

BIOCONTROL POTENTIAL AND MECHANISM OF *BACILLUS* SPP. AGAINST PHYTOPATHOGENS: A MINI REVIEW

Abstract: The biocontrol potential of *Bacillus* spp. clearly indicated by several authors are presented crop wise. The understanding of mechanism of action of *Bacillus* spp. against phytopathogens is essential to support their biocontrol potential. In this regard the various mechanism of action of *Bacillus* is elaborately discussed with suitable subheadings along with their plant growth promoting ability. The available literature presented below as review strongly emphasized their distinct biocontrol potential coupled with plant growth promoting effect through their various behavioural, biological, biochemical, induction of resistance, role of antimicrobial peptide genes etc.

Keywords: Antibiosis, antimicrobial peptide genes, *Bacillus*, biocontrol potential, mechanism of action

Introduction

The scanning of literature strongly emphasised that the endophytic *Bacillus* spp. have tremendous biocontrol potential and possess desirable attributes of bioagents. The edophytes proved to be amenable to use as biopesticide against plant nematodes, fungal and bacterial plant pathogens.

Mechanisms of *Bacillus*

Several researchers reported the different types of mechanism of *Bacillus* spp. in the management of nematode, fungal and bacterial pathogens as follows.

Inhibition in egg hatching of nematodes

The culture filtrate of *B. firmus* significantly reduced egg hatching of

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Meloidogyne Incognita (Mendoza *et al.*, 2008). Similar viewpoints put forth by Perry (1989) and Jones *et al.* (1998) revealed that some endophytic bacteria inhibiting egg hatch of potato cyst nematode through altering the behaviour of nematodes. In the crop rice also Padgham and Sikora (2006) reported that the plants treated with the

bacterium *B.megaterium* reduced the attraction of *M.graminicola*. In support of the above findings Reitz *et al.* (2000) also demonstrated that the B43 strain of *B.sphaericus* reduced the egg hatch of *G.pallida* by 30 per cent. Hence it is hypothesised that the high density of bacteria found over the root could affect nematode attraction by (1) altering root exudates patterns, making the root less attractive (2) producing metabolites with nematode repellent activity and (3) producing high amounts of carbon dioxide which attracts nematodes towards plant roots.

Mortality of juveniles

Toxin proteins produced by *B. thuringiensis* (*Bt*) are the most broadly used natural insecticides in agriculture against root knot nematodes. The nematicidal effects of spore/crystal proteins of ten *B. thuringiensis* isolates studied *in vitro* against *M. incognita* exhibited the highest nematicidal activity with the mortality range of 86-100 per cent. In addition, ammonium sulphate cut off fraction of vegetative cultures of the most potent isolates of *Bt* 7, *Bt* 7N, *Bt* Soto and *Bt* Den examined *in vitro* for their nematicidal effects also showed similar results with 80 to 100 per cent mortality of nematodes in general (Mohammed *et al.*, 2008). Toxic metabolites like bacillopeptidase and subtilin E from *B. subtilis* and lactamase from *B. cereus* (Katz and Demain, 1977 and Simonen and Paiva, 1993) and non-cellular extracts (Gokte and Swarup, 1988) were attributed for the high degree of juvenile mortality of root knot nematode. The exposure of three nematodes *viz.* root knot, burrowing and cyst nematodes to the pure culture filtrate of *B. firmus* resulted in significant mortality of juveniles of the above nematodes. The bioactive compounds of secondary metabolites of the bacterium were reported to be responsible for its larvicidal effect according to Mendoza *et al.* (2008).

Nematode penetration

There was report on the reduction in the rate of penetration of sedentary and migratory endoparasitic nematodes in plants treated with endophytic *Bacillus* spp. The reniform nematode *Radopholus reniformis* penetration in tomato was delayed by 44.5 per cent due to *B. subtilis* (isolate Bs) cell suspension with 10^{10} cells / ml (Niknam *et al.*, 2003). **Nematode reproductive potential**

Racke and Sikora (1992) found that reproductive potential of *G. pallida* significantly reduced following treatment with *B.sphaericus* B43.

Nematode multiplication

The multiplication rate of *R. reniformis* reduced significantly when the *B. subtilis* isolate Bs suspension used as soil drench a week before nematode inoculation in tomato (Niknam *et al.*, 2003).

