

Breeding for salt tolerance

Salt Tolerance: refers to the ability of plants to prevent, reduce or overcome injurious effects of soluble salts present in their root zone.

It is a global problem as saline and alkali soils are found in almost all the countries of the world, more in Semi Arid Tropic (SAT) of world.

Problem of salinity can be overcome by two ways:

1. Soil reclamation : costly, time consuming and shortlived
2. Resistant varieties: less costly, more effective, long lasting require longer period to develop.

Behavior / characteristics of plants to salt:

1. Land races more tolerant than High yielding varieties. Tolerant plants varieties are found in salt affected areas
2. Salt tolerance capacity differs from species to species. Also genetic differences exist among cultivars for their salt tolerance capacity.
3. Different crop plants show differential response to salinity

Salinity Crops

- a. Highly tolerant crops Sugarbeet, sunflower, barley (grain), cotton, date palm, asparagus
 - b. Moderately Tolerant crops Barley (Forage), rye, sorghum, wheat, safflower, soybean
 - c. moderately sensitive Rice, corn, foxtail millet, cow pea, peanut, sugarcane, tomato, potato, sweet potato, radish, alfalfa, cabbage
 - d. Extremely sensitive Citrus, straw berry, melon, peas, other legumes, apple, rajmabean, carrot, okra, onion (orange)
4. Higher ploidy level crops are more tolerant than lower ploidy level crops. Eg. Hexaploid wheat more tolerant than tetraploid
Tetraploid Brassica more tolerant than diploid Brassica
 5. In rice tall, coarse grained, late maturing varieties- more tolerant
 6. In sugarcane different strains have differential tolerance Barley more tolerant than wheat.

Symptoms of plants to salt stress :

1. Retardation / cessation of growth
2. Necrosis
3. Leaf abscission
4. Loss of turgor
5. Ultimate death of plant

Mechanisms of salt tolerance:

2 types of mechanisms

1. **Salt Tolerance** : Plants respond to salinity stress by accumulating salt, generally in their cells or glands and rootsetc.

2. **Salt avoidance** : plants avoid salt stress by maintaining their cell salt concentration unchanged either by water absorption eg. Rice, chenopodiaceae family or by salt exclusion eg. Tomato, soybean, citrus, wheat grass

Glycophytes (Non-halophytes) plants owe their resistance primarily to avoidance. Eg. Barley
Halophytes (plants that grew in salty or alkaline soils) show tolerance by ion accumulation mechanism

Breeding methods

Breeding methods are same but breeding strategies are

1. Breeding for yield potential should have greater emphasis than breeding for salt resistance per se (As screening is done on the basis of yield reduction in stress environment as compared to non-stress Environment.).
2. Selection should be done in stress target environments (As abiotic stress resistance is an important part of Environ. Fitness & is bound to be location specific i.e. it is related to narrow adaptation).

Screening Techniques

Common methods are

1. Sand culture by using nutrient solution in sand & irrigation with saline water
2. Solution culture by using solution culture tanks (Hydroponic culture)
3. Microplot techniques by using small microplots

Microplot Techniques: By using small microplots of size 6 x 3 x 1 m (CSSRI, Karnal, Haryana) at central soil salinity Research Institute.

Then Multilocation Trial (MLT) conducted over seasons to get more reliable results.

Genotypes which survive better under salinity are considered tolerant & tested further.

Selection criteria

1. Germination (%) in saline medium
2. Dry matter accumulation (seedling / plant dry wt.) / Early vigour
3. Leaf senescence or death – Estimated by total dead leaf area or No. of dead leaves
4. Leaf necrosis
5. Leaf ion content
6. Osmoregulation (Determined as maintenance of turgor under stress) Measured as proline or CHO accumulation or accumulation of glycine, betaine etc.
7. Yield – Economic yield

Problems

1. Creation of reliable controlled salinity Env.
2. Scoring for salinity resistance
3. Genetic control – it is complex & polygenic
4. Mechanisms of resistance poorly understood. Salinity may have interaction with other stresses.

COLD TOLERANCE

When temperatures remain above-freezing *i.e.* $>0^{\circ}\text{C}$ to $<10^{\circ}\text{C}$ - 15°C it is called chilling When temperature remain below freezing *i.e.* $<0^{\circ}\text{C}$ it is called Freezing.

A. Chilling Resistance:

Chilling sensitive plants are typically tropical plants. Temperate plants are generally tolerant to chilling injury.

Effects of chilling stress on plants :

1. Reduced germination
2. Poor seedling establishment
3. Stunted growth
4. Wilting
5. Chlorosis
6. Necrosis
7. Pollen sterility
8. Poor fruit set / seed formation
9. Reduced root growth
10. Locked open stomata
11. ABA accumulation

At subcellular level

12. Reduces membrane stability
13. Poor chlorophyll synthesis (affected)
14. Reduced photosynthesis & respiration
15. Toxicity due to H₂O₂ formation

A. Chilling Tolerance

Ability of some genotypes to survive / perform better under chilling stress than other genotypes is called chilling tolerance. It is because of chilling hardening, i.e. an earlier exposure to a near chilling temperature for a specified period as a result of which chilling tolerance of the concerned plants increases.

