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Stress Tolerance in Chilli (*Capsicum annum* L.):

A Focus on Abiotic Factors

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Abiotic stresses such as drought, heat, cold, and salinity significantly limit the production of chilli (*Capsicum annum* L.), a crucial crop for both spices and vegetables worldwide. In chilli plants, these environmental conditions cause major physiological, biochemical, and molecular changes that hinder growth, hinder photosynthesis, and result in severe production losses. This article thoroughly summarises the state of the art about the impacts of the main abiotic stresses on chillies, including how they specifically affect plant physiology, including water relations, gas exchange, and nutrient uptake, as well as the oxidative damage and metabolic abnormalities that occur. It also critically analyses the different breeding methods used to improve chilli's resistance to abiotic stress, from traditional screening and selection of germplasm to sophisticated molecular breeding techniques like Marker-Assisted Selection (MAS) and the incorporation of multi-omics technologies (genomics, transcriptomics, proteomics, and metabolomics). The article also emphasises how genetic engineering and advanced genome editing technologies, especially CRISPR/Cas9, can be used to accurately alter chilli genomes for increased resilience.

Keywords: Chilli, hot pepper, heat stress, cold stress, chilling injury, breeding, molecular breeding.

Introduction

A highly prized spice and vegetable crop grown all over the world is chilli (*Capsicum annum* L.). The increasing yearly demand for chilli peppers and the corresponding financial benefits are what entice farmers to cultivate them. It is renowned for both its high nutritional value, which includes vitamins, carotenoids, and antioxidants, and its strong flavour, which is derived from capsaicin. Capsaicin, the main component in chilli peppers, has a number of health benefits. The health benefits of consuming chilli peppers are highlighted by the thermogenic properties of capsaicin, which may improve digestion and nutrition absorption (Cheng *et al.*, 2023). The nightshade family (Solanaceae) includes the genus *Capsicum*, which includes chilli plants (Ahmad *et al.*, 2024). Their strong flavour is the main reason behind their cultivation. Among the most widely consumed types of chilli peppers are the cayenne, jalapeño, serrano, and Thai kinds.

The biggest producer of chillies in India is Andhra Pradesh, which is followed by Telangana, Madhya Pradesh, Karnataka, and Odisha. The most widely grown Indian chilli types are Bhut Jolokia, Kashmiri, Guntur, Jwala, Kantari, Byadagi, Ramnad, Dhani, Tomoto, Madras Puri, Khola, and Dalle Khursani. It is very important to use the genetically distinct genotypes for breeding programs in chilli, specially in case of abiotic stress resistance (Nehru *et al.*, 2003). Abiotic stressors that impair plant development and reproduction include traits like salinity, heat, cold, and drought. Thus, it is essential to use abiotic stress breeding to create chilli varieties that are more resilient to these circumstances in order to guarantee food security and sustainable agricultural production. As a result, the degree of genetic variation represented by the yield-contributing qualities in the germplasm is crucial to the success of a chilli breeding.

