



# **Pointing Out Opportunities to Increase Grassland Pastures Productivity via Microbial Inoculants: Attending the Society's Demands for Meat Production with Sustainability**

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Abstract: Estimates are that land area occupied by grass pasture far exceeds that of other crops; at least half are at some stage of degradation. The use of elite plant-growth-promoting microorganisms (PGPM) as inoculants represents an important strategy to achieve qualitative and quantitative improvements in forage biomass, increasing the productivity and sustainability of livestock production. Several studies have reported the benefits of PGPM in grass pastures, with an emphasis on bacteria of the genera Azospirillum, Pseudomonas, Bacillus, rhizobia, and on arbuscular mycorrhiza fungi (AMF). The main grasses studied are Urocholoa (syn. Brachiaria), Megathyrsus (syn. Panicum), Paspalum, Cynodon, and Agropyron. Several microbial processes associated with improvements in root and shoot growth, nutrient content in biomass, and other benefits have been described. Promotion of plant growth has been associated with the synthesis of phytohormones and enzymes regulating several steps of plant development, nutrient mineralization by release of microbial molecules, biological nitrogen fixation, nutrient uptake facilitation by means of molecules such as exopolysaccharides, amongst others. Outstanding benefits of increased root growth, resulting in higher uptake of water and nutrients, either by phytohormones released by bacteria or by expanding root surface by AMF, have been reported. Biocontrol is another important property of PGPM, by a variety of mechanisms, including the synthesis of antimicrobial molecules, lytic enzymes, siderophores, and the release of specific inhibitory compounds such as hydrogen cyanide. Although improvements in forage management can enhance microbial performance, as shown for AMF, in general, inoculation with elite strains positively impacts growth parameters. Globally, the use of microbial inoculants has significantly increased in the past few years, but their application is still modest on grass pastures, which are generally degraded and would need special use of microbial inoculants for reclamation. Efforts towards increasing the use of PGPM in pastures can have deep positive environmental, economic, and social impacts worldwide.

**Keywords:** AMF; biofertilizers; biopesticides; inoculants; mycorrhiza; plant growth-promoting microorganisms; PGPM; PGPB; phytohormones

# 1. Introduction

The world's growing population requires increasing food production, but the expansion of new land areas for cropping has reached its maximum, whereas land degradation advances. According to the United Nations Environment Program [1], two billion people have been affected by land degradation, and more than 12 million hectares (Mha) of arable land are annually lost to desertification. Livestock is a major sector-driven land use and, according to Ritchie et al. [2], the combination of pastures for grazing and the land used



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**Copyright:** © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). to grow crops for animal feeding represents 77% of the global farming land. Meat consumption has increased rapidly since the 1980s, and considering the human population growth on the planet, estimated at 9.8 billion people by 2050, the increase in livestock production to meet this demand is inevitable. However, estimates are that about 49% of pasture ecosystems are under some level of degradation [3].

Land use for livestock production varies by country and can account for up to half of the total land area in continental regions [2]. In Brazil, beef livestock is a major economic activity, with pasturelands amounting to about 2.5-times of the area for other crops [4]. According to the Brazilian Association of Meat Exporting Industries (ABIEC, Associação Brasileira de Indústrias Exportadoras de Carnes), considering the inputs, services, and revenue of the whole meat business chain, livestock was responsible for 10% of the Brazilian total Gross Domestic Product (GDP) in 2019 [5]. Brazil is the largest world beef exporter, with 2.2 million tons and 14.4% of the international market share [6] and has the second-largest cattle herd, with 214.89 million heads [7]. This herd is distributed over 173 million hectares, equivalent to about 20% of the national territory, with 126 Mha composed of cultivated pastures [8,9].