Nematode fecundity

The culture fluid, cell-free supernatant and cell-pelleted residues of each of the four isolates of *B. thuringiensis* viz. Bt 7, Bt 7N, Bt Soto and Bt Den were evaluated for their nematicidal activities *in vivo* using tomato plants as host. The results showed that both crude suspension and cell free supernatant of isolate Bt7N reduced the root knot nematode number of egg masses by 78 and 77 per cent respectively and number of eggs by 84 and 76 per cent compared to untreated control (Mohammed *et al.*, 2008)

Inhibitory effect on fungal / bacterial pathogens

Ruichen *et al.* (2014) stated that the endophytic bacterium *B. subtilis* strain Y-1 isolated from apple had effect to arrest the hyphal growth of *Fusarium* sp., *Rhizoctonia* sp, *F. oxysporum*, *F. moniliforme*, *F. proliferatum*, *F. solani* and *R. solani* *in vitro* and it was to the extent of 64.90 per cent in apple. Certain isolates viz. Rb29, Rb6, Rb12, Rb4, and Rb15 of *Bacillus* spp. capable of producing more volatile metabolites inhibited *F. oxysporum* f.sp. *cubens* mycelial growth by 40 per cent *in vitro* (Zeimet *et al.*, 2013).

Antibiosis

Identification of three lipopeptides antibiotics viz. surfactin, fengycin and iturin A in butanolic extracts from cell-free culture filtrates of some strains of *B. subtilis* were responsible to affect *Podosphaera fusca* causing powdery mildew in cucurbit. In this study Romero *et al.* (2007) pointed out that antibiosis could be a major factor involving the biocontrol activity of the bacterium.

Antibiotic production

The scanning of literature revealed adequate information on the influence of antibiotics of *Bacillus* spp. in the management of fungal / bacterial plant pathogens but it is almost nil in respect of nematodes.

Influence of antimicrobial peptide genes in general

Many species of *Bacillus* were capable of producing a wide variety of secondary metabolites that are diverse in their structure and function. The production of metabolites with antimicrobial activity is important to control plant diseases (Silosuhet *et al.*, 1994). In general *Bacillus* spp. express antagonistic activities by suppressing the pathogens and numerous reports covering this aspect both *in vitro* and *in vivo* were already documented by several authors (Arrebola *et al.*, 2010; Chen *et al.*, 2009; Joshi and McSpadden Gardener, 2006).

Hence if a *Bacillus* has to perform well under field conditions it should possess genes like surfactin for sustainable performance against plant diseases according to Ongena and Jacques (2008).

The metabolites of *Bacillus* spp. can be ribosomal compounds such as subtilin (Zuber *et al.*, 1993), subtilosin A (Babasakiet *et al.*, 1985), tas A (Stover and Driks, 1999) and sublancin (Paik *et al.*, 1998). A variety of nonribosomally produced small lipopeptides are belonging to the surfactin family: surfactin and lichensysins (Kluge *et al.*, 1988); the iturin family: iturin A, C, D and E, bacillomycin D, F and L and mycosubtilin (Maget-Dana and Peypoux, 1994) and the Fengycin family: fengycin and plipastatins (Vanittanakomet *et al.*, 1986). Zwittermycin A is belonging to aminopolyol group as reported by Milner *et al.* (1996).

Iturin

Antibiotics of iturin family showed strong antifungal and haemolytic activities with limited antibacterial activity (Nishikioriet *et al.*, 1986). All the 21 isolates of *B. subtilis*, *B. cereus*, *B. thuringiensis*, *B. licheniformis*, *B. mycoides* and *B. amyloliquefaciense* evaluated by Athukorala *et al.* (2009) were showed positive reaction for the antibiotic iturin A. Antibiotics from iturin family showed strong antifungal and haemolytic activities with limited antibacterial activity (Maget-Dana and Peypoux, 1994).¹ Iturin had a broad antifungal spectrum and serves as a potential agent for the biological control of plant diseases (Maget-Dana and Peypoux, 1994). Iturin A produced by *B. subtilis* had strong antimicrobial action in suppressing *Pythium ultimum*, *Rhizoctoniasolani*, *Fusarium oxysporum*, *Sclerotium sclerotiarum* and *Macrophominaphaseolina* (Constantinescu, 2001).