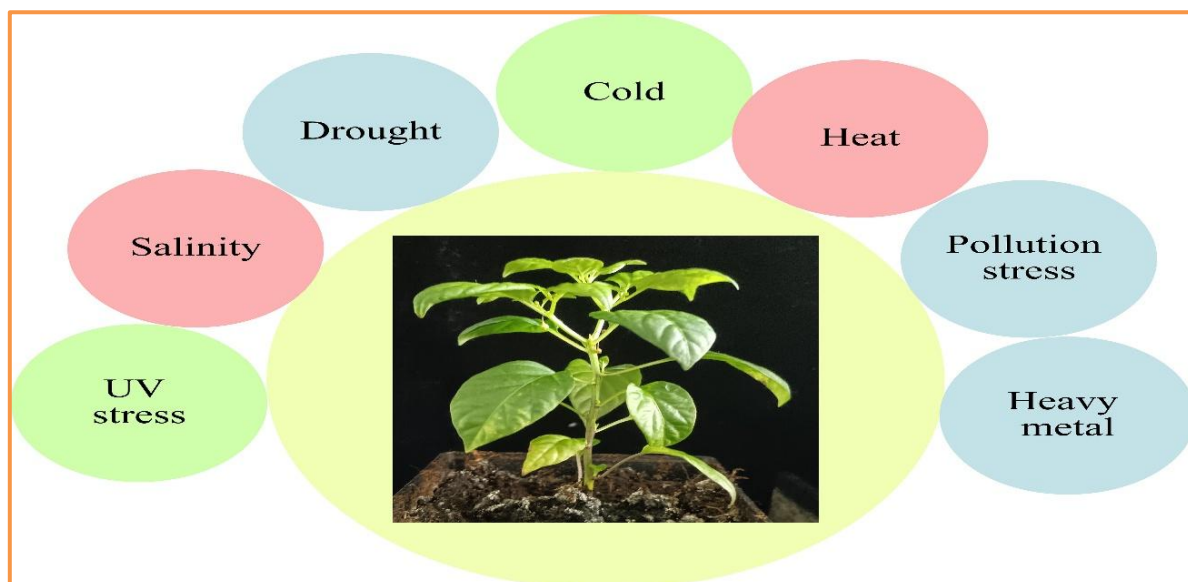
Need of stress tolerance

As an important horticultural crop, chilli (*Capsicum annuum* L.), is very much prone to abiotic (non-living) stresses, which are made worse by intensive agricultural techniques and shifting climatic circumstances. Due to several environmental issues that have a substantial impact on their growth, productivity, and total output, chilli crops must be able to withstand stress. An essential tactic to guarantee steady and sustained chilli production globally is the development of stress-tolerant cultivars. Numerous environmental factors restrict the growth and development of chilli crops, which eventually affects productivity. Global food security is seriously threatened by the decline in crop productivity, particularly in light of the growing population and the shrinking amount of arable land.

For both plant biologists and agronomists, one of the most important topics is how plants react to different environmental stimuli. Salt, drought, and heat stress are the most significant and prevalent environmental stresses that impact plant growth and development. Traditional breeding methods are ineffective due to the complex traits linked to stress tolerance; therefore, innovations are required to close the gap between the world's food supply and demand. In this field, the creation of novel and efficient techniques is essential.

Key physiological indicators to identify plant stress conditions

S. No.	Indicators	Description
1	Relative Water Content (RWC):	RWC is a reliable indicator of the plant's water status and is significantly influenced by drought stress.
2	Leaf Area and Expansion Rate:	Drought stress causes a significant decrease in the leaf area of chilli plants.
3	Leaf Temperature:	Leaf temperature increases significantly under heat stress
4	Osmotic Adjustment:	Salt-affected plants wilt due to increased accumulation of soluble salts, leading to osmotic stress
5	Chlorophyll Content:	Chlorophyll content is significantly higher under non-saline conditions and reduces with imposed salinity
6	Leaf Water Content:	A considerable decrease in leaf water content is observed in chilli plants subjected to drought stress
7	Nutrient Uptake:	Drought stress leads to a considerable decrease in nutrient uptake.
8	Growth Parameters:	Low-temperature stress causes deleterious effects at the vegetative growth stage of plants.
9	Chlorosis (Yellowing of leaves):	Yellowing of leaves is a common symptom in response to cold stress
10	Photosynthesis:	Combined stresses can significantly impact photosynthesis.



