

20. PLANT GROWTH REGULATORS

Plant growth regulators or phytohormones are organic substances produced naturally in higher plants, controlling growth or other physiological functions at a site remote from its place of production and active in minute amounts. Thimmann (1948) proposed the term *Phyto hormone* as these hormones are synthesized in plants. *Plant growth regulators* include auxins, gibberellins, cytokinins, ethylene, growth retardants and growth inhibitors. Auxins are the hormones first discovered in plants and later gibberellins and cytokinins were also discovered.

Hormone

An endogenous compound, which is synthesized at one site and transported to another site where it exerts a physiological effect in very low concentration. But ethylene (gaseous nature), exert a physiological effect only at a near a site where it is synthesized.

Classified definition of a hormone does not apply to ethylene.

Plant growth regulators

- Defined as organic compounds other than nutrients, that affects the physiological processes of growth and development in plants when applied in low concentrations.
- Defined as either natural or synthetic compounds that are applied directly to a target plant to alter its life processes or its structure to improve quality, increase yields, or facilitate harvesting.

Plant Hormone

When correctly used, is restricted to naturally occurring plant substances, there fall into five classes. Auxin, Gibberellins, Cytokinin, ABA and ethylene. Plant growth regulator includes synthetic compounds as well as naturally occurring hormones.

Plant Growth Hormone

The primary site of action of plant growth hormones at the molecular level remains unresolved.

Reasons

- Each hormone produces a great variety of physiological responses.
- Several of these responses to different hormones frequently are similar.
- The response of a plant or a plant part to plant growth regulators may vary with the variety of the plant.
- Even a single variety may respond differently depending on its age, environmental conditions and physiological state of development (especially its natural hormone content) and state of nutrition. There are always exceptions for a general rule suggesting the action of a specific growth regulator on plants.
- There are several proposed modes of action in each class of plant hormone, with substantial arguments for and against each mode.

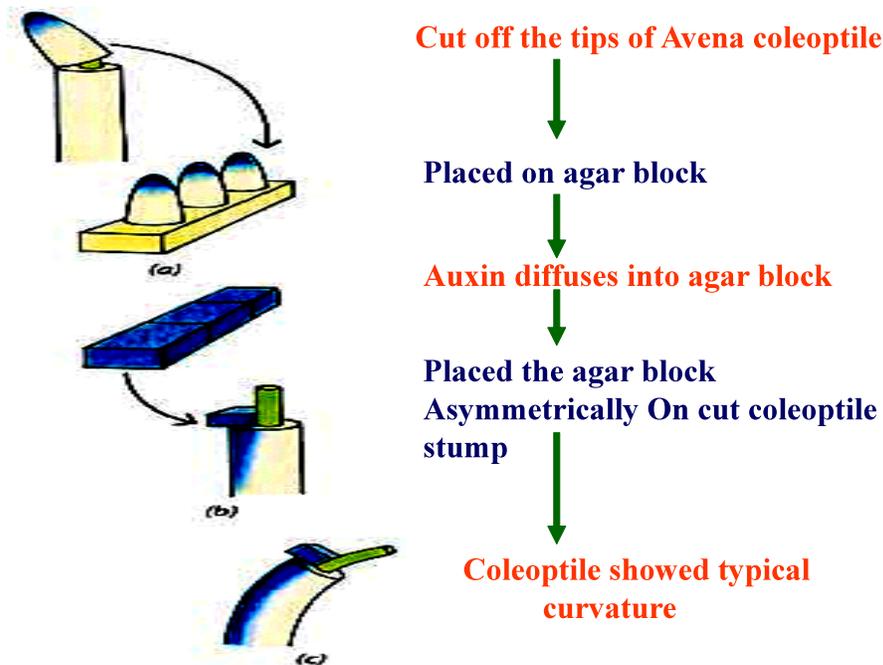
Hormone groups

- | | | |
|--------------|---|--|
| Auxin | - | Substances generally resembles IAA and has the ability to stimulate the elongation of coleoptiles. |
| Gibberellins | - | are diterpenoids, which have the ability to elongate the stem of green seedlings especially certain dwarf and rosette types. |
| Cytokinin | - | Usually substituted Adenines, which resembles zeatin (Naturally occurring cytokinin in <i>Zea mays</i>) and have the ability to stimulate cytokinesis in cultures of tobacco cells. |
| Ethylene | - | Gaseous regulator that stimulate is diametric growth in the apices of dicot seedlings. |
| Inhibitors | - | are regulators of growth, which originally depress the |

