



# COLD STRESS

Dathan C S

## **STRESS**

An adverse force or a condition, which inhibits the normal functioning and well being of a biological system such as plants.

## **VARIOUS TYPES ARE**

Cold, heat, drought, flood, salinity, etc.

# Low temperature stress

Chilling stress - when plants are exposed to a low temperature above 0 °C.

Freezing stress – when plants are exposed to a low temperature below 0 °C

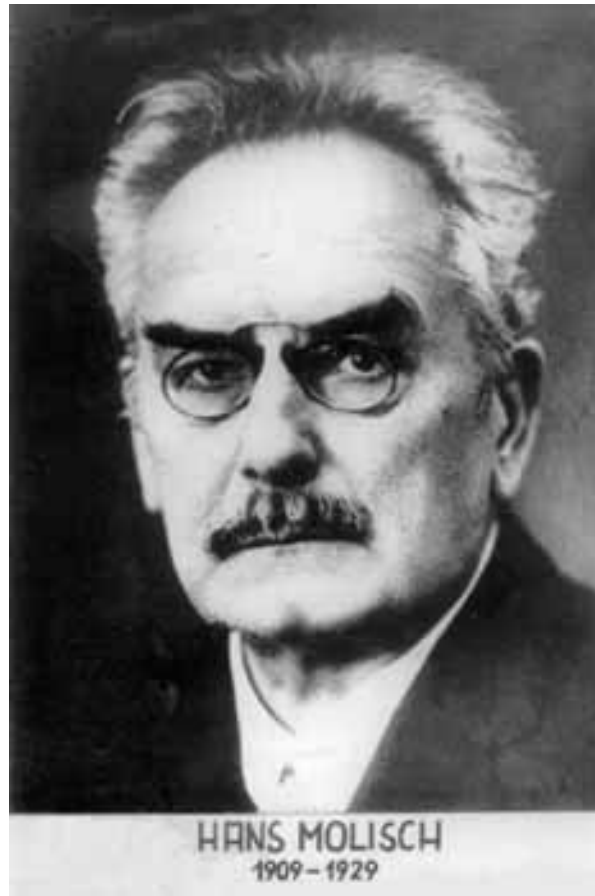


# CHILLING STRESS

Plants may develop physiological disorders when exposed to low but non-freezing temperatures.

The German plant physiologist Molisch suggested the term 'chilling injury' as long ago as 1897 to describe this phenomenon.

The international "star of plant physiology" Hans MOLISCH. He studied at the university of Vienna. In 1894, he became professor in Prague and in 1909, in Vienna, he began research into "frost-resistance" at the cellular level. As a microbiologist, he was working with purple and luminescent bacteria, and has been innovative in photosynthesis research and chemotaxonomy.



# Plant at Chilling Stress



# Chilling Injury

- Plant chilling injury refers to an injury that is caused by a temperature drop to below to 10 to 15°C but above the freezing point.
- Among crops, maize, Phaseolus bean, rice, tomato, cucumber, sweet potato, and cotton are chilling sensitive. Passiflora, Coleus, and Gloxinia are examples of susceptible ornamentals.
- The most common site implicated for chilling injury is the plasma membrane.
- The consequences of this change may lead to cell leakage or disruption.
- Changes in membrane permeability are often invoked as a cause of the loss of cell turgor

# Symptoms of Chilling injury

- **Cellular changes** : Changes in membrane structure and composition, decreased protoplasmic streaming, electrolyte leakage and plasmolysis.
- **Altered metabolism** : Increased or reduced respiration, depending on severity of stress, production of abnormal metabolites due to anaerobic condition.
- **Common Symptoms**
  - Reduced plant growth and death
  - Surface lesions on leaves and fruits
  - Abnormal curling, lobbing and crinkling of leaves
  - Water soaking of tissues
  - Cracking, splitting and dieback of stems
  - Internal discolouration (vascular browning)
  - Increased susceptibility to decay
  - Failure to ripen normally
  - Loss of vigour (potato lose the ability to sprout if chilled)









# Symptoms of Chilling injury

- **Most common symptoms of chilling stress is**

Rapid wilting followed by water soaked patches which develop into sunken pits that reflect cells tissue collapse. Following warming, the sunken pits usually dry up, leaving necrotic patches of tissues on the leaf surface.

