

# Salt Tolerance and Cultivation Challenges of Jojoba (*Simmondsia chinensis*): A Review

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**Abstract:** Jojoba is considered to be salt tolerant, but little is known about the extent of its tolerance and the relative influence on ion uptake and ethylene production and other factors are affecting the mode of actions of jojoba salt tolerant. According to many research has done before including our article about the glyoxalase I gene from jojoba which exhibits moderate tolerance to soil salinity, withstanding salinity levels up to about 8-10 ds/m without significant adverse effects on growth and productivity. However, higher salinity levels can lead to reduced growth rates and lower yields. A main focus in plant biology has been the response of plants to salt stress and the mechanisms of salt tolerance. Abiotic stress significantly increases the level of Methylglyoxal (MG) and Glyoxalase I (Gly I) gene activity, the first of two enzymes involved in cellular detoxification. Abiotic stress causes large losses in agricultural production globally. The glyoxalase I sequences from the jojoba plant were quickly and easily identified as salt tolerance genes. The glyoxalase I gene was isolated, amplified by PCR using gene-specific primers, and sequenced from the jojoba plant. It was then compared to glyoxalase I sequences from organisms. The expression profiles of salt-related genes in the leaf transcriptome of Jojoba (*Simmondsia chinensis*) were analyzed to decipher the molecular mechanisms underlying salt stress tolerance in these species. RNA-Seq data revealed numerous differentially expressed genes most of which were upregulated under salt (NaCl) stress conditions. In a previous studies jojoba seeds were sown on germination media treated with varying concentrations of seawater, specifically 2000, 3000, and 5000 ppm of final solute concentration. Salt-tolerant jojoba cultivars were selected and established using *in vitro* culture. After selection on germination media with different solute levels, shoot tips from the seedlings were grown on multiple-shoot induction media.

**Keywords:** Jojoba Plant, Abiotic Stress, Physiological Responses, Methylglyoxal, Glyoxalase I

## Introduction

The woody, perennial, dioecious jojoba (*Simmondsia chinensis* L.), which belongs to the Simmondsiaceae family, is commonly farmed in semi-arid climates. It is also known by names such as deer nut, coffee berry, wild hazel, and oat nut. The jojoba plant is native to the hills of Arizona, southern California, and northwest Mexico. Over the past thirty years, jojoba cultivation has expanded to various countries, including Saudi Arabia, Egypt, Tunisia, Mexico, Chile, and Argentina (Al-Obaidi et al., 2017). Today, over 18,500 hectares of jojoba crops are grown worldwide. Jojoba seeds contain approximately 65% oil, characterized by a light golden liquid colour, as shown in Figure (1) by Hussain et al. (2011). One of the most detrimental environmental

stresses is salinity, which reduces crop development and yield in rainfed and semi-rainfed regions across the globe in a variety of soil types. The negative salinity's detrimental impact on plant growth is caused by the absence of plants' access to water, which results in osmotic stress and hinders beneficial functions (Hasanuzzaman and Fujita, 2022).

Jojoba is cultivated in several countries and is considered a promising oil crop for various reasons. The waxy seed oil has been used to cure wounds, relieve headaches, and reduce irritation in the neck due to its anti-inflammatory, antibacterial, and antifungal qualities. In addition to its medicinal qualities, jojoba seed oil is extensively used in the cosmetic, bioenergy, and pharmaceutical sectors. What sets jojoba oil apart from

other plant-based oils is its unique chemical composition. In addition, jojoba leaves also contain flavonoids, which are antioxidants with potential applications in treating cancer, inflammation, and asthma (Abdel-Mageed et al., 2014). Farmers cultivating jojoba need to develop effective biotechnology platforms to improve cultivation practices and enhance production cycles. Over the past 20 years, numerous attempts have been made to cultivate jojoba in vitro using various molecular biology methods. However, significant efforts are still required to achieve results that effectively address cultivation challenges (Al-Obaidi et al., 2017).