Different types of abiotic stresses seen in chilli: (Zheng et al., 2023)

Different approaches for abiotic stress tolerance in chilli

- 1. Selection and Screening of Germplasm:-** For traditional breeding operations, it is essential to identify and use desired genes and traits from existing germplasm collections. Also, in order to identify natural sources of abiotic stress tolerance, breeders investigate the genetic variety within chilli germplasm. This also helps to increase the effectiveness of identifying promising genotypes, traits that are heritable and associated with desired characteristics—like stress resistance—are chosen. Finding the most resistant cultivars is aided by assessing genotype performance in conditions that mimic certain stresses, such as low nitrogen or water stress.
- 2. Marker Assisted Selection (MAS):-** By using molecular markers associated with desired features, MAS makes it possible to select plants with higher yield and abiotic stress tolerance more quickly and precisely. This method works very well for finding target genes. DNA Markers are crucial for MAS to identify DNA markers linked to characteristics like economic significance and resilience to biotic and abiotic stressors.
- 3. Genomic Selection (GS):-** GS is a marker-assisted selection technique that predicts an individual's genomic estimated breeding values (GEBV) by using genome-wide marker data, thus increasing selection efficiency.
- 4. CRISPR/Cas9 Gene Editing:-** This technology enables exact nucleotide-level changes, such as replacing or changing alleles, silencing, or introducing new genes at specific locations. Because of its precision, affordability, and time efficiency, it has become a potential technique for enhancing plants' ability to withstand abiotic stress. In crops including rice, maize, and chilli, CRISPR/Cas9 has been effectively used to alter genes for increased tolerance to a variety of abiotic challenges like drought, salinity, and extremely high temperatures. Additionally, it permits multiplex genome editing, which targets several genes at once.
- 5. Transcriptomics:-** This approach examines how genes (mRNAs and sRNAs) express themselves under stress, identifying genes that are differently regulated and essential for plant adaptation. Understanding how plants react to environmental stressors like heat, cold, drought, and salinity can be gained through transcriptome analysis.
- 6. Proteomics:-** Studies on proteomics offer a comprehensive view of the metabolic pathways and protein networks involved in stress tolerance processes. It aids in locating master regulator proteins that are essential to pathways involved in the response to abiotic stress.
- 7. Genetic engineering:-** It is the process of creating new features and attributes that are challenging to accomplish through traditional breeding. It focusses on finding and

overexpressing important genes, like those involved in signal transduction, detoxification, or osmoprotection, that have a positive impact on stress tolerance.

8. **Conventional breeding:-** Methods such as heterosis breeding, estimation of gene action is useful for selection of new elite hybrids which can tolerate stress conditions. General combining ability and Specific combining ability are the measure which are widely used for this approach (Barhate *et al.*, 2023). This helps to choose the best hybrid combination to be suggested for future breeding Programmes (Shaniware *et al.*, 2024).

Importance of breeding for stress tolerance

The goal of creating stress-tolerant chilli varieties is to create a more robust and sustainable agricultural system, not only to reduce losses. Following are some importance:

- ❖ **Food Security:** As the world's population continues to grow, crop varieties that can flourish in challenging environments are necessary to guarantee a steady supply of food. Stress-tolerant chillies help achieve this objective by lowering crop failure and stabilising yields.
- ❖ **Economic Stability for Farmers:** Farmers face severe financial difficulty as a result of crop losses brought on by stress. Farmers' lives can be improved by stress-tolerant cultivars that yield more consistent harvests.
- ❖ **Sustainable Agriculture:** Sustainable agriculture methods include lowering water use through drought-tolerant cultivars and relying less on chemical inputs for disease and insect management.
- ❖ **Adaptation to Climate Change:** Extreme weather events are becoming more frequent and intense due to climate change. A proactive adaptation technique to guarantee that chilli agriculture continues to be viable in a changing climate is breeding for stress tolerance.
- ❖ **Genetic Improvement:** By incorporating several desired features into new chilli varieties through stress tolerance breeding, quality attributes such as pungency, flavour, and nutritional content can be improved as well as overall plant performance.
- ❖ **Advancement in breeding technologies:** Because stress tolerance is so complicated, modern breeding methods such as genetic engineering, proteomics, and genomics are required. These methods make the breeding process more accurate and efficient by speeding up the discovery and introgression of stress-responsive genes and QTLs.

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