- **Chilling symptoms in fruits vary and include**

- Sunken pits in cucumber
- Browning of skins and degradation of pulp tissue in banana
- Blackheart of pine apple (Wilson, 1987)

# Chilling Injury

- Occurs at Low temperature but nonfreezing temperatures
- Chilling injury occurs in
  - Tropical and subtropical plants at 10 °C to 25°C
  - Temperate plants at 0 to 15°C
- Chilling Effect is manifested by physiological and cytological changes
- Cytological changes may be reversible or irreversible depending upon time of exposure to low temperature

# Affects on plants

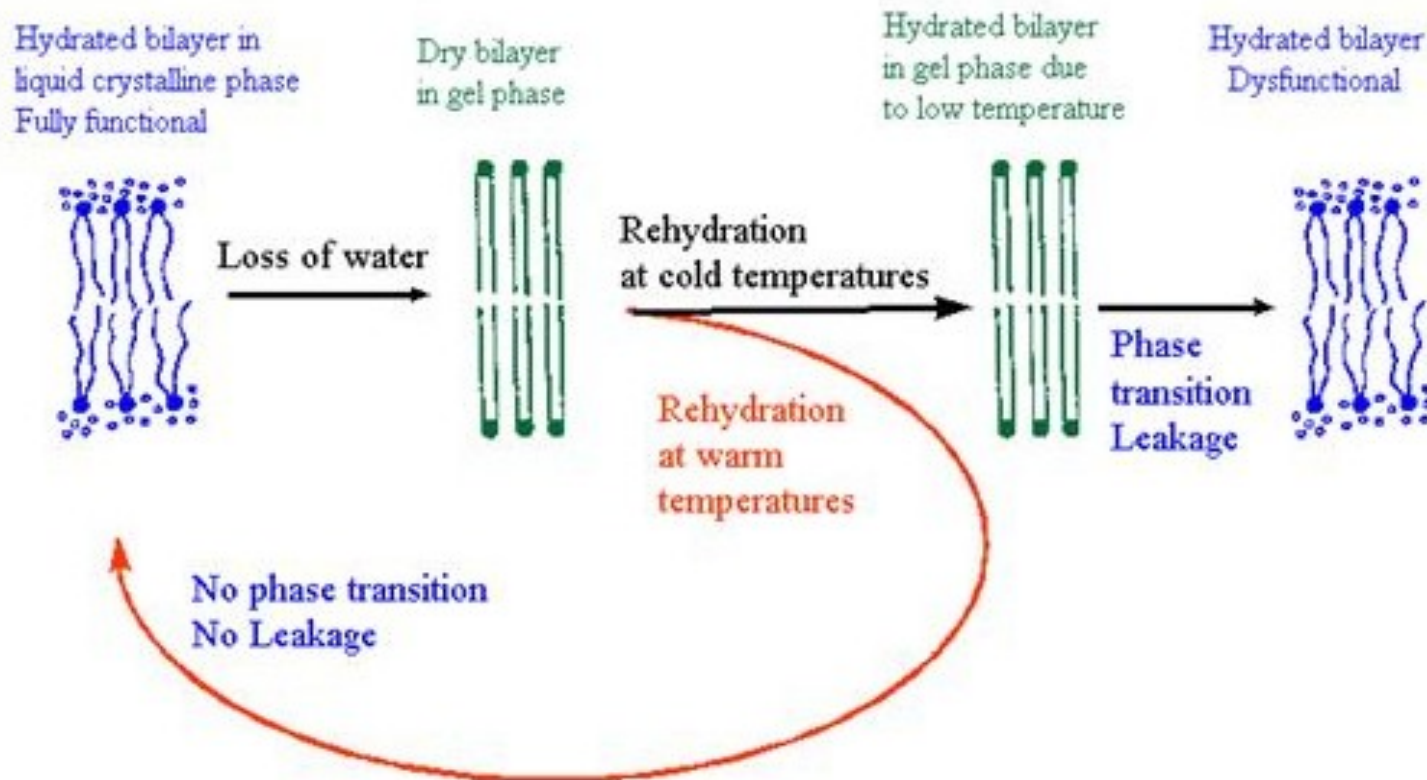
Chilling injury causes several metabolic or physiological dysfunctions to the plant including

