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Rhizosphere competence and applications of plant growth-promoting rhizobacteria in food production – A review

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ABSTRACT

Sustainable food production, among other non-intensive production systems, involves the important interactions of myriads of plant growth-promoting microorganisms, plant, soil, soil fauna, and production of utilizable carbon in the rhizosphere capable of enhancing soil health, plant growth, and protection that lead to increased crop productivity. Plant growth-promoting rhizobacteria (PGPR) including symbiotic rhizobia and free-living rhizobacteria possess traits that help to enhance plant growth due to their many modes of action that start with the ability to colonize both the intracellular and extracellular rhizosphere niche in their search for a carbon source and reduction in the free use and quantity of agrochemicals. In the past few decades, the focus on developing biosafety agro-products has shifted from agrochemical-based applications to a more sustainable system without posing negative impacts on the soil microflora or fauna. The present review focuses on the application of PGPR inoculants on soils and seeds to improve biological nitrogen fixation, solubilization of phosphate, and secretion of phytohormones required for growth, especially application in a pressured environment. We discuss how PGPR enhances nutritional regulation and hormonal balance in plants, bacterial taxa enrichment, and improvement of carbon sources utilization beneficial for plant growth. We highlight the antagonistic and synergistic interactions with microorganisms within the rhizosphere and beyond in bulk soil which indirectly boosts plant growth rate and induces resistance against phytopathogens. While soil-borne pathogens continually oppose the functions of these microorganisms, PGPR has improved diverse strategies in the form of agro-compatibility, root colonization, nutrient, iron, and space competition, systemic resistance, antibiotics synthesis, lytic acid, hydrogen cyanide, and siderophore production for advanced food production. Finally, we highlighted the roles of PGPR in phytoremediation, techniques of applying microbial inoculants to enhance plant growth and commercialization of PGPR products and the challenges developing countries have to defeat.

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Introduction

Intensive agricultural practices that include the prolonged use of inorganic fertilizers and chemical pesticides have led to huge agricultural and ecological damage. The excessive use of chemical fertilizers exerts damaging impacts on soil microorganisms, altering the fertility status of soil and polluting the environment. The progressive increase in human population is projected at more than 9 billion by 2050 worldwide. According to the United Nations, Africa's share of the global population is predicted to grow from 17% in 2020 to 26% by 2050 and 39% by 2100, while Africa's most populous nation, Nigeria population is expected to surpass that of the United States by the 2050 [1,2].

Sustainable food production has become the main need to improve food security globally, particularly in developing nations. Ensuring food security is one of the challenging tasks faced by many nations in developing countries. Such challenge faced by mankind in the past decades has resulted in an impending negative effect on the human race which needs to be addressed without delay [3]. One of the problems in addressing food security is that the ecosystem is already under stress and more people are undernourished. This makes it very difficult to conquer the challenge of food security in many developing nations [4].

Moreover, the majority of farmers especially in developing nations practice substantive farming, but nowadays the rise in mechanized farming practice and the use of agrochemicals as external inputs are out of control, which adversely affect the soil environment and biodiversity [5,6]. Crop yield in the majority of the developing nations is the lowest compared to the developed nations of the world. Many of these developing countries lose crops worth billions of US dollars yearly; the intensive agricultural practices destroy the ecosystem, land degradation, diseases, and pest infestation. Still, some countries in developing countries such as Nigeria, South Africa, Zambia, Ethiopia, Mozambique, and Liberia rely on agriculture and have experienced an increase in their gross domestic product (GDP) growth in agriculture [7].

Although the advantageous outcomes of chemical-based fertilizers and pesticides are instantaneous, the indiscriminate and prolonged usage of these harmful chemicals in agriculture is dangerous [8]. Artificial chemical inputs in the form of chemical fertilizers and pesticides destroy the soil structure, resulting in the depletion of available nutrients, contamination of water resources that alter the soil biological population, and degradation of the environment [9,10]. They ultimately pose negative impacts on crop yield and increased susceptibility towards biotic and abiotic stresses. Furthermore, humans are exposed to these harmful chemicals through the possible entrance from the food chain as heavy metals [11]. The resultant effects of these dangerous chemicals used for agricultural purposes over time cause illnesses including cancers in humans and also pollute the environment [8].

The progressive increase in food production is essential for the development and continuity of developing countries and poor economies. Recently, many new agricultural technological innovations have evolved and should be implemented by policymakers because the growth and development of nations would rely on advanced and environmentally friendly technologies such as hybrid disease-resistant species, modern management practices, and climatic modified seeds [9,12]. The acceptance of these technologies by farmers is quite slow, due to some negative consequences over time, high cost, and dependability. With respect to these concerns, the use of novel agricultural beneficial microorganisms for sustainable agriculture will be an alternative strategy to increase crop yield without having any long-term negative effects on the ecosystem [13,14].

Rhizosphere competence refers to the ability of any microorganism to grow, function, and compete with others for critical nutrients and exudates generated by plant roots, as well as their ability to colonize the root surface of host plants in rhizospheric soil [9,12]. Most indigenous soil microorganisms, in particular rhizobacteria residing in the vicinity of plant roots, perform different beneficial interactions with plants and play critical roles in sustainable agriculture by improving soil quality and health, and making nutrients readily available to plants [15]. These distinctive groups of microorganisms possess characteristics to stimulate plant growth due to their interactions with the rhizosphere and are called plant growth-promoting rhizobacteria (PGPR) [16,17]. PGPR interacts and carefully selects their mutual partners through natural selection, creation of host specificity, and sensitivity to environments where microbial diversity is few [18]. PGPR improves plant growth through different mechanisms such as increasing the availability of nutrients to plants and producing essential metabolites without influencing the environment negatively. Additionally, PGPR stimulates the solubilization of phosphorus and other nutrients, increases iron acquisition in plants through the production of siderophore, produces antibiotics and other compounds that prevent the growth of phytopathogens, bioremediation of contaminated soils, can help plants tolerate abiotic stress such as extreme temperatures, salinity and drought, and also mitigate the adverse effects of various stresses [9,10].

Most PGPRs can survive adverse environmental conditions such as drought, salinity stress, unavailability of nutrients, and high heavy metal contamination [9]. PGPR triggers major biological functions in the soil but also have been engineered to improve crop productivity through healthy competition in the soil and breakdown of nutrients [8,19]. Plant growth-promoting rhizobacteria (PGPR) have been found to have numerous beneficial effects on plant growth, health, and development, as well as on the environment. The application of PGPR in sustainable agriculture and environmental remediation has the potential to improve crop nutrition, increase crop production, enhance crop yield, and disease management, and mitigate environmental hazards caused by the misuse and overuse of chemicals. However, the efficacy of PGPR in promoting plant growth and development depends on the particular bacterial strain and the conditions in which it is used. Consequently, the selection of a specific strain is critical in obtaining maximum benefits in terms of improved plant growth and development [10,16,17]. This review aims to present the various mutualistic interactions of PGPR with plant roots in the rhizosphere, its major plant growth-promoting traits, and mechanisms of action, as well as the commercialization of PGPR products and the challenges developing countries have to overcome, are also discussed.

Rhizosphere: the 'epicenter' of microbial activities

The rhizosphere region connects the plant roots to the soil from which organic and inorganic substances and signaling molecules are transferred into other soil regions allowing lasting mineralization processes controlled by agricultural important microorganisms and their activities [20,21]. The rhizosphere houses numerous groups of organisms interacting with the plants and other

Table 1
Some commercially produced PGPR, mechanisms of action, and plant responses.

PGPR	Product/ Country	Plant	Mode of action	Plant response	References
Paenibacillus helianthi sp. nov., *Streptomyces* sp., *B. subtillis*	BIOVED, AgBio, USA, Hungary, Italy	Sunflower	Nitrogenase activity; nitrogen fixation	Increased nutrient content and dry weight	[26–28]
P. thivervalensis, Azospirillum, A. brasilense, Paenibacillus polymyxa, Lipoferum, Pseudomonas putida, Serratia marcescen, P. fluorescens,	AtEze, Spot-Less, BlightBan, frostban, Bio-save- USA, India, Iran, Pakistan, Brazil	Maize (*Zea mays*)	The hydrolytic enzyme, phosphate solubilization, Siderophore and IAA production	Increase tolerance to salt; Increase plant leaf and height	[10,29]
Pantoea agglomerans RK-92, , *B. megaterium* TV-91C, *B. subtilis* TV-17C, *B. megaterium* TV-87A, *Bacillus megaterium* TV-3D, *B. megaterium* KBA-10	EcoGuard, Kodiak, Subtilex, Yeild-Shield, Companion, Bioyield, -USA, Turkey	Cauliflower (*Brassica oleracea L.*)	Nitrogen fixation, Phosphate solubilization, IAA production	Nitrogenase activity, phosphate solubilization, IAA production, Increased mineral nutrient uptake	[29]
Bacillus, B. amyloliquefaciens, Bacillus subtilis, B. pumilus, P. fluorescens, Serratia marcescens	BIOVED, HiStick N/T, Mepplus– USA, Brazil, India, Egypt, Hungary	Tomato (*Solanum lycopersicum*)	Siderophore and IAA production, phosphate solubilization Nitrogen fixation, antiviral activity	Increased crop yield and nutrient uptake, nutrient content, biologically controls viral pathogens	[29]
B.cereus, E. cloacae, B acillus drentensis, Providencia vermicola	Sonata, Serenade- USA, Saudi Arabia, India,	Mung bean (*Vigna radiata)*	N-fixing and phosphate solubilization Siderophore and IAA production,	Increases nutrient uptake, Enhance gaseous exchange, photosynthetic	[30]
Ama-2, *Rhizobium, B. pumilus* Sol-1, *Brevundimonas* Kro13, *Alcaligenes* sp. Mal-4, *Kluyvera ascorbata* SUD165, *P. putida, Ochrobactrum,*	Egypt, Pakistan, Bangladesh		Activities of nitrogenase, ACC-deaminase, and acetylene reducing	pigments, and water retention. Improved growth and seed yield, Increases the stimulation of mung bean plant growth reduces Pb and Cd uptake decreases the toxic effect of chromium to seedlings by reducing Cr (VI) to Cr (III)	[30]
Bacillus spp., *B. amyloliquefaciens*	Germany, USA, Iran, China	Cotton (*Gossypium)*	Antibiotic activity; N-fixing and phosphate solubilization, Production of plant hormones (auxins, cytokinins, and gibberellins)	General improves growth and biocontrol of phytopathogens	[31]
Bacillus simplex T7, *Pseudomonas putida* BA-8	Ecosoil-USA, Turkey	Grapes (*Vitaceae family*)	Antibiotic activity, IAA production, phosphate solubilization, N- fixation	Promote grafting capacity at nursery conditions	[26,29]
B. polymyxa (LAB/BP/01) *B. subtilis*	Inomix ® biostimulant Spain	Cereals	Production of antimicrobial compounds and metabolites induced plant resistance to pathogens, uptake of nutrients	Improve plant growth and control of pathogens	[27]
Azospirillum sp.	Nitrofix ®Cuba	Wheat, barley, carrot, maize, cabbage	Secretion of hormones, nitrogen fixation	Enhance nutrient and water uptake, stimulate root development, generally promote plant growth	[32]
B. subtillis BR62, *Rhizobium* sp. base formulation *Peanibacillum durus* PD74	Micosat F® cereal Italy Mamezo®, Processing Seeds®, Hyper Coating Seeds ® Japan	Tomato, soybean, cereals, beet, legumes, sunflower	Nitrogen fixation, HCN production, produce enzymes that can destroy the cell wall of pathogenic fungi	Increase plant height, shoot dry weight and nodules	[21]
Azotobacter, P. fluorescens, phosphobacteria	Greenmax Agro Tech Life, Gmax, Biosink -India	A wide range of plants	Nitrogen fixation Solubilize phosphate, produce siderophore, alkaline phosphatase and IAA	Increase crop yield, nutrient uptake, resistant phytopathogens	[33]

microorganisms, such as archaea, bacteria, fungi, and their environment [22]. Most microorganisms found in the rhizosphere improve plant growth through different mechanisms.

