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RESEARCH ARTICLE

PLANT HORMONES SYNTHESIZED BY MICROORGANISMS AND THEIR ROLE IN BIOFERTILIZER-A REVIEW ARTICLE.

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Abstract

Plant growth promoting rhizobacteria (PGPR) are the soil bacteria inhabiting around/on the root surface and are directly or indirectly involved in promoting plant growth and development via production and secretion of various regulatory chemicals in the vicinity of rhizosphere Various species of bacteria like *Pseudomonas*, *Azospirillum*, *Azotobacter*, *Klebsiella*, *Enterobacter*, *Alcaligenes*, *Arthrobacter*, *Burkholderia*, *Bacillus* and *Serratia* have been reported to enhance the plant growth directly by either assisting in resource acquisition (nitrogen, phosphorus and essential minerals) or modulating plant hormone levels, or indirectly by decreasing the inhibitory effects of various pathogens in the forms of biocontrol agents. The ability to synthesize growth stimulating phytohormones by numerous prokaryotic and eukaryotic microorganisms including numerous soil bacteria and fungi is reviewed, with emphasis on their effect on plant physiology and development. A phytohormone is an organic substance synthesized in defined organs of the plant that can be translocated to other sites, where it triggers specific biochemical, physiological and morphological responses. The commonly recognized classes of phytohormones, regarded as the "classical five", are: the auxins, gibberellins, cytokinins, abscisic acid and ethylene. Several PGPR are reported to produce IAA, gibberellic acid and cytokinins in the rhizospheric soil and thereby play a significant role in increasing the root surface area and number of root tips in many plants. Plant hormones contribute to the coordination of diverse physiological process in plants, including the regulation of quiescence and seed germination, root formation, fluorescence, branching, tillering, and fruit ripening. They increase plant resistance to environmental factors and induce or suppress the expression of genes and the synthesis of enzymes, pigments and metabolites (Arsad and Frankenberger, 1991; Kulaeva and Kuznetsov, 2002).

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Discovery of plant hormones:-

Auxins: The discovery of auxins during the nineteenth century was outcome of experiments on phototropism and geotropism (reviewed by Moore, 1979). In 1880 Charles Darwin reported on the phenomenon by which the plants bent towards the sunlight. In 1926, the Dutch botanist Frits W. Went discovered auxin and described its bioassay for its quantitative detection by "Avena coleoptile curvature test". Although Went had succeeded in isolating auxin, he was not able to purify the active compound to establish its chemical structure. In 1934, the biochemist Kogl Haage-Smit and Erxleben obtained an active substance from urine, indole-3-active acid (IAA), which was found to be identical to auxin. Finally K. V. Thimann isolated IAA from cultures of the fungus *Rhizopus suinus* in 1935.

Gibberellins:-

Gibberellins was discovered by E. Kurosawa in 1926(Moore, 1979). In Japan disease called foolish seedling or bakanne in maize and rice seedling treated with spent-culture medium from fungus *Gibberella fujikuroi* was reported. Yabuta and Sumiki, in 1938 isolated and crystallized two biologically active substances, which they named as gibberellins "A and B". In 1956, gibberellins were shown to be natural compound of the plant tissues both by West and Phinney in USA and by Radley in England. Up to now about 125 different GAs (Gibberellins) have been characterized (Crozier *et al.*, 2001).

Cytokinins:-

The discovery of the cytokinins occurred in 1955,when Skoog isolated a substance called kinetin from an autoclaved sample of DNA, and demonstrated it to be active *in vitro* in promoting mitosis and cell division in tobacco callus tissue. Although kinetin is an artifact derived from 2-deoxyadenylate, its biological activity resembles that of zeatin (Z), a native inducer of plant cell division that was isolated from immature maize seedling 1963.

Ethylene:-

Ethylene is also known as "ripening hormone" was identified some 50 years ago (Burg, 1962). Many soil bacteria code for enzyme aminocyclopropane deaminase (ACC-deaminase) that degrades a key intermediate in ethylene accumulation by plants (Penrose and Glick, 2003).

