Plant Physiology (Biol 511)

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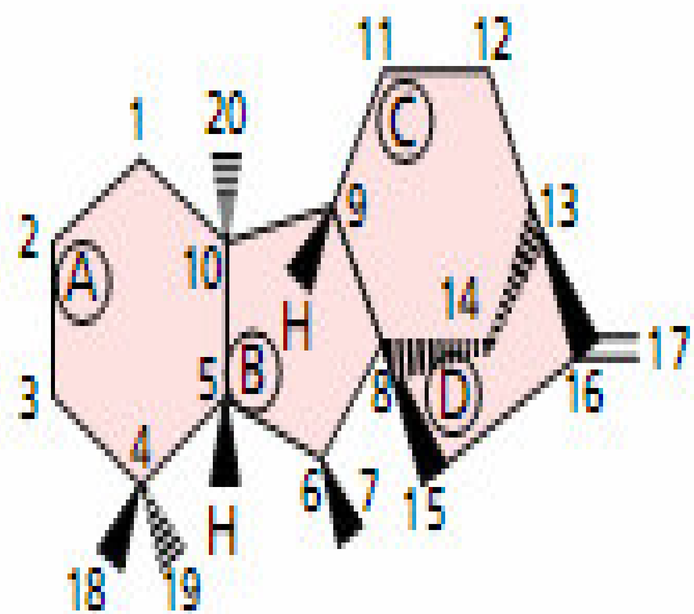
GIBBERELLINS

- Gibberellins were first discovered in Japan in 1930's from diseased rice seedlings that grew excessively tall
- This disease is known as *bakanae*, meaning foolish seedling, caused by *Gibberella fujikuroi* (imperfect stage/asexual is *Fusarium moniliforme*)
- The extract of fungus applied to rice seedlings caused the same symptoms of *bakanae* disease as the fungus itself demonstrating that a definite chemical substance is responsible for the disease

- Yabute and Hayashi (1930’s) subsequently isolated an active compound from this fungus and named it as **gibberellins**
- As of 2002, 125 gibberellins were discovered
- All gibberellins are derived from the *ent*-gibberellane skeleton
- The *ent*-gibberellane skeleton has 4 rings- A, B, C & D with 20 C atoms having methyl (CH_3) group at 7, 17, 18, 19 and 20 C positions
- All gibberellins are acidic and are named GA (gibberellic acid) with number subscripts e.g. GA1, GA3, GA4, GA7, GA9, GA32
- The subscripts number shows the order of their discovery

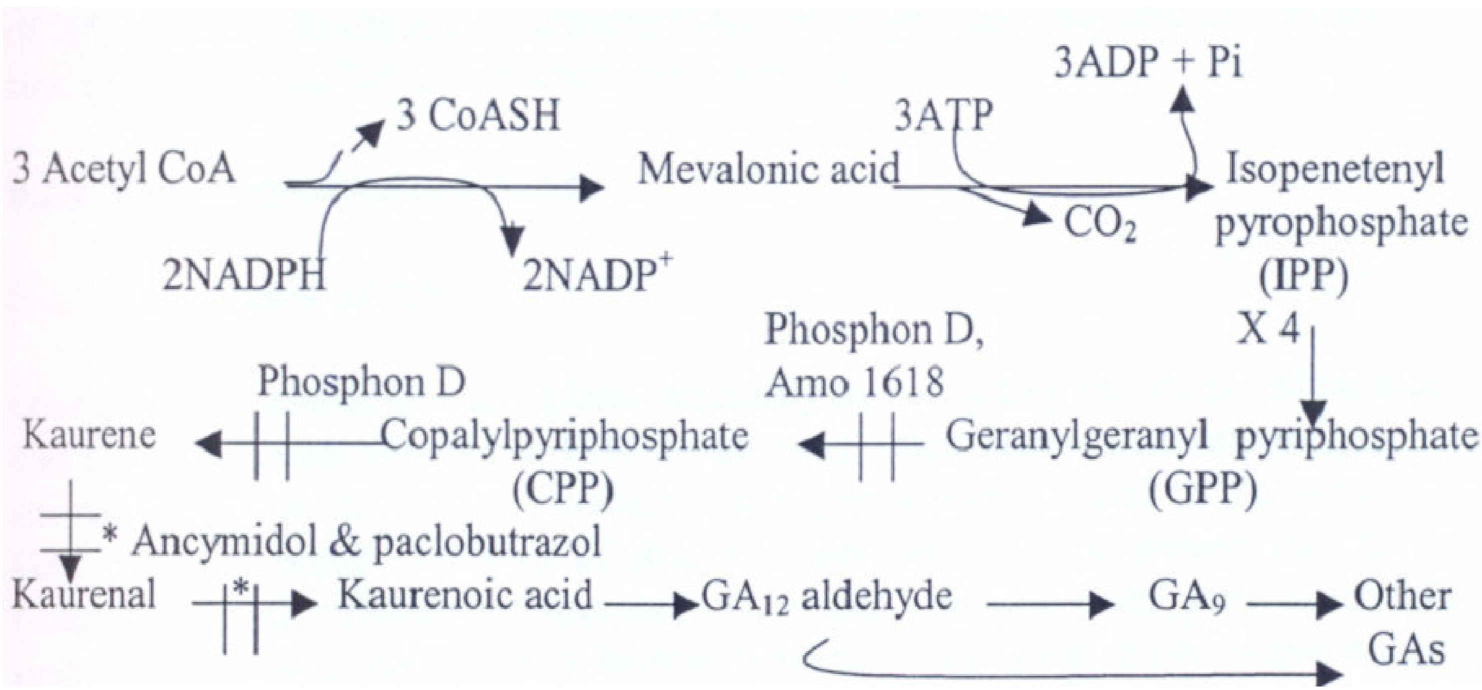
All gibberellins are based on the *ent*-gibberellane skeleton



ent-Gibberellane structure

Biosynthesis of GA

- Gibberellins constitute a large family of diterpene acids and are synthesized by a branch of the terpenoid pathway - from acetate units of acetyl CoA through mevalonic acid pathway below



- Researchers have determined the entire GA biosynthetic pathway in seed and vegetative tissues of several species by feeding various radioactive precursors and intermediates and examining the production of the other compounds of the pathway.
- Plastid, endoplasmic reticulum & cytosol take part in GA biosynthesis

Site of GA biosynthesis & transport

- The GAs are mostly synthesized in:
  - young immature seeds
  - Young developing leaves
  - Shoot apex
  - Roots
- Mature seeds contain almost no GAS
- GA biosynthetic enzymes and GAs are specifically localized in young, actively growing buds, leaves, and upper internodes
- Both phloem and xylem participate in GAs transport

