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A review

Advances in research on the use of *Brevundimonas* spp. to improve crop and soil fertility and for soil bioremediation

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ABSTRACT

Biofertilizers or biological fertilizers maintain soil fertility by fixing atmospheric nitrogen, solubilizing P and K, producing plant growth substances and antibiotics as well as biodegradation of organic matter in the soil that enriches the root rhizosphere. Microbial biofertilizers are eco-friendly and less expensive alternatives to chemical fertilizers. The key components of healthy soil are populations of plant growth promoting rhizobacteria (PGPR) which play multiple beneficial and ecological roles in the rhizosphere soil. PGPR colonizes rhizosphere or plant roots, resulting in phytostimulation, biofertilization and biocontrol either directly and/or indirectly. Another important role of PGPR is its ability to decontaminate soils through a process called soil bioremediation. Recently, the known rhizobacteria environmentally friendly biofertilizers for sustainable agriculture are those belonging to *Brevundimonas* spp., which play a significant role in improving crop production and soil health.

1. Introduction

Unreasonable and indiscriminate use of agrochemicals has damaged soil health during the time. This has harmful effects as well as an impact on the environment, including the soil microbial community. Biofertilizers have been seen as promising alternatives in contrast to chemical fertilizers, primarily due to their positive effects on reducing the risk of environmental damage and human health [1]. Eco-friendly approaches inspire a wide range of applications of biofertilizers who effective solutions can be contributing toward higher agronomic yield.

Biofertilizers are organic and biodegradable. They contain microbial living cells of different microorganisms which can enhance and increase the nutrient uptake by the plants, such as N, P, K, antibiotics, hormones like auxins, cytokinins, and vitamins which enrich root rhizosphere and the soil by improving its quality by mobilizing plant nutrients from unusable to usable form through a biological process.

Biofertilizers amendment was very significant when applied either on the topsoil or through seed inoculations because those substances contain living microorganisms that can colonize the rhizosphere of the plant and increase the supply or availability of primary nutrients viz., nitrogen and phosphorus besides improving biological fixation of atmospheric nitrogen and enhance phosphorus availability and/or growth stimulus to the target crop [2-3].

Biofertilizers can fix atmospheric nitrogen in the range of 20-200 kg/ha/year, solubilize P in the range of 30-50 kg P_2O_5/ha/year; mobilize P, Zn, Fe, and Mo to varying extents. They also help host plants to resist diseases and withstand stress conditions by different mechanisms, which vary depending upon the type of biofertilizer agent involved [4-5-6].

Numerous species of soil bacteria that colonize mainly in the rhizosphere of plants are known as plant growth promoting rhizobacteria (PGPR) such as *Azotobacter*, *Azospirillum*, *Bacillus*, and *Pseudomonas* [7]. The present review highlights the role of rhizobacteria *Brevundimonas* spp. as PGPR on plant growth and

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productivity, nutrient profile, plant protection and crop improvement.

This review aims to present an overview of the current knowledge and information about nitrogen fixation, phosphate solubilization in the rhizosphere and phytoremediation of crops. Information gathered through this paper review may yield new insights into *Brevundimonas*/plant interaction development.

2. The genus Brevundimonas

Brevundimonas is a genus of Alphaproteobacteria, belonging to the family Caulobacteraceae. The term "Brevundimonas" was first used by Segers et al. [8] in 1994, when two species of Pseudomonas initially named *P. diminuta* and *P. vesicularis* were renamed *Brevundimonas diminuta* and *Brevundimonas vesicularis*. In 1999, several species of *Caulobacter* were transferred to *Brevundimonas* and the genus description was emended markedly [9] (Figure 1).

Molecular methods, based on 16 S rRNA gene sequences, have enabled the resolution of the taxonomic entities and especially the discovery of many new species of *Brevundimonas*. Dahal and Kim [10], Kang et al. [11], Tsubouchi et al. [12] are among the taxonomists who have recently focused on this genus.

At the time of writing, it comprises 38 species (listed in figure 1) with validly published names within the *Brevundimonas* genus (http://www.bacterio.net/brevundimonas.html).

The genus Brevundimonas is one of the most diverse and ubiquitous bacterial genera whose species have been isolated worldwide in all kinds of environments, such as soils, water and plant roots [13 – 16]. Brevundimonas has the potential to be used for a wide range of activities, including soil bioremediation [17], water pollutant treatment [18], and plant growth promotion for sustainable agriculture [15, 19].