Abscissic acid:-

Abscissic acid was discovered around 1960 as the hormone causing abscission of fruits and dormancy of buds.

Production and role of phytohormones:-

There are two sources of phytohormones naturally available for the plants: endogenous production by the plant tissues and exogenous production by the associated microorganisms (Kumar and Lonsane, 1989; Arshad and Frankenberger, 1991; Costacurta and Vanderleyden 1995; Patten and Glick 1996). Many plant associated microorganisms are themselves capable of synthesizing phytohormones (given in table 1) which are necessary as mediator in communications between the plant host and its microflora. The ability to form plant hormones is believed to be a major property of rhizospheric, epiphytic and symbiotic bacteria that stimulate and facilitate plant growth called as plant growth promoting rhizobacteria (PGPR) strains (Kameneva and Muronets, 1999; Karvchenko *et al.*, 2004; Suzuki and Oyaizu, 2003). On the other hand, certain free-living microorganisms (i.e. those that form no association with plants in the course of the life cycle) are also capable of synthesizing plant hormones.

Table.1:- Phytohormones produced by plants and microorganisms and their effect on plant morphology and development

Auxin Production

S.no.	Plant endogenous production or Causative agent	Observed effect on plant	Reference
1.	**Plant**		
	Zea mays *Arabidopsis thaliana*	Cell enlargement, root initiation, vascular differentiation and apical dominance	Ostin *et al.*,1999 Bartling *et al.*, 1994
2.	**Fungus**		
	Pisolithus thaliana	Plant growth promotion	Frankenberger and Poth,1987
3.	**Bacteria**		
	Azospirillum	Decrease of root length, increase	Tien *et al.*,1979

	Rhizobium *Bradyrhizobium*	of root hair development	Atzorn *et al.*,1982 Badenosch-Jones *et al.*, 1982
	Klebsiella	Increase in root branching and root surface	El-Kawas and Adachi,1999
	Azospirillum, Gluconobacter, Herbaspirillum	Corn seedling inoculated showed an increase on free active IAA and IBA	Fuentes-Ramirez *et al.*,1993 Bastian *et al.*,1998 Fallik *et al.*,1989
	Pseudomonas syringae pv savastanoi *Agrobacterium* *Erwinia herbicola pv gypsophilae*	Induction of gall and tumor formation	Comai and Kosugue, 1980 Liu *et al.*,1982 Manulis *et al.*,1998
	Cyanobacteria Nostoc	Symbiotic tissue of Gunnera	Sergeeva *et al.*,2002

Gibberellins production

S.no.	Plant endogenous production or Causative agent	Obsereved effect on plant	Reference
1.	**Plant**		
	Arabidopsis Thaliana *Oryza sativa* *Zea mays* *Pisum satvium*	Seed germination development and reproduction of plants, floral development	Kobaysahi *et al.,* 1994 Helliwell *et al.*,2001 Spray *et al.,* 1996
2.	**Fungus**		
	Gibberellia fujikuroi	Bakanae effect in maize, rice and other plants	Rojas *et al*.,2001 Fernadez-Martin *et al*., 1995
3.	Bacteria		
	Azospirllum brasilense *Azospirllum lipoferum* *Azospirllum brasilense*	Reversion of dwarfism in maize and rice Promotion of shoot elongation, growth and root hair density	Cassan *et al*.,2001 Fulchieri *et al*.,1993

Cytokinins production

S.no.	Plant endogenous production or Causative agent	Observed effect on plant	Reference
1.	**Plant**		
	Arabidopsis thaliana	Cell division ,chloroplast differentiation, photosynthesis, senescence and nutrient metabolism	Takei *et al*.,2001
2.	**Bacteria**		
	Azospirillum *Pseudomonas syringe* pv *savastanoi* *Agrobacterium tumefaciense* *Erwinia hericola*	Plant growth promotion Induction of gall and tumor Formation	 Roberto and Kosuge,1995 Lichter *et al*.,1995

Auxins:-

Auxin represents one of the most important plant hormones regulating many aspect of plant growth and development throughout the plant cell cycle from cell division,cell elongation and differentiation to root initiation, apical dominance, tropistic response, flowering ,fruit ripening and senescence (stress to stress factors)