Factors affecting GAs biosynthesis

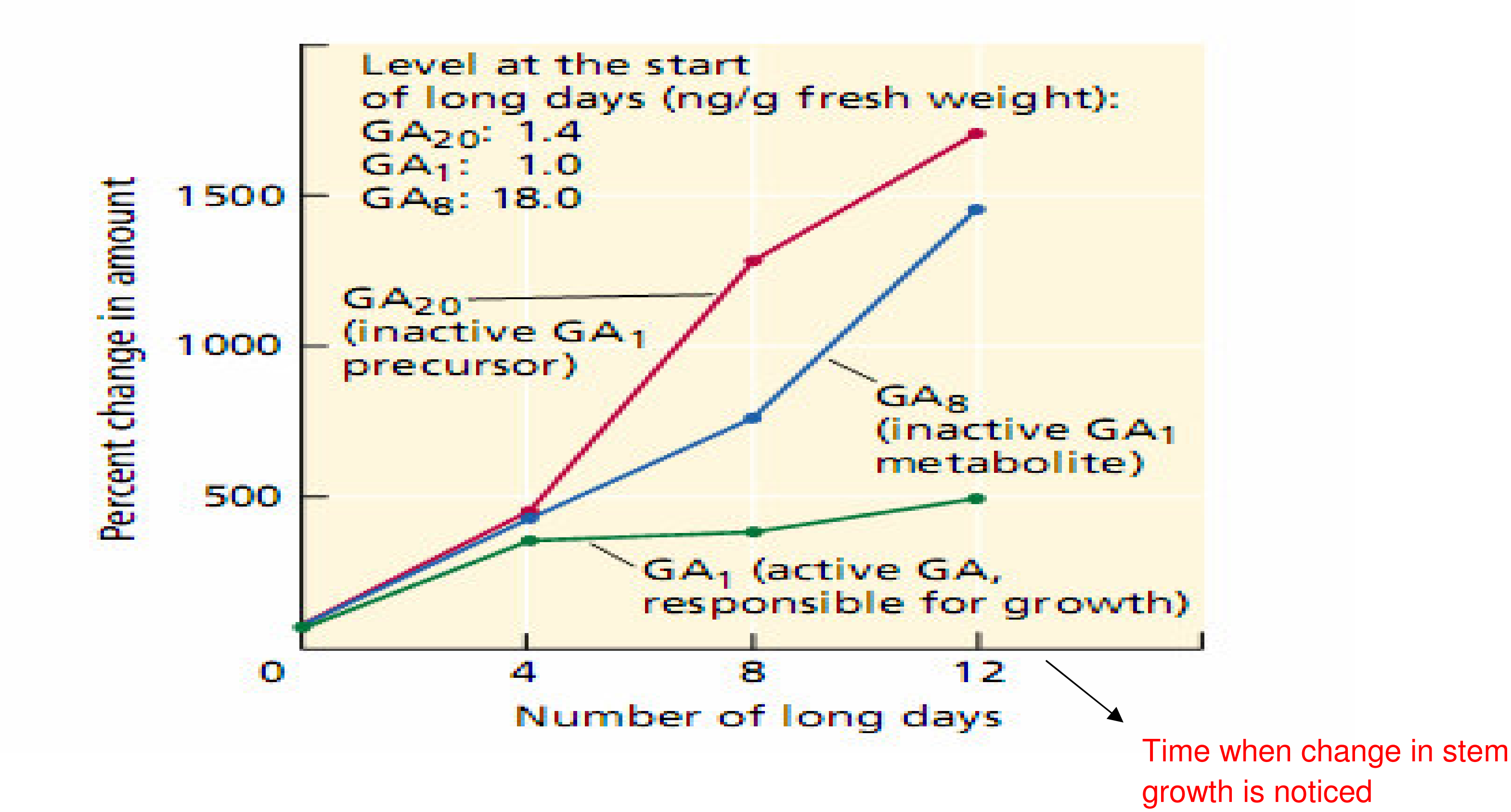
i- GA Regulates Its own metabolism

- Endogenous GA regulates its own metabolism by either switching on or inhibiting the transcription of the genes that encode enzymes of GA biosynthesis and degradation (feedback and feed-forward regulation, respectively)
- In this way the level of active GAs is kept within a narrow range, provided that precursors are available and the enzymes of GA biosynthesis and degradation are functional

ii-Environmental Conditions Can Alter the Transcription of GA Biosynthesis Genes

- Light promotes germination of seed due to increases in GA1 levels
- Photoperiod regulates GA biosynthesis. Plants that require long day to flower show increased rate of GA synthesis when shifted from short days to long days. In some instance, the requirement for long day can be substituted by exogenous GA application

- Tuberization of potatoes is promoted by short days. Potato plants were grown under either long days or short days. The formation of tubers in short days is associated with a decline in GA1 levels



- The fivefold increase in GA1 is what causes growth in spinach exposed to an increasing number of long days. Noticeable stem elongation starts at about 14 days

❖Temperature effects

- Cold temperatures are required for the germination of certain seeds (stratification) and for flowering in certain species (vernalization)
- GAs can substitute for the cold treatment

Degradation of GAs

- GAs are degraded as follows:
- i. GA3 is normally degraded slowly but during active growth GAs are rapidly metabolised by hydroxylation to inactive products
- ii. GAs become readily conjugated and remains inactive
- iii. Conversion of highly active GA to less active one, e.g. GA4 to hydroxylated GA34

Physiological roles of GAs

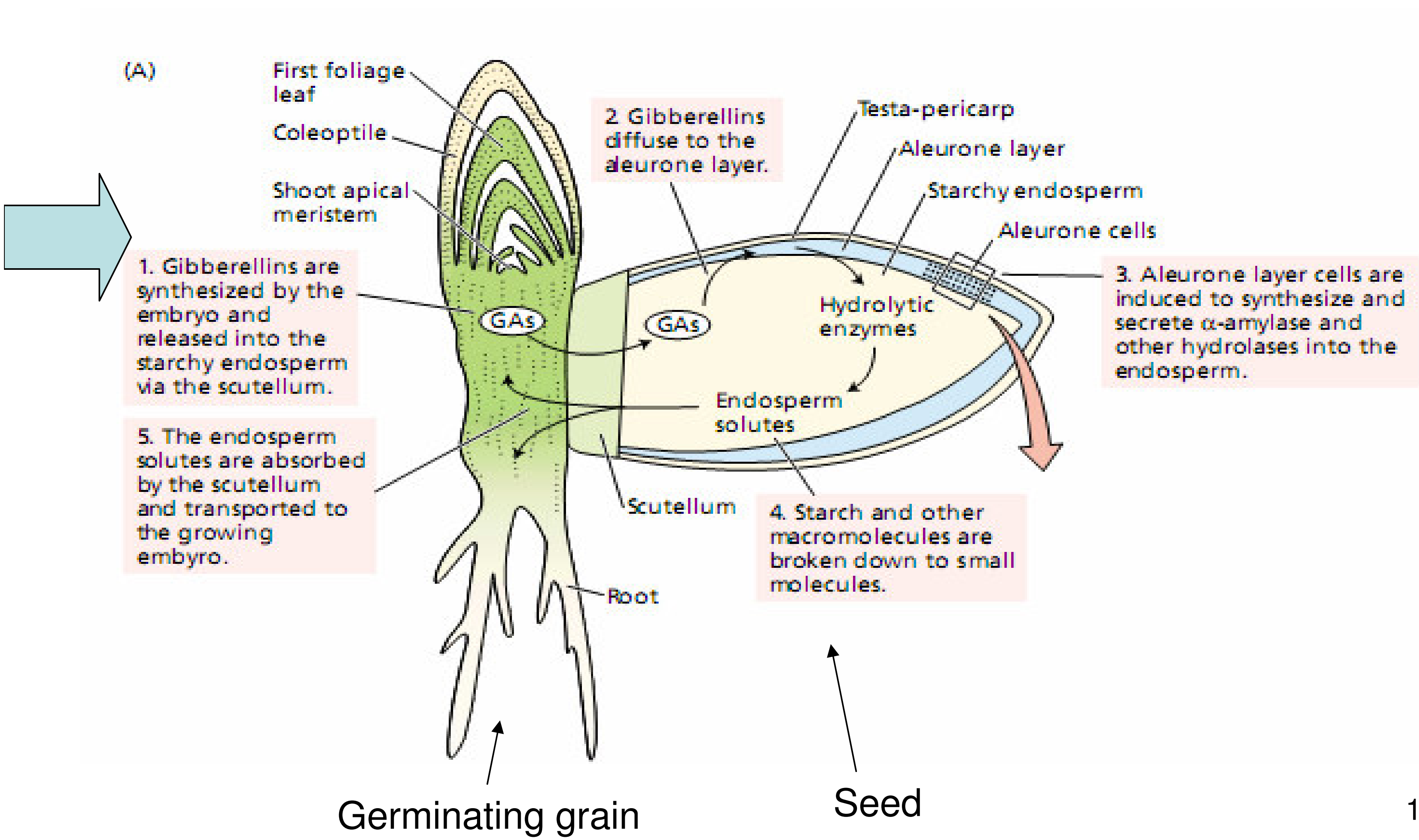
- **Growth of plants:** GAs promote growth of intact plants primarily by elongating the stems

