with 0.72 mg/kg Se and pH of 4.72, was used for pot experiments. Two PGPRs, *Lelliottia sp.* KY49 and *Enterobacter sp.* EG16, with the IAA producing capacity of 63.8 mg/L and 9.42 mg/L, and siderophore producing capacity of 0.15 and 0.78, respectively, were inoculated to pak choi (*Brassica rapa ssp.* chinensis). Plants without inoculations were used as the control. The plants were harvested after 21 days. The fresh weight was measured directly, and roots were analyzed using a WinRHIZO system (EPSON V750). The Se concentration in frozen-dried shoots and roots was determined by HG-AFS (LC-AFS8530). A five-step sequential extraction method was used to further investigate the Se fractionations in soil by HG-AFS: soluble Se (SOL-Se), exchangeable Se and carbonate-bound Se (EXC-Se), iron (Fe)/manganese (Mn) oxide-bound Se (Fe/MnO-Se), organic matter-bound Se and elemental Se (OM-Se) and residual Se (Res-Se). Soil pH was measured by pH meter and soil organic matter (SOM) were determined by the dichromate acid oxidation method. Each experiment was performed on five biological replicates. The significant differences among the experimental groups were tested using Duncan method by SPSS 20.0 (IBM, United States).

Results and Discussion: The growth rate of pak choi was as high as 104% (EG16) and 130% (KY49), and the average root volume increased by 36.6% (EG16) and 30.4% (KY49), compared to their corresponding control groups. The Se concentration of shoots and roots reached 0.12 mg/kg and 0.45 mg/kg, increasing by 64.9% and 70.0%, respectively, when EG16 was inoculated. Only 0.05 mg/kg Se and 0.21 mg/kg Se were detected in the shoots and roots, and there was no significant difference between the KY49 inoculated and uninoculated soils. The direct bio-Se (SOL-Se and EXE-Se) concentration of EG16-inoculated soils increased from 54.2 µg/kg to 67.9 µg/kg, whereas the direct bio-Se concentration only increased by 4.0 µg/kg for the KY49-inoculated soils. In addition, the concentration of OM-Se and RES-Se declined for the inoculated soils, indicating the bioavailable Se forms might originate from transformation of the two Se fractions. Moreover, the strain EG16 achieved a higher bio-Se transformation efficiency than strain KY49, probably a larger siderophore production capacity. Meanwhile, the content of SOM increased synchronously with that of direct bio-Se but asynchronously with that of OM-Se. Therefore, the increase of direct bio-Se concentration was partially related to that of soil OM. Furthermore, Pearson's correlations showed an extremely significant positive correlation (p < 0.01) between the plant-Se and bio-Se (r = 0.87), implying the increase of soil Se bioavailability contributed more in plant Se biofortification by PGPRs.

Conclusions: The PGPRs promoted pak choi growth primarily related to IAA secretion capacity. Furthermore, PGPRs enhanced Se bioavailability by boosting the transformation of OM-Se and RES-Se to bioavailable Se forms and consequently facilitated plant Se accumulation. This promotive effect mainly depended on siderophore producing capacity of PGPRs.

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Screening, Identification and Evaluation of Selenium- and Zinc-Enriched Probiotic Strains

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Introduction: Micronutrient deficiencies, particularly of selenium (Se) and zinc (Zn), remain a significant global health concern. Conventional food supplementation typically delivers nutrient elements separately and lacks synergistic interaction with probiotic functions. However, the concurrent enrichment of both Se and Zn within a single probiotic strain remains largely unexplored, hindering the development of multifunctional nutraceuticals.

Methods: Lactic acid bacteria were isolated from fermented vegetables (pickled cucumber, bamboo shoots, bell pepper, white radish) ^[1]. Strains tolerant to Se and Zn were screened using gradient concentration assays (0 - 300 mmol/L), and their bioaccumulation capacities were quantified via ICP-MS ^[2,3]. Probiotic properties, including antibiotic susceptibility, gastrointestinal survival, hemolytic safety, and antioxidant activities, were also evaluated.

Results and Discussion: Several *Lactobacillus plantarum* strains capable of tolerating high Se (20 mM) and Zn (50 mM) concentrations were screened. These strains exhibited efficient Se bioaccumulation (1.8 - 2.5 g/L biomass) with maximum accumulation of Se by the strains (>90%), suggesting active conversion into Se nanoparticles. Concurrently, selected strains demonstrated significant Zn precipitation (>15%), likely through biosorption or nanoparticle synthesis. Notably, biomass remained stable under metal stress, indicating metabolic resilience. The dual enrichment capability and probiotic functionality of these strains position them as promising candidates for multifunctional nutraceuticals. These strains may then address both micronutrient deficiencies and gut health synergistically, a significant advancement over conventional single-element supplements.

Conclusions: Dual Se-Zn-enriched *Lactobacillus plantarum* strains were successfully screened, demonstrating high tolerance and efficient bioaccumulation. Their metabolic stability and probiotic functionality position them as innovative candidates for multifunctional supplements.

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Effects of Different Chemical Forms of Selenium on Se-Biofortification in Wheat Grains

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Introduction: "Hidden hunger" is a globally public health challenge. FAO statistics show that over 2 billion people lack micronutrients, with selenium (Se) deficiency being a major issue for over 1 billion. Selenium is an essential micronutrient for humans, playing roles in antioxidant capacity, immune regulation, and degradation of misfolded proteins. Its deficiency is linked to many diseases, and therefore, Se is increasingly used in medicine and clinical care strategies. In Se- deficient areas, plant - derived Se is a primary source for humans and animals. Plants can absorb inorganic Se forms and transform them into organic Se, but the forms and content vary by plant species and variety. Wheat is a staple crop worldwide with the high Se enrichment potential. Thus, boosting the content of beneficial forms of Se in wheat is a critical strategy to address Se deficiencies in humans^[1-4].

Materials and methods: Wheat varieties Zhengmai 9023 (ZM9023) and Xinong 1376 (XN1376) were chosen to study the Seenrichment effects of different Se forms on wheat grains. The Se chemical forms selected were categorized into inorganic and organic types. Selenium was applied to wheat plants via foliar spraying during the booting - heading stages under both greenhouse and field conditions. For total Se determination, pretreated samples were digested in a MARS 5 microwave system and analyzed using an Optima 7300 DV ICP - OES. For Se speciation, liquid chromatography - atomic fluorescence spectrometry (LC-AFS) was used. This analysis was performed in Dr. Yuan's Lab at Xi'an Jiaotong-Liverpool University.

Results and discussion: Selenium content in wheat grains differed depending on soil properties, wheat variety, and Se form applied. Selenium content in field-grown wheat grains was about 50% lower than that in greenhouse-grown wheat. Under different Se treatments, Se content in XN1376 grain was 1-2 times higher than that in ZM9023. Compared to the control, both inorganic and organic Se application increased Se content in grains, which increases with Se doses. However, the increase range varied with Se forms. In general, the Se content in wheat grains for different Se forms followed the order of MSG (methylselenized glucose) > Na₂SeO₃ > MeSeCys > MSL (methylselenized lactide). Na₂SeO₃ and MeSeCys are the naturally occurring chemical forms of Se in plants. Wheat plants had a significantly higher absorption and accumulation of Na₂SeO₃ than MeSeCys. For the two newly synthesized methylated Se compounds, the plant absorption capacities were completely different. The MSG treatment had the highest enrichment effect while the MSL treatment had the poorest, indicating that different chemical complex linked to the Se methyl (CH₃Se-) group significantly affected Se absorption.

Conclusions: This new organic MSG form had significant advantage in Se-enrichment, offering a new and eco-friendly Sebiofortification fertilizer.

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Enhancing Food Security: Advances in Plant Nutrition with Selenium

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Introduction: Selenium (Se) is a vital nutrient that enhances antioxidant defense, strengthens agricultural sustainability, and improves food security, supporting the UN's Sustainable Development Goals (SDGs). Agronomic biofortification using fertilizer containing Se reduces oxidative stress, boosts plant resilience, and increases mineral bioavailability, promoting better human nutrition. Optimizing Se uptake through agronomic biofortification ensures efficient and cost-effective strategies for emerging countries. Identifying the dose-response of fertilizer application can be an effective strategy for biofortification programs and establishing safe Se doses to combat nutritional deficiencies ^[1].

Methods: This study integrates a meta-analysis on Se biofortification experiments in Brazil. Foliar Se application (0, 10, 25, 50, 100, and 150 g/ha) was performed using cowpea ^[2]. Physiological and biochemical analyses and mineral elemental concentrations were examined using X-ray fluorescence spectroscopy ^[2].

Results and Discussion: The foliar application of selenium (Se) at a rate of 50 g/ha resulted in yield increases of 53% and 41% in cowpea plants over two consecutive years compared to untreated controls. Foliar application proved to be the most effective method for enhancing Se concentrations in both leaves and grains. However, applications exceeding 50 g/ha led to toxicity, manifesting as necrosis in the leaves. A foliar Se application rate of 10 g/ha was identified as the safest approach to achieving the recommended daily Se intake of 55 μ g/day in both years, in accordance with Codex Alimentarius guidelines. Selenium was deposited and accumulated within the endosperm, particularly near the embryo region (Fig. 1). Agronomic biofortification with Se improved the nutritional quality of cowpea grains by increasing concentrations of essential minerals such as calcium (Ca), magnesium (Mg), sulfur (S), iron (Fe), and zinc (Zn). This study provides valuable insights into the agronomic biofortification of cowpea plants with Se as a strategic approach to enhancing food quality and potentially improving human health. Agronomic biofortification plays a crucial role in increasing crop nutrient content, thereby strengthening human nutrition and plant resilience while promoting agricultural productivity. Selenium biofortification represents a sustainable solution for food security, malnutrition mitigation, and resilient agricultural practices.



Figure 1. Localization of Se in cowpea seeds using X-ray fluorescence spectroscopy.

Conclusions: Foliar Se application up to 50 g/ha increases the yield of cowpea plants and is an effective strategy to biofortify to a safe level. In emerging countries like Brazil, these studies are driving initiatives to address Se deficiency in affordable foods, such as cowpea, which might enhance human health.

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Effect of Foliar Application of Selenium on Speciation, Elemental Accumulation, and Nutritional Composition of Sweet Maize

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Introduction: A significant factor contributing to Se deficiency in human populations is its low content in food, which directly affects Se intake in humans. Crop-Se biofortification has been recognized as the primary entry point of Se into the food chain, with cereals and vegetables accounting for approximately 70% of human Se intake ^[1]. Sweet maize (Zea mays ssp. Saccharata) is widely cultivated across temperate, tropical, and subtropical regions due to its high nutritional value and desirable sweet kernel taste. Despite the growing interest in Se biofortification, limited studies have focused on speciation and trace element content in sweet maize kernels. Therefore, it is crucial to investigate the impact of foliar Se application on Se accumulation, elemental composition, and kernel quality in sweet maize.

Methodology: A field experiment was conducted from July to October 2024 at the Wangying Village Experimental Base in Chuzhou, Anhui Province, China. A randomized complete block design with three replications was used, consisting of four selenite foliar treatments: 0 (CK), 20 g/ha (Se1), 40 g/ha (Se2), and 60 g/ha (Se3), resulting in 12 experimental plots, each measuring 48.75 m^2 (7.5 m × 6.5 m). The treatments were uniformly applied at the tasselling stage, with a 0.2% Tween-20 surfactant incorporated into the spray solutions to improve adhesion and absorption. Analysis of SOD, POD, CAT and MDA were performed. Sweet maize nutritional quality, including soluble sugar, crude protein, crude fat, and vitamin C, was assessed following the Chinese National Standard GB 5009.168-2016 and GB/T 5009.6-2016, while sugar content was evaluated using a Brix refractometer. Speciation, macro- and micro-elements, including N, P, K, Mg, Zn, Cu, Fe, and Mn, were assessed using ICP-MS. One-way ANOVA was used to evaluate differences among the means with the Tukey test (α =0.05).

Results And Discussion: The findings of biochemical analysis demonstrated that Se1 treatment significantly enhances SOD, POD, and CAT while reducing MDA levels. In contrast, at a high rate (Se3), SOD, POD, and CAT declined, while MDA levels increased compared to CK. Regarding micro- and macro-elements levels, Se1 resulted in a significant increase in Mg by 5.4%, P by 6.3%, N by 8.0%, Zn by 17.5%, Mn by 12.5%, Fe by 5.1%, and Cu by 10.7% compared to CK. However, at Se3, a significant decline in both macro- and micro-elements was observed. Similarly, Se1 demonstrated substantial enhancements in nutritional quality, showing significant variations in sugar content (4.4%), soluble sugar (4.5%), vitamin C (19.8%), crude fat (15.6%), and crude protein (8.0%) compared to CK. Conversely, a significant reduction in crude protein (2.7%), crude fat (20.7%), and vitamin C (16.1%) was observed at Se3. Moreover, the predominant Se species in kernels was exclusively organic Se, with SeMet accounting for 82.3%, SeCys 12.6%, and MeSeCys 5.1%.

Conclusions: Foliar application of selenite at optimal rates significantly boosted enzymatic activity, enhanced nutrient uptake, and improved nutritional quality. In contrast, high rates of Se led to phytotoxic effects, resulting in a decline in our measured parameters. These results show the effectiveness of optimizing Se application rates as a sustainable strategy to address global Se deficiency while improving the performance of maize growth and yield.

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Effects of Sowing Rate on Selenium Accumulation and Translocation of Different Winter Wheat Varieties in Selenium-enriched Area

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Introduction: Dietary trace element intake like selenium (Se) has gained increasing attention worldwide. In China, wheat is a staple food for the Chinese population. Hence, clarifying Se absorption and understanding the mechanisms related to plant uptake of Se is of significant scientific and nutritional value in winter wheat cultivars.

Methods: In 2021–2023, we conducted a split-plot experiment at Hongtong Wheat Research Institute using two varieties (Shinong 086, Yannong 1212) as main plots and three sowing rates (300, 375, 450 kg/hm²) as subplots. In the study, we analyzed soil Se content^[1], speciation^[2], organ-specific accumulation^[3], and grain or flour Se concentration^[4] at flowering and maturity.

Results and Discussion: The variety Yannong 1212 was exposed to a higher soil water-soluble/exchangeable Se than Shinong 086. The sowing rate of 375 kg/hm² increased soil exchangeable Se by 11.0 - 21.1% and grain Se accumulation by 18.6 - 27.1% compared to the sowing rate of 450 kg/hm² applied to Shinong 086. Root-to-shoot translocation coefficients peaked at 375 kg/hm², with Yannong 1212 achieving the maximum bio-enrichment of Se. The optimal plant density increased flour organic Se by 3.4 - 11.1%. Grain Se correlated positively with the yield, flowering-stage soil water-soluble Se, and the maturity-stage bioavailable Se (p < 0.05).

Conclusions: Yannong 1212 out-performed Shinong 086 in yield and grain Se content. A sowing rate of 375 kg/hm² optimized root uptake of soil nutrients, enhanced root-to-grain Se translocation efficiency, and promoted grain Se accumulation, thereby increasing grain Se content and aligning with high-yield Se-enriched wheat production objectives.

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Selenium Application Enhances the Accumulation of Flavones and Anthocyanins in Bread Wheat (*Triticum aestivum* L.) Grains

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Introduction: Selenium (Se) is a trace element essential for human health. Selenium biofortification in wheat increases not only Se in grains, but also vital health-promoting compounds like anthocyanins. However, it is not clear whether flavonoids other than anthocyanins are enhanced by Se biofortification, and what is the mechanism underlying the accumulation of flavonoids in wheat grains. Herein, we investigated the foliar application of Se on flavones and anthocyanins in wheat.

Methods: Foliar application of Se (as selenite) in wheat was conducted ten days after flowering. Metabolite profiling and transcriptome sequencing were performed ten days after the Se treatment^[1,2].</sup>

Results and Discussion: The significant increases in the total contents of Se and anthocyanins were observed in mature wheat grains. Metabolite profiling identified 27 significantly increased flavonoids in immature wheat grains treated with Se relative to control. Furthermore, catechins were also found to increase in Se-biofortified wheat grains. Integrated analysis of metabolites and transcriptome revealed that Se application enhanced the accumulation of flavones, anthocyanins and catechins by increasing the expression levels of seven key structural genes in flavonoid biosynthesis (two TaF3Hs, two TaDFRs, one TaF3'5'H, one TaOMT and one TaANR). Furthermore, 182 transcription factors (including MYBs and bHLHs) showed significantly differential expression levels in the Se-applied wheat grains, which could be involved in gene regulation induced by the Se treatment.

Conclusions: This study shows that Se biofortification increases the accumulation of tricin, dihydroquercetin, anthocyanins and other flavonoids in wheat grains. Thus, Se biofortification is a practical approach to efficiently improve the contents of selenoproteins and other health-promoting compounds such as flavonoids in wheat grains. Our study provided insights into the underlying molecular mechanism that led to the improved flavonoid accumulation in Se-biofortified wheat grains and laid a theoretical foundation for further development of wheat health products.

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Selenium Use in Agroecosystems within the Framework of the 4R Nutrient Stewardship

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Introduction: Selenium (Se) is a vital micronutrient for both animals and humans, and it has recently been acknowledged for its benefits to plants ^[1]. For humans, selenium offers antioxidants, anti-inflammatory, anti-cancer, antiviral, and anti-aging properties. It is estimated that 1 in 7 people worldwide ^[2], as well as about 14% of the world's soils, are deficient in Se.

Methods: The 4R Nutrient Stewardship is a straightforward concept – apply the right source of nutrients at the right rate, at the right time, and in the right place – but implementing it is knowledge-intensive and site-specific. Here, we addressed the challenges of Se use in plant and human/animal nutrition^[3], highlighting selected studies that demonstrate a sustainable approach to Se use in agricultural systems.

Results and Discussion: Ensuring the efficacy of Se use in agriculture for biofortification purposes or as a beneficial element that enhances plant growth and increases tolerance to biotic and abiotic stresses – is challenging, because soil and plant characteristics must be considered. Selenium is chemically similar to sulfur and is absorbed by plants through sulfur transporters in the roots. Selenium primarily exists in soils as selenate or selenite. As anions, they undergo reactions with positively charged soil colloids that are abundant in tropical agroecosystems- the world's frontier in food, feed, and fiber production ^[4].

Conclusions: By addressing the 4R Nutrient Stewardship concept for Se use in agroecosystems, we aim to optimize the use of the world's limited Se resources, while considering the economic, social, and environmental dimensions of nutrient management to enhance soil, plant, and human health.

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Development and Implementation of an Assessment Framework for Identifying Natural Selenium-enriched Tea Growing Regions

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Introduction: Selenium (Se)-enriched tea has gained increasing market recognition due to its health benefits. However, it remains a challenge to identify suitable production regions capable of yielding natural Se-rich tea within vast Se-rich soil areas ^[1]. This study aims to develop a scientific evaluation methodology for screening natural Se-enriched tea production areas by comprehensively considering soil properties and tea plants' Se absorption and accumulation capacity.

Methods: This study selected Lincang City and Pu'er City, the primary tea-producing regions in Yunnan Province, as the research area. A total of 256 paired soil-tea samples were collected, and their Se content was determined using standard methods such as atomic fluorescence spectrometry. Four evaluation parameters were adopted: soil pH, organic matter content, soil Se concentration, and the Se enrichment coefficient in tea leaves ^[2]. A fuzzy mathematical model was employed as the evaluation method, combined with GIS spatial analysis technology for empirical application and suitability zoning of production areas.

Results and Discussion: The pH of tea garden soils ranged from 3.82 to 8.46. In Lincang, 30.0-87.5% of soils, and in Pu'er, 17.6-80.0% of soils, fell within the optimal pH range for tea plants (4.5 - 5.5). Organic matter content was generally abundant (2.1 - 91.8 g/kg), though lower levels were observed in some eastern regions. Soil Se content exhibited significant variation (0.02 - 1.24 mg/kg), with higher concentrations in western areas. The Se enrichment coefficient in tea leaves varied widely (0.026-17.500), indicating substantial differences in Se uptake capacity among tea plants. Methodological Innovation: This study innovatively integrates fuzzy mathematics and GIS technology to establish an objective evaluation system for Se-rich tea production areas by quantifying indicator weights and membership functions, overcoming the subjectivity of traditional qualitative assessments ^[3]. Validation: Empirical results demonstrate strong agreement between the evaluation outcomes and actual production conditions. Western production areas were rated as optimal due to favorable soil conditions (pH 4.5 - 5.5, organic matter >30 g/kg, Se 0.4 - 3 mg/kg), confirming the system's reliability. Future Improvements: Although the current system is practical, it relies on a limited number of indicators (only four) and involves complex calculations. Future research should incorporate additional factors (e.g. soil texture) and validate the system in more regions to enhance its applicability.

Conclusions: Spatial analysis revealed higher suitability in western regions, while eastern areas were deemed unsuitable due to alkaline soils, low organic matter, and insufficient Se levels. Validation demonstrated a significant correlation between the evaluation results and tea compliance rates (GH/T 1135-2017). The proposed method effectively identifies suitable production zones for Se-rich tea. For unsuitable areas, improvement measures such as pH adjustment, organic fertilizer application, and cultivation of high-accumulation tea varieties are recommended ^[4]. This study provides a scientific foundation for the development of the Se-rich tea industry.

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Standard Threshold Determination and Agricultural Utilization of Selenium-Rich Soils in Anhui Province

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Introduction: Anhui Province, as a key component of the selenium (Se)-rich belt in the middle-lower Yangtze River region, holds strategic importance in rural revitalization and health industry development through standardized utilization of its selenium-rich soil resources. This research integrates 1:250,000-scale land quality geochemical survey data with crop and rhizosphere soil investigations in representative areas, systematically revealing the spatial distribution patterns, genetic mechanisms, and bio-enrichment characteristics of selenium-rich soils across the province. The study established definitive threshold standards for Anhui's selenium-rich soils: strongly acidic (pH \leq 5.5, Se \geq 0.40 mg/kg), acid-neutral (5.5 < pH \leq 7.5, Se \geq 0.34 mg/kg), and alkaline (pH > 7.5, Se \geq 0.31 mg/kg) soils. The preparation and publication of "Technical Requirements for Evaluation of Selenium-Rich Soil" (T/SSSAP 006-2024) has addressed the provincial standardization gap in selenium-rich soil assessment, while providing robust scientific foundation for optimizing Se resource utilization and advancing premium agricultural development.

Methods: This study systematically integrated 1:250,000-scale land quality geochemical survey data to construct a comprehensive geochemical database for Anhui Province. Field investigations were conducted across 10 representative areas (including Chizhou and Xuancheng), collecting crop samples, rhizosphere soils, and vertical soil profiles. Advanced analytical techniques (XRF and ICP-MS) were employed to quantify Se and other critical geochemical indicators. The selenium-rich soil threshold was scientifically determined through cumulative frequency analysis and iterative elimination methods, while principal component analysis was applied to elucidate the genetic sources and enrichment mechanisms of Se in the studied soils.

Results and Discussion: The study established pH-dependent thresholds for selenium-rich soil classification: strongly acidic soil ($pH \le 5.5$) with Se ≥ 0.40 mg/kg, acid-neutral soil ($5.5 < pH \le 7.5$) with Se ≥ 0.34 mg/kg, and alkaline soil (pH > 7.5) with Se ≥ 0.31 mg/kg. Rice demonstrated the highest Se enrichment rate among tested crops, highlighting cereal crops' superior suitability for selenium-rich agricultural development. These findings were formalized in the newly released "Technical Requirements for Evaluation of Selenium-rich Soil" (T/SSSAP 006-2024) standard.

Conclusions: This study successfully established a pH-based threshold standard system for selenium-rich soils (strongly acidic: Se $\geq 0.40 \text{ mg/kg}$; acid-neutral: Se $\geq 0.34 \text{ mg/kg}$; alkaline: Se $\geq 0.31 \text{ mg/kg}$), confirming geological high background as the primary Se source. The findings address the critical gap in selenium-rich soil evaluation standards for Anhui Province, offering both scientific guidance for optimal utilization of selenium-rich land resources and strategic support for national "Healthy China" and rural revitalization initiatives.

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Research Progress on Application of Selenium-enriched Yeast

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Introduction: Selenium (Se) is an essential trace element critical for human and animal health. Organic Se can be synthesized via physicochemical methods that enhance its bioavailability; however, microbial biotransformation represents the most efficient and eco-friendly approach for organic Se production ^[1]. Currently, predominant Selenium-enriched microbial strains include Saccharomyces cerevisiae (yeast), lactic acid bacteria (LAB), *Bacillus subtilis*, and *Aspergillus oryzae*, among others. Yeast, a eukaryotic microorganism, is widely utilized in fermented foods such as fruit wines, meat products, and cereal-based bread. As a feed additive, Se-enriched yeast not only elevates Se content but also enhances the nutritional quality of livestock products.

Results and Discussion: Supplementing dairy cow feed with 5 mg/kg Se-enriched yeast significantly increased selenomethionine (SeMet) and selenocysteine (Sec) levels in milk ^[2]. Similarly, Se-enriched yeast ameliorated atrazine-induced oxidative stress and muscle deterioration in quails by suppressing inflammatory pathways ^[3]. Additionally, Se-enriched yeast attenuated lipopolysaccharide (LPS)-triggered inflammation and oxidative stress while maintaining key regulatory pathways for milk fat and protein synthesis in porcine mammary epithelial cells ^[4]. Beyond animal nutrition, Se-enriched yeast has been employed in fermented beverages to diversify Se supplementation and improve product quality. A mutagenesis-derived engineered yeast strain (6#) can achieve a total Se content of 2617 mg/kg with 94.5% organic Se incorporation. Fermentation, this strain enhanced polyphenol, flavonoid, and volatile aroma profiles in apple cider, yielding total and organic Se concentrations of 20.67 µg/L and 15.39 µg/L, respectively ^[5]. In bakery applications, Se-enriched yeast influences dough rheology. As Se content increased, the gas production rate of Se-enriched yeast slowed down, the dough's viscoelasticity decreased, and the dough became softer ^[6]. Owing to its anti-inflammatory, immunomodulatory, and antioxidant properties, Se-enriched yeast holds therapeutic potential. For example, a metabolic engineering strategy combining Se-enriched yeast with biomimetic nanozymes to synthesize carbon quantum dots will enable dual-function therapy via reactive oxygen species (ROS) scavenging and vascular-bone metabolic axis modulation ^[7]. Industrial applications of Se-enriched yeast span active yeast cells, Se-containing proteins, Se nanoparticles (SeNPs), and yeast-derived metabolites, with uses in feed, food, nutraceuticals, and pharmaceuticals.

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Comprehensive Comparison of Se Accumulation, Translocation and Effects on Plant Growth in Vegetable, Legume and Cereal Species

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Introduction: Selenium (Se) is an essential micronutrient for humans and animals, while its deficiency is globally widespread, . , posing a significant threat to public health. To mitigate this issue, several primary dietary crops, including cereals, legumes and vegetables, have been proposed for Se biofortification. Nevertheless, the efficiency of Se uptake from soil, root to shoot translocation, and accumulation in plant tissues, varies significantly among plant species, resulting in differing biofortification efficiencies. A systematic and comparative evaluation of the fate of Se in various crops is therefore crucial for identifying species with high Se uptake efficiency and accumulation capacity. The results are expected to provide a scientific basis for the selection of optimal crop species for practical biofortification applications, particularly in regions with Se-deficient soils, and ultimately enhance dietary Se intake in humans and animals through agricultural intervention.

Methods: Pot experiments were conducted under greenhouse conditions using soils with Se concentration ranging from 0 to 5.0 mg/kg. Crops of pak choi (*Brassica rapa subsp. chinensis*), lettuce (*Lactuca sativa*), wheat (*Triticum aestivum*), maize (*Zea mays*), and beans (*Phaseolus vulgaris*) were selected to represent vegetables, cereals and legumes species. After harvest, plant growth parameters, Se concentrations and Se distribution within plant tissues were measured. Mantel test, principal component analysis (PCA), and partial least-squares path modeling (PLS–PM) were employed to comprehensively compare crops-specific response to Se. X-ray fluorescence (XRF) was used to determine the distribution of Se in plant tissues.

Results and Discussion: The results demonstrated that pakchoi exhibited the highest Se concentration in the crops, reaching up to 732 ± 137 mg/kg in shoots, which are 2.0 - 4.5 times greater than that of wheat, the crop with the lowest Se concentration. In pakchoi and beans, the Se taken up by the crop was primarily translocated to the aboveground parts, with the translocation factor (TF) ranging from 1.5 to 4.6, whereas it was mainly retained in the roots of other crops. High soil Se (≥ 3 mg/kg) significantly reduced the total dry biomass of pak choi and lettuce by 38-50% and 22-54%, respectively, compared to untreated soil in which beans and wheat biomass increased by 14-30 % and 3-29%, respectively. Mantel test and PCA analysis indicated that Se mainly affected the growth of the vegetables, which showed a different response to Se application, compared to the other crops. PLS–PM showed that soil Se application indirectly affected shoot Se through regulating root Se (path coefficient > 0.76) ^[1]. XRF analysis indicated that Se was primarily distributed in the petioles and leaf veins, as well as in the axial roots of pak choi, suggesting Se translocation through vascular tissues. The high Se accumulation and efficient translocation in pak choi highlights its potential as a preferable candidate for Se biofortification.

Conclusions: *Brassicaceae* pak choi demonstrated the highest capacity for Se uptake and accumulation, as indicated by its significantly elevated Se concentrations, followed by beans and maize. The efficient Se uptake and accumulation in the edible aboveground tissues of pakchoi suggest it is an optimal crop for Se biofortification. Moreover, its high uptake of Se not only enhances Se biofortification efficiency and address Se deficiency, but it also reduces Se fertilizer loss in the environment.

