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Role of GA3 in Commercial Flower Crops

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A B S T R A C T

Plant hormones are a group of naturally occurring, organic substances which influence physiological processes at low concentrations. There are six classical groups of plant hormones includes auxins, gibberellins, cytokinins, ethylene, abscisic acid and brassinosteroids which occur naturally. The use of plant hormones in ornamental crops is more prevalent than in edible crops. The most common types of hormones used are gibberellins, ethylene and their antagonists. Gibberellic acids are one among them which are used to enhance stem elongation of many cut flowers and to promote bud break, thus producing more flowering shoots. They are commonly used to promote flowering of long day plants and of autonomous-flowering plants of the *Araceae*. The largest use of plant hormones in ornamental crops is GA antagonists, which are used in pot plant production to achieve more compact and attractive structure and to promote flowering in certain woody ornamentals (Halevy, 1995) Gibberellic acids are known to coordinate and control various phases of growth and development including flowering at optimum concentrations. Exogenously applied GAs act through the alteration in the levels of natural hormone thus modifying the growth and development of flower crops.

Introduction

Plant Hormones

These are organic substances produced naturally in the plants, controlling growth and other physiological functions at a site of remote from its place of production and active in minute concentration. (Thimman, 1948). Plant hormones may be natural or synthetic one.

Plant growth regulators are classified as

Promoters -causes a faster growth. Those are auxin, gibberellin, cytokinin, brassinosteroides.

Inhibitors-Reduce the growth of plants. Ex-ethylene, ABA, jasmonic acid

Retardants-Inhibit the growth by blocking some specific steps in biosynthetic pathway.

Ex-Paclobutrazol, Phoshon D

6 recognized groups of growth regulators…….

These are the most important agents involved in cording the growth of the plant as a whole. Depending upon the plants, they act either singly or in association.

- Auxins –Regulates apical dominance
- Gibberellins –Induces the stem elongation in dwarf plant.
- Cytokinins –Regulates cell division.
- Ethylene –Induces the fruit ripening
- Abscisic acid -Enhances the rate of leaf senescence
- Brassinosteroids –Induces cell differentiation.

History

- The origin of gibberellins can be traced to Japanese plant pathologists who were investigating the causes of the "bakanae" (foolish seedling) disease.
- Symptoms of the disease are pale yellow, elongated seedlings with slender leaves and stunted roots.
- 1898- The first paper on the cause of bakanae was published by Shotaro Hori demonstrated that the symptoms were induced by infection with a fungus belonging to the genus Fusarium, probably *Fusarium heterosporium* Nees.
- In 1912, Sawada published a paper entitled "The Diseases of Crops in Taiwan" in which he suggested that the elongation in rice seedlings infected with bakanae fungus might be due to a stimulus derived from fungal hyphae.
- 1926, Eiichi Kurosawa found that culture filtrates from dried rice seedlings caused marked elongation in rice and other sub-tropical grasses. He concluded that bakanae fungus secretes a chemical that stimulates shoot elongation, inhibits chlorophyll formation and suppresses root growth
- 1930, Wollenweber - named the imperfect stage as *Fusarium moniliforme* (Sheldon) and the perfect stage, *Gibberella fujikuroi* (Saw.) Wr. ("Fujikuroi" and "Saw.") -- Yosaburo Fujikuro and Kenkichi Sawada
- 1935, Teijiro Yabuta and Sumuki, first isolated a crystalline compound from fungal strains (*Gibberella fujikuroi*) provided by Kurosawa.Yabuta named the isolate as gibberellin
- 1955, members of Sumuki group, succeeded in separating the methyl ester of gibberellin A into 3 components and named gibberellins A1, A2, and A3. (Takahashi *et al.,*)
- Gibberellin A3 was found to be identical to gibberellic acid and structure was proposed for this in 1956.
- 1991 it was determined that gibberellic acid was ubiquitous in plantsand are considered to be widespread among angiosperms, gymnosperms, ferns, algae, fungi, and bacteria. (Arteca, 1996)

Nature of Gibberellin

- Terpenoids – compounds made up of 5- carbon isoprenoid building blocks, joined head to tail.
- The GAs is diterpinoids that are formed from four such isoprenoid units.
- All GAs possess either a tetracyclic *ent*-gibberellane skeleton containing 20 carbon atoms (C_{20}-GAs) or a 20-nor-*ent*-gibberellane skeleton which has only 19 carbon atoms (C_{19}-GAs)
- C_{19}-GAs has lost carbon 20 and, in place, possess a five-member lactone bridge that links carbons 4 and 10.
- GA_1, GA_3 and GA_4 - promote vegetative growth, are the most active gibberellins and the most widely used.
- Gibberellic acid - the first gibberellin to be structurally characterized is GA_3.
- A carboxyl group at carbon-7 is a feature of all GAs and is required for biological activity.
- GAs with 3-β-hydroxylation; 3-β, 13-dihydroxylation; 1, 2-unsaturation are generally more active.
- There are 136 known GAs identified from plants, fungi, and bacteria.
- Chemical formula of gybberellic acid - $C_{19}H_{22}O_6$.
- When purified, it is a white to pale-yellow solid.
- Gibberellic acid is a simple gibberellin, a pentacyclic diterpene acid promoting growth and elongation of cells.
- It affects decomposition of plants and helps plants grow if used in small amounts.