- Cabbage, a long-day plant remains as a rosette in short days, but it can be induced to bolt and flower by applications of gibberellin. In the case illustrated, giant flowering stalks were produced

- **Seed and bud dormancy:** GAs overcome the seed and bud dormancy substituting the cold temperature treatment, long day or red light (eg grape, apple, many biennial vegetables, etc)
- **Flowering:** GAs can substitute for long day requirement in some species, and vernalization (eg cabbage)

- **Mobilization of foods and mineral nutrients in seed storage cells:**
- Degradation/ hydrolytic breakdown of food reserves in germinating seeds is assisted by GAs
- **Parthenocarpy in fruits:** GAs specially GA4 and GA7 cause parthenocarpic (seedless) fruits in some species
- **Delayed senescence or aging in leaves and citrus fruits**

Structure of a barley grain and the functions of various tissues during germination



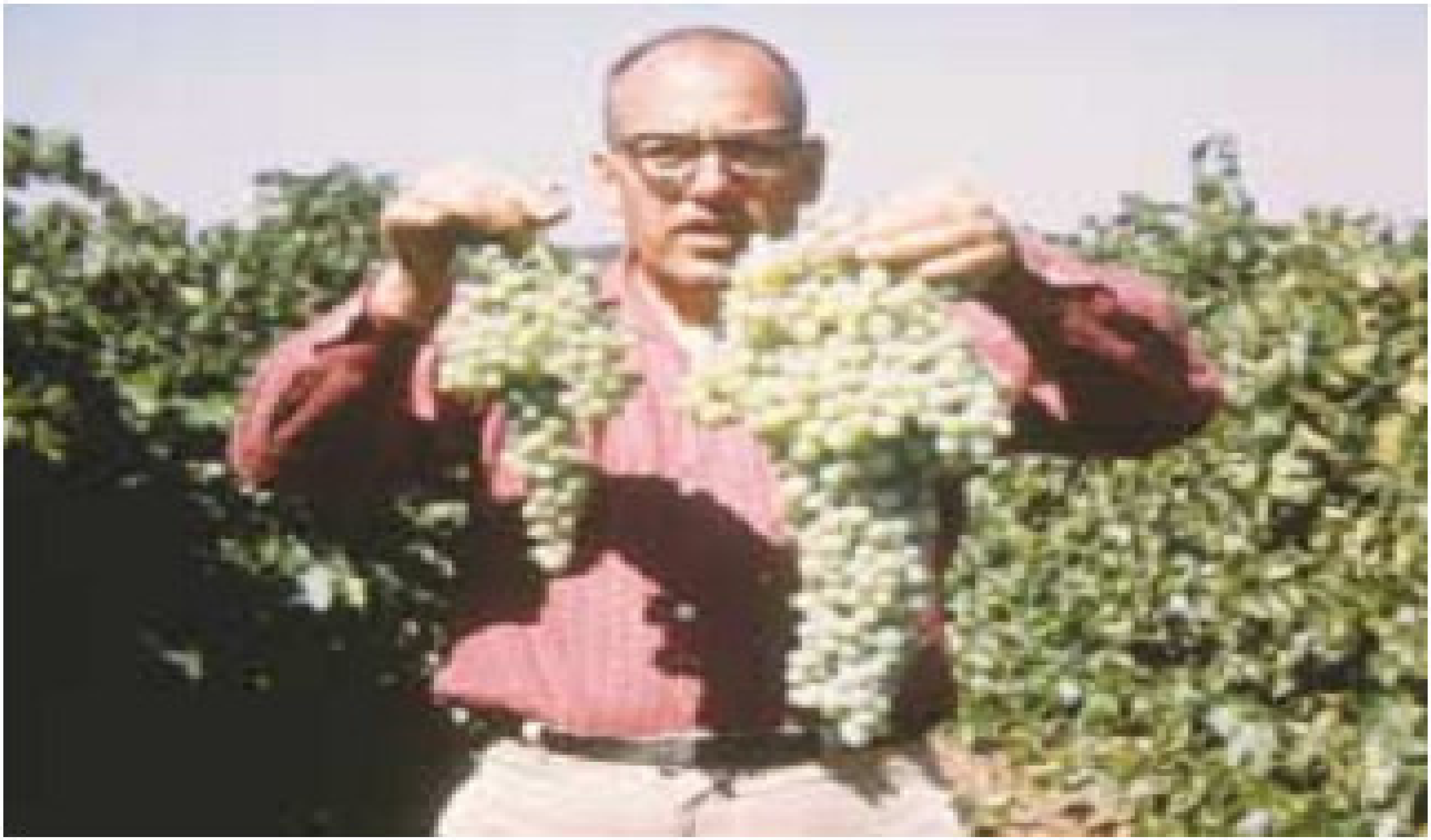
Possible Mechanisms of GA Action

1. GAs stimulate cell division & cell elongation in the shoot apex
2. GAs increase cell-wall plasticity
3. GAs promote cell growth because they increase hydrolysis of starch, fructan (a polymer composed of units of fructose) and sucrose into glucose and fructose

Commercial uses of GAs

1. GA is sprayed on grape at bloom and fruit-set for elongation of bunches in order to produce bigger berries with little or no fungal infection
2. A mixture of GA4 and GA7 is used to enhance the seed production in Pinaceae (pine) seed orchard
3. Breweries use GA to increase the rate of malting
4. Use of GA in naval orange orchard to prevent several rind disorders (splitting, creasing, cold pitting, puffing) and to have firmer rinds due to delay in senescence
5. GAs are used in sugarcane plantations to increase the sugarcane growth and sugar yields

- Gibberellin induces growth in Thompson’s seedless grapes. The bunch on the left is an untreated control. The bunch on the right was sprayed with GA during fruit development.

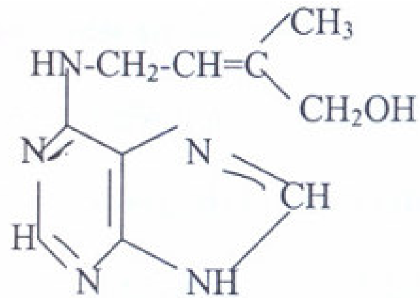


CYTOKININS

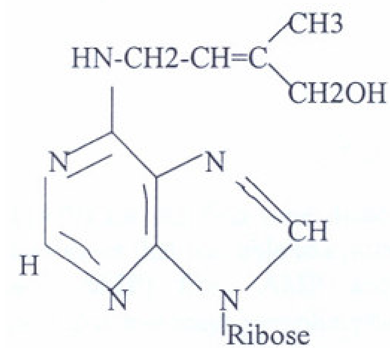
- THE CYTOKININS were discovered in the search for factors that stimulate plant cells to divide (i.e., undergo cytokinesis)
- Cell division (cytokinesis) promoting hormones
- The most active cell division promoting substances are kinetin, zeatin and zeatin riboside

- Cytokinins have effects on many other physiological and developmental processes, including leaf senescence, nutrient mobilization, apical dominance, the formation and activity of shoot apical meristems, floral development, the breaking of bud dormancy, and seed germination
- Cytokinins also appear to mediate many aspects of light-regulated development, including chloroplast differentiation, the development of autotrophic metabolism, and leaf and cotyledon expansion