Fengycin

The antibiotic fengycin is specifically effective against filamentous fungi and inhibits phospholipase A2 (Nishikioriet *et al.*, 1986). Liang *et al.* (2009) identified antifungal compound fengycin responsible for the growth suppression of *F.*

moniliforme. *B. subtilis* SR146 isolated from Tunisian salty soil showed antifungal property against several species of *F. culmorum*, *F. graminearum*, *F. oxysporum*, *F. melonis*, *F. equiseti* and *F. solani*. Absolute inhibition of *F. culmorum* spores germination was observed with the strain SR146. The compounds which responsible for antifungal activity were purified and characterized. The GC-MS analysis of the compounds showed high similarity coefficient to fengycin. This result was confirmed by PCR through the detection of the *fen A* gene of the fengycin operon (Hanene *et al.*, 2012).

Surfactin

Surfactin showed antiviral and antimycoplasmal activities (Vollenbroichet *al.*, 1997a; Vollenbroichet *al.*, 1997b). All the 21 isolates of *B. subtilis*, *B. cereus*, *B. thuringiensis*, *B. licheniformis*, *B. mycoides* and *B. amyloliquefaciens* tested by Athukorala *et al.* (2009) were found to be positive for the antibiotic surfactin gene.

Zwittermycin A

Zwittermycin A had structural similarities to polyketide antibiotics with broad spectrum of action against various microbes (Silo-suhet *al.*, 1998). The diverse biological activity of these antibiotics caused the suppression of oomycetous disease of plants and responsible for the insecticidal activity of *B. thuringiensis* also (Emert *et al.*, 1999). In addition, the biocontrol activity of *Bacillus* strains against multiple plant pathogens have been widely reported and well documented (Kloepper *et al.*, 2004 and Correa *et al.*, 2009). Silo-suhet *al.* (1998) reported that zwittermycin A is having a broad spectrum activity against certain gram-negative bacteria and eukaryotic microorganisms. There are reports that *B. subtilis*, *B. cereus*, *B. licheniformis* and *B. amyloliquefaciens* were effective against plant and fruit diseases caused by soilborne, aerial or post-harvest fungal diseases (Broggini *et al.*, 2005; Szczech and Shoda, 2006 and Yoshida *et al.*, 2002). *B. subtilis* H-08-02, *B. cereus* L-07-01 and *B. mycoides* S-07-01 showed strong antifungal activity against *F. graminearum*. Detection of antibiotic production by a particular bacterium is important in determining its capacity to be a good biocontrol agent for the management of plant diseases (Fernando *et al.*, 2002 and Ramarathnamet *al.*, 2007).

Combined influence of surfactin and iturin A

The biocontrol agent *B. subtilis* produced several classes of broad spectrum lipopeptides antibiotics effective against many plant pathogens as reported by Ongena and Jacques. (2008) and Nagorska *et al.* (2007). Both surfactin and iturin A

were serving as surfactants with a hydrophilic ring of seven amino acids and a long, hydrophobic hydrocarbon tail. The hydrocarbon tail penetrates pathogen cell membranes while the amino acid end stays in the soil solution. This action creates openings in cell membranes is related for inhibiting the growth of many pathogens (Ongena and Jacques., 2008). The strain YC300 identified as *Paenibacilluskoreensis* had strong antifungal activity against *F. oxysporum*, and *Colletotrichum lagenarium*, *S. sclerotiorum*, *R. solani* and *B. cinerea* (Chung *et al.*, 2000). Asaka and Shoda (1996) observed a significant suppressive activity of iturin A against plant pathogens compared to surfactin. Surfactin is an acidic cyclic lipopeptide produced by strains of *B. subtilis* are being used as a biosurfactant (Maget-Dana and Ptak, 1995). Surfactin and iturin A were the most common among lipopeptide antibiotics produced by *Bacillus* spp. The specific surface and membrane active properties of the surfactin help bacteria to form biofilm. Therefore, surfactin is thought to perform developmental functions rather than defense functions in the environment. Surfactin also induced a strong membrane destabilizing action at concentrations even below its critical micellar concentration and induced the formation of ion channels in lipid bilayers (Heerklotz and Seelig, 2007). Phae *et al.*, (1990) reported that more than 23 types of plant pathogens were suppressed *in vitro* by iturin A and surfactin producing *B. subtilis* isolate. Surfactins have been reported to be powerful surfactants due to their excellent surface activities. Surfactins can largely reduce the surface tension of water from 72 to 27 milli Newton / meter at a concentration of 10 M (Peypoux *et al.*, 1999). Compared with conventional surfactants, the surfactin had the additional advantages of antiviral (Itokawa *et al.*, 1994) and antibacterial property (Beven and Wroblewski, 1997).