- disruption of the conversion of starch to sugars (amylolytic activity)
- decreased carbon dioxide exchange
- reduction in net photosynthesis
- the destruction/degradation of chlorophyll

# Affects on plants

- **Cellular Membranes**

- The first symptom of chilling injury is the phase transition from liquid crystalline phase to solid gel state
- Increase in permeability of plasmalemma results in leakage of organic and inorganic substances



# Affects on plants

## Cellular Membranes(contd..)

- Plasmolysis: Plasmalemma- pressed against the tonoplast and deformed into the vacuole as sac like intrusions
- Formation of crystalline deposits in root cells, epidermal, mesophyll and vascular cells of leaves -leading to tonoplast disruption.
- Tonoplast injury is irreversible
- During hardening at low or above zero temp the lipid bodies accumulate in cytoplasm in close association with plasmalemma.

## Lipid composition

- Ratio of Unsaturated to saturated fatty acids is higher in chilling resistant plants
- Increase in activity of fatty acid de-saturases was found in chilling resistant plants



# Cytological Changes

- Swelling of plastid membranes and mitochondrial membranes
- Swelling of chloroplast thylakoids
- Decrease in size and no. of starch grains
- Grana disintegration and increase in size and no. of plastoglobules
- Mitochondria with reduced cristae and transparent matrix
- Mitochondria – double the volume
- Extensive dilation and vesiculation of smooth ER cisternae

# Cytological Changes

- Rough ER completely disappears *ie.*, Ribosomes are lost from the membrane
- Dilated Vesicular ER cisternae – Accumulation of cryoprotective substances
- Transformation of rough ER into vacuolated smooth ER-represents early stage of chilling
- Since ER-most dynamic structure-full reversibility of ER ultrastructure is possible
- Swelling of dictyosomes
- Longer exposure to chilling-disintegration of dictyosomes

## Causes of Chilling injury

- The primary cause has been found to be the opening (and locking) of the leaf stomata when the permeability of the roots to water is low.
- The leaves lose water faster than it can be replaced and they become dehydrated.
- In some plants, the stomata behave properly at chilling temperatures and the injury is said to be metabolic.
- A decrease in respiration, photosynthesis and fatty acid synthesis may all contribute to the chill-starvation of some plants.

# Freezing stress

When plants are exposed to a low temperature below 0°C. Freezing damage occurs primarily due to the formation of ice crystals, which damage cell structure when the temperature falls below 0°C.



# Varying Severity of Frost/Freeze Injury

(Planted Apr. 5, Image May 28)

Both plants  
at leaf stage V5

Plant #1

Plant #2







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# Freezing stress

Freezing injury in plants can be from two sources:

## 1. Freezing of soil water

The soil water that is available to plants is found in the porous regions between soil particles. It freezes at about  $-2^{\circ}\text{C}$ , depriving the plant of its source of water.

## 2. Freezing of the fluids within the plant.

Freezing of water within the plant can cause disruption of structure and function of cells and tissues. Ice usually forms first in the cell walls and intercellular spaces. Damage occurs when ice crystals grow and puncture into the cytoplasm.

# Symptoms of Freezing injury

- Desiccation or burning of foliage
- Water-soaked areas that progress to necrotic spots on leaves, stems or fruit and death of sections of the plant or the entire plant.
- Close examination of woody plants several days or weeks after freezing may reveal a dead or weakened root system or split bark on stems or branches.
- Obvious symptoms on plant foliage may not be present until after the plant has been stressed by warm temperatures.
- Wilting and/or desiccation, as caused by direct drought stress.

