

Review Article

MITIGATION OF ARSENIC CONTAMINATION THROUGH BIOTECHNOLOGICAL APPROACHES IN RICE

Abstract

Arsenic poisoning negatively impacts plants, soil, water, and human health, posing a serious threat to sustainable agriculture. Pesticides, fertilizers, and industrial processes that contain arsenic are widely used, which leads to soil contamination and reduces soil fertility and productivity. The drinking of groundwater contaminated with arsenic affects around 300 million people globally. Prolonged exposure to arsenic has been linked to a number of health concerns, including as cancer, developmental abnormalities, and skin sores. Concerns have been raised over the possible health effects of arsenic, which is mostly exposed to consumers through the consumption of contaminated food. Rice is one of the most impacted crops from arsenic-polluted irrigation water and soil, making it a common vehicle for dietary intake of arsenic. In present review, the challenges related to arsenic toxicity and its possible solutions in rice have been discussed.

Keywords: Arsenic, Brassinosteroid (BR), Salicylic acid (SA), QTL, CRISPR-Cas9

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Introduction

Arsenic toxicity poses a significant threat to sustainable agriculture by adversely affecting soil, water, plants, and human health (Prasad et al., 2020). The widespread use of arsenic-containing pesticides, fertilizers, and industrial processes contributes to the contamination of soil, compromising its fertility and overall productivity. Moreover, arsenic can leach into groundwater, further contaminating water sources used for irrigation and exacerbating the risk of crop uptake. Crops such as rice, leafy vegetables, and root crops are particularly susceptible to arsenic accumulation, presenting a direct threat to food safety. The consumption of arsenic-contaminated crops can lead to various health issues, including skin lesions, cancer, and neurological problems, underscoring the interconnectedness of environmental and human well-being. Additionally, the economic sustainability of agriculture is at stake due to potential trade restrictions on arsenic-contaminated agricultural exports.

Arsenic (As), a metalloid, ranked first in a list of 20 major hazardous substances by the Agency for Toxic Substances and Disease Registry and United States Environmental Protection Agency (Roy and Saha., 2002). Due to its high toxicity, it is also known as 'king of poison' (Shaji et al., 2021). Recent study showed that nearly 108 countries are affected by arsenic contamination in groundwater (with concentration beyond maximum permissible limit of 10 ppb recommended by the World Health Organization (WHO) (Shaji et al., 2021). The highest among these are from Asia (32) and Europe (31), followed by regions like Africa (20), North America (11), South America (9) and Australia (4). More than 300 million people worldwide are affected by the consumption of arsenic contaminated groundwater (Kumar et al., 2021). The South and Southeast Asian Belt is considered as the most arsenic polluted areas including India, Bangladesh, Nepal, Vietnam and China (McArthur, 2019; Shaji et al., 2021). In India, 20 states including West Bengal, Jharkhand, Bihar, Uttar Pradesh, Assam, and 4 Union Territories have so far been affected by arsenic contamination in groundwater (Shaji et al., 2021). Bihar has shown an upsurge in cases affected by arsenic poisoning (Kumar et al., 2021).

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The adverse human health effects resulting from chronic As consumption include increased risk of cancers, hyperkeratosis, birth defects, cardiovascular disease, neurotoxicity, and diabetes (Phan et al., 2010; Flanagan et al., 2012; Kumar et al., 2021). Skin lesions are a common sign of arsenic poisoning and indicate prolonged exposure. Even more concerning, the International Agency for Research on Cancer has categorized arsenic as a Group 1 human carcinogen, connecting it to malignancies of the skin, lungs, bladder, and other organs. Prolonged exposure to arsenic has also been linked to developmental defects, cardiovascular disorders, and neurological impairments. The negative health effects are more severe in areas where arsenic poisoning is common, which emphasizes how urgently this public health issue has to be addressed.