IAA (indole acetic acid) exhibits the greatest activity although plants are known to contain other auxins, most of them also belonging to indole derivatives (structurally similar to IAA). This indole may be precursors or products of the transformation of IAA. The ability to synthesis IAA was detected in many rhizospheric and epiphytic bacteria: *Azospirillum* spp., *Agrobacterium* spp., *Azotobacter* spp. *Alcaligenes* spp., *Entrobacter* spp. *Erwinia* spp., *Acetobacter* spp., *Rhizobium* spp., *Bradyrhizobium* spp., and *Herbasirillum* spp. (Cacciari *et al*., 1989; Datta and Basu, 2000). The formation of IAA is widespread among bacteria of the genera *Pseudomonas, Bacillus* and *Xanthomonas* spp. (Wilkinson *et al*.,1994; Olyunina and Shabaev,1996; Tsavkelova *et al*, 2005; Park *et al*.,2005;Mordukhova *et al*.,1991) as well as Methylobacteria (*Methylobacterium, Methylovorus, Aminobacter* and *Paracoccus*) (Ivanova,2001) Biosynthesis of auxins was noted in *Achromobacter, Flavobacterium, Arthrobacter, Klebisella, Rhodococcus, Mycobacterium, Sphingomonas, Stenotrophomonas,* Micrococuccus (Belimov *et al*.,1999). Many *Streptomycetes* and certain respresentatives of *Archaea (*e.g. thermophylic sp. *Sulfolobus acidocaldaris)* also synthesise IAA(White,1987) The capacity of IAA biosynthesis was found in representatives of free living and symbiotic cyanobacteria of the genera *Nostoc, Chlorogloeopsis, Calothrix, Plectonema, Gloeothece, Anabeana, Cylindrospermum* and *Anabaenopsis.*

Among phototropic eukaryotes IAA formation was noted in algae of the genera *Chlorella, Dunaliella* and *Fucus* (Basu *et al*., 2002) Yeast of the genus *Saccharomyces* and *Micromycetes* belonging to the genera *Fusarium, Rhizoctonia, Rhizopus, Absidia, Aspergillus, Pencillium, Monilia, Phoma, Pythium, Trichoderma* and *Actinomucor* also produce IAA(Gunasekaran and Weber, 1972). Mycorrhizal fungi of the genera *Laccaria, Pisolithus, Amanita, Rhizopogon, Paxillus* and *Hebeloma.* In plant cells IAA is largely formed by *de novo* synthesis from tryptophan, which undergoes either oxidative deamination (via formation of indole-3-pyruvic acid) or decarboxlation (via formation of tryptamine, with indole-3-acetic aldehyde as an intermediate). In case of microorganisms, the known pathways of IAA biosynthesis are also related to tryptophan metabolism.

1. IAA formation via indole -3-pyruvic acid and indole-3-acetic aldehyde is found in majority of microorganism, such as the phytopathogenic bacterium *Erwinia herbicola*, saprophytic species of the genera *Agrobacterium* and *Pseudomonas,* certain representatives of *Bradyrhizobium, Azospirillum, Rhizobium Klebsiella* and *Enterobacter; Methylobacteria* ; the symbiotic nitrogen fixing cyanobacterium *Nostoc* sp. ; the yeast *Saccharomyces uvarum* and phytopathgenic micromycetes of the genera *Fusarium, Rhizoctonia* and *Colletotrichum* (Furukawa,1996; Thakur, and Vyas, 1983)
2. The conversion of tryptophan into indole-3-acetic acid aldehyde may involve an alternative pathway in which tryptamine is formed. This pathway is believed to operate in pseudomonades and azospirilla, anundentified mycorrhizal fungus of orchid (*Ophrys lutea* Cav.) (Barroso, 1986) and the cyanobacterium *Chlorogloea fritschii* (Ahmad, and Winter, 1969).
3. IAA biosynthesis via indole-3-acetamide formation takes place in phytopathogenic bacteria *Agrobacterium tumefaciens, Pseudomonas syringe* and *E. herbicola;* certain streptomycetes; saprophytic pseudomonades *Pseudomonas putida* and *Pseudomonas fluorescence.*
4. IAA biosynthesis that involves tryptophan conversion into indole-3-acetonitrile is found in plant, *Alcaligenes fecalis* and possibly, the cynobacterium *Synechocystis* sp.
5. The tryptophan-independent pathway, more common in plants, is also found in microorganisms (*Azospirillium* and C*ynobacteria*)