Bacillomycin D

Bacillomycin D which is a member of the iturin family along with mycosubtilin and iturin A is made of one β -amino fatty acid and seven α -amino acids exhibited a strong antifungal activity against *Aphelenchus flavus* and a broad range of plant pathogenic fungi. The bacillomycin D has been reported to inhibit aflatoxin production by *A. flavus* and *A. parasiticus* (Ono and Kimura, 1991). Biosynthesis of bacillomycin D is independent of the ribosomal process and the enzymes responsible for bacillomycin D production were complex peptide synthetases (Besson and Michel, 1992). The bacillomycin D and fengycin jointly contributed to the inhibition of conidial germination of *Monilinia fructicola* and fengycin played a

major role in suppressing mycelial growth of the fungal pathogens viz. *Magnaportheorizae*, *R. solani* and *Botrytis cinerea*. Microscopic observations demonstrated that the hyphae of the pathogenic fungi treated with bacillomycin L showed abnormal growth and enlargement in conidia and constricted germ tube. Cellular leakage was also observed when bacillomycin L used in high concentration (Luo *et al.*, 2010).

Influence against diseases of specific crops

Tomato

The production of antibiotics like iturin and surfactin of *B. subtilis* strain RB14 suppressed the damping off disease of tomato (Asaka and Shoda, 1996). The colonization of plant roots by *B. subtilis* is associated with surfactin production and biofilm formation which protect *Arabidopsis thaliana* from *Pseudomonas syringae* on tomato (Bais and Vivanco, 2004). Asaka and Shoda (1996) observed that antibiotics like iturin A and surfactin, produced by *B. subtilis* RB 14 suppressed damping off of tomato seedlings caused by *R. solani*.

Cucurbits

The production of mixtures of bacillomycin, fengycin and iturin A by *B. subtilis* has been related to the control of powdery mildew caused by *Podosphaera fusca* on cucurbits by Romero *et al.* (2007). Similarly the production of bacilysin, iturin and mersacidin by *B. Subtilis* (ME488) was reported to be responsible for the suppression of *Fusarium* wilt of cucumber. Thus a single strain of *B. subtilis* was found to be effective against both Oomycetous and Dueteromycetous fungi (Chaung *et al.*, 2008). Accordingly the strains of *Bacillus* that score positive reaction for AMP biosynthetic genes were more effective to inhibit the growth of *R. solani* and *Pythium ultimum* than other *Bacillus* isolates that lack one or more of AMP genes (Joshi and MC SPadden, 2006). Involvement of iturin and fengycin antibiotics from four *B. subtilis* strains viz. UMAF6614, 6616, 6639 and 8561 were reported in the suppression of powdery mildew of cucurbits caused by *Podosphaera fusca*. (Romero *et al.*, 2007)

Beans

The fengycin produced by *B. subtilis* was effective against damping-off of bean caused by *Pythium ultimum* (Ongena *et al.*, 2005).

Pepper

The bacilysin, iturin and mersacidin of *B. subtilis* (ME488) were effective for the management of *Phytophthora* blight of pepper (Romero *et al.*, 2007).

Apple

Ongenaet *al.* (2005) described the role of secreted lipopeptides and more particularly of fengycin against grey mould of apple.

