

Microbes Knocking Louder Roles for Next Green Revolution: A Meta-Analysis

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ABSTRACT

Feeding the ever-growing population has always been a daunting challenge to researchers, however on the contrary, it opened the floodgate of innovative technologies, especially of late, with the introduction of meta-genomics unfolding those genetic traits of microbes, which otherwise remained unexplored for a very long time. Learning negative consequences of previous green revolution in terms of various chronic environmental issues, phyto-microbiome analysis (microbiome bioprospecting) leading to identification of many plant growth promoting microbes, disease suppressive antagonists including entomopathogenic microbes and agrochemicals residues cleansing microbes, preferably in a bioformulation mode have evoked a very strong claim to be stitched into modern day agriculture, claimed close to chemical residue free organic as well as natural agriculture, a pre-requisite to stake claim for next green revolution. Such microbial interventions are likely to ensure three major paradigm shifts in agriculture viz., i. over-use of pesticides to residue free precision farming, ii. traditional to intelligent farming and iii. input-intensive (high cost of production) to climate-smart solutions, all collectively look forward towards more sustainable development of agriculture. All these issues have been systematically analysed through bibliometric evidences and envisioned the future course of action to realise yet another green revolution through microbial interventions.

Keywords: Green revolution, precision farming, microbiomes, bioformulations, organic agriculture, agrochemicals, climate-smart

PROLOGUE

The global population has been dramatically rising, increasing the needs of food. Towards the end of 1800s, the world population rose to 1 billion and by the end of 1930 it reached 2 billion and now it is almost 8 billion. In 1944, Borlaug, an American Plant Pathologist and wheat breeder developed semi-dwarf high yielding varieties resistant to disease and lodging. The high yielding varieties produced astounding results in Mexico exporting half a million tons of wheat produced. Following Borlaug's success in Mexico, the governments of India and Pakistan requested for Borlaug's assistance and thus, agricultural revolution started in Asia (Britannica, 2022). Much of the success that took place during the first green revolution was caused by the combination of high rates of investment in crop research, intensive agriculture and appropriate policy support. Thus, green revolution helped to overcome a phase of famine and malnutrition which otherwise a huge population of world had to face. But, with such advantages, green

revolution also left several negative consequences in the post-green revolution decades, being in news in either electronic or print media.

Intensive agriculture saved barren lands from conversion to agricultural lands. Although, populations had more than doubled, the production of cereal crops tripled during this period, with only a 30% increase in land area cultivated. Green revolution driven intensification is a major source of greenhouse gas emissions and driver of climate change. Nitrogenous fertilizers like ammonium nitrate, calcium ammonium nitrate, urea, and urea ammonium nitrate are its major sources (Wood and Cowie, 2004). On the other hand, phosphate fertilizers such as mono- and diammonium phosphates, single superphosphates, and tri-superphosphates result in CO₂ emissions (Davis, 1999); whereas NPK fertilizers were observed to release CO₂ and N₂O, and most of the CO₂ was liberated during production of ammonia, and 100% N₂O was produced during nitric acid production (Kongshaug, 1998). CO₂ emissions increased by 1% in fruit and vegetable, 2.4%

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milk, 5.65% rice, 45.54% mutton meat production in CO₂eq/ kg (Vetter *et al.*, 2017). Biomagnifications of persistent chemicals in soil affect human health in different ways. As we consume the pesticidal residues along with raw fruits and vegetables they get circulated through blood and reach our brain cells and neurons leading to generation of diseases *viz.*, neurological imbalance, infertility in humans etc. High yielding varieties are more responsive to fertilizers and pesticides and the increase in fertilizers and pesticides significantly decreased the biodiversity of different ecosystems (terrestrial and aquatic) and has rapidly increased rate of extinction since the last 50 years (Prashar and Shah, 2016).

Agricultural biodiversity is not just a raw material for industrial agriculture; it is also the key to food security and sustainable agriculture. With the loss of biodiversity, options for long-term sustainability and agricultural self-reliance will lose too. Among the huge diversity of microbial communities in nature, some microbes help in promoting growth to plants which are termed as "plant growth-promoting microorganisms" (PGPM) while some others induce resistance to the plants against phytopathogens termed as biocontrol agents. Thus, PGPM and antagonistic microbes are good biofertilizers and biopesticides for plants, commonly termed Plant beneficial microbes (PBM).