However, the contribution of this pathway to IAA biosynthesis is insignificant and the mechanisms are largely unknown. In plant, IAA binds to sugars, amino acids and protein, forming storage (inactive) forms which release the phytohormone when it is needed (the physiological activity is recovered shortly thereafter). Omission of tryptophan from the culture medium decreases the level of IAA synthesis by the culture's microorganisms. Exogenous tryptophan may augment auxin biosynthesis by an order of magnitude or higher, this being the reason why the yield of the phytohormone in the most active strains exceeds 80-100mg IAA per 1 ml culture medium (Tsavkelova *et al*.,2003).

Gibberellins:-

A substance including excessive extension of rice sprouts, first isolated from the phytopathogenic fungus *Fusarium moniliforme* in the 1930s, was given the name gibberellins (after the perfect stages of *Fusarium monilifomra, Gibberella fujikoroi*) (Escamilla *et al*.,1999). Gibberellins, classified with diterpenes, consist of isoprene residues that usually form four rings (A, B, C and D). Gibberellic acids (GAs)**,** GA_3, GA_7, GA_1, and GA_4 are the best studied phytohormones of this group; they exhibit maximum biological activity and are the most widespread in nature. Gibberellins amount to more than 100 compounds, constituting the largest class of phytohormones, which are found

in both plants and microorganisms. Certain compounds are classified with gibberellins based solely on their characteristic biological activity (they have a different structure). First and foremost, gibberellins affect the division and elongation of the cells constituting the intercalary meristem, although stimulation of fluorescence, activation of the synthesis of membranes and amylolytic enzymes, and other effects have also been described. The ability to synthesize gibberellins is inherent in all groups of microorganisms (Badenoch-Jones *et al.*, 1982). Gibberellins are formed by epiphytic and rhizospheric bacteria (representatives of the genera *Azotobacter, Arthrobacter, Azospirillum, Pseudomonas, Bacillus, Acinetobacter, Flavobacterium, Micrococcus, Agrobacterium, Clostridium Rhizobium* and *Xanthomonas*) (Grappelli and Rossi, 1981; Cassan *et al*., 2001).

The Gibberellins (GAs) are complex molecules of tetracarbocyclic diterpernes. GAs numbering is not related to their structure. Molecules, whose structure has been elucidated, are numbered in approximate order of their discovery. There is continuing interest in the biosynthic origin of the GAs since some of them have important activities in plants. The most important GA in plant is GA_1, primarily responsible for stem elongation. In *Gibberellia,* GAs biosynthesis is catalyzed by enzymes falling into three classes: terpene cyclases catalyze the synthesis of ent-kaurene from geranylgernayl diophosphate; cytochrome P450 monooxygenase catalyze the steps of the pathway from ent-kaurene to GA_{12}; and soluble dioxynenase catalyze the final steps of the pathway.
The ability of *Azospirillum lipoferum* and *Azospirillum brasilense* to metabolize $GA_{20}GA_1$ in rice (Oryza sativa L.) seedlings suggests that an enzyme similar to that operating in plants (2-oxyglutarate-dependent dioxygenase) is involved in gibberellins biosynthesis in these bacteria (belonging to the genera *Azotobacter, Pseudomonas* and *Lactobacillus*) certain yeast strains, and mycelia rhizospheric fungi.GA increases the growth rate and promotes nitrogen fixation in cyanobacteria of the genus *Anabaena* and it also stimulates the formation of lytic enzymes in certain bacteria and fungi (Vinklarkova and Sladky,1978; Barea,1974).