Gibberellin Biosynthetic pathway

- The GA biosynthetic pathway can be divided into 3 stages, each residing in different cellular compartment: the plastid, the endoplasmic reticulum, and the cytosol

Stage i) Formation of terpenoid precursors and ent-kaurene in plastids

Stage ii) Oxidations to form GA_{12} and GA_{53} on ER through GA_{12} aldehyde

Stage iii) Formation of all other GAs from GA_{12} or GA_{53} in cytosol

MVA (Mevalonic acid) pathway is a cytosolic pathway. This is the source of IPP for GA biosynthesis in the fungus Gibberella zeae.

MEP is a plastidic pathway occurs in the green parts of plants and algae.

Ex: arabidopsis, IPP used in GA biosynthesis comes

predominantly from IPP synthesised in plastids by this pathway.

Stage i) GGPP is converted via *ent*-copalyl diphosphate (CPP) to the tetracyclic hydrocarbon *ent*-kaurene, which is then modified by sequential oxidations on C-19, C-7 and C-6 to produce GA_{12}-aldehyde.

GA_{12}-aldehyde is formed from *ent*-kaurene via *ent*-kaurenoic acid and *ent*-7_-hydroxykaurenoic acid by the action of cytochrome-P450-dependent mono-oxygenases. The reactions occur on membranes outside the plastid.

Stage ii) Occurs in endoplasmic reticulum Mono-oxygenases also catalyse the further oxidation of GA_{12}-aldehyde at C-7 to give GA_{12} and the 13-hydroxylation of GA_{12} to GA_{53}. These last two intermediates are substrates for the final stage of GA biosynthesis.

Stage iii) Occurs in the cytoplasma. GA12 and GA53 are converted by GA 20-oxidase (GA20ox) to GA_9 and GA_{20}, respectively, by oxidation of C-20 to an aldehyde followed by the removal of this C atom and the formation of a lactone. The bioactive GAs, GA_4 and GA_1, are then formed from GA_9 and GA_{20}, respectively, by the action of GA_3- hydroxylase (GA3ox). In some species, GA_9 and GA_{20} are also converted to GA_7 and GA_3, respectively, via 2, 3-didehydroGA_9 and GA_5.

Gibberellin Transport in Plant

- Non – polar in nature, it does not appear to be polar unlike auxin but it moves along with phloem translocated the organic material according to source and sink relationship. Studies conducted by application of radio labelled Gas either to the stem or coleoptile section. Later it has been detected in both phloem and xylem saps but xylem transportation is not clear. In phloem lateral translocation is seen.
- The GAs are not translocate in pants as free molecules

Probably in bound form as gibberellin-glycosides

Bioassay

Barley Endosperm Test

It is most important and widely used bioassay of gibberellin. By this method a little as 0.2ngm og GA can be detected within 24 hours. Barley aleyoron layer (half seed without containing a embryo) is incubated in a solution of GA for 24 hours. The gibberellin stimulates amylase activity which causes hydrolysis of starch to glucose

Dwarf Maize and Pea Bioassay

Dwarf pea has taken for bioassay, these strains are mutants and are believed to be dwarf because they have neglizible amount of endogenous gibberellins. Application of exogenous gibberellins overcome the dwarfness of the seedlings by increasing the length of the stem.

Gibberellins – Now a day's more sophisticated biochemical techniques are available to detect and quantitate the plant hormone from small amount of tissues. Those are

Effect of environment on Gibberellin Biosynthesis

✓ Light
✓ Temperature

Both alter the levels of active GAs by affecting gene transcription for specific steps in the biosynthesis pathway.

Light quality, quantity and duration can all influence GA biosynthesis and metabolism. And this varies with crop to crop.

Seed germination-GAs induces the production of enzymes that are needed to digest the endosperm, which would otherwise form a mechanical barrier to root emergence.

Ex: Arabidopsis – seed germination is stimulated by exposure to light, which acts to promote production of bioactive Gas.

Physiological Effects of Gibberellins

Stimulates Cell Division

GA affect the cell division *and* cell elongation – It induce cyclin dependent kinase gene expression leading to increase the mitotic activity thus transcription of those genes regulating the transmission from G1 to S and G2 to S. It induces the intercalary meristem.GA may influence the gene expression of an expansin named OsEXP4. It

activates genes responsible for cyclin dependent kinase affecting the cell cycle at G1.