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Use of contaminated groundwater for crop irrigation may result in the accumulation of As in agricultural soils, eventually resulting in decreased crop yields and impaired human health (Brammer and Ravenscroft, 2009; Kumar et al., 2021). The threat of As to humans is usually exacerbated in countries that have high population densities, use groundwater as their primary drinking water source, and rely heavily on large quantities of irrigation for agriculture, such as those within South and Southeast Asia. However, contamination of agricultural soils from arsenic in groundwater is truly a global problem, with geographically dispersed countries (e.g., Mexico, Chile, Argentina, Greece, and the USA) experiencing

varying degrees of soil contamination (Smedley and Kinniburgh, 2002; Sahoo and Kim, 2013; Shaji et al., 2021).

Rice (*Oryza sativa* L.) arsenic contamination is a pressing concern in global food safety, as rice is known to accumulate higher levels of arsenic compared to other staple crops (Upadhyay et al., 2020; He et al., 2021). The rice plant effectively absorbs arsenic from the soil, especially inorganic arsenic, which results in higher amounts in the grain. Eating rice exposes one to arsenic over time, which has been related to cancer, developmental disorders, and skin sores, among other health problems. The natural ability of the plant to take up arsenic, in combination with many agronomic, geochemical, and hydrological factors, leads to As accumulation in rice (Zhao et al., 2010). The management of the rice plant frequently consists of controlled flooding in order to eliminate competition from weeds, reduce herbicide needs, and increase yields (IRRI 2015). To create flooded conditions, large quantities of irrigation water may be used in both dry and wet seasons, depending on the ability of a rice paddy to receive adequate natural saturation. Flooding changes natural soil hydrodynamics, results in high loadings of arsenic in soil, if contaminated groundwater is utilized for irrigation.

South and Southeast Asia are the areas of greatest concern for As accumulation in rice-paddy soils following irrigation with contaminated groundwater. Throughout the densely populated river basins that drain the Himalayas, rice is a staple crop, and natural arsenic contamination of groundwater has been used extensively for irrigation of dry-season rice for several decades (Fendorf et al., 2010; Chakraborty et al., 2015). The Bengal Delta of Bangladesh and India, one of the most severely impacted and extensively studied areas, As concentrations in irrigation water applied to rice-paddy fields frequently exceed 50 µg/L (Williams et al., 2006; Khan et al., 2010; Chakraborty et al., 2015), reaching as high as 1800 µg/L (Alam et al., 2003). As a consequence, soil arsenic concentrations in polluted fields can reach up to 95 mg/kg, and rice grains may have As concentrations of up to 1835 µg/kg (Khan et al., 2010). Rice has the ability to accumulate arsenic, amassing concentrations ten times higher than other cereals such as wheat (Karagas et al., 2019). High concentrations of iAs have also been found in rice-based products, including those consumed by infants and young children (Davis et al., 2017). Rice constitutes approximately 66 % of the caloric intake in the region and up to 50 % of the daily arsenic consumption for some people (Liao et al., 2010). Rice is a major contributor to arsenic intake in humans (Williams et al. 2005; Meharg et al. 2009; Gilbert-Diamond et al. 2011), because rice is mainly cultivated in anaerobic paddy soil, where arsenite [As(III)] is more available (Takahashi et al.2004).

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The identification and **introgression of QTLs** related to arsenic are among the preferred strategies for arsenic mitigation in crop plants. The genome wide association (GWA) mapping for arsenic in shoots and grains of rice grown under continually flooded (CF) and alternate wetting and drying (AWD) has identified a large number of 74 individual QTLs for arsenic, with six QTLs showing stability across years and/or water treatments (Norton et al., 2019). Recently, a machine-learning Bayesian network approach along with high-resolution GWAS were performed to identify QTL for grain-arsenic and straight head disorder **(StHD)** resistance (Pinson et al., 2021). For each As-associated trait, several QTL (ranging from 9 to 33) were identified (Pinson et al., 2021). Using a doubled haploid population created by anther culture of F1 plants from a hybrid between a Japonica cultivar CJ06 and an Indica cultivar TN1, the quantitative trait loci (QTLs) linked to arsenic accumulation in rice were mapped (Zhang et al., 2008). The map revealed four QTLs for the concentrations of arsenic. At the seedling stage, a single QTL for As concentrations in roots was found on chromosome 3, and a single QTL for As concentrations in shoots was mapped on chromosome 2 with a 24.4% phenotypic variation. Two QTLs with 26.3 and 35.2% phenotypic variance, respectively, were discovered on chromosomes 6 and 8 at maturity for As concentrations in grains (Zhang et al., 2008).