Cytokinins:-
Cytokinins are adenine derivatives. The first compound exhibiting cytokinin activity was isolated from the semen of herring. Subsequently, a factor responsible for control of cell division was isolated from maize (*Zea mays* L.); hence named as zeatin. The second natural cytokinin to be identical was isopentenyadenine; this compound was a minor base in serine tRNA of yeast. Studies with slime mold *Dictyostelium discoideum* revealed that 5'-AMP was a direct precursor of isopentenyl adenosine 5'-phosphate. The enzyme catalyzing this conversion, dimethylallyl diphosphate: 5'-AMP transferase (or isopentenyl transferase) was also found in cell-free extract from maize kernels, and from tobacco callus tissue cultures that became cytokinin-autonomous. Recently several genes encoding the isopentenyl transferase have been identified from *Arabidopsis thaliana*. A corresponding enzyme from the bacterium *Agrobacterium tumefaciens*, encoded by the ipt gene, has been studied in depth at the molecular level, and the same gene was also found in *Pseudomonas syringae* pv. s*avastanoi*, where it is named ptz.

Depending on the chemical structure of their molecules (i.e. on the nature and position of a substituent in the purine ring), cytokinins exhibit diverse physiological activities. The multiplicity of functions performed by cytokinins allows them to regulate a wide range of physiological responses: activation of cellular RNA and protein synthesis, stimulation of plant cell division, promotion of the branching (tillering), interruption of the quiescence of dormant buds, activation of seed germination, regulation of chloroplast formation, stabilization of photosynthetic apparatus under the conditions of water stress and augmentation of general resistance of the cells to a variety of adverse environmental factors(Chernyad'ev,2005). Cytokinins are capable of potentiating the activities of RNA polymerase and matrix chromatin, and thereby affect protein synthesis. Microorganisms are capable of synthesizing kinetin, zeatin, isopentenyladenine and some other cytokinin derivatives.

Cytokinins are formed by rhizobacteria (belonging to the genera *Rhizobium, Azotobacter ,Azospirillum, Arthrobacter, Bacillus* and *Pseudomonas*) and certain streptomycetes (Upadhyaya *et al*.,1991). The ability to synthesize cytokinins is inherent in methylotropic and methanotropic bacteria (belonging to the genera *Methylobacterium, Methylomonas, Methylopila, Methyloarcula, Methylophylus, Methylobacillus,Methylovorus, Xanthobacter, Paracoccus, Blastobacter, Hyphomicrobium* and *Methylosinus*)(Shepelyakovskaya *et al*.,1999). PGPR strains (i.e. *Pseudomonas fluorescence* and phytopathogenic bacteria *Agrobacterium tumefaciens, Erwinia herbicola, Pseudomonas solanacearum, P. syringae* and *Rhodococcus fascians*) from cytokinins, in addition to auxins.

The synthesis of cytokinins is tRNA –dependent in *Pseudomonas aeruginosa, Rhizobium* spp., *Rhodococcus fascians*, and the fungus *Taphrina cerasi* (Gray, 1996). In the majority of cases, however, tRNA degradation

produces inactive *cis*-isomers of zeatin, whereas the active *trans*-isomers are formed by *de novo* biosynthesis. On the other hand, tRNA of the diazotropic bacterium *Azotobacter vinelandii* was found to contain 2-methylthioribosyl-*trans*-zeatin.

Ethylene:-

Ethylene biosynthesis by plants originates from methionine. The first step is the synthesis of S-adenosyl-methionine, followed by its conversion into 1-aminocyclopropane-1-carboxylic acid (ACC). ACC is the direct precursor of ethylene. The ACC oxidase, formerly known as the ethylene-forming enzyme **(**EFE), was first characterized in apple. Ethylene production has been also reported for bacteria and fungi.