Lytic enzymes

The lytic enzymes of *B. subtilis* strain RB14 was capable of suppressing damping off disease of tomato according to Asaka and Shoda, (1996). *In vitro* studies revealed that *M. javanica* eggs and juveniles were inhibited by the crude antibiotics of *B. alvei* NRC14 and its effect was positively correlated with the concentration of the same. The strain producing lytic enzymes *viz.* chitinase, chitosanase, proteases as well as other potential bioactive metabolites were reasoned for the inhibitory effect of the bacterium (Abdelaziz *et al.*, 2013). The strain *B. alvei* (NRC14) producing mycolytic enzymes *viz.* chitinase, chitosanase, β -1,3 glucanase as well as cellulases, proteases and potential bioactive compounds were effective to suppress several plant diseases, insect pests and plant parasitic nematodes due to its insecticidal and antimicrobial properties (Shadia, 2013). Terefe *et al.* (2008) indicated that the *B. subtilis* strain Tolr-MA has an ability to produce proteolytic enzymes.

Huang *et al.* (2009) reported that chitinolytic bacterium *B. cereus* 28-9 isolated from lily plant in Taiwan exhibited biocontrol potential on *Botrytis* leaf blight of lily as demonstrated by a detached leaf assay and dual culture assay. At least two chitinases (ChiCW and ChiCH) were excreted by *B. cereus* 28-9. The ChiCW encoding gene was cloned and moderately expressed in *Escherichia coli* DH5 α . Near homogenous of ChiCW was obtained from the periplasmic fraction of *E. coli* cells harboring ChiCW. Further *in vitro* assay showed that the purified ChiCW posed inhibitory activity on conidial germination of *Botrytis elliptica*, a major fungal pathogen of lily leaf blight.

Inducing systemic resistance (ISR)

One of the promising strategies for the management of nematodes is use of resistant inducers. The resistance inducers or elicitors can take the form of a chemical compound or a live organism whose function is to activate the plant's defense mechanisms (Arieiraet *al.*, 2013, Wilson, 1989; Vanpeeret *al.*, 1991; Drobyet *al.*, 1991; Leeman *et al.*, 1995). Plant growth promoting rhizobacteria (PGPR)

belonging to *Bacillus* spp. are being exploited commercially in the field of plant protection to induce systemic resistance against various pests and diseases. The *Bacillus* strains have resulted in increased efficacy by inducing systemic resistance against several pathogens attacking the same crop. Seed treatment with PGPR like *Bacillus* spp. causes cell wall structural modifications and biochemical / physiological changes leading to the synthesis of proteins and chemicals involved in plant defense mechanisms. Lipopolysaccharides, siderophores and salicylic acid are the major determinants of PGPR mediated ISR. The performance of PGPR has been successful against certain pathogens, insects and nematodes under field conditions as proved by Ramamoorthy *et al.* (2001).

The crop protection resulting from ISR elicited by *Bacillus* spp. has been reported against leaf spotting fungal and bacterial pathogens; systemic viruses, a crown-rotting fungal, stem-blight fungal, damping off, blue mould and late blight diseases in addition to root-knot nematodes. Reductions in field populations of three insect vectors have also been observed earlier (Kloepper *et al.*, 2004). The induction of systemic resistance through the use of *Bacillus* spp. has been demonstrated in different crops as follows.

In tomato

In an experiment on the management of *Fusarium* wilt of tomato, two strains *viz.* *B. fortis* IAGS162 and *B. subtilis* IAGS174 were found to trigger the defence enzymatic activities *viz.* Peroxidase (PO), Polyphenoloxidase (PPO) and Phenylammonialyase (PAL) and phenolic content which helps for the induction of systemic resistance against fungal pathogens (Akram *et al.*, 2013).

The systemic resistance induced by *B. subtilis* (strain Bs) was effective against *Rotylenchulus reniformis* in tomato (Niknamlet *et al.*, 2003). The elicitation of ISR by the specific strains of *B. amyloliquefaciens*, *B. subtilis*, *B. pasteurii*, *B. cereus*, *B. pumilus*, *B. mycoides* and *B. sphaericus* has been demonstrated under controlled and field conditions on tomato, bell pepper, muskmelon, watermelon, sugarbeet, tobacco, cucumber, loblolly pine and two tropical crops *viz.* long cayenne pepper and green kuangfutsoi (Vanloon, 1983).

In chilli

The induction of systemic resistance by *Bacillus* spp. was responsible for the management of blight disease of chilli as reported by Ahmed *et al.* (2000).