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Effect of Selenium Fertilizer Application in China and Analysis of Influencing Factors

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China faces a significant issue of uneven selenium (Se) distribution in soil, with selenium-deficient areas accounting for 72% of the nation's land area. Selenium fertilizers, as an important means of crop Se biofortification, are influenced by multiple factors such as soil properties, fertilizer types, and climatic factors. Current research indicates significant regional variation and inconsistent conclusions, necessitates a systematic literature analysis to integrate national data, and reveals the mechanisms of Se fertilizer efficiency, and optimize regionalized fertilization strategies. This study integrates multi-source literature data across the country and aims to: (1) identify the key drivers of crop Se enrichment efficiency; (2) analyze the interaction effects between Se application parameters and environmental factors; and (3) propose regionally adaptive Se fertilizer management strategies. Over 260 valid observation values were extracted from more than 130 Chinese and English literature sources over the past decade (2015 - 2025), covering different crops such as grains, vegetables, and fruits. A structured dataset was constructed, including the following dependent variables: grain Se content (mg/kg), yield increase (%); independent variables: Se fertilizer type (inorganic or organic), Se application rate (g/ha), application method, soil properties (pH, organic matter, background Se), and climatic factors (annual-average temperature, precipitation). Methods such as variable correlation analysis, multiple regression modeling, and group model construction were used to analyze the relationships between variables and construct a relationship diagram of Se fertilizer application effects and also explore the main influencing factors. The analysis of Se fertilizer application effects and influencing factors in China over the past decade shows that Se fertilizers can effectively increase crop Se content and improve nutritional quality without significant negative impacts on yield. Their effects are regulated by multiple factors: regarding Se fertilizer types. For example, organic Se fertilizers are safe and have significant quality improvement effects, while inorganic Se fertilizers act quickly but require controlled application to prevent overuse. In regard to the application methods, combining foliar spraying and soil basal application can achieve synergistic effects, understanding that soil conditions and crop types are also core influencing factors. Additionally, supporting measures such as split application, combined use of phosphorus and Se, and optimized planting density, can further enhance the efficiency of Se fertilizer. The research indicates that by precisely matching Se fertilizer types, application strategies, and regional environments, a balance between Se fortification and sustainable agricultural development can be achieved, providing a scientific basis for precise management in functional agriculture.

Progress Research of Selenium Stable Isotopes in the Earth's Surface Environment

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Introduction: Selenium (Se) is an essential trace element for life but becomes toxic in excess. Its migration and transformation behavior within Earth's surface environments is crucial for ecosystems and human health. Recent advances in Multi-Collector Inductively Coupled Plasma Mass Spectrometry (MC-ICP-MS) analytical techniques have established Se stable isotopes (typically denoted as $\delta^{82/76}$ Se or $\delta^{82/78}$ Se) as a powerful tool for tracing Se sources, elucidating its biogeochemical cycling mechanisms, and reconstructing paleoenvironmental conditions ^[1].

Methods: This study systematically reviews recent advances in Se stable isotope applications within surficial environmental research. It focuses on key geochemical and biological processes governing Se isotope fractionation, including: (1) Redox reactions: Transformations between different selenium species (e.g. Se(VI), Se(IV), Se(0), Se(-II)); (2) Biological metabolic processes: Se uptake, assimilation, dissimilatory reduction, methylation (volatilization), and demethylation by microorganisms and plants; (3) Abiotic adsorption/desorption processes. Concurrently, the study summarizes application examples utilizing selenium isotopes to trace sources, migration pathways, and transformation mechanisms of Se across diverse surficial reservoirs (e.g. soils, rivers, lakes, oceans, the atmosphere, and organisms).

Results and Discussion: Research indicates that redox reactions of Se are the primary drivers causing significant isotopic fractionation. For example, during the reduction of Se(VI) to Se(IV) or Se(0), the products typically enrich light isotopes (e.g., ⁷⁶Se), while the residual Se(VI) enriches heavy isotopes (e.g. ⁸²Se). The fractionation magnitude ($\Delta^{82/76}$ Se) can reach -10% to - 15% or even higher, depending on specific bacterial strains, electron donors, and environmental conditions ^[2-4]. In soil environments, Se isotopes effectively trace selenium sources (e.g. parent rock weathering, atmospheric deposition, agricultural fertilization) and its migration and speciation transformations within the soil-plant system. Soil type, pH, Eh conditions, and organic matter content all influence Se speciation and isotopic composition in soils, thereby affecting its bioavailability. In aquatic systems (rivers, lakes, oceans), Se isotopic composition is governed by a combination of watershed geology, anthropogenic inputs (e.g. mining effluent, agricultural runoff), hydrochemical conditions, and biological activity ^[5]. The isotopic signature of atmospheric selenium aids in differentiating contributions from various sources such as volcanic activity, fossil fuel combustion, and biological volatilization, providing a basis for assessing the impact of atmospheric selenium deposition on ecosystems.

Conclusions: The Se stable isotope system provides a unique and powerful means for gaining deeper insights into the complex behavior of Se within Earth's surface systems. However, current research still faces several challenges: the coupled effects of multiple fractionation mechanisms (e.g. redox, biological processes, adsorption) are difficult to precisely resolve; the extremely low Se concentrations in some environmental samples impose exceptionally high demands on analytical precision; and systematic studies on the isotope fractionation factors for certain specific processes are still lacking.

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Selenium in Organic Paddy Rice Soil

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Introduction: Sustainable soil management can unleash soil biodiversity potential and increase environmental, economic and social well-being through SOILGUARD project funded by EU Horizon 2020 Research & Innovation Program. The project focused on assessing the impact of agricultural practices on soil fertility in different regions of the world. In Thailand, research has been conducted to determine agricultural soil quality in comparing between continuous organic and conventional paddy rice plantation systems that were associated with different levels of soil degradation. Thus, effects of both organic and conventional cultivation practices on soil Se content were also examined in 2022 to 2023.

Methods: Sixteen soil sampling sites of organic and conventional paddy rice were collected in Chiang Rai Province, Thailand. A total of 32 soil samples were collected from those areas having high, medium, and low levels of soil degradation that were classified based on soil organic carbon content and soil texture after rice harvesting in November 2022. The total Se content in soils were measured using ICP-MS. Soil pH, electrical conductivity (EC), available phosphorus, and exchangeable potassium were also analyzed. The soil parameters were also correlated with soil Se content via statistical correlation analysis.

Results and Discussion: The total Se content in soil was averaged as $160 \ \mu g/kg$ and $200 \ \mu g/kg$ in organic and conventional farming, respectively. In organic paddy rice soils Se concentration varied significantly, showing higher soil Se content in low soil degradation level than in high soil degradation level as observed in conventional rice paddy soil. Selenium in soil is influenced by soil texture. Generally, soils with higher clay content tend to have higher Se concentrations. This is because clay particles have a greater capacity to adsorb and retain Se. Conversely, sandy soils, with lower clay content, typically have lower soil Se concentrations. Selenium can also be associated with organic matter. Soils with higher organic matter content may have higher Se concentrations. There was a significant correlation between total soil Se and soil organic matter content (r=0.52).

Conclusions: Selenium in organic soils were significantly different with conventional paddy rice. The total Se content in soil was average about 160 μ g/kg and 200 μ g/kg in organic and conventional farming, respectively. In organic paddy soil with a high level of soil degradation (showing coarse soil particles and low organic carbon content) soil Se content was lower than in the soil with a low level of soil degradation with fine soil particles and high organic carbon.

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Driving Factors of Selenium Distribution in Rice and Rhizosphere Soils in China: A Hotspot Analysis Approach

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Introduction: Selenium (Se) is one of the most important trace elements for human health. However, China is a relatively Sedeficient country, making the development and utilization of Se-rich land resources of great significance. Rice is widely cultivated across China, and the Se content in rice grains directly affects dietary Se intake. Nevertheless, the driving factors influencing soil and grain Se concentrations and the bioconcentration factor of Se in rice across different planting regions remain unclear.

Methods: This study integrated over 11,908 data points from published literature and our research database. It employs hotspot analysis, geographically weighted regression, and big data machine learning techniques to systematically investigate the geochemical characteristics of Se, its ecological effects, and the driving factors of Se bio-concentration in rice grains at a national scale.

Results and Discussion: Nationwide, the average Se contents in rhizosphere soil and rice grains are 0.375 mg/kg and 0.051 mg/kg, respectively. 1 High soil Se values are associated with the distribution of black rock series and metal sulfide deposits. Hotspot analysis with varying bandwidths reveals that at an 800 km bandwidth, the BCF-Se hotspots and cold spots in rice grains can be categorized into four major regions: the Southwestern Karst Coldspot Zone (low BCF-Se), the Yangtze River Middle-Lower Plain Hotspot Zone (high BCF-Se), the Northeastern Plain Coldspot Zone (low BCF-Se), and the Southeastern Hilly Region (non-significant BCF-Se variation). At the national scale, soil organic carbon (Corg) and soil sulfur content are identified as the primary driving factors influencing rice grain BCF-Se. In the Southwestern Karst Region, soil pH and the widespread development of iron-manganese nodules (Mn and TFe₂O₃) are key drivers that inhibit Se uptake by rice. In the Southeastern Hilly Region, factors such as the soil aluminum-to-silicon ratio (Al/Si), temperature, precipitation, and elevation significantly affect BCF-Se. In the Yangtze River Middle-Lower Plain, flat terrain and the weathering-leaching coefficient (ba) serve as unique drivers influencing the mobilization of bioavailable Se and its accumulation in rice grains. In the Northeastern Plain, high Corg content leads to strong Se adsorption/complexation, reducing its bioavailability.

Conclusions: The Se contents in rhizosphere soils and grains in China are influenced by multiple factors. Hotspot analysis confirms that soil organic carbon and sulfur content are the main nationwide drivers of Se bioconcentration in rice grains, while each geographic region exhibits distinct characteristic drivers. This study contributes to the theoretical understanding of Se's biogeochemical behavior in surface environments and provides theoretical and practical insights for the regional planning of Serich rice cultivation in China.

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Dissolved Organic Matter Could Activate Selenium and Inactivate Cadmium in Natural Selenium-Cadmium-Rich Paddy Soils

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Introduction: Currently, cadmium (Cd) contamination problems exist in many natural selenium (Se)-rich soils, which severely limits crop cultivation in these areas. The present study aims to explore the effects of applying agricultural residues-derived dissolved organic matter (DOM) on the fate of Se and Cd in natural Se-Cd-rich soils. This study aimed to explore the effects of DOM derived from different agricultural residues enhance Se bioavailability while reducing Cd bioavailability in natural Se-Cd-rich pappy soils, and to find the possible drivers for DOM application to increase Se content and reduce Cd content in grains.

Methods: The different agricultural residues derived-DOMs will be applied in the natural Se-Cd-rich paddy soils and the responses of Se and Cd fractions transformation and bioavailability will be studied. Two-period pot experiments were performed to examine the transformation and bioavailability of Se and Cd fractions in response to the different agricultural residues derived-DOM, including chicken, ox and earthworm manures, rice straw, and straw mixed with each of the other three manures. Soil Se fractions were analyzed using the classical sequential extraction method ^[1]; soil Cd fractions were determined using the BCR sequential extraction protocol ^[2]; soil properties were analyzed ^[3]; and plant total Se and Cd were analyzed per Chinese GB5009.93-2017 and GB5009.15-2014, respectively.

Results and Discussion: The application of agricultural residue derived-DOMs application can increase soil chemical properties (soil pH, organic matter content and cation exchange capacity). The application of 2% chicken manure derived DOM had the greatest impact and enhanced the Se content in rice grains by 46.14% and decreased the Cd content in rice grains by 47.22%. Structural equation modeling (SEM) analysis revealed that this phenomenon was primarily attributed to the enhancement of the soluble Se fraction content (+54.81%). Conversely, the reduction in Cd content of grains was mainly due to the decreased oxidizable Cd fraction content (-20.97%) in the soil.

Conclusions: Overall, we concluded that the application of agricultural residues derived from DOM could activate Se and inactivate Cd in natural Se-Cd-rich paddy soils, in which the chicken manure derived-DOM gave the best effect. The increase in Se content in rice grains is primarily attributable to the capacity of DOM to modify soil chemical properties (specifically SOM, and soil CEC), thereby facilitating the transformation of diverse Se fractions into SOL-Se, consequently enhancing Se bioavailability. Conversely, the decline in Cd content in rice grains is primarily attributed to elevated soil pH and SOM levels following DOMs application, triggering the transformation of OXI-Cd into stable fractions, thereby reducing Cd bioavailability.

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Machine Learning-based Prediction of the Bioavailability of Selenium in Soil

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Introduction: Selenium (Se) is an essential trace element for the human body, and the soil-plant system serves as the primary pathway for human Se intake. The bioavailability of Se within the soil-plant system plays a crucial role in agronomic biofortification strategies^[1]. In recent years, machine learning technology has provided a new paradigm for accurate prediction of soil Se bioavailability based on its powerful nonlinear fitting ability^[2]. The purpose of this study was to use machine learning techniques to explore the main drivers of soil available Se content and predict soil available Se content.

Methods: a total of 2619 sample points from 12 provinces/autonomous regions in China were analyzed. These points cover a wide geographical range (103.3°E to 131.9°E longitude and 21.8°N to 47.8°N latitude) and encompass diverse land-use types, including drylands, paddy fields, tea plantations, orchards, and plantations. Multi-model analysis was utilized to investigate the influencing factors and prediction of soil available Se content.

Results and Discussion: Based on 11 influencing factors, such as soil total Se, pH, and soil organic carbon, we constructed a random forest model. The results showed that the model composed of these factors could explain 76.45% of the changes in soil available Se content. Among them, the total Se content of soil was the dominant factor controlling the available Se content of soil (importance = 34.15%, p < 0.05), followed by pH (importance = 22.60%, p<0.05) and the ratio of fungi to bacteria (importance = 22.46%, p < 0.05). This observation may be due to the fact that although Se in soil can exist in a variety of forms and valence states, the proportion of available Se varies from a small range ^[3]. Using the optimal model further, we evaluated the available Se



content in soil in 12 provinces/autonomous regions of China, based on 2619 samples (Fig. 1). The results showed that the soil available Se content in these areas was generally higher in the southeast (> 60 μ g/kg in Zhejiang) and lower in the northeast (< 20 μ g/kg in Heilongjiang & Liaoning). In addition, soil available Se content was also higher in the southwest of Hubei (> 60 μ g/kg). A high degree of fit between the predicted Se content and the measured Se content, implying that it was feasible to use total Se and some soil biochemical variables to predict soil available Se content.

Figure 1. Available Se content in soil of 12 provinces/autonomous regions in China.

Conclusions: The dominant factor influencing soil available Se is the total Se content in the soil. As the total soil Se increases, the concentration of available Se may increase significantly, although the proportion that is available can vary. Predictions of soil available Se levels indicate an overall distribution pattern characterized by higher concentrations in the southeast and lower concentrations in the northeast. Furthermore, using total Se combined with certain soil biochemical variables shows promise for predicting soil available Se content.

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Selenium-Enriched Cereals: A Comprehensive Study on Biofortification, Bioaccessibility, and Anti-Inflammatory Efficacy in Functional Food Applications

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Introduction: Selenium (Se) is a trace element crucial for maintaining cellular homeostasis, with roles in antioxidant defense, immune modulation, and reducing inflammation. It exists predominantly in organic forms like selenomethionine (SeMet) in selenium-enriched crops. Addressing Se deficiencies through biofortified crops cultivated in seleniferous regions presents an effective strategy to improve dietary Se intake. However, understanding the bioaccessibility and biological efficacy of Se in these matrices is essential.

Methods: A multi-method approach was employed, involving: (1) Selenium extraction and quantification from wheat, maize, and rice matrices using fluorescence spectrometry. (2) Analysis of Se bioaccessibility during simulated gastric and gastrointestinal digestion to understand its release from protein fractions such as albumins, globulins, glutelins, and prolamins. (3) Cell culture studies with RAW264.7 murine macrophages exposed to Se-rich extracts (± rMETase enzyme treatment) to evaluate anti-inflammatory and antioxidant responses. Key inflammatory markers (e.g. COX-2, mPGES-1) and antioxidant enzymes (e.g. GPx-1) were measured at the transcriptional and protein levels.

Results and Discussion: Cereals grown in seleniferous soils contained significantly higher Se levels compared to non-seleniferous controls (wheat: 122.9 μ g/g, maize: 26.5 μ g/g, rice: 19.7 μ g/g). Selenium was more bioaccessible during intestinal digestion than gastric digestion, with glutelin and prolamin fractions contributing the most to bioavailable Se. Selenium supplementation downregulated pro-inflammatory markers (COX-2, mPGES-1) and upregulated anti-inflammatory enzymes (GPx-1, H-PGDS) in macrophages. Enzyme-treated Se matrices (e.g. SeW + rMET) exhibited enhanced bioavailability and stronger biological effects. Selenium release and efficacy varied among cereals, with wheat demonstrating superior antioxidant and anti-inflammatory potential. The findings demonstrate the potential of Se-enriched cereals as functional foods capable of mitigating inflammatory responses. The enzymatic treatment of Se-rich matrices significantly enhances Se bioavailability, making them effective dietary supplements. However, matrix variability and Se concentration need careful regulation to minimize Se toxicity in the environment.

Conclusion: Selenium-enriched cereals provide a dual benefit of addressing Se deficiency and offering therapeutic potential against inflammation. Future studies should focus on optimizing processing techniques and developing tailored supplementation strategies for diverse populations.

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Effects of Soil Amendments on Selenium and Cadmium Accumulation in Rice in High Geochemical Background Areas

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Introduction: Selenium (Se) is an essential micronutrient with antioxidant and immune functions, while cadmium (Cd) is a toxic metal linked to kidney damage, bone disorders, and cancer [3]. In some regions, especially parts of China, soils derived from specific parent rocks are simultaneously enriched in Se and contaminated with Cd, posing challenges for crop safety and human health. Rice (*Oryza sativa* L.) is both a major dietary source of Se and a key pathway for Cd exposure. Its uptake of Se and Cd is influenced by soil properties and element speciation. Soil amendments, such as organic matter, chemical stabilizers, and biochar, can alter metal availability, reduce Cd uptake, and enhance Se bioavailability. The outcomes vary with site conditions and amendment types.

Methods: Field trials were conducted in Changshan County, Zhejiang, where paddy soils derived from Cambrian black shale are enriched in Se and Cd. Five amendments were tested: polyferric sulfate (PFS), leaf extract + PFS (LF), laccase-modified leaf extract (LL), laccase-modified leaf extract + PFS (LLF), and Fe–Mn-modified biochar (FMBC). Amendments were applied to the 0-20 cm soil layer at 0.2% (w/w), except FMBC at 0.5%. Untreated plots served as controls (CK). Each 4×4 m plot was isolated with plastic sheets to prevent cross-contamination. Amendments were mixed into the soil two weeks before sowing. Initial soil values were: total Cd 2.40 mg/kg, DTPA-Cd 0.88 mg/kg, and total Se 1.10 mg/kg. At maturity, soil and plant samples were collected. DTPA-Cd was measured for bioavailability; Cd fractions were extracted using the Tessier method. Se and Cd concentrations were analyzed following national standards.

Results and Discussion: The application of iron–manganese-modified biochar (FMBC) significantly increased soil pH, whereas other amendments had minimal effects. The high alkalinity of FMBC (pH 10.65) likely contributed to neutralizing acidic components in the soil. All amendments reduced soil redox potential (Eh), which may be attributed to anaerobic conditions induced by flooding, promoting the release of Fe^{2+} and Mn^{2+} , strong reducing agents that lower Eh. Additionally, increased soil moisture under treatments may have further contributed to Eh reduction. The amendments also enhanced soil CEC to varying degrees and effectively reduced the concentration of available Cd. The treatments shifted Cd speciation by decreasing the proportion of exchangeable Cd and increasing Fe–Mn oxide-bound and organically bound fractions, suggesting adsorption and complexation processes that stabilized Cd in less bioavailable forms. Concurrently, all amendments significantly increased the proportion of water-soluble Se. Furthermore, Cd content in brown rice was reduced by 63.88 - 87.63%, with FMBC showing the greatest reduction. In contrast, Se content increased by 6.11 - 63.35%, with PFS exhibiting the most pronounced enhancement.

Conclusions: This study assessed five soil amendments for remediating Se-rich, Cd-contaminated paddy soils in a high-Cd geological area. All amendments effectively enhanced Se availability and reduced Cd accumulation, though with varying efficacy. FMBC was most effective in reducing soil Cd, while PFS most significantly increased Se availability. These results offer a scientific foundation and practical guidance for the safe cultivation of rice in regions with coexisting Se enrichment and Cd contamination, contributing to improved food safety and sustainable agriculture.

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Selenium Impacted the Feedback of Soil Microbiota to Elevated Atmospheric CO₂ Stress

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Introduction: Closely tied to soil microbiota, selenium (Se) serves as a crucial element in improving microbial community organization ^[1,2]. Meanwhile, increased atmospheric CO_2 , a major factor in shifting climate patterns, profoundly affects soil ecosystem processes driven by microbial activity. However, the impacts of Se on the soil microbiota under elevated CO_2 levels has received little attention in previous studies. This research aimed to investigate the regulatory role of Se in shaping soil bacterial communities during short-term elevated CO_2 stress.

Methods: Soil samples were gathered each week from the elevated CO₂ level experiment. Soil properties including pH and moisture were measured. In addition, amplicon sequencing was performed on the DNA extraction of the gathered samples. The α -diversity and composition of the microbiota were determined using R software ^[3,4].

Results and Discussion: The soil pH from the elevated CO₂ treatment was observed to be significantly lower compared with that from the ambient CO₂ treatment (p < 0.05). However, the moisture between the two treatments showed no significant differences. Meanwhile, the α -diversity showed more sensitivity to the elevated CO₂ level compared to the microbial composition. The changes in the microbial diversity might relate to the change in soil pH under the elevated CO₂ level.

Conclusions: The elevated CO_2 level resulted in a noticeable decline in the values of soil pH and microbial diversity, yet the influence was slight after the application of Se.

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Study on the High-efficiency Utilization Threshold of Selenium in Paddy Soil

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Introduction: Selenium (Se) is an essential trace element for animals and humans, but its effect is, however, dependent on concentration. The Se ingested by the human body mainly comes from soil and enters the human body through the food chain, which has an impact on human health. There are various forms of Se in soil, among which available Se is a key parameter determining the mobility of Se in soil. In previous research studies, we also found that Se-enriched soil may not necessarily produce Se-enriched rice. In addition, although increasing soil and Se content through Se biofortification strategies can significantly increase the total Se content of rice, the proportion of inorganic Se is significantly higher than that of rice grown in Se-enriched soil, which causes potential risks for human health. Therefore, we aim to explore the corresponding soil Se content levels for producing high-quality Se rich rice with a high proportion of organic Se.

Methods: This study focused on Se-enriched soils of different origins in Zhejiang Province. Based on soil geochemical surveys and analyses of Se in rice, the content, forms, and migration transformation of Se in the soil rice system were elucidated. In addition, a species sensitivity model was used to derive the threshold for efficient utilization of Se in soil.

Results and Discussion: The difference in soil Se levels were due to geological background. There were significant differences in the correlation between Se content in rice and soil among Se-enriched soils of different origins. The Se forms in rice were mainly organic Se, and the differences of Se form characteristics in rice were mainly determined by the available Se content in soil. With the increase of available Se content in soil, the proportion of inorganic Se in rice showed an upward trend, but the proportion of selenocysteine gradually decreased and the proportion of selenomethionine gradually increased. Furthermore, the Se bioconcentration factors varied among different rice varieties, and the Se bioaccumulation factor of indica rice was significantly higher than that of japonica rice. Above all, the accumulation of Se in rice is a very complex process, influenced by both soil physicochemical properties and rice varieties.

Conclusions: Based on survey and analysis data in Zhejiang province, the soil total Se threshold for rice crops to reach a 90% Seenriched level was 0.68 - 5.13 mg/kg, and the soil available Se threshold was 0.06 - 0.43 mg/kg. When the available Se content in the soil was greater than 0.08 mg/kg, there was a 75% probability that the forms of Se in rice were all organic Se.

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Soil Available Selenium Content Regulated the Distribution Characteristics of Selenium Speciation in Rice Grains

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Introduction: Selenium (Se) is an essential micronutrient element, and Se soil bioavailability and the distribution characteristics of Se speciation in rice grains directly affect the nutritional quality and health effects of rice ^[1]. The migration and transformation mechanism and regulation of soil available Se to grain have become a research hotspot in the field of functional agriculture. The purpose of this study was to elucidate the effect of soil available Se content on the distribution characteristics of Se speciation in rice grains, and to provide a scientific basis for improving the quality and nutrient use efficiency of Se-rich rice.

Methods: In this study, Ruian City (RA) and Zhuji City (ZJ) in Zhejiang Province were selected as the study areas. Through natural investigation experiments, soil and corresponding rice grain samples were randomly collected. A total of 38and 50 samples were collected from the RA and ZJ areas, respectively. The available Se fractions in the soil samples were extracted using ammonium bicarbonate-diethylenetriamine-penta acetic acid. Selenium content and speciation analysis were determined by AFS and HPLC-ICP-MS, respectively. Data were analyzed by variance analysis (ANOVA) with SPSS Statistics 26. Data visualization used Origin 2019.

Results and Discussion: The available Se content in the soil of RA and ZJ ranges from 0.02 to 0.07 mg/kg and 0.04 to 0.42 mg/kg, respectively, with average values of 0.03 ± 0.01 mg/kg and 0.09 ± 0.06 mg/kg, respectively. Analysis of Se speciation in grains



showed that organic Se was the main speciation in grains from both regions. Our results also found that with an increase of soil available Se content, the proportion of selenocysteine (SeCys) decreased gradually ($R^2 = 0.24$, p < 0.05), and the proportion of selenomethionine (SeMet) gradually increased ($R^2 = 0.17$, p < 0.05) (Fig. 1). This observation is consistent with the previous research results ^[2], indicating that the addition of Se in the soil significantly increased the content of SeMet in rice grains, while reducing the content of SeCys^[2]. This is mainly due to the soil available Se content determines the total amount of Se that can be absorbed and utilized by rice crops, which in turn affects the distribution characteristics of Se speciation in rice grains^[3].

Figure 1. Relationship of the soil available Se concentration to the proportions of selenium speciation in rice grains.

Conclusions: Soil available Se content is closely related to the distribution of Se speciation in rice grains. With the increase of soil available Se content, the proportion of SeCys and SeMet in rice grains changed significantly, which confirmed that soil available Se content was the key controlling factor regulating the distribution of rice Se speciation.

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Research and Application of Grade Focus Fast-detection Based for Selenium-rich Soil

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Introduction: Traditional soil selenium (Se) surveys are conducted by testing soil samples. However, due to the complex nature of soil matrix, testing costs are relatively high. Furthermore, soil that meets Se-rich standards doesn't necessarily produce Se-rich crops^[1]. Therefore, the proposed GFFB (Grade Focus Fast-detection Based) method was to utilize XRF to detect total Se in grains for creating a stepwise rapid survey of soil Se enrichment in the study area.

Methods: Se-rich soil areas suitable for producing Se-rich rice were delineated based on grain Se content. Samples were collected from 6.67 hectares of non-Se-biofortified paddy fields in Shitai County, Anhui Province. The area was divided into 24 units, each with four sampling points. Firstly, bulk samples per unit were analyzed for total grain Se using atomic fluorescence spectrometry. Secondly, those units meeting the standard (GB/T 22499-2008) were subjected to rapid X-ray fluorescence spectrometry on two additional samples. Finally, units requiring further sampling were re-identified based on a coefficient of variation (CV) $\geq 20\%$ among samples within each unit.

Results and Discussion: The investigated 96 samples exhibited a mean Se content of 37.6 μ g/kg, a median of 35.0 μ g/kg, and a range of 15.5–75.5 μ g/kg. Approximately 38.00% of samples met the Se-enriched rice standard (GB/T 22499-2008). The coefficient of variation (CV) among units ranged from 2.77% to 42.18%, with 8 units exceeding CV > 20%^[2]. Interpolation of unit data generated the rice Se distribution map (Fig. 1a). For the 10 units meeting the Se-enriched standard, we further analyzed Se



42.1876, with 8 units exceeding $CV > 2076^{13}$. Interpolation of unit 0 units meeting the Se-enriched standard, we further analyzed Se content at individual sampling points to produce a hierarchical focus map (Fig. 1b). Based on CV values, supplementary sampling points were measured and plotted (Fig. 1c). The predictive map generated by Radial Basis Function Radial Basis Function (RBF) interpolation demonstrated high consistency with the full-sample map in characterizing Se-enriched zones ^[3]. Notably, this investigation method more precisely excludes areas incapable of producing Se-enriched rice, enhances the utilization efficiency of naturally Se-rich soils, and demonstrates superior accuracy and cost-effectiveness compared to comprehensive

sampling.

Figure 1. Distribution map of Se-rich rice grains created based on the RBF interpolation method.

Conclusions: This study based on GFFB method integrated RBF interpolation, we found that a high prediction accuracy was still achieved with a 43.3% reduction in sampling points, thus enabling accurate Se soil mapping through rice grain bioindicators. The GFFB method maximizes the accuracy of identifying selenium-rich areas, while reducing costs. Future work should expand validation across diverse agroecosystems and optimize for multi-crop use.

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Selenium and Trace Element Profiles in Wheat Cultivated in Saline-Alkaline Soils

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Introduction: The total area of saline-alkali farmland in China reaches 115 million mu, accounting for about 4% of the global saline-alkali soil area ^[1]. Generally, these areas are subjected to dual stresses of salt and alkali ^[2] and possess the soil characteristic of "high salt content" (Fig. 1, left). Soil salt ion analysis reveals that Cl^- is the dominant anion, while Na⁺, Ca²⁺ and Mg²⁺ are the primary cations constituting the major salt components in these soils (Fig. 1, right).



Figure 1. Soil salinity content and distribution in the profile (left) and ions content in the salt-salinity soils (right).

Results and Discussion: Wheat, as a staple food crop in China, is grown in cover millio-mu of saline-alkali land, where specific wheat varieties have developed strong mineral nutrient enrichment capabilities through adaptive evolution. Research in the Yangtze River Delta region indicates a significant positive correlation between soil carbonate content and selenium levels in wheat (r = 0.68, p < 0.01)^[3]. Huanghua dryland-alkali wheat is rich in trace elements such as Ca²⁺, K⁺, Fe²⁺, and Zn²⁺. This synergistic enrichment may be attributed to the high concentrations of cations and enhanced micronutrient availability in saline-alkali soils. The synergistic enrichment of calcium, potassium, iron, zinc, and other elements in saline-alkali wheat further enhances its nutritional value. For example, Nanpi alkali wheat demonstrates outstanding nutritional properties, with calcium, protein, and polyphenol content exceeding average test values. Additionally, certain saline-alkali wheat varieties are rich in antioxidants such as anthocyanins, which provide health benefits including anti-inflammatory and anti-aging effects^[4].