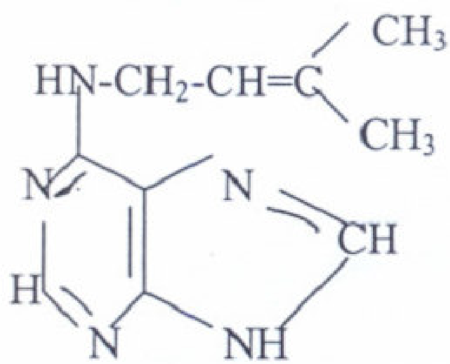
Structures of some of the naturally occurring and synthetic cytokinins



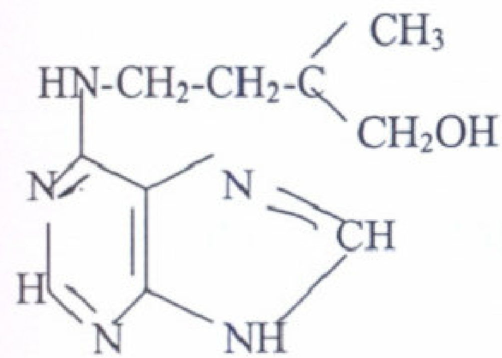
Zeatin



Zeatin riboside or ribosyl zeatin



Isopentenyl adenine



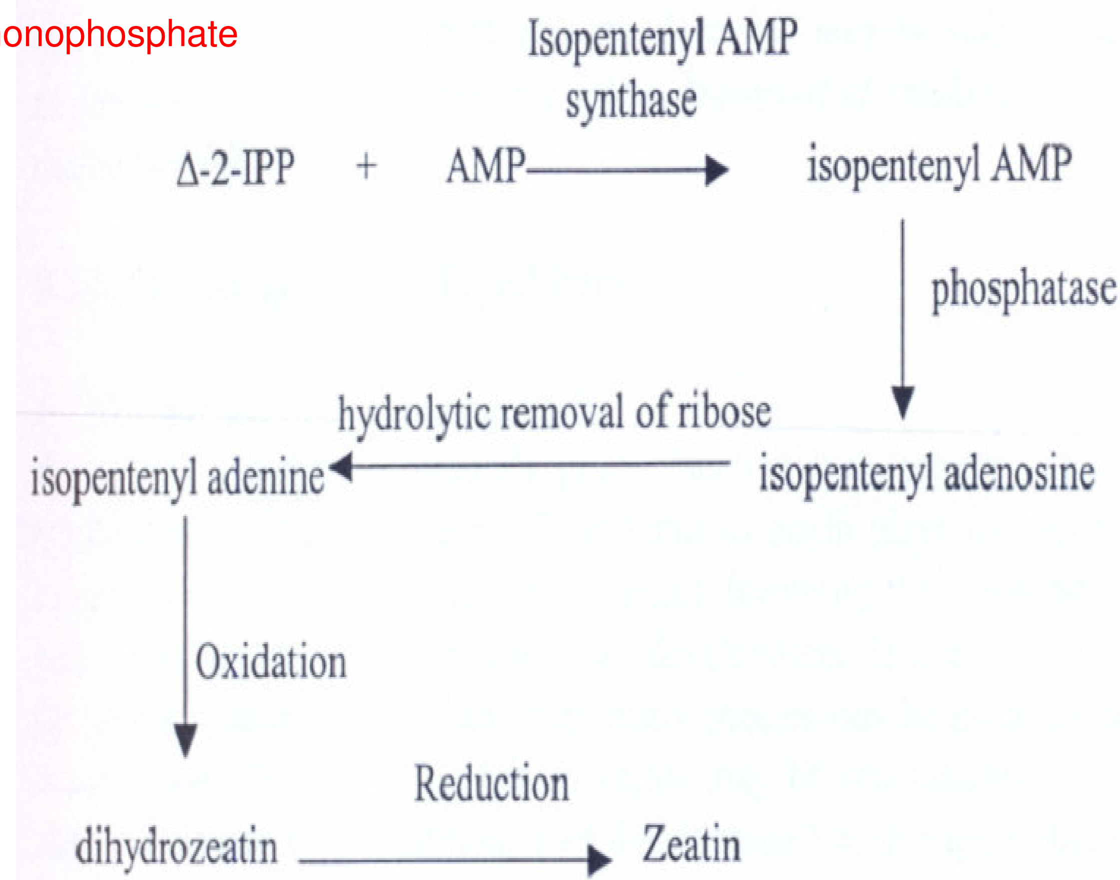
Dihydrozeatin

Biosynthesis of cytokinins

- Biosynthetic pathway of cytokinins

IPP- Isopentenyl pyrophosphate

AMP – adenosine monophosphate



Degradation and inactivation

- (i) Cytokinins are degraded by cytokinin oxidase
- (ii) Cytokinins may be inactivated by forming conjugates

Site of cytokinin synthesis and transport

- Young organs - seeds, fruits, leaves and root tips are sites of cytokinin synthesis
- Root tips are the major sites of cytokinin synthesis & are transported to growing meristems, seeds, fruits and leaves through xylem
- Sieve tubes (phloem) transport limited amount of cytokinins

Physiological role of cytokinins

- **Cell division and organogenesis**
- Cytokinin's prime function is to promote cell division or cytokinesis
- In addition, ratio of cytokinin to auxin plays an important role in organogesis in tissue culture: higher ratios favouring the development of buds, stems and leaves and lower ratios root development
- **Delayed senescence**
- **Lateral bud development**
- **Cell expansion in dicotyledons and leaves**
- **Suppresses the growth of roots**

- Tobacco plants over-expressing the gene for cytokinin oxidase
- The plant on the left is wild type
- The two plants on the right are over-expressing two different constructs of the Arobidopsis gene for cytokinin oxidase: AtCKXJ and AtCKX2
- Shoot growth is strongly inhibited in the transgenic plants

ETHYLENE (C_2H_4)

- **"Certain gases to stimulate fruit ripening"** had been recognized for many centuries
- Russian botanist Dimitry N. Neljubov (1901) observed that dark-grown pea seedlings growing in the laboratory exhibited symptoms that were later termed the triple response: reduced stem elongation, increased lateral growth (swelling), and abnormal, horizontal growth
- When the plants were allowed to grow in fresh air, they regained their normal morphology and rate of growth
- Neljubov identified ethylene, which was present in the laboratory air from coal gas, as the molecule causing the response