**Fig. 1:** Jojoba plant cultivated in arid lands

## Salt Tolerance in Jojoba

### *Salinity Levels*

Salinity is one of the most damaging environmental stresses that significantly reduce agricultural output and quality worldwide. Due to both anthropogenic and natural processes, more than 20% of the world's arable land suffers from the hardship of salt stress, and the number of these regions is steadily rising (Arora, 2019). However, because of the rising need for irrigation water requirements over the past 20 years, this difficulty has gotten much worse in dry and semi-arid countries (Acosta-Motos et al., 2017).



**Fig. 2:** Cultivation of jojoba plant in high salt lands

The global environmental issue of water and soil salinization is particularly problematic in arid and semi-arid regions, where over-irrigation exacerbates the situation (Parida and Das, 2005). The physiological effects of salinity include growth, oxidative stress, photosynthesis, germination, and water imbalance; these effects can ultimately result in plant death or a reduction in crop yield, biomass, or harvest index. Current irrigation techniques and climate change are likely to worsen this issue. To mitigate the harmful effects of salinity on plants, a variety of tactics have been employed, including the implementation of appropriate farm management techniques and the selection or cultivation of salt-tolerant plant species (Majeed et al., 2020).

## Mechanisms of Tolerance

### *Physiological Adaptations*

Plants under salt stress exhibit a range of physiological and metabolic alterations, including delayed seed germination behaviour, suppression of other biosynthetic processes, photosynthetic inhibition, and reduced growth. Crops respond differently to salinity: Halophytes thrive and reproduce well in saline environments, while glycophytes generally experience stunted growth and reduced total production. As a result, the accumulation of  $\text{Na}^+$  and  $\text{Cl}^-$  in the leaf has a slower impact at higher osmotic pressures in the root-soil interface. This results in decreased growth of the shoots together with decreased expansion of the leaves and suppression of the establishment of lateral buds. In response to salt stress, plant cells adjust to low soil water potential by redistributing ions and accumulating suitable solutes (Munns, 2011). However, jojoba plants demonstrate a notable tolerance to salt Figure (2).

### *Morphological Adaptations*

In response to environmental stressors, plants typically develop secondary metabolites as a defence mechanism. The diversity of plant-specific metabolic products varies depending on the type of stress Jojoba plants have developed adaptive traits to cope with salinity: Their root system can explore deeper soil layers to access less saline water, and their leaves have thick cuticles to minimize water loss and reduce salt uptake (Hasanuzzaman and Fujita, 2022).

**Ion Regulation:** Jojoba regulates the uptake and distribution of sodium ( $\text{Na}^+$ ) and chloride ( $\text{Cl}^-$ ) ions within its tissues to avoid toxic concentrations, often sequestering these ions in older leaves or vacuoles.

## Impact of Salinity

### *Germination*

High salinity can impact seed germination rates, with studies indicating that salinity levels above 4-6 ds/m can

significantly reduce germination. The length of the roots and rate of rooting of jojoba plantlets grown at different salinities did not differ significantly, according to Roussos et al. (2005). Similarly, their study revealed comparable outcomes for seedlings raised in a range of salinity conditions, demonstrating no discernible difference in the quantity and length among seedlings that germinated and grew in different salinity environments Figure (3).



**Fig. 3:** Germination of jojoba plant in high salinity lands

### Growth

Elevated salinity levels can lead to reduced plant height, leaf area, and biomass, these effects being more pronounced during the early stages of growth. Under *in vitro* salt stress, jojoba undergoes anatomical changes, including increased thickness of the mesophyll in leaves and enlargement of the cortex, pith, and xylem vessel diameters in stems. While cell density decreases in leaves and stems, it increases in roots. These anatomical alterations, influenced by *Azospirillum Brazilense*, may contribute to enhanced salinity tolerance in inoculated jojoba plants, helping them withstand the damaging effects of saline stress (Gonzalez et al., 2021).

Where Different salinity levels (0, 30, or 60 mM NaCl) were applied to wheat plants during seeding, and three exogenous aspartic acid (Asp) foliar spray application amounts (0, 0.4, 0.6, or 0.8 millimolar), the study's findings showed that salt stress inhibited growth. Characteristics such as shoot biomass, leaf area, and shoot length, in addition to Endogenous indole acetic acid and pigments involved in photosynthesis. NaCl strain decreased the amount of beta carotene, flavonoids, and carbs overall lycopene (Sadak et al., 2022).