BENEFICIAL MICROBES

One gram of soil harbors 10⁸⁻⁹ bacteria, 10⁵⁻⁸ actinomycetes, 10⁵⁻⁶ fungi population count but only 1-2 % of the micro-organisms in soil have been identified and the rest 99% is still unculturable (Srivastava *et al.*, 2015; Bora *et al.*, 2019). Hence, there is a huge number of microbes still to be exploited, probably the count is larger than those already isolated (Bora and Bora, 2010; Bora *et al.*, 2021b; Nasreen *et al.*, 2020). With the recent developments in molecular fields, metagenomics is one such efficient approaches which enables to screen and identify all the bacteria, viruses, fungi, nucleic acid present in a particular habitat without the need of culturing these microbes (Bora and Bora, 2008; Srivastava *et al.*, 2022). In *Rhizobium*, the three genes: *nod-nif-fix* were identified which plays the complete role of Symbiotic Nitrogen Fixation in leguminous

plants. Invasion of *Rhizobia* in plants and formation of root hair and nodules are encoded by the *nod* genes (Moulin *et al.*, 2004).

The species *Trichoderma* is one of the most studied and extensively used biocontrol agents (Bora *et al.*, 2020a; 2020b; Bora and Bora, 2021). Different species of *Trichoderma* which exhibit biocontrol activity against a wide group of phytopathogens either by secreting some enzymes or coiling hyphae of the later are *T. viride*, *T. harzianum*, *T. asperellum*, *T. atroviride*, *T. hamatum*, *T. longibrachiatum*, *T. reesei* (Sharma *et al.*, 2020; Saikia *et al.*, 2021). *Tri5* gene was isolated and characterised from *T. harzianum* and other isolates which is responsible for synthesizing the enzyme trichothecene. Trichothecene enzyme can successfully inhibit pathogen *Fusarium* by inhibiting protein and DNA synthesis in their cells. *Tri5* gene was inserted into pGEM-T vector, cloned, and expressed. (Gallo *et al.*, 2004) Also, another gene, *cre1* was isolated from *T. harzianum* which causes repression of cellulose and xylanase encoding genes which causes cell wall degradation of several phytopathogenic fungi. The gene was cloned in pTZ57R/T plasmid vector and inserted into *E. coli* DH 10B to study its expression (Saadia *et al.*, 2008; Bora *et al.*, 2022).

Researchers across the world have been exploring micro-organisms from highly polluted areas capable of degrading agricultural pesticides. In the recent years, researches are done to study the genes responsible for encoding various enzymes capable of converting the harmful pesticides to inert forms. Five bacterial species *viz.*, *Pseudomonas* sp., *Pseudomonas putida*, *Micrococcus lylae*, *Pseudomonas aureofaciens*, and *Acetobacter liquefaciens* were isolated from agricultural soil which successfully degraded malathion to malathion monocarboxylic and dicarboxylic acids. LC/ESI-MS analysis was done to confirm the degradation due to carboxylesterase activity (Bora *et al.*, 2016; Rahman *et al.*, 2020; Sharma *et al.*, 2021; Bora *et al.*, 2021a).

INCREASING USAGE OF BIOPESTICIDES

Biofertilizers are gaining importance worldwide as it helps maintaining proper health of soil, minimization of environmental pollution and cut down the use of chemical pesticides

(Saikia *et al.*, 2020; 2022). The global biofertilizer market was worth US\$ 2.0 Billion in 2019 and is projected to reach USD 2.7 billion by 2027, expanding at a CAGR of 12.8% (www.researchandmarket.com). Innovative companies *viz.*, Monsanto Bio AG, Symborg, Agrinos AS, Camson Bio Technologies Ltd, Bio Works Inc., Lallemand Inc. which were once indulged in production of inorganic fertilizers are making strides in developing microbial biofertilizers for plants.

About 20–40% annual reduction of the global crop yield is caused by pests and plant diseases (FAO (2015)). To overcome such huge loss, high number of chemical pesticides has been applied to the crop field during green revolution. This exacerbated the scenario by addition of xenobiotics such as DDT and methyl parathion, which are persisting in the soil as non-degradable moieties. Such issues must be mitigated through sustainable approaches before endorsing the concept of next green revolution (Arora *et al.*, 2020). Microbial inoculants have shown influential results in sublimating phytopathogenic biotic stress and reclaiming the quality and quantity of agricultural production. Plant pathologists across the world have identified a lot of antagonistic microbes and the number of research works and publications on biopesticides has been increasing since 1989 till date (Lahlali, *et al.*, 2020)). Also, many agricultural firms are manufacturing biopesticide products. The leading ones in the market are Byer, Certis, Dow, Monsanto, Isagro, Andermalt biocontrol, Marrone Bio innovations. The global biopesticides market is projected to grow at a CAGR of 14.7% from an estimated value of USD 4.3 billion in 2020 to reach USD 8.5 billion by 2025 (Khakimov *et al.*, 2020).