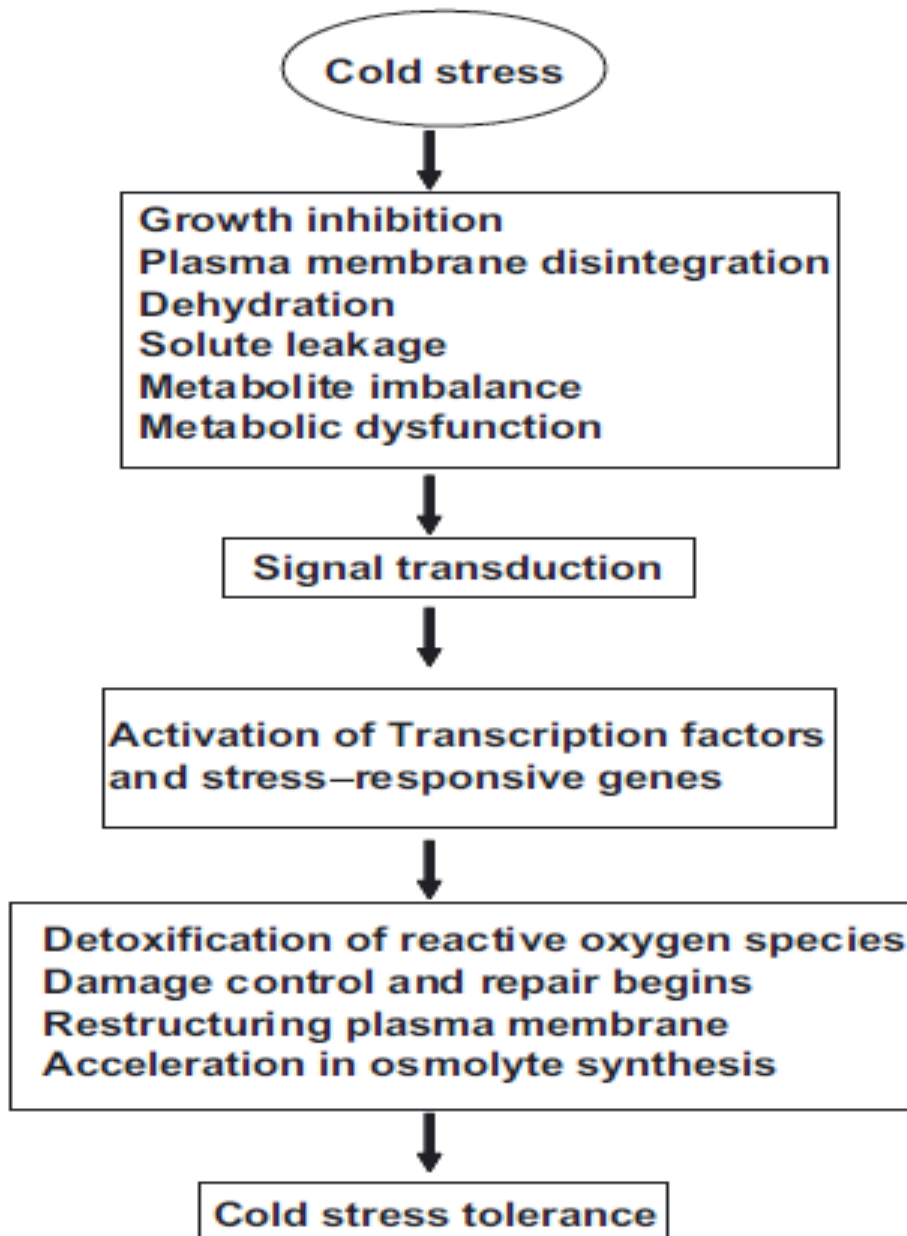
# **Formation of ice intracellularly may be due to**

- By internal nucleation (certain large polysaccharides /proteins serve as nucleating agents to form ice)
- By penetration of external ice crystals into the cells
- Intracellular ice formation is very lethal which causes immediate disruption of cells.
- It spreads from cell to cell through plasmodesmata
- Formed in the cell wall adjacent to the intercellular spaces
- Originates spontaneously from centers of nucleation in the cytoplasm.