Most of the Brazilian beef cattle's production occurs extensively in pastures, which depends on the forage quality and productivity [10]. The main forage grasses in these pastures are *Urochloa* (syn. *Brachiaria*) (usually referred to as brachiaria), followed by *Megathyrsus* (syn. *Panicum*), but there are also species of *Cynodon*, *Hemarthria*, *Andropogon*, *Avena*, *Pennisetum*, and *Paspalum* [11]. Based on the area used for seed production, 72.2% of the planted pasture area is occupied by *Urochloa* spp., and 25.7% by *Megathyrsus maximus* (syn. *Panicum maximum*) [9].

As in many countries with extensive pastures, inadequate management, lack of fertilization, and low use of technologies, in addition to the high occurrence of weeds and pathogens in the tropical environment, have led to the increasing degradation of pastures in Brazil. According to Dias-Filho [10], about 50% of the Brazilian pastures are considered degraded, 30% are in degradation processes, and only 20% are in good condition. Degraded pastures have low plant production and animal support capacity as well as soil physical, chemical, and biological degradation, which results in environmental degradation with negative effects on water resources, increased greenhouse gas (GHG) emissions, and desertification [10,12].

Carbon sequestration is another critical point to be considered in pasturelands. The topsoil represents the largest stock of Earth's C and contains approximately twice the C in vegetation and the atmosphere [13]. The CO<sub>2</sub> emissions from the soil come from the respiration of roots and soil organisms, and when respiration is greater than deposition, C is lost to the atmosphere [14]. Changes in land use, especially the conversion of forests into pastures or agricultural systems, represent the major source of CO<sub>2</sub> emissions in Brazil [15]. As the amount of soil C in areas of degraded pastures is lower than in well-managed areas [16], reclamation of pastures delineates a crucial scenario for C sequestration in the soil. For example, in Portugal, a recovery program of 42,000 hectares of degraded pastures over two years resulted in the sequestration of approximately 910 Gg (thousand tons) of CO<sub>2</sub> equivalents [17].

In addition, pasture degradation affects the economic sustainability of animal production. Considering only the fattening phase of the herd, meat productivity in a degraded pasture is around 2 at sign  $ha^{-1} yr^{-1}$  (1 at sign ~15 kg, used for cattle pricing in Brazil), while in a well-managed pasture, 12 at sign  $ha^{-1} yr^{-1}$  can be obtained [12]. With the drop in pasture productivity due to degradation, the stocking capacity and animal weight gain are reduced, leading many farmers to open new areas, resulting in significant economic and environmental losses [18]. For example, comparing zootechnical indices (birth rate, birth rate until weaning, age at the first calf, age at slaughter, and stocking) between good pastures and degraded pastures, for each hectare of recovered pasture, approximately three hectares could be released for other purposes (e.g., agricultural, forestry planting, or conservation) without reducing production [10].

With increasing political and general public pressures on sustainable food production systems, the development and use of technologies that allow increased production with less environmental impact are essential [19]. In this context, microbiological inoculants, also known as biofertilizers in some countries, are agricultural inputs composed of living microorganisms that can help the growth and development of plants by a variety of processes and have contributed to increasing the economic and environmental sustainability of food production systems [20]. Due to their benefits, the use of inoculants is now widespread globally, and impressive increases are expected in the following decade. In this scenario, Brazil is an important example, where the commercialization of inoculants went from 18.1 million doses in 2008 to 70 million in 2018 [20,21], and will soon overtake 100 million doses currently. Inoculants in Brazil have been used mainly in legume grain crops, with almost 90% for the soybean [Glycine max (L.) Merrill] crop [20]. However, in the past decade, the development of inoculants containing the plant-growth-promoting bacterium (PGPB) Azospirillum brasilense for cereals [22–24] was rapidly adopted by farmers, so that nowadays, over 10 million doses have been annually commercialized, mainly for the maize (Zea mays L.) and wheat (Triticum aestivum L.) crops [23], in addition to co-inoculation of soybean together with *Bradyrhizobium* spp. [24].