Conclusions: Soil is the foundation of agricultural production, and soil fertility is its core attribute. Therefore, wheat cultivation should be placed on the integration of utilization and conservation, with improving soil fertility and enhancing land productivity in saline-alkali areas.

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Effects of Dissolved Organic Matter on Selenium Availability and Regulatory Mechanisms in a Selenium-Enriched Red Soil-Navel Orange System

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Introduction: Selenium (Se) is an essential trace element for humans and a beneficial element for plants. In Se-enriched red soils, the widespread presence of high-content iron oxides exhibits strong affinity toward selenate and selenite ions in the soil^[1], constituting a major factor limiting Se bioavailability in selenium-enriched red soils. How to enhance Se bioavailability while developing Se-enriched farmland represents an important scientific question. Organic matter plays a crucial role in regulating the stability of iron-Se complexes and improving Se bioavailability in Se-enriched red soils. However, the effects of different organic matter sources - especially the most reactive dissolved organic matter (DOM) - on the stability of iron-selenium complexes in selenium-enriched red soils and the regulatory mechanisms controlling Se availability in soil-plant systems remain unclear.

Methods: This study investigated the Gannan selenium-enriched red soil-navel orange system through pot experiments. Total soil Se, Se speciation, DOM three-dimensional fluorescence, modern spectroscopic characterization techniques, and Se contents were periodically determined in various plant parts. We investigated DOM's effects on navel orange seedling growth and Se accumulation, along with the regulatory mechanisms of Se bioavailability in the soil-navel orange system.

Results and Discussion: Enhancement of Soil Se Availability by DOM: DOM significantly improves soil Se availability by driving the reductive dissolution of iron oxides. Rapeseed cake-derived DOM is more effective than cow manure-derived DOM in promoting the reductive dissolution of iron-selenium complexes, thereby releasing Se from the complexes and enhancing its bioavailability. Regulation of Se Absorption and Distribution in Plants by DOM: Cow manure-derived DOM systematically enhances Se accumulation efficiency in plants, with particularly pronounced effects in roots and secondary organs (such as secondary stems and leaves). This observation may be attributed to the abundance of aromatic protein-like substances in cow manure DOM, which facilitates short-term Se activation. In contrast, rapeseed cake-derived DOM is more conducive to the



transport of selenium to aboveground parts, which may be related to the higher proportion of humic acid-like substances in rapeseed cake DOM, which improves the soil environment and promotes longdistance Se transport within plants. Compositional Characteristics and Functional Differences of DOM: Three-dimensional fluorescence spectroscopy analysis reveals that rapeseed cake-derived DOM significantly increases the proportion of humic acidlike substances, while cow manure-derived DOM is rich in aromatic protein-like substances. These compositional differences determine the distinct roles of DOM in Se activation and transport processes.

Conclusions: This study not only reveals the regulatory mechanisms of DOM on Se transport but also provides an important theoretical basis for selecting appropriate DOM types and dosages to improve Se accumulation efficiency in crops, thereby promoting the sustainable utilization of selenium resources.

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Selenium Transport - Relevance of Selenoprotein P (SELENOP) for Human Health

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A large proportion of the world's population must be considered selenium (Se) deficient, but a clear definition of Se deficiency is lacking. Recent research based on observational and intervention studies has shown that the transporter selenoprotein P (SELENOP) can serve as a meaningful biomarker for selenium status and as a suitable indicator of Se supply and health-relevant deficiency at the population level. On a personal level, SELENOP indicates the acute bioavailability of the trace element and reliably reflects the Se intake from food or during supplementation. In disease, its decline correlates to severity and inversely to survival odds. Based on analytical studies, the presentation will highlight the importance of avoiding low SELENOP levels in the blood to reduce the risk of mortality, as observed in a large prospective study of healthy European adults. In addition, the importance of SELENOP for avoiding health decline in cardiovascular disease or reducing mortality risk in cancer will be presented. Finally, the effectiveness of supplementation to increase low SELENOP concentrations is discussed. All in all, it appears that nature, the environment and time have shaped SELENOP into an efficient transporter that ensures the supply of those cells that most urgently require the essential trace element due to their high metabolic activity and need for protection.

Selenium Status and Cancer Risk: Results from a Large European Cohort Study

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Introduction: Selenium (Se) may help prevent cancer development by countering oxidative and inflammatory processes implicated in carcinogenesis. We previously reported in the large, European Prospective Investigation into Cancer and Nutrition (EPIC) cohort that a higher Se status (as assessed by prediagnostic circulating concentrations of Se and its major transport protein, Selenoprotein P; SELENOP) was associated with lower risks of colorectal cancer (CRC)^[1] and hepatocellular cancer, ^[2] and that several common genetic variants in selenoprotein and Se metabolic pathway genes may affect CRC development ^[3]. However, there does not appear to be the same relevance for hormonal-related cancers, where we have observed no marked associations of Se status with either prostate ^[4] or breast cancer ^[5]. Although, we did show evidence that genetic variants in selenoprotein or wider Se-pathway genes were also associated with altered risks for these cancer types ^[4.5]. Additionally, the activity of the redox selenoenzyme glutathione peroxidase 3 (GPx3), as a third Se status biomarker, was associated with lower risk of premenopausal breast cancer ^[5].

Methods: In EPIC, we recently examined the association of gastric cancer (GC) risk with plasma Se status (Se, SELENOP, and GPx3) from 668 nested case-control pairs, while SNP variants across several selenoprotein and Se biosynthesis genes were also assessed in available DNA from most of these subjects. Blood samples and dietary/lifestyle questionnaire data were taken years before cancer diagnosis in the case group. Multivariable-adjusted logistic regression was used to calculate odds ratios (ORs) and 95% confidence intervals (CIs) for the association of Se status biomarkers and SNPs with GC risk.

Results and Discussion: Higher plasma Se concentrations were statistically significantly associated with a lower GC risk (4th versus 1st quartile, OR = 0.59, 95% CI: 0.40-0.85, $p_{trend} = 0.008$), with stronger associations observed in women (as we also found for CRC). SELENOP concentrations indicated a non-statistically significant inverse association, while GPx3 activity showed no apparent association with GC risk. Genetic analyses identified nominally significant associations ($p \le 0.05$) between SNPs in *SELENOS*, *GPx2*, and *SEPHS1* and GC susceptibility, although none retained significance after multiple testing adjustment. Of these, three *SELENOS* SNPs were also associated with altered GC risk in interaction with Se status.

Conclusions: Together, these findings highlight that an adequate Se status may help prevent the development of gastrointestinal cancers, and that selenoprotein (plus related Se pathway) gene variants could influence cancer risk alone or in combination with Se status. The results align with the hypothesis of a Se status in many European subjects that is insufficient for robust Se-mediated cancer prevention. However, they require validation in populations with varying Se status / availability.

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Selenium and Coenzyme Q₁₀ Supplementation Improves Thyroid Hormone Function, Cardiovascular Mortality, and Quality-of-life in an Elderly Population Low in Selenium

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Introduction: Selenium (Se) is essential for the metabolism of thyroid hormones. The selenoenzymes iodothyronine deiodinases (DIO) I, and II activate prohormone T4 to T3 in peripheral tissues, while DIO II and III inactivate T4 and T3. Selenoenzymes glutathione peroxidases and thioredoxin reductases protect the thyroid against oxidative cellular injury during synthesis of thyroid hormones. An insufficient Se intake and status may disturb thyroid hormone balance, with less peripheral conversion of T4 to T3 and an increased TSH concentration. The aim of the present sub-study of an elderly population low in Se and Coenzyme Q_{10} (Co Q_{10}) was to examine associations between Se and thyroid hormones in serum, and to determine if intervention with Se and Co Q_{10} might beneficially influence the thyroid hormone concentrations and further cardiovascular (CV) mortality and health-related quality-of-life (Hr-QoL).

Methods: The study population was recruited from a municipality in Sweden where all inhabitants aged 70-88 were invited in a prospective randomized double-blind placebo-controlled trial. 443 of them agreed to participate in a dietary supplementation project that involved taking 100 μ g Se yeast and 100 mg coenzyme Q₁₀ twice daily for four years. Free T3 (fT3), free T4 (fT4), reverse T3 (rT3), TSH were determined at inclusion and after four years. CV mortality was evaluated after 10 years, and Hr-QoL was evaluated after 48 months using Short Form 36 (SF-36). Pre-intervention plasma Se was low with a mean concentration of 67 μ g/L in the study population^[1].

Results and Discussion: The participants with the lowest Se concentration at baseline had significantly higher levels of TSH and lower levels of fT3 compared to subjects with the highest Se concentrations. Supplementation with Se and CoQ10 increased fT3 and rT3, decreased fT4, and diminished the increased TSH levels in comparison with placebo treatment (p=0.03, all). In the placebo group, TSH and fT4 values above the median were associated with a significant increase in 10-year CV mortality, while this difference disappeared in those supplemented. Similarly, TSH> median and fT3<median was associated with a decline in mental Hr-QoL measures vs. TSH < and fT3> median in the placebo group during four years of follow-up in the dimensions of "vitality", "bodily pain", "social function" and in "physical composite scores", but this difference was removed in the supplemented group. Thus, Se and CoQ₁₀ supplementation significantly increased fT3 levels in association with better health outcome ^[2].

Conclusions: The initial moderately deficient Se status was associated with an impaired thyroid function and the changes in thyroid hormone levels upon supplementation can be explained by increased deiodinase activity that converts thyroxine (T4) to T3 locally supporting thyroid hormone activity. Supplementation with Se and CoQ_{10} not only improved thyroid hormone function but also had beneficial clinical effects with reduced CV mortality and improved Hr-QoL outcomes. Our findings should stimulate clinicians treating patients with thyroid disorders to along with thyroid hormones assessment also determine Se concentrations, in order to optimize treatment.

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Selenium Supplementation Strategies in Sheep: Effects on WB-Se Species

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Introduction: The focus of our selenium (Se) research is to improve health, prevent diseases, and increase productivity in sheep flocks across Oregon, where soils are deficient in Se. Deficiencies in the Se content of forages are reflected in whole-blood (WB) Se concentrations in sheep consuming these forages. WB-Se concentrations from 56 farms in Oregon showed that 16 % had deficient WB-Se concentrations in 100% of the ewes tested, and 23 % had > 50 % of ewes tested with WB-Se deficiency. Thus, subclinical Se deficiency is quite common in Oregon flocks of sheep, even when owners are providing their animals some form of Se supplement. We have previously shown that feeding selenate-fertilized forages not only increased WB-Se concentrations in sheep and cattle, but also improved their productivity and health. Other methods of Se supplementation include giving selenomethionine (SeMet) from Se-yeast (mixed with water and administered as a drench, in boluses, or as topdressing on feeds), and providing inorganic Na selenite in salt-mineral mixtures. We hypothesized that the form of Se supplement administered can influence Se species in blood, and that different Se species may confer unique health benefits.

Methods: Blood samples were collected from ewes on 4 farms known from the prior survey study. All ewes had adequate total WB-Se concentrations. Farms differed in Se supplementation strategies: ewes from farm #1 grazed Se-biofortified pastures, were drenched weekly with SeMet enriched yeast, and had access to free choice salt-mineral mix containing 200 ppm Na selenite. Ewes from farm #2 grazed Se-fortified pastures and received mineral mix containing ≥ 24.2 mg/kg Se from Se-yeast and selenite. Ewes from farms #3 and #4 had access to free choice salt mineral mix containing ≥ 24.2 mg/kg Se from Se-yeast and selenite. Ewes from farms #3 and #4 had access to free choice salt mineral mix containing 165 mg/kg and 90 mg/kg sodium selenite, respectively. To measure WB-Se, blood was mixed with an indium solution as an internal standard and digested in concentrated H₂O₂ and distilled nitric acid in plastic tubes at 90 °C until a clear digest formed. The solution was diluted 122-fold prior to ICP-MS analysis at the WM Keck Collaboratory for Plasma Spectrometry at Oregon State University. The analyses were conducted on a ThermoScientific iCAP-RQ quadrupole ICP-MS configured with a robust skimmer cone insert and operated in standard (STDR) and kinetic energy discrimination (KEDR) modes. ⁷⁷Se was analyzed in KEDR mode to mitigate interference. Selenium species in WB samples were extracted using a methanol, chloroform, and water extraction to isolate 60% MeOH soluble Se compounds for subsequent characterization. The SAX-HPLC–ICPMS Se speciation method was used to quantify the four most abundant Se species: inorganic selenate and selenite, and the proteinogenic selenoamino acids SeMet and SeCys, the latter which eluted as the dimer selenocystine (SeCys2). Using total WB-Se concentrations, quantities of each of the Se species were determined.

Results and Discussion: Total WB-Se and percentage of Se species (means) for sheep from each farm described above are shown in the table below. Selenium is incorporated into the amino acid methionine in forages or yeast to make SeMet, and when this is consumed by livestock, WB-Se appears mainly in the form of Se-Met. Conversely, consuming inorganic Se sources results in much higher concentrations of WB-SeCys. Proteins and enzymes containing SeMet act as better redox catalysts and antioxidants than their methionine-sulfur-containing analogues, although not as strong as SeCys-containing glutathione peroxidases or thioreductases.

	WB-Se	SeCvs2	SeO ₃	SeMet	SeO ₄	SeCvs2	SeO ₃	SeMet	SeO ₄
Farm ID	(ng/mL)	5	5			(ng/mL)	(ng/mL)	(ng/mL)	(ng/mL)
1	457	16.8%	0.4%	78.6%	4.2%	76.7	1.9	359.4	19.0
2	353	26.9%	0.6%	63.1%	9.4%	94.7	2.1	222.7	33.3
3	362	78.8%	1.6%	15.3%	3.4%	285.2	5.8	55.6	12.3
4	212	70.7%	1.0%	22.6%	3.2%	149.8	2.0	47.9	6.8

Conclusions: Selenium supplementation in the form of SeMet results in greater WB-SeMet species as a percentage of total WB-Se. Because of its organic form, SeMet is less toxic than inorganic forms. SeMet can be incorporated into body proteins and released with protein turnover providing greater body reserves of Se. In addition, the redox and antioxidant activity of SeMet-containing proteins could have major physiological consequences ^[1].

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Molecular Mechanisms of Selenium Deficiency Induced Oxidative Stress in Cartilage Injury of Kashin-Beck Disease

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Introduction: Kashin-Beck disease (KBD) is a food-sourced osteoarthropathy caused by environmental factors, environmental response genes, and their interaction with selenium (Se) deficiency is an environmental risk factor for KBD^[1]. Studies showed that oxidative stress and the Notch signaling pathway play vital roles in regulating cartilage development and occurrence, as well as chondrocyte proliferation and differentiation. However, the exact mechanisms in KBD remain unknown^[2]. This study aims to explore the mechanism of Se deficiency-induced oxidative stress in the pathogenesis of KBD and further search for new molecular targets for the early diagnosis, prevention, and treatment of KBD.

Methods: The Se contents in the flour and plasma samples were determined using the hydride atomic fluorescence spectrometry (AFS) assay technique. The mRNA levels in KBD patients and cultured chondrocytes were detected via qRT-PCR. The chondrocyte injury models were established by Se deficiency and tert-butyl hydroperoxide (tBHP), respectively. The viability of the chondrocytes was detected using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay. Apoptosis and necrosis rates were assessed using Hoechst 33342/PI and Annexin V-FITC/PI, respectively.

Results and Discussion: The findings revealed that the Se levels in both the flour and plasma of the KBD group were significantly lower than those in the control group, indicating Se deficiency in both the internal and external environments of KBD patients. In injury models induced by Se deficiency, GPX activity was markedly decreased, while ROS levels were elevated, confirming the presence of oxidative stress. In contrast, Se supplementation led to a notable increase in GPX activity and a reduction in ROS levels, suggesting that antioxidant capacity was effectively restored under Se supplementation. Further analysis showed that the Notch signaling pathway (NOTCH1/JAG1/HES1) was significantly upregulated in KBD patients. Correlation analysis revealed a negative association between *NOTCH1* expression and the anti-apoptotic gene *BCL2*, and a positive association with the pro-apoptotic gene *BAX*, suggesting that activation of the Notch/Hes1 pathway is closely linked to chondrocyte apoptosis in KBD. Consistently, the expression of the Notch signaling pathway increased, and chondrocyte apoptosis and necrosis increased in injury models induced by Se deficiency and tBHP, while Se supplementation could reverse this effect, which was like the results observed in KBD patients.

Conclusions: Decreased plasma Se levels in KBD patients may be related to low dietary Se intake. Selenium deficiency contributes to cartilage injury in KBD by enhancing oxidative stress and subsequently activating the Notch signaling pathway, and appropriate Se supplementation could reverse it.

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Dietary Supplementation with Selenium Yeast Mitigates Diquat-Induced Oxidative Damage in Oviductal Magnum of Hens

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Introduction: Oxidative stress can lead to decreased growth and reproductive performance in poultry. Nuclear factor erythroid 2-related factor 2 (Nrf2), a critical transcription factor, regulates antioxidant protein synthesis, neutralizes reactive oxygen species (ROS), and mitigates toxic metabolite accumulation, thereby enhancing cellular antioxidant capacity ^[1]. Key enzymes such as total superoxide dismutase (SOD) and catalase (CAT), produced via Nrf2 activation, scavenge excess ROS and attenuate oxidative stress severity ^[2]. Furthermore, the Nrf2 pathway modulates antioxidant defense systems by activating the antioxidant response element (ARE) of hemeoxygenase-1 (HO-1) ^[3]. Organic Se (Se) (e.g. Se yeast, SeY), a low-toxicity organic source, demonstrates superior bioavailability and enhances systemic health outcomes compared to the inorganic Se ^[4]. And the SeY integrates into yeast cellular proteins, enabling efficient absorption and tissue storage to prevent short-term deficiencies. Although selenium's capacity to reduce oxidative damage is well-established in mammalian systems, its effects on avian reproductive organs remain underexplored. This study investigates the protective mechanisms of dietary SeY supplementation against magnum oxidative damage in Kangle yellow hens, aiming to elucidate organic selenium's role in reproductive health.

Methods: A total of 60 healthy 180-day-old Kangle yellow hens with similar body weights (averaging 1.1 ± 0.1 kg) from the same batch were randomly divided into four treatment groups, each group containing 15 healthy adult hens. After 14 days of dietary treatment, all groups received a single intraperitoneal injection (0.9% NaCl or DQ) and continued their respective diets for an additional 3 days. On day 17, hens were euthanized via carotid artery exsanguination, and magnum tissues of the oviduct were collected. The total experimental duration was 17 days. Enzyme activity assays, H.E. staining, Immunohistochemistry, qRT-PCR and Western blot were used in this study.

Results and Discussion: The results showed that dietary supplementation with SeYn effectively alleviated DQ-induced oxidative damage in the oviductal magnum. This alleviation included the mitigation of damaged mucosal structure, reduced abundance of mucosal proliferating cells, and decreased expression of genes related to reproductive performance. Specifically, dietary supplementation with SeY counteracted DQ-induced oxidative stress by activating the Nrf2/HO-1 signaling pathway, enhancing the activity of antioxidant enzymes such as catalase (CAT) and total superoxide dismutase (T-SOD), and reducing the concentration of malondialdehyde (MDA). Moreover, dietary supplementation with SeY inhibited DQ-induced mucosal cell apoptosis by regulating the expression of apoptosis-related genes and proteins (Bax, Bcl-2, and Caspase3), as well as inflammation-related factors (NF- κ B and p53). Additionally, dietary supplementation with SeY restored proliferative capacity of mucosal cells by promoting the expression of the proliferation-related protein PCNA. The findings signify that dietary supplementation with SeY effectively safeguards the oviductal magnum against DQ-induced damage through mechanisms involving the reduction of oxidative stress, inhibition of apoptosis, promotion of proliferation, and the preservation of reproductive performance in hens.

Conclusions: Utilizing qRT-PCR, HE staining, immunohistochemistry, and Western blot analyses, this study demonstrated that SeY alleviates DQ-induced oxidative damage in the magnum tissue of Kangle yellow hens, reduces apoptosis, and restores the expression of reproductive regulatory genes.

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Selenium and Toxic Elements in Chinese Marketed Selenium-enriched Rice: A Health Risk Perspective

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Introduction: Selenium (Se) is an essential micronutrient, with rice being a major dietary source. However, Se-enriched rice may co-accumulate toxic elements, raising potential health concerns. In this study, 54 Se-enriched rice samples were collected from 14 provinces in China to quantify Se, arsenic (As), and cadmium (Cd) concentrations, and to assess their associated exposure and health risks.

Methods: The concentrations of Cd and As were determined by the high-precision portable X-ray fluorescence spectrometer (HDXRF) (XR700, JP Scientific, China), while the concentration of total Se was determined by the high-precision portable X-ray fluorescence spectrometer (HDXRF) (JP500, JP scientific, China)^[1].

Results and Discussion: The average concentration of Se was 0.13 ± 0.14 mg/kg, with a range from < LOD to 0.59 mg/kg in Seenriched rice, indicating a wide variation of Se concentration in Se-enriched rice in China (Fig. 1). Among the 54 Se-enriched rice samples, 46.3% samples of Se concentration in rice satisfied the standard (0.04 - 0.30 mg/kg), while there were 37.1% samples below the lower limited value (0.04 mg/kg). The average concentration of As was 0.11 ± 0.04 mg/kg, ranging from 0.03 to 0.20 mg/kg, and the concentration of Cd ranged from < LOD to 0.39 mg/kg, with an average value of 0.073 ± 0.071 mg/kg. The average EDI of Se was $33.3 \pm 34.8 \ \mu g/day$ and ranged from 0.59 to 154.4 $\mu g/day$, of which 20.4% population satisfied the recommended lower intake level of 60 $\mu g/day$ through Se-enriched rice consumption. All mean HI values were lower than 1, suggesting a low health hazard for As and Cd via Se-enriched rice consumption.



Figure 1. The geographical distribution of the sampling sites and the distribution of As, Cd, and Se concentrations in Se-enriched rice in China, respectively. The units were milligrams per kilogram.

Conclusions: The average concentrations of As and Cd in Se-enriched rice did not exceed the limit values of national food safety standards. Moreover, most of the Se concentration met the standards for Se-enriched rice within the range of 0.04 to 0.3 mg/kg. We noted that several samples of As and Cd were over the safety limit for the high geological background.

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Effect of Yeast Selenium and Boron on Digestive Organ Development in Broilers

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Introduction: Selenium (Se) Yeast selenium (Se) has been demonstrated to significantly enhance growth performance and antioxidant capacity in broilers. When combined with appropriate boron (B) supplementation, it further promotes the development of immune system ^[1]. The impact of their combined application on the development of digestive organs and the underlying mechanisms remains unclear. This study investigates the effects of dietary yeast Se supplemented with graded B levels on the development of the proventriculus, gizzard, and intestinal segments in broilers.

Methods: A total of 1,200 one-day-old healthy Arbor Acres (AA) broilers were randomly allocated into five groups using a completely randomized design: a control group and four experimental groups (I–IV), with 24 replicates per group and 10 birds per replicate. The control group was fed a basal diet, while Group I received the basal diet supplemented with 0.4% yeast Se. Groups II–IV were further supplemented with 5, 10, and 15 mg/kg B, respectively, in addition to the yeast Se. The trial lasted for 42 days.

Results and Discussion: The At 21 days, compared with the control group, Group II exhibited significant increases (p<0.05) in liver index, proventriculus index, and ileum index. Group III showed significantly higher (p<0.05) duodenum, jejunum, and ileum weights. By 42 days, compared with the control group, Groups II and III displayed significantly higher liver weight and liver index (p<0.05). Group II exhibited significantly increased proventriculus weight (p<0.01) and gizzard index (p<0.01), while Groups I–III showed elevated proventriculus index (p<0.05). Group III demonstrated significantly higher jejunum and ileum weights (p<0.05). When compared to Group I at 42 days, Groups II and III had significantly higher liver weight (p<0.05), and Group II showed significant increases in proventriculus weight, gizzard weight, gizzard index, and duodenum weight (P<0.05). These results indicate that yeast Se significantly enhanced the 42-day proventriculus index and moderately improved ileum length, while positively influencing liver and proventriculus, gizzard, duodenum, and jejunum weights at 42 days, though early-stage effects were less pronounced. The findings suggest that yeast Se combined with moderate B levels promotes digestive organ development, whereas excessive B may exert detrimental effects, corroborating previous research ^[2]. The synergistic mechanism may involve bioactive components enhancing hepatic glutathione-S-transferase (GST) activity, while suppressing GST-free radical activity, thereby improving digestive function.

Conclusions: The study demonstrates that the combined supplementation of yeast Se with optimal B levels significantly enhances liver, proventriculus, and gizzard weights, as well as organ indices, in 42-day-old broilers, thereby promoting digestive organ development and improving digestive function. The most effective dosage combination was observed with 0.4% yeast Se supplemented with 5 - 10 mg/kg B.

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The Mechanism of Selenium Metabolism-Related Genes in Patients with Osteoarthritis Based on Bioinformatics

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Introduction: Recent studies have shown that nano-selenium (Se) particles in hydrogel systems can address joint issues ^[1], improve subchondral osteosclerosis, reduce inflammation and pain, and delay osteoarthritis (OA) progression ^[2]. This study aims to explore the molecular mechanism of Se metabolism-related genes in OA via bioinformatics, screen key diagnostic markers, and analyze their association with the immune microenvironment.

Methods: The data were from GSE51588, GSE57218, GSE55235, GSE1919 and GSE253198. Selenium metabolism genes were identified from literature and GeneCards. Limma packet analyzed the difference between OA and normal group, and WGCNA identified disease-related genes. DEG1, DEG2, WGCNA and Se metabolism genes were taken for GO/KEGG intersection gene enrichment and PPI network analysis. Key genes were selected by LASSO regression and SVM-RFE, and their respective diagnostic value and relationship with the immune response were evaluated. In addition, single cell trajectory analysis and cell communication were conducted.

Results and Discussion: 23 intersection genes closely related to OA and Se metabolism were identified. GO/KEGG enrichment showed that the characteristic genes regulated extracellular matrix, oxidative stress and other pathways. PPI network revealed 57 interactions in 20 of the 23 intersection genes. CIBERSORT showed that the proportion of M2 macrophages and activated fibroblasts increased, and endothelial cells decreased in OA. Five Se metabolism genes, including COL1A2, GPX3, MMP9, TGFB3 and TNFSF11, were screened by machine learning. These 5 genes have high diagnostic reference value and are significantly related to Th17 cells, Treg cells and other immune cells. Single cell analysis showed that fibroblasts had the highest Se metabolism score, and the pathways of HYPOXIA and CHOLESTEROL_HOMEOSTASIS were inhibited in OA. Fibroblasts strongly interacted with macrophages through the PDGFC-PDGFRA ligand receptor pair. COL1A2 and GPX3 play a role in the process of cell differentiation.

Conclusions: Selenium metabolism genes COL1A2, GPX3, MMP9, TGFB3 and TNFSF11 can be potential diagnostic markers for OA. COL1A2 and GPX3. Importantly, they regulate immune microenvironment and intercellular communication in OA progression, offering new ideas for OA research and therapy.

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Prevotella copri Improves Selenium Deposition and Meat Quality in the Longissimus Dorsi Muscle of Fattening Pigs

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Introduction: Selenium (Se) is an essential trace element that plays a vital role in determining meat quality. Although the gut microbiota has been implicated in Se metabolism, its potential influence on Se accumulation in muscle tissue remains unclear. This study aimed to identify specific gut microbial taxa associated with Se deposition in muscle and to evaluate their effects on meat quality.

Methods: Colonic microbial compositions were compared between fattening pigs with high (HSE) and low (LSE) Se content in the longissimus dorsi (LD) muscle, revealing a significantly higher abundance of *Prevotella copri* in the HSE group. To investigate the role of *P. copri*, sixteen fattening pigs with an average initial body weight of 61.02 kg were fed either a basal diet or a basal diet supplemented with *P. copri* (1.0×10^{10} CFU/kg feed) for 45 days. Selenium and selenoprotein P (SelP) levels in serum and muscle ^[1], antioxidant indices in serum (GPX, T-AOC, SOD, MDA), and meat quality parameters (drip loss, pH, and color)^[2] were assessed. Colonic contents were analyzed by 16S rRNA gene sequencing ^[3].

Results and Discussion: Selenium content was positively correlated with SelP levels in the LD muscle of fattening pigs. Dietary supplementation with *P. copri* significantly increased Se and SelP concentrations in both serum and LD muscle. Moreover, *P. copri* enhanced serum antioxidant capacity, as evidenced by increased glutathione peroxidase activity and decreased malondialdehyde levels. Meat quality was also improved, as indicated by reduced drip loss, higher pH at 24 hours postmortem, and increased redness (a value). The intestinal abundance of *P. copri* was positively associated with Se deposition in muscle, suggesting that *P. copri* promotes Se accumulation, strengthens antioxidant defenses, and contributes to improved meat quality in fattening pigs.

Conclusions: This study demonstrates that a higher intestinal abundance of *P. copri* is significantly associated with increased Se and SelP levels in the LD muscle of fattening pigs. Dietary supplementation with *P. copri* enhances Se status and improves meat quality, highlighting its potential as a novel nutritional strategy to optimize pork quality.

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Enhancing Male Reproductive Cell Viability: The Role of Se-Methylselenocysteine and Potential Molecular Mechanisms in Male Reproductive Cells

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Introduction: Male infertility, a critical global health issue, impacts roughly 8-12% of couples globally, with male factors contributing to approximately half of these cases^[1]. Abnormal spermatogenesis is a major cause of male infertility, and excessive reactive oxygen species (ROS) and associated oxidative stress, are significant contributors. Selenium (Se), an essential trace element, plays a crucial role in normal spermatogenesis and male fertility. Different Se species, including inorganic Se, such as Se (VI) and Se (IV), and organic Se, including methylselenocysteine (MeSeCys), selenocysteine (SeCys), selenocystine (SeCys₂), and selenomethionine (SeMet), have varying bioavailability within the human body. Among these, MeSeCys is the major Se species in *Brassicaceae* selenium-enriched plants^[2] and has been shown to have beneficial effects on male reproductive improvement, antitumor, antioxidant, anti-inflammatory and cognitive deficits improvement. This study sought to explore the effect of MeSeCys on male germ cell lines GC-1spg (GC-1) and GC-2(spd)ts (GC-2), which mimic early stage of spermatogenesis, to provide a theoretical foundation for the advancement of MeSeCys-enriched foods and their application in promoting male reproduction.