# Membrane changes

- Intact cellular membranes act as effective barrier to the propagation of ice. This depends upon temperature and cold hardening .Cellular Membranes are more susceptible to freezing damage than soluble enzymes.
- Plasmalemma is the major site of lethal injury. Leakage of ions from thawed tissues occurs due to protoplasmic swelling and alteration in permeability to  $K^+$ ions.

# Signal transduction



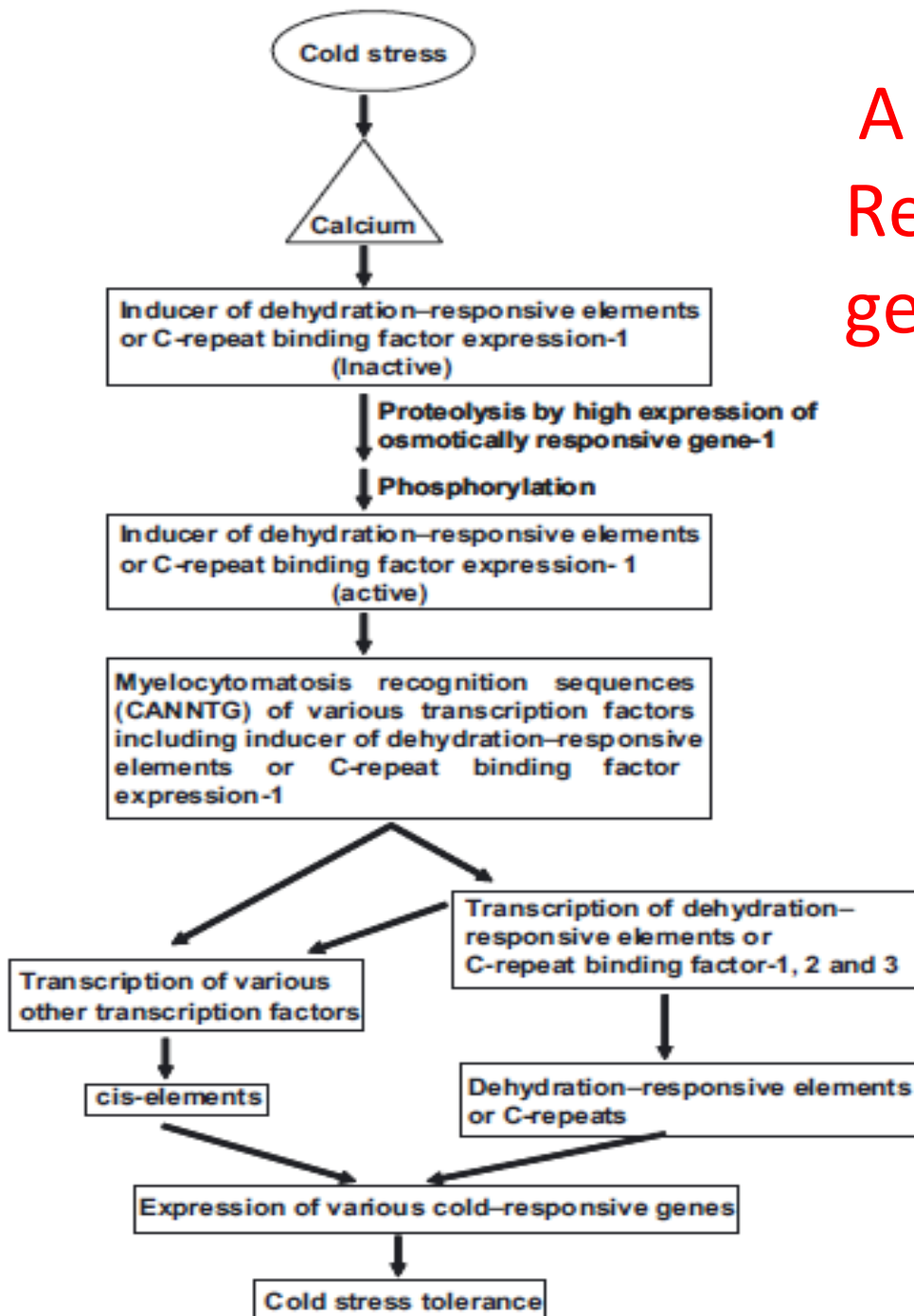
Fundamental responses of plants during cold stress exposure. Cold stress exposure causes various physiochemical disturbances, leading to growth inhibition. Cold stress response is perceived by plants through a signal transduction that leads to the activation of transcription factors and cold-responsive genes. Such transcription factors and genes control the damage due to cold stress and help in providing tolerance to plants.