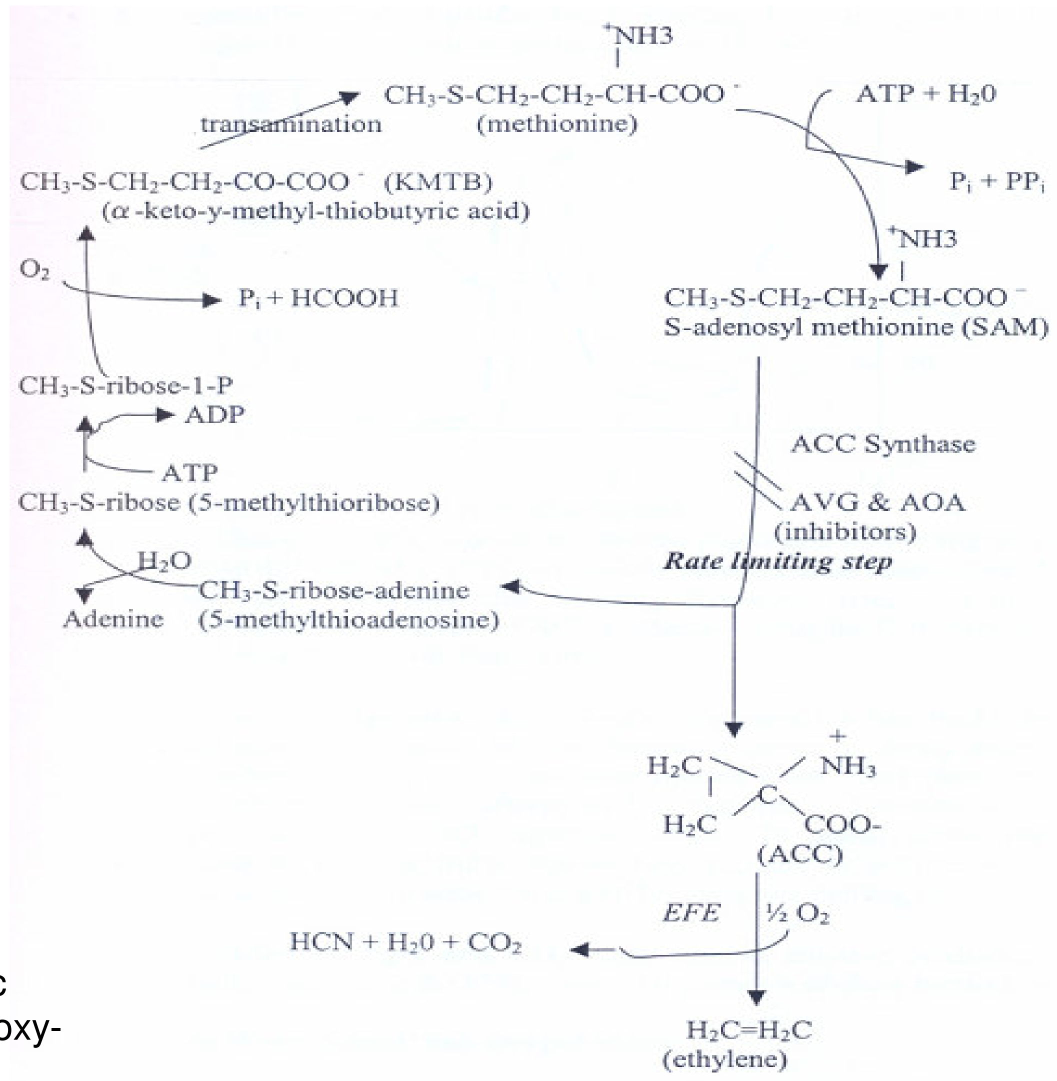
- Triple response of etiolated pea seedlings. Six-day-old pea seedlings were treated with 10 ppm ethylene (right) or left untreated (left). The treated seedlings show a radial swelling, inhibition of elongation of the epicotyl, and horizontal growth of the epicotyl

Biosynthesis of Ethylene

- Ethylene can be produced by almost all parts of higher plants, although the rate of production depends on the type of tissue and the stage of development
- In general, meristematic regions and nodal regions are the most active in ethylene biosynthesis
- Ethylene production also increases during leaf abscission and flower senescence, & during fruit ripening
- Wounding, physiological stresses such as flooding, chilling, disease, and temperature or drought stress induces ethylene production

- Ethylene is biosynthesised through ***ACC (1-amino-cyclopropane- 1-carboxylic acid) pathway*** (Fig below)
- Methionine (amino acid) is the primary precursor of ethylene

ACC (1-amino-cyclopropane-1-carboxylic acid) biosynthetic pathway of ethylene in plant tissues

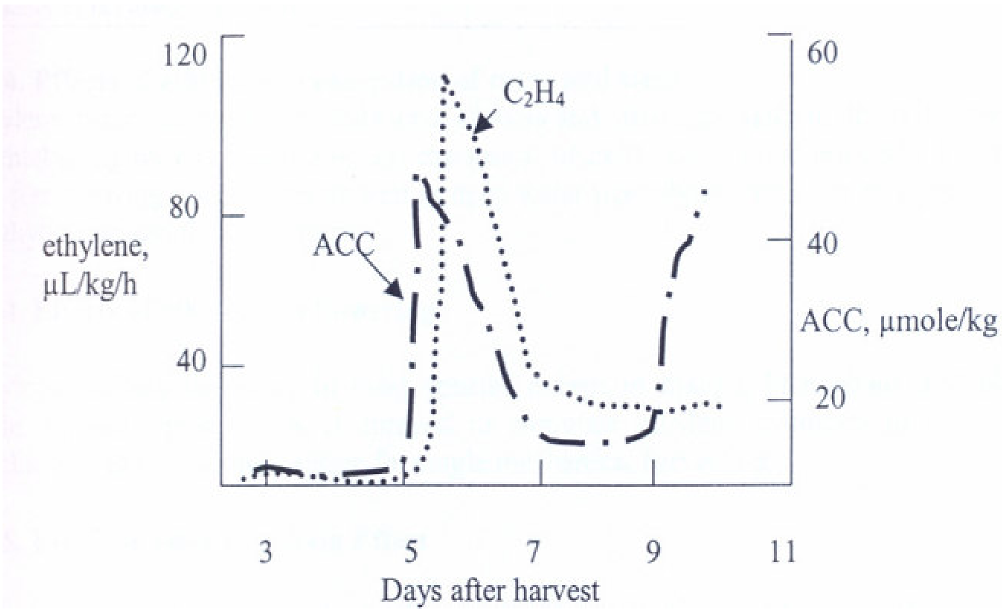


1-amino-cyclopropane-1-carboxylic acid

AOA = aminooxyacetic acid; AVG = aminoethoxy-vinylglycine

Control of Ethylene Biosynthesis

- **IAA induced ethylene production:** IAA induces ethylene production several folds by inducing the synthesis of ACC synthase
- **By wounding:** Wounding also induces ethylene production several folds by inducing the synthesis of ACC synthase.
- **Drought stress:** induces/ accelerates ethylene production
- **ACC controlled ethylene production:** In ripening climacteric fruits, ACC again limits the ethylene production as in Fig. below
- **Regulation by light and CO_2**: Light inhibits ethylene production; whereas CO_2 promotes ethylene production



Physiological roles

- Under water-logged: chlorosis of leaves, decreased stem elongation, increased stem thickness, wilting, epinasty, and/or eventual leaf abscission, decreased root elongation, adventitious root formation, increased susceptibility to pathogens

- Causes inhibition of elongation of roots and stem especially in dicots followed by thickening by enhanced radial expansion of cells
- Inhibits flowering in most species except in mango, bromeliads and pine apple

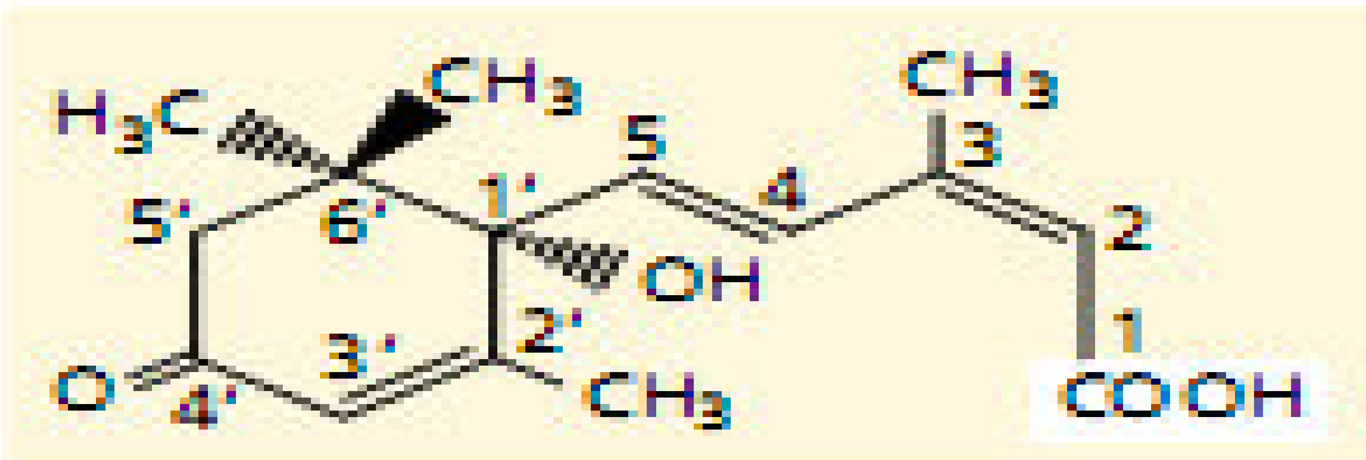
- Epinasty or downward bending of the tomato leaves (right), is caused by ethylene treatment. Epinasty results when the cells on the upper side of the petiole grow faster than those on the bottom