Methods: Cell viability and proliferation was assessed using the CCK-8 and EdU assays. RNA sequencing and bioinformatics analysis were employed to identify differentially expressed genes and pathways. Cell transfection was performed to knockdown or overexpress specific genes. Q-PCR and western blotting were used to analyze gene and protein expression levels.

Results and Discussion: Of the five common Se species (MeSeCys, SeCys₂, SeMet, SeCys, and Na₂SeO₃), MeSeCys demonstrates the most potent effect in enhancing GC-1 and GC-2 cell viability, with GC-1 cells viability reaching 179.24% and GC-2 cells viability reaching 126.81% at a concentration of 75 µmol/L after 24 h of treatment. MeSeCys raised intracellular GSH levels in both cells, with a 41.51% increase in GC-1 cells and a 35.91% increase in GC-2 cells, correlating with greater viability in GC-1 cells. RNA-sequencing, bioinformatics analysis, and cell experiments were combined to identify the common and specific mechanisms underlying the significant increase in GC-1 cell viability. To be specific, in GC-1 and GC-2 cells, MeSeCys enters via the SLC7A11 or LRP8 channel, and upregulates selenoprotein synthesis and boosts GSH synthesis through the glutathione metabolism pathway, enhancing cell viability by improving redox balance. Specially, in GC-1 cells, MeSeCys activates the MAPK-mTOR pathway, increasing *Aft4* expression to further regulate GSH and intracellular redox balance, resulting in higher cell viability. These results indicate that MeSeCys has a positive impact on male reproductive cells.

Conclusions: This study shown that MeSeCys enhance male reproductive cell viability. It modulates selenocompound and glutathione metabolism in both male reproductive cells, and specially regulates intracellular redox balance via MAPK-mTOR signaling in GC-1 cells. These experimental findings not only reveal the molecular mechanisms underlying the contributions of MeSeCys on male reproductive cells but also underscore its potential as a promising nutritional supplement for improving male reproductive health.

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Protective Effect of Selenium-Yeast on Diquat-induced Oxidative Stress and Cell Apoptosis in Kangle Chicken

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Introduction: Selenium (Se) has antioxidant properties that can activate the antioxidant system, reduce oxidative damage, and enhance the oxygen-carrying capacity of erythrocytes. Is there a potential role of Se of protecting chickens exposed to residual herbicide in animal feed? This question is important to address since Diquat (1,1'-ethylene-2,2'-bipyridine dibromide) is the third most-commonly used toxic herbicide for modern agriculture in the world.^[1]

Methods: 180-day old Kangle chickens with similar body weights were randomly divided into different treatment groups: I: control, II: oxidative stress, III: oxidative stress + Se, IV: Se. Tissue samples from the liver, lung, kidney, and small intestine were collected at the end of the experiment. ELISA assays were conducted to detect the activities of superoxide dismutase, malonaldehyde (MDA), glutathione peroxidase (GSH), and catalase (CAT). The pathological changes in tissues were measured, qPCR and western blot determined the expression of apoptosis-related genes and proteins were conducted. Effects of Se-yeast on oxidative stress and apoptosis in liver, lung, kidney, and small intestine of Kangle chicken were determined.

Results and Discussion: The results indicated that compared to Group II, Group III significantly increased SOD activity in the liver, lung and ileum, GPx activity in liver and lung, and CAT activity in the liver, lung, kidney, jejunum, and ileum. Tissue dissection and HE staining of the collected chicken organs indicated that daily feeding of the Se diet improved liver, kidney, and small intestine lesions. qPCR results demonstrated that the relative expression of the apoptotic genes NF- κ B, Caspase 3, and Bax was down-regulated, while the expression of the Bcl-2 gene was up-regulated in each tissue. Western blot analysis demonstrated that Se decreased the expression of related apoptotic proteins in liver and lung tissues. Therefore, adding Se-yeast into chicken feed can alleviate oxidative stress caused by intraperitoneal injection of diquat, and enhance the activities of antioxidant enzymes in chicken, which can inhibit cell apoptosis by reducing the relative expression of apoptotic proteins, thereby lessening the body damage caused by oxidative stress.

Conclusions: Dietary supplementation with 0.5 mg Se/kg as Se-yeast can alleviate oxidative stress caused by intraperitoneal injection of 15 mg/kg Diquat. The practice can enhance the activity of antioxidant enzymes, increase the level of antioxidants, and reduce the amount of oxidized products in Kangle chickens, which mitigate the organ damage caused by oxidative stress.

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Study on the Protective Effect of Selenium Enriched Yeast Against Cadmium Induced Liver Injury in Chickens

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Introduction: Selenium (Se)-enriched yeast (SeY) is one of the most bioavailable organic Se compounds. SeY possesses significant antioxidant properties and can scavenge excess reactive oxygen species (ROS) and protect cells from structural damage and apoptosis by increasing the body's antioxidant capacity ^[1]. With the acceleration of industrialization and agricultural modernization, the heavy metal cadmium (Cd) poses a serious threat to poultry health through environmental pollution and residual contamination in feed ingredients ^[2]. This study aimed to investigate the antagonistic effect of SeY against Cd-induced liver injury by establishing a Cd-induced liver damage model in chickens.

Methods: A total of 180 chickens were randomly assigned to six groups, with 6 replicates in each group. The blank control group (CON) received oral gavage of 0.9% physiological saline solution. The model group received oral gavage of 35 mg/kg/day cadmium chloride (CdCl₂). The SeY-alone group received 0.6 mg/kg/day SeY. The low, medium, and high-dose SeY groups received 0.3 mg/kg/day, 0.6 mg/kg/day, and 0.9 mg/kg/day SeY, respectively. On day 7, 2 hours after of all groups except the CON group received an oral gavage of 35 mg/kg/day CdCl₂ solution. During the trial, chicken growth was monitored, daily body weight was recorded, and average weight gain was calculated. Twenty-four hours after Cd administration, chickens were euthanized via cardiac puncture. Liver tissue was collected to calculate the liver index. Serum levels of alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (AKP) were measured using commercial assay kits. Liver tissue samples were also subjected to histopathological examination.

Results and Discussion: Following CdCl₂ administration, chickens in the model group exhibited depression, sluggishness, and reduced appetite. Compared with the CON group, the model group showed a highly significant decrease in average weight gain and a highly significant increase in liver index. Serum levels of ALT and AST were also significantly higher in the model group than in the CON group. Histopathological examination revealed extensive inflammatory infiltration, severe ballooning degeneration of hepatocytes, and widespread hepatic necrosis. Compared to the model group, the low dose SeY group showed a significant increase in average weight gain, while the medium and high dose groups exhibited highly significant increases. The medium dose SeY group showed a significant reduction in liver index. All SeY treatment groups significantly reduced serum ALT levels and highly significantly reduced serum AST levels. Pathological changes in liver tissue were also markedly ameliorated by SeY supplementation. These results indicate that SeY effectively alleviates Cd induced liver injury by modulating organ indices, regulating serum transaminase levels, and intervening in the pathological changes induced by Cd exposure in liver tissue.

Conclusion: This study confirms that SeY can suppress the elevation of organ indices and mitigate liver function damage caused by Cd exposure in chickens. SeY intervention reversed the histopathological changes induced by Cd exposure. These findings provide an experimental basis for SeY in the prevention and treatment of Cd poisoning in poultry and offer new insights into the role of SeY in counteracting heavy metal toxicity.

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Twenty-year Clinical Practice Report on Pharmaceutical Grade Selenium Yeast

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Introduction: Selenium (Se) is an essential trace element for the human body, and it exerts various biological roles in the form of selenoproteins in the human body. The main selenoprotein families include glutathione peroxidase, thioredoxin reductase, and iodothyronine deiodinase. Glutathione peroxidase has the function of oxidoreductase, which can protect cells from oxidative stress. Thioredoxin reductase is a component of the cellular redox system and exists in various tissues and organs, playing a crucial role in cell growth and differentiation. Iodothyronine deiodinase catalyzes the conversion of T4 to T3, which is the main source of T3 in the body.

Methods: In the past two decades, significant progress has been made in the application of Se yeast Capsules (NDCH20052532, Wuhu Huaxin Biopharmaceutical Co., Ltd) has been active in the prevention and treatment of thyroid diseases and in the management of precancerous lesions. In this regard, Se yeast capsules treatment has been recommended in multiple expert guidelines for treating thyroid diseases.

Results and Discussion: Clinical studies have shown that low Se increases the incidence rate of thyroid diseases, and Se supplementation can reduce the risk of thyroid diseases globally. A large amount of empirical evidence shows that Se supplementation can reduce the levels of TPOAb and TGAb, and improve the condition of patients [1]. The decrease in blood Se levels is closely related to tumors. Selenium yeast capsules can prevent cancer and cancer, prevent the progression of gastrointestinal precancerous diseases, and prevent the occurrence of cancers such as gastric cancer, liver cancer, and colorectal cancer^[2].

Conclusions: For patients with thyroid diseases, it is recommended to supplement selenium yeast capsules to reduce antibody levels and improve the patient's condition. For patients with precancerous diseases, supplementing selenium yeast capsules can prevent the occurrence of cancer. Selenium yeast capsules have definite clinical efficacy and high safety.

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Eco-Friendly Synthesis and Biomedical Applications of Exopolysaccharide-Capped Selenium Nanoparticles: Antimicrobial, Antioxidant, and Therapeutic Insights

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Introduction: The growing demand for sustainable and effective biomedical materials has led to innovative approaches to combat bacterial infections and promote healing. Selenium nanoparticles (SeNPs) represent a promising solution due to their potent antimicrobial, antioxidant, and anticancer properties. These nanoparticles, synthesized using eco-friendly biological methods, demonstrate biocompatibility and stability, making them suitable for various applications in health care. This study explores the development and functionalization of SeNPs using exopolysaccharides (EPS) derived from the endophytic fungus *Nigrospora guilinensis*, focusing on their antimicrobial, antioxidant, and anticancer properties ^[1,2], along with their integration into biomedical substrates like cotton gauze for wound healing applications.

Methods: SeNPs were synthesized using EPS as a capping agent and characterized using techniques like UV-Vis spectroscopy, FTIR, XRD, TEM, and DLS to evaluate their size, morphology, and surface properties. Cotton gauze (CG) was cationized with chitosan (CH) to enhance the adhesion of SeNPs, producing EPS-SeNPs@CH_CG functionalized gauze. The bioactivity of EPS-SeNPs was tested against Gram-positive and Gram-negative bacteria (*Staphylococcus aureus* and *Escherichia coli*) and fungal strains using protein and polysaccharide leakage assays and antioxidant assays like DPPH and ABTS. The viability of peripheral blood mononuclear cells (PBMC) and HepG2 cancer cells was assessed using the MTT assay to determine the biocompatibility and anticancer potential of the SeNPs.

Results and Discussion: EPS-SeNPs exhibited a spherical morphology with an average size of 43.7 nm and stable physicochemical properties, demonstrating a high surface charge and hydrodynamic stability across pH ranges. The functionalized cotton gauze (EPS-SeNPs@CH_CG) showed enhanced antibacterial activity, with significant anti-adhesion and sustained release of Se, effectively reducing bacterial adherence and growth. EPS-SeNPs displayed dose-dependent radical scavenging activity, indicating robust antioxidative properties. EPS-SeNPs exhibited low toxicity toward PBMCs and dose-dependent anticancer effects against HepG2 cells, underscoring their therapeutic potential. The EPS-SeNPs@CH_CG demonstrated dual antimicrobial and antioxidant capabilities, positioning it as a superior alternative to conventional wound dressings. The biocompatibility of EPS-SeNPs highlights their potential for broader biomedical applications. The green synthesis method using fungal EPS ensures sustainability and cost-efficiency, addressing concerns over toxicity and scalability in nanoparticle production.

Conclusion: EPS-capped Se nanoparticles represent an innovative step toward integrating nanotechnology with health care solutions. Their application in functional wound dressings and potential as therapeutic agents in cancer treatment, showcases their versatility and efficacy. Future research should focus on clinical trials and exploring synergistic effects with other bioactive compounds.

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Selenium Nanoparticles in Food Supplementation: Monitoring Needs and Analytical Challenges

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Introduction: Selenium (Se) nanoparticles (SeNPs) are emerging as a promising form of Se supplementation. Recent advances favor biosynthetic production methods utilizing microorganisms or plant-based systems—such as fungi, yeast, algae, and bacteria—to reduce ionic Se and stabilize the resulting nanoparticles. These biological approaches offer advantages by reducing production costs, minimizing toxicity, and improving biocompatibility, as they avoid the use of synthetic reducing and stabilizing agents. Natural extracts not only support the formation of SeNPs but also enhance their stability and biological functionality. In biosynthetic production systems, nanoparticle size can be controlled by modifying Se concentrations and incubation time. There is a pressing need for robust analytical techniques to detect and characterize SeNPs within complex organic matrices, differentiating them from other Se forms (e.g. selenite, selenate, organic selenium). This demand arises from the growing interest in SeNPs as next-generation Se supplements and increasing regulatory requirements for the monitoring of food and feed products derived from microbial or plant origins, as well as from metabolic studies involving SeNPs.

Methods: Single-particle inductively coupled plasma mass spectrometry (spICP-MS) has emerged as a powerful technique for the determination and characterization of Se nanoparticles (SeNPs) in biological matrices. This method enables the detection, sizing, and quantification of SeNPs by aspirating a dispersion of intact particles into the plasma, where each particle is atomized and detected as a discrete signal proportional to its selenium content ^[1]. However, in complex biological matrices, sample preparation remains a critical step; protocols must effectively liberate SeNPs from the matrix without causing their dissolution or aggregation ^[2].

Results and Discussion: This study will highlight the current needs in the monitoring of SeNPs in food and feed supplements, as well as studies related to their biosynthesis and transformation in the gastrointestinal digestive system, and discuss the potential of spICP-MS in this context ^[1-3]. Special attention will be paid to the sample preparation schemes employed for the extraction of SeNPs from complex biological matrices. This will be illustrated by the results of the projects carried out in the laboratory of the Institute of Analytical and Physical Chemistry of the National Research Council in Pau, France.

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Advances in Characterizing the Selenium Metabolome across Microorganisms, Plants, and Animals

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Introduction: The selenium (Se) metabolome is diverse, species-specific and varies significantly across biological kingdoms. Microorganisms, plants, and animals each produce unique sets of Se-containing metabolites depending on their metabolic capabilities, environmental exposure, and physiological roles of Se. Understanding the diversity and biosynthetic pathways of complex Se metabolites is crucial for harnessing their potential in health, agriculture, and environmental sustainability.

Methods: Electrospray mass spectrometry (MS) and inductively coupled plasma (ICP) MS have significantly improved the sensitivity and specificity of Se species detection, enabling detailed large-scale profiling of Se metabolites in complex biological matrices.^[1] In particular, high-resolution accurate mass spectrometry (HRAM) using Orbitrap and Fourier Transform Ion Cyclotron Resonance (FT-ICR) mass analyzers provide exceptional resolving power: Orbitrap systems can achieve resolving powersup to 500,000 for transient signals while FT-ICR systems can surpass this in direct infusion mode. Such resolutions allow the separation of Se-containing metabolites with very similar masses, which is crucial given the chemical diversity and complexity of the selenometabolome in microorganisms. These capabilities have been exploited for resolving Se-containing metabolites from isobaric interferences and distinguishing between different isotopologues and elemental compositions (e.g. ⁸⁰Se vs. ³⁴S + ¹⁶O).

Results and Discussion: Recent methodological advances are discussed to profile low-molecular-weight selenocompounds in diverse biological systems, including yeast, bacteria, plants, and animals. In microorganisms like *Saccharomyces cerevisiae* and *Bifidobacterium longum*, Se is biotransformed into a complex array of metabolites, including selenoamino acids and organoselenium compounds. The strain and enrichment procedure influence the distribution of Se species. In plants such as rice and kale, Se uptake and metabolism are affected by both genetic and agronomic factors. Se-enriched rice cultivars contain unique selenosugars and amino acid conjugates, while kale sprouts fortified with benzoselenoate-based compounds, produce chemoprotective metabolites. Supplementation with inorganic Se leads to increased formation of glutathione-, cysteine-, and methyl-conjugated selenosugars in animal models, including rats and turkey poults. The structural elucidation of these metabolites, e.g. Se-N-acetylgalactosamine derivatives, including their binding to proteins via S-Se bonds, will also be presented.

Conclusions: HRAM spectrometry using Orbitrap and FT-ICR is indispensable for selenometabolome research, offering the resolution, accuracy, and sensitivity needed to unravel the complexity of Se metabolism in microorganisms, from routine quantitation to novel metabolite discovery and metabolic pathway elucidation. The expanding knowledge of the microbial Se metabolome opens new avenues for biotechnological applications, such as the production of Se-enriched nutraceuticals, biofortification, and the bioremediation of Se-contaminated environments.

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Switchable Fluorescent Molecular Tools Containing Selenium

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Introduction: Selenium (Se) is isovalent with sulfur (S) and allows for continued novel and specific small molecule design. Our work in Se chemistry relates mainly to fluorophores and Se as an atom directly connected to the fluorophore core as a substituent form of PhSe. A Se center serves in versatile ways in chemosensing and in theranostics; our lab (Molecular Logic Gate Laboratory) has carried out this project for more than a decade.

Methods: While synthetic chemistry with Se might be more challenging than that for S, we tried to implement Se (e.g. -SePh) as a direct substituent on chromophore systems using different chromophores and using many standard synthetic chemistry and characterization methods. For example, the strategy to change its oxidation state (valence state) and also to serve as a part of an organic leaving group, allows for the formation of interesting different molecular tools compatible with biological systems, which align with NMR characterization.

Results and Discussion: We have synthesized and characterized a variety of chromophores (fluorophores) that contain Se as a substituent. The chromophore systems include BODIPY, coumarin, mycophenolate precursors, etc., with the newest chromophore explored being that stemming from 2-(phenylselanyl)-1H-indole-3-carbaldehyde.

Conclusions: Our interest in bringing S/Se/Te containing molecules as fluorescent molecules into existence, has allowed us to study Se as a reactive center in different ways where the reaction at Se has many effects.

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Selenium Speciation Distribution and Bioavailability in the Soil-maize System in a Typical High-Se Area

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Introduction: Selenium (Se) is essential for human health. The main sources of Se are obtained through food crops. Many populations worldwide face Se deficiencies but high-Se areas offer potential for Se-enriched agriculture and the production of Se-enriched plant products. Plant uptake of Se depends not only on total soil Se but significantly on its chemical forms (speciation/fractionation) in the soil, which affects bioavailability. Studies have investigated Se dynamics, including fractionation and accumulation in maize and in soil-crop systems in high-Se regions of China^[1]. This study investigated Se distribution and its transfer in the soil-maize system in a typical high-Se region (Ziyang, China), which included analyzing soil Se fractionation, Se content in maize parts, and Se species in grains. The objectives were to link soil Se forms to uptake and provide a basis for Se-enriched production.

Methods: Rhizosphere soil and maize plant samples (root, stem, leaf, grain; n=11 for each) were collected from Naore Village, Ziyang. Samples were processed (air-dried/sieved soil, washed/dried/powdered plants). Total Se was determined by Atomic Fluorescence Spectrometry. Soil Se fractionation used a four-step five-state sequential extraction method ^[2]: water-soluble (F1), exchangeable (F2), organic-bound (F3), acid-soluble (F4), residual (F5). Selenium species, including SeCys₂, MeSeCys, Se(IV), SeMet, Se(VI) in enzymatically extracted grains were analyzed by HPLC-ICP-MS ^[3]. In addition, soil pH and total organic carbon (TOC) were measured. Statistical analyses (descriptive statistics, correlation, regression) were performed using Minitab 16.

Results and Discussion: Soil showed high total Se (avg 16.45 mg/kg), predominantly in residual (71.13%) and acid-soluble (21.33%) fractions, which were significantly correlated with total soil Se. Maize accumulated highest Se in leaves (14.88 mg/kg) > roots (7.44) > grains (5.65) > stems (3.53). Se-enriched grains safely met Se-enriched food standards. Regression analyses showed soil bioavailable fractions (F1+F2+F3) significantly correlated with maize leaf Se, indicating these labile forms drive uptake of Se. In grains, Se was mainly present in organic forms: SeMet (85.79%) and SeCys₂ (9.16%), with minor MeSeCys/inorganic species. Total grain Se significantly correlated with organic Se species, suggesting the efficient translocation of Se to edible parts or grains.

Conclusions: Se-enriched soils in Ziyang contain Se mainly in recalcitrant forms, but labile fractions (water-soluble, exchangeable, organic-bound) of soil Se are critical for plant uptake. Maize accumulates Se effectively and produces Se-enriched grains with bioavailable organic forms (SeMet, SeCys₂). This study indicates that soil bioavailable Se is important for the enrichment and capacity of organic Se synthesis and translocation in maize, supporting the development of Se-enriched agriculture in high soil Se regions.

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Efficient Nano-Selenium Production by *Cupriavidus necator* via ARTP Mutagenesis

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Introduction: Nano-selenium (Se) known for its high bioactivity and low toxicity holds significant potential for applications in health care, disease treatment, biopharmaceuticals, and Se-enriched agriculture. Although microbial synthesis of nano-Se offers a green and stable alternative, existing strains often fall short in achieving high yields of nano-Se. This study investigates the bioreduction of Na₂SeO₃ by *Cupriavidus* sp. GX-5 and explores its potential for nano-Se production through *Atmospheric and Room Temperature Plasma* (ARTP) mutagenesis. The aim is to develop mutant strains with enhanced capacities for Na₂SeO₃ conversion and nano-Se yield.

Methods: The study utilized *Cupriavidus* sp. GX-5, a strain originally isolated from a lead-zinc mine. The ARTP mutagenesis technique was employed to induce genetic mutations, optimizing the mutagenesis time through lethal rate analysis ^[1, 2]. Mutant strains were screened using solid and liquid LB media containing sodium selenite ^[3]. The conversion rate of sodium selenite was assessed using atomic fluorescence spectrometry. The morphological and structural characteristics of nano-selenium were examined using scanning electron microscopy and X-ray diffraction.

Results and Discussion: The mutant strains F9-1 and F33-1, obtained through six cycles of 50-second ARTP mutagenesis, demonstrated significant improvements in Na₂SeO₃ conversion rates. Compared to the original strain, these mutants exhibited increased minimum inhibitory concentrations of sodium selenite by 20% and 22.2%, respectively. The conversion rates of Na₂SeO₃ in the culture medium in the first two days were 8.78% and 5.53% higher than those observed with the original strain. After 10 days of continuous culture, the conversion rates reached 82.61% and 79.69%, respectively. SEM analysis revealed that the nano-Se particles were spherical, with a uniform size distribution ranging from 100 to 500 nm and an average particle size of 190 nm. XRD results indicated that the nano-Se was amorphous, suggesting a high level of bioactivity. The genetic stability of the mutant strains was confirmed through multiple generations of cultivation.

Conclusions: This study confirms that *Cupriavidus* sp. can effectively reduce sodium selenite to produce nano-Se. The ARTP mutagenesis technique successfully yielded two high-yield nano-Se strains, F9-1 and F33-1, which exhibited enhanced sodium selenite conversion rates and genetic stability.

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Nanopriming with Selenium Nanoparticles Reprograms Seed Germination, Antioxidant Defense, and Phenylpropanoid Metabolism to Enhance *Fusarium graminearum* Resistance in Maize Seedlings

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Introduction: *Fusarium graminearum* threatens maize through stem rot and deoxynivalenol (DON) contamination. Conventional approaches like chemical seed coatings are constrained by limited long-term efficacy and environmental concerns such as soil residue accumulation. In contrast, selenium nanoparticle (SeNP) priming provides a sustainable and economically viable strategy, minimizing ecological footprints through rapid elemental bioconversion, while enhancing disease resistance in agricultural systems. However, nanotechnology applications synchronizing seed enhancement with pathogen resistance remain underexplored. The study aimed to establish SeNP as a nano-priming agent for improving seed vigor and disease resistance in maize.

Methods: Multi-omics integration (metabolomics/transcriptomics) and physiological profiling established the optimal diseasesuppressing SeNP dose (50 mg/L) through dose-response screening, enabling mechanistic evaluation of F. graminearum resistance against water controls.

Results and Discussion: SeNP priming of seeds enhances starch-to-sugar conversion to elevate energy reserves, while in seedlings this treatment activates JA-mediated phenolic acid biosynthesis - synergistically establishing dual resistance that suppresses *F. graminearum* infection severity by 26.3% and DON production by 42.5% (p<0.05) (Fig 1).



Figure 1. Mechanisms of growth promotion in maize by SeNP priming and its elicited resistance against F. graminearum

Conclusions: This study found that 50 mg/L SeNPs priming can enhance the quality and antioxidant capacity of maize, reduce the contamination of *F. graminearum* and DON, and provide an eco-friendly alternative to chemical treatment for mycotoxin control.

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Effects and Mechanism of Selenobacteria on Selenium Transformation in Se-rich Soil and Selenium Absorption in Chinese Flowering Cabbage

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Introduction: The "soil-plant-human" pathway is recognized as the most efficient route for Se biofortification. Guangxi, China, possesses the nation 's largest Se-rich soil region, but the acidic nature of these soils results in low Se bioavailability. Microorganisms, especially selenobacteria, have the potential to transform soil Se species and enhance plant Se uptake. However, the genetic basis and mechanisms underlying Se transformation by selenobacteria remain poorly understood. This study aimed to isolate highly Se-tolerant microorganisms from Guangxi's Se-rich soils and evaluate their soil Se species and valencies transformation ability. Importantly, we examined their ability to promote Se absorption in Chinese flowering cabbage, and we performed whole-genome sequencing of two selenobacteria strains with different to Se tolerance identify candidate genes for Se utilization.

Methods: Soil samples were collected from multiple Se-rich locations in Guangxi. Se-tolerant microorganisms were isolated and screened by gradient Se concentration on selective media. Highly tolerant bacterial strains were identified using 16S rDNA sequencing. Two representative selenobacteria, *Bacillus cereus* YLB1-6 and *Bacillus altitudinis* YLB2-1, were selected for pot experiments with Chinese flowering cabbage to assess their effects on Se transformation and plant uptake. Soil Se species were extracted using the continuous leaching method ^[1], and valencies were extracted following the method by Wang et al. ^[2] The Se species and valencies were determined by hydride generation-atomic fluorescence spectroscopy. The whole genome sequencing of the two strains was conducted using PacBio SequelII, and bioinformatic analyses identified Se-related genes^[3].

Results and Discussion: Sixty-two Se-tolerant strains were isolated, with bacteria showing the highest Se tolerance (up to 30,000 mg/mL). Eight highly tolerant bacterial strains were identified as *Bacillus licheniformis, B. pumilus, B. cereus, B. altitudinis,* and *Serratia marcescens*. Pot experiments showed that inoculation with selenobacteria significantly increased available Se in soil and Se accumulation in the aboveground parts of Chinese flowering cabbage. YLB1-6 exhibited a stronger capacity for Se transformation and plant Se uptake than YLB2-1. Genome analysis revealed that YLB1-6 contained 46 Se utilization-related genes, including peroxiredoxin, thioredoxin reductase, methionine sulfoxide reductase, seryl-tRNA synthetase, yqeB, yqeC, and nitrite reductase, while YLB2-1 had 38 such genes but lacked yqeB and yqeC. The greater number and diversity of Se-related genes in YLB1-6 likely explain its superior Se transformation capability. These findings provide new microbial and genetic resources for the development of Se biofortification strategies.

Conclusions: This study successfully isolated and characterized highly Se-tolerant selenobacteria from Se-rich soils in Guangxi. We demonstrated their ability to enhance soil Se availability and plant uptake and identified key genes involved in Se transformation through whole-genome sequencing.

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Microbial-Driven Selenium Biotransformation - from Multidimensional Microbiome Modulation to Functional Precision Applications

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Introduction: Selenium (Se) is an essential trace nutrient for humans and animals, with deficiency linked to Keshan disease, impaired immunity, and increased cancer risk. Application of conventional inorganic Se supplements (e.g. sodium selenite) face limitations such as low bioavailability and a narrow toxicity window. Microorganisms offer a promising solution by efficiently converting inorganic Se into low-toxicity, high-bioactivity organic forms (e.g. selenamino acids) or other specific Se species. This capability can contribute to the development of safe and efficient Se-enriched products.

Methods: Research in microbial Se enrichment primarily revolves three core strategies: Strain Isolation and Optimization: Selenium-tolerant/Se accumulating strains are isolated and screened from high-Se environments ^[1]. The organic Se conversion efficiency is maximized through directed enrichment and domestication, alongside systematic optimization of cultivation conditions (temperature, pH, Se concentration, carbon and nitrogen sources). Microbial Community Engineering: Complex communities are constructed using a "bottom-up" partial combinatorial screening strategy. Interactions among community members and their impacts on collective functions, e.g. biofilm formation are thoroughly analyzed through phenotypic observation ^[2], growth kinetics monitoring, antagonism assays, and conditioned media analysis. Concurrently, methods integrating antibiotic resistance markers with selective cultivation or morphological trait tracking are developed to achieve absolute quantification of specific strains within mixed biofilms. Engineered Probiotic Design: Genetically engineered strains capable of synthesizing multifunctional nanoparticles are developed with genetic engineering techniques ^[3]. This approach integrates Se delivery with targeted therapeutic functions such as antioxidant and anti-inflammatory activities ^[4].

Results and Discussion: Microorganisms serve dual roles as efficient Se converters and versatile producers/delivery carriers of functional bioactive compounds (e.g. probiotics, selenium nanoparticles, organic acids, vitamins). Optimized cultivation conditions significantly enhance organic Se conversion yields, while microbial community analyses reveal intricate interactions governing biofilm dynamics and functional outcomes. Engineered probiotics further demonstrate multi-functionality by co-delivering Se with therapeutic molecules (e.g. antioxidants), enabling synergistic integration of nutritional supplementation, probiotic effects, antioxidant/anti-inflammatory activity, plant growth promotion, and detoxification. This convergence underscores their broad utility in functional foods, agricultural applications, and disease intervention strategies.