3. *Brevundimonas* as a biofertilizer and a crop enhancer

3.1. Potential benefits of biological nitrogen fixation

Nitrogen is the major factor yield-limiting element in many agricultural production systems under most conditions. This importance is translated by the raised cost of nitrogen fertilizer, which is the most important purchased material input in crop production. Since we became aware of the energy cost from nitrogen fertilizers, we give more importance to the biological fixation of nitrogen [20].

Crop plants can use about 50% of the applied fertilizer N, while 25% is lost from the soil-plant system through leaching, volatilization, denitrification, and due to many other factors causing not only an annual economic loss of US$ 3 billion but also pollution to the environment [20, 21].

Nitrogen (N), an essential constituent for synthesizing amino acids, nucleic acids, vitamins, proteins, and chlorophyll, is an important mineral necessary for the living and growth of plants. Despite 80% of the atmospheric air consisting of nitrogen, plants cannot directly assimilate this gaseous form. Plants assimilate the available nitrogen in the soil through their roots in the form of ammonium ($NH4^+$) and nitrates ($NO3^-$) [20].

Since the main limiting nutrient for crops is nitrogen, it becomes important to find alternatives to reduce and optimize the use of chemical N fertilizers applied to crops. Many studies have reported the transfer of nitrogen between the roots of different crops and nitrogen-fixing bacteria [19, 22 – 26].

The process through which atmospheric nitrogen (N2) is converted to ammonia (NH3) is referred to as biological nitrogen fixation (BNF), and the nitrogen-fixing bacteria are known as “Diazotrophs”. Since introducing of the concept of endophytic diazotrophs in 1988 by Döbereiner, many new endophytic diazotrophs have been isolated. The three major types of bacteria that fix the nitrogen are rhizospheric, endophytic, and nodulating bacteria [27].

In 1904, Hiltner was the first to define the rhizosphere. This zone harbors many microorganisms having a higher activity around the roots of crops. Among these, the native bacteria called plant growth-promoting rhizobacteria (PGPR) live surroundings the root or on its surface and play a crucial role in soil health and plant growth [28].

The mechanism by which PGPRs promote the growth of plants can be either direct or indirect. There are several ways the PGPRs may directly facilitate the proliferation of their plant hosts by solubilizing minerals like phosphates in a form that can be used by the plant, synthesizing phytohormones like auxins that trigger plant cell growth and proliferation and fixing atmospheric nitrogen and supply it to the plant.

The biofertilizers based upon PGPR are economically cheap and environmentally safe. Rhizosphere competence and root colonization are important features of any candidate strain for biofertilizer production. Rhizobacteria contain various plant-beneficial traits and their inoculation can sustainably increase crop yield and productivity.

The nitrogen-fixing potential is an important attribute for PGPRs; this ability was reported in many studies. More specifically, PGPRs with nitrogen-fixing ability is reported to be a valuable source of nitrogen for sustainable crop production as well as to maintain soil fertility. Several groups of soil and root associated nitrogen-fixing microorganisms such as *Brevundimonas* sp. [15]; *Brevundimonas diminuta* [29]; *Enterobacter asburie* and *Bacillus pumilus* [26]; *Pantoea dispersa*, *Chryseobacterium indologenes*, *Pseudomonas geniculata*, *Stenotrophomonas pavanii*, *Stenotrophomonas maltophilia*, and *Stenotrophomonas acidaminiphila* [30]; *Pseudomonas koreensis* and *Pseudomonas entomophila* [24], *Rhizobium tropici*, *Bradyrhizobium diazoefficiens* and *Bradyrhizobium japonicum* [23]; *Gluconacetobacter diazotrophicus*, *Herbaspirillum seropedicae*, *Herbaspirillum rubrisubalbicans*, *Paraburkholderia tropica*, *Nitrospirillum amazonense* [22]; *Bacillus megaterium* and *Bacillus mycoides* [25], have been found to colonize different crops and stimulate plant growth either directly or indirectly. Dos Santos et al. [31] demonstrated that Diazotrophs can modify nitrogen uptake by improving the root architecture of plants and can also affect the enzymes responsible for N utilization.

