Advancing Soybean Resilience: The Role of Induced Polyploidy to Abiotic Stress Tolerance

Abstract

Abiotic stress is one of the major constraints affecting the productivity of soybeans. It reduces germination ability, seedling growth, and development of the reproductive parts. Physiological traits, such as membrane and enzyme structure and functionality, are also negatively impacted due to the increased levels of reactive oxygen species (ROS). Polyploidy is known for creating diversification among plants, playing a key role in enhancing tolerance capacity against abiotic stress. These variations help plants survive under harsh conditions by modifying several morpho-physiological, molecular, and biochemical traits. However, polyploidy's role in enhancing tolerance to abiotic stress has been less explored in leguminous crops, particularly in soybeans. Additionally, no proper in-vitro or in-vivo techniques have been successfully employed to induce polyploidy in soybean and other legumes. Soybean, also known as *soja bean* or *soya bean*, has a history of polyploidy, which might be related to its tolerance mechanisms. This review paper discusses the limitations impacting soybean productivity under extreme environmental conditions and the role of synthetically developed polyploids in mitigating these abiotic stresses.

Keywords: abiotic stress tolerance; morphological traits; physiological traits; Polyploids; productivity; soybean

1. Introduction

The cultivated soybean (*Glycine max* L. Merill) which is an important crop belonging to the leguminous family is a paleopolyploid species (2n= 40) resulted from the polyploidization and domestication events that happened around 59 million years ago **(Yuan and Song, 2023**). Polyploidy induction is a key evolutionary mechanism that aided in creating diversification among plants **(Soltis *et al.* 2015).** These variations can morphological, physiological and genetical attributes **(Kim *et al.* 2015; Mangena and Mushadu, 2023)**.

Soybean also known by the name *soja bean* or *soya bean*, is a multipurpose legume plant which is extensively grown for its edible beans **(Saikia *et al.* 2020; Sharma 2021).** Millions of individuals benefit from soybeans, which are high in dietary proteins and oil, as well as vitamins, minerals, and energy. The ancient procedure of plant evolution, which involves a series of polyploidy events and the diversity of soybean, might have helped to boost both its agronomical and nutritional traits **(Sedivy *et al.* 2017).** Apart from having nutritional sources, soybean being a part of the legume family continues to be an important biological component of many ecosystems by engaging in symbiotic relationship with rhizobial bacteria in their root systems to fix nitrogen from the atmosphere. Dependency on the human-made nitrogen fertilisers like urea is decreased due to this biological phenomenon.

Polyploidy is a genetic situation where an organism possesses more than two complete sets of homologous chromosomes within the same nucleus or where the whole genome gets duplicated. **Ramsey and Schemske (1998)** demonstrated somatic doubling and gametic non-reduction as the two pathways that can lead to polyploidy conditions. Several synthetic treatments, such as colchicine, which is frequently used in plant breeding programs to induce polyploidy, can cause mutagenesis effects, resulting in the doubling of the genome number **(Eng *et al.* 2019; Shariatpanahi *et al.* 2021)**.

Polyploidy has also been reported in other leguminous crops following chemical treatments such as the use of colchicine as reported in cowpea **(Essel *et al.* 2015)** and faba bean **(Nagat *et al.* 2020)**. While colchicine has significantly induced polyploidy in soybeans and other leguminous crops like faba beans and cowpeas, its potential for varietal improvement and mitigating abiotic stress tolerance in these crops remains largely unexplored. Abiotic stress refers to environmental conditions that adversely affect growth and production in crops such as soybean, factors such as salt, drought, and high temperatures **(Oshunsanya *et al.* 2019; Yadav *et al.* 2020)**

2. Literature Review

2.1 Abiotic Stress as Challenges of Soybean Production

Getting a better yield is the aim of every plant breeder. Depletion of abiotic factors like water, light, soil nutrients and others are influencing the yield and productivity of crops, as emphasized by **Kumar (2020).** This stress adversely affects the afflicted plants' morphological, physiological, and molecular responses, which in turn impacts the crop output, biomass, and yield quality **(Hussain *et al.* 2018).**