Benefits of the inoculation of legume pastures, especially alfalfa (*Medicago sativa* L.), with nitrogen-fixing rhizobia have been well known for a long time [25]. Conversely, although grasslands represent the great majority of the global areas with pastures, research, and use of microbial inoculants for pasture grasses are still inexpressive. In Brazil, approximately five years ago, the first inoculant carrying *A. brasilense* for *Urochloa* spp. [26] reached the market and started to call the farmers ´ attention. Great benefits have been reported since then, highlighting that the use of elite strains as microbial inoculants can improve pastures, promoting qualitative and quantitative increases in forage biomass and, consequently, increasing the productivity and sustainability of livestock production [20,26,27].

This review highlights the benefits of using inoculants for forage grasses, exploring the microorganisms already used in commercial inoculants and others with the potential to benefit pastures. As Brazil is a major country both in terms of the percentage of area with grassland pastures [9] and in the use of microbial inoculants [20,23], the country will be used as an example.

## 2. Microbial Inoculants

Microbial inoculants, also called biofertilizers in some countries, are products composed of microorganisms that can stimulate plant growth and development and are marketed in the liquid or solid form [20,28]. The first commercial inoculant was produced in 1856 in the United States of America. The first inoculant industry in Brazil was established in 1956 in the southern state of Rio Grande do Sul, initially focused on inoculants for soybean and subtropical forage legumes, such as alfalfa and clovers (*Trifolium* spp.); from the 1960s onwards, production was primarily focused on soybeans [20].

In Brazil, as in probably all countries that have used inoculants for more than 30 years, the preferred vehicle was peat, but the environmental impacts caused by the exploitation of peatlands and the increased costs of importation gave way to inoculants with liquid formulations. The first liquid inoculant was registered in Brazil in 2000, and currently, this vehicle represents more than 70% of the national market [20].

Soybean is the leading crop in the use of inoculants worldwide. In Brazil, the largest world soybean producer, annual inoculation was adopted in 80% of the total area cropped in 2019/2020 [21], leading to savings in N-fertilizer estimated at about US\$ 20 billion per year [20]. As commented in the introduction section, the first inoculant for grasses in Brazil was launched in 2009 for maize and wheat, with great acceptance by the farmers [23]. However, although the area of grasslands in the country is far greater than that used for other crops [5,9], the first commercial inoculant for this sector was available only in 2016, carrying the strains Ab-V5 (=CNPSo 2083) and Ab-V6 (=CNPSo 2084) of *A. brasilense*,

for seed inoculation of brachiarias, *Urochloa brizantha* (A.Rich.) RDWebster, and *Urochloa decumbens* (Stapf) RDWebster [26].

In addition to their economic importance, inoculants play an important environmental role, as they allow the complete replacement of N-fertilizers in legume crops such as soybeans [20,29], and the partial replacement in grasses such as maize [30]. Inoculation with *A. brasilense* can provide 20–40 kg ha<sup>-1</sup> of N per growing season in grasslands [26,27,31], with significant environmental benefits, as the use of each kg of N-fertilizer used results in the emission of approximately 10.5 kg of CO<sub>2</sub> equivalents [32]. It is also worth mentioning that in the soil, N-fertilizer derivatives, mainly nitrate (NO<sub>3</sub><sup>-</sup>), with high solubility and mobility, infiltrate and reach the water table, resulting in groundwater contamination, or are leached into water bodies where they exacerbate eutrophication [33].

The use of microorganisms with biocontrol activity in inoculants has also been discussed to reduce the use of pesticides in agriculture [34]. According to de Moraes [35], the use of pesticides in Brazil rose from 58,000 tons (or megagrams, Mg) in 1991 to 375,000 Mg in 2015, representing 9.2% of global use. Pesticides can threaten human and animal health, contaminate soil and water, and lead to the selection of increasingly resistant pathogens. Microbial biocontrol agents which have antagonistic mechanisms against pathogens, such as parasitism, competition for nutrients and space, and antibiosis [34,36], are a viable alternative.