Auxins

Auxins are a group of phytohormones produced in the shoot and root apices and they migrate from the apex to the zone of elongation. Auxins promote the growth along the longitudinal axis of the plant and hence the name (auxein : to grow). The term, auxin was introduced by Kogl and Haagen- Smit (1931). Went (1928) isolated auxin from the *Avena coleoptile* tips by a method called *Avena coleoptile or curvature test* and concluded that no growth can occur without auxin. Auxins are widely distributed through out the plant however, abundant in the growing tips such as coleoptile tip, buds, root tips and leaves. Indole Acetic Acid (IAA) is the only naturally occurring auxin in plants. The synthetic auxins include,

Avena Curvature Test



IBA : Indole Butyric Acid

NAA : Naphthalene Acetic acid

MENA: Methyl ester of Naphthalene acetic acid

MCPA: 2 Methyl 4 chloro phenoxy acetic acid

TIBA : 2, 3, 5 Tri iodo benzoic acid

2, 4-D : 2, 4 dichloro phenoxy acetic acid

2, 4, 5-T: 2, 4, 5 – Trichloro phenoxy acetic acid

Natural auxins may occur in the form of either *free auxins*- which freely move or diffuse out of the plant tissues readily or *bound auxins*- which are released from plant tissues only after hydrolysis, autolysis or enzymolysis.

Physiological effects of auxin

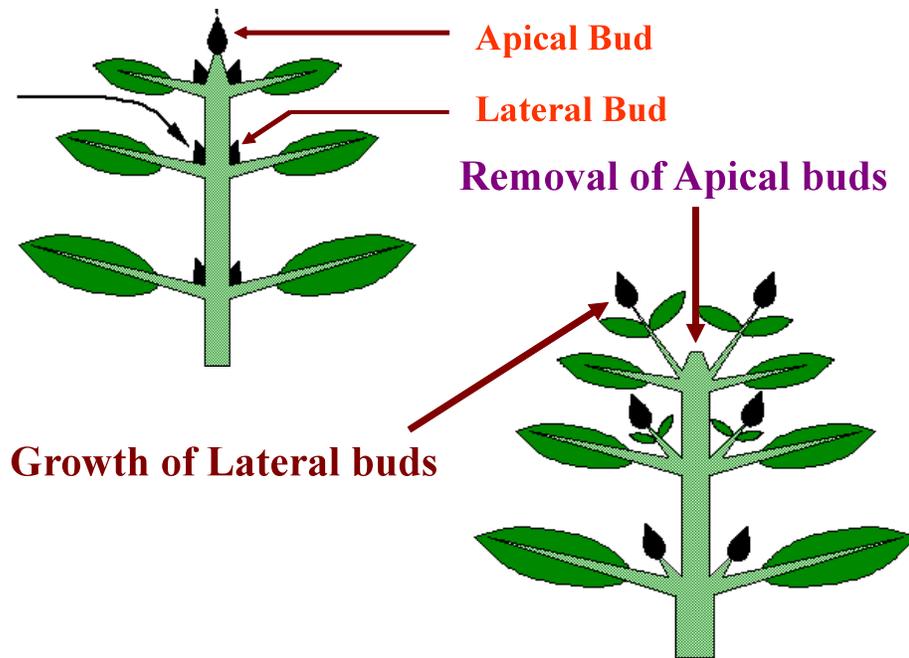
1. Cell division and elongation

The primary physiological effects of auxin are cell division and cell elongation in the shoots. It is important in the secondary growth of stem and differentiation of xylem and phloem tissues.

2. Apical dominance

In many plants, if the terminal bud is intact and growing, the growth of lateral buds just below it remains suppressed. Removal of the apical bud results in the rapid growth of lateral buds. This phenomenon in which the apical bud dominates over the lateral buds and does not allow the lateral buds to grow is known as *apical dominance*.

Skoog and Thimmann (1948) pointed out that the apical dominance might be under the control of auxin produced at the terminal bud and which is transported downward through the stem to the lateral buds and hinders the growth. They removed the apical bud and replaced it with *agar* block. This resulted in rapid growth of lateral buds. But when they replaced the apical bud with agar block containing auxin, the lateral buds remained suppressed and did not grow.



3. Root initiation

In contrast to stem, the higher concentration of auxin inhibits the elongation of roots but the number of lateral roots is considerably increased i.e., higher concentration of auxin induces more lateral branch roots. Application of IAA in lanolin paste (lanolin is a soft fat prepared from wool and is good solvent for auxin) to the cut end of a young stem results in an early and extensive rooting. This fact is of great practical importance and has been widely utilized to promote root formation in economically useful plants which are propagated by cuttings.