The microbes undergo several mechanisms to inhibit the pathogens. Some produce HCN, antibiotics, bacteriocins, lytic enzymes, VOCs and cry-protein/ δ endotoxins against the pathogens and insect. While some others trigger ISR in plants and activates their defense system to help combating phytopathogens. Also, some antagonistic bacteria release biosurfactants CLPs (cyclic lipopeptides) and exopolysaccharides forming a protective biofilm layer across the plants and microbes to protect them from pathogens and chelate nutrients. Another group of microbes excludes availability of nutrients (N, P, K, Fe)

and increase niche competition inhibiting the growth of phytopathogens. A single microbe may undergo different mechanisms to combat the pathogens depending upon the pathogen and host-pathogen interaction (Table 2). Since early research have been focused on isolating beneficial microbes from rhizospheric soil. Endophytes are another group of microorganisms from nature which engage mutually to their host plants and can cause benefits to the plants through numerous ways. Endophytic microbes have been shown to obtain nutrients in soils and transfer nutrients to plant, increase plant growth and development, reduce oxidative stress of hosts, protect plants from disease, deter feeding by herbivores and suppress growth of competitor plant species (Srivastava *et al.*, 2015).

Systemic acquired resistance (SAR) and induced systemic resistance (ISR) are the two forms of induced resistances. SAR is induced by the pathogen infection which is mediated by salicylic acid and associated with the accumulation of pathogenesis-related (PR) proteins. While ISR is induced by some non-pathogenic microbes and is mediated by jasmonic acid or ethylene this is not associated with the accumulation of PR proteins (Bora *et al.*, 2026; Srivastava *et al.*, 2022). Microbe- or pathogen-associated molecular patterns (MAMPs/PAMPs) *viz.*, flagellin is an essential structure that is conserved and recognized by receptor proteins in plants to induce the plant immune system. Endophyte *Bacillus cereus* triggers a much stronger defense reaction ISR than the pathogen *Botrytis cinerea* on *Arabidopsis thaliana* through an enhanced accumulation of the PR1 protein expression on time, hydrogen peroxide accumulation, and callose deposition. Endophytes also induce several cell wall modifications, such as deposition of callose, pectin, cellulose, and phenolic compounds, leading to the formation of a structural barrier at the site of a potential attack by phytopathogens. Similarly, they induce defense-related proteins such as peroxidases, chitinases, and β -1,3-glucanases.

A wide range of endophytic actinomycetes have a potential to produce antimicrobial compounds (Table 3), which probably contributed to be more competitive with other microbes in the host plants.

Table 1: Response different microbial species on growth, yield and quality parameters of different field and horticultural crops

Sr.No	Involvement of microbial species	Crop	Response parameters	References
1.	<i>Azospirillum</i> , phosphate solubilizing bacteria and vesicular-arbuscular mycorrhiza	Tomato	Increase in yield parameters with cost: benefit ratio of 1:3.76 with application of <i>Azospirillum</i> + 75% N + 100% PK.	Premsekhar and Rajashree (2009)
2.	<i>Bradyrhizobium japonicum</i>	Soybean	Significant enhancement in yield with application of <i>B. japonicum</i> and EM (effective microorganisms) in farmyard manure amendment with 45-47% increase in shoot biomass and number of pods/plant.	Javaid <i>et al.</i> (2010)
3.	<i>Pseudomonas</i> , <i>Bacillus</i> and yeast	Rice in sandy soil	Inoculation with BioGro containing a <i>Pseudomonad</i> , two <i>Bacilli</i> and a soil yeast significantly increased grain and straw yield by 3–5%.	Phan <i>et al.</i> (2011)
4.	<i>Pseudomonas</i> , <i>Azospirillum</i> and <i>Agrobacterium</i>	Wheat	Most of the growth characteristics were higher in treatments receiving biofertilizer supplemented with ½UN and full PM (200 mg N kg ⁻¹).	Abbasi & Younsa (2012)
5.	<i>Pseudomonas</i> sp., <i>Pseudomonas putida</i> , <i>Pantoea agglomerans</i>	Cucumber	<i>Pseudomonas</i> sp. produced the highest yield.	Isfahani <i>et al.</i> (2012)
6.	<i>Azotobacter</i> , <i>Azospirillum</i> , <i>Phosphobacteria</i> , <i>Rhizobium</i>	Black gram	The maximum germination percentage, fresh and dry weight, no. of pods/plant, seed yield per plant, hundred seed weight and root nodule plant were increased with the inoculation of <i>Rhizobium</i> with <i>phosphobacteria</i> treatment.	Selvakumar <i>et al.</i> (2012)
7.	<i>Azotobacter</i> , <i>Azospirillum</i> , PSB	Bittergourd	Maximum increase in vegetative growth, yield and different quality parameters.	Masih <i>et al.</i> (2020)