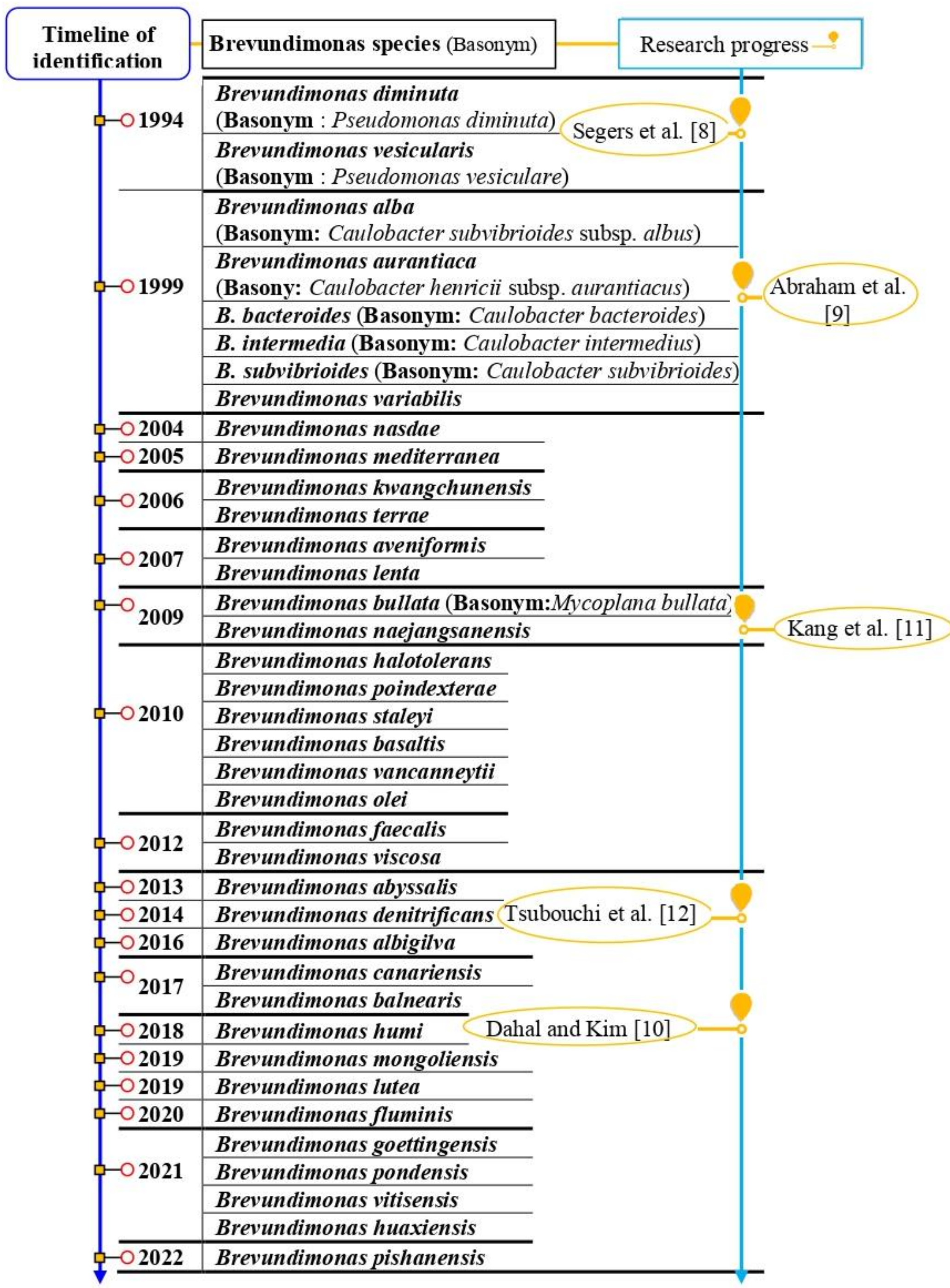

Fig 1. Timeline of identification history and research progress of *Brevundimonas* species.

Colonization of the rhizosphere and survival under varying environmental conditions by effective plant growth promoting rhizobacteria (PGPR) inoculant necessitates the presence of a diverse set of traits. IAA quantification is considered an important trait in the characterization of plant-associated bacteria. Roots exudates contain tryptophan which endophytic bacteria can consume by as a precursor for IAA production increases root size and distribution, resulting in better nutrient uptake from the soil [32]. The diazotrophs *Brevundimonas* spp. can produce phytohormones such as indole-3-acetic acid (IAA). The phytohormones increase root hair production, thereby improving nutrient uptake from the soil during stress tolerance [15, 29, 33].