4. Prevention of abscission

Natural auxins prevent the formation of abscission layer which may otherwise result in the fall of leaves, flowers and fruits.

5. Parthenocarpy

Auxin can induce the formation of parthenocarpic fruits (fruit formation without pollination and fertilization). In parthenocarpic fruits, the concentration of auxin in the ovaries is higher than in the ovaries of plants which produce fruits only after fertilization. In

the later cases, the concentration of the auxin in ovaries increases after pollination and fertilization.

6. Respiration

Auxin stimulates respiration and there is a correlation between auxin induced growth and respiration. Auxin may increase the rate of respiration indirectly through increased supply of ADP by rapidly utilizing ATP in the expanding cells.

7. Callus formation

Besides cell elongation, auxin may also be active in cell division. In many tissue cultures, where the callus growth is quite normal, the continued growth of such callus takes place only after the addition of auxin.

8. Eradication of weeds

Some synthetic auxins especially 2, 4- D and 2, 4, 5-T are useful in eradication of weeds at higher concentrations.

9. Flowering and sex expression

Auxins generally inhibit flowering but in pine apple and lettuce it promotes uniform flowering.

Distribution of auxin in plants

In plants, auxin (IAA) is synthesized in growing tips or meristematic regions from where; it is transported to other plant parts. Hence, the highest concentration of IAA is found in growing shoot tips, young leaves and developing auxiliary shoots. In monocot seedling, the highest concentration of auxin is found in coleoptile tip which decreases progressively towards its base.

In dicot seedlings, the highest concentration is found in growing regions of shoot, young leaves and developing auxiliary shoots. Within the plants, auxin may present in two forms. i.e., *free auxins* and *bound auxins*. Free auxins are those which are easily extracted by various organic solvents such as diethyl ether. Bound auxins on the other hand, need more drastic methods such as hydrolysis, autolysis, enzymolysis etc. for extraction of auxin. Bound

auxins occur in plants as complexes with carbohydrates such as glucose, arabinose or sugar alcohols or proteins or amino acids such as aspartate, glutamate or with inositol.

Biosynthesis of auxin (IAA) in plants

Thimann (1935) found that an amino acid, tryptophan is converted into Indole 3 acetic acid. Tryptophan is the primary precursor of IAA in plants. IAA can be formed from tryptophan by two different pathways.

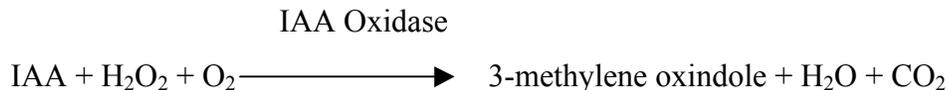
1. By deamination of tryptophan to form indole-3-pyruvic acid followed by decarboxylation to form indole-3-acetaldehyde. The enzymes involved are tryptophan deamination and indole pyruvate decarboxylase respectively.
2. By decarboxylation of tryptophan to form tryptamine followed by deamination to form indole-3-acetaldehyde and the enzymes involved are tryptophan decarboxylase and tryptamine oxidase respectively. Indole 3-acetaldehyde can readily be oxidized to indole 3-acetic acid (IAA) in the presence of indole 3-acetaldehyde dehydrogenase.

Transport of auxin in plant

The transport of auxin is predominantly polar. In stems, polar transport of auxin is basipetal i.e., it takes place from apex towards base. Polar transport of auxin is inhibited by 2, 3, 5 Triiodobenzoic acid (TIBA) and Naphthyl thalamic acid (NPA). The substances are called as antiauxins.

Destruction / Inactivation of auxin in plants

Auxin is destroyed by the enzyme IAA oxidase in the presence of O₂ by oxidation.



Rapid inactivation may also occur by irradiation with x-rays and gamma rays. UV light also reduces auxin levels in plants. Inactivation or decomposition of IAA by light has been called as photo oxidation.

Mechanism of Action

IAA increases the plasticity of cell walls so that the cells stretch easily in response to turgor pressure. It has been suggested that IAA acts upon DNA to influence the production of mRNA. The mRNA codes for specific enzymes responsible for expansion of cell walls. Recent evidences indicate that IAA increases oxidative phosphorylation in respiration and enhanced oxygen uptake. The growth stimulation might be due to increased energy supply and it is also demonstrated that auxin induces production of ethylene in plants.

Gibberellins

Discovery

A Japanese scientist Kurosawa found that the rice seedlings infected by the fungus *Gibberella fujikuroi* grow taller and turned very thin and pale. An active substance was isolated from the infected seedlings and named as Gibberellin.