Soybeans are an essential crop in agriculture, known for their diversity and economic value. The nutrient-rich seeds of soybean and the oil extracted from them are the main factors that determine its economic worth **(Rennie and Tanner, 1989).** However, abiotic factors such as drought, severe temperatures, and soil salinity can negatively influence soybean growth, development, and productivity, possibly threatening the quantity and quality of commercially valuable seeds and oil. **Staniak *et al.* (2023)** reported that abiotic stress causes about a 30 per cent reduction in soybean seed yield.

In India, Soybean is grown as a warm-season crop during two main seasons: spring (Feb-June) and kharif (July-Oct). The ideal temperature for soybean germination is around 30°C mentioned by **Tyagi and Tripathi, 1983.** Soybeans become sensitive in their germination period when the temperature falls below the ideal temperature. So **Alsajri (2018) and Staniak *et al.* (2021)** reported that low temperatures in soybean cultivars slowed down germination and delayed emergence, but when the temperature was raised, the plants emerged quite promptly. Lower temperatures can also have an impact on the flowering stage **(Staniak *et al.* 2021; Jähne *et al.* 2019).** Reduction in pollen grains and stigma number in soybean flowers because of the cold stress is also mentioned in the study reported by **Ohnishi *et al*. (2010)**. **Ohnishi *et al.* (2010)** also demonstrated that cold temperatures could result into poor pod set due to pollen development abnormalities. Cold temperatures negatively impact multiple physiological processes, including membrane fluidity, nutrition and water absorption, protein and nucleic acid structure, and metabolic rate **(Chinnusamy *et al.* 2007; Nouri *et al.* 2011)**. When soybeans are subjected to cold stress, it primarily impacts how they produce protein, and their cell walls operate **(Nouri *et al.* 2011)**.

The production of soybeans is also impacted by water scarcity. Several soybean accessions show decreased root length and dry biomass buildup **(Thu *et al.* 2014; Kunert *et al.* 2016)**. **Istanbuli *et al.* (2022)** demonstrated how drought stress affected the nodule development in legume crop chickpea. Most studies in soybean have been based on how drought affects the traits of shoot parts and flowers, but only a few reports are available **(Ferguson *et al.* 2010; Kunert *et al.* 2016)**. As discussed previously, leguminous crops like soybean are also helpful in increasing nitrogen content in the soil, by the process known as nitrogen fixation, where plants form mutualistic partnership with the rhizome bacteria and forms root nodules and within these nodules, nitrogen gets fixed. Drought stress diminished the rise in soybean plant height and leaf area expansion, with the inhibitory effects becoming more prominent as the drought became more severe and lasted long reported by **Dong *et al.* (2019)**. Extreme scarcity or lack of water can disturb the physiological processes such as the equilibrium of oxidation-reduction processes within cells, potentially leading to the accumulation of detrimental oxidative stress consequences over time **(Tardieu *et al.* 2018)**. When plants experience drought conditions, their stomata (pores) tend to close, leading to reduced levels of carbon dioxide inside the leaves, resulting in the generation of reactive oxygen species (ROS) because of insufficient amounts electrons in the photosynthetic machinery and a triggered photo-respiratory mechanism **(Tossi *et al*. 2022).** Increased ROS can disrupt cellular components like membranes, proteins, and nucleic acids (DNA and RNA), oxidize and degrade important biomolecules like lipids, leading to membrane leakage and loss of cellular compartmentalization **(Webster, 2012)**.