Mechanisms of chilling tolerance:

1. Membrane lipid unsaturation
2. Reduced sensitivity of photosynthesis
3. Increased chlorophyll accumulation
4. Improved germination
5. Improved fruit / seed set
6. Pollen fertility

Sources of chilling Tolerance:

1. Late adopted breeding populations eg. maize
2. Germplasm(eg. That collected from high altitude, low temperature geographic regions)
3. Induced mutants for cold tolerance
4. Cold tolerant somaclonal variants
5. Related wild species eg. Tomato

Selection criteria

Based on -

1. Germination test
2. Growth under stress (measured as plant dry matter accumulation)
3. Chlorophyll Loss under chilling stress eg. rice, cucumber, tomato (measured as seedling colour)
4. Membrane stability : (Assayed in terms of solute leakage from tissues)
5. Photosynthesis : Chilling injury to photosynthesis is assayed as variable chlorophyll fluorescence at 685nm
6. Seedling mortality
7. Seed / Fruit set
8. Pollen fertility (assayed during injury at PMC)

B. Freezing Resistance

Freezing injury / Frost injury / cryo injury

Freezing Stress: Dormant state is conducive to freezing resistance, while resistance in actively growing tissue is rare: Thus Freezing resistance largely involves surviving freezing stress in such a manner as to enable subsequent regrowth when the temperature rises.

As water in plants cools below 0°C, it may either

1. Freeze *i.e.* form ice or
2. Super cool without forming ice.

Effects of freezing stress

1. Ice formation : Two types
Intercellular ice formation
Intracellular ice formation

Intercellular Ice formation: Initiation of ice formation on plant surface is sufficient to induce freezing of the internal (intercellular & xylem vessels etc.) water in most plant species.

Intracellular ice formation: It is more lethal may be due to physical disruption of subcellular structure by ice crystals. Intracellular ice formation is the major and terminal freezing stress.

Extracellular ice formation in a cases the conc entrations of extracellular solutes, the re by water is withdrawn from the cells during extracellular ice formation. This creates water- stress in the frozen tissue / plant.

2. Membrane disruptions:

- Freezing causes disruptions is and / or alter the semipermeable properties of plasmamembrane
- Loss of solutes from the cells occur
- Cells remain plasmolyzed even after thawing which is often called as frost plasmolysis
- Cells may become highly turgid due to uptake of excess water.

3. Supercooling:

Cooling of water below 0°C without ice crystal formation is called supercooling

- In plants water may cool down to -1 to -15°C in herbaceous sps and to -40 to -45°C in hardy trees.
- This becomes possible apparently because internal ice-nucleators are absent in such cases.
- This is regarded as an important. Mechanism of freezing avoidance

4. Stress due to external factors : Consequent to freezing

- 1) Ice sheet formation below and above the ground causes reserve depletion anoxia etc. in plants.
- 2) Tissues killed during freeze-thaw are highly prone to pathogen attacks
- 3) Auto toxicity may occur

Mechanism of Freezing Resistance :

The ability of a genotype to survive freezing stress and to recover and re grow after thawing is known as freezing resistance. Freezing resistance is a complex trait involving physiological, chemical & physical processes at the tissue and cell level.

Mechanism of Freezing resistance.

1. **Freezing avoidance** : The ability of plant tissues / or genes (but the whole plants) to avoid ice formation at sub zero temperature is called freezing avoidance

Supercooling is a mechanism of freezing avoidance it is controlled by

1. Lack of ice-nucleators
2. Small cell size
3. Little or no intercellular space
4. Low moisture content
5. Barriers against external nucleators
6. Presence of antinucleators

2. **Freezing Tolerance** : Ability of plants to survive the stresses generated by extra cellular ice formation and to recover and regrow after thawing is known as freezing tolerance. The various components of freezing tolerance are as follows:

- 1) Osmotic adjustment
- 2) Amount of bound water
- 3) Plasma membrane stability
- 4) Cell wall components properties
- 5) Cold-responsive proteins

Eg. ABA

Sources of freezing tolerance

1. Cultivated varieties
2. Germplasm lines
3. Induced Mutations
4. Related wild species Eg. Wheat *Agropyron* sps; rye
Barley – *H. jubatum*, *H. brachyantherum* x *H. bogdanii*, *H. jubatum* x *H. compressum*
Oats – *Avena sterilis*
5. Transgenes: Eg. chemical Synthesized antifreeze protein gene, ala3, into tobacco

Selection criteria:

Based on

1. Field survival
2. Freezing test in laboratory
3. Cryofreezing
4. Osmoregulation

Problems in breeding for freezing tolerance

1. Freezing Tolerance is a complex trait & involves several components. So, it is not readily measurable under field conditions
2. Breeding work under field conditions is highly influenced by other environmental factors and biotic stresses
3. Due to large G X E for the trait field survival shows poor heritability
4. Freezing tolerance also shows a large GXE interaction which limits progress under selection
5. Laboratory tests are yet to be developed to screen large breeding population