- GA promotes internodal elongation in genetically dwarf mutant. The effect of exogenous GA on wild type (labeled as normal) and dwarf mutant (d1 maize)**.** Gibberellins stimulates dramatic stem elongation in dwarf mutant, but has little or no effect on the tall, wild type plant.
- Rosette species are plants in which the first formed internodes do not elongate under certain growing conditions like growing of long day plant under short day condition. Bolting and flowering can occur either plant treated with GA or exposed to long day condition.

Gibberellins promotes seed germination

GA stimulates the mobilization of nutrient reserves during the germination of seed. GAs is mainly produced by the embryo and its scutellum.

These are released into the endosperm and aleuron layer where they induce the production and secretion of a number of hydrolyzing enzymes and break the reserves in the endosperm.

Stage i) GA moves from the embryo to aleuron layer where it stimulates the synthesis of amylase, protease enzymes.

Stage ii) Amylase and protein together digest the starch to glucose which is metabolize to metabolic demand of the growing embryo.

Gibberellins Induce flowering in ornamental plants

Multiple developmental pathways for flowering in *Arabidopsis*

Photoperiodism, the autonomous (leaf number) and vernalization (low temperature) pathways, the energy (sucrose) pathway, and the gibberellin pathway. The photoperiodic pathway is located in the leaves and involves the production of a transmissible floral stimulus, FT protein. In LDPs such as *Arabidopsis,* FT protein is produced in the phloem in response to CO protein accumulation under long days. It is then translocated via sieve tubes to the apical meristem. In SDPs such as rice, the transmissible floral stimulus Hd3a protein accumulates when the repressor protein, Hd1, is not produced under short days, and the Hd3a protein is translocated via the phloem to the apical meristem.

In *Arabidopsis,* FT binds to FD, and the FT/FD protein complex activates the *AP1* and *SOC1* genes, which trigger *LFY* gene expression. *LFY* and *AP1* then trigger the expression of the floral homeotic genes.

The autonomous (leaf number) and vernalization (low temperature) pathways act in the apical meristem to negatively regulate *FLC,* a negative regulator of *SOC1*. The sucrose and gibberellin pathways, also localized to the meristem, promote *SOC1* expression.

- *Impatiens balsamina* L. treated with GA_3 under continuous illumination. Flower can be seen in the treated plants. The Gibberellin pathway activates the expression of genes involved in flower formation both directly, through the activation of LFY and FT genes, and indirectly, through the positive regulation of SOC1 gene.
- In many conifers, juvenile phase which may last upto 20 years, can be shorten by treatment with GA3 and much younger plants can be induce to enter the reproductive or cone producing phase precociously.

Anti-gibberellins

- Since the 1950s, a number of synthetic compounds known as growth retardants have been developed, also known as anti-gibberellins.
- Found commercial use in the ornamental plants
- ex: potted plants – as foliar spray or soil drench
- It reduce the stem elongation, resulting in plants that are shorter and more compact, with darker green foliage
- Flower size is unaffected as individual growth retardants block specific steps in gibberellin biosynthesis
- AMO-1618 and Phosphon D : inhibit enzyme involved in the synthesis of kaurene
- Ancimidol : blocks the oxidation of kaurene to kaurenoic acid
- BX-112 : blocks the 3β-hydroxylation of GA_{20} to GA_1.

Case study

Experiments are carried out to study the influence of GA3 on growth, flowering and quality of different flower crops.

Table.1 Commercial uses of GA3 in flower crops

Crop	GA_3	Results
Tuberose cv. Prajwal	150 ppm	Enhancing all the vegetative, floral and bulbous characteristics
Marigold cv. Pusa Narangi Gainda	300 ppm	Early flowering and enhanced yield attributes
China aster	200 ppm	Enhancing the growth, early flowering and yield characteristics
Tuberose cv. Single	150 ppm	Increasing results on growth, flowering and flower yield
Gladiolus cv. White Prosperity	25-50 mg L^{-1}	Improving the vase life and quality of gladiolus cut flowers
Cut rose cv. First Red	100 ppm	Enhancing the floral attributes, vase life and also colour development
Tulip	400 ppm	Increasing blooming period and quality bulb production
Annual chrysanthemum	200 ppm	greater results for seed yield, seedling dry weight and seedling vigour index