#### 3. Plant Growth-Promoting Microorganisms

Plant-growth-promoting microorganisms (PGPM) colonize the rhizosphere, root surface, phyllosphere, and internal tissues of vegetative plant organs and promote plant growth by different mechanisms [37]. An impressive and increasing number of plant growth-promoting mechanisms have been reported for PGPM, ranging from facilitating nutrient uptake to biocontrol activity; they are discussed in this review and are summarized in Figure 1.



**Figure 1.** Main plant-growth-promoting (PGP) traits associated with the main bacteria used in studies or in commercial products for pastures with grasses, *Azospirillum, Pseudomonas, Bacillus, Rhizobium*, and arbuscular mycorrhizal fungi.

The main microbial processes related to plant-growth promotion include: biological nitrogen fixation (BNF); synthesis of phytohormones; (e.g., auxins, cytokinins, gibberellins, abscisic acid (ABA), and ethylene); of enzymes such as ACC (1-aminocyclopropane-1-carboxylic acid)-deaminase (which reduces ethylene levels in plants); nutrient mineralization and mobilization; solubilization of phosphorus (P); iron (Fe), and potassium (K); molecules linked to the induction of plant systemic resistance; antibiosis mechanisms [22,26,37,38].

The ability to fix atmospheric nitrogen (N<sub>2</sub>) depends on the presence of the nitrogenase enzyme complex in the bacterium, which is composed of two subunits, Fe-protein, and MoFe-protein. In the BNF process, the Fe-protein is responsible for receiving and transferring electrons, while the MoFe-protein is the site with adequate conditions for N<sub>2</sub> reduction. The complex can reduce N<sub>2</sub> into NH<sub>3</sub>, which is converted at physiological pH into NH<sub>4</sub><sup>+</sup>, which will then be transported and assimilated by the plant [25,39].

The production of phytohormones by PGPM is one of the main factors responsible for stimulating plant growth. Auxins play a critical role in the formation of lateral roots, root hairs, shoot architecture, responses to light and gravity, and vascular development. On the other hand, gibberellins are known to stimulate cell elongation, break the dormancy of buds in stems, and promote germination and development of primordial leaves and fruits [40–42], while cytokinins act in embryogenesis, cell division, differentiation, and formation of lateral roots, and thus may affect the water and nutrient availability by the roots [43]. ABA plays a fundamental role in the seed germination process and acts on water and saline stress tolerance by inducing stomatal closure and accumulation of amino acids and soluble sugars [44]. Studies show that ethylene acts synergistically with auxin in root growth, root hair proliferation, regulation of hypocotyl elongation, and apical hook formation [45]. In addition, some PGPMs have the ability to decrease ethylene levels produced by plants during stress situations, which is beneficial, as high concentrations of ethylene inhibit plant growth due to induction of senescence [46]. The production of ACC-deaminase enzyme degrades the aminocyclopropane carboxylic acid, a precursor of ethylene [47].

Acquired systemic resistance is a plant protective mechanism against pathogens and can be constitutive or induced [48]. This protection can be induced by molecules (elicitors) produced by PGPM that act as signalers that regulate the expression of genes related to plant defense, such as salicylic acid, jasmonic acid, and ethylene [49,50]. There are also reports about the involvement of nitric oxide in mechanisms related to drought and salinity tolerance [51].

Soil microorganisms play important roles in the cycle of nutrients such as P, K, S, and Fe [52]. The increase in P availability occurs by the release of organic acids, chelation of phosphate-bound cations, or lowering of the pH in the rhizosphere [53], while P mineralization is mediated by the action of phosphatases, phytases, or lyases [54,55]. The solubilization of K occurs by the production of low molecular weight molecules such as citric, oxalic, tartaric, succinic, and malic acids [56]. The availability of S occurs by a complex process carried out by some microorganisms in which elemental-S is oxidized to sulfate [57], or organic forms are mineralized by the action of sulphatases. The increased availability of Fe relies on the ability of many organisms to reduce iron oxides [58], as Fe availability and solubility increase in the reduced state [59]. Many microorganisms can change the oxidation status of reduced Fe when using it as a substrate for energy generation or using the oxidate status as a final electron acceptor in anaerobic respiration, facilitating its availability [60].