Biosynthesis of gibberellins in plants

The primary precursor for the formation of gibberellins is acetate.

Acetate + COA → Acetyl COA → Mevalonic acid → MA pyrophosphate → Isopentanyl pyrophosphate → Geranyl pyrophosphate → GGPP → Kaurene → Gibberellins.

Physiological effects of gibberellins

1. Seed germination

Certain light sensitive seeds eg. Lettuce and tobacco show poor germination in dark. Germination starts vigorously if these seeds are exposed to light or red light. This requirement of light is overcome if the seeds are treated with gibberellic acid in dark.

2. Dormancy of buds

In temperate regions the buds formed in autumn remain dormant until next spring due to severe cold. This dormancy of buds can be broken by gibberellin treatments. In potato also, there is a dormant period after harvest, but the application of gibberellin sprouts the tuber vigorously.

3. Root growth

Gibberellins have little or no effect on root growth. At higher concentration, some inhibition of root growth may occur. The initiation of roots is markedly inhibited by gibberellins in isolated cuttings.

4. Elongation of internodes

The most pronounced effect of gibberellins on the plant growth is the elongation of the internodes. Therefore in many plants such as dwarf pea, dwarf maize etc gibberellins overcome the genetic dwarfism.

5. Bolting and flowering

In many herbaceous plants, the early period of growth shows rosette habit with short stem and small leaves. Under short days, the rosette habit is retained while under long days bolting occurs i.e. the stem elongates rapidly and is converted into polar axis bearing flower primordia. This bolting can also be induced in such plants by the application of gibberellins even under non-inductive short days.

In *Hyoscyamus niger* (a long day plant) gibberellin treatment causes bolting and flowering under non-inductive short days. While in long day plants the gibberellin treatment usually results in early flowering. In short day plants, its effects are quite variable. It may either have no effect or inhibit or may activate flowering.

6. Parthenocarpy

Germination of the pollen grains is stimulated by gibberellins; likewise, the growth of the fruit and the formation of parthenocarpic fruits can be induced by gibberellin treatment. In many cases, eg. pome and stone fruits where auxins have failed to induce parthenocarpy, the gibberellins have proven to be successful. Seedless and fleshly tomatoes and large sized seedless grapes are produced by gibberellin treatments on commercial scale.

7. Synthesis of the enzyme α - amylase

One important function of gibberellins is to cause the synthesis of the enzyme α - amylase in the aleurone layer of the endosperm of cereal grains during germination. This enzyme brings about hydrolysis of starch to form simple sugars which are then translocated to growing embryo to provide energy source.

Distribution of gibberellins in plant

Gibberellins are found in all parts of higher plants including shoots, roots, leaves, flower, petals, anthers and seeds. In general, reproductive parts contain much higher concentrations of gibberellins than the vegetative parts. Immature seeds are especially rich in gibberellins (10-100 mg per g fresh weight).

In plants, gibberellins occur in two forms free gibberellins and bound gibberellins. Bound gibberellins usually occur as gibberellin – glycosides.

CYTOKININS (Kinetin)

Kinetin was discovered by Skoog and Miller (1950) from the tobacco pith callus and the chemical substance was identified as 6-furfuryl aminopurine. Because of its specific effect on *cytokinesis* (cell division), it was called as cytokinins or kinetin. The term, cytokinin was proposed by Letham (1963). Fairley and Kingour (1966) used the term, *phytokinins* for cytokinins because of their plant origin. Chemically cytokinins are kinins and they are purine derivatives.

Cytokinins, besides their main effect on cell division, also regulate growth and hence they are considered as natural plant growth hormones. Some of the very important and commonly known naturally occurring cytokinins are Coconut milk factor and Zeatin. It was also identified that cytokinin as a constituent of t-RNA.

Naturally occurring cytokinins

Cytokinins can be extracted from coconut milk (liquid endosperm of coconut), tomato juice, flowers and fruits of *Pyrus malus*; fruits of *Pyrus communis* (Pear), *Prunus cerasiferae* (plum) and *Lycopersicum esculentum* (bhendi); Cambial tissues of *Pinus radiata*, *Eucalyptus regnans* and *Nicotiana tabacum*; immature fruits of *Zea mays*, *Juglans* sp. and *Musa* sp; female gametophytes of *Ginkgo biloba*; fruitlets, embryo and endosperms of *Prunus persica*; seedling of *Pisum sativum*; root exudates of *Helianthus annuus* and tumour tissues of tobacco. According to Skoog and Armstrong (1970), at least seven well established types of cytokinins have been reported from the plants.