Numerous studies indicate that Brevundimonas bacteria have been isolated from a wide range of environments and frequently exhibit the ability to promote plant growth [15, 17, 19, 29, 33, 34]. *Brevundimonas* spp. as plant-growth-promoting bacteria have already been reported in the literature, as in the case of *Brevundimonas diminuta* [35].

Although, *Brevundimonas* spp. have been used as PGPR in enhancing the growth of wheat plants. Under pot culture conditions, isolate *B. diminuta* (AW7) promoted root and shoot growth of wheat seedlings, increasing the dry biomass of shoots and roots relative to untreated controls. A significant enhancement was recorded in root weight, panicle weight and plant height with 64.2%, 38.9% and 29.14%, respectively [29].

According to Kumar and Gera [15], the bacterial strain isolated from the sugarcane (*Saccharum* L.) rhizosphere, *Brevundimonas* sp. MDB4 has the nif*H* gene as well as exhibited nitrogenase activity with 47.90 nmol ethylene mg^{-1}protein h^{-1} indicating its ability to fix nitrogen. In the same case, Rana et al. [29] reported that *Brevundimonas diminuta* (AW7) exhibited a nitrogenase activity with 18.29 nmol ethylene mg^{-1}protein h^{-1}. The evaluation of other plant growth promoting activities of the MDB4 revealed that it produced ammonia (0.954 ± 0.006 $lgmL^{-1}$) and indole-3- acetic acid (364.1 ± 0.750 $lgmL^{-1}$) [15].

In pot experiments, it was envisaged that inoculation of cotton with diazotrophic PGPR *Brevundimonas* sp. MDB4 enhances plant growth by supplying nitrogen. It showed stimulatory effects on plant height and dry weights of root and shoot with 68.41, 58.44 and 64.81 %, respectively [15].

3.2. Phosphate Solubilizers

Phosphorus (P) is the structural component of nucleic acids, phospholipids, and adenosine triphosphate. It is an essential and key nutrient for the metabolic and biochemical pathways for various reactions [36]. Soil is a storehouse of several phosphate forms, including inorganic and organic phosphate. Insoluble forms of P are found relatively in higher amounts. The availability of this element depends on its solubility in both monobasic ($H_2PO_4^{-}$) and dibasic (HPO_4^{-2}) forms that may be enhanced by the root activity of plant and microorganisms available in the soil [37].

Using rock phosphate as a phosphate fertilizer, with its solubilization by microbes through the production of organic acids, has become a valid alternative to chemical fertilizers [38]. Several studies have shown that phosphate-solubilizing microorganisms solubilize the fixed P in the soil resulting in higher crop yields [39].

Brevundimonas spp. is also considered as a fertilizer when it makes insoluble phosphorus available to plants. Numerous reports found that different strains of *B. diminuta* were able to solubilize insoluble phosphates [29, 40, 41].

Significant increases in the growth and yield of important crops in response to inoculation with species of *Brevundimonas* spp. have been reported [29, 42, 43]. In most cases, the application of Brevundimonas in pot trials showed a statistically significant increase in several crop production parameters such as root length, leaf length, or plant weight. Specifically, *Brevundimonas* spp. TN37 can significantly improve the biomass of shoot and root fresh, total nitrogen contents of shoot, and root length seed when inoculating potato plants in a pot due to the ability of this bacterium to fix nitrogen and P-solubilizing. *Brevundimonas* sp. TN37 was found to be a root-colonizer of potato forming strong root-associations. The bacterial cell density was high in the rhizosphere, where root hairs were preferred sites for early colonization [19].

Brevundimonas sp. (PS-4) endophyte bacterium in the roots of *Saccharum munja* L. can produce hydrolytic enzymes, IAA, and solubilize phosphate [33]. It was also found that this bacterium is capable of attaching to rhizospheric sludge, which increases the development of the plant under stressful environments.

However, *B. diminuta* and *B. vesicularis* endophyte bacterium isolated from peach and pear rootstock cultures significantly influence the host plant growth. It showed positive results for their potential to promote plant growth according to indole acetic acid (IAA) production, nitrogen fixation and phosphate solubilization [41].

3.3. Phytoremediation and Soil Health

Several factors can affect the health of the soil, among them the presence of toxic substances and residues of pesticides and chemical fertilizers. Soil health is the capacity of soil to function as a vital living system within an ecosystem. It supports plant and animal productivity and maintains or improves water and air quality, while fostering plant health [44].