Phytohormone-like substances formed by microorganisms affect not only the plant host, but also the producer microorganisms, which undergoes the necessity of holistic approaches to the plant and its associated micro biota as components of a single system. Some researchers believe that excretion of IAA by bacteria grown under unfavorable condition may have considerable functional importance as a factor increasing the probability of forming plant-microorganism association. This conclusion based on the finding that the amount of IAA reaches maximum values during the steady-state stage of development, characterized by nutrient depletion. The role of hormones as regulatory substances should therefore be viewed on abrader scale, because they act as intermediaries not only in processes confined to plant tissues, but also in communications of diverse organisms inhabiting the same ecological niche. Each participant of such a community has an intrinsic biochemical activity and pursues its own ends; both pathogenic and symbiotic microorganisms, however, excrete the same phytohormones. The difference in the resulting effect is not infrequently reduced to the concentration of a phytohormone. In this particular this case, microorganisms populating the root surface and capable of excreting phytohormones gets advantages in its colonization (Maor *et al.,* 2004).

Role of microbial phytohormones in plant growth promotion as biofertilizers:-

Plant growth regulators are widely used as stimulants of seed and tuber germination, accelerators of root formation and fruit ripening, agents for controlling plant growth and development, and means of increasing crop productivity. Growth stimulators accelerate and augment root formation (in plants where root age poses problems or does not occur under normal conditions) and regulate quiescence. phytohormones are used against viral infections and diseases caused by phytophogenic fungi. The use of phytohormones is indispensive in all cases where a whole plant is grown from an *in vitro* plant tissue culture.

1. **PGP Effect on Crops of Agronomic Importance:-** The observed PGP (plant growth promotion) effect include modifications of the root morphology after inoculation with *Azospirillum*, such as a dramatic increase of length and density of roots hairs, increase in root branching and root surface area, which led to an enhanced uptake of water and minerals. All these effects have been tentatively attributed to the production phytohormones such as IAA, gibberellins and kinetin by the bacteria (Jain and Patriquin, 1984).
2. **Use of low IAA producers:-** The effect of *Azosirillum* inoculation on the plant is concentration dependent leading to the promotion or inhibition of root growth (Dobbelaere *et al.,* 1999). Thus inoculation with *Azospirillum* mimics typical growth response induced by auxins, which are inhibitory of plant growth at high concentrations and stimulatory at lower levels.
3. **Use of plant hormone producer microorganisms as inoculum for many crops:-** Microbial phytohormones exert beneficial effects when plant seeds, seedlings etc. are treated with cultures and /or suspensions of producer microorganisms. Seeds treatment with soil rhizobacteria *Azospirillum, Bejerinckia, Rhizobium, Agrobacterium, Bacillus, Pseudomonas, Mycobacterim, Arthrobacter, Methylovorus,* and *Flavobacterium* strongly stimulates the germination capacity and germination in seed, also increasing the growth and crop productivity in mature plants(Dileep,1998). The augmentation of the growth rate correlates with the increase in ability of the bacteria to colonize the plant and the amount of the phytohormones formed (Zvyagintsev, 1995). Strains of rhizobacteria producing small amounts of auxins increased considerably the development of wheat (*Triticum aestivum* L.) and its crop productivity. Inoculation with cytokinin-producing methylobacteria of transgenic tobacco plants characterized by altered morphology (rootlessness) restored root formation and the effects of the microorganism culture on seed germination and plant development were similar to those of the phytohormones or the culture liquid of methylobacteria. Treatment of dwarf rice (which lacks the ability to synthesize gibberellins) with *Azospirillum lipoferum* and *A. brasilense* resulted in a pronounced stimulation of plant growth. This effect was due to the ability of the bacteria to metabolize exogenous GA20 (gibberellins 20) into the biologically active GA1 (Tudzynski, 1999). Moreover, , industrial production of gibberellins for agriculture relies primarily on the cultivation (on an industrial scale) of the fungus *Fusarium moniliforme* ,the perfect stage of which (*Gibberella*