Plants typically secrete and discharge a significant amount of carbon fixed by photosynthesis, and the soil microbial community and their roots use the majority of these discharged materials known as root exudates, which include primary metabolites (organic acids, sugars, inorganic ions, vitamins, amino acids, enzymes, purine, and gaseous molecules) as a source of food [9,10]. Collection of these root exudates has been performed using both qualitative and metabolomic approaches, including sampling root exudates from plants grown on soil and obtaining root exudates from in vitro cultures or hydroponic grown plants. However, in root exudates the composition of soil will be instantly changed which is attributed to their adsorption to the soil particles as well as microbial degradation, actually, the exudation from soil microbial communities also modifies plant exudation profile [23]. Due to the challenges of root exudation from plants grown in the field and the low reproducibility of the process many factors influence the exudation process such as biotic and abiotic stress conditions like toxic ions, such as trace elements (heavy metal ions), extreme temperature, high salinity (generating both ion toxicity and osmotic stress), nutrient starvation (P deficiency) and water status, has led to other approaches which exclude the extension of soil-culture [7].

Plant culture in artificial field conditions can modify exudate composition, factors such as aeration in vitro culture or hydroponic cultures in temporary immersion systems (ITS) have impacted on root exudate composition and plant development. The main benefit of these in vitro sterile systems is the reproducible exudation pattern which is important to understand the effects of stress factors and nutrient deficiency [24]. Hence, some researchers highly recommend this method for exudate collection to prevent the presence of rhizosphere microorganisms with the ability to affect the composition of root exudate [25]. In some cases, root exudates are collected from in vitro or hydroponic cultures using plants grown in the soil while in most studies root exudation is from in vitro or hydroponic cultures, where the liquid medium containing compounds is obtained. In respect to this, root exudates are collected by various physicochemical processes, which include through sorption filters buried in the ground [26] or extraction processes with different solid or chemical-phase extraction. The in vitro methods allow the culture of excised organs such as shoots or roots or entire plants. However, this can influence root exudate composition, because many studies have reported that root exudation is affected by the presence of shoots in the culture, as they are in control of the CO_2 absorption by photosynthesis. In some plants such as watermelon [27], and a combination of eggplant and tomato [28], the modification in the aerial tissues and as changing the scion of grafted plants were reported to affect root exudation processes.

Moreover, traditional stationary methodologies of culture for obtaining root exudates from in vitro-cultured plant tissues. Recently, many automated TIS have evolved, some allow forced ventilation, CO_2 enrichment, culture hairy roots and cell suspensions. The first and one of the most commonly used systems is RITA® (Recipient à Immersion Temporaire Automatique) which was originally developed for plant mass propagation and also has been approved for metabolites from root culture capable of reducing hyperhydricity challenges in plant material grown in vitro. The major advantage is that ITS substantially increases metabolite and biomass production, hence their use for obtaining specific metabolites from root exudates. Some metabolites obtained from root cultures using TIS are betalains from *Beta vulgaris* (beet) having colorant and antioxidant properties [24].

These plants' exudates have various available nutrients, playing a significant role in attracting different groups of microorganisms to the rhizosphere [19]. Besides, the available nutrients in plants, they also depend on the interactions among microorganisms in the rhizosphere using these exudates, thereby creating a connection between microorganisms and plants [29]. The capability of microorganisms to colonize the host plant roots and improve soil health is determined by the efficiency and success of PGPR as an inoculant for crops [30]. Examples of commercially available PGPR are listed in Table 1. Only microbial species that can strive and survive the competition in the rhizosphere are effective in expressing many genes and communication between cells through quorum sensing (a cell-to-cell microbial communication process that uniquely controls various physiological and metabolic activities) can emerge as promising PGPR [18].

Beneficial plant-microbial interactions that exist in the rhizosphere between the plant roots and the microorganisms can be mutualistic (both organisms benefit from each other), commensalistic (one of the organisms benefits, while the otherobtains neither benefit nor harm), and amensalistic (one of the organisms is harmed while the other is unaffected) forming the bases for rhizobacteria to improve crop yield [6]. However, for rhizobacteria to be classified as PGPR, it must be able to colonize the root surface, to can survive, proliferate, and compete with other indigenous and nonresident microbiota [34]. Above all, it should promote plant growth [35]. In our recent study, *Firmicutes* (17 to 51%), *Proteobacteria* (18 to 36%), and *Actinobacteria* (7 to 38%) were the most abundant PGPR characterized in the sunflower rhizosphere soil. These rhizosphere microbiomes are part of the complex food web that utilizes the large quantity of nutrients released by the plant and have been widely recognized to enhance plant health and growth [34]. Similar studies have postulated that plants may modify the rhizosphere microbiome to their advantage by selectively stimulating microorganisms with distinctive features that are beneficial to plant growth and health [5,6]. In the rhizosphere, many bacterial species have been recorded to symbolically interact with host plants as a result release organic plant growth-promoting substances in the form of exudates as important nutrients for soil bacterial growth and metabolic roles. This has been achieved by studying various microbial species, and their functional roles through next-generation sequencing techniques, the plant cultivars, and the rhizosphere. PGPR demonstrates different mechanisms of action generally to promote plant growth either directly or indirectly, or both [36].

PGPR improves plant growth directly by enhancing nutrient acquisition, mainly essential minerals such as phosphorus, vitamins, and nitrogen from the environment. Furthermore, other growth factors such as enzymes, water, production of growth hormones, solubilization of phosphorus, and iron sequestration by siderophore in the absence of pathogens [37,38]. Indirect mechanisms reduce the effects of plant pathogens by protecting plants from phytopathogens. Indirect mechanisms such as competition, production of antibiotics, antifungal metabolites, production of hydrogen cyanide, siderophore, and induced systemic resistance indirectly promote plant growth [39].

Kloepper and Schroth [16] postulated how PGPR alters the rhizosphere niche; the authors buttressed the diverse effects of PGPR on plant growth and development upon inoculation [36]. The application of microbial inoculants, such as PGPR is beneficial for stimulating plant roots for growth, pest control, plant stress control, and destruction of plant diseases leading to agricultural productivity [37,38]. Plants naturally can fight against diseases, but the application of microbial inoculants as biocontrol agent help advance these abilities.

Besides, numerous live microorganisms, such as *Rhizobium, Anabaena*, *Pseudomonas*, and *Azotobacter* species are used as microbial inoculants to improve nutrient uptake and nutrient pool. These microbial inoculants can be incorporated into soil, seeds, and plants for enhancing biofertilization and biostimulation serving as an eco-friendly and substitute to chemical fertilizers [39,40]. The synergistic effect caused by microbial inoculants, and resident microorganisms improves plant nutritional activities that involve breaking down, solubilization, and mobilization of nutrients, hence increasing crop yields have reduced the use of chemical fertilizers. The acceptance and application of microbial inoculant as plant improving agent is spreading widely in many developed countries where agriculture is the key driver of the economy [41]. Nevertheless, despite the numerous benefits of PGPR, there are some toxic effects associated with the use of PGPR, for example, when PGPR is used in contaminated soils may lead to the release of heavy metals into the environment [8]. It is important to note that the toxic effects of PGPR are not well documented, and more research is needed to fully understand the potential risks associated with their use. The overall benefits of using PGPR in agriculture and environmental remediation outweigh the potential risks, but caution should be exercised when selecting and using a particular bacterial strains [9,10,16,17].

Direct mechanisms of plant growth promotion by PGPR

Biofertilization

Biofertilization refers to the application of microbial inoculants on soil, plant surfaces, or seeds, which is important to enhance crop production. The microbial inoculants colonize the rhizosphere and supply essential nutrients required for plant use resulting in improved plant growth. The application of microbial inoculant improves the soil texture, such as soil porosity and soil aggregate structure, etc. These biofertilizers with relatively low density have no negative impacts on the environment and are non-toxic in even high concentrations without having any adverse effects on crops and underground water because they do not contain heavy metals such as arsenic, mercury, lead, vanadium, and cadmium. Studies have proved that these heavy metals could alter the composition and diversity of soil microbial population and nutrient cycling [42]. Ultimately, these chemicals could find their way into the food chain causing diseases such as stomach cancer [43].

Biofertilizers are formulated products of microbial inoculants in a liquid or powdered carrier containing PGPR as the active ingredient which, when applied to the seeds or into the furrows,enhances plant growth and increases yield, in a wide range of agricultural crops [42]. Many studies have documented the critical role biofertilizers play in maintaining soil health, promoting long-term soil fertility, increasing organic farming, and reducing the use and application of chemical fertilizers [39,40]. To use biofertilizers, there must be an efficient formulation of microbial inoculants, proper selection of suitable carriers, and construction of adequate application methods.

In a study by O Babalola and A Akindolire [44] which characterized PGPR from rhizospheres of eight food crops plants in South Africa, different PGPR strains belonging to *Burkholderia gladioli, Vibrio fluvialis, Serratia plymuthica, Pseudomonas fluorescens, B. cepacia, P. putida, S. ficaria, P. luteola, Erwinia* spp, *Aeromonas hydrophila, Rahnella aquatilis, Acinetobacter baumannii, A. calcoaceticus, Escherichia vulneris, Proteus penneri, Ewingella americana,* and *Shigella* spp. were being used with productive effects as biofertilizers. All these strains produced hydrogen cyanide and ammonia to indicate their functions as biofertilizers in increasing crop productivity

Nitrogen fixation

Nitrogen (N) is one of the most essential nutrients for plant growth and development that ultimately increases crop productivity. Although atmospheric air is composed largely of N gas (N_2) at 78 %, N is one of the most limiting nutrients for crop production worldwide [45]. This is because atmospheric nitrogen (N_2) is very stable due to the strong triple bond between the two N atoms that require a large amount of energy to break, and thus plants cannot convert it to its usable form [46]. Only a few prokaryotic organisms called diazotrophs have the enzymatic machinery to break the strong bond that held the two N atoms. Atmospheric N_2 is biologically fixed through symbiotic means by the root nodulating Rhizobia group or non-symbiotically by the free-living PGPR strains such as *Azospirillum* and *Azotobacter* spp. and endophytes such as Cyanobacteria (*Gluconoacetobacter diazotrophicus, Anabaena, Noctoc* and *Azocarus*) provide 7 and 80 kg N_2 per hectare annually [47]. This process of converting atmospheric N_2 into a usable form of N by the enzymatic action of diazotrophs described above is called Biological Nitrogen Fixation (BNF).

The BNF is an economically beneficial and eco-friendly alternative to chemical fertilizers and accounts for up to 290 million tonnes of N per year providing N to plants [48]. For many years, rhizobia inoculants have been used for cultivating legumes as biofertilizers. Like the developed world, many sub-Saharan African countries such as South Africa and Namibia use rhizobia inoculants for N uptake. Due to its economic benefits, rhizobia have been widely used as a substitute for chemical fertilizers frequently used to boost the production of legumes [7].