Despite certain negative effects, jojoba and *Azospirillum brasilense* can withstand high NaCl concentrations (Gonzalez et al., 2015) determine whether inoculation with *A. brasilense* mitigated the adverse effects of salt on a number of jojoba plant clonal lines. Shoots were grown in an auxin-free medium with 0, 40, 80, 120, or 160 mM NaCl and infected or not with 107 cfu of *A. brasilense* Cd or Az39 for 45 days after being

induced in half-strength Murashige–Skoog media with 24.6  $\mu$ M or 49.2  $\mu$ M indole-3-butyric acid for 6 days. The root index, shoot and root biometric parameters, and rooting percentage were ascertained. On the other hand, some crops like barley are cultivated under salt stress (4 g NaCl/L).

### Yield

Salinity stress can reduce the number of flowers and seeds produced, directly impacting yield. Oil content and quality may also be affected under high salinity conditions.

### Breeding and Selection

Ongoing research aims to develop more salt-tolerant jojoba cultivars through breeding and selection. Identifying and propagating genotypes with naturally higher tolerance can enhance the sustainability of jojoba cultivation in saline environments. Jojoba is typically propagated from seeds or cuttings. A significant challenge with seed propagation is that jojoba is a dioecious plant, making it difficult to determine the plant's sex until it flowers, which can take 3-4 years. Vegetative propagation can be achieved by rooting semi-hardwood cuttings or micropropagation. *In vitro* culture allows the propagation of plants of known sex and high productivity. Many protocols have been developed using different growth regulators and culture conditions to facilitate this process (Bashir et al., 2007; Llorente and Apóstolo, 1998; Mills et al., 1997; Tyagi and Prakash, 2004).

### Genetic Improvement

Breeding programs aim to develop jojoba varieties with enhanced salt tolerance, higher yield, and better oil quality. Genetic studies have identified traits associated with stress tolerance that can be targeted in breeding efforts. Biotechnological approaches, including genetic engineering and marker-assisted selection, hold promise for accelerating the development of improved jojoba cultivars. Yadav *et al.* (2005) investigated that Glyoxalase I uses the cytotoxic by-product Methylglyoxal (MG), which is mostly made from triose phosphates, as a substrate. The estimation of MG levels in plants, which has not been previously reported, is the subject of this study. In response to salinity, drought, and cold stress, we demonstrate that MG concentrations in different plant species vary from 30 to 75 microns and increase by 2 to 6 times. Transgenic tobacco that lacked glyoxalase I exhibited increased MG buildup, which prevented seeds from germinating, when compared to the untransformed plants, MG levels in the glyoxalase I overexpressing transgenic tobacco did not rise in response to stress; nevertheless, both the untransformed and transgenic plants experienced a drop in MG levels when exogenous GSH was added. When GSH was

applied exogenously, MG levels in WT were reduced to 50%, while in transgenic plants, they were shown to have decreased five times. These results show that glyoxalase I and GSH concentration play a significant role in preserving MG levels in plants under both abiotic and normal stressors. Consequently, the glyoxalase I gene was extracted from the jojoba plant, amplified by PCR using gene-specific primers, and sequenced. It was then compared to other glyoxalase I sequences in other plants, such as *Brassica napus*, ID: KT720495.1, and *Brassica juncea*. IDs: Y13239.1, DQ989209.2 for *Arachis hypogaea*, and AAL84986 for *A. thaliana* L. (Mohasseb et al., 2020). Additionally, genes linked to cell wall remodelling, lowering chlorophyll content, building up reduced sodium ( $\text{Na}^+$ ) and chloride ( $\text{Cl}^-$ ) concentrations, and elevated lignin levels serve to help a plant remain intact while it is under salt stress (Alghamdi et al., 2021).

Jojoba's moderate salt tolerance makes it a viable crop in semi-arid and arid regions where soil salinity poses a challenge. However, effective management of salinity through appropriate agricultural practices and the development of more tolerant cultivars is essential for maximizing the crop's potential and ensuring sustainable production. Due to these traits, jojoba is an attractive option for arid and semi-arid regions, providing economic benefits while requiring minimal water resources. Its ability to thrive in harsh conditions makes it a sustainable and resilient choice for cultivation in areas prone to salinity. These studies collectively demonstrate the potential of cultivating jojoba in saline environments using both traditional and biotechnological approaches. By employing in vitro selection, harnessing beneficial microbial associations, and understanding the genetic basis of salt tolerance, researchers are advancing the development of more resilient jojoba crops suitable for harsh climates.