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Phytohormones play a key role in mediating cellular responses to various stress stimuli (Vos et al., 2015; Ku et al., 2018). The protective role of salicylic acid (SA) in plants against abiotic and biotic stress are well documented (Prasad et al., 2009; Noriega et al., 2012; Kumari et al., 2022; Sahni et al., 2021; Sahni and Prasad 2022). Recent studies showed the modulation of genes related to heavy metal stress in plants (Noriega et al., 2012; Singh et al., 2015). The exogenous application of SA was found effective in mitigating arsenate (AsV) toxicity in rice by lowering translocation to the shoots (Singh et al., 2015). The amount of As in the shoot was positively connected with the expression of *OsLsi2*, an efflux transporter that moves As from the root to the shoot as arsenite (AsIII) (Singh et al., 2015).

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Brassinosteroid (BR), a plant steroidal hormone, is one of the recognized phytohormones that is important for plant growth and development. It also modulates stress responses and can shield plants from heavy metal poisoning (Sahni et al., 2016; Xu et al., 2018). According to Xu et al. (2018), there is preliminary evidence that treating rice plants cultivated in hydroponic culture with BRs prevents arsenic accumulation. Recently, we have unequivocally demonstrated that the application of Brassinosteroid, **BR mediated suppression of *OsLsi1* and *OsLsi2* had a beneficial effect in reducing the arsenic accumulation in straw and grain of**

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rice plant (Chaudhary et al., 2023). Further, BRs lowered (by up to 50%) the uptake of arsenic in plants grown in naturally arsenic-contaminated soil (Chaudhary et al., 2023). Overall, BR therapy had a net positive effect on rice plant arsenic absorption, highlighting BR's higher-level modulation of this feature in plants' hierarchical response system. A solid foundation for creating BR-based arsenic mitigation techniques in rice is provided by gene expression analysis and the decreased absorption of arsenic in rice seeds and straw in EBR-treated rice plants compared to mock-treated rice plants.

In recent years, Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) and CRISPR-associated protein 9 (Cas9) has become the method of choice for genome editing in crop plants including rice (Zafar et al., 2020; Kumari et al., 2022). The well-developed rice transformation protocol facilitates faster adoption of gene editing technology in rice (Prasad et al., 2008, 2009; 2016). The emerging reports in targeting genes associated with arsenic uptake, translocation and detoxification of arsenic in rice has paved the way of developing low arsenic mitigation in rice using gene editing (Chen et al., 2017; Fiaz et al., 2019; Chaudhary et al., 2023). The role of *OsLsi1* and *OsLsi2* in arsenic translocation in rice plants are well described (Chen et al., 2017; Singh et al., 2015; Chaudhary et al., 2023). The editing of *OsLsi1* and *OsLsi2* genes in rice might be helpful in reducing the arsenic accumulation in straw and grain of rice plant without yield penalties.

Conclusion

Addressing arsenic toxicity in agriculture requires a comprehensive approach, including the development of arsenic-resistant crop varieties and adopting new biotechnological tools. The management practices and use of phytohormones have the potential to significantly lower the arsenic uptake in rice. Further, the plant breeding approaches amalgamated with genes will pave the way for developing arsenic-free rice.

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Comment [AP10]: It is better for terms and abbreviations, you make it in the form of a table so that it is easy to understand.

Comment [AP11]: You should explain the use of new biotechnology tools that you are referring to.

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