The mechanism of N fixation requires a complex enzyme system such as nitrogenase mainly encoded by the *nif* genes. The *nif* genes are structural genes involved in the Fe protein activation, donation of an electron, activation of iron-molybdenum, and stimulation of plant regulatory genes needed for the synthesis and function of enzymes [49]. The *nif* genes are primarily found as a cluster of around 20–24 kb with about seven operons of 20 proteins variants. The advantageous impacts of *nif* genes have been well documented. Some

studies have also proved that *nif* genes can improve N fixation through genetic engineering. However, others argued how N fixation traits of plants could be enhanced and harnessed [7,50]. Nevertheless, the focus of these methods is still on promoting plant growth and sustaining N levels in the soil. A diazotroph is any microorganism that can grow without any source of external N. Diazotrophic microorganisms provide to the host plant fixed N that can be exchanged for fixed carbon which is then released as root exudates [51, 52].

Mineral solubilization and nutrient uptake

For a plant to grow optimally there is a need for adequate mineral nutrient supply. However, even with the presence of mineral nutrients in the soil, plants may still show deficiencies because of the unavailability and insolubility of essential mineral nutrients to support plant growth. PGPR solubilizes mineral nutrients such as potassium (K), iron (Fe), and phosphorus (P), and then makes it accessible for plant use [1].

P is the second major important mineral nutrient after N required for plant growth [53,54]. P is abundant in soil in both organic and inorganic forms but is insoluble and the uptake of available P by plants is minimal [40]. PGPR triggers plant growth by enhancing P solubilization. Plants absorb insoluble P forms such as monobasic ($H_2PO_4^-$) and diabasic ions (HPO_4^{2-}) ions. Unavailable P in the soil are in insoluble forms such as phosphotriesters (organic), apatite (inorganic), phosphor monoesters, and soil phytate or inositol phosphate [40]. Therefore to eradicate the challenge of P deficiency in soils, phosphatic fertilizers are frequently applied to the soil by some farmers [55]. To solve the problem, phosphate solubilizing activity of PGPR is important. PGPR mineralizes and solubilizes inorganic phosphorus by improving the root system. Hence, provide the available forms of P to the plants serving as potential biofertilizers [9, 10].

PGPR solubilizes inorganic phosphate by secreting low molecular weight organic acids, such as carboxylic acids, lactic acid, succinic acid, glycolytic acid, formic acid, fumaric acid, and propionic acid which lower the pH in the root area, consequently releases the confined forms of phosphate such as Ca phosphate in calcareous soil [56]. Generally, organic phosphorus mineralization occurs by various enzymes such as phytase and C-P lyases phosphatases that catalyze the hydrolysis of phosphoric esters [42].

Additionally, other mineral nutrients such as iron, and zinc are made available through the biosynthesis of compounds capable of promoting plant growth. Although mineralization and phosphate solubilization can be impacted by environmental factors, they play a critical role in agricultural sustainability, especially in soils short of P [57]. Another essential macronutrient is K. Naturally, the accumulation of K in soils is minute and the majority of the K is found in silicate minerals or insoluble rocks. The short supply of K in soil results in poor development of plant roots, tiny seeds, and low crop yields generally present as a concern to sustainable crop production. The search for solutions to alleviate the shortfall of K in soils has led to the innovation of environmentally friendly and economical options for K uptake by plants and to sustain the K in soils for improving crop production [29,41,58].

The solubilization of K occurs through the secretion of organic acids by PGPR. Interestingly, PGPR such as *Bacillus, Pseudomonas, Burkholderia*, and *Paenibacillus* species together with K solubilizing activity has been reported to provide the available forms of K to plants [30,55,59]. These microbial communities are referred to as potassium-solubilizing microorganisms (PSM) because they release unavailable K and make it accessible to plants. PGPR as potential biofertilizers can reduce the use of chemical fertilizers and eco-friendly presents an approach for crop production.

Biostimulation, production of plant growth regulators and bioactive compounds

The process of modifying the soil environment, during which PGPR is excited to produce substances capable of promoting plant growth is called biostimulation [58]. Biostimulants are biochemical substances that promote plant growth in the absence of pesticides, nutrients, or soil improvers [60]. The chemical structure is closely related to that of natural plant hormones and the mode of action is being estimated as the production of plant hormones or phytohormones [61]. Recently, PGPR based stimulants have been widely applied in agricultural practices globally, in many African countries. PGPR based stimulants directly or indirectly improve crop quality by enhancing nutrient uptake and tolerance to stresses such as salinity and drought [49,62]. Across the global markets, different PGPR formulations have been registered and are commercially available. Some of the commercialized species are *Enterobacter, Pseudomonas, Variovarax, Serratia, Bacillus, Azobacter,* and *Azospirillium* [60]. Although, their use in the cultivation of plants still presents a small fraction worldwide [57].

Organic compounds required in minute concentrations capable of manipulating the physiological processes and regulating plant growth are called phytohormones or plant growth regulators (PGRs) [63]. Phytohormones are phytostimulants that are chemical compounds required for the physiological and biochemical development of plant growth. *Pseudomonas, Bacillus, Acetobacter, Xanthomonas, Bradyrhizobium, Penicillium,* and *Aspergillus* are some genera of PGPR capable of producing phytohormones [16,64,65].

Globally today, agricultural systems are frequently impacted by different stresses (biotic and abiotic) which often affect plant growth and crop yield. Yearly, farmers experience 30-50% losses due to stress [64]. These stresses can be human induced or inherent. The common abiotic stresses are drought, temperature, salinity, and accumulation of heavy metals. Hence, stimulates plants and PGPR to produce the phytohormones. The phytohormones are found in minute amounts that exert control on the morphological, physiological, and biochemical processes, even the structure and genetic expressions in plants, and facilitate plant-microbial interactions. Therefore, it has had notable effects on soil microbial composition, soil fertility, and nutrient compositions, though the production of these organic chemicals is efficiently managed [29].

PGPR can enhance plant growth and development in stressed and natural conditions, therefore using competent microorganisms could advance agricultural sustainability and environmental stability. In stressed soil environments, PGPR produces specific low

molecular compounds such as rhizobiotoxine around the rhizosphere capable of controlling phytopathogens by suppressing ethylene production. Moreover, to increase survival under stressed conditions, particular bacteria transcription proteins regulate gene expression in plants [29,47,64]. *Arthrobacter* sp. AF-163 produced phytochemicals capable of improving drought stress and crop growth in sunflower plants by altering membrane integrity and reactive oxygen species movement and also built up glycine, proline, and betaine. Bacteria modulate the positive responses causing distinctive gene expression related to the activation and expression of transcription factors, ethylene and salicylic biosynthesis in stressed conditions [53].

PGPR such as *P. putida, Glucunobacter* sp.*, Agrobacterium* sp.*, Alcaligenes piechaudii, Micrococcus luteus,* and *Streptoverticillum* sp. synthesized high levels of ethylene, IAA, and hydrogen cyanide to reduce salt stress and enhance sunflower plant root growth, as a result improved root structure development, N content and nodule biomass [31,38,66]. PGPR producing auxin, cytokinins, and gibberellins ameliorated antioxidant enzymes of different plants' rhizosphere such as groundnut, lettuce and canola under gnotobiotic conditions. Hence, the mechanisms of action improved signaling molecules that affect gene expression, plant growth and yields [63, 67]. Various PGPR such as *Serratia, Rhizobium*, and *Pseudomonas* species used as bioinoculants to cultivate common crops such as rice, sunflower, and legumes produced phytohormones involved in nodule formation and elongation of the root system [2,68,69]. *A. thaliana* seedlings treated with ACC tolerated drought when compared to the uninoculated seedlings, and also increased in root hair length. Further investigation revealed that the mechanism is dependent on ethylene [50].

Sunflower plant's soil treated with *Costus* sp. triggered the production of phytohormones (auxins, cytokinins, and gibberellins) as a result boosted the plants' growth compared with chemical fertilizers [70]. Nonetheless, PGPR improves plant stress tolerance and growth by reducing the use and damaging effects of inorganic fertilizers on soil health leading to environmental and agricultural sustainability.

Siderophore production

Among the essential minerals abundant in the rhizosphere soil needed by plants is Fe, which is not readily available for plant and bacterial use. Generally, ferric iron (Fe3+) is the major form of Fe on Earth but is insoluble. Plants and microorganisms require a sufficient amount of Fe, which is of concern in the rhizosphere where there is a high competition of Fe by plants and microorganisms. To alleviate this problem and make Fe readily available in the root region, PGPR secretes low molecular weight and high-affinity iron-chelating compounds called siderophores [54].

Ever since the report by Kloepper and Schroth [16] on the occurrence of enhanced plant growth by siderophores produced by *P. fluorescence*, the role of siderophores produced by innumerable PGPR strains in plant growth promotion has been reported in many studies [39,71]. Fig. 1 below indicates an *in-vitro* detection of siderophore production by PGPR isolates using the universal chemical (CAS-agar plate) assay. PGPR strains *P. fluorescence* KBS6-17, *Serratia marcescens* KBS-6H and *Enterobacter sokazaki*-NAS6B are shown to produce siderophores confirmed by the change in the color of the blue CAS agar media into yellow and orange after inoculation and incubation for 5 hours at room temperature [54].

Siderophores are grouped based on their functional capability as carboxylates and hydroxamates [58,72]. PGPR produces different types of siderophores, such as ferroxamine and Pseudobactin that chelate in plant rhizosphere and as a pointer to show that plants are

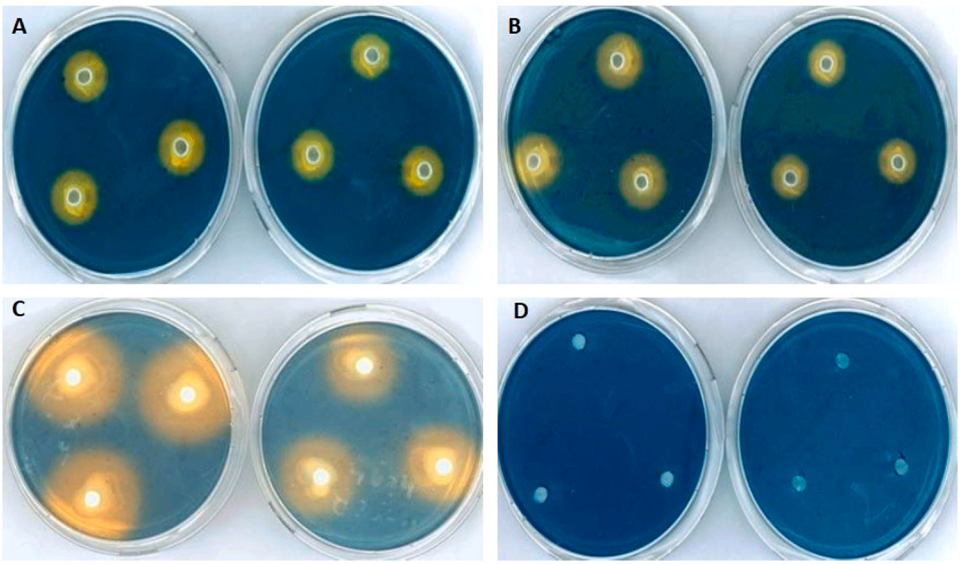

Fig. 1. *In-vitro* production of siderophores on CAS-agar plates by PGPR strains *P. fluorescence* KBS6-17 (A), *S. marcescens* KBS-6H and *E. sokazaki*-NAS6B. The production of siderophores is indicated by a change in the color of the CAS agar into yellow and orange when the siderophores sequester the Fe in the medium Source: [70].

capable of Fe uptake, which barely makes Fe accessible to pathogenic microflora and halt pathogens from obtaining Fe [20]. Furthermore, PGPR species such as Streptomyces spp. reportedly produce siderophores comparable to those produced by the plants [29,31,33]. Siderophores are known to be synthesized by microorganisms under iron deficiency stress conditions hence, to assure the growth and development of their cells. These microorganisms upsurge soluble iron in the soil, stimulate the chelation of iron, and make it accessible for themselves and also for the plants [73,74]. Sunflower plants inoculated with PGPR produced siderophores by stimulating plant cell division that enhanced root growth compared to uninoculated plants [75]. The ferric siderophore complex produced by *P. putida* and *P. fluorescens* is taken up by plants, resulting in the restriction of microorganisms from accessing the Fe in plant roots and other regions [58]. The siderophores synthesized by numerous PGPRs may be very useful for stimulating the absorption of iron in an iron-deficient environment by plants [76]. Also, the PGPR presence in soil induces the production of siderophores and helps plants reduce the toxicity of heavy metals in the stressed environment [77].