In potato

Gunther *et al.* (1998) established the systemically induced resistance by *B. sphaericus* mechanism in the management of *Globodera pallida* in potato. The ability of *B.sphaericus*(strain B43) and *Agrobacterium radiobacter*(strain G12) isolated from the potato rhizosphere to induce systemic resistance was said to be responsible for reducing the rate of root penetration and population of juveniles of potato cyst nematode *G.pallida* by Gunther *et al.* (1998).

In apple

The bacterium *B.subtilis*strain Y-1 isolated from apple induced systemic resistance through altered activities of super oxide dismutase against *Fusarium* sp, *Rhizoctonia* sp, *F. oxysporum*, *F. moniliforme*, *F. proliferatum*, *F. solani* and *R. solani* in apple according to Ruichenget *al.* (2014).

Induced biochemical changes

Biochemical changes due to nematode infestation and their antagonists in plant system was well documented by several authors (Tayal and Agrawal .1982 and Ganguly and Dasgupta, 1979). Information on this line is much useful to know the biochemical mechanism of bioagents used for the management of phytonematodes. Plant metabolism and hypersensitivity reaction in plants were explained as possible mechanism of bioagents towards the invading pathogenic organisms like nematodes. It has been reported that polyphenols and polyphenol oxidase together in an oxidative process resulted in browning reaction. Increase in polyphenol oxidase after the entry of nematodes is attributed for the triggering of phenol oxidation process as defence mechanism (Maraite.1973).

It is well known that accumulation of phenols in plant system is imparting resistance to invading plant pathogens including nematodes. The use of many biocontrol agents including PGPR resulted in accumulation of phenol as biochemical changes in favour of plants and against invading pathogens like nematodes (Pitcher *et al.*,1960). Abdelaziz *et al.*, (2013) reported that the release of high level of reducing sugar by *B.alvei* strain NRC14 is responsible for the nematicidal action of the bacterium against eggs and juveniles of *M. javanica*. The higher production of indoleacetic acid and hydrogen cyanide by the isolates viz. B5, B11, B4 and B1 of *Bacillus* spp. was related to their biocontrol potential in the management of *M.incognita* in tomato by Singh and Siddiqui. (2010). The biocontrol potential of two strains of *Bacillus* viz. *B. fortis* 162 and *B. subtilis* 174 against *Fusarium* wilt of tomato was studied under laboratory conditions. In this study the quantification of

total phenolic compounds and defense related proteins viz. PPO, PAL and PO by calorimetric methods was made. The results showed that the strain *B. subtilis* 174 exhibiting higher biocontrol potential resulted in higher content of phenolic compounds and enhanced defence enzymatic activities of PPO, PAL and PO compared to strain *B. fortis* 162 (Akram and Anjum.,2011).

Three isolates of *B. subtilis* viz. EXB-123, ENB-24 and S-9 proved to be promising bacterial antagonists against chilli anthracnose pathogen, *C. capsici* were subjected to bioassay study. It clearly indicated the higher level of phenolic compounds and the activities of defense related enzymes like PO, PPO and PAL following the use of *B. subtilis* against *C. capsici* in chilli. Among the three above bacterial antagonists tested, the *B. subtilis* EXB-123 ranked first in inducing biochemical changes as defence mechanism in chilli (Ramanujam *et al.*, 2012). The biochemical changes induced by *B. subtilis*, *B. firmus* and *B. coagulans* reported to be effective against *M. javanicum* in eggplants were observed at 15 and 45 days after inoculation. The results indicated that all three species of *Bacillus* were capable of accumulating phenolic compounds and enhancing defence enzyme activities of PO, PAL, guaiacol peroxidase, catalase, ascorbate peroxidase and declining super oxide dismutase (Abbasi *et al.*, 2014).

Plant growth promoting ability

The PGPR are known to enhance plant growth and health through their direct or indirect mechanisms. The plant health could be improved by controlling a range of plant pathogens including bacteria, fungi and nematodes. The use of PGPR recently named as plant probiotics to control plant pathogens is receiving increasing attention as they may represent an alternative approach to chemical pesticides (Shadia. 2013).