In recent years, the heavy metal pollution of farmlands has become very severe and caused a real problem [45].

Indeed, Pulleman et al. [46] reported that microbial communities provide many potential indicators for the sustainable management of soils, which is why further efforts are therefore required to investigate biodegradable bacteria to expand their application in the bioremediation of chemical pollutants. Biofertilizers not only exhibit plant growth promotion but are also effective in bioremediation by detoxifying detrimental pollutants such as pesticides and heavy metal pollutants.

Phytoremediation is a promising technology that uses plants and their associated bacteria to clean up contaminants from the environment. Enhancing these relations can be a core part of this technology [47]. In recent years, phytoremediation by using PGPRs has been highly solicited to clean up toxic metals from soil [48]. Henceforth, the suitable employment of these species can boost metal phyto-availability while reducing toxic effects, stimulating the host plant's ability to generate more biomass while storing significant concentrations of metals and decreasing metal toxic effects [33].

Previous studies clearly showed that the role of the microbial community during phytoremediation of plant growth promotes activities like the production of siderophore, nutrient solubilization, etc. [49, 50].

It is well established that microbes-assisted heavy metals uptake is primarily carried out by some of the important genera of bacteria used in bioremediation, include *Kluyvera*, *Bacillus*, *Rhodococcus*, *Flavobacterium*, *Methylobacterium*, *Ochrobactrum*, *Azotobacter*, *Burkholderia*, *Brevibacillus*, *Brevundimonas*, *Variovox*, *Xanthomonas*, *Pseudomonas*, *Methanobacteria*, *Ralstonia*, and *Deinococcus*, etc. [51 – 53]. Brevundimonas species are ubiquitous in the environment. Interestingly, many studies show that several strains of the genus Brevundimonas could eliminate some hazardous organic compounds and metals. For instance, *B. diminuta* has the potential to degrade oils [18], *B. vesicularis* could remove nickel and copper [54], as well as *Brevundimonas* sp. HgP1 and HgP2 were able to eliminate mercury [55].

In addition, a novel bacterial strain identified as *Brevundimonas naejangsanensis* J3 isolated from agricultural soils has the potential to effectively detoxify dimethachlon from contaminated soils. Free cells and enzymes of this strain could rapidly degrade dimethachlon to simple products that would be completely mineralized [56].

According to Rathi and Yogalakshmi [17], inoculation of sunflowers with the rhizobacteria *Brevundimonas diminuta* MYS6 enhanced the vegetative growth and biomass of these plants, subsequently increasing copper uptake in their vegetative tissues. The high efficiency of copper bioremediation by *B. diminuta* MYS6 indicates the potential application of the strain for ecological and cost-effective purposes.

Singh et al. [57] found that inoculation of *Brevundimonas dimunta* (IITISM22) increased the rate of Hg^{II} accumulation in Indian mustard plants (*Brassica juncea* L.). After bacterial inoculation, plant biomass and growth metrics, as well as nutrient absorption capacity, were augmented.

Brevundimonas diminuta (NBRI012) has also been reported to be arsenic resistant with PGPR characteristics, which help to promote rice (*Oryza sativa* L.) plant growth under heavy metals stress environment [42].

4. Conclusion

Using of effective nitrogen-fixing and phosphate-solubilizing microorganisms is an opportunity for improving crop production in addition to maintaining soil structure and fertility. The use of PGPRs as inoculant biofertilizers is an efficient approach to replacing chemical fertilizers and pesticides for sustainable crop cultivation. Scientists have long sought out the potential of metal-tolerant rhizobacteria as a cost-effective and environmentally friendly phytoremediation agent for heavy metals. This review, the role of *Brevundimonas* in nitrogen fixation, phosphate solubilization and phytoremediation of several crops has been presented. In its last years, *Brevundimonas* as root bacteria heads the list of PGPRs assessed worldwide in recent experiments in numerous studies. Recent studies on the improvement and biostimulation of plant growth and crop yield aim to replace chemical fertilizers with biofertilizers based on *Brevundimonas* in order to reduce pollution caused by agrochemicals. There is overwhelming evidence in the literature indicating that *Brevundimonas* could be a true success story in sustainable agriculture. We could conclude that *Brevundimonas* species act as a green technology in addressing issues such as agricultural soil pollution and fertilization in various crops which opens prospects for sustainable agriculture.

Conflict of Interest

The authors declare that they have no conflict of interest

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