Antibiosis mechanisms have also been associated with PGPM, via synthesis of molecules such as cyclic lipopeptides, phenazine, tropolone, and surfactins. They can produce lytic enzymes, such as chitinases, which suppress pathogens, and siderophores that chelate Fe in the rhizosphere, hindering the growth of other microorganisms, especially pathogens that have a lower ability to compete for Fe [61]. In addition, siderophores have the ability to chelate Fe, facilitating its absorption by plants [62].

Some PGPM synthesize exopolysaccharides that help root colonization, retain moisture and nutrients, and protect against pathogens. In addition, they can colonize the root surface and use nutrients, preventing the proliferation of harmful microorganisms and producing signaling molecules such as lumichrome, protecting the plant against biotic and abiotic stresses [61].

PGPM has been found associated with several species of cereals and forage grasses, with a large number of reported genera, with emphasis on *Pseudomonas*, *Bacillus*, *Rhizobium*, and *Azospirillum*, in addition to mycorrhizal fungi [22,26,63], which will be emphasized in this review.

# 4. Plant Growth-Promoting Bacteria (PGPB)

#### 4.1. Azospirillum spp.

Regarding the inoculation of grasses, the species *A. brasilense* has stood out. In Brazil, especially in the past decade, there are numerous reports of yield increases in plants of agronomic interest due to inoculation with *A. brasilense* [22,26,27,64–69]. For example, inoculation with strains Ab-V5 and Ab-V6 of *A. brasilense* increased yields by 27% in maize and 31% in wheat [22]. Later, in a meta-analysis based on 103 field experiments with maize, inoculation with these two strains increased grain yield and root mass by 5.4% and 12.1%, respectively [69]. Inoculation with strains Ab-V5 and Ab-V5 and Ab-V6 of *A. brasilense* also increased yields in other crops such as rice (*Oryza sativa* L.) and sugarcane (*Sacharum* spp.) [65,67].

Azospirillum spp. may contribute to the plant's N nutrition by the BNF process [70,71] that can reach 20–40 kg ha<sup>-1</sup> of N [31], allowing a reduction in the use of N-fertilizers [67]. Inoculation of *A. amazonense* (syn. *Nitrospirillum amazonense*) *A. brasilense*, and *A. lipoferum* in *U. brizantha* BRA 003719, *U. humidicola* BRA 005011, and *U. brizantha* BRA 000591 grown in concrete cylinders was responsible for providing 26.2%, 19.6%, and 24.8% of the plant accumulated N, corresponding to 7.02, 3.08, and 8.43 kg ha<sup>-1</sup> of N, respectively [72]. In field trials with ten cultivars of *M. maximus*, the inoculation with *A. lipoferum*, *A. brasilense*, and *A. amazonense* supplied 5 to 10 kg ha<sup>-1</sup> of N by the BNF process [73]. In another study performed under greenhouse conditions, inoculation with *A. brasilense* provided 40 kg ha<sup>-1</sup> of N to M. maximus and *Digitaria decumbens* [74].