- Ripening of climactric fruits (also in non-climacteric fruits for change of colour of fruit skins)

Climacteric and nonclimacteric fruits	
Climacteric	**Nonclimacteric**
Apple	Bell pepper
Avocado	Cherry
Banana	Citrus
Cantaloupe	Grape
Cherimoya	Pineapple
Fig	Snap bean
Mango	Strawberry
Olive	Watermelon
Peach	
Pear	
Persimmon	
Plum	
Tomato	

ABSCISIC ACID (ABA)

- Abscisic acid (ABA), a growth inhibitor, seed maturation & anti-stress signals
- Naturally occurring structure of ABA
- Tranlocated in the vascular tissue (phloem & xylem)

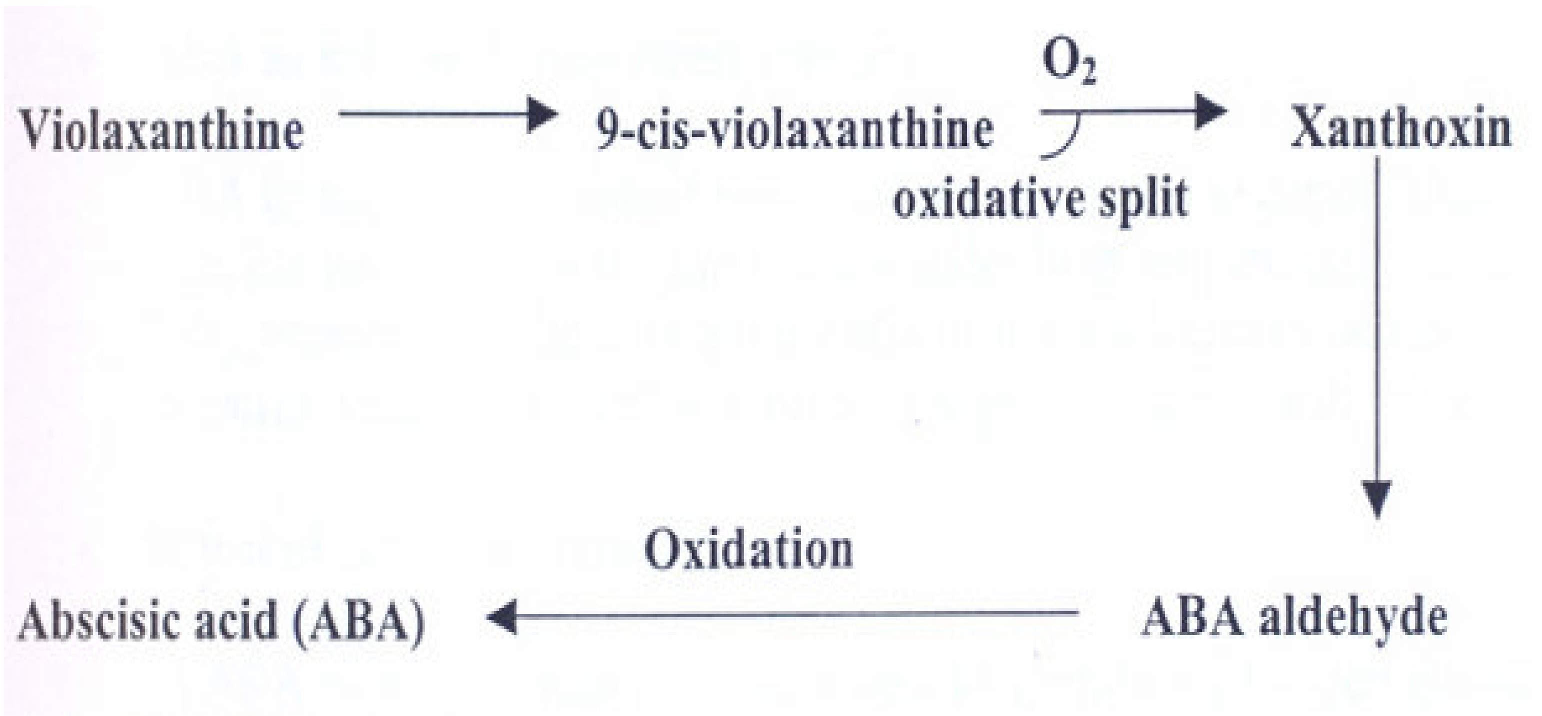


(S)-*cis*-ABA
(naturally occurring active form)

Boisynthesis of ABA

- Biosynthesis of ABA occurs indirectly by degradation of certain (40-C) carotenoids present in plastids — in chloroplast in leaves and in roots, fruits, seed embryos and other non-green plant parts, in other chromoplasts, leucoplasts or proplastids
- A tentative pathway of ABA is depicted in Fig below wherein all reactions up to xanthoxin probably occur in plastids but subsequent reactions in cytosol.

Tentative ABA synthetic pathway



Degradation and/or inactivation

- Inactivation of ABA occurs in two ways:
- (i) By forming ABA-glucose ester wherein carboxyl group of ABA attaches with glucose
- (ii) By oxidation to form phaseic acid or dihydrophaseic acid

Transport

- ABA is transported in/through xylem and phloem as well as also in surrounding parenchyma cells with no polarity

Physiological roles

❖ ABA mediated stomata closure:

- ABA shows dramatic rise (20- to 40- folds; up to 8 fg or 8 x 10^{-15} g/cell) in its content within half an hour of wilting
- Root's ABA sends signal to leaves for stomata closure
- Photosynthesis nearly stops and shoot growth becomes restricted; however, roots continue to grow until they too become dry
- ***ABA causes stomata closure by inhibition of an ATP-dependent H+ pump in plasma membrane of guard cells thus shutthing off K+ influx in guard cells.***

❖ABA as salt- and cold- stress tolerator

- ABA levels increase under saline conditions and also under chilling or freezing temperatures (or occasionally under high temperature conditions too)
- Salinity induces production of low molecular mass proteins called osmotin which accumulates and protect against salt-stress

❖ Effect of ABA on dormancy

- (i) ABA causes dormancy of bud or seeds by inhibiting the cell growth
- (ii) Direct application of ABA on non-dormant bud slows down or stops the growth but does not cause development of bud scales and other features of dormant buds
- (iii) Exogenous ABA has proven potency in causing seed dormancy in some cases. ABA is inhibitor of seed germination slowing down the rate of germination, reducing the elongation rate of radicals and plumules

❖ABA vis-à-vis Abscission

- It was originally isolated as an abscission-causing factor
- However, it has since become evident that ABA stimulates abscission of organs in only a few species and that the primary hormone causing abscission is ethylene
- On the other hand, ABA is clearly involved in leaf senescence, and through its promotion of senescence it might indirectly increase ethylene formation and stimulate abscission

Mode of action:

- (i) Effect on plasma membrane of roots
- (ii) Inhibition of protein synthesis
- (iii) Specific activation and deactivation of certain genes (transcription effects)

Chapter 6. PLANT WATER RELATIONS

❖ Importance of water

- (i). Water is a universal solvent: medium of nutrient uptake and transport, an integral part of protoplasm/cytoplasm constituting >98% wherein all metabolic reactions take place.
- (ii). Water is the reactant and by-product of photosynthesis as under:

$$6\,CO_2 + 12\,H_2O \xrightarrow[\text{Chlorophylls}]{\text{Solar energy}} C_6H_{12}O_6 + 6\,O_2 + 6\,H_2O$$

- (iii). Water keeps cells turgid: open stomata, ererct plant structures and respiration.
- (iv). Water has very high specific heat (4.184 J/g/℃) & thermal conductivity. Hence water absorbs and redistribute large amount of heat energy without correspondingly large fluctuations in temperatures. Hence it has high buffering capacity with respect to temperature fluctuations.