Conclusions: Microbial systems transcend their role as mere "Se-converting factories," emerging as versatile platforms for producing and delivering bioactive compounds. Methodological advances in strain isolation, community engineering, and genetic design enable efficient synthesis of low-toxicity, high-value Se products. The synergy between Se enrichment and multi-functional benefits (probiotic, therapeutic, agricultural) positions microorganisms as pivotal tools for next-generation applications in nutrition, health, and sustainable agriculture.

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Mechanism Study of Trichoderma-Synthesized Selenium Nanoparticles in Inducing Rice Resistance against Sheath Blight

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Introduction: Biosynthesized nano-selenium, a green and biocompatible antibacterial agent, is synthesized simply and efficiently. It enhances plant disease and abiotic stress resistance, promotes growth, and improves agricultural product quality. This study aims to explore new synthesis methods for biosynthesized nano-selenium and investigate its effects on rice sheath blight, as well as its influence on related physiological and biochemical indicators during disease resistance induction.

Methods: Bio-synthesized nano-selenium (Bio-SeNPs) was produced by fermenting *Trichoderma* sp. strain TY-1 in a seleniumrich medium. These Bio-SeNPs were characterized with SEM-EDS and their particle size distribution was analyzed using Origin software. Plate inhibition assays determined the EC_{20} of Bio-SeNPs against the rice sheath blight pathogen via DPS software. Additionally, the impact of this Bio-SeNPs concentration on induced resistance enzymes (CAT, MDA, H₂O₂, O₂⁻) was assessed using ultraviolet spectrophotometry. GraphPad Prism was used to plot bar charts and analyze significant differences (P < 0.05).

Results and Discussion: Scanning Electron Microscopy (SEM) point-scan images revealed that **Trichoderma* sp.* strain TY-1 can reduce sodium selenite to generate selenium nanoparticles (SeNPs), which are primarily distributed on the surface of the mycelium with a particle size of 156 ± 39.64 nm, meeting the size requirements for nanomaterials ^[2]. This process demonstrates the feasibility of synthesizing nano-selenium using **Trichoderma** TY-1, providing technical support for the development of biocontrol-based nano-selenium synthesis. Additionally, plate inhibition assays confirmed that Bio-SeNPs exhibit inhibitory effects on the growth of the rice sheath blight pathogen. Data analysis using DPS software determined the EC₂₀ of Bio-SeNPs against the rice sheath blight pathogen to be 141.59 mg/L, with increasing concentrations of Bio-SeNPs correlating with stronger inhibitory effects. Following foliar application, Bio-SeNPs enter the plant and modulate the activity of antioxidant enzymes. By enhancing the activities of defense-related enzymes such as CAT and MDA, Bio-SeNPs induce the defense response, reducing intracellular levels of H₂O₂ and O₂⁻, thereby directly scavenging reactive oxygen species. This maintains a dynamic balance among various antioxidant systems within the cell, ultimately mitigating the infection and occurrence of rice sheath blight ^[1].



Figure 1. A: Synthesis and size distribution profile of biogenic selenium nanoparticles. B: The effect of bio-SeNPs on the antioxidant enzyme activity of *Rhizoctonia solani*. C: Antibacterial experiment of biosynthesized nano-selenium.

Conclusions: The application of Bio-SeNPs significantly inhibits the growth of the rice sheath blight pathogen (*Rhizoctonia solani*) through multiple mechanisms, while enhancing the antioxidant and defense systems of rice plants, synergistically combating rice sheath blight. Foliar application of Bio-SeNPs holds substantial potential as a novel strategy for controlling rice sheath blight, offering a promising approach for the development of environmentally friendly nano-Se-based pesticides to manage this disease.

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Preparation and Applications of Self-emulsifying Powder Delivering Selenium and Peptide

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Introduction: Selenium (Se) is an essential trace element for the human body. Selenoproteins in the human body have physiological functions such as anti-oxidation, immunity regulation, tumor prevention and treatment, and free radical scavenging. Selenium polysaccharide in plants is an organic compound composed of Se and polysaccharide that combines the activities of both and has the advantages of low toxicity and high bio- availability. This study aims to synthesize a nano self- emulsifying powder composed of Se polysaccharide-peptide composite to enhance the bioavailability of Se in organisms. The study employs endogenous soybean protein isolate as an amphiphilic nonionic surfactant, mild medium-chain triglycerides as the oil phase of the self-emulsifying system, and Se polysaccharides conjugated with olypeptides via the Maillard reaction as the bioactive component. After emulsification, the elements are freeze-dried to prepare the aforesaid self-emulsifying powder delivering selenium and peptide. The prepared self-emulsifying powder exhibits uniform stability and excellent reconstitution properties. The prepared selenium-peptide nano self-emulsifying powder exhibits homogeneous stability and excellent redispersibility. This formulation can effectively reduce the transport barrier of macromolecules across gastrointestinal epithelial cell membranes, facilitate penetration through the intestinal aqueous boundary layer, and significantly enhance selenium-polysaccharide absorption without causing any gastrointestinal irritation. Consequently, the formulated product may eventually be available for eventual commercialization.

Methods: The self-emulsifying selenium-polysaccharide powder was prepared by: (1) hydrolyzing soybean protein iso- late (4 g/56 mL H₂O, pH 7, 120 °C/30 min) as surfactant; (2) formulating primary emulsion with seaweed selenium-polysaccharide (100 mg), PEG2000 (25 mg), and caprylic/capric triglycerides (1 mL); (3) combining components with sodium oleate (125mg) under homogenization (1000 bar/20 cycles) and freeze-drying. Nanoemulsion size was verified by TEM/laser analysis. Biocompatibility was tested via HCT-116 cell MTT assay (4 h incubation), while release profiles were assessed in simulated gastric/intestinal fluids (14 kD dialysis, 37°C) with Cremophor EL, nanoemulsions, confirming no payload-induced aggregation. TEM imaging verified mono-disperse spherical nanostructures. The formulation exhibited exceptional biocompatibility, maintaining > 90% cell viability in HCT-116 assays. In vitro released studies demonstrated pH-responsive kinetics: sustained gastric release (< 20%)

Results and Discussion: The results from Laser particle analysis and TEM revealed that the size distributions of Se polysaccharide peptide complexes were between 100 - 300 nm. The formulation exhibited exceptional biocompatibility, maintaining > 90% cell viability in HCT-116 assays. In vitro release studies demonstrated pH-responsive kinetics: sustained gastric release (< 20% at 2 h, pH 1.2) contrasted with rapid intestinal liberation (> 80% at 4 h, pH 6.8), indicating targeted intestinal delivery.

Conclusions: The solid nano-self-emulsifying formulation of selenium-polysaccharide-peptide complexes demonstrates superior solubility, biocompatibility, and bioavailability. During gastro-intestinal transit, the system effectively penetrates the intestinal aqueous boundary layer, while maintaining structural integrity. This design protects oil-phase-encapsulated Se polysaccharides from gastric degradation (pH 1.2), while enabling rapid payload release (>80% within 4 h) in intestinal conditions.

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Boosting Selenium Accumulation in Rice Grains by Enhancing NRT1.1B-Mediated MeSeCys Translocation

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Introduction: Selenium (Se) deficiency affects over one billion people worldwide. Enhancing Se accumulation in rice grains through biofortification represents an effective strategy to boost dietary Se intake. Rice (*Oryza sativa* L.), a staple food for half of the global population, predominantly consists of selenomethionine (SeMet). SeMet has lower bioavailability compared to methyl-selenocysteine (MeSeCys), which is found in rice roots and exhibits superior anticancer and antioxidant properties ^[1,2]. However, the mechanisms by which MeSeCys is transported to grains remain unclear. NRT1.1B, a member of the nitrate transporter 1/peptide transporter (NRT1/PTR) family, regulates nitrate transport and has been previously shown to transport SeMet^[1,3]. In this study, we demonstrate that NRT1.1B functions as a bidirectional vascular transporter through dual pathways, facilitating the uptake, transport, storage, and redistribution of MeSeCys in rice, thereby enhancing Se concentration in rice grains.

Methods: Rice seedlings were cultured in Kimura nutrient solution (pH 5.5) containing 2 μ mol/L MeSeCys for 3 d and 6 d, respectively. We determined Se concentrations in roots, leaf sheaths, and leaf blades. Rice seedlings were grown in Kimura nutrient solution containing 2 μ mol/L MeSeCys for 3 weeks before flowering and then cultured without MeSeCys until the rice seedling reached maturity. We then determined Se concentrations in root, node, internode, leaf sheath, leaf blade, and grain tissues.

Results and Discussion: MeSeCys uptake was energy-dependent and required a maintained proton gradient. Heterologous expression of *NRT1.1B* in *Saccharomyces cerevisiae* yeast cells (pYES2-*NRT1.1B*) significantly enhanced MeSeCys uptake, demonstrating that NRT1.1B may stimulate the transport activity of MeSeCys. Furthermore, transgenic rice lines overexpressing *NRT1.1B* significantly enhanced MeSeCys uptake in both roots and leaves, while the *nrt1.1b* mutant exhibited significant reductions. In roots, NRT1.1B facilitated the entry of MeSeCys into epidermal and cortical cells, allowing efficient radial transport into root xylem vessels. Similarly, foliar uptake depends on NRT1.1B-mediated transmembrane transport in mesophyll cells. More importantly, overexpression of *NRT1.1B* significantly enhanced Se translocation when roots were supplied with MeSeCys. This effect led to an increased Se concentrations in leaves of *NRT1.1B* overexpressing lines, NT31 and NT72, compared to the wildtype. Additionally, NRT1.1B facilitated the storage and remobilization of MeSeCys in rice through both xylem and phloem systems. The significant increase in Se concentrations in leaf sheaths and internodes of *NRT1.1B* overexpressing lines, in contrast to the decline in the *nrt1.1b* mutant, demonstrates that NRT1.1B facilitates the loading of xylem from root parenchyma cells and supports long-distance upward transport through the xylem system.

Conclusions: This study demonstrates that NRT1.1B facilitates the uptake, transport, storage, and redistribution of MeSeCys in rice. Specifically, NRT1.1B optimizes the bidirectional dual vascular translocation of MeSeCys, thereby enhancing grain Se concentrations.

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Melatonin and Selenium Boost Strawberry Defense against *Botrytis*: Dual Omics Reveal Key Mechanisms

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Introduction: Selenium (Se) is a beneficial trace element for plants and humans, known to inhibit fungal pathogens like *Alternaria tenuis* and *Fusarium sp.*, and has been used to control postharvest diseases such as *Botrytis cinerea* and *Penicillium expansum*^[1-3]. Melatonin (MT), a phytohormone, regulates plant growth and stress responses, and its derivative Mt-23 exhibits antifungal activity by targeting fungal MAPKs, suppressing diseases like rice blast and gray mold^[2]. The combined effect of MT and Se on suppressing gray mold in strawberries remains unexplored. This study investigates their synergistic role using physiological, transcriptomic, and metabolomic approaches to uncover the underlying resistance mechanisms^[1].

Methods: Strawberry fruits (Fragaria×ananassa 'Red Face') were harvested, sterilized, and inoculated with *Botrytis cinerea* after pretreatment with a composite solution of Se (25 mg/L) and MT (0.5 mmol/L). Mycelial growth inhibition assays were conducted on PDA media with varying Se/MT concentrations. Antioxidant/disease-related enzyme activities were analyzed. Gene expression (PR1, PR2, NP24) was quantified via qRT-PCR.

Results and Discussion: The Se and MT treatments significantly inhibited Botrytis cinerea growth in tomato fruits, with the



and W1 treatments significantly innoted *Bonyus cherea* growth in tomato nuits, with the combined Se+MT treatment showing the highest suppression (74%). While Se alone reduced lesion area by 51% and directly inhibited fungal colony growth, MT alone had minimal antifungal effects but enhanced plant defense enzymes (SOD, POD) and pathogenesis-related (PR-1, PR-2, PR-24) gene expression (Fig. 1). The synergistic effect of Se+MT improved antioxidant activity (SOD, APX) and delayed fruit decay, though MT antagonized Se-induced GSH accumulation. Selenium likely disrupts fungal membrane integrity, while MT acts as a defense signaling molecule, priming SA-dependent resistance pathways. Optimal disease control was achieved with sequential application (Se followed by MT), suggesting MT modulates Se toxicity while amplifying plant immunity. These findings show that Se controls fungal diseases like *Sclerotinia* rot and gray mold by altering soil microbial community ^[1] and directly inhibiting pathogen spore germination ^[3], while MT enhances plant resistance by upregulating defense-related genes with Se also inducing MT biosynthesis to synergistically bolster plant immunity ^[4].

Figure 1. Enzymes including SOD, POD, CAT, PPO, APX, GSH, PAL variation Se+MT (A) and transcriptome abundance changes with Se and MT (B).

Conclusions: Selenium and MT synergistically combat gray mold by inhibiting *Botrytis cinerea* and boosting tomato's antioxidant and defense systems, providing an eco-friendly disease control strategy.

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DL-alanine Promotes the Colonization of *Pseudomonas aeruginosa* and Their Synergistic Enrichment of Selenium and Decrease of Cadmium Absorption by *Brassica napus*

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Introduction: The coexistence of selenium (Se) and cadmium (Cd) seriously limits the development of Se-enriched agriculture. The plant growth promoting bacteria *Pseudomonas aeruginosa* is well known for its high tolerance to Cd and its participation soil Se cycling. DL-alanine as an amino acid exuded by plant roots, significantly promotes *P. aeruginosa* biofilm formation. Therefore, the main aims of the present study were: (1) investigate the colonization of *P. aeruginosa* in the roots of *B. napus*; (2) identify the colonization signal substances and their effect on enhancing bacterial colonization; and (3) study the effects of the combination of signal substances and *P. aeruginosa* on the Se and Cd fractions in soil, the expression levels of genes encoding Se and Cd transporters in roots, and on the functional group changes of cell precipitation and extracellular polymeric substances (EPS).

Methods: Pot experiments with DL-alanine and *Pseudomonas aeruginosa* were constructed to explore their effects on both the availability of Se and Cd in soil and the uptake of Se and Cd in plants. FTIR and XPS analysis were used to investigate the biofilm formation and morphological changes of Se and Cd in the cultured strains. The pathway of Se and Cd conversion by *Pseudomonas aeruginosa* and DL-alanine was further validated by gene expression analysis^[1].

Results and Discussion: The results showed that DL-alanine significantly boosts *P. aeruginosa* biofilm formation and root colonization. The combined treatment of DL-alanine and *P. aeruginosa* increased the Se content in the shoots by 55.8% and decreased the Cd content in the shoots and roots by 66.3% and 67.9%, respectively. The direct reason for this result is that the available Se in the rhizosphere soil increased by 32% and the available Cd decreased by 10%. Further investigation showed that DL-alanine promotes the transformation of Se(0) to Se(-II) and the formation of CdSe nanoparticles by *P. aeruginosa*, which enhances the availability of Se and reduces that of Cd. Gene expression analysis revealed that the expression levels of Se-related genes were upregulated, while those of Cd transport genes were downregulated. This study provides a theoretical basis for safer agricultural practices.

Conclusions: This study demonstrated that in an environment with coexistence of Se and Cd, DL-alanine and *P. aeruginosa* can promote the transformation of Se(0) to Se(-II) and the formation of CdSe nanoparticles, thereby promoting plant Se enrichment and Cd reduction.

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Enhancing Antioxidant Capacity and Modulating Sensory Traits by Nanoselenium Foliar Biofortification on Field Leek Moss

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Introduction: Leek moss (*Allium tuberosum* flowering stalk) is valued for its pungent flavor and crisp texture. However, strategies to improve their nutritional value and edible traits remain limited. Selenium (Se), an essential micronutrient, is known to boost antioxidant defense in plants, yet its effects on sensory quality remain unclear. This study aimed to investigate the efficacy of nano-selenium (nano-Se) in enhancing antioxidant capacity, nutritional composition, and modulating sensory properties in leek moss under field conditions.

Methods: A randomized block field trial was conducted using foliar applications of nano-Se (0, 2.5, 5.0, 7.5, 10, 15 mg/L) and sodium selenite. Antioxidant capacity (T-AOC, SOD, POD, CAT), GSH, MDA, and nutrient metabolites were evaluated using spectrophotometry and UPLC-MS/MS. Transcriptomic and WGCNA analyses were performed to investigate regulatory mechanisms. Three consecutive harvests were analyzed to assess treatment persistence. Statistical analysis was performed using ANOVA and multivariate tools^[1,2].

Results and Discussion: Nano-Se at 7.5 - 10 mg/L significantly improved antioxidant status: T-AOC increased by 45%, enzyme activities by 20–35%, and GSH by 50%, while MDA decreased by 25%. Metabolomics revealed elevated flavonoids (+ 32%), phenolic acids (+ 26%), and vitamins C/B5, alongside suppressed sulfur volatiles (ACSO and allicin at 27% of control). Transcriptome analysis indicated activation of phenylpropanoid and MAPK signaling pathways, downregulation of CSO biosynthesis genes (*AtuGGT*, *AtuFMO*, *AtuAIN*), and upregulation of lignin synthesis genes (*CCR*, *CAD*, *COMT*), explaining regulated flavor balance and firmer texture.

Conclusions: Nano-selenium acts as an effective biofortification agent that enhances antioxidant capacity and selectively modulates flavor and texture traits in leek moss. Compared to sodium selenite, nano-Se provided superior and more sustained effects across multiple harvests, offering a promising strategy for high-quality, sustainably produced leafy vegetables.

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Nano-Se and GSH Combined Foliar Application Enhances Cucumber Resistance to *Botrytis cinerea* by Promoting Cucurbitacin Biosynthesis through JA Signal Transfer Activation

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Introduction: *Botrytis cinerea* is a major necrotrophic fungal pathogen responsible for significant yield losses in cucumber cultivation. Conventional chemical control methods raise environmental concerns, driving the search for sustainable alternatives. This study investigates whether the combined foliar application of nano-selenium (Nano-Se) and glutathione (GSH) can enhance cucumber resistance to *B. cinerea* through activation of the jasmonic acid (JA) signaling pathway and subsequent induction of cucurbitacin biosynthesis.

Methods: Cucumber seedlings were treated with Nano-Se (10 mg/L), GSH (100 mg/L), or a combination of both (SeG). Following inoculation with *B. cinerea*, disease severity was assessed. Physiological parameters, antioxidant enzyme activities, phytohormone levels, and metabolite accumulation were analyzed. Integrated transcriptomic and metabolomic approaches were employed to elucidate underlying molecular mechanisms.

Results and Discussion: SeG treatment significantly promoted vegetative growth and photosynthetic capacity and also enhanced the activities of key antioxidant enzymes in the ASA-GSH cycle, including APX (81.7%), MDHAR (43.4%), and GSH-Px



(60.5%). Disease severity was reduced by 41.9% in SeG-treated plants. JA accumulation increased by 44.1%, along with the upregulation of JA biosynthetic genes such as LOX4 (430%) and AOC2 (120%). Notably, cucurbitacin levels were significantly elevated, and key genes involved in its biosynthesis - including OSC (107.5%), P450 (440.8%), and ACT (414.0%) - showed marked upregulation. Furthermore, the expression of cucurbitacin biosynthesis genes was significantly induced by exogenous JA application, suggesting that JA functions as an upstream regulatory signal for cucurbitacin production. SeG also enhanced the accumulation of phenylpropanoid compounds and activated defense-related pathways, collectively contributing to enhanced resistance against B. cinerea.

Conclusions: Nano-Se and GSH synergistically enhance cucumber defense against *B. cinerea* by activating JA signaling and promoting cucurbitacin and phenylpropanoid biosynthesis. Notably, JA and cucurbitacin function cooperatively as part of the defense mechanism, with JA acting as an upstream signal to induce cucurbitacin production, thereby strengthening resistance to pathogens. This coordinated response significantly reduces disease severity. These findings suggest that SeG is a promising biostimulant strategy that balances plant growth and defense for sustainable crop protection.

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Identification and Functional Analysis of the MMT Gene Family in Wheat

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Introduction: In plants, S-adenosyl-L-Met:L-Met S-methyltransferase (MMT) is the key enzyme catalyzing the methylation of Se-Met to Se-methyl Se-Met (MeSeMet) (Figure 1). Emerging studies have demonstrated the broad-spectrum antimicrobial activity of methylated selenium (Se) compounds against phytopathogens ^[1]. Wheat is one of the global staple foods and has been subjected to various pests and diseases. However, neither MeSeMet has been detected nor has *TaMMT* been cloned in wheat. It is still not clear about the possible pleiotropic effects of *TaMMT* on phytopathogen resistance, nutritional quality, yield, and other agronomic traits. In this study, we conducted a comprehensive analysis of the MMT gene family in wheat, employing both bioinformatics and experimental methods to identify and clone *TaMMT* genes in wheat.

Methods: BLAST tools were used to identify wheat MMT homologous genes based on the MMT sequences from the model plant *Arabidopsis thaliana*. A total of 19 MMT candidates were identified and annotated. The TBtools software was employed to visualize the chromosomal locations of the identified MMT genes. The basic physicochemical properties of the MMT proteins were predicted using the ProtParam website. Semi-quantitative and qRT-PCR analyses were conducted to validate the mRNA expression of the MMT candidate genes in wheat varieties Zheng9023 and Xinong1376 treated with sodium selenite. The promoters and coding sequences of the MMT genes were amplified and compared across different wheat varieties. Vectors containing the MMT genes were constructed and transformed into Agrobacterium tumefaciens for plant transformation.

Results and Discussion: Gene identification and localization: The identified MMT genes were distributed across different wheat chromosomes. The proteins exhibited diverse physicochemical properties, with molecular weights ranging from 9,673.03 to 124,422.69 Da and isoelectric points from 4.54 to 5.71. Expression patterns: *TaMMT_1A*, *TaMMT_1B*, and *TaMMT_1D* showed higher expression levels compared to other *TaMMT* genes. *TaMMT_1A* was highly expressed in both Zheng 9023 and Xinong 1376, while *TaMMT_1B* was not expressed in Xinong1376. The expression of these genes was responsive to Se treatment, with *TaMMT_1A* and *TaMMT_1D* showing higher expression levels at 2 days post-treatment. Promoter and CDS sequence comparison: The promoter and CDS sequences of *TaMMT_1A* were highly conserved across the tested varieties, while *TaMMT_1D* showed significant sequence variations, especially in Xinong1376. Vector construction and transformation: The vectors containing *TaMMT_1A*, *TaMMT_1B*, and *TaMMT_1D* were constructed and positive transgenic lines were achieved.

Conclusions: The study provides a comprehensive identification of *TaMMT* gene family in wheat, and the potential roles of *TaMMT_1A*, *TaMMT_1B*, and *TaMMT_1D* in wheat development and stress responses remain to be investigated.

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Selenoprotein W Targets PKM2 to Regulate Macrophage Polarization and Pyroptosis Crosstalk to Induce Porcine Kidney Fibrosis

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Introduction: Selenium (Se) is an essential trace element for the human body, and its biological functions are mainly accomplished by regulating selenoproteins. Selenoprotein W (SelW) is a kind of small molecule selenoprotein that plays an important role in the body's antioxidant function. Fibrosis is a characteristic of all chronic kidney diseases. It is a pathological process in which inflammation leads to the necrosis of parenchymal cells in organs and excessive extracellular matrix deposition. Pyroptosis and macrophage polarization are important links involved in the inflammatory response in fibrosis. Energy metabolism reprogramming leads to the release of mitochondrial mtROS, and oxidative stress can cause pyroptosis and M1 polarization.

Methods: In this study, the Se-deficient porcine kidney tissues, SelW knockdown treatment LLC-PK1 and 3D4/21 cell models were established. The functional impairment of the kidneys was determined by H&E staining and the detection of urea nitrogen and creatinine contents. The interaction between SelW and PKM2 was confirmed by CoIP and molecular docking. The energy metabolism-related indicators and oxidative stress levels in renal tissues and cells were detected. Real-time fluorescence quantitative PCR (qPCR), western blotting, and immunofluorescence techniques were applied to evaluate the mRNA and protein levels of markers related to pyroptosis, fibrosis, and macrophage polarization. In addition, a co-culture system of LLC-PK1 and 3D4/21 was constructed using transwell chambers to explore the crosstalk effect between pyroptosis and M1 polarization.

Results and Discussion: Selenium deficiency leads to morphological damage and functional disorders of pig kidneys, and at the same time, the expression of SelW in the tissues is significantly reduced. The co-immunoprecipitation experiment confirmed the interaction between SelW and PKM2. After Se deficiency or SelW knockdown, the TCA process was blocked. The expression of glycolysis genes (HK-2, PFKM, PKM2, and LDHA) was decreased, the activity of mitochondrial respiratory chain complex enzymes (NDUFB8, SDHB, UQCRC2, MTCO1, and ATP5A) was decreased, and the expression of mitochondrial fusion proteins (OPA1, MFN1, and MFN2) was reduced. In contrast, the expression of fission proteins (FIS1, DRP1, and MFF) was elevated. PKM2 activator TEPP-46 alleviated the mitochondrial damage, indicating that SelW targets PKM2 can regulate energy metabolic reprogramming in porcine kidney. In addition, Se deficiency or SelW knockdown, also decreased CAT and GPX activities, and increased H_2O_2 and MDA content, with a significant increase in mtROS production. Selenium deficiency or SelW knockdown, resulted in upregulation of pyroptosis-related gene (Caspase-1, GSDMD, NLRP3, ASC, and IL-1 β) expression, upregulation of M1 polarization markers (CD86, TNF- α , and IL-1 β), downregulation of M2 markers (CD163, IL-10, and Arg-1), and activation of fibrosis-related indicators. Crosstalk between cellular pyroptosis and macrophage polarization was confirmed to exacerbate kidney fibrosis by the addition of the pyroptosis inhibitor MC9950 and the M1 polarization inhibitor Prostaglandin E3.

Conclusions: Selenium deficiency regulates energy metabolism through SelW-targeted PKM2, and reprogramming induces mtROS-mediated pyroptosis-M1 polarization crosstalk to promote the occurrence of kidney fibrosis in pigs. This study expounds a new toxicological mechanism of Se deficiency causing kidney fibrosis in pigs, providing a new perspective for the research of animal nutritional deficiency diseases and also offering a reference for comparative medicine.

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Foliar Selenium Application Enhances Antioxidant Metabolism, Yield and Nutritional Quality of Grape Plants

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Introduction: Selenium (Se) is recognized as a beneficial element for plants, largely due to its antioxidant properties that strengthen cellular defense mechanisms. In regions such as the São Francisco Valley in Brazil, agricultural crops are frequently exposed to high temperatures, leading to significant stress that can impair plant growth, productivity, and overall physiological integrity. Elevated temperatures often trigger the excessive production of reactive oxygen species (ROS), causing oxidative damage to plant tissues. Foliar application of Se has been shown to mitigate heat stress by enhancing antioxidant activity and reducing ROS accumulation, thereby supporting better plant development and crop performance. Given these potential benefits, the objective of this study was to evaluate the effects of different foliar Se application rates on antioxidant regulation and the productivity of crops grown under high-temperature conditions.

Methods: The experiment was carried out in the São Francisco Valley, in Petrolina - PE, Brazil, using a randomized block design with four replicates per treatment, totaling 24 plots. Grape vines of the ARRA 15 variety were used. Foliar Se application was performed using six rates (0, 4, 8, 12, 16, and 20 g Se ha⁻¹) with Se-bisglycinate as the Se source and a spray volume of 500 L ha⁻¹. Antioxidant metabolism and Se concentration was evaluated in our previous publication ^[1].

Results and Discussion: Selenium application reduced lipid peroxidation compared to the control treatment as indicated by lower MDA (malondialdehyde) content. This reduction was most pronounced at Se doses of 8 and 20 g/ha, which led to MDA decreases of 16.6% and 18.1%, respectively, relative to the control. In addition, all Se treatments resulted in a reduction of H₂O₂ content. Specifically, Se doses of 4 and 8 g/ha increased the specific activities of APX and CAT by 276.8% and 97.8%, respectively. A significant enhancement in total soluble proteins (TSP) was also observed, with an increase of up to 255% under the 20 g/ha Se treatment. Furthermore, Se accumulation in grape berries reached up to 10.94 μ g/kg, as shown in Table 1. Foliar Se application at 12 g/ha dramatically increased grape berry yield due to ROS scavenging enhancement.

Table 1. Sampling dates and corresponding measurements of MDA, H₂O₂, SOD, CAT, APX, PST, productivity, and Se concentration.

Treatments	MDA	H_2O_2	SOD	CAT	APX	TSP	Yield	Se concentration
g ha ⁻¹ Se	nmol g ⁻¹ FW	µmol g ⁻¹ FW	U mg ⁻¹ protein	nmol min ⁻¹ mg protein	µmol min ⁻¹ mg protein	mg g ⁻¹ FW	t ha⁻¹	µg kg⁻¹ FW
0.00	1.88 ± 0.06 a	$0.51\pm0.02\ a$	$125.51 \pm 1.42 \ a$	$660.25 \pm 12.66 \text{ d}$	$36.40\pm1.04~cd$	$1.87\pm0.17~f$	$31.08\pm4.45\ b$	$0.00^* \pm 0.22 \text{ d}$
4.00	$1.70\pm0.05\ b$	$0.38\pm0.01\ d$	$109.31\pm1.54~b$	$1101.52 \pm 12.34 \ b$	137.15 ± 9.75 a	$2.24\pm0.04~e$	$41.96\pm 6.63\ ab$	$5.03\pm0.27\ c$
8.00	$1.57\pm0.05\ c$	$0.43\pm0.01\ c$	$74.11\pm0.97\ c$	1306.52 ± 24.83 a	$115.06\pm4.50\ b$	$3.34\pm0.07\ d$	$50.67 \pm 2.76 a$	$5.41\pm0.42\ c$
12.00	$1.71\pm0.03\ b$	$0.47\pm0.01\ b$	$59.07 \pm 0.51 \ d$	725.33 ± 10.33 c	47.09 ± 1.30 c	$4.19\pm0.03\ c$	$52.71\pm4.38\ a$	$8.28\pm0.81~b$
16.00	$1.65\pm0.03\ bc$	$0.39\pm0.01\ d$	49.86 ± 0.44 e	733.81 ± 12.55 c	34.03 ± 3.78 d	$4.92\pm0.08\ b$	$49.58 \pm 5.61 \text{ a}$	9.54 ± 0.25 ab
20.00	$1.54\pm~0.04~c$	$0.36\pm0.02\ d$	$36.61 \pm 0.70 \; f$	$684.29 \pm 19.68 \text{ d}$	$16.65 \pm 1.68 e$	6.63 ± 0.06 a	$43.08\pm2.20\ a$	10.94 ± 1.03 a
CV (%)	2.98	3.17	1.48	2.05	8.07	2.48	11.18	9.88

*Not detectable at the method detection limit of 3 µg/kg

Conclusions: Foliar Se application at concentration of 12 g/ha enhanced antioxidant metabolism and increased grape yield by 63% and Se concentration in the berry at safe levels for human consumption.