Besides cold and drought stress, agriculture productivity has been affected particularly by salinity stress, due to its broad spectrum and strong effects **(Amirjani *et al.* 2010)**, and which is subsequently affecting soybean productivity too. High salt concentration affects around 7% of the world's soil and 20% of the total arable area **(Rasool *et al.* 2013).** Salinity stress in plants is caused by an excessive accumulation of soluble salts like $CaCl_2$, $MgCl_2$, Na_2SO_4, KCl and especially $NaCl_2$ in the soil or water **(Tavakkoli, 2011)**. Excessive salt uptake by plants reduces the osmotic potential, leading to ion toxicity, which ultimately damages cell membranes and organelles (**Okon, 2019; Mangena, 2023**)**.** Salinity stress also exhibit stunted growth and reduced overall size in plants. **Amirjani (2010)** performed a study on soybean cultivars where they were exposed to 0, 50, 100 and 200 nM NaCl and when these salinity levels were slowly increased to 50, 100 nM, the plants showed significant decrease in their plant height and fresh weight. This finding shows that elevated salt stress has a detrimental effect on the soybean plant growth and development. Chlorophyll content, carotenoid content and the size of stomata are also affected by saline conditions **(Yarsi *et al.* 2017; Sharif *et al.* 2018).** In a research activity demonstrated by **Ghassemi-Golezani *et al.* (2011)** some genotypes showed reduced chlorophyll florescence as chlorophyll were destroyed due to ion- toxicity caused by saline environment. Reduction in chlorophyll content restricts photosynthetic activity and ultimately causes poor pod growth and decreased yield. **Figure 1** illustrates all these various consequences of abiotic stresses like heat, cold, drought, and salinity on soybean.

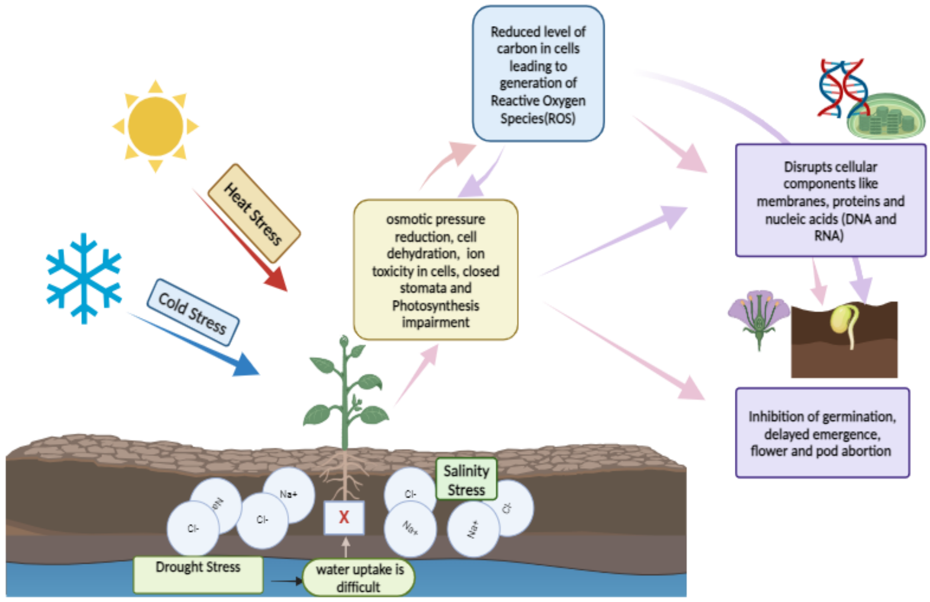

Figure 1. A Diagrammatic representation of the influence of the various abiotic stresses on the growth and development of soybean, including other leguminous and non-leguminous crops, (**Tavakkoli, 2011; Staniak *et al.* 2021; Mangena, 2023).** Created with www.biorender.com

Abiotic stress conditions are increasing day by day; it's an inevitable change. To thrive under such conditions, plants including soybean have undergone many anatomical, morpho-physiological, and biochemical adaptations **(Hasanuzzaman *et al.* 2016; Kashyap *et al.* 2021).** One such evolutionary strategy that plants have developed to cope with abiotic stress is polyploidy as emphasized earlier.