#### Cultivation of Jojoba Under High Salinity

To boost agricultural output and feed the world's expanding population, it is imperative to conduct the required research on salt stress and soil pollution prevention. An increasing number of people are interested in environmentally sustainable salt tolerance strategies as the world's hunger crisis and salinity stress worsen. There are numerous more methods for cultivating crops that can withstand salt besides irrigating with fresh water. It was effective to choose jojoba plants that could tolerate saline levels of 5000 ppm. However, plantlets derived from the 5000 ppm salt level showed less development and multiplication under 2000 and 3000 ppm salinity compared to those from lower salt levels. The seedlings selected at 2000 and 3000 ppm successfully developed in the multiple-shoot induction and rooting media. The plantlets' high growth and multiplication were not supported by their

chlorophyll and carotenoid content, even though, as shown in, the plantlets at 2000 ppm showed higher values in all growth and multiplication parameters during the multiple-shoot induction stage Figure (4).



Fig. 4: Tissue culture of jojoba in various concentrations of NaCl

Conversely, plantlets from 3000 ppm salinity demonstrated high concordance levels of chlorophyll A and carotenoid content, although they performed slightly worse than those from 2000 ppm salinity throughout the multiple-shoot induction stage. Only plantlets that were initially selected at salinities of 3000 ppm and control were able to root in rooting media that had salinities matching those levels. Plantlets picked at varying salt levels have not shown the same development and yield metrics as plantlets coming from the 2000 ppm, demonstrated enhanced  $\text{Na}^+$  absorption without affecting the uptake of  $\text{K}^+$  and  $\text{Ca}^{2+}$  as shown in Figure (5) (Alyousif et al., 2023). On the other hand, Treatments with cysteine were helpful in reducing the negative effects of salt stress on the soybean plant. The most successful therapy for improving the soybean plant's ability to withstand salinity was forty milligrams of cysteine per litre (Sadak et al., 2020).



Fig. 5: Rooting of jojoba plant

Jojoba's dioecious nature leads to significant variability at the clonal level, affecting both morphological traits and yield characteristics. Research highlights the importance of selecting desirable male and female plants and their most compatible combinations to improve yields. For the continued development of jojoba as a commercial crop, it is crucial to identify the factors contributing to the considerable variability among genotypes. Employing a multidisciplinary approach, incorporating molecular genetics, functional genomics, plant reproductive biology, biochemistry and agronomy, could provide valuable insights into identifying genotypes with consistent yields across various production systems. The jojoba industry must address the challenge of enhancing productivity and product quality to advance its commercial viability (Bala and Laura, 2015).

## Conclusion

Jojoba's moderate salt tolerance and adaptability to arid environments make it a valuable crop for regions with saline soils and limited water resources. Implementing effective soil and water management practices, coupled with ongoing genetic improvement, can mitigate cultivation challenges and enhance the productivity and resilience of jojoba plantations. Research and innovation in agronomic practices and biotechnological interventions are crucial for the sustainable development of jojoba as a commercial crop. Moreover, the mechanisms of salt tolerance and the response of plants to salt stress have been the focus of plant biology. Methylglyoxal (MG) and Glyoxalase I (Gly I) gene activity, the first of two enzymes involved in cellular detoxification, are both markedly elevated in response to abiotic stress. Globally, abiotic stress results in significant losses in agricultural productivity. It was simple and rapid to identify the jojoba plant's glyoxalase I sequence as salt tolerance genes. From the jojoba plant, the glyoxalase I gene was extracted, amplified by PCR using primers unique to the gene, and sequenced. After that, it was contrasted with organisms' glyoxalase I sequences.

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## Ethics

This study was approved by the university human research ethics committee and all procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

## Conflicts of Interest

The author declare no conflict of interest. The funders had no role in the study's design, in the collection, analyses, or interpretation of data, in the writing of the manuscript, or in the decision to publish the results.

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