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Functional Analysis of the Selenocysteine Methyltransferase (TaSMT) in *Fusarium graminearum*

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Introduction: Selenium (Se), an essential trace element for humans, exhibits dual characteristics of both nutritional benefits and toxicity. Among various forms of Se, methyl-selenium demonstrates the highest efficacy in suppressing wheat Fusarium head blight (FHB)^[1]. Selenocysteine methyltransferase (SMT) is the enzyme responsible for the methylation of Selenocysteine (SeCys) to Se-methylselenocysteine (MeSeCys) in plants. *Fusarium graminearum* is one major causal agent of FHB ^[2]. The main purpose this study is to dissect the effects of transformation of TaSMT into *F. graminearum* on pathogen deoxynivalenol (DON) production and virulence.

Methods: The disease severity was estimated by measuring the proportion of symptomatic spikelets and DON content in a spike 21 days post inoculation.

Results and Discussion: Three transformants exhibited normal growth and colony morphology. However, fungal biomass was significantly increased in PDA plate inoculated with the *ProRP27:TaSMT-5B* stains in comparison with those inoculated with PH-1. No significant differences in PSS and DON levels were detected between wild-type and *ProRP27:TaSMT-5B*-infected wheat heads. However, when 60 mg/L sodium selenite solution was applied to wheat plants 24 hours before inoculation, the PSS, as well as DON production, were significantly induced in the resulting *ProRP27:TaSMT-5B* mutants when compared with the wild type. These observations suggest that *TaSMT-5B* enhances the pathogenicity and DON biosynthesis of *F. graminearum*, which was probably Se-dependent through its metabolic conversion of inorganic Se into organic forms.

Conclusions: The high efficacy of MeSeCys in suppressing *F. graminearum* on PDA plates and the enhanced pathogenicity and DON synthesis of *F. graminearum* TaSMT-5B transformants, indicate that *F. graminearum* may perceive MeSeCys through fungal flagella or other surface receptors.

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Selenium Transport and Absorption in *Brassica napus*: The Role of the *SMT* Gene

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Introduction: Selenium (Se) is an essential trace element for humans, possessing a variety of physiological functions, *Cruciferae* has a high capacity for Se uptake, transport and transform, making it a desirable source for dietary Se. *Brassica napus* L, belonging to the *Cruciferae* family, has a particularly outstanding ability to efficiently transform methy-selenocysteine (MeSeCys) compared with other crops, and has attracted much attention as a high-quality Se-enriched crop ^[1]. *SMT* (Selenocysteine methyltransferase) is a key enzyme in the plant Se metabolism pathway, capable of converting selenocysteine to methylselenocysteine ^[2]. This conversion not only alleviates the toxicity of inorganic Se but also enhances Se accumulation in plants. As a crucial enzyme for Se detoxification, this process is vital for plant Se tolerance and accumulation. The function of this gene has been elucidated in species such as *Astragalus* M, *Camellia sinensis* L, *Brassica oleracea* L, and *Solanum lycopersicum* L. ^[3-7]. However, research on *SMT* in rapeseed is limited, and its function remains unclear. Therefore, we initiated functional studies on the *BnaSMT* gene in rapeseed.

Methods: We conducted a gene family analysis, utilized Arabidopsis hmt mutant materials, created *BnaSMT* Arabidopsis heterologous expression lines, and developed *BnaSMT* yeast heterologous expression strains.

Results and Discussion: Firstly, through gene family analysis, we revealed a high degree of phylogenetic relatedness between *BnaSMT* and *AtHMT*. Secondly, we ascertained that the mutation of *hmt* in Arabidopsis thaliana led to a significant reduction in Se accumulation. Heterologous expression of *BnaSMT* in the *hmt* mutant alleviated the deficiency in Se accumulation, restoring it to wild-type levels without compromising growth. Lastly, heterologous expression of *BnaSMT* in yeast strains significantly enhanced their capacity for Se accumulation.

Conclusions: These studies demonstrate that the heterologous expression of *BnaSMT* can enhance the capacity for Se accumulation. Therefore, we developed transgenic rapeseed materials, which will be subjected to subsequent phenotypic evaluation. This study elucidates the crucial role of this gene in Se biofortification in rapeseed, providing theoretical support for the development of selenium-enriched rapeseed varieties.

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Selenium Deficiency-Induced Th1 Polarization and Inflammation in Porcine Thymus via SelM-Targeted LATS2 Regulation of the Hippo/mtROS/NF-κB Axis

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Introduction: Selenium (Se) deficiency can cause immune organ damage and immune dysfunction. It has been shown that SelM is related to biological processes such as oxidative stress, energy metabolism, and immune response. The thymus is an important organ for the development and maturation of T cells. Mitochondrial biosynthesis and energy metabolism, as well as NF- κ B pathway, are involved in the regulation of T cell differentiation. As an important regulator of cell proliferation, death and differentiation, Hippo pathway also plays an important role in regulating mitochondrial function and metabolism.

Methods: In this study, a SelM-deficient porcine thymus model and a SelM knockdown porcine thymus primary lymphocyte model were established. HE staining, co-immunoprecipitation test, transcriptomology, immunofluorescence, qRT-PCR, Western blotting, fluorescent probe staining, commercial kits and LC-MS were used. We observed changes in pathological morphology, seloproteins, Hippo pathway related indicators (LATS2, YAP, p-YAP, TAZ and p-TAZ), sulfur transfer pathway related indicators (CBS, CSE, AHCY, Cys, Met, Ser and Hcy), mitochondrial iron-sulfur cluster related indicators (NFS1, ISCU and FXN), and TCA cycle.. In addition, we detected forrelated indicators (IDH3A, PDHA1, ACO1, ATP, pyruvate, citrate, α -ketoglutarate and acetyl CoA), NF- κ B pathway related indicators (P65, p-P65, I κ B α , p-I κ B α , Ikk- β and p-Ikk- β), Th1/Th2 differentiation related indicators (T-bet, IFN- γ , STAT4, GATA3, IL-4, IL-5, IL-13 and STAT6) and inflammatory cytokines related indicators (IL-1 β , IL-6 and TNF- α).

Results and Discussion: After Se deficiency or SelM knockdown, the levels of SelM, YAP, and TAZ were decreased, while the levels of LATS2, p-YAP and p-TAZ were increased. Co-immunoprecipitation assay confirmed that SelM interacted with LATS2 to activate the Hippo pathway and promoted the nuclear translocation of YAP and TAZ. After Se deficiency or SelM knockdown, the expression of sulfur transfer pathway related indicators decreased, the content of Cys was significantly decreased, and the content of Hcy was accumulated. The inhibition of LATS2 confirmed that SelM knockdown inhibited sulfur transfer-dependent cysteine synthesis through activation of Hippo pathway. After Se deficiency or SelM knockdown, the expression of mitochondrial iron-sulfur cluster related indicators decreased, the contents of TCA cycle products ATP, citrate and α -ketoglutaric acid decreased, while the contents of pyruvate and acetyl-coa increased. The release of mtROS was increased. LATS2 inhibitor GA-017 treatment or Cys supplementation confirmed that the cysteine deficiency caused by SelM knockdown inhibited mitochondrial iron-sulfur cluster synthesis, impaired mitochondrial function, and increased the release of mtROS by activating the Hippo pathway. Furthermore, Se deficiency or SelM knockdown activated the NF-kB pathway, and supplementation of Cys in SelM knockdown cells confirmed that SelM knockdown activated the NF-kB pathway by inhibiting the trans sulfur pathway dependent cysteine synthesis and resulting in mtROS. After Se deficiency or SelM knockdown, the expression of inflammatory factors was upregulated, the expression of Th1 differentiation indicators was up-regulated, and the expression of Th2 differentiation indicators was down-regulated. The addition of Cys, MitoPQ and NF-KB pathway inhibitor BAY11-7082 confirmed that SelM induced inflammation and Th1 cell polarization in thymus tissue through the Hippo/mtROS/NF- κ B pathway.

Conclusions: Selenium deficiency led to downregulation of SelM and activation of the Hippo pathway by targeting LATS2, which in turn inhibited transculturation pathway-dependent cysteine synthesis, leading to impaired synthesis of mitochondrial iron-sulfur clusters as well as reduced levels of TCA cycling and ATP synthesis, accumulation of mtROS, activation of the NF- κ B pathway, and ultimately inflammation and polarization of Th1 cells.

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Selenium Deficiency Induces Aberrant Th17 Differentiation in Porcine Spleen via GPX3-Targeted PIG3 Regulation of the Mitochondrial DNA Release

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Introduction: Th17 cells, a critical subset of $CD4^+$ T cells, play pivotal roles in host defense against extracellular pathogens and the pathogenesis of autoimmune diseases. Studies have demonstrated that selenium (Se) deficiency impairs the responsiveness of T lymphocytes to mitogenic stimulation. The loss of glutathione peroxidase 3 (GPX3) disrupts lymphocyte development, proliferation, and differentiation. The p53-inducible gene 3 (PIG3) protein, an NADPH-dependent reductase, is involved in regulating oxidative stress responses and DNA damage repair.

Methods: The present study established a selenium-enriched porcine kidney cell (PK-15) model and employed coimmunoprecipitation, immunofluorescence co-localization, and protein-protein molecular docking to validate the targeted interaction between GPX3 and PIG3. Concurrently, the selenium-deficient porcine splene injury model, the selenium-deficient porcine splenic lymphocyte model, and the GPX3-knockdown porcine splenic lymphocyte model, combined the GPX3knockdown and PIG3-overexpression porcine splenic lymphocyte model and the exogenous autophagy flux inhibitor Bafilomycin A1 (Baf-A1) intervention model. Using transmission electron microscopy, antioxidant function assays, immunofluorescence staining, confocal laser microscopy, transcriptomics, Western blotting and qRT-PCR, we observed ultrastructural changes in spleen tissues. We measured oxidative stress levels, evaluated mitochondrial function (NDFB8, SDHB, UQCRC2, MTCO1 and ATP5A1), mitochondrial dynamics (Mfn1, Mfn2, OPA1, Drp1, Mff, Fis1, PTEN, Parkin, Pink-1, Beclin-1, p62 and LC3II) and mtDNA release, and assessed the expression of Th17 cell differentiation-related markers (IL-17A, IL-23R, RORγt and CCR6).

Results and Discussion: After Se deficiency or GPX3 knockdown, the following effects were observed: 1) levels of GPX3 and PIG3 were decreased, and the direct interaction between GPX3 and PIG3 was confirmed by protein interaction analysis, Co-IP and immunofluorescence colocalization; 2) activities of antioxidant enzymes (CAT, GSH-PX and T-SOD) were decreased, ROS and H₂O₂ were accumulated, and oxidative stress was triggered; 3) function of mitochondrial electron transport chain was impaired, ATPase activity and ATP production were inhibited, and mitochondrial function was impaired; 4) mitochondrial dynamics became imbalanced, characterized by downregulation of fusion genes and upregulation of fission genes, along with elevated expression of mitophagy-related genes, and the mCherry-GFP-LC3B dual fluorescence assay further demonstrated impaired mitophagy, and 5) mtDNA leakage occurred into the cytoplasm; and the differentiation of Th17 cells was increased. In this regard, PIG3 overexpression effectively alleviated the above results caused by GPX3 knockdown, whereas treatment with the autophagy inhibitor Baf-A1 abolished the protective effects of PIG3. Furthermore, Baf-A1 treatment demonstrates that GPX3 knockdown-induced mitophagy impairment exacerbates mtDNA release, thereby promoting abnormal Th17 cell differentiation.

Conclusions: Selenium deficiency led to the accumulation of ROS through GPX3 targeting PIG3, which in turn led to increased mitochondrial fission and mtDNA leakage. At the same time, selenium deficiency blocked the normal occurrence of mitophagy by GPX3 targeting PIG3, and the accumulated mtDNA could not be removed by mitophagy pathway, which further aggravated the leakage of mtDNA and finally led to abnormal active differentiation of Th17 cells in porcine spleen.

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Calcium Ion Regulates Selenium Uptake, Subcellular Distribution, and Gene Expression in Barley During Germination

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Introduction: Agronomic Se biofortification of cereals is a pragmatic solution, but biological limitations restrict the uptake and grain accumulation of Se. Calcium (Ca²⁺), as an essential nutrient and signalling molecule, influences transporter activity and redox balance. Recent studies suggest that Ca²⁺ may modulate Se metabolism, although the underlying mechanisms and optimal dosages remain unclear. We investigated whether adjusting the concentration of external Ca²⁺ during barley (*Hordeum vulgare* L.) germination can increase Se accumulation, while preserving seedling vigour and grain quality. This approach offers a simple, low-cost method for Se biofortification in Se-deficient regions.

Methods: Barley seeds were surface-sterilised and soaked for 48 h in solutions containing 150 μ M selenite alone or supplemented with calcium chloride at concentrations of 0.5, 1.0, 2.0, 4.0 or 8.0 mM. The seeds were then germinated at 18 °C under 95% relative humidity. Triplicate samples were taken on day 1 for biomass, respiration and signalling molecules and dissected into rootlets, acrospires, husk and endosperm. Total, inorganic and organic Se concentrations were quantified using hydride-generation atomic fluorescence spectroscopy; sub-cellular and protein-fraction Se by differential extraction. The relative expression of nine Se-related transport and metabolism genes was analysed by qRT-PCR. Data were statistically evaluated by one-way ANOVA, with $\alpha = 0.05$.

Results and Discussion: The addition of Ca²⁺ significantly influenced both the uptake and subcellular distribution of Se during barley germination. Supplementation with 0.5-4 mM Ca2+ to 150 µM selenite increased total Se uptake by 15-39 %, with the highest concentration (10.13 mg kg⁻¹ DW) observed at 4 mM Ca²⁺, a 39% increase relative to the selenite-only control. While moderate levels of Ca2+ maintained seedling vigor, 8 mM Ca2+ depressed fresh weight and respiration by 14 % and 33 %, respectively, indicating that excessive Ca²⁺ intensified Se stress. Tissue mapping revealed that Ca²⁺ redirected Se from the storage husk/endosperm to the metabolically active acrospires at Ca concentrations of 0.5 mM, whereas translocation was limited at 4 mM. This observation revealed a threshold beyond, which Se transport factor values declined. At the subcellular level, Ca²⁺ promoted the sequestration of Se into the cell wall and cytoplasmic fractions, thereby alleviating the burden of excessive Se on sensitive organelles. Protein-fraction analysis further showed that 2 mM Ca²⁺ shifted Se into the nutritionally valuable globulin and glutelin fractions, whereas 0.5 mM and 8 mM favored the enrichment of the albumin and gliadin fractions. At the molecular level, Ca²⁺ upregulated Se transporter HvPT and assimilation genes HvSiR (3.6-fold) and HvCS (1.3-fold), facilitating the conversion of inorganic into organic forms. However, high level of Ca^{2+} downregulated HvSULTR and HvPTR, restraining further influx and peptide transport. Simultaneous increases in hydrogen peroxide (H₂O₂), nitric oxide (NO), and proline were strongly correlated with Se translocation factors. These results indicate the involvement of a $Ca^{2+}-H_2O_2$ -NO signaling network that mitigates oxidative stress, while promoting Se mobilization. Together, these findings demonstrate that fine-tuning the concentration of external Ca^{2+} between 0.5 and 2 mM maximizes Se enrichment by enhancing uptake, safe compartmentalization and gene-level assimilation. However, excess Ca²⁺ counteracts these benefits by hindering growth and restricting Se transport.

Conclusions: Moderate levels of external Ca^{2+} boosted Se enrichment in barley by enhancing transporter expression, accelerating conversion to organic forms, and safely compartmentalizing Se. However, higher levels of Ca^{2+} reversed these benefits and impaired seedling vigor. Therefore, harnessing the Ca^{2+} -Se interaction provides a practical and scalable strategy to enhance Se biofortification without compromising early-stage crop growth.

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Selenium's Role in the Unified Theory of Soft Electrophile Toxicities

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Introduction: Exposures to neurotoxicants such as mercury (Hg), cadmium (Cd), and other metallic or organic soft electrophiles (E^*) cooperate in sequestering selenium (Se), the most potent intracellular nucleophile ^[1,2]. While uptake rates, tissue kinetics, and distributions of E^* vary in relation to exposure, their thermodynamic affinities for Se ensure their contributions cooperate in Se binding. Although individual E^* concentrations may not approach toxic levels, their combined concentrations may be sufficient to inhibit selenoenzyme synthesis and activities. Loss of essential selenoenzyme activities may result in subtle pathological consequences such as increased oxidative damage.

Overview: The physiological activities of Se occur through enzymes that employ selenocysteine (Sec), the 21st genetically encoded amino acid. The human genome includes 25 genes coding for Sec in the primary structure of enzymes that employ it in their active sites to catalyze reactions which require its unrivaled redox capacity. Over half of these selenoenzymes are involved in preventing and reversing oxidative damage while others control thyroid hormones, calcium activities, immune responses, and perform other vital functions. Electron poor E*s initially bind with cysteine (Cys) an abundant (~300 mM) nucleophile whose thiol (pKa 8.3) readily forms Cys-E* adducts. Since the Cys moieties of certain molecules are selenoenzyme substrates, Cys-E* adducts function as suicide substrates which orient E* for direct interactions with the active site Sec ^[1,2]. The higher affinities of E* for Se result in its transfer to form Sec-E*, irreversibly inhibiting the enzyme's activity. While organic E* binding affinities are far lower than metallic E*, their exposure sources and concentrations are far more abundant.

Future Directions: To reliably assess hazards associated with E^* exposures, the stoichiometries and binding affinities of individual E^* will need to be defined to establish their aggregate exposure risks ^[2]. Populations with poor dietary Se status will clearly be far more vulnerable to the effects of Se sequestration as a result of high E^* exposures. Further studies will need to assess potential adverse synergies that may arise when high E^* :Se stoichiometries accompany exposures to toxicants that independently initiate free radical cascades or produce reactive oxygen and reactive nitrogen species.

Discussion: The combined effects of exposures to Se-binding components of environmental, pharmaceutical, and dietary E^* require study since the loss of selenoenzyme activities would accentuate oxidative damage to tissues and contribute to accumulating DNA damage. Because somatic mutations contribute to the etiologies of many ^[3,4] if not most or all degenerative diseases, defining the effects of concurrent E^* exposures on selenoenzyme synthesis and activities may enhance the ability of preventative medicine to reduce disease pathogenesis and support healthy aging.

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Impact of Selenium Forms on Cadmium and Nickel Accumulation and Their Mutual Interactions in Rice (*Oryza sativa* L.)

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Introduction: Rice (*Oryza sativa* L.) cultivation increasingly faces concerns over metal contamination, particularly the accumulation of cadmium (Cd) in grains ^[1], which can exceed legal safety thresholds. In recent times, increasing attention has been paid to nickel (Ni) as an additional contaminant in rice grain ^[2]. While Cd has been extensively studied, knowledge of Ni behavior and its interaction with Cd in rice remains limited ^[3]. Selenium (Se), commonly used in rice biofortification ^[4], has shown potential to reduce toxic metal accumulation in some crop species. Therefore, this study investigates the interplay between Ni and Cd accumulation in rice and examines the influence of different Se forms, selenate (Se(VI)) and selenite (Se(IV)) on these interactions.

Methods: Rice plants were grown hydroponically and treated for one week under various treatments combining Cd (5 or 20 μ M), Ni (20 μ M), and Se (5 μ M) as either Se(VI) or Se(IV). Parameters assessed included plant biomass, lipid peroxidation, elemental accumulation, and the tissue distribution of Se and Ni via μ XRF 2D imaging.

Results and Discussion: Exposure to Cd and Ni independently reduced leaf and root biomass by 40-50% and triggered oxidative stress. Their combined exposure did not further diminish leaf growth; however, Cd significantly reduced root Ni accumulation (by approximately 50%) under equimolar conditions, likely due to competition at shared uptake sites. The addition of Se partially alleviated the toxicity of Ni and low Cd levels, particularly enhancing root growth compared to plants treated with Cd and Ni without Se supplementation, suggesting an antioxidant role for Se. However, co-treatment with high Ni and Cd concentrations impaired Se uptake, reducing Se(IV) accumulation in roots by 60% and Se(VI) in leaves by 77%, and intensified oxidative damage when Se(IV) was present. The Se form influenced its protective effect on rice: Se(VI) more effectively reduced Ni accumulation, while Se(IV) was more efficient in lowering Cd (by 45–75%) and Ni in the absence of Cd (by 50%). μ XRF revealed that in Se(VI)-treated plants, Se was diffusely distributed in the central cylinder, cortical parenchyma, and possibly rhizodermal cells of roots, and accumulated along veins and leaf tips. In Se(IV)-treated plants, Se accumulated more in roots and less in leaves. Nickel was mainly localized in xylem vessels and lateral root emergence zones, absent in cortical parenchyma. Nickel increased in roots and slightly in leaves of Ni-treated plants but decreased with the presence of Cd. Consistent with elemental analysis obtained via ICP-MS, both Se forms reduced Ni levels, especially in roots when combined with Cd, and Cd also reduced Se accumulation, particularly in leaves.

Conclusions: Selenium supplementation in most cases could alleviate metal-induced stress and limit Cd and Ni accumulation in rice in both leaves and roots, although the simultaneous presence of both metals may compromise Se uptake and Se biofortification efficiency.

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Effects of Manure-biochar, Foliar Selenium, Silicon, and Zinc on Cadmium Accrual, Soil Microbial Diversity, Root Metabolites, and Health Risk in Rice

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Introduction: The Food and Agriculture Organization reported that rice contributed to over 8% of worldwide crop production in 2019^[1]. Rice's substantial yield and nutritional content renders it a favored choice among farmers and consumers^[2]. Cadmium (Cd) accrual in rice poses several plant defects and also human health risks, as rice is a staple food in many regions^[2]. Several strategies have been proposed to mitigate Cd uptake, including the use of organic amendments^[2]. Manure-biochar (BC) reportedly improves soil health while Se, Si, and Zn are known for their protective roles in plants^[2-4]. This study uses a co-application of treatments approach to evaluate their synergistic effects on Cd accumulation in rice, soil microbial diversity, and root metabolite profiles. We hypothesize that the co-application of manure - BC with foliar Se and additional foliar Si and Zn may enhance growth, alter the soil microbial community, influence root metabolite profiles, and reduce Cd health risk.

Methods: The study applied four treatments: CK (control), T1 (2% manure-BC + 4 mg/L foliar Se), T2 (2% manure-BC + 4 mg/L foliar Se + 100 mg/L foliar Si), and T3 (2% manure-BC + 4 mg/L foliar Se + 100 mg/L foliar Zn) to evaluate their effects on rice growth, Cd accumulation, soil microbial diversity, root metabolite and health risks.

Results and Discussion: At maturity, T1-T3 showed significant increases in pH and cation exchange capacity compared to CK and a notable reduction in redox potential. T3 achieved the highest number of panicles and panicle weight, exhibited the greatest increase in superoxide dismutase activity and reduction in malondialdehyde content. HMA2 and LCT1 were notably downregulated in T1–T3, with T3 exhibiting the lowest expressions. T3 modulated key metabolites in the rice roots. Cadmium concentrations in plant tissues were significantly lower in T1-T3 compared to CK, with T3 showing the greatest reduction. The grain Cd content of T3 was 52.73% lower than T1 and 72.63% less than CK. Moreover, T1, T2, and T3 increased the Chao1 and Shannon indices. T3 also resulted in the highest increases in starch and protein content and the lowest ADI and HQ.

Conclusions: The results suggest that T3 promotes rice growth and quality by mechanisms such as downregulating Cd transport genes, modulating key root metabolites, and enhancing soil microbial diversity. This combination of treatments holds potential for improving rice production and reducing the health risks associated with Cd exposure.

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Reassessing Cd Toxicity in Se-Coexistent Soil-Crop Systems

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Introduction: In China, cadmium (Cd) contamination in agricultural soils is regulated under two thresholds: a risk screening value of 0.3 mg/kg and a risk intervention value of 1.5 mg/kg^[1]. For food safety, maximum permissible Cd levels are set at 0.1 mg/kg in cereals (excluding rice), 0.2 mg/kg in rice, and 0.05 mg/kg in fresh vegetables ^[2]. Notably, current regulatory frameworks for Cd exposure risk do not account for potential nutrient interactions, particularly with selenium (Se), a well-documented Cd antagonist. While extensive evidence confirms that Se mitigates Cd toxicity in soil-crop systems through antagonistic effects ^[3], regulatory guidelines fail to incorporate this critical biointeraction between Se and Cd, leaving Cd risk assessments incomplete in naturally Se-rich regions where the co-existence of Se and Cd is prevalent.

Methods: Field studies: Rhizosphere soil, root, stem, and leaf samples were systematically collected from soil-corn systems across natural Se-Cd co-enriched regions during both grain-filling and mature growth stages in corn (*Zea mays* L.). Laboratory experiments: Hydroponic cultivation of *Cardamine hupingshanensis* (Se-Cd hyperaccumulator) was conducted to investigate Se-Cd interactions. Animal test: Mice were utilized *in vivo* to assess the modulation of Cd toxicity by Se, with particular focus on organ-specific effects.

Results and Discussion: Field data revealed that soil bioavailable Se:Cd molar ratios ≤ 0.7 enhanced Cd accumulation, while ratios > 0.7 significantly inhibited Cd uptake in corn without impacting Se bioavailability ^[4]. Hydroponic studies confirmed this molar ratio-dependent relationship, demonstrating optimal Se:Cd molar ratio (1:1) in growth media minimized Cd translocation and biotransformation, while reducing phytotoxicity of Cd. Results from mouse trials showed that *Cardamine hupingshanensis* (source of selenocystine (SeCys₂)) effectively attenuated Cd-induced hepatotoxicity through regulation of SLC7A11/GPX4 signaling pathways and suppression of lipid peroxidation ^[5]. Concurrently, SeCys₂ exhibited dual osteoprotective effects—modulating renal FGF23 expression, restoring serum 1,25(OH)₂D₃/PTH homeostasis, and reshaping gut microbiota profiles to counter Cd-induced bone resorption ^[6].

Conclusions: This systematic investigation demonstrates that traditional Cd risk assessments significantly overestimate toxicity in Se-coenriched food systems. The identified Se:Cd molar ratio thresholds and species-specific mitigation mechanisms necessitate urgent revision of food safety standards for Cd, particularly in China's naturally Se-rich agricultural regions. Future regulatory frameworks should integrate biogeochemical interaction parameters to enable precision risk management.

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Mechanisms of Selenium Enrichment in Wheat and Synergistic Regulation by Nitrogen and Nitric Oxide in Selenium-Rich Regions of China

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Introduction: The heterogeneous distribution of soil selenium (Se) and suboptimal grain Se enrichment efficiency in wheat presents critical challenges across China's primary wheat-producing regions.

Methods: This study synthesized literature data (from 2001 to 2021, at 123 sampling sites) with field experiments (conducted in 2018 – 2023 in the Se-rich region of Shanxi, China) to systematically elucidate the soil-grain Se relationship and investigate the mechanisms underlying Se utilization in black-grained wheat (BGW) under nitrogen (N) and exogenous nitric oxide (NO) regulation^[1].

Results and Discussion: Key findings revealed the following: (1) While 21.14% of sampling sites exhibited Se-rich soils (Se \geq 400 µg/kg), only 13.83% of wheat grains met the Se-rich standard (>150 µg/kg), with a significant positive soil-grain Se correlation (p < 0.05). Distinct Se-enriched regions were identified in Changzhi and Hongtong in Shanxi, and Xiantao in Hubei. (2) Black-grained wheat grains demonstrated 10.8–15.3% higher Se content (181.42–192.37 µg/kg) compared to common wheat (163.91 - 166.60 µg/kg), with purple-grained BGV Jizi 496 achieving the highest proportion of organic Se (94.97%) and high root-to-grain translocation efficiency. Despite a 7.2 - 12.5% yield reduction, BGW exhibited superior Se absorption capacity. (3) Optimal N application (192 - 216 kg/ha) enhanced soil-available Se (water-soluble + exchangeable forms: 12.45 - 12.66%) and elevated grain Se content (168.85 - 194.59 µg/kg) by modulating leaf enrichment coefficients (+0.8) and root-to-grain translocation, while maximizing yield (6304 - 7922 kg/ha). (4) Exogenous NO alleviated high-Se stress (\geq 50 µmol/L), reducing leaf Se accumulation by 17.43 - 41.67%, upregulating Se detoxification genes (HvMMT1, GSH1; 1.5-2.3-fold), and improving photosynthetic efficiency (23.6% increase in net rate) and antioxidant capacity (18.9% decline in H₂O₂, 34.5% increase in T-AOC).

Conclusions: This study proposes an integrative strategy combining BGW cultivation, optimizing N management (192 - 216 kg/ha) and NO application to enhance Se biofortification and stress resilience. The integrated strategies provide theoretical and practical frameworks for functional wheat production and sustainable agriculture in Se-rich regions.

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Dynamic and Stable Core Microbiota Assist Plants in Enriching Selenium and Reducing Cadmium Absorption

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Introduction: The coexistence of selenium (Se) and cadmium (Cd) seriously limits the development of Se-enriched agriculture. Mechanisms by which plants regulate rhizosphere microbes to manage Se/Cd availability under coexisting beneficial and harmful elements in plant different growth stages remain unclear. Therefore, the main aims of the present study were to (1) investigate the dynamic changes of rhizosphere microbiome and the chemical forms of Se and Cd changes across the different developmental stages; (2) study whether there is a group of rhizosphere microbiotas that are highly related to the availability of Se and Cd and participate in the rhizosphere Se and Cd cycle; and (3) construct a synthetic microbial consortium from the rhizosphere microbiotas that regulate the availability of Se and Cd in soils.