2.2 Induced Polyploidy for Mitigating Abiotic Stress in Soybean

Polyploidy is a major force in plant evolution, speciation and crop domestication by creating novel genomic compositions and enhancing heterosis **(Fort *et al.* 2016; Nieto *et al.* 2020)**. **Koenen *et al.* (2021)** reported that with the advent of allopolyploid hybrids, which include several chromosomal sets derived from distinct species, corresponded with huge worldwide climatic and geographical shifts, resulting in a mass extinction event for many species.

The process of evolution and domestication that led to polyploid soybean involved alterations both at molecular and phenotypic level. The deviations in gene composition and expression patterns identified in polyploid resulted in a variety of beneficial physiological responses, increasing tolerance and allowing for optimal growth and production under abiotic stress conditions as explained by **Rao *et al.* (2020)**. Similarly, these changes might have contributed to the soybean's increased tolerance to fluctuating environmental conditions, enabling it to adapt and thrive in diverse environments. Polyploidy has been associated with improved salt tolerance in sorghum **(Ceccarelli *et al.* 2006),** citrus **(Saleh *et al.* 2008),** wheat **(Du *et al.* 2023)** and several other non-leguminous crops.

In relation to this, there are some mutagenic agents such as colchicine is particularly used to produce synthetic polyploids that may provide a viable alternative to biotechnology-based strategies for crop improvement. Colchicine is an alkaloid derived from the bulbs of the *Colchicum autumnale* plant, also known as meadow saffron, having molecular formula $C_{22}H_{25}NO_6$ **(Ben-Chetrit, 2019).** These artificially induced polyploid plants have larger shoot and root cells when compared to diploid plants **(Tal and Gardi, 1976) (Figure 2),** and these bigger root cells promotes better water use efficiency from soil and also not only size but the numbers of stomata and xylem vessel facilitating better water movement and avoiding stress conditions as reported by **Tossi *et al*. (2022). Figure 2** demonstrates the potential of colchicine-induced polyploids in stress tolerance and other crop improvement traits over diploids **(Tal and Gardi, 1976; Mangena and Mushadu, 2023)**. **Mangena and Mushadu (2023)** discussed the use of colchicine-induced polyploids as a method to introduce genetic variation associated with abiotic stresses, particularly drought stress tolerance. In a research study performed by **Mangena (2021)** two soybean genotypes, TGx1835-10E and Dundee, were subjected to imbibition (soaking) in solutions containing varying concentrations of colchicine (0.0% (control), 0.1%, 0.5%, and 1%) and significant changes in the germination, seedling growth and morphology were observed and also reported that with prolonged period of imbibition affected the polyploidy induction. Some other constraints have also been observed, that can hamper proper induction of polyploidy plants when using colchicine, namely earlier embryo abortion, lack of endosperm, seed sterility etc. **(Rodrangboon *et al.* 2002; Carbajal *et al.* 2019)**.