Indirect mechanisms of plant growth promotion by PGPR

Biocontrol of phytopathogens

Globally, diseases damage many crops and before harvest, a large percentage is lost. Many farmers use chemicals to control pests and this is becoming a concern due to health implications, hence PGPR offers a potential solution. Presently, awareness and acceptance of biopesticides are on the rise globally. Biocontrol is the use of microorganisms to diminish the frequent occurrence of plant diseases. Biopesticide selectively controls various organisms capable of causing diseases in plants without being toxic. PGPR is capable of controlling various phytopathogenic microorganisms and uses at least one of the following mechanisms including competition with pathogens for nutrients and specific ecological niches at the root region, the production of antibiotics and anti-fungicides, lytic enzyme, hydrogen cyanide production, induced systemic resistance, and signal interference [31,66]. Some biocontrol agents are highlighted in Table 2.

PGPR as a biocontrol agent exerts these mechanisms of action to antagonize pathogenic bacteria, fungi, and nematodes depending on their severity. *Pseudomonas fluorescence* is the most studied biocontrol agent because of its antagonistic actions against many phytopathogens [69]. Besides, PGPR manipulates the involvement and application of important rhizobacteria and their metabolites in suppressing the impact of phytopathogens, as a result, promotes plant health and minimizes loss during harvest [49,60]. Recently, farmers' interests and preferences have shifted to the application of integrated alternative pest control strategies that are viable, inexpensive, and eco-friendly, including spore-forming *Bacillus* and other microorganisms with biocontrol ability [58]. Likewise, Singh, et al. [78] reported that all ACCd producing PGPR isolates identified from sunflower soil had an antagonistic effect on *Fusarium oxysporum* that causes *Fusarium* wilt in the sunflower. Furthermore, the ACCd producing PGPR exhibited a minimum of four plant growth-promoting traits, including the production of phytohormones (IAA) and provided mineral nutrients, such as P and N fixation that significantly improved seed and vegetative growth parameters of the sunflower plant compared to the control.

PGPR produces antimicrobial secondary metabolites and siderophores to chelate Fe and conduct a multifactorial process that stimulates induced systemic resistance and relies on various compounds, including volatiles and c-LP surfactin [69]. The combined impacts of the strategies mentioned earlier would increase crop yield through adequate bioformulation of spores from viable PGPR and concentrated culture supernatants with antimicrobial metabolites.

Induction of systemic resistance

Some PGPR triggers physical or chemical changes that are related to plant defense, a process referred to as induced systemic

Table 2
Some PGPR use as a biocontrol agent against diseases and pathogens of different plants.

Plant	Disease/pathogen	PGPR	References
Sunflower	Sunflower wilt fungus	*Pseudomonas cepacia*	[27,28]
Beans	*Sclerotium rolfsii*	*Pseudomonas cepacia*	[30]
Maize	Corn earworm; *Fusarium miniforms*	*P. maltophila* *Enterobacter agglomerans, P. cepacia* strain 406 and 526	[32]
Rice	Rice sheath blight Rice root nematode *Rhizobium solani*	*Bacillus cereus, Streptomyces* spp., combined with *P. fluorescens* and *Burkholderia* sp., *P. fluorescens* Fp7 and Pf1	[56,74]
Tomato	Cucumber mosaic virus; Root knot nematode	*B. pumilus, B. amyloliquifacians* strain IN 937a; *P. chitinolytica*	[29,75]
Wheat	Septoria tritici Other diseases	*P. aeruginosa* strain Leci; *P. putida* strain BK8661 *Rhodococcus, Pseudomonas, Bacillus, Penicillium*, and *Beauveria* species	[53]
Barley	Powdery mildew	*B. subtilis*	[32]
Sugar beet	*Fusarium oxysporum, Rhizopus stolonifer, Phona beta, Pythium ultimum,* Cyst nematode	*P. fluorescens* F113 *P. fluorescens*	[32]
Mung bean	Root-knot and rot	*P. aeruginosa, B. subtilis*	[7,30]
Sugar cane	Red rot	*P. fluorescens other* PGPR	[53]

resistance (ISR) through the action of various phytohormones [79]. It is the mechanisms by which PGPR protects plants from phytopathogenic infection by eliciting the production of different metabolites such as jasmonic acid, salicylic acid, and ethylene which is released in the form of stress response signaling within plants. The production of these chemicals within the plants stimulates-stimulates their resistance ability against infection by various phytopathogens [36]. PGPRs such as *Azospirillum, Bacillus, Enterobacter,* and *Pseudomonas* species have been used as active ingredients in many biofertilizer products by farmers worldwide to activate induced systemic resistance. Such inoculants containing this PGPR are often applied on the seed coat before planting to initiate ISR against pathogens such as *Pseudomonas syringae* and *Erwinia trachephila* that cause leaf spot and bacterial wilt, respectively [53]. Common ISR elicitors are siderophore, MAMPs (when formed by pathogens are called Pathogen-Associated Molecular Patterns: PAMPs) are microbial molecules secreted by microorganisms, like chitin, flagellin, and lipopolysaccharides [79], cyclic lipopeptides, antibiotics (2, 4-diacetylphoroglucinol) and some volatile compounds such as acetoin, ketones, 2,3-butanediol, alkanes, alcohols, sulfides, alkanes, esters and sesquiterpenes [38,47].

Antibiotics production

Soil-borne pathogens are known for their harmful effects on plant health and crop yield. Globally, many crops are destroyed due to the devastating impacts of pathogens, which reduce crop yield, the severity is more in Africa where crop yield is reduced to approximately 40% and harvests further to 20% [19,41]. Recently, antibiotic production has been among the research areas extensively studied for the biocontrol of phytopathogens and has been the most effective treatment to suppress phytopathogens globally [58].

Antibiotics are low molecular weight compounds, among their numerous functions is to inhibit the proliferation of pathogenic microorganisms and their metabolic activities [80]. Antibiotic production is the major antagonistic activity to inhibit phytopathogen growth, thus playing significant roles in disease control and also can be used as biocontrol agents [66]. In recent times, antibiotics produced by PGPR have been employed to suppress the negative impacts of pathogens in the plant root environment. Such antibiotics include phycocyanin, oomycin-A, pyrrolnitrin, and 2,4-diacetyl phloroglucinol [39].

Studies have recorded that PGPR produces butyrolactones, zwittermycin A, xanthobaccin phenazine-1-carboxylic acids, pyrrolnitrin, kanosamine, iturins, fengycins phenazine, circullin, colistin, polymyxin, and 2,4-diacetylphloroglucinol (2,4-DAPG) with biocontrol effects on phytopathogens, such as *Fusarium oxysporum, Rhizoctonia solani, Podosphaera fusca,* and *S. griseoviridis* that cause plant diseases including leaf and seedling blight in rice, *Fusarium* root rot and tomato wilt [3,38,68].

These biochemicals are produced by *Bacillus, Pseudomonas* strains, *Stenotrophomonas* sp., and *Streptomyces* spp. as active chemicals against phytopathogens [20,40,81]. Some plant species naturally produce antibiotics that control plant diseases and are also produced by soil microorganisms, for example, Phloroglucinols [33,38,75,82]. The biocontrol agents control plant diseases and have contributed to increasing crop productivity.

Competition for nutrients and binding sites

Additionally, poor soil health is a concern globally, PGPR capable of producing biochemical substances with inhibitory effects on phytopathogens, has been used to improve soil health in many African countries such as Nigeria, Benin, and South Africa [2,44]. The inoculated PGPR as a biocontrol agent competes and in most cases out-compete the phytopathogens either for nutrients or binding sites in the host plant root region [49]. Such competition acts by constraining phytopathogens from binding to the plants, therefore making it relatively impossible to proliferate and infect the plant. For example, nutrient competition is the biocontrol mechanism of *Pythium aphanidermatum* [40].

Apart from inherent growth and availability of sufficient nutrients in the rhizosphere, some properties help PGPR compete favorably with phytopathogens and enhance their survival. These properties include the presence of flagella, chemotaxis, liposaccharides, and the use of synthesized exudates [58]. In the ecological niche competition, PGPR dominates the niche and prevents colonization by phytopathogens until the available nutrients, such as Fe are exhausted.

Recently, PGPR inoculants have been reported to have antifungal activity and compete against pathogenic fungi such as *Fusarium oxysporum* [83]. Similarly, the competitiveness of *Bacillus megaterium* in improving tomato plant growth has also been reported [75]. PGPR most times works in close association with other biocontrol mechanisms to halt the actions of phytopathogens [47,71,84].

Hydrogen cyanide production

Many PGPR produce hydrogen cyanide (HCN) in a process called cyanogenesis. Some of these PGPRs can be inoculated into the soil to stimulate the production of HCN [33]. HCN is a volatile, broad-spectrum antimicrobial compound implicated in the biocontrol of phytopathogens associated with Fluorescent *Pseudomonads* [38]. The cyanide ion obstructs the action of metalloenzymes, especially copper composed of cytochrome c oxidase. Secondary metabolite synthesized by Gram-negative bacteria is formed from glycine and catalyzed by the enzyme HCN synthase which is encrypted by biosynthetic genes such as *henA, henB,* and *henC* [59].

Moreover, HCN stops the electron transport to the target cells and disrupts the energy supply leading to the death of organisms [47]. The HCN interrupts the proper activities of enzymes and inhibits natural receptor mechanisms. Some strains of Fluorescent *Pseudomonads* produce HCN and they have been implicated in the elimination of soil-borne pathogens [75]. PGPR such as *P. fluorescens* strain CHAO, *Bacillus, Stenotrophomonas, Brevibacterium*, and *Pseudomonas* species isolated from rhizosphere soil produced HCN that inhibited the growth of pythium [33]. PGPR produces HCN in soil capable of altering the plant's physiological activities by stimulating

the formation of root hair [80]. Rhizobacteria are undeniably important in preserving soil fertility and improving plant growth and development. This improvement in growth occurs through a variety of pathways, however, some other studies show the opposite [85, 86]. Cyanide generation, for example, is recognized to be a feature of certain Pseudomonas species. Cyanide produced by bacteria is seen as both a growth promoter and a growth inhibitor in this context. Likewise, cyanide functions as a biocontrol agent against certain plant diseases [87], although it can also have negative effects on plant growth [88].