CELL-FREE BIOCONTROL STRATEGIES IN PLANT DISEASE MANAGEMENT

Use of cell-free inoculants has paved a new way in precision agriculture. Cell-based bioproducts have certain limitations under field conditions. It is difficult to maintain the proper colony count of microbial colonies in the products for a longer period time and to maintain their structural and functional integrity under diverse environmental conditions in field (Rahman *et al.*, 2020; Bora *et al.*, 2022). So, application of microbial metabolites along with cell-based inoculants will help in protecting and enhancing the functionality of PBM under field conditions. Microbial metabolites, *viz.*, phytohormones, enzymes, osmo-protectants, EPS, flavonoids and biosurfactants are some of the bioproducts which can serve the cause and give propitious results.

Phytohormones produced by microbes are one of the best stimulators for plants. Incorporating the precursors of these hormones

will reflect significant growth-promoting traits. Evidently, Zahiret *et al.* (2010) revealed that L-tryptophan application to mung bean along with *Rhizobium phaseoli* strain enhanced the auxin biosynthesis supporting increased nodulation, growth, and yield. Biosurfactants are another important group of microbial inoculants paving the way to microbial green revolution by improving soil quality through removal of heavy metals and hydrocarbon contaminants, antimicrobial activity against plant pathogens (Nihorimbere *et al.*, 2011; Khare *et al.*, 2011), quorum sensing, facilitating important plant-microbe communication (Berti *et al.*, 2007; Rosenberg and Ron, 1998), and adjuvants in fungicides, insecticides, and herbicides. Jeneil Biotech Inc. company, USA, is manufacturing a rhamnolipid (purified from *P. aeruginosa*)-based product ZonixBiofungicide™ substantial for biocontrol against downy mildew, late blight, black rot, and all phytophthora and pythium diseases (Thavasiet *et al.*, 2011). Microbial

Table 2: Multiple roles of microbes in plant health in field as well as horticultural crops