Biosynthesis

It is assumed that cytokinins are synthesised as in the case of purines in plants (nucleic acid synthesis). Root tip is an important site of its synthesis. However, developing seeds and cambial tissues are also the site of cytokinin biosynthesis. Kende (1965) reported that cytokinins move upwards perhaps in the xylem stream. However, basipetal movement in petiole and isolated stems are also observed. Seth *et al* (1966) found that auxin enhances kinetin movement (translocation) in bean stems.

Physiological effects of cytokinins

1. Cell division

The most important biological effect of kinetin on plants is to induce cell division especially in tobacco pith callus, carrot root tissue, soybean cotyledon, pea callus etc.

2. Cell enlargement

Like auxins and gibberellins, the kinetin may also induce cell enlargement. Significant cell enlargement has been observed in the leaves of *Phaseolus vulgaris*, pumpkin cotyledons, tobacco pith culture, cortical cells of tobacco roots etc.

3. Concentration of apical dominance

External application of cytokinin promotes the growth of lateral buds and hence counteracts the effect of apical dominance

4. Dormancy of seeds

Like gibberellins, the dormancy of certain light sensitive seeds such as lettuce and tobacco can also be broken by kinetin treatment.

5. Delay of senescence (Richmand - Lang effect)

The senescence of leaves usually accompanies with loss of chlorophyll and rapid breakdown of proteins. Senescence can be postponed to several days by kinetin treatment by improving RNA synthesis followed by protein synthesis.

Richmand and Lang (1957) while working on detached leaves of *Xanthium* found that kinetin was able to postpone the senescence for a number of days.

6. Flower induction

Cytokinins can be employed successfully to induce flowering in short day plants.

7. Morphogenesis

It has been shown that high auxin and low kinetin produced only roots whereas high kinetin and low auxin could promote formation of shoot buds.

8. Accumulation and translocation of solutes

Plants accumulate solutes very actively with the help of Cytokinin and also help in solute translocation in phloem.

9. Protein synthesis

Osborne (1962) demonstrated the increased rate of protein synthesis due to translocation by kinetin treatment.

10. Other effects

Cytokinins provide resistance to high temperature, cold and diseases in some plants. They also help in flowering by substituting the photoperiodic requirements. In some cases, they stimulate synthesis of several enzymes involved in photosynthesis.

11. Commercial applications

Cytokinins have been used for increasing shelf life of fruits, quickening of root induction and producing efficient root system, increasing yield and oil contents of oil seeds like ground nut.

Ethylene

Ethylene is the only natural plant growth hormone exists in gaseous form.

Important physiological effects

1. The main role of ethylene is it hastens the ripening of fleshy fruits eg. Banana, apples, pears, tomatoes, citrus etc.
2. It stimulates senescence and abscission of leaves
3. It is effective in inducing flowering in pine apple
4. It causes inhibition of root growth
5. It stimulates the formation of adventitious roots
6. It stimulates fading of flowers
7. It stimulates epinasty of leaves.

Abscisic acid

Addicott (1963) isolated a substance strongly antagonistic to growth from young cotton fruits and named Abscissin II. Later on this name was changed to Abscisic acid. This substance also induces dormancy of buds therefore it also named as Dormin.

Abscisic acid is a naturally occurring growth inhibitor.

Physiological effects

The two main physiological effects are

1. Geotropism in roots
2. Stomatal closing
3. Besides other effects

1. Geotropism in roots

Geotropic curvature of root is mainly due to translocation of ABA in basipetal direction towards the root tip.

2. Stomatal closing

ABA is synthesized and stored in mesophyll chloroplast. In response to water stress, the permeability of chloroplast membrane is lost which results in diffusion of ABA out of chloroplast into the cytoplasm of the mesophyll cells. From mesophyll cells it diffuses into guard cells where it causes closing of stomata.

3. Other effects

- i. Including bud dormancy and seed dormancy
- ii. Includes tuberisation
- iii. Induces senescence of leaves fruit ripening, abscission of leaves, flowers and fruits
- iv. Increasing the resistance of temperate zone plants to frost injury.

Growth retardants

There is no. of synthesis compounds which prevent the gibberellins from exhibiting their usual responses in plants such as cell enlargement or stem elongation. So they are called as anti gibberellins or growth retardants. They are

1. Cycocel (2- chloroethyl trimethyl ammonium chloride (CCC)
2. Phosphon D – (2, 4 – dichlorobenzyl – tributyl phosphonium chloride)
3. AMO – 1618
4. Morphactins

5. Maleic hydrazide