PGPR mitigation of abiotic stresses in plants

In many agricultural fields globally, crop productivity and yield are affected by many abiotic stresses that adversely affect crop production. Abiotic stresses such as drought, salinity, metal toxicity, acidic soils and nutrient deficiency (fertility stress) limit the growth and productivity of various economically important crops in manyagricultural fields globally. Apart from the intensive agricultural practices such as the prolonged use of external chemical inputs in many agricultural fields, the spreading impact of the global climate change causes numerous abiotic stresses on major crops worldwide. The predominant abiotic stresses such as drought, high soil salinity, nutrient deficiency, acidic soils and metal toxicity significantly reduce the growth and yield of most cultivated crops [89–91].

There are many mechanisms by which PGPR reduces the negative impacts of such abiotic stresses in plants and hence promotes growth indirectly. PGPR has been used as a biofertilizers in the past few decades as an alternative strategy to reduce dependence on chemicals while improving plant growth and yield in a sustainable way [92]. Many of these PGPRs have developed various mechanisms to exert beneficial effects on abiotic stress in plants. One of the mechanisms PGPR uses to alleviate abiotic stress in soils is by producing the enzyme ACC deaminase. Upon inoculation of PGPR with this ability, the ACC deaminase lowers the plant ethylene level, resulting in longer roots and providing relief frommany abiotic stresses. Examples of PGPR that significantly alleviated different abiotic stresses in numerous agriculturally important crops include *Bacillus polymyxa* under nutrient-deficient stress, *Azosprillum brasilense* on drought and salt stress, *P. fluorescence* on salt stress, *Pseudomonas fluorescence, P. putida* and *Azosprillum lipoferum* on drought to mention a few [91]. Furthermore, Bradyrhizobium elkanii produces rhizobitoxine, which has a twofold impact. It can relieve the deleterious effect of stress-induced ethylene production on nodulation because it is an inhibitor of ethylene synthesis [93]. Rhizobitoxine, on the other hand, is classified as a plant toxin since it causes foliar chlorosis in soybeans [94]. So far, the preceding discussion has demonstrated that, while PGPR is very successful in promoting plant growth and development, a few bacterial species can limit growth. However, this negative influence may occur only under certain conditions and by certain features. As a result, selecting a certain strain is critical for obtaining the greatest benefits in terms of better plant growth and development.

Fig. 2 illustrates the mechanisms by which certain plant growth-promoting rhizobacteria elicit induction of systemic tolerance (IST) in plants. In this interaction, various types of abiotic stresses such as drought, salinity, and fertility stress could be mitigated by the production of certain bioactive compounds by the PGPR residing on the root surfaces. For instance, the secretion of cytokinins by the

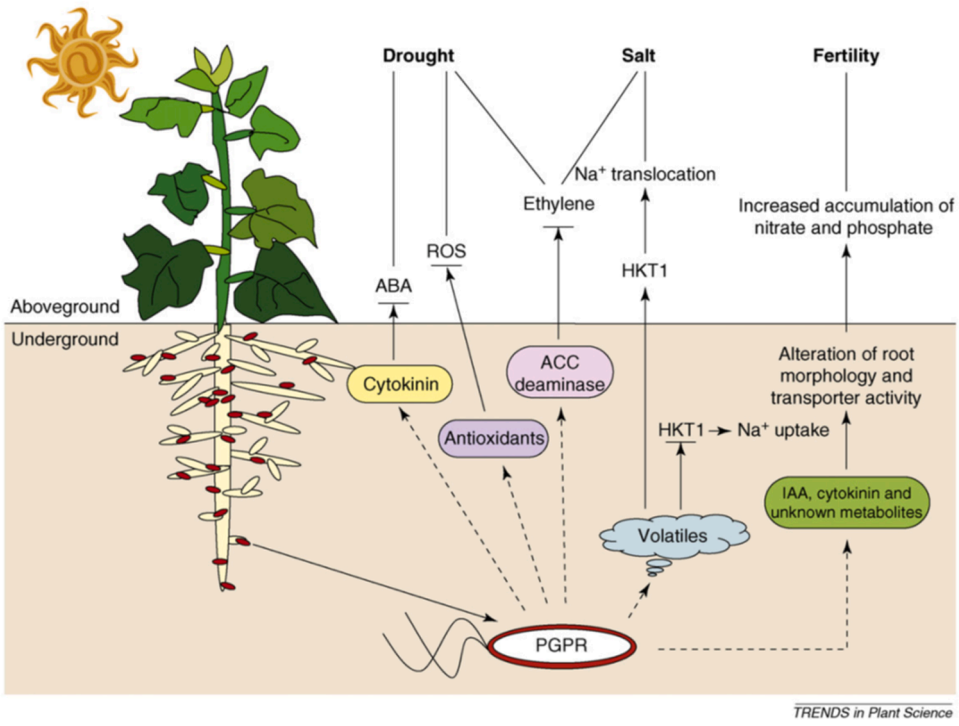

Fig. 2. A schematic representation of how PGPR is involved in the mitigation of various abiotic stresses including drought, salinity, and nutrient deficiency (fertility) stress and elicit induced systemic tolerance (IST) in plants. PGPR stands for plant growth promoting rhizobacteria. Source: [70].

bacteria can result in the accumulation of abscisic acid (ABA) in the leaves, which elicits stomatal closure under drought stress. Whereas the secretion of antioxidants and the enzyme ACC-deaminase by PGPR results in the degradation of reactive oxygen species (ROS) and that of the ACC precursor respectively [95]. These processes elicit induced systemic tolerance (IST) in the plants against drought and salt stress conditions and promote plant growth indirectly [38,47].

Lytic enzyme production

PGPR synthesizes extracellular enzymes known as lytic enzymes. These enzymes hydrolyze polymeric compounds such as protein, cellulose, chitin, and hemicellulose as an important form of biocontrol [69]. Lytic enzyme as a multifunctional naturally occurring protein protects plants from climatic factors, abiotic, and biotic conditions [58]. Lytic enzymes are secreted by PGPR to directly suppress the activities and growth of different phytopathogens, which are of concern. The presence of lytic enzymes in soil degrades a different range of compounds from the plant, and also destroys or digests fungal pathogen cell wall structure. The application of PGPR in soil activates the most critical eco-friendly mechanism (hydrolytic enzyme production) for the suppression of soil pathogens [38].

Similarly, PGPR produces and stimulates beneficial enzymes such as lipases, chitinases, proteases, and glucanases involved in the lysis of the cell wall. These enzymes degrade plant residues and nonliving organic matter to obtain carbon nutrition. In an experiment, *Myxobacteria* produces a lytic enzyme that has shown effectiveness against fungal phytopathogens [36]. Plant diseases such as bipolaris leaf spot induced by *Pythium* sp. and *Bipolaris* sp. are reportedly suppressed by glucanase and beta-1, 3-glucanase produced by *Lysobacter and Lysobacter enzymogenes* strain C3, respectively [5]. Furthermore, *Serratia marcescens* produces chitinase that has an antagonistic effect on *Sclerotium rolfsii.* Nevertheless, hydrolytic enzymes directly control phytopathogens and save plants from different biotic and abiotic stresses affecting sustainable agriculture [66].

Role of PGPR in phytoremediation

Green technology is an emerging eco-friendly and inexpensive strategy to improve agricultural soil contaminated with heavy metals which results in advanced soil health [20]. Phytoremediation involves the use of specific plant cultivars, heavy metal tolerant, and plant growth-promoting microorganisms [21]. Phytostabilization is a major strategy that makes up phytoremediation that uses a particular plant cultivar to stabilize heavy metal contaminants, consequently reducing their bioavailability in soil [77].

For soil remediation to be considered efficient, it requires plants to intercept, take uptake up, accumulate, sequestrate, stabilize, and translocate heavy metal contaminants [71]. Despite the potential of cleaning up heavy metals in the soil, phytoremediation is faced with some factors that can slow down or stop the process [71]. For instance, such factors are nutrient availability, type of pollutant, plant species, enzyme class, soil component, and pH [77]. The plant microbial community determines the success of the phytoremediation process. Likewise, microbial biomass, soil enzymes, and microorganisms are major mediators of biological processes and even serve as an indicator of heavy metal stress [20,39].

When heavy metal tolerant PGPR is inoculated into polluted soil, it stimulate plants to perform tasks such as heavy metal sequestration, recycling nutrients, controlling pathogens, mineralization, sustaining soil structure, and detoxifying chemicals. As a result, the plants release exudates including amino acids, phytohormones, vitamins, and proteins for the microorganism [15]. The soil enzyme action responds quickly to metal stress, for example, catalase can degrade hydrogen peroxide (H_2O_2) and stop it from contaminating soil organisms [37,96].

Correspondingly, enzyme activities involved in soil organic carbon (C) (saccharase and β-glucosidase), P (acid phosphatase), and N (urease) cycles are considered important indices that present as the potential for soil enzyme synthesis by microbiomes [21,37]. Microbial community diversity reduces with increasing levels of heavy metals such as zinc (Zn) and cadmium (Cd) in soil [97,98]. In a heavy metal polluted soil, *Trigonella foenum-graceum* and *Brassica juncea* with PGPR inoculated into the soil showed rapid degradation of phenanthrene. This was a result of the high microbial population and exudates secreted by plants and root-associated microbial consortium [21].

PGPR such as *Pseudomonas* sp. GHD-4 improved soil enzyme activities and enriched microbial community diversity by degrading lead (Pb) and reducing its concentrations in soil [99]. Heavy metal contaminated rhizosphere soil inoculated with PGPR has been reported to have greater microbial diversity compared to uninoculated soil [39]. PGPR involved in phytoremediation not only benefits plant growth but also improves soil quality [96].

Techniques of applying microbial inoculants to enhance plant growth

Microbial inoculants are made up of live microorganisms, mainly bacteria such as rhizobia or fungi capable of improving plant growth and development. Besides, it could be a pure or mixed culture. Similarly, exists in solid or liquid form [100]. A good carrier for preparing microbial inocula should provide a conducive environment for PGPR to live and grow, and also, have long cellular viability at room temperature for two months and above. A suitable carrier is characterized by being ecologically friendly, easy to apply, stable, and nontoxic toinoculants and plants; easy to sterilize using gamma-irradiation or autoclave, readily available and inadequate form. In addition, have good pH buffering capacity, moisture absorption capacity, easy to process, and should be free from lump-forming materials.

The carrier type determines the application of microbial inoculants. Solid microbial inoculant materials such as peat are bead-like or granules are rich in organic matter and are used for microorganism immobilization [101]. In addition, liquid inoculants are microbial cultures in broth form rich in cell protectors and nutrients. Moreover, liquid inoculants can be suspensions in organic oils or

mineral or oil in water suspensions. In seed inoculation, liquid or powder microbial inoculants are used to coat the seeds. Likewise, other methods of liquid inoculant applications are in-furrow or foliar application or spray on soil [100].

Again, other materials and techniques for carrying microorganisms are through lyophilization, industrial and agricultural residues, and polymers for cell encapsulation [102]. Carriers from agricultural and industrial residues such as wheat bran, clay, sawdust, rice bran, lignite powder, and peat can be augmented with inorganic or chitin containing media such as betonies, vermiculite, silicate, and kaolin for soil or seed inoculation [103]. The use of different microbial inoculants around the world and in Africa over the past decades has increased, resulting in higher crop yields.