Sr.No.	Bioagent	Diseases/Pathogen	Response parameters	Reference
1.	<i>Trichoderma hamatum</i>	Radish and pea	Soils treated with <i>Trichoderma hamatum</i> showed lower densities of <i>R. solani</i> and <i>Pythium</i> over untreated soil.	Harman <i>et al.</i> (1980)
2.	<i>T. harzianum</i>	Bean, cotton and tomato	The wheat bran preparation of <i>T. harzianum</i> controlled <i>S. rolfsii</i> more efficiently than a conidial suspension of the same antagonist in beans, cotton, or tomatoes, and significantly increased the yield of these crops.	Elad (1980)
3.	<i>Trichoderma harzianum</i>	<i>Pythium aphanidermatum</i> in cucumber, peas, tomato, peppers and gypsophila	<i>Trichoderma harzianum</i> mixed with rooting mixture or soil efficiently controlled damping off induced in cucumber, peas, tomato, peppers and gypsophila.	Sivan (1984)
4.	<i>Trichoderma harzianum</i> and <i>Streptomyces rochei</i>	<i>Phytophthora</i> root rot of pepper	A combination of two compatible micro-organisms, <i>Trichoderma harzianum</i> and <i>Streptomyces rochei</i> , both antagonistic to the pathogen <i>Phytophthora capsici</i> , controlled the root rot in pepper.	Ezziyyani <i>et al.</i> (2007)
5.	<i>T. viride</i> and <i>P. fluorescens</i>	<i>Phytophthora infestans</i> causing late blight of potato	<i>T. viride</i> (AUDPC=260) and <i>P. fluorescens</i> (AUDPC=765.1) significantly controlled the disease.	Zegeye and Santhanam (2011)
6.	<i>Bacillus subtilis</i> and <i>Trichoderma asperellum</i>	<i>Pythium aphanidermatum</i> in tomatoes	The damping-off on seeds coated with <i>B. subtilis</i> and <i>T. asperellum</i> was reduced by 20.19% and 24.07% respectively.	Kipngeno <i>et al.</i> (2015)
7.	<i>Bacillus</i> and <i>Pseudomonas</i> spp.	Gray mold and <i>Alternaria</i> rot on blueberry fruit	Various bacterial antagonists reduced mycelial growth of <i>B. cinerea</i> and <i>A. alternata</i> by up to 42% and 27%, respectively.	Kejela <i>et al.</i> (2017)
8.	<i>P. fluorescens</i> , <i>P. aeruginosa</i> , <i>P. putida</i> , <i>P. cepacia</i>	Tomato early blight caused by <i>Alternaria solani</i>	The plots treated with <i>P. fluorescens</i> , <i>P. aeruginosa</i> , <i>P. putida</i> , <i>P. cepacia</i> showed maximum control of early blight in town.	Joseph <i>et al.</i> (2017)
9.	<i>Pseudomonas</i> spp.	<i>Colletotrichum gloeosporioides</i> and <i>Fusarium oxysporum</i> in coffee	The isolate named as PT11 showed maximum inhibition of <i>Colletotrichum gloeosporioides</i> (70%) and <i>Fusarium oxysporum</i> (72%) ($P < 0.05$) in <i>Coffea arabica</i> L.	Kejela <i>et al.</i> (2017)

osmoprotectants, lipochitooligosaccharides (LCO), and flavonoids are also being employed as inoculants to enhance stress tolerance, symbiotic association, and nodulation, respectively (Kaya *et al.*, 2013; Morel *et al.*, 2016). Lipase, protease, laccase/ligninase, cellulose, glucanase, and chitinase are some of the important microbial enzymes which can dissolve or degrade the cell wall of phytopathogens. Again, defense enzymes in microbes viz., peroxidase, polyphenol oxidase, phenylalanine ammonia lyase, chitinase and β -1,3-glucanase are promoters to Induced Systemic Resistance in plants. These biocontrol mechanisms of the microbial enzymes make them a good choice as biocontrol tools. So, large-scale industrial production of these

enzymes could be useful in making good quality of biocontrol products in the form of biopesticides (Bora *et al.*, 2016a ;2016b).

Enhancing the commercial production of microbial enzymes needs selective approaches so that maximum yield can be obtained with low capital cost. Use of low-cost raw materials as substrates, improved bioprocess technologies, and bioengineering of microbes are some of the multifaceted approaches that can be harnessed for maximum production of microbial enzymes (Lynd *et al.*, 2002; Sukumaran *et al.*, 2005). Fermentation and bioprocessing technologies are an essential component of mass production of microbial enzymes (Kumar *et al.*, 2011). Submerged fermentation (SmF) is preferred for bacterial enzymes because these microbes

require higher water potential in comparison to fungi (Chahal, 1983). Solid-state fermentation (SSF) is mostly implemented for fungal enzyme production and requires less water potential

(Babu and Satyanarayana,1996). Sooner, their use may expand in the manufacturing of the new generation biocontrol products showing host-specific and broad-spectrum activity.

Table 3: Microbes mediated involvement of secondary metabolites against disease management

Sr.No.	Endophytic microbe	Chemical compound	Host crop	Functional traits	Reference
1.	<i>Streptomyces</i> sp.	Fistupyrene	<i>Allium fistulosum</i>	Inhibition in incidence of <i>Alternariabrassicicola</i>	Igarashi <i>et al.</i> (2000)
2.	<i>C. quercina</i>	Cryptocandin	<i>Oryza sativa</i>	Inhibition in incidence of <i>Pyriculariaoryzae</i>	Li <i>et al.</i> (2000)
3.	<i>Pestalotiopsismicrospora</i>	Pestacin and Isopestacin	<i>Terminalia spp.</i>	Increase in antioxidant	Harper <i>et al.</i> (2003)
4.	<i>Streptomyces</i> sp.	β -1,3-glucanases, chitinases and cellulases	<i>Oryza sativa</i>	Inhibition in incidence of <i>Xanthomonas oryzaepvoryzae</i>	Hastuti <i>et al.</i> (2012)
5.	<i>Bacillus safensis</i> B21	Iturin A2 and Iturin A6	<i>Oryza sativa</i>	Inhibition in incidence of <i>Magnaporthe grisea</i>	Rong <i>et al.</i> (2020)