Figure.1

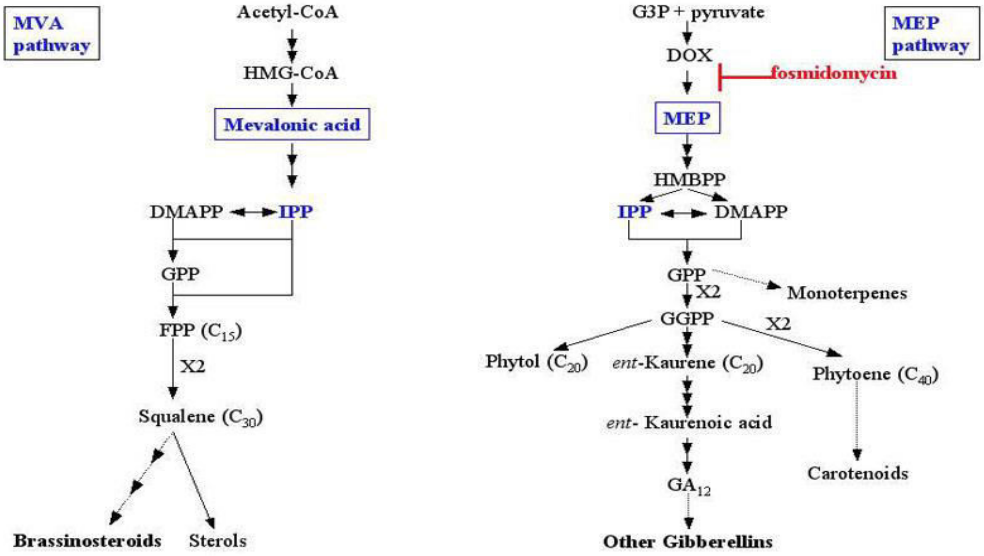
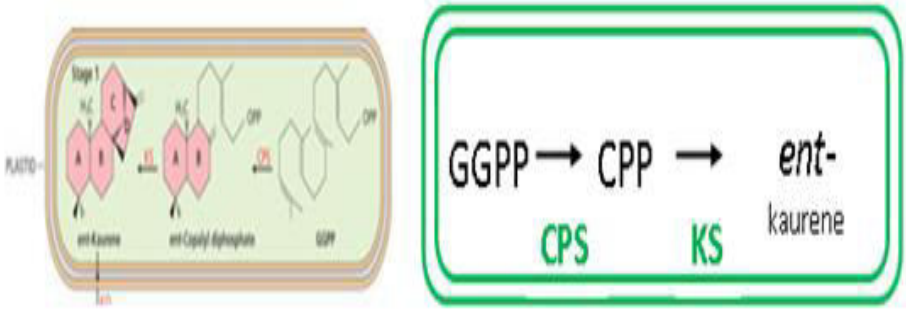


Figure.2



Tuberose

GA3 at 150 ppm proved to be best concentration in enhancing all the vegetative (plant height, number of leaves and sprouting of bulbs), floral (spike length, number of florets/spikes, floret length) and bulbous characteristics in tuberose. GA3 also resulted in early

flowering and more durable flowers which are the major contributing traits for floriculture industries. Better performance of tuberose with application of GA3 might be due to efficient nutrient uptake, enhancing source and sink potential by promoting photosynthetic enzymes, leaf area, more trapping of light for increasing photosynthetic rate, proper metabolism of antioxidant enzymes to normal level. (Rani and Singh, 2013, Haryana)

Marigold

GA 300 ppm resulted in the early flower bud initiation, opening of first flower and maximum duration of flowering, flower stalk length, number of flowers per plant weight of flower, weight of flower per plant and flower yield per hectare followed by GA 200 ppm. (Mithilesh *et al.,* 2014)

Rose

Gibberellic acid at higher concentration of 100 ppm as a pre-harvest spray exerted a significant influence on crop growth and recorded highest mean values for plant height (76.18cm), stalk length (60.98 cm), stem girth (1.66 cm) and total chlorophyll content (1.826 mgg-1). Similarly the application of gibberellic acid at100 ppm level drastically increased the quality traits viz., mean flower diameter (6.89cm), anthocyanin content (0.1970 OD value) and vase life (2.6 days). Likewise the earliest flowering (40.00 days) was also obtained from pre-harvest spray of gibberellic acid at 100 ppm (Muthukumar *et al.,* 2011). GA constituent a group of tetracyclic diterpene, response of the plant to GA3 application is very critical and varies among the plants species, organs, and tissues. It needs to be optimizing the GA concentration to improve all attributes like growth parameters, flowering, and yield, vase life of the flower crops.

Author Contribution

Anuradha R. Wadagave: Investigation, formal analysis, writing—original draft. R. P. Sateesh,: Validation, methodology, writing—reviewing. Deepa U. Pujar:—Formal analysis, writing—review and editing. D. R. Jhanavi: Investigation, writing—reviewing.

Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethical Approval: Not applicable.

Consent to Participate: Not applicable.

Consent to Publish: Not applicable.

Conflict of Interest: The authors declare no competing interests.

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