Methods: Pot experiments with representative soils containing different levels of Se and Cd were set up to perform nondestructive continuous sampling of rhizosphere soil. The rhizosphere soil of the local main crop *Brassica napus* was collected at six different stages of its growth cycle. Microbiome analysis, metagenomics, genomic analysis, strain isolation, and functional validation were performed to investigate the dynamic changes of rhizosphere microorganisms, the relationship between key metabolic processes and the availability of Se and Cd in rhizosphere^[1]. Finally, synthetic microbial community related to Se-Cd availability and key glutathione-related-amino acids were constructed by rhizosphere microbial isolation, and their effects on enhancing Se enrichment and reducing Cd in plants were determined.

Results and Discussion: Result showed that a significant negative correlation was observed between the rhizosphere available Se and Cd content across plant whole growth cycle and we identified a group of core microbiota that were highly positively correlated with available Se and negatively correlated with available Cd. Genomics and metagenomics analyses revealed that the core microbiota had as higher substrate preference for amino acids related to the glutathione metabolic pathway. Key glutathione-related-amino acids and synthetic microbial community significantly improved the expression of glutathione anabolism and related amino acid transport genes and enhanced Se uptake and reduced Cd absorption in plants grown in various Se-Cd-rich soils. This study provides insights into the mechanisms of root-associated microbes responding to complex soil nutrients during plant growth.

Conclusions: The availability of Se and Cd in the rhizosphere can be affected by regulating the rhizosphere microbial abundance during plant growth in Se and Cd associated areas. The glutathione metabolism of microorganisms significantly contributed to the activation of Se and passivation of Cd. Inoculation of amino acids related to glutathione metabolism pathway and specific groups of rhizosphere microorganisms could significantly promote plant Se uptake and reduce plant Cd uptake.

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Synergistic Regulation of Selenium and Cadmium in Rice: Achieving a Balance between Antioxidant Stress Response and Quality Enhancement

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Introduction: Selenium (Se), a critical trace element with established antioxidant and chemopreventive functions, is an essential micronutrient exclusively derived from diet due to human inability to synthesize it endogenously^[1]. As a staple for over half the global population, particularly in China, rice (*Oryza sativa* L.) serves as a primary dietary Se source^[2]. However, its high cadmium (Cd) accumulation capacity threatens food safety, given Cd nephrotoxic, osteotoxic, and carcinogenic effects. While prior studies highlight Se-mediated Cd toxicity mitigation, the mechanistic impacts of varying Se/Cd concentration combinations on rice physiology, grain quality, and Cd partitioning remain unclear. This study employs controlled pot experiments to investigate their regulatory effects on antioxidant enzymes, grain traits, and Cd accumulation, aiming to decipher Se-driven detoxification pathways and inform Se-biofortified, Cd-safe rice systems.

Methods: The rice pot experiments were conducted in greenhouses of Yichun University's Agronomy Base, using CdCl₂·2.5H₂O for Cd stress and humic acid-chelated Se. A 4×4 factorial design included 4 Cd levels (4-32 mg/kg) and 4 Se rates (0-25 L/ha), with 4 replicates. Antioxidant enzymes (POD, SOD, CAT) were assayed via colorimetric, NBT photoreduction, and UV absorption methods. Grain quality parameters (amylose, protein, amino acids, gel consistency) were measured using titration, biuret, HPLC-MS, and GB/T 22294-2008 ^[3]. Element contents (Se/Cd) were determined by HG-AFS and GFAAS (GB 5009.93-2017/GB 5009.15-2014) ^[4]. Data were analyzed via SPSS 29.0 and visualized in Origin 9.0, establishing a systematic methodology for Cd-Se interaction research in rice.

Results and Discussion: Under single Cd stress, rice exhibited biphasic antioxidant enzyme (SOD, POD, CAT) activities: initial activation to counteract ROS, followed by decline beyond Cd tolerance thresholds. Selenium supplementation reduced amylose content, enhancing grain palatability ^[5], while significantly increasing protein content and gel consistency (GC) under low/medium Cd stress. Even under high Cd stress, low-dose Se has improved protein levels and GC, though high-dose Se decreased GC. Single Cd stress depleted threonine and lysine, but Se supplementation restored essential amino acid profiles. A soil Cd concentration of 8 mg/kg with 20 L/ha Se application optimized grain Se enrichment while minimizing Cd accumulation, demonstrating a dose-dependent strategy for Se-biofortification to concurrently enhance nutritional quality and mitigate heavy metal toxicity in rice.

Conclusions: This study systematically analyzed Cd stress and Se supplementation effects on rice physiology and quality. Single Cd exposure affected antioxidant enzyme activity and grain quality. A low Se concentration mitigated Cd toxicity, while a high Se concentration showed synergistic toxicity. There was a dose-dependent element interaction. The optimal combination offers guidance for agriculture.

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Selenium Inhibited Cadmium in Rice Cultivated in Mildly Cadmium-Contaminated Soil

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Introduction: Soil cadmium (Cd) pollution has become a major environmental issue that threatens food security and human health ^[1]. In geological surveys, it was found that there may be an antagonistic effect between selenium (Se) and Cd in natural crops ^[2]. To investigate the inhibitory effect of Se and Cd accumulation in rice, we conducted a pot experiment on mildly Cd contaminated soil (total Cd 0.523 mg/kg) to explore the effect of exogenous Se application on the absorption and transport of Se and Cd in rice.

Methods: A pot experiment was conducted in Jiangsu China in 2024. The total Se content in the soil background is 0.267 mg/kg, and the Se/Cd molar ratio is 0.72. By applying sodium selenite to the soil during the basal fertilizer period, the soil Se/Cd molar ratio is increased by 0 (CK), 0.5 (S1), 1.0(S2), and 1.5(S3). During the rice heading stage, late flowering stage, milk ripening stage, and maturity stage, rice roots, stems, leaves, panicles, shells, rice and other organs were collected to detect their biomass, Se and Cd content, and we analyzed the accumulation characteristics and interaction effects of Se and Cd.

Results and Discussion: Moderately increasing the soil Se/Cd molar ratio can significantly improve rice growth. When the soil Se/Cd molar ratio increased by 1.0, that is, increased to 1.72 (1.0 + 0.72 background), the biomass of rice increased by 36.5% compared to the CK group, and the plant height increased by 5.8%, indicating that moderate Se can promote rice development by alleviating Cd stress. Regarding Se content in rice, Se application can significantly increase the Se content in various organs of rice, and the distribution of Se is relatively uniform in each organ. The order of Se content in each organ from high to low is: leaves > roots > stems > shells > rice. The Se content in rice grains significantly increased with the increase of Se application rate. The S1, S2, and S3 groups increased by 339%, 575%, and 922% respectively compared to the CK group. Exogenous Se has a significant inhibitory effect on the migration of Cd in rice. The best Cd reduction effect was observed when the molar ratio of Se to Cd increased by 0.5 (S1), especially during the booting and flowering stages, the Cd content in roots, stems, and leaves decreased by 36.1%, 32.3%, and 57.4%, respectively, compared to the CK group. Under the condition of a soil Se/Cd molar ratio of 1.72, the Cd content in rice grains decreased from 0.270 mg/kg to 0.183 mg/kg, a decrease of 32.1%, which meets the national food safety standard of China (<0.2 mg/kg). At the same time, the Se content in the grains increased to 0.292 mg/kg, meeting the standard for Se enriched rice (\geq 0.15 mg/kg). The dual requirements of reducing Cd and increasing Se have been achieved.

Conclusions: Selenium application with special Se/Cd molar ratio could inhibit Cd accumulation in rice cultivated in mildly Cdcontaminated soil. and the optimal molar ratio of Se to Cd has been determined to be about 1.72. Further experiments are needed in the future to explore the physiological regulation mechanism of Se/Cd interaction. These research results provide reference for the remediation of Cd contaminated paddy soil and the production of Se rich rice.

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Absorption of Selenium and Sulfur and Their Synergistic Competition Mechanism in Rapeseed

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Introduction: Selenium (Se) is a non-metallic but essential trace element for the human body. Plants are an important dietary source of Se for animals and humans. The level of Se accumulation in plants depends on the soil Se content and the level of competing S compounds. Selenate and selenite are the main forms of Se available for plant absorption depending on the chemical properties of the soil. Selenium is a chemical element belonging to the Chalcogenide group, which shares a common metabolic pathway with sulfur in plants. To promote the Se enrichment ability of crops and cultivate products with high nutritional quality, it is of great significance to elucidate the cooperative competition mechanism of Se sulfur (S) absorption and their respective utilization by plants.

Methods: The low glucosinolate and erucic acid rapeseed variety 9D632 was used in this study. Hydroponic cultivation was performed according to the procedures described by Wang et al. ^[1]. In the low sulfate (LS, containing 29.05 μ M sulfate) and half-sulfate (HS, containing 0.5 mM sulfate) groups, all or part of the MgSO₄ was replaced with MgCl₂. Na₂SO₄ was added to generate double sulfate (DS, containing about 2.0 mM sulfate) and quadruple sulfate (QS, containing about 4.0 mM sulfate) solutions. After six days of growth, either 10 or 100 μ M selenite was added to each nutrient solution group. Plant Se concentrations were determined after harvest using the hydride generation atomic fluorescence spectrometry (HG-AFS) method. The compositions and concentrations of different Se species were detected using high performance liquid chromatography-inductively coupled plasma mass spectrometry (HPLC-ICP-MS; Agilent 7900, Singapore).

Results and Discussion: In our study, we found that reducing the sulfate supply increased the Se content in the ground part of rapeseed seedlings by more than 6 times. Our analysis of Se morphology shows that at normal sulfate supply levels (1.0 mM sulfate), most of the root-absorbed selenite is converted to organic forms of Se compounds, and selenate and methylselenocysteine (MeSeCys) are the main Se compounds for root-to-ground transport. We also found that reducing sulfate supply enhanced genes associated with selenate distribution and key rate-limiting steps in organoselenium biosynthesis, including the aquaporin gene (*NIP2;1*), sulfate transporter gene (*SULTR2;2 SULTR4;1* and *SULTR4;2*), and APS reductase gene (*APR2*).

Conclusions: This study indicates that removing the majority of sulfate from a selenite hydroponic solution can enhance the rootto-shoot transport efficiency of Se, leading to a greatly improved accumulation of Se in the above-ground organs. A low sulfur environment may also promote the biosynthesis of organic Se from inorganic sources. These physiological findings suggest that it is feasible to produce desired Se-rich food or raw materials by reducing the contents of inorganic Se sources. Moreover, we were surprised to find that MeSeCys is the main Se species translocated from root to shoot. Further studies are necessary to determine the molecular and physiological mechanisms underlying the transporting and degradation of MeSeCys. These potential findings could then be applied to the production of even higher Se-accumulating crops.

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Concentration-Dependent Phosphorus Effects on Selenite Biochar Adsorption: Site Competition Driving Monolayer-to-Multilayer Transition

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Introduction: Selenium (Se) deficiency affects $\sim 72\%$ of agricultural land in China, necessitating exogenous Se application in agriculture. Interactions between phosphate (P) and selenite Se(IV) on soil interfaces influence Se bioavailability. However, the interaction mechanisms in the soil remain unclear. Biochar has been widely used in remediation of polluted or nutrient-deficient soils and may serve as a model interface. This study investigates how P concentrations modulate Se(IV) adsorption on biochar, which may be useful for developing more efficient strategies for Se-enriched crop cultivation.

Methods: This study deciphers the concentration-dependent role of phosphorus in regulating Se(IV) adsorption onto biochar through batch experiments. Corn straw-derived biochar was pyrolyzed (500°C, N₂ atmosphere). Batch adsorption experiments assessed Se(IV) adsorption under three Se gradients (low: $8.5-11.5 \ \mu g/L$; mid: $85-115 \ \mu g/L$; high: $850-1150 \ \mu g/L$) and four P concentrations (0, 30, 300, 3000 \ \mu g/L). Solutions (pH 5.5) were agitated (150 rpm, 25°C, 12 h), filtered (0.45 \ \mu m), and analyzed via atomic fluorescence spectrophotometry. Adsorption isotherms were fitted to Langmuir and Freundlich models. Biochar was characterized pre-/post-adsorption using SEM, TEM, XRD, and FT-IR.

Results and Discussion: At low Se: P exhibited dual effects: promotion (\leq 3%) at Se \leq 10 µg/L but inhibition (\leq 3.1%) at higher Se concentrations. The 3000 µg/L P showed the strongest effect. Mid Se: All P concentrations inhibited adsorption (0.42-2.61% reduction), with inhibition intensity scaling with P concentration. At high Se: Initial inhibition weakened as Se increased; maximum inhibition (3.40%) occurred at 3000 µg/L P. Isotherm modeling revealed a transition from monolayer (Langmuir-dominant, R² > 0.98 in low-Se controls) to multilayer adsorption (Freundlich-dominant, R² > 0.95 in high-Se systems) under P influence. Characterization (XRD, FT-IR) confirmed unchanged biochar crystallinity and functional groups, indicating physical adsorption dominated by pore occupation and site competition. Higher P concentrations intensified competition for adsorption sites.

Conclusions: Phosphorus concentrations critically regulate Se(IV) adsorption on biochar via site competition, with effects shifting from promotion to inhibition as Se levels increase. Low Se favors P-facilitated adsorption, while mid/high Se systems show P-dependent inhibition. The physical nature of adsorption implies that biochar's porous structure governs Se retention. Optimizing P/Se ratios in carbon-based fertilizers could enhance Se bioavailability in deficient soils.

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Selenium and Cadmium Integration Reduces Rice Blast Severity and Alters its Biochemical and Molecular Responses

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Introduction: Selenium (Se) is an essential trace metal or metalloid that plays an important role in human health. Rice is considered as a staple food for more than half of the world population and is an important source of Se. The nutritional quality of rice is affected by various biotic and abiotic stresses. In this study, sodium selenite and cadmium chloride were applied to rice plants to investigate the antioxidative system (SAR) SOD, POD, CAT, MDA, PAL, and PPO activities compared to application of water-soluble Se fertilizer, in rice shoots, antioxidant and disease resistance related gene expressions, and evaluation of Se on rice quality.

Methods: Various experiments have evaluated the efficacy of Se against the rice blast disease. An antifungal test was performed to evaluate the efficiency of combined application of Cd and Se on *M. oryzae* growth. For transcriptomic analysis RNA-extraction and qRT-PCR analysis were performed. The SOD, POD, CAT activity, and MDA content was analyzed by following the previously described protocol with slight modification^[1]. Selenium contents in rice grains were analyzed using ICP-MS. One-way ANOVA was used for variance of analysis with Duncan test p<0.05). DPS 7.0 was used for data processing and multiple comparisons.

Results and Discussion: The combined application of Na₂SeO₃+Cd (50 mg/L+50 μ mol/L) significantly inhibited the mycelial growth by 53.84 %. The results showed that water-soluble Se fertilizer treatment significantly increased the activities of SOD, POD, CAT, PAL, and PPO. With time, the activities of POD, SOD, and CAT increased as compared to untreated check. This experiment used the qRT-PCR method to analyze the expression of two disease-related and antioxidant genes, OsPR1a and OsPAL1. The results indicated that the disease-related genes OsPR1a and OsPAL1 gene expression significantly up-regulated. Chen et al. ^[3] explained that Se ions play a pivotal role in increasing the expression of disease-related and antioxidative genes to enhance resistance against *Rhizoctonia solani*, which causes sheath blight disease in rice plants. At higher concentration, 250mL/667m² Se content in the grains peaked at 0.3557 mg/kg (Fig. 1).



Figure 1. Impact of Se fertilizer on gene expressions (A), Se1 (50 µmol/L Cd+50 mg/L Na₂SeO₃), and Se2 (50 µmol/L Cd+500-fold selenium fertilizer) (B), Se content in Rice grains (C).

Conclusions: Overall, the study highlights the water-soluble Se fertilizer showed the increase of antioxidative enzymes (SOD, POD, CAT, PPO, and PAL) activities, while reducing the level of MDA, and up-regulation of OsPR1a and OsPAL1 gene expression. The combine application of Na₂SeO₃ and CdCl₂ reduced rice blast severity and activated the defense system.

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Selenium Alleviates Cadmium Stress in Rice by Enhancing Antioxidant Defense and Photosynthetic Activity

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Introduction: The physiological responses of plants play a key role in the uptake of heavy metals such as cadmium (Cd) and the alleviation of associated stress by selenium (Se)^[1]. Selenium is a beneficial element for higher plants, and its role in mitigating heavy metal toxicity has been widely documented ^[2]. However, under systematically designed concentration gradients of Cd and Se, the regulatory effects of Se on Cd uptake, translocation, and related physiological responses in rice remain insufficiently understood. This study investigated the effects of varying Se and Cd concentrations on Se and Cd accumulation in rice, antioxidant enzyme activities, and on photosynthetic performance.

Methods: A pot experiment was conducted using weathered black shale soil with six levels of Se including 0, 0.4, 0.8, 1.2, 1.6, and 2.0 mg/kg, and four levels of Cd (0, 2, 4, and 8 mg/kg), with a total of 24 treatment combinations. Each treatment had three replicates. When rice seedlings reached a height of 5 - 6 cm, Se and Cd solutions were injected into the water-saturated soil using syringes. Plant growth was observed throughout the experiment, and key physiological parameters were recorded and analyzed.

Results and Discussion: The Se application alleviated Cd stress in rice. Notably, Se concentrations of 0.8 and 1.2 mg/kg significantly inhibited Cd uptake in rice. Since Cd^{2+} is primarily absorbed via mineral element transporters and channels, Se may competitively interfere with plant uptake of Cd^{2+} , thereby reducing Cd accumulation in plant tissues ^[3]. Compared to control, the Se treatments at 0.8 and 1.2 mg/kg more effectively activated plant antioxidant defense system, as evidenced by increased activities of catalase (CAT) and ascorbate peroxidase (APX), which could enhance plant's tolerance to Cd toxicity. Additionally, the Se application significantly improved the net photosynthetic rate and photochemical efficiency (Fv/Fm). The photosynthetic rate was highest under moderate Cd stress with the Se treatment of 0.8 - 1.2 mg/kg.

Conclusions: With the Se treatment range of 0.8 to 1.2 mg/kg, the Se application effectively reduced Cd accumulation in rice and alleviated its toxic effects by activating the antioxidant defense system and improving photosynthetic activity. Overall, Se mitigates Cd stress in rice through the enhancement of both antioxidant capacity and photosynthetic performance.

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Selenium Absorption-Transformation Mechanisms and Se-Cd Interactions in Selenium-Enriched Asparagus

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Introduction: Selenium (Se) biofortification has received growing interest in enhancing the nutritional quality of crops ^[1]. Asparagus (*Asparagus officinalis* L.) is a known Se accumulator and a major export vegetable in China. However, selenium-rich soils are often co-contaminated with toxic cadmium (Cd), which poses a challenge for safe biofortification. This study aimed to investigate the absorption, translocation, and enrichment of Se in asparagus under varying Se and Cd soil conditions, and to explore the interaction between the two elements. We hypothesized that exogenous Se application can reduce Cd accumulation in asparagus, while enhancing Se uptake and translocation.

Methods: Pot experiments were conducted in soils with different native Se and added Cd treatment levels at 0, 1, 2, and 5 mg/kg. Asparagus plants were treated with different types and concentrations of Se fertilizers (inorganic, organic, and nano-Se). Concentrations of Se and Cd in roots, stems, and leaves were measured using ICP-MS. Translocation and enrichment coefficients were calculated. Statistical analyses were conducted by one-way analysis of variance (ANOVA) followed by Duncan's test at $\alpha = 0.05$.

Results and Discussion: Selenium accumulation was highest in leaves, followed by stems and roots. Translocation from stem to leaf was efficient, while soil-to-root migration was limited. Low-level Se application of 1 mg/kg significantly enhanced Se accumulation in different plant tissues compared to the highest Se treatment of 5 mg/kg. Exogenous Se effectively reduced Cd uptake, especially in stems, showing up to 83.5% reduction. Under high Cd treatments, Se displayed a non-linear interaction, while showing stronger mitigation effects with low concentration treatments. These findings demonstrate the potential of having low Se fertilization for safe Se biofortification in Cd-contaminated soils.



Figure 1. Migration (a) and enrichment (b) coefficients of Se in different organs of asparagus. Means with different letters are significantly different between different organs for each treatment at p < 0.05.

Conclusions: Selenium biofortification in asparagus is most effective at low Se concentrations and Se primarily accumulates in leaf tissue. Exogenous Se can alleviate cadmium-induced toxicity and reduce Cd accumulation in edible tissues. These results offer a promising strategy for producing high value Se-enriched vegetables on moderately Cd-contaminated farmland, contributing to world food safety and soil utilization.

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Sulfur-Mediated Iron Plaque Enhances Selenium Uptake and Reduces Cadmium Accumulation in Rice in Cd-Contaminated Seleniferous Soil

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Introduction: Cadmium (Cd) contamination commonly coexists with selenium (Se) in naturally seleniferous soils due to inherent geochemical associations, which substantially constrain the sustainable exploitation and agricultural utilization of Se resources. Although sulfur fertilization is widely used in flooded paddy soil to reduce Cd bioavailability via sulfide-induced immobilization, its effects on Se biogeochemistry remain poorly understood. Although previous studies have reported that sulfur fertilization under dryland conditions can reduce Se accumulation in wheat ^[1], the effect of sulfur on Se dynamics under flooded conditions remains unclear. As a redox-sensitive element, Se responds strongly to environmental redox fluctuations, and the distinct redox characteristics of upland versus flooded soils may lead to divergent outcomes in Se speciation and bioavailability. Which limits the development of effective strategies for optimizing Se management and improving rice Se biofortification in paddy ecosystems. Therefore, this study aimed to investigate the effects of sulfur application on Se bioavailability in Cd-contaminated seleniferous soils and to assess its impact on Se and Cd accumulation in rice.

Methods: A soil pot experiment was conducted with the application of two forms of sulfur, elemental sulfur (S⁰) and sulfate (S(VI)), at a rate of 200 mg/kg. Soil and plant samples were collected at the seedling stage (20 days after rice transplanting) and the tillering stage (40 days after transplanting). Soil Se bioavailability was assessed using the diffusive gradients in thin films (DGT) technique ^[2]. Se and Cd contents in rice were measured after nitric–perchloric acid digestion using hydride generation-atomic fluorescence spectrometry (HG-AFS) for Se and inductively coupled plasma optical emission spectrometry (ICP-OES) for Cd. Sequential extraction was used to isolate different iron oxides from the iron plaque on rice root surfaces, and iron concentrations were determined by o-phenanthroline colorimetry ^[3]. The ratio of ferric iron (Fe³⁺) to ferrous iron (Fe²⁺) in the iron plaque was determined by X-ray photoelectron spectroscopy (XPS). The speciation of Se adsorbed onto the iron plaque was analyzed by high-performance liquid chromatography coupled with atomic fluorescence spectrometry (HPLC-AFS).

Results and Discussion: Compared with the control, S(0) significantly increased rice biomass by 43.12% and 40.17% at the seedling and tillering stages, respectively, while S(VI) increased biomass by 11.49% and 28.29%. S(0) also markedly enhanced shoot Se content (by 413% and 335%) and reduced shoot Cd content (by 34.76% and 45.49%). In roots, S(0) increased Se content (by 84.22% and 7.17%) and decreased Cd content (by 22.27% and 42.18%). In contrast, S(VI) had no significant effect on Se accumulation but significantly reduced Cd levels in both shoots and roots. S(0) also reduced DGT-extractable Se in soil by 34.79% and 18.07%, while S(VI) had no significant effect. Both treatments promoted iron plaque formation, with S(0) increasing plaque content by 57.48% and 41.25%, and S(VI) by 16.97% and 10.66%. XPS analysis showed that sulfur application lowered the proportion of Fe(III) in iron plaques, which were mainly composed of ferrihydrite and lepidocrocite. Both treatments reduced Se and Cd bound to the plaques. Under S(0) treatment, Se was mainly present as Se(IV), potentially explaining its increased accumulation in rice. Se content was positively correlated with iron plaque formation, while Cd content showed a negative correlation.

Conclusions: The addition of sulfur (S0) significantly enhanced Se accumulation and reduced Cd uptake in rice grown in Cdcontaminated seleniferous soils, primarily through the modulation of iron plaque formation on roots, demonstrating sulfur amendment as an effective and environmentally sustainable strategy for utilizing seleniferous soils while mitigating heavy metal risks.

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Effects of Nitrogen Fertilizer on Soil Selenium Availability, Selenium Metabolism and Yield of Triticale in Selenium-Enriched Area

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Introduction: To investigate the effects of nitrogen (N) fertilization on selenium (Se) accumulation and translocation in seleniumenriched triticale production.

Methods: The field experiment was conducted at the Hongtong Wheat Science and Technology Courtyard in Shanxi from 2021 to 2023. Six N application rates (0, 60, 120, 180, 240, 300 kg/hm²) were set to investigate the impacts on soil Se availability, triticale Se accumulation, and yield formation. Soil samples (0- 20 cm) were collected at anthesis and maturity to determine total Se content ^[1], Se speciation ^[2], soil pH ^[3], and other physiological indices. Fresh flag leaves were sampled every 5 days after anthesis to measure activities of Se-metabolizing enzymes, including ATP-sulfurylase (ATPS), methionine-S-methyltransferase, and selenocysteine methyltransferase, to analyze the effects of N rates on Se metabolism.

Results and Discussion: Results showed that exchangeable Se was the most bioavailable form, and Fe-Mn oxide-bound Se could be converted to available Se under suitable conditions. N application influenced soil Se availability by regulating soil pH and organic carbon. Compared with 240 - 300 kg/hm², 180 kg/hm² N increased the proportion of exchangeable Se by 31.9 - 76.4% and Fe-Mn oxide-bound Se content by 4.2 - 16.6% at anthesis and maturity. Nitrogen significantly affected Se accumulation and enzyme activities: 240 kg/hm² N enhanced ATPS activity by 1.0 - 58.1%, methionine synthase activity by 2.6 - 97.2%, cysteine methyltransferase activity by 4.5 - 52.1%, and γ -glutamyl-cysteine synthetase activity by 4.9 - 74.8% in post-anthesis flag leaves.

Conclusions: Applying 180 - 240 kg/hm² N improves soil Se availability, enhances Se transport coefficients in vegetative organs, promotes post-anthesis Se accumulation, and increases grain Se content. We recommend this additional N as an optimal cultivation technique for selenium-enriched triticale in the region.

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Tomographic Mapping of Selenium in the Meristems of Hyperaccumulator Plant *Neptunia amplexicaulis*

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Introduction: *Neptunia amplexicaulis* is a selenium hyperaccumulator endemic to a small area of north-west Queensland, Australia. From its discovery in the 1950's, this plant has shown the capacity to accumulate > 4000 μ g Se/g ^[1]. When grown in controlled glasshouse conditions, this plant has demonstrated accumulation of up to 13 6000 μ g Se/g in its youngest leaves specifically ^[2]. Understanding where selenium (Se) is distributed within plant tissues is an essential component of understanding how and why this species is able to hyperaccumulate Se. This far, others have demonstrated that Se is transported around the plant and concentrated in phloem sinks (developing young leaves, root tips, flowers and seeds), while being cycled out of phloem sources (developed leaves)^[2].

Methods: Detailed elemental mapping of meristem tissue requires the use of Synchrotron X-Ray Fluorescence Microscopy-Computed Tomography ^[3]: virtual slices were taken of apical meristems and taproot tips of young Se-dosed *N. amplexicaulis* seedlings the DESY (the German Synchrotron) P06 beamline. Additionally, XANES mapping of root tissue and analysis of Sedosed *Neptunia heliophila* (non-accumulator comparison species) was conducted.

Results and Discussion: Selenium is located ubiquitously in the developing tissue of meristems, but once tissue begins to develop, Se is highly concentrated in the epidermis of developing leaf tissue and the ground meristem of the root tip before differentiating into the developing xylem tissues. XANES mapping shows a strong concentration of organic-Se species within root tips, suggesting this may not only be the site of uptake, but also metabolism of Se, in contrast to the classical models of hyperaccumulator metabolism^[4].

Conclusions: The translocation and metabolism of Se in *Neptunia amplexicaulis* is a unique, novel perspective from both hyperaccumulator and non-accumulator plant systems. Synchrotron tomographic methods provide a powerful tool for examining details of Se-distribution which, paired with molecular approaches, can elucidate the mechanisms of hyperaccumulation in this species.

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Advancing the Analysis of Volatile Selenium Species in High-humidity and Low-volatility Paddy Systems: A Novel Approach for Speciation and Quantification

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Introduction: Selenium (Se) volatilization represents a crucial component of the Se biogeochemical cycle in paddy systems. However, current existing methodologies for capturing volatile Se (VOSe) in high-humidity and low-volatile paddy systems are insufficient. Therefore, there is a strong need to accurately assess the speciation, sources, and fluxes of VOSe to comprehensively evaluate the biogeochemical cycle of Se in paddy systems. In this study, we developed a sensitive and accurate method for both the speciation and quantification of volatile DMSe and DMDSe from soil and rice. We captured and measured VOSe in a highhumidity and low-volatility soil–rice system from Enshi of China, known as the "Se Capital of the World," and identified another potential plant-based volatile organic Se, demonstrating the accuracy and applicability of this method for analysing VOSe in paddy systems.

Methods: In this study, we optimized the field pre-concentration method and indoor analysis method respectively. The detailed information has been shown previously^[1].

Results and Discussion: Initially, the efficacy of preconcentration for VOSe sample collection in paddy field was enhanced by optimizing the sampling apparatus, which significantly reduced water vapor by 43.5 % and increased the Se concentration by 37.7 % within 6 h sampling. Subsequently, HPLC–ICPMS analysis, after screening and optimizing chromatographic columns and mobile phases, had detected limits of 1.2 pg (picogram) and 1.4 pg for DMSe and DMDSe, respectively. For validation, VOSe was quantified in the paddy systems, and DMSe and DMDSe volatilized at 1 measured at 16.55 ± 9.94 ng/m²/h and 124.49 ± 120.34 ng/m²/h, and from rice plants at 38.38 ± 29.85 ng/m²/h and 72.54 ± 94.66 ng/m²/h, respectively. Additionally, volatile H₂Se and potential plant-based volatile organic Se species were found form rice plants.