MICROBES AS SOIL DECONTAMINANT

Green revolution has provoked several adverse effects to the environment due to indiscriminate use of pesticides, herbicides, and nitrogen fertilizers. Application of these products promotes the accumulation of toxic compounds in soils. Crop plants can absorb these compounds from soil, representing a latent problem to the human and plant health. It has been demonstrated that some pesticides can be absorbed from soil by potatoes (Juraskiet *al.*, 2011) and the highly recalcitrant compound TNT can be absorbed by maize plants (Van Dillewijn *et al.*, 2007). Although European countries have prohibited use of such pesticides in their lands, but many regions in the world are continuing to use them. Microorganisms can carry out both the plant growth promotion and the bioremediation of contaminated soils derived from intensive farming. PBM are good rhizomediators as they can decompose phenols, petroleum, pesticides, xenobiotics in the rhizosphere during a plant-microbe interaction. *B. unamae* has the potential to fix nitrogen, produce phytohormones, siderophores and other inhibitory substances, ACC-deaminase activity, and degradation of toxic aromatic compounds (Caballero-Mellado *et al.*, 2007; Onofre-Lemus *et al.*, 2009).

Bioremediation is an innovative technology that is frequently being used for cleaning-up of polluted sites. The process can be carried out by using indigenous

microorganisms or by adding an enriched culture of microorganisms (Pandey and Fulekar, 2012). A huge number of micro-organisms have been isolated which are capable of degrading organochlorine, organophosphate, carbamate, pyrethroid pesticides secreting different enzymes. A group of micro-organisms including *Ochrobactrum* sp, *Castellaniella* sp, *Variovorax* sp, *Pseudomonas* sp, *Psychrobacter* sp, *Sphingomonas* sp, *Enterobacter* sp, *Bacillus pumilus*, *Burkholderia* sp, *Acinetobacter radioresistens*, *Pseudomonas fredenksbergensis*, *Serratia liquefaciens*, *Serratia marcescens*, *Pseudomonas putida*, *Burkholderia gladioli*, *Micrococcus* sp, secreting enzymes Organophosphorus hydrolase (OHP), Organophosphorus anhydrolase (OPAA)), Laccase, *Aspergillus* enzyme (A-OPH), Penicillium enzyme (P-OPH) that degrades organophosphate pesticides viz., Lgepal CO-210, Lgepal CO-520, chloropyriphos, Isoproturon, Fenitrothion, Methyl parathion, Dimetoate, Malathion, Diazinon, Prophenofos, Propiconazole, Diuron (Singh and Walker, 2006; Ortiz-Hernández *et al.*, 2013). Another group of micro-organisms *Alcaligenes eutrophus*, *Acetobacter aerogenes*, *Penicillium miczynskii*, *Aspergillus sydowii*, *Trichoderma* sp, *Penicillium raistrickii*, *Trichoderma viridae*, *Pseudomonas* sp, *Micrococcus* sp, *Arthrobacter* sp, *Bacillus* sp, *Pseudomonas aeruginosa*, *Burkholderiacepaeia*, *Arthrobacter* spKW, *Aspergillus niger*, *Flavobacterium* sp, *Pleurotus ostreatus*, *Streptomyces* sp, *Ganoderma australe*,

Phanerochaete chrysosporium secretes dehydrogenase enzyme capable of degrading organochlorine pesticides such as DDT, Dieldrin, Endosulfan, 1,4-Dichlorobenzene, Lindane, DDE, DDD, Heptachlor epoxide, Heptachlor O, Toxaphene O, Aldrin, Endrin O, Dieldrin O. *Achromobacter*, *Pseudomonas*, *Flavobacterium*, *Mesorhizobium*, *Ralstonia*, *Bacillus*, *Arthrobacter*, *Rhodococcus*, *Mesorhizobium*, *Ralstonia*, *Bacillus* secretes carbofuran hydrolase which degrades carbamate pesticide Carbofuran, EPTC (Huang *et al.*, 2018). *Pseudomonas*, *Serratia*, *Aspergillus niger* degrades pyrethroid with enzymes Pyrethroid hydrolase, Carboxyl esterase. (Singh and Walker, 2006; Ortiz-Hernández *et al.*, 2013, Bhatt *et al.*, 2019)