fujikoroi) produces considerable amounts of diverse gibberellins. (Polyanskaya *et al*., 2002). Bacterial treatment of seed makes it possible to achieve germination of germination-resistant seed of rare, decorative or industrially important plants. Thus, bacteria of the genera *Pseudomonas, Bacillus, Xanthomonas, Rhodococcus* and *Micrococcus*, which all form auxins, strongly stimulate symbiotic germination of the seeds of tropical orchids and accelerate their development under greenhouse conditions. It should be taken into account that the beneficial effects of bacterial treatment depend on a variety of factors, including the activity of the strain, the concentration of the cells, the amount of phytohormones in the culture liquid , the quantity of the dry preparation of the stimulating microorganism, the duration of the treatment, the species of the plant ,the state of the indigenous microflora at the time of seeding , the characteristics of the soil and the general conditions of the agro technological complex. The introduction of bacterial inoculums is more successful if the strains are isolated from the rhizoplane or rhizosphere of mature plants of the same species (Lalande *et al*., 1989). It is not frequent that stimulation of plant growth and development by PGPR (plant growth promoting rhizobacteria) strains of bacteria is underlain not only by phytohormone formation, but also by their capacity for nitrogen fixation, improvement of plant nutrition (water and mineral) and prevention or suppression of phytopathogen growth; the latter effect is due to excretion by PGPR strains of bactericidal and fungicidal substances (Glick and Pasternak, 1998).

4. **Sugar cane promotion:-** Up to 80% of the total N incorporated into several sugar cane cultivars can be attributed to BNF (biological nitrogen fixation). In addition, the growth promotion can be driven by a hormone-dependent mechanism. Under N-sufficient growth condition, plants inoculated with *Gluconacetobacter diazotrophicus*, either as the wild type or a nifD mutant are approximately 20% taller than non inoculated plants. These results suggested that *Gluconacetobacter diazotrophicus* could benefit sugarcane by two ways: by transfer of bacterial nitrogen fixed and as well as via phytohormones production (Sevilla *et al*, 2001).
5. **Gain in root length associated to ACC-deaminase:-** Ethylene plays an inhibitory role on root elongation. A role for the ACC-deaminase in preventing ethylene effect was shown in inoculation experiments of canola roots by *E.cloacae*. The plant growth promotion effect is linked to the lowering of plant ethylene levels by the bacterial ACC-deaminase.

Conclusion:-

Understanding of IAA, Gibberellins and cytokinins metabolism calls for further identification and analysis of the intermediates, enzyme and genes involved in their biosynthesis, as well as in the isolation of mutants defective in each pathway. Although the production of phytohormones at the free living state is well established in many microorganisms, there is still insufficient evidence for their synthesis in their natural habitats. The ecological significance of phytohormones production by bacteria would be more convincing if it could be demonstrated that bacterial phytohormones production occurs while bacteria colonize the root system. As both the plant and the bacteria synthesize and secrete auxins, gibberellins and cytokinins is difficult to address the contribution of one particular hormone as responsible of the effects observed. Thus the possibility that the host plant directs the bacterium to produce IAA through Trp present in root exudates is intriguing and speculative at this point.

Plant-associated microflora is the richest source of microorganisms synthesizing phytohormones. The bulk of evidence shows that phytohormones formed by fungi, algae and bacteria are structurally identical to their plant counterparts. In future, the use of transcriptional (or other type) fusion for the analysis of the differential expression of the bacterial genes involved in phytohormones biosynthetic pathways in association with the host plant should generate important information.

In recent years, a number of studies on inoculation of cereal crops (wheat, maize, sugar cane, sorghum and sunflower) and horticulture crops with PGPR have been performed. Beneficial effects such as increases in nitrogen content and yield have been reported in Belgium, Israel, France, Argentina, Uruguay, Mexico, USA and South Africa. Success of field experiments depends of many parameters, such as the strain used, concentration of bacterial inoculum, viability of bacteria during storage, carrier employed, appropriate inoculation methodology, and soil characteristics. The identification of many traits and genes related to the beneficial effects of inoculated bacteria shall result in a better understanding of the performance of bioinoculants in the field. It will also provide a strategy to design genetically modified strains with improved PGP effects. This multiplicity of effects of phytohormones determines the function of the plant-microorganism community as a whole. The productive efficiency of a specific PGPR may be further enhanced with the optimization and acclimatization according to the prevailing soil conditions. In future, they are expected to replace the chemical fertilizers, pesticides and artificial growth regulators which have numerous side-effects to sustainable agriculture. Further research and understanding of mechanisms of

PGPR mediated-phytostimulation would pave the way to find out more competent rhizobacterial strains which may work under diverse agro-ecological conditions.

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