- (v). Unique melting and vaporizing energy of water:
- (a) High heat of fusion (6.0 kJ/mol) required to change the state of water without change in temperature
- (c) High heat of vaporization (~ 44kJ/mol at 25 ℃) and hence gives cooling effect
- vi) Density of water in liquid form is greater than the one in ice form. Hence ice floats on the surface of lakes or ponds; a unique and extremely vital characteristics for aquatic flora and fauna in temperate climates.

Concept of Water Potential

- Water potential (ψ) is the chemical potential of water in a system compared to with the chemical potential of pure water at atmospheric pressure and the same temperature
- The ψ of pure free water at the atmosphere and at a temperature of 298°K (25 °C) is 0 mPa
- ψ may be determined from the sum of hydrostatic pressure (P) and osmotic pressure (π)

$$\psi = P + \pi$$

- The status of water in soil, plants and the atmosphere is commonly described in terms of ψ
- It is measured in mega Pascal (MPa - a pressure unit equivalent to 10 bars)
- Osmotic pressure (ψπ) is the chemical potential of water in a solution due to the presence of dissolved minerals or the reduction in ψ of a system due to solutes.
- It has -ve values and higher concentration of solutes results in lower the water potential (ψw)

- Hydrostatic pressure (ψp) may be -ve or +ve depending on the physical pressure on the water in a system
- For example, water in turgid root cortical cells or leaf mesophyll cells is under +ve turgor pressure exerted against cell wall, whereas water in dead xylem vessels of a rapidly transpiring plant is typically under suction tension (-ve pressure)
- Total water potential (ψw) can be a +ve or -ve value depending on the algebric sum of its components ($\psi \pi + \psi p$)

- When dealing with water potential in soils (ψsw), an additional term matric potential (ψm) is used
- The matric potential (ψm) refers to the force with which water is adsorbed onto the surfaces such as cell wall, soil particles or colloids
- The matric potential (ψm) has always -ve values because forces tend to attract water in place, relative to water in absence of adsorptive surface
- The matric potential (ψm) becomes more -ve as the film of water becomes thinner

- **Water in Soil-Plant-Atmosphere Continuum**
- **Water in soil**
- Water moves in soil along the gradient i.e. from regions of higher to that of lower free energy
- Water movement may be lateral, upward or downward
- Movement of water in soil comprises three phases:
- (i) infiltration involving filling of all pore spaces until saturation
- (ii) redistribution and equilibrium allowing percolation/drainage of excess water along gravity (gravitational water) and retention of water in micro pores (field capacity) against gravity
- (iii) withdrawal due to evapo-transpiration

- Soil water is available to plant between two limits **Field capacity and Permanent Wilting Point** in soil
- **Field capacity** refers to the upper most limit of available water in soil and is the water content of soil after the drainage of gravitational water
- Soil water tension at field capacity varies from 0.1 to 0.3 atm depending upon soil types

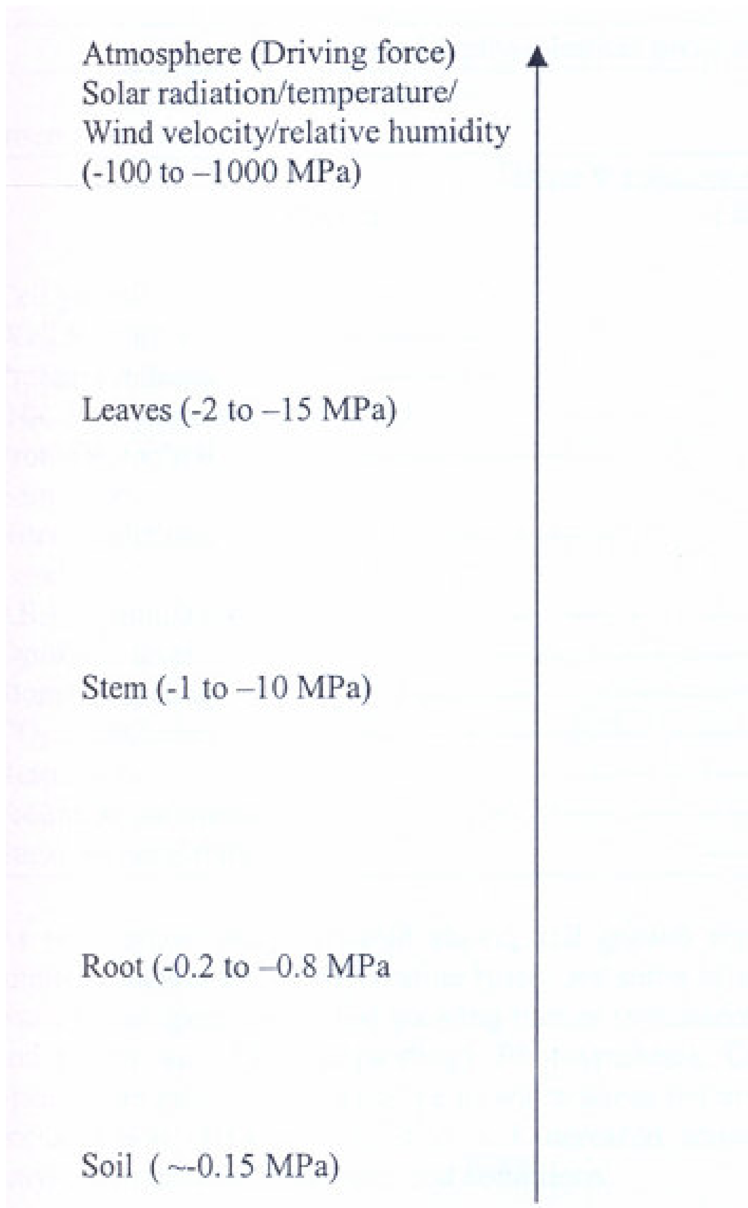
- **Permanent wilting point** is the water content of soils at which plants can no longer extract water from soil resulting into irreversible wilting of plants
- Soil water tension at permanent wilting point ranges from 7 to 32 atm depending upon soil types but normally 15 atm tension correspond to the Soil water content at permanent wilting point
- **Available soil water** lies between **Field Capacity (FC) and Permanent Wilting Point (PWP)**