Application methods of PGPR in field

The applications of PGPR in field condition have been studied extensively and reported. A variety of PGPR have been used as soil bioinoculants intended to enhance the supply of nutrients to plants. Diverse species of PGPR such as Clostridia, Rhizobium, Bacillus, Azotobacter, Klebsiella, and Azospirillum sp. have successfully been inoculated in the fields to understand the mechanisms and effectiveness of PGPR to increase plant development and productivity. The beneficial effects of inoculations of PGPR (Rhizobium species including Sinorhizobium, Mesorhizobium, Allorhizobium, Bradyrhizobium etc.) has significantly increase plant vegetative and yield by nitrogen-fixing symbiosis with leguminous plants [9]. Additionally, Clostridia was reportedly isolated from rice soils and their interaction also increased after re-inoculating straw to fields which elevated the C to N ratio in the soil. Azospirillum has reportedly increase wheat yields in both field and greenhouse conditions. *Azospirillum lipoferum* increased rice yield to approximately 1.8 t ha−1 in a field experiment [104]. ^{15}N tracer techniques in a field study showed that *Azospirillum lipoferum* and *Azospirillum brasilense* contributed 7–12% of wheat plant nitrogen [105].

Many field studies have confirmed that *Azospirillum diazotrophicus* isolated from rhizosphere soil increased sugarcane plant growth by improving N to over 200 kgN ha^{-1} $year^{-1}$ [103]. Acetobacter-sugarcane system has now become an active experimental model and the diazotrophic character (nif+) is a significant module of this system [106]. Azoarcus sp. BH72 isolated from Kallar grass (Leptochloa fusa Kunth) colonized rice rhizosphere soils which reportedly improved microbial interactions and positively increased rice yield in both field and laboratory conditions [107]. In field trials, many species of family Enterobacteriaceae include diazotrophs mainly those isolated from the rhizosphere of agricultural plants with plant growth-promoting activity colonizes sugarcane, sorghum, rice, maize and other crops [86]. Also, improvement of soybean yield, seed size, fat content and drought stress by microbial inoculation correlated with enhancement in soil microbial assortment in the rhizosphere since it was previously reported by Igiehon et al. [108] that microbial communities could be affected by Rhizobium and mycorrhizal fungi inoculation as well as fertilizer amendment.

Commercialization of PGPR products and challenges in developing countries

The green revolution has sadly introduced chemical pesticides, herbicides, and inorganic fertilizers into agricultural soils that ledto huge damage to the soil environment in the form of pollutants. To resolve this problem, there is a need to design microbial consortia that can help to solve aspects of plant growth potential and bioremediation [11].In many developed countries including the United States of America, Australia, and Europe, and some developing countries like Brazil, Argentina, and India, there is a well-documented success in the commercialization of PGPR products as biofertilizers and biocontrol agents [15,21,52]. Despite the numerous benefits of PGPR inoculants, their development and their commercial application in sustainable crop production and protection in developing nations, particularly in Africa are still very scarce [15].

Developing PGPR into new microbial inoculants depends on initial laboratory screening assays that are aimed at detecting particular PGPR traits such as phosphate solubilization, N fixation, auxin biosynthesis, siderophore production, ACC deaminase activity, antibiotic production to mention a few [38,52]. According to Vacheron et al. [109], auxin production by PGPR can have both beneficial and negative impacts on plant development. It is vital to keep in mind that the concentration of auxin determines its effectiveness. For example, at low concentrations, it promotes plant development, whereas, at high quantities, it inhibits root growth [110]. Screening of PGPR for these plant growth-promoting traits under *in-vitro* assays does not necessarily result in isolates that can improve plant growth under field conditions [14,15,102].

A major challenge in the commercialization of PGPR is developing consortia of highly efficient microorganisms with high rhizosphere colonization and persistence [67]. PGPRs are frequently inoculated on plant materials but without a suitable carrier or quantities, they do not allow effective colonization of the rhizosphere under field conditions, especially due to competition with indigenous soil macro and micro fauna [11]. Additional challenge emanates from soil management practices such as the fumigation of soil with broad-spectrum biocidal fumigants that changes the biological community structure of the soil including introduced microbial inoculants [67,111].

The formulation of bioinoculants exceptionally for particular soil conditions reduces environmental challenges. Also, training farmers and workers on how to expertly apply bioinoculants on crops is critical in the advancement and distribution of beneficial inocula [21]. The need to educate the general population and farmers on the advantages of formulated products containing beneficial microorganisms in agriculture and to explore the full richness of fertile land for sustainable food production is crucial [63].

Conclusion

There is a need for sufficient effort to be made to use and apply existing scientific knowledge for a better understanding of soil-plant-microbial associations and their modes of action in nutrient acquisition and plant growth promotion. With the recent

increase in the awareness and demand to use PGPR for a sustainable agroecological system, the need to substitute agrochemicals with biological products made up of single or a consortium of elite PGPR is very crucial.

The application of PGPR in sustainable food production has the potential to boost crop production, enhance crop nutrition, yield, and control of diseases, and reduce environmental hazards associated with chemical usage. The efficacy of PGPR in boosting plant growth and development is dependent on the specific strain of bacteria and the conditions under which it is used. As a result, selecting a certain strain is critical for obtaining the greatest benefits in terms of better plant growth and development. There are many benefits of microbial inoculant products with PGPR as active ingredients. These include increased mineral nutrients availability and biocontrol through antibiosis, and ISR, competition for essential nutrients such as Fe through the chelation effect of siderophores (the competition must be carefully stated and understood by farmers for improved crop productivity). Genetic engineering of PGPR is an important constituent in modern food production that will mitigate soil contaminants, environmental alteration, and destruction of soil flora and fauna when properly harnessed specifically in developing economies.

Further research on the long-term impacts of PGPR on soil health and ecosystem functioning is recommended to fully comprehend PGPR's application for sustainable food production. More research is needed to properly comprehend the possible benefits and drawbacks of PGPR in terms of sustainable food production. Furthermore, the discovery of new PGPR strains that are more efficient and stable under varied environmental conditions can aid in improving PGPR's effectiveness in encouraging plant growth and development. Likewise, researching the interactions of PGPR with other microorganisms in the rhizosphere and beyond in bulk soil can aid in better understanding the mechanisms by which PGPR stimulates plant growth and development. Also, we recommend the advancement of new methods for the application of PGPR in agriculture, which can help improve the efficiency and effectiveness of PGPR in promoting plant growth and development. Still, promoting the implementation of PGPR in agriculture, given the expected benefits of PGPR in terms of biofertilization, biocontrol, and bioremediation, all of which have a beneficial effect on crop productivity and ecosystem functioning, encouragement should be given to its implementation.

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CRediT authorship contribution statement

Blessing Chidinma Igiehon: Conceptualization, Validation, Data curation, Formal analysis, Writing – original draft, Writing – review & editing. **Olubukola Oluranti Babalola:** Funding acquisition, Supervision, Validation, Writing – review & editing. **Ahmed Idris Hassen:** Supervision, Validation, Writing – review & editing.

Declaration of competing interest

The authors declare no conflicts of interest.

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References

[1] BS Adeleke, OO Babalola, The endosphere microbial communities, a great promise in agriculture, Int. Microbiol. l (2020) 1–17.
[2] ET Alori, OO Babalola, Microbial inoculants for improving crop quality and human health in Africa, Front. Microbiol. 9 (2018) 1–12.
[3] T Obeholster, S Vikram, D Cowan, A Valverde, Key microbial taxa in the rhizosphere of sorghum and sunflower grown in crop rotation, Sci Tot Environ 624 (2018) 530–539.
[4] NO Igiehon, OO Babalola, Rhizosphere microbiome modulators: contributions of nitrogen fixing bacteria towards sustainable agriculture, Int. J. Environ. Res. Public Health 15 (2018) 1–25.
[5] MG Şeker, I Şah, E Kırdök, H Ekinci, YÖ Çiftçi, Ö Akkaya, A hidden plant growth promoting bacterium isolated from in vitro cultures of fraser photinia (Photinia× fraseri), Int. J. Agric. Biol. 19 (6) (2017) 1511–1519.
[6] PE Busby, C Soman, MR Wagner, ML Friesen, J Kremer, A Bennett, M Morsy, JA Eisen, JE Leach, JL Dangl, Research priorities for harnessing plant microbiomes in sustainable agriculture, PLOS Biol. 15 (3) (2017) e2001793.
[7] NO Igiehon, OO Babalola, BR Aremu, Genomic insights into plant growth promoting rhizobia capable of enhancing soybean germination under drought stress, BMC Microbiol. 19 (2019) 1–22.
[8] L Tang, Y Shi, Y Zhang, D Yang, C Guo, Effects of plant-growth-promoting rhizobacteria on soil bacterial community, soil physicochemical properties, and soil enzyme activities in the rhizosphere of alfalfa under field conditions, Diversity 15 (4) (2023) 537.
[9] A Basu, P Prasad, SN Das, S Kalam, R Sayyed, M Reddy, H El Enshasy, Plant growth promoting rhizobacteria (PGPR) as green bioinoculants: recent developments, constraints, and prospects, Sustainability 13 (3) (2021) 1140.
[10] Q Saeed, W Xiukang, FU Haider, J Kučerik, MZ Mumtaz, J Holatko, M Naseem, A Kintl, M Ejaz, M Naveed, Rhizosphere bacteria in plant growth promotion, biocontrol, and bioremediation of contaminated sites: a comprehensive review of effects and mechanisms, Int. J. Mol. Sci. 22 (19) (2021) 10529.