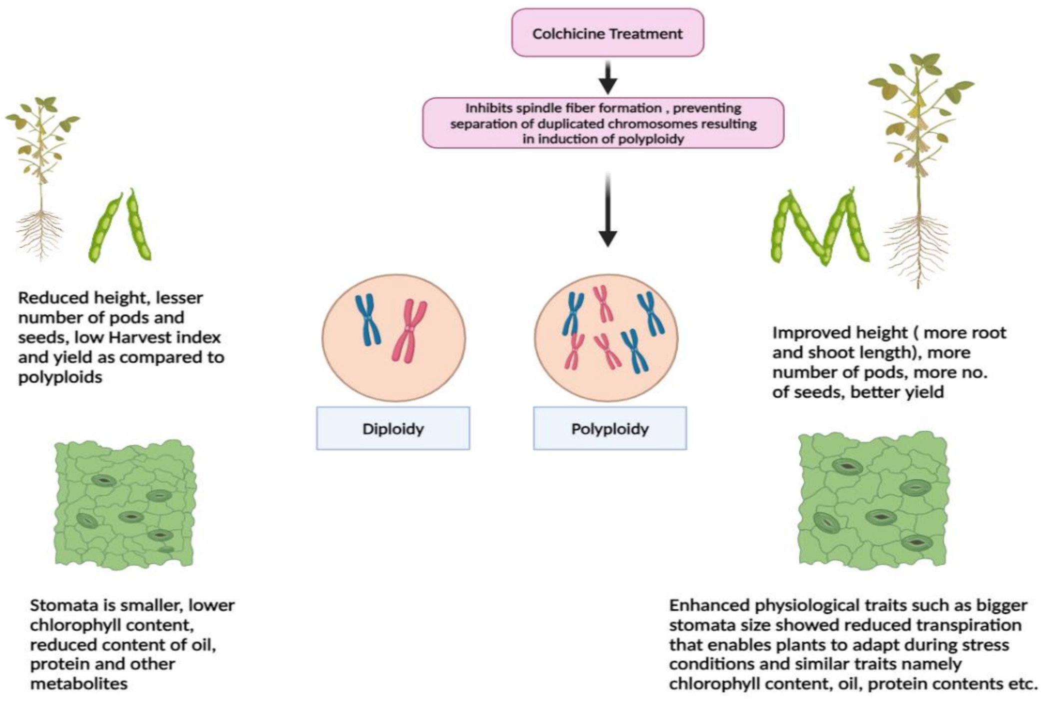

Figure 2. Demonstrating the potential of colchicine-induced polyploids in stress tolerance and other crop improvement traits over diploids **(Tal and Gardi, 1976; Mangena and Mushadu, 2023)**. Made with www.biorender.com

Numerous, other mutagenic agents have been also utilized and should be practiced more to enhance agronomic parameters in soybean such as gamma rays and EMS (Ethyl methane sulfonate) **(Pavadai *et al.* 2009; Gopinath and Pavadai, 2015)**.

Several mechanisms to mitigate abiotic stress have been adopted by plants. Soybeans use complex defence systems against abiotic stress by transforming stress signals into altered gene expression, which triggers processes that allow adaptation and tolerance to these circumstances **(Mangena, 2023).** Further, plants when exposed to different environmental conditions such as drought, harsh temperatures, or elevated salt levels have been identified with higher amounts of proteins that neutralise oxidative damage **(Komatsu *et al.* 2013; Raza *et al.* 2016)**. These antioxidant proteins aid in the removal of reactive oxygen species, preventing oxidative damage to essential components within plant cells **(Hossain *et al*. 2012; Raza *et al.* 2016)**. Heat shock proteins (HSPs) play a crucial function in dealing with harsh environmental conditions **(Kim *et al.* 2014; Wu *et al.* 2022)**. These diversified stress response mechanisms, facilitated by antioxidants and proteins, highlight the complex molecular adaptations that soybean uses for abiotic stress tolerance. However, there is no absolute proof that describes polyploids as a tool in bringing tolerance mechanism in soybean and other leguminous crop.

3. Conclusion and Future Insights

Polyploidy is an evolutionary phenomenon that might be used to mitigate abiotic stress tolerance in soybeans and other leguminous crops. The contribution of induced polyploidy on the development of abiotic stress resistance in soybeans is still little studied and needs to be explored. This review without any doubt explains the role of polyploidy as a potential strategy in mitigating stress conditions in soybean, but there has been less proof suggesting that the tolerant mechanism found in this crop is mainly facilitated by polyploidy. More importantly, better protocols based on in-vitro and in-vivo should be facilitated to generate successful artificial polyploids that are suited to adverse environments, it will promote further research in the application of biotechnology, such as comparative studies between diploids and synthetically developed polyploids, along with the role of gene expression changes and particularly epigenetic modifications associated with polyploidy will help us in understanding its role in mediating stress responses and adaptations in soybean. Overall, there has been enough evidence that genetic redundancy and increased heterozygosity resulting from the polyploidy mechanism, have improved water use efficiency, antioxidant defence mechanisms, and the expression of stress-responsive genes by bringing physiological, molecular and biochemical adaptations in plants.

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