Soil bio-reclamation: Due to diverse metabolic capabilities of bacterial cells, they are capable of survive in extreme conditions from warm and humid to cool dry places, densely populated with vegetation to deserts and barren sites, even in the highest latitudes. As such, microbes use different enzymatic pathways which are a part of their metabolic activities to convert the toxic substances in soil to less toxic compounds. There are different mechanisms by which microbes can successfully degrade the toxic elements. Some micro-organisms are capable of forming biominerals with the toxic elements in environment either inside or outside their cells (Moraga *et al.*, 2017). Some excrete toxic components which sequester and eliminate toxic agents from their interior and excrete chelating compounds to immobilize or solubilize toxic substances in the exterior membrane (Gupta and Diwan, 2016). Another important strategy among the bacterial population is forming of the biofilm or exopolymeric substances (EPS) around their cell which help them remove toxins from the environment by immobilizing or flocculating them from a solution (Garrett *et al.*, 2008)

Biological water reclamation: In the municipal wastewater treatment plants, gram-negative bacteria belonging to Proteobacteria are predominant group (21-65%). Among which Betaproteobacteria is the most abundant class and are largely responsible for the elimination of organic elements, excess nutrients, hazard chemicals, heavy materials. Other groups of bacteria present are *Bacteroidetes*, *Acidobacteria* and *Chloroflexi* (Wang *et al.*,

2012). The ones which are found most abundantly are *Candidatus*, *Tetrasphaera*, *Microthrix*, *Trichococcus*, *Rhodoferrax*, *Hyphomicrobium* and *Rhodobacter* (McIlroy *et al.*, 2015). Among fungi, Ascomycetes are the most common, accounting for 6.3 to 7.4% of micro-organisms.

Wastewater treatment is usually done in two stages- Primary and secondary treatment. Biological wastewater treatment with the application of microbial biodegraders is done during the secondary treatment (Mittal, A. (2011). It occurs in three environmental conditions: aerobic, anaerobic, and anoxic. According to market forecast 2025, the market size of biological wastewater treatment is USD 8.7 billion in 2020 and is projected to reach USD 11.1 billion by 2025, at a CAGR of 5.1% from 2020 to 2025. The aerobic segment accounted for a larger share in the market, while the anaerobic segment is expected to grow at a higher CAGR during the forecast period. The large share of the aerobic segment is due to its low capital cost, simple design, and efficiency (Markets and Markets, 2020).

EPILOGUE

Green revolution resulted through the adoption of newer technologies including high yielding varieties of cereals, irrigation expansion and judicious use of chemical fertilizers, well supported by replicated package of practices superseding the old traditional practices. However, the post-green-revolution brought some negative consequences of late, especially issues relating health of natural resources and environment, thereby, challenging the sustainable development of agriculture to great an extent. The next question comes into mind is, how to sustain the growth of agriculture at a pace matching with current rate of population growth. Of the many options, microbial technology is considered standalone alternative to safeguard soil-plant-human health in a continuum mode. Biopesticides research and development, inclusive of microbial inoculants such as enzymes, secondary metabolites, biosurfactants, phytohormones, flavonoids extracted from the beneficial microbes are gradually paving a new way towards next green revolution. With an additional safeguard, sustaining the next green revolution would possibly be not so difficult as residue free green technology.

Certain quality issues like the original and true to type of biocontrol strains, efficacy, minimum spore concentration, physical parameters of formulated products and free from contaminants and chemical pesticides would come in the way of largescale popularisation. But, such interventions could find a much better popularity with initiation of many of the start-ups and biopesticides development as small scale industry, besides little difficulty in the label claim during the tedious process of registration of well proven biopesticides. Very often biopesticides

response in field is way low over *in-vivo* crop responses, warranting more robust R&D to popularise biopesticides from lab-to-land. Shortcomings related to production of microbial enzymes such as lack of efficient strains, high production costs, inadequate formulation design, and instability at various conditions also need to be overcome. More research is needed towards integrating biological agents into production system of developing countries, considering farmers as 'policy takers' rather than 'policy makers'.

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