[11] C Sergaki, B Lagunas, I Lidbury, ML Gifford, P Schäfer, Challenges and approaches in microbiome research: from fundamental to applied, Front. Plant Sci. 9 (2018) 1–12.
[12] OO Babalola, Beneficial bacteria of agricultural importance, Biotechnol. Lett. 32 (11) (2010) 1559–1570.
[13] EA Gomes, UG Lana, JF Quensen, SM de Sousa, CA Oliveira, J Guo, LJ Guimarães, JM Tiedje, Root-associated microbiome of maize genotypes with contrasting phosphorus use efficiency, Phytobiomes 2 (3) (2018) 129–137.
[14] J Singh, M Sharma, S Singh, R Bano, A Mahawar, Effect of organic and inorganic sources of npk and bio-fertilizer on enhancement of growth attributes and chlorophyll content of sweet potato, Int. J. Curr. Microbiol. Appl. Sci. 7 (9) (2018) 3659–3667.
[15] C Keswani, H Dilnashin, H Birla, SP Singh, Regulatory barriers to agricultural research commercialization: a case study of biopesticides in India, Rhizosphere 11 (2019) 1–4.
[16] J Kloepper, M Schroth, Relationship of in vitro antibiosis of plant growth-promoting rhizobacteria to plant growth and the displacement of root microflora, Phytopathology 71 (10) (1981) 1020–1024.
[17] LA de Andrade, CHB Santos, ET Frezarin, LR Sales, EC Rigobelo, Plant growth-promoting rhizobacteria for sustainable agricultural production, Microorganisms 11 (4) (2023) 1088.
[18] M Lyu, X Li, J Xie, PM Homyak, L Ukonmaanaho, Z Yang, X Liu, C Ruan, Y Yang, Root–microbial interaction accelerates soil nitrogen depletion but not soil carbon after increasing litter inputs to a coniferous forest, Plant Soil 444 (1-2) (2019) 153–164.
[19] M Lang, S Bei, X Li, TW Kuyper, J Zhang, Rhizoplane bacteria and plant species co-determine phosphorus-mediated microbial legacy effect, Front. Microbiol. 10 (2019) 1–12.
[20] N Tirry, NT Joutey, H Sayel, A Kouchou, W Bahafid, M Asri, N El Ghachtouli, Screening of plant growth promoting traits in heavy metals resistant bacteria: prospects in phytoremediation, J. Genet. Eng. Biotechnol. 16 (2) (2018) 613–619.
[21] R Urana, A Dahiya, P Sharma, N Singh, Effects of plant growth promoting rhizobacteria on phytoremediation of phenanthrene contaminated sodic soil, Polycycl. Aromat. Compd. 12 (2019) 1–10.
[22] BC Nwachukwu, AS Ayangbenro, OO Babalola, Elucidating the rhizosphere associated bacteria for environmental sustainability, Agriculture 11 (1) (2021) 1–18.
[23] BC Nwachukwu, OO Babalola, Perspectives for sustainable agriculture from the microbiome in plant rhizosphere, Plant Biotechnol. Rep. 15 (2021) 259–278.
[24] M Regueira, E Rial, B Blanco, B Bogo, A Aldrey, B Correa, E Varas, C Sánchez, N Vidal, Micropropagation of axillary shoots of *Salix viminalis* using a temporary immersion system, Trees 32 (2018) 61–71.
[25] SI Zandalinas, RM Rivero, V Martínez, A Gómez-Cadenas, V Arbona, Tolerance of citrus plants to the combination of high temperatures and drought is associated to the increase in transpiration modulated by a reduction in abscisic acid levels, BMC Plant Biol. 16 (1) (2016) 1–16.
[26] V Vives-Peris, C de Ollas, A Gómez-Cadenas, RM Pérez-Clemente, Root exudates: from plant to rhizosphere and beyond, Plant Cell Rep. 39 (1) (2020) 3–17.
[27] N Ling, W Zhang, D Wang, J Mao, Q Huang, S Guo, Q Shen, Root exudates from grafted-root watermelon showed a certain contribution in inhibiting *Fusarium oxysporum* f. sp. niveum, PLoS One 8 (5) (2013) e63383.
[28] N Liu, B Zhou, X Zhao, B Lu, Y Li, J Hao, Grafting eggplant onto tomato rootstock to suppress *Verticillium dahliae* infection: the effect of root exudates, Hort. Sci. 44 (7) (2009) 2058–2062.
[29] V Jeyanthi, S Kanimozhi, Plant growth promoting rhizobacteria (PGPR)–prospective and mechanisms: a review, J. Pure Appl. Microbiol. 12 (2) (2018) 733–749.
[30] C Mosimann, T Oberhänsli, D Ziegler, D Nassal, E Kandeler, T Boller, P Mäder, C Thonar, Tracing of two *Pseudomonas* strains in the root and rhizoplane of maize, as related to their plant growth-promoting effect in contrasting soils, Front. Microbiol. 7 (2017) 1–14.
[31] E Tassi, L Giorgetti, E Morelli, J Peralta-Videa, J Gardea-Torresdey, M Barbafieri, Physiological and biochemical responses of sunflower (*Helianthus annuus* L.) exposed to nano-CeO2 and excess boron: modulation of boron phytotoxicity, Plant Physiol. Biochem. 110 (2017) 50–58.
[32] M Naveed, LK Brown, AC Raffan, TS George, AG Bengough, T Roose, I Sinclair, N Koebernick, L Cooper, CA Hackett, PD Hallett, Plant exudates may stabilize or weaken soil depending on species, origin and time, Eur. J Soil Sci. 68 (6) (2017) 806–816.
[33] A Ambrosini, FH Sant'Anna, J Heinzmann, G de Carvalho Fernandes, E Bach, LMP Passaglia, *Paenibacillus helianthi* sp. nov., a nitrogen-fixing species isolated from the rhizosphere of *Helianthus annuus* L, Anton Leeuw. 111 (2018) 2463–2471.
[34] OO Babalola, BC Nwachukwu, AS Ayangbenro, High-throughput sequencing survey of sunflower soil, Microbiol. Resour. Announc. 10 (8) (2021) e01331-20.
[35] KA Odelade, OO Babalola, Bacteria, fungi and archaea domains in rhizospheric soil and their effects in enhancing agricultural productivity, Int. J. Environ. Res. Public Health 16 (20) (2019) 3873.
[36] B Kumari, M Mallick, MK Solanki, AC Solanki, A Hora, W Guo, Plant growth promoting rhizobacteria (PGPR): modern prospects for sustainable agriculture. Plant Health Under Biotic Stress, Springer, Switzerland, 2019, pp. 109–127.
[37] MS Arif, SM Shahzad, M Riaz, T Yasmeen, T Shahzad, MJ Akhtar, L Bragazza, A Buttler, Nitrogen-enriched compost application combined with plant growth-promoting rhizobacteria (PGPR) improves seed quality and nutrient use efficiency of sunflower, J. Plant Nut. Soil Sci. 180 (4) (2017) 464–473.
[38] R Backer, JS Rokem, G Ilangumaran, J Lamont, D Praslickova, E Ricci, S Subramanian, DL Smith, Plant growth-promoting rhizobacteria: context, mechanisms of action, and roadmap to commercialization of biostimulants for sustainable agriculture, Front Plant 9 (2018) 1–17.
[39] W Ju, X Jin, L Liu, G Shen, W Zhao, C Duan, L Fang, Rhizobacteria inoculation benefits nutrient availability for phytostabilization in copper contaminated soil: drivers from bacterial community structures in rhizosphere, Appl. Soil Ecol.. 150 (2020) 1–13.
[40] R Zhang, JM Vivanco, Q Shen, The unseen rhizosphere root–soil–microbe interactions for crop production, Curr. Opin. Microbiol. 37 (2017) 8–14.
[41] AA Adedeji, MM Häggblom, OO Babalola, Sustainable agriculture in Africa: plant growth-promoting rhizobacteria (PGPR) to the rescue, Sci. Afr. 9 (2020) 1–14, https://doi.org/10.1016/j.sciaf.2020.e00492.
[42] Y Wan, Q Huang, Q Wang, Y Yu, D Su, Y Qiao, H Li, Accumulation and bioavailability of heavy metals in an acid soil and their uptake by paddy rice under continuous application of chicken and swine manure, J. Hazard. Mater. 384 (2020) 121293.
[43] BS Adeleke, OO Babalola, Oilseed crop sunflower (*Helianthus annuus*) as a source of food: Nutritional and health benefits, Food Sci. Nut 8 (2020) 1–19.
[44] O Babalola, A Akindolire, Identification of native rhizobacteria peculiar to selected food crops in Mmabatho municipality of South Africa, Biol. Agric. Hortic. 27 (3-4) (2011) 1–17.
[45] AJ Valentine, VA Benedito, Y Kang, Legume nitrogen fixation and soil abiotic stress: from physiology to genomics and beyond, Annu. Plant Rev. online 42 (2018) 207–248.
[46] W Hassan, S Bashir, F Ali, M Ijaz, M Hussain, J David, Role of ACC-deaminase and/or nitrogen fixing rhizobacteria in growth promotion of wheat (*Triticum aestivum* L.) under cadmium pollution, Environ. Earth Sci. 75 (3) (2016) 267.
[47] L Hu, CA Robert, S Cadot, X Zhang, M Ye, B Li, D Manzo, N Chervet, T Steinger, MG Van Der Heijden, Root exudate metabolites drive plant-soil feedbacks on growth and defense by shaping the rhizosphere microbiota, Nat. Commun. 9 (1) (2018) 1–13.
[48] J Mishra, R Singh, NK Arora, Plant growth-promoting microbes: diverse roles in agriculture and environmental sustainability. Probiotics and Plant Health, Springer, Switzerland, 2017, pp. 71–111.
[49] H Etesami, DK Maheshwari, Use of plant growth promoting rhizobacteria (PGPRs) with multiple plant growth promoting traits in stress agriculture: action mechanisms and future prospects, Ecotoxicol. Environ. Saf. 156 (2018) 225–246.
[50] OO Babalola, AI Sanni, GD Odhiambo, B Torto, Plant growth-promoting rhizobacteria do not pose any deleterious effect on cowpea and detectable amounts of ethylene are produced, World J. Microbiol. Biotechnol. 23 (6) (2007) 747–752.
[51] A Majeed, MK Abbasi, S Hameed, S Yasmin, MK Hanif, T Naqqash, A Imran, *Pseudomonas* sp. AF-54 containing multiple plant beneficial traits acts as growth enhancer of *Helianthus annuus* L. under reduced fertilizer input, Microbiol. Res. 216 (2018) 56–69.
[52] M Prasad, R Srinivasan, M Chaudhary, M Choudhary, LK Jat, Plant Growth Promoting Rhizobacteria (PGPR) for sustainable agriculture: perspectives and challenges. Plant Growth Promoting Rhizobacteria Amelioration in Sustainable Agriculture, Elsevier, United Kingdom, 2019, pp. 129–157.