Conclusions: This study combined *in situ* moisture removal with IEC–HPLC–ICPMS analysis to develop and validate a method for determining VOSe species in paddy soil–rice systems. The HNO₃ preconcentration method for capturing DMSe and DMDSe in the field proved to be both swift to deploy and cost-effective, thereby enhancing the analysis of VOSe in intricate environments. Furthermore, this method facilitated the first systematic analysis of VOSe species in paddy soil–rice system, revealing that plants and soil volatilized not only DMSe and DMDSe but also H_2 Se and other VOSe species. Comprehensively, this prospective work provides a practical approach for characterizing the environmental behavior of VOSe in fields with high-humidity, low-volatility and collection difficulties, contributing to a more comprehensive understanding of the biogeochemical cycle of Se in terrestrial ecosystems.

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Development of Novel, Sustainable Biobased Composite Adsorbents for the Removal of Selenium from Wastewaters

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Introduction: Various methods have been developed to remove Se oxyanions from wastewater. Among them, sorption-based technologies are particularly advantageous due to their ease of application, low energy requirements, and high removal efficiencies at low concentrations. Despite this, many developed materials still face significant limitations, including difficulties to cope with competing anions present in the wastewater, high synthesis costs, energy-intensive production processes, potential toxicity of reagents used in their fabrication, and challenges in scaling up production. These drawbacks underscore the need for further research and development to create sustainable, cost-effective, and environmentally friendly materials capable of efficiently removing Se under real-world wastewater conditions, particularly in the presence of high sulfate concentrations.

Methods: three biobased composite materials were synthesized: (1) hydroceramsite coated with layered aluminum oxyhydroxide and iron(III)-alginate (HC@LAO-IA), (2) porous ferric hydroxide cross-linked alginate beads (Fe(OH)₃-Alg), developed by incorporating sodium carbonate (Na₂CO₃) into the traditional preparation process of ferric alginate beads (Fe(III)-Alg) to generate additional internal pores within the beads, and (3) an improved porous multiphase iron-based pea starch hydrogel (IPMIPS-hydrogel), incorporating ferric iron (Fe³⁺) and nanoscale zero-valent iron (nZVI) in pea starch gel. All developed materials were evaluated as adsorbent for Se removal from both simulated and real wastewater samples in batch, as well as column experiments. Details on the production processes of the materials and the adsorption tests are extensively described by Ren^[1].

Results and Discussion: HC@LAO-IA exhibited superior removal efficiencies (≥99.9%) for both Se(IV) and Se(VI) in simulated and actual wastewater containing multiple competing anions (Cl⁻, NO₃⁻, CO₃²⁻, PO₄³⁻, and SO₄²⁻), performing especially well in high-sulfate wastewaters across a wide pH range of 3 - 10. Column experiments using real metallurgical wastewater (1.18 mg/L Se and 2577 mg/L SO₄²⁻) achieved 92% Se removal within 6 hours. HC@LAO-IA retained approximately 82% of its initial removal efficiency after four reuse cycles. However, the synthesis of this material required high-temperature, long-duration hydrothermal processes, increasing costs and environmental impacts and limiting scalability. Consequently, the next step focused on simplifying the synthesis process and developing a more cost-effective porous material, while maintaining high removal efficiency and ultimately leading to the fabrication of Fe(OH)₃-Alg beads. These beads effectively removed Se(IV) from aqueous solutions and significantly improved the removal of Se(VI) compared to conventional Fe(III)-Alg materials. Under optimal conditions in synthetic wastewaters, the beads achieved removal efficiencies of > 99.8%, reducing residual contaminant concentrations to < 0.02mg/L for Se(IV) and 0.01 mg/L for Se(VI). The beads consistently maintained high removal efficiencies and low residual concentrations across a pH range of 3-9, temperatures 10-40°C, and in the presence of competing anions, including sulfate, at concentrations up to 100 mg/L. Despite these improvements, the removal efficiency for Se, particularly Se(VI), remained suboptimal in real wastewater samples. Furthermore, notable pH decreases were observed during adsorption, increasing treatment costs due to the need for pH adjustments. These challenges prompted the development of the IPMIPS-hydrogel, which efficiently removed Se(IV) and Se(VI) from simulated wastewater, reducing their concentrations to 0.08 mg/L and 0.07 mg/L. The hydrogel demonstrated stability across a pH range of 2-10 for Se(IV) and 2-6 for Se(VI), with temperature-dependent enhancement in Se(VI) removal, likely due to increased reduction and adsorption. Even under sulfate-rich conditions - where sulfate concentrations were 1000 times higher than Se - the IPMIPS-hydrogel maintained removal efficiencies exceeding 99.2%. In sulfate-rich real wastewater, the hydrogel exhibited exceptional resistance to competitive ion interference, with SO4²⁻ concentrations surpassing Se by 2000-fold, while maintaining effluent Se levels consistently below 0.05 mg/L throughout a 10-hour column operation. Mechanistic analysis indicated that Se removal was driven by surface complexation, hydrogen bonding, electrostatic interactions, and selenate reduction mediated by Fe²⁺ and nZVI.

Conclusions: Several development steps finally led to the synthesis of an improved porous multiphase iron-based pea starch hydrogel (IPMIPS-hydrogel), achieving near-complete Se removal in high-sulfate wastewater.

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Production of Methylselenocysteine in *Saccharomyces cerevisiae* LG6 by Continuous Fermentation

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Introduction: Methylselenocysteine (SeMCys), a bioactive seleno amino acid, is a highly bioavailable form of organic selenium (Se) with significant importance in human health. Organic Se compounds, such as SeMCys, play a crucial role in antioxidant defense, immune function, and cancer prevention, making them essential for human nutrition. *Saccharomyces cerevisiae* was initially selected for Se yeast fermentation, and as an adapted mutant strain, *S. cerevisiae* LG6, was obtained through continuous fermentation. Following optimization, the organic Se production rate and intracellular Se contents were significantly enhanced. The total organic Se in the yeast reached 7500 mg/kg, the highest level reported in Se yeast production worldwide.

Method: S. cerevisiae LG6 was employed to produce SeMCys. Glucose media supplemented with varying concentrations of sodium selenite were tested to optimize medium composition and fermentation parameters. The maximal specific growth rate (μ_{max}) and the specific Se consumption rate (SSCR) were determined to be 0.162 / h and 0.47 mg / g h, respectively. Based on the μ_{max} and SSCR, the dilution factor was optimized during the continuous fermentation process, and the final production dilution rate was set at 0.11/ h.

Results and Discussion: At the optimal dilution rate of 0.11/ h, the maximal organic Se production rate reached 17.88 mg/ L h. Speciation analysis of the Se-enriched yeast showed that 58% of total Se content were in the form of SeMCys. The final concentration of SeMCys reached 5.746 mg/g (Fig. 1).



Figure 1. Effect of dilution rate on the organic selenium production rate and the biomass productivity.

Conclusion: The research findings demonstrate the potential of *S. cerevisiae* LG6 as an efficient platform for producing SeMCysenriched yeast. This product could serve as a valuable dietary supplement to address Se deficiency related human health issues.

Optimization of Selenium Speciation Detection Using Multi-Dimensional Chromatography Coupled to Atomic Fluorescence Spectrometry in Hyperaccumulator and Non-hyperaccumulator Plants

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Introduction: Traditional strong anion exchange (SAX)-ICP-MS identified predominantly selenocystine (SeCys₂) in the Se hyperaccumulator *Cardamine hupingshanensis*^[1]. Recent findings suggest selenolanthionine (SeLan), rather than SeCys₂, is the primary form of Se in the plant ^[2], highlighting the need to optimize the current analytical method for speciation analysis.

Methods: We developed a multi-dimensional chromatographic approach combining SAX-AFS and strong cation exchange (SCX)-AFS, applied to the hyperaccumulator *C. hupingshanensis* and non-hyperaccumulator wheat, with optimized sample preparation protocols, including extraction, enzymatic hydrolysis conditions, etc.

Results and Discussion: We employed our multi-dimensional chromatographic strategy, enabling the precise separation and quantification of seven Se species (Se⁴⁺, Se⁶⁺, SeLan, SeMet, SeCys₂, MeSeCys, and γ -GluMeSeCys). Our findings revealed that *C. hupingshanensis* predominantly accumulates SeLan in its water-soluble fraction, whereas wheat primarily contains enzymatically hydrolyzed SeMet. Notably, sample preparation protocols exerted a profound influence on Se speciation outcomes. Direct enzymatic hydrolysis disrupted the quantification of SeLan in *C. hupingshanensis*, while exhibiting negligible effects on wheat. Furthermore, enzymatic hydrolysis parameters, including enzyme concentration, temperature, pH, and duration, were found to critically impact Se speciation accuracy.

Conclusions: This study underscores the superiority of multi-dimensional chromatography over traditional single-column techniques for Se speciation analysis. The distinct Se distribution patterns between hyperaccumulator and non-hyperaccumulator plants highlight the necessity of tailored methodologies, paving the way for a refined understanding of Se biochemistry and its ecological implications.

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Selenium Alleviates Cotton Heat Stress by Antioxidant Metabolism Regulation

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Introduction: Heat stress is a major challenge to agriculture negatively affecting yield of crop species. High temperatures dramatically impair photosynthesis and growth of cotton plants with most critical effects occurring from 30°C to 40°C. Heat stress condition also stimulates overproduction of reactive oxygen species (ROS) disturbing cellular homeostasis. Selenium (Se) plays a crucial role in alleviating both thermal and oxidative stress by enhancing the activity of antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), and ascorbate peroxidase (APX). This study evaluated optimal rates of foliar Se application to alleviate heat stress in young cotton plants.

Methods: The experiment was carried out under growth chamber conditions with a controlled temperature at 38/25 °C day/night for 3 days. Young cotton plants (variety TMG-51) were sprayed once, with four Se (Sodium Selenete) concentrations (0, 10, 25, and 50 g/ha) two days before exposure to heat stress. After 3 days of Se treatment, cotton leaves were collected and immediately frozen into liquid nitrogen to evaluate oxidative and antioxidant metabolism measured by malondialdehyde (MDA) ^[1], hydrogen peroxide (H₂O₂) ^[2], superoxide dismutase (SOD) ^[3], ascobate peroxidate (APX) ^[4], catalase (CAT) ^[5], total soluble protein (TSP) ^[6] according to described methods.

Results and Discussion: Foliar Se application rate of 10 g/ha decreased MDA concentration by 29% and H_2O_2 concentration by 14%, indicating less oxidative stress in cotton leaves (Table 1). Total soluble protein in leaf tissue was increased by application rate of 10 g Se/ha, resulting in downregulation of specific activities of SOD, CAT and APX in order to scavenge ROS overproduction in cell tissues. These results suggest that low concentration foliar application of Se is a viable strategy to alleviate heat stress in young cotton plants, and result shows improvement in plant growth and development.

Treatments	MDA	H2O2	SOD	APX	CAT	TSP
g ha ⁻¹	nmol g ⁻¹ FW	µmol g ⁻¹ FW	U mg ⁻¹ protein	µmol min ⁻¹ mg ⁻¹ protein	µmol min ⁻¹ mg ⁻¹ protein	mg g ⁻¹ FW
0	$10.47\pm0.13~a$	$0.21\pm0.01~b$	$43.72 \pm 2.60 \text{ a}$	$39.81 \pm 2.36 \text{ d}$	342.91 ± 27.92 c	$0.57\pm0.03~b$
10	$7.40\pm0.12~\text{c}$	$0.18\pm0.01\ c$	$37.21\pm2.09\ b$	$51.04\pm3.36~\mathrm{c}$	$270.01 \pm 9.00 \ d$	$0.67\pm0.03\ a$
25	$8.08\pm0.10~b$	$0.21\pm0.01\ b$	$43.30\pm1.98~b$	$65.61 \pm 2.49 \text{ b}$	$388.50 \pm 14.47 \text{ b}$	$0.57\pm0.02\ b$
50	$10.45\pm0.18\ a$	$0.26\pm0.01\ a$	$53.88\pm4.47~c$	77.51 ± 1.78 a	552.41 ± 36.31 a	$0.46\pm0.03~\text{c}$
CV (%)	0.37	0.01	5.36	4.63	44.22	0.06

Table 1. Response of oxidative and antioxidant metabolism in cotton plants fertilized with Se under heat stress.

Conclusions: Foliar Se application rate at 10 g/ha enhanced total soluble protein and downregulated activities of antioxidant enzymes, resulting in less oxidative stress of young cotton plants grown under heat stress condition.

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An Atlas of Plant Selenium Metabolism: A Model to Help Interpret Modernomics Data in Selenium Research

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Introduction: Research on the uptake and metabolization of selenium (Se) by plants goes back almost a hundred years, with a special interest in Se-hyperaccumulator plant species. The large corpus of plant Se studies have identified many metabolic processes and gene (products) involved in plant Se physiology and various Se-hyperaccumulating species are now identified ^[1]. In recent decades, plant science has benefited from advanced techniques in genomics, transcriptomics, proteomics, metabolomics, ionomics and high-throughput phenotyping. These large omics data collection methods allow for a more comprehensive and systematic approach to studying plant biology, yet the interpretation of these whole-genome / untargeted approaches becomes more complex at the same time. Thus far, a comprehensive model of the entire plant Se metabolism has not been drawn up yet. We hypothesize that creating such model can aid interpretation of the current Se literature, as well as be extremely useful in future studies.

Methods: In our research, we have constructed a comprehensive working model for the Se physiology of plant; the SeATLAS (Fig. 1). The model is drawn studies on the following: (1) the Se physiology in plants, especially Se-hyperaccumulators; (2) the sulfur (S) metabolism of plants (since Se and S have many biochemical similarities) and (3) the (bio)chemistry of Se *in vitro* or in animals, bacteria and fungi. In addition, this systematic literature review includes a bioinformatic analysis comparing *Arabidopsis thaliana* transcriptome data from S-deficient and SeO₄-exposed plants and an analysis of published ionomic data from 1000+ plant species. Furthermore, new RNA-sequencing data from *Neptunia amplexicaulis* (a Se-hyperaccumulator species) and *Neptunia heliophila* (non-accumulator) exposed to hydroponics conditions with or without Se, are analyzed and interpreted based on the SeATLAS.

Results and Discussion: Some major takeaways from the literature analysis are: (1) most naturally grown plants contain extremely low Se levels, except for Se accumulators that can contain macronutrient-level Se concentrations; (2) Se and S metabolism have similarities but unique pathways exist respectively for both; (3) the plant species and Se conditions heavily determine whether Se is beneficial or toxic to plants; (4 exposure of plants to Se leads to a (futile) S-deficiency response; and 5) Se tolerance is achieved by avoiding two distinct types of Se-toxicity: direct chemical (oxidative) stress of inorganic species and S-replacement by selenocysteine (SeCys). *N. amplexicaulis* transcriptome analysis showed a major role for root-based methionine biosynthesis in the Se-hyperaccumulation syndrome, which is in agreement with insights from the SeATLAS that non-SeCys organic selenocompounds are most favorable for Se accumulation.

Conclusions: By compiling a consensus model of plant Se metabolism that includes 500+ metabolites and genes, the interpretation of complex omics datasets is simplified. By building an updatable model of plant Se physiology, current and future scientific data are used to their best extent.

Acknowledgement:

Reference

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Figure 3. A miniature preview of the plant Se accumulation, tolerance, localization & assimilation scheme (SeATLAS)

Toxic Effects of Selenium on Life Span of C. elegans

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Introduction: With the increase of industrial activities and agricultural fertilization, the accumulation of Se in soil has aroused extensive attention. As the most abundant metazoan in soil, nematodes play a crucial role in energy flow and material cycling, and thus it is an ideal model organism for evaluating soil Se ecotoxicology. *Caenorhabditis elegans* is a free-living nematode, abundant in soil ecosystems, with a short life cycle, easy culture, clear developmental and genetic background ^[1]. and has been widely used in ecotoxicology studies. *C. elegans* was used to explore the effects of short-term Se exposure on life span, aiming to provide a scientific basis for evaluating the potential soil ecological risk of Se.

Methods: In this study, the model organism *C. elegans* was selected as subject with *Escherichia coli* OP50 as food. The nematodes were grown in NGM dished in a biochemical incubator at 20°C, and pregnant adult nematodes were synchronized to L4 stage by lysis method ^[2] for the following experiments. Sodium selenite was added into the NGM medium to prepare the Se treatments of 0, 0.4, 3, 5, and 10 mg/kg, and each in three replicates. Fifty synchronized L4 stage larvae were selected to each petri dish. The time of inoculation was the starting time of the nematode life span, and the time when all the nematodes died was determined as the life span. The time showing half of the mortality rate was defined as the median lethal time.

Results and Discussion: The median lethal time and life span of *C. elegans* were significantly different under different concentrations of Se exposure. The median lethal time of *C. elegans* in the control group was 9.00 ± 1.73 days. Compared with the control group, the median lethal time of *C. elegans* at 0.4 mg/kg and 3.0 mg/kg Se concentrations showed an insignificant downward trend. The median death time of *C. elegans* exposed to 5.0 mg/kg and 10.0 mg/kg Se was 4.33 ± 0.58 days, which was significantly shorter than that of the control group by 51.89%. The life span of *C. elegans* in the control group was 28.33 ± 2.08 days, while no significant effect on its life span was observed on those exposed to Se concentration of 4.0 mg/kg. The life span of *C. elegans* at 3.0 mg/kg, 5.0 mg/kg and 10.0 mg/kg, was significantly shortened by 15.28%, 21.18% and 32.93%, respectively, compared with the control group. Similar results were also reported in a different study.^[3]

Conclusions: The survival rate is one of the most important parameters for assessing the ecotoxicological effects of chemicals on nematodes. This study showed that Se exposure influenced the life span of *C. elegans*, which is Se concentration-dependent. The lifespan of *C. elegans* was significantly reduced when exposed to the Se concentration of 3.0 mg/kg or higher. Therefore, with agricultural Se fertilization, it is important to determine the background value of soil Se first so that the application rate of Se fertilization can be determined accordingly to avoid having excessive Se input and to protect soil nematodes from Se toxicity.

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Altered Key Diazotrophic Community Composition Decrease Nitrogenfixing Potential in Legume Rhizospheres

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Introduction: Micronutrient deficiencies, often termed "hidden hunger", pose a major challenge to achieving the United Nations' second Sustainable Development Goal of ensuring food security and improving nutrition by 2030. It is estimated that approximately 3 billion people worldwide are affected by hidden hunger, a figure that may rise due to soil degradation (e.g. depletion of selenium (Se), iron, and calcium) and reduced atmospheric inputs (e.g. gaseous iron, zinc, Se dioxide, and dimethyl selenide) ^[1]. Among these micronutrients, Se has gained considerable attention in recent years as an essential trace element for human health. Selenium deficiency is associated with severe health outcomes, including Keshan disease and increased cancer risk. Currently, nearly 1 billion people suffer from insufficient Se intake. Low soil Se levels and limited plant uptake often result in inadequate Se concentrations in food, posing risks to human and animal health. Studies suggest that the Se content in staple foods should not fall below a critical threshold of 100 µg/kg to meet basic human requirements ^[2].

Methodology: We conducted experiments using pots filled with yellow-brown soil. The initial soil properties were as follows: soil organic carbon (SOC) 9.52 g/kg, total nitrogen (TN) 0.96 g/kg, available nitrogen (AN) 90.12 mg/kg, total phosphorus (TP) 0.60 g/kg, total potassium (TK) 17.44 g/kg, total Se 0.26 mg/kg, available Se 22.34 μ g/kg, and pH 7.17. Each pot (PVC material, 20 cm diameter, 45 cm height) was filled with 2 kg of soil, and one soybean plant (variety: Zhong Huang 13) was planted per pot. Four treatments were established, each with three replicates, resulting in a total number of 12 pots. The four treatments: Control (CK); No fertilizers applied, not irrigated and weeds were manually removed regularly. Chemical fertilization (CF): Nitrogen, phosphorus, and potassium applied at rates of 75 kg N/ha, 150 kg P₂O₅/ha, and 75 kg K₂O/ha, respectively. Selenium was added in the form of selenite at a concentration of 0.15 mg Se/kg. The treatment combining chemical fertilization with selenium addition (CFSe) was applied. Fertilizers used included urea (46% N), superphosphate (15% P₂O₅), and potassium chloride (60% K₂O). We applied analytical-grade sodium selenite (Na₂SeO₃) as source of Se. For statistics, one-way analysis of variance (ANOVA) combined with Tukey's HSD post-hoc test was employed, and the Tukey test assessed the significance of these differences at p < 0.05.

Results and Discussion: The Se addition, despite not altering α -diversity, significantly reduced *nifH* gene copies. Selenium treatment increased Cyanobacteria (approximately 1.64%) and Clostridia (approximately 3.54%) proportions. Because Clostridia are anaerobic microbes involved in sulfate/selenite reduction, their co-enrichment with Cyanobacteria may further destabilize rhizobia-legume symbiosis.

Conclusions: Our results demonstrate that CF, Se, and CFSe treatments reduce nifH gene copies and suppress α -diversity of diazotrophs. Network and Mantel analyses highlight that these treatments impair nitrogen-fixing potential by altering key microbial modules. However, nifH-based profiling cannot fully capture all nitrogen-fixing communities. Hence, future studies should employ absolute quantitative metagenomics approaches to assess comprehensively exogenous selenium's impacts on rhizosphere diazotrophs and their nitrogen-fixing capacity.

Acknowledgement: Shiyu Hu and Jianrong Zhao made equal contributions.

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Preparation, Structure Features and Antitumor Activity of a Novel Selenized Polysaccharides from *Cordyceps cicadae*

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Introduction: Selenium (Se) is both an essential trace element and plays an important role in the field of tumor therapy ^[1]. Selenopolysaccharide, as an organic Se compound, is not clearly related to its structural features and functions. The objective of this study was to investigate the toxicity of selenized polysaccharides from *Cordyceps cicadae* (SCP1) on hepatocellular carcinoma, and to elucidate the structural features of selenopolysaccharide. This study will provide a theoretical basis for the development of selenopolysaccharide in natural health foods or antitumor drugs.

Methods: The heteropolysaccharide (CP1) was extracted and purified from *Cordyceps cicadae* mycelium, and SCP1 was obtained by nitric acid-sodium selenite (NA-SS). The effects of CP1 and SCP1 on the toxicity, apoptosis and cell cycle of hepatocellular carcinoma HepG2 and Huh7 were investigated by microplate Reader, fluorescence microscopy and flow cytometry. Additionally, the structures of CP1 and SCP1 were analyzed using high-performance liquid chromatography (HPLC), gas chromatography–mass spectrometry (GC–MS), Fourier-transform infrared spectroscopy (FTIR) and nuclear magnetic resonance spectroscopy (NMR).

Results and Discussion: SCP1 had a significantly stronger inhibitory effect on HepG2 and Huh7 than CP1, with maximum inhibition rates of 63.5% and 69.1%, respectively. After SCP1 induction, the proportion of HepG2 and Huh7 cells in S phase increased from 27.4% to 49.1% and 11.2% to 22.3%, respectively. These results suggest that SCP1 inhibits cell proliferation by blocking the normal progression of hepatocellular carcinoma cells in the S phase. Apoptosis analysis revealed a decrease in the proportion of viable HepG2 cells from 96.3% to 43.4% and an increase in the corresponding apoptosis rate from 3.1% to 51.0%. Similarly, the proportion of viable Huh-7 cells decreased from 97.7% to 52.2%, and the corresponding apoptosis rate increased from 0.9% to 44.7%. Additionally, the expression of the DNA repair enzyme PARP and the apoptosis-inhibiting protein Bcl-2 decreased, while the expression of the pro-apoptotic protein Bax and cleaved caspase-3 increased. These results demonstrate that selenopolysaccharide SCP1 can induce apoptosis in hepatocellular carcinoma cells. The physicochemical analysis revealed a 5.6% reduction in total sugar content and a 2.8% increase in uronic acid content in SCP1, suggesting nitric acid oxidized some sugar units to uronic acid after selenization. The molecular weight of SCP1 significantly increased from 28.9 kDa to 123.2 kDa. indicating Se reacted with hydroxyl groups, forming selenite bonds and enhancing structural complexity ^[2]. However, the monosaccharide composition (galactose, glucose, mannose) remained constant. NMR and methylation analyses confirmed the primary structure of CP1 is α -Galp -(1 \rightarrow [2)- α -Manp -(1]₅ \rightarrow [2)- α -Galp-(1]₂ \rightarrow 2,6)- α -Manp -(1 \rightarrow , while FR-IR spectroscopy identified new peaks at 1078 cm⁻¹ (Se=O asymmetric stretching) and 852 cm⁻¹ (C-O-Se bending), confirming Se was successfully bound to the CP1 chain.

Conclusions: In this study, a heteropolysaccharide (CP1) was extracted and purified from *Cordyceps cicadae* and characterized for its monosaccharide composition, molecular weight, and glycosidic bond linkage. After selenization, Se ester groups were successfully introduced into the polysaccharide chain of CP1 to obtain SCP1. In addition, we demonstrated that SCP1 could significantly inhibit the proliferation of hepatocellular carcinoma cells by inducing apoptosis and affecting cell cycle progression. Nonetheless, further studies are necessary to elucidate the application and the anti-tumour molecular mechanisms of SCP1.

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Bioreduction and Volatilization of Selenium by Delftia acidovorans R25

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Introduction: Selenium (Se) is an essential trace element for the life activities of humans and animals, but its safety threshold is low, and excessive environmental Se can pose toxicity risks. Environmental microorganisms play a key role in the biogeochemical cycling and transformation of Se, effectively reducing ecological toxicity by reducing highly toxic selenate [Se(VI)] or selenite [Se(IV)] into low-toxic Se nanoparticles (SeNPs) and methyl Se. However, the role of Se-reducing bacteria in the global Se cycle and their potential in high Se environmental remediation require further investigation.

Methods: *Delftia acidovorans* R25 isolated from seleniferous soil was used as the material, and the Se tolerance range of strain R25 was determined by setting the concentration gradient experiment. The time rule of the synthesis of elemental Se or volatile Se by strain R25 was studied by time kinetics experiment. The elemental Se was identified by TEM-EDX and the volatile Se was identified by GC-MS. The effect of nitrate, ammonium salt and phosphate on the synthesis of volatile Se by strain R25 was studied by single factor experiment. The removal and recovery of Se by strain R25 was simulated in the fermenter.

Results and Discussion: R25 can metabolize Se(IV) to synthesize red nano Se, and mainly synthesize elemental Se in the log phase of growth under high Se conditions (>1 mM). Strain R25 can metabolize Se(IV) and SeNPs to synthesize volatile Se. The volatile Se products include DMDSe, DMSeO2 and triselenothane, among which DMDSe has the highest abundance. And MeSeCys is an intermediate product of volatile Se synthesis. R25 first reduces Se(IV) to SeNPs, and then the step of synthesizing volatile Se from SeNPs is the rate limiting step of Se volatilization. The phenomenon that this elemental Se is an intermediate product of the strain using Se(IV) to synthesize volatile Se was also observed in *Corynebacterium sp.*^[1], *E. cloacae* SLD1a-1 ^[2], *P. stutzeri* NT-I ^[3] and *M. trichosporium* OB3b ^[4]. Strain R25 mainly synthesized volatile Se at the stationary phase under low Se conditions (<0.5 mM). Selenium volatilization of Strain R25 increased under the conditions of phosphate 0.25 mM, pH 9 or temperature 31°C. However, nitrate (<12 mM) and ammonium (<2 mM) inhibited Se volatilization of methylselenic acid.

Conclusions: Strain R25 mainly synthesized elemental Se in the log phase of growth and volatile Se in the stationary phase of growth. The threshold of the detoxification pathway of elemental Se in strain R25 was higher than that of volatile Se. The volatile Se products synthesized by strain R25 include mainly DMDSe. In the process of synthesizing volatile Se, SeNPs and MeSeCys are intermediate products. The step of synthesizing volatile Se from SeNPs is the rate limiting step of R25 Se volatilization. Phosphate (<0.25 mm) promoted the Se volatilization of R25, and strain R25 simulated the removal of Se (IV) and SeNPs from Se containing water and recovered them in the form of methylselenic acid.

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Plant and Soil Microbial Interaction for the Enhancement of Selenium Volatilization

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Introduction: Biogenic volatilization is an important ecological process of selenium (Se) biogeochemistry ^[1]. Earlier studies showed that rabbitfoot grass (*Polypogon monspeliensis* (L.) Desf.) grown in constructed Se treatment wetlands is a particularly efficient Se volatilizer ^[2,3]. The treatment wetland with rabbitfoot grass can remove 35-49% of total monthly Se mass input to the atmosphere in the San Joaquin Valley, California ^[3]. This study aimed at quantifying the potential effects of plant and soil microbial interaction on Se volatilization. The specific research objectives were to (1) determine effects of manipulating soil conditions on Se volatilization in soil-plant systems, (2) identify soil bacteria associated with rabbitfoot grass showing higher capacity of volatilizing Se and (3) demonstrate the enhancement of Se volatilization in a soil-Indian mustard (*Brassica juncea* L.) system through inoculation of the soil with the identified best Se-volatilizing bacterial strain.

Methods: The partitioning of Se volatilization between the soil-root compartment and shoots in the soil-plant system was determined, with manipulation of the soil conditions. Soil bacteria were isolated from topsoil and rhizosphere soils of rabbitfoot grass, and the bacterial colonies were characterized via PCR-DGGE and DGGE band analysis prior to their identification using 16S rDNA sequencing technique. The rates of Se volatilization were determined using a volatilization chamber system ^[4].

Results and Discussion: The soil-root compartment is the major source of biogenic volatilization in the soil vegetated with nonhyperaccumulator species. Manipulating soil conditions such as soil water content and application of plant litter or manure can significantly increase the rates of Se volatilization. *Bacillus cereus* produced over 500-fold more volatile Se in a culture medium treated with 15 µg Se/mL (equal mixture of SeO₄²⁻, SeO₃²⁻ and selenomethionine) than any of the other eight identified bacterial strains. Inoculation of Indian mustard vegetated soil with the best Se volatilizing bacterial strain *B. cereus* resulted in a significant (p < 0.05) increase in Se volatilization during a 7-day time period, compared to the soil-plant system without inoculation of *B. cereus*. Thus, inoculation of the soil with *B. cereus* substantially enhanced Se removal via biogenic volatilization in the soil-Indian mustard system^[4].

Conclusions: Biogenic Se volatilization in soil-plant systems can be significantly manipulated under different soil conditions. Inoculation of rabbitfoot grass associated *B. cereus* in the soil substantially enhancing Se volatilization in the soil-Indian mustard system, demonstrating the importance of plant and soil microbial interaction for the Se volatilization process.

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