The principal mechanisms of *Bacillus* spp. are attributed to the production of growth stimulating phytohormones viz. Indole acetic acid and gibberellic acid and solubilization and mobilization of phosphate, siderophore production leading to the promotion in plant growth and thereby imparting tolerance against plant pathogens (Richardson *et al.* 2009; Idris *et al.* 2007; Gutierrez-Manero *et al.* 2001 and Whipps.2001) The plant growth promoting ability of *Bacillus* is detailed below crop wise.

Tomato

The plant growth characters of tomato challenged with *R. reniformis* increased following the application of *B. subtilis* strain Bs (Niknamlet *et al.*, 2003).

Cucumber

The *B. subtilis* strain BACTO is capable of improving the growth and yield of cucumber plants besides managing fungal disease caused by different pathogens (Utkhedeet *et al.*, 1992).

Safflower

Similarly Liang *et al.* (2009) reported that seed treatment with *B. polymixa*, increased the seedling height of safflower. Thus the *Bacillus* spp. has played both the role of crop protection as well as crop improvement.

Maize

In maize four isolates of *Bacillus* spp. produced IAA ranging from 53.1 to 71.1 ppm optimally. In this study conducted by Lwin *et al.* (2012) it is observed that all the isolates had different optimum IAA production periods and strain R1 was the best IAA producer strain with 121.1 ppm.

Chickpea

The strains BHUPSB13 of *B. subtilis*, BHUPSB17 of *Paenibacilluspolymyxa* and BHUPSB19 of *B. boronophilus* induced production of IAA, phosphate solubilization and ammonia production in chickpea. Hence Yadav *et al.* (2010) opined that the above isolates will be useful as biofertilizers to enhance the growth and productivity of chickpea.

Paddy

The efficiency of *B. subtilis* isolates designated as BS 1-10 was studied for IAA, GA, and siderophore production in addition to phosphate solubilisation. The study carried out by Sivasakthi *et al.* (2011) revealed the maximum phosphate solubilisation with the isolate BS-8 in paddy.

Glory lily (*Gloriosa superba*)

Phytohormones are plant growth regulators which have stimulatory effects on plant growth. In medicinal crop, *Glorios superba* the plant growth promoting rhizobacteria including *Bacillus* spp. were able to produce IAA and GA as reported by Megala *et al.* (2013).

Siderophore production

Rajendran *et al.* (2008) found that *Bacillus* strains NR4 and NR6 were able to produce siderophores and the rhizobial bioinoculant IC3123 was able to cross utilize under iron starved conditions. The above bioinoculant showed enhanced growth in the presence of the *Bacillus* isolates indicating that siderophore mediated interactions might be underlying mechanism of beneficial effect of the strains on nodulation by IC3123.

Competition for nutrients, space and niche exclusion

Competition for resources such as nutrients and oxygen occurs generally in soil among soil inhabiting organisms. Root inhabiting microorganisms compete for suitable sites over the root surfaces. Thus the competition for nutrients especially for carbon is assumed to be responsible for the well-known phenomenon of fungistasis (Alabouvette *et al.*, 2006; Paulitz *et al.*, 1992 and Baker *et al.*, 1991). Competition for trace elements such as iron, copper, zinc, manganese etc. also occurs in soil. For example, iron is an essential growth element for all living organisms and the scarcity of its bioavailable form in soil habitats results in a furious competition between pathogens and their antagonists (Loper and Henkels, 1997). Suppression of soilborne plant pathogens through competition for niche and nutrients has been demonstrated for some beneficial bacteria such as *Pseudomonas* spp. (Haas and Défago, 2005). In this regard the experimental proof available with regard to *Bacillus* is meagre. However, the competitive phenomena are speculated to occur with this bacterium under natural rhizosphere conditions by the above authors.

Others

The ability of *Bacillus* spp. for the fixation of nitrogen, degradation of cellulose, starch, pectin and protein in addition to production of various types of antimicrobial compounds were explained as probable mechanism of the bacterium for the management of plant pathogens (Turner *et al.*, 2004). The bacterium *B. subtilis* can improve the plant growth by producing biologically active substances or by transforming unavailable mineral and organic compounds into available forms to plants (Broadbent *et al.*, 1977). Thus it may partially compensate the losses caused by plant parasitic nematodes besides increasing crop yield.

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