Soil-water-plant-atmosphere continuum

More -ve

Direction of water flow

Less -ve

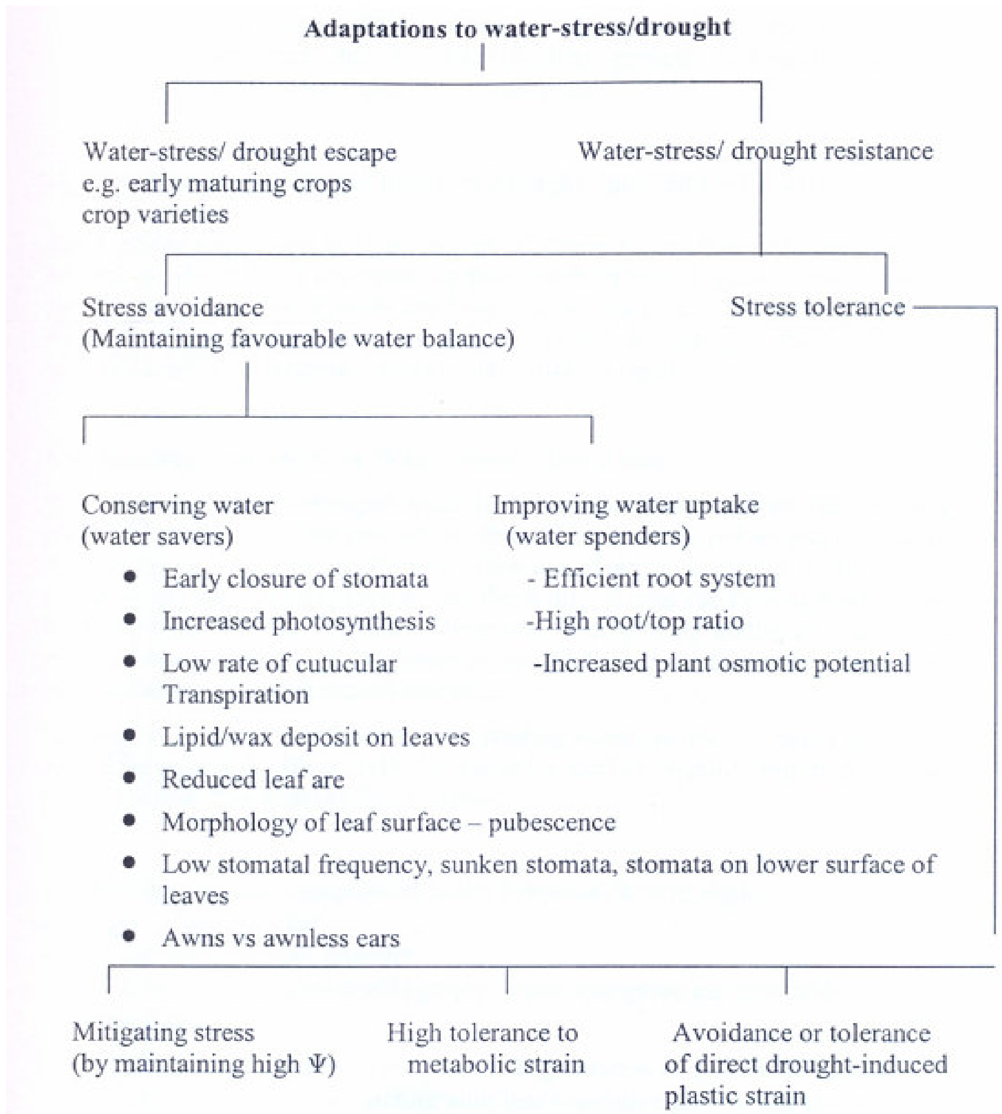


Some of the physiological effects of water-stress in crop plants are summarized below

	Metabolic/physiological processes sensitive to stress
	Very sensitive — Relatively insensitive
	Tissue Ψ required to affect: 0 MPa — -1.0 MPa — -2.0MPa
Cell growth	---------- - - - - -
Wall Synthesis	------------------
Protein synthesis (NR, Phenylalanin Lyase, PAL)	------------------
Protochlorophyll Formation	--------------------
Nitrate reductase Level	--------------------
ABA accumulation	- - - ---------------------
Cytokinin level	-----------------------------------
Stomatal opening	- - - -- - - - -
CO_2 assimilation	- - - -- - - - -
Respiration	- - - ---------------------------
Proline accumulation	- - - - ----------------------------
Sugar accumulation	---------------------------------

- Differential resistance/tolerance to soil water deficit is genetic:
- (i) Rooting characteristics: extensive root growth with large surface area and root hairs and indeterminate root growth habits.
- (ii) Leaf surface with modifications to reduce or minimize the transpiration loss e.g. reduced leaf area, low stomata no., sunken stomata, waxy cuticles, hairy leaves and leaf anatomy modifications
- (iii) C4 vs C3 plants: In general, C4 plants are more efficient and tolerant to water stress

Crop plants adaptations to water-stress or dry conditions



Chapter 7. PHOTO-PHYSIOLOGY

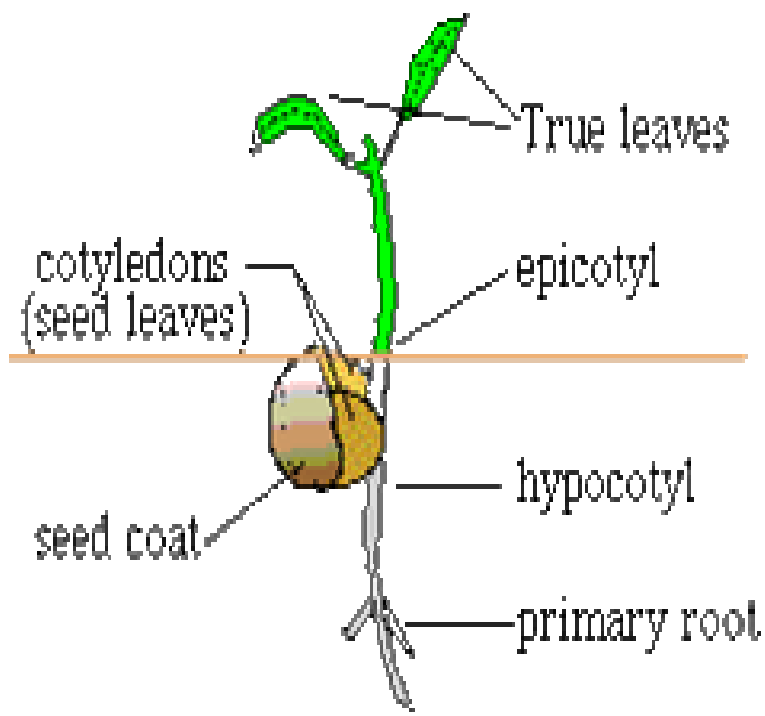
Photomorphogenesis

- Light is an important factor controlling growth and development in plants
- Light causes numerous effects on plant growth and development independent of photosynthesis
- Photomorphogenesis is the control of growth and morphology (shape, flowering, etiolation, *etc.*) by light
- It is mediated by several light receptors: PSII, cryptochrome, phototropin, zeaxanthin and phytochrome

- **Photoreceptors**
- Currently four types of photoreceptors are known to absorb light causing photomorphogenesis. Note that Light Harvest Complex I & II also absorb light during light reaction of the photosynthesis. Four types of photoreceptors are:
- (i) **Phytochromes** absorb red and far-red lights strongly. Phytochromes also absorb blue light. Two major types of phytochromes are known, isolated and characterized.
- (ii) **Cryptochromes** are a group of similar unidentified pigments that absorb blue light and long wave light (UV-A region 320 — 400 nm wave length). Photoreceptor cryptochromes are named because of their special importance in cryptograms (non-flowering plants).
- (iii) **UV-B photoreceptors**: One or more unidentified compounds (technically not pigments) absorb UV radiation of 280 — 320 nm wavelength.
- (iv) **Protochlorophyllides a** absorb red and blue light and are reduced to chlorophyll a.

- Photomorphogenetic light initially causes a small change in cells, but this change is somehow amplified greatly so that large morphological events occur. These events vary with the kinds of cells involved, their position in the plant, and their age. Many genes must eventually become activated, and others deactivated during photomorphogenesis.

- On the contrary some effects are not easily detected. In such cases, light in low doses sometimes triggers initial developmental processes resulting into visible effect later. Still in other cases, rather triggering initial development process, light serves as modulator but in both cases small amount of light energy is involved. Whatever the case, light initially cause small changes in target cells and these changes are somehow amplified greatly causing large morphological events. These events vary with the kind of cells involved, their position in plants and their age. in many cases (not in all cases) gene activation represents part of amplification process. For light to control development of plant, plants absorb light with the help of photoreceptors causing structural developments called photomorphogenesis


True leaves
cotyledons
(seed leaves)
epicotyl
hypocotyl
seed coat
primary root