[53] A Majeed, Z Muhammad, H Ahmad, Plant growth promoting bacteria: role in soil improvement, abiotic and biotic stress management of crops, Plant Cell Rep. 37 (12) (2018) 1599–1609.
[54] XY Zhang, XM Jia, R Zhang, ZL Zhu, B Liu, LY Gao, YX Wang, Metabolic analysis in *Malus halliana* leaves in response to iron deficiency, Sci. Hortic. 258 (2019) 108792.
[55] T Orikowska, K Nowak, B Reed, Bacteria in the plant tissue culture environment, Plant Cell Tissue Organ. Cult 128 (3) (2017) 487–508.
[56] XY Xiao, MW Wang, HW Zhu, ZH Guo, XQ Han, P Zeng, Response of soil microbial activities and microbial community structure to *vanadium* stress, Ecotoxicol. Environ. Saf. 142 (2017) 200–206.
[57] I Fabiańska, N Gerlach, J Almario, M Bucher, Plant-mediated effects of soil phosphorus on the root-associated fungal microbiota in *Arabidopsis thaliana*, New Phytol. 221 (4) (2019) 2123–2137.
[58] OS Olarewaju, AS Ayangbenro, BR Glick, OO Babalola, Plant health: feedback effect of root exudates-rhizobiome interactions, Appl. Microbiol. Biotechnol. 103 (3) (2019) 1155–1166.
[59] CJ Castelle, JF Banfield, Major new microbial groups expand diversity and alter our understanding of the tree of life, Cell 172 (6) (2018) 1181–1197.
[60] GF Andrade, FP Paniz, AC Martins Jr, BA Rocha, AK da Silva Lobato, JL Rodrigues, P Cardoso-Gustavson, HP Masuda, BL Batista, Agricultural use of Samarco's spilled mud assessed by rice cultivation: a promising residue use? Chemosphere 193 (2018) 892–902.
[61] M Saleem, HN Asghar, ZA Zahir, M Shahid, Impact of lead tolerant plant growth promoting rhizobacteria on growth, physiology, antioxidant activities, yield and lead content in sunflower in lead contaminated soil, Chemosphere 195 (2018) 606–614.
[62] MC Enebe, OO Babalola, The influence of plant growth-promoting rhizobacteria in plant tolerance to abiotic stress: a survival strategy, Appl. Microbiol. Biotechnol. 102 (18) (2018) 7821–7835.
[63] J Itelima, W Bang, I Onyimba, E Oj, A review: biofertilizer; a key player in enhancing soil fertility and crop productivity, J Microbiol Biotechnol Rep 2 (2018) 22–28.
[64] SH Youseif, FH Abd El-Megeed, SA Saleh, Improvement of faba bean yield using rhizobium/agrobacterium inoculant in low-fertility sandy soil, Agronomy 7 (2017) 1–12.
[65] M Barquero, R Pastor-Buies, B Urbano, F González-Andrés, Challenges, regulations and future actions in biofertilizers in the european agriculture: from the lab to the field. Microbial Probiotics for Agricultural Systems, Springer, Switzerland, 2019, pp. 83–107.
[66] US Oak, Amrita Kumar Vinay, Microbial intervention in agriculture and environment, vol 3. Microbial Interventions in Soil and Plant Health for Improving Crop Efficiency, Springer, New York City, United States of America, 2019.
[67] T Chaudhary, M Dixit, R Gera, AK Shukla, A Prakash, G Gupta, P Shukla, Techniques for improving formulations of bioinoculants, 3 Biotechol 10 (2020) 1–9.
[68] R Garrido-Oter, RT Nakano, N Dombrowski, K-W Ma, TA Team, AC McHardy, P Schulze-Lefert, Modular traits of the rhizobiales root microbiota and their evolutionary relationship with symbiotic rhizobia, Cell Host Microbe 24 (1) (2018), 155-167. e155.
[69] S Mukhtar, S Mehnaz, MS Mirza, KA Malik, Isolation and characterization of bacteria associated with the rhizosphere of halophytes (*Salsola stocksii* and *Atriplex amnicola*) for production of hydrolytic enzymes, Braz. J. Microbiol. 50 (1) (2019) 85–97.
[70] RK Jadhav, L Anil, Attempt of Costus arbuscular mycorrhizal inoculants association with leguminous foliage whey on chlorophyll of sunflower (*Helianthus annus* L.) plants, J. Plant Stress Physiol. 6 (2020) 18–23.
[71] SB Singh, H Gowtham, M Murali, P Hariprasad, T Lakshmeesha, KN Murthy, K Amruthesh, S Niranjana, Plant growth promoting ability of ACC deaminase producing rhizobacteria native to Sunflower (*Helianthus annuus* L.), Biocatal. Agric. Biotechnol. 18 (2019) 1–9.
[72] ET Alori, BR Glick, OO Babalola, Microbial phosphorus solubilization and its potential for use in sustainable agriculture, Front. Microbiol. 8 (2017) 1–8, https://doi.org/10.3389/fmicb.2017.00971.
[73] L Zhao, Y Wang, S Kong, Effects of *Trichoderma asperellum* and its siderophores on endogenous auxin in Arabidopsis thaliana under iron-deficiency stress, Int. Microbiol. 23 (2020) 501–509.
[74] Z Lei, YQ Zhang, Effects of phosphate solubilization and phytohormone production of Trichoderma asperellum Q1 on promoting cucumber growth under salt stress, J. Integ. Agric. 14 (8) (2015) 1588–1597.
[75] K Lindström, SA Mousavi, Effectiveness of nitrogen fixation in rhizobia, Microb. Biotechnol. 0 (0) (2019) 1–22.
[76] C Colombo, G Palumbo, JZ He, R Pinton, S Cesco, Review on iron availability in soil: interaction of Fe minerals, plants, and microbes, J. Soils Sedim. 14 (2014) 538–548.
[77] AS Ayangbenro, OO Babalola, A new strategy for heavy metal polluted environments: a review of microbial biosorbents, Int. J. Environ. Res. Public Health 14 (1) (2017) 1–16.
[78] SB Singh, H Gowtham, M Murali, P Hariprasad, T Lakshmeesha, KN Murthy, K Amruthesh, S Niranjana, Plant growth promoting ability of ACC deaminase producing rhizobacteria native to Sunflower (*Helianthus annuus* L.), Biocatal. Agric. Biotechnol. 18 (2019) 101089.
[79] FJ Romera, MJ García, C Lucena, A Martínez-Medina, MA Aparicio, J Ramos, E Alcántara, M Angulo, R Pérez-Vicente, Induced systemic resistance (ISR) and Fe deficiency responses in dicot plants, Front. Plant. 10 (2019) 287.
[80] S Deng, HML Wipf, G Pierroz, TK Raab, R Khanna, D Coleman-Derr, A plant growth-promoting microbial soil amendment dynamically alters the strawberry root bacterial microbiome, Sci. Rep. 9 (1) (2019) 1–15.
[81] SA Lee, Y Kim, JM Kim, B Chu, JH Joa, MK Sang, J Song, HY Weon, A preliminary examination of bacterial, archaeal, and fungal communities inhabiting different rhizocompartments of tomato plants under real-world environments, Sci. Rep. 9 (1) (2019) 1–15.
[82] H Li, JQ Su, XR Yang, YG Zhu, Distinct rhizosphere effect on active and total bacterial communities in paddy soils, Sci. Tot. Environ. 649 (2019) 422–430.
[83] A Ambrosini, FH Sant'Anna, R de Souza, M Tadra-Sfeir, H Faoro, SM Alvarenga, FO Pedrosa, EM Souza, LM Passaglia, Genome sequence of Bacillus mycoides B38V, a growth-promoting bacterium of sunflower, Genome Announc. 3 (2) (2015) e00245-00215.
[84] SK Singh, PP Singh, A Gupta, AK Singh, J Keshri, Tolerance of heavy metal toxicity using PGPR strains of *Pseudomonas* species. PGPR Amelioration in Sustainable Agriculture, Elsevier, United Kingdom, 2019, pp. 239–252, vol 140.
[85] Saharan B, Nehra V (2011) Plant growth promoting rhizobacteria: a critical review. v. 2011.
[86] P Vejan, R Abdullah, T Khadiran, S Ismail, A Nasrulhaq Boyce, Role of plant growth promoting rhizobacteria in agricultural sustainability—a review, Molecules 21 (5) (2016) 573.
[87] O Martínez-Viveros, MA Jorquera, D Crowley, G Gajardo, M Mora, Mechanisms and practical considerations involved in plant growth promotion by rhizobacteria, J. Soil Sci. Plant Nutr. 10 (3) (2010) 293–319.
[88] AW Bakker, B Schippers, Microbial cyanide production in the rhizosphere in relation to potato yield reduction and Pseudomonas spp-mediated plant growth-stimulation, Soil Biol. Biochem. 19 (4) (1987) 451–457.
[89] S Daryanto, L Wang, PA Jacinthe, Global synthesis of drought effects on food legume production, PLoS One 10 (6) (2015) e0127401.
[90] M Naveed, B Mitter, TG Reichenauer, K Wieczorek, A Sessitsch, Increased drought stress resilience of maize through endophytic colonization by Burkholderia phytofirmans PsJN and Enterobacter sp. FD17, Environ. Exp. Bot. 97 (2014) 30–39.
[91] C Dimkpa, T Weinand, F Asch, Plant–rhizobacteria interactions alleviate abiotic stress conditions, Plant Cell Environ. 32 (12) (2009) 1682–1694.
[92] N Rascovan, B Carbonetto, D Perrig, M Díaz, W Canciani, M Abalo, J Alloati, G González-Anta, MP Vazquez, Integrated analysis of root microbiomes of soybean and wheat from agricultural fields, Sci. Rep. 6 (2016) 28084.
[93] R Vijayan, P Palaniappan, S Tongmin, P Elavarasi, N Manoharan, Rhizobitoxine enhances nodulation by inhibiting ethylene synthesis of *Bradyrhizobium elkanii* from Lespedeza species: validation by homology modelingand molecular docking study, World J. Pharm. Sci. 2 (2013) 4079–4094.
[94] K Xiong, J Fuhrmann, Comparison of rhizobitoxine-induced inhibition of β-cystathionase from different bradyrhizobia and soybean genotypes, Plant Soil 186 (1996) 53–61.
[95] J Yang, JW Kloepper, CM Ryu, Rhizosphere bacteria help plants tolerate abiotic stress, Trends Plant Sci. 14 (1) (2009) 1–4.
[96] W Peng, X Li, J Song, W Jiang, Y Liu, W Fan, Bioremediation of cadmium-and zinc-contaminated soil using Rhodobacter sphaeroides, Chemosphere 197 (2018) 33–41.

[97] N Khan, P Zandi, S Ali, A Mehmood, M Adnan Shahid, J Yang, Impact of salicylic acid and plant growth promoting rhizobacteria on the drought tolerance and phytoremediation potential of *Helianthus annus*, Front. Microbiol. 9 (2018) 2507.
[98] S Tiwari, C Lata, Heavy metal stress, signaling, and tolerance due to plant-associated microbes: an overview, Front. Plant Sci. 9 (2018) 1–12.
[99] Y Xu, Y Ge, J Song, C Rensing, Assembly of root-associated microbial community of typical rice cultivars in different soil types, Biol. Fert. Soils 56 (2019) 1–12.
[100] MS Santos, MA Nogueira, M Hungria, Microbial inoculants: reviewing the past, discussing the present and previewing an outstanding future for the use of beneficial bacteria in agriculture, AMB Express 9 (1) (2019) 1–12.
[101] LG Moretti, E Lazarini, JW Bossolani, TL Parente, S Caioni, RS Araujo, M Hungria, Can additional inoculations increase soybean nodulation and grain yield? J. Agron. 110 (2) (2018) 1–7.
[102] LG Moretti, CA Crusciol, EE Kuramae, JW Bossolani, A Moreira, NR Costa, CJ Alves, IM Pascoaloto, AB Rondina, M Hungria, Effects of growth-promoting bacteria on soybean root activity, plant development, and yield, Agron. J. 112 (1) (2020) 1–11.
[103] J Fukami, MA Nogueira, RS Araujo, M Hungria, Accessing inoculation methods of maize and wheat with *Azospirillum brasilense*, Ann. Microbiol. Exp. 6 (1) (2016) 1–13.
[104] F Cassan, Diaz-Zorita, *Azospirillum* sp. in current agriculture: from the laboratory to the field, Soil Biol. Biochem. 103 (2016) 117–130.
[105] N Ilyas, K Mumtaz, N Akhtar, H Yasmin, RZ Sayyed, W Khan, HA El Enshasy, DJ Dailin, EA Elsayed, Z Ali, Exopolysaccharides producing bacteria for the amelioration of drought stress in wheat, Sustainability 12 (2020) 8876.
[106] CA Anaya, E Huber-Sannwald, Long-term soil organic carbon and nitrogen dynamics after conversion of tropical forest to traditional sugarcane agriculture in East Mexico, Soil Till Res. 147 (2015) 20–29.
[107] H Chauhan, DJ Bagyaraj, G Selvakumar, SP Sundaram, Novel plant growth promoting rhizobacteria—prospects and potential, Appl. Soil Ecol. 95 (2015) 38–53.
[108] NO Igiehon, OO Babalola, C Xavier, T Baldwyn, Effects of rhizobia and arbuscular mycorrhizal fungi on yield, size distribution and fatty acid of soybean seeds grown under drought stress, Microbiol. Res. 242 (2021) 1–16.
[109] J Vacheron, G Desbrosses, ML Bouffaud, B Touraine, Y Moënne-Loccoz, D Muller, L Legendre, F Wisniewski-Dyé, C Prigent-Combaret, Plant growth-promoting rhizobacteria and root system functioning, Front. Plant Sci. 4 (2013) 356.
[110] H Xie, J Pasternak, BR Glick, Isolation and characterization of mutants of the plant growth-promoting rhizobacterium *Pseudomonas putida* GR12-2 that overproduce indoleacetic acid, Curr. Microbiol. 32 (1996) 67–71.
[111] S Gopalakrishnan, A Sathya, R Vijayabharathi, RK Varshney, CL Gowda, L Krishnamurthy, Plant growth promoting rhizobia: challenges and opportunities, 3 Biotech 5 (4) (2015) 355–377.