# Chilling tolerance

- The acquisition of chilling tolerance is associated with huge changes in metabolite contents, such as the accumulation of soluble sugars, dehydrins, RNA-chaperones, and an increase in reactive oxygen species (ROS) detoxification activities.
- These changes in cellular components are due to a transcriptome rearrangement. They mean that chilling has been perceived and transduced to the nucleus.
- Chilling is not perceived by a single mechanism in plants but at different sensory levels, that are the very biological processes disturbed by the temperature downshift. Once perceived, chilling stress is transduced.
- An increase in cytosolic calcium is the major transducing event that will then regulate the activity of many signalling components, including phospholipases and protein kinases. This will end in changes in gene expression.
- The best-documented pathway leading to gene induction in response to cold is the CBF (C-repeat binding factor) pathway.

# A Transcription Factor Regulates cold-Induced gene Expression

Involvement of various transcription factors in the induction of cold-responsive genes during cold stress. Many cold stress-induced genes are activated by transcriptional activators called C-repeat binding Factors.



# Prevention & Protection

## Cold hardening

- Cold hardening alters the behavior of the stomata so that they close under the same conditions; the root permeability is also increased.
- Cold hardening affects the lipid content of cell membranes and has been found to lower the optimum temperature for photosynthesis and respiration.





# Prevention & Protection

Some methods to avoid chilling and freezing effects artificially:

## SOIL BANKING

- It consists of placing a mound of soil around the tree's trunk to protect the bud union and trunk from cold.
- One of the most efficient cold protection methods for young trees and has been used with success for many years





# Prevention & Protection

## Wrapping

- Most tree wraps can be attached anytime during the year and left on the tree throughout the year or even for several years.
- Wraps should be properly positioned and fastened around the trunk for best results.
- It is important to cover the entire lower trunk, especially at the base.



# Prevention & Protection

## Sprinkling

- It can be used to moderate temperatures above freezing because of sensible heat in water and can maintain plant leaf temperature at 1 to 2°C degrees or more.
- Water applied to aisles of shade structures or greenhouses increases the moisture content of the air and soil surrounding the plants, thus slowing the rate of temperature drop.
- The water absorbs heat during the day which is released slowly at night.



# Prevention & Protection

## Fogging

- Fog also retards the loss of heat from soil and plant surfaces to the atmosphere
- Fog can provide up to 4°C (8°F) of protection outdoors during radiational cooling
- Applying ground water with an average temperature of 21°C (70°F) to a greenhouse can create a ground fog if the ground surface is several degrees cooler than the water.
- The applied water adds heat to the plant environment and/or buffers temperature change by increased humidity.
- Temperatures can be elevated as much as 5°C (9°F) in these unheated structures





# Prevention & Protection

## Supplementary Heat supply

### **Air movement method:**

- Wind machines have been used for many years in citrus and vegetable industries and recently in the ornamental industry as a means of cold protection
- Air movement also helps distribute and circulate heat added by orchard heaters or other sources.

### **Warm water movement:**

- Circulation of warm water (43 to 54°C, 110 to 130°F), not hot water, in enclosed growing and/or storage areas is effective to prevent cold injuries

# SURVIVAL STRATEGIES

## Anti freeze proteins (AFP)

- Declines rate of ice crystal growth
- Lowers the efficiency of ice nucleation sites
- Lowers temp. at which ice forms

## Osmoprotectants

- Osmolytes- quarternary amines, amino acids, sugar alcohols
- Balances the osmotic potential of externally increased osmotic pressure

## ABA

- Plants develop freezing resistance when treated with exogenous ABA

# Thank u