Moreover, these bacteria are able to synthesize and release phytohormones [37,75,76], in addition to inducing plant tolerance to abiotic and biotic stresses due to the production of molecules such as jasmonic and salicylic acid, and stimulating the activity of detoxifying enzymes like catalases, superoxide dismutase, and ascorbate peroxidase, as well as prolin, an osmoregulator, in both shoots and roots [20,37,77,78]. Some strains are also capable of phosphate solubilization [79]. Among these benefits, the production of phytohormones is probably the most studied and important for promoting plant growth. The synthesis of indole-3-acetic acid (IAA) by *Azospirillum* spp. [76,80] favors the growth of plant root tissues and may triplicate root growth, increasing its capacity for uptake of water and nutrients from soil [22,81,82]. In addition to IAA, molecules like gibberellins (GAs) [83,84], ethylene [85], cytokinins [86,87], and ABA [76,84,85] can contribute to plant growth.

Despite the vast majority of inoculations in grasses being carried out in important agricultural crops, it is worth mentioning that *A. brasilense* was first isolated in Brazil from the rhizosphere of the pasture *D. decumbens* Stent [88]. Another example of strain isolated from the rhizosphere of pastures is of UAP55, from *Brachiaria* (syn. *Urochloa*) mutica (Forssk.) Stapf in Mexico, which increased wheat grain yield by 43%, 15%, and 11%, in combination with different doses of N (60, 90, and 120 kg ha<sup>-1</sup> of N, respectively), compared with the non-inoculated controls [89]. Another important *A. brasilense* strain commonly used is Sp7, isolated from the forage grass *Cynodon dactylon* (L.) Pers., and its variant strain Cd [90,91].

In Brazil, evaluations of *A. brasilense* strains Ab-V5 and Ab-V6 in pastures started in 2011 with *U. brizantha* and *U. decumbens*. Field trials were performed with seed inoculation and a basal level of 40 kg ha<sup>-1</sup> of N. Inoculation resulted in average increases in shoot biomass by 17.3% for *U. brizantha* and 12.5% for *U. ruziziensis*, while N accumulated in shoots increased by 25% [26]. The study resulted in the first commercial inoculant for pastures with brachiarias in 2016, and since then, several field trials have been performed and confirmed the positive effect of Ab-V5 and Ab-V6 strains on grass pastures in Brazil [92–96].

As most pastures in the world, including Brazil, are established with perennial grasses, farmers demanded studies to investigate the feasibility of leaf spray inoculation. Greenhouse and field experiments were performed with brachiarias inoculated with strains Ab-V5 and Ab-V6 via seeds or leaf spray; all treatments received 40 kg ha<sup>-1</sup> of N at sowing and half received a second application with 40 kg ha<sup>-1</sup> of N 30 days after emergence [27]. Under greenhouse conditions, inoculation with *A. brasilense* impressively increased root traits, including biomass, tissue volume and density, total and specific length, and the incidence of root hairs in *U. brizantha* and/or *U. decumbens* (syn. *Urochloa eminii*) [27]. Following, field trials were performed with *U. ruziziensis* (syn. *Urochloa eminii*), and the benefits of seed inoculation at the pasture establishment, or leaf spray in established pastures were confirmed, either when they received a basal level of 40 kg ha<sup>-1</sup> of N, or when receiving another application of 40 kg ha<sup>-1</sup> of N 30 days after seedling emergence. On average, shoot biomass increased by 22%, in addition to 13% of N and 10.4% of K concentrations in leaves [27].

## 4.2. Pseudomonas spp.

*Pseudomonas* are Gram-negative bacteria with a very versatile metabolism, commonly found in soil and water. They are considered aerobics but can use nitrate as a final acceptor of electrons during anaerobic respiration. The optimal temperature for growth is between 25 and 30 °C [97].

The main species referred to as a growth promoter in agriculture is *Pseudomonas fluorescens*. Among the main properties of *Pseudomonas* species are their abilities to produce auxins, notably IAA [97]. There are also reports of the production of cytokinins, gibberellins, and ACC-deaminase [98]. In a study carried out by Hungria et al. [27], inoculation of *Pseudomonas fluorescens* strain CNPSo 2719 (=CCTB 03) via seeds or leaf spray increased the total length and the number of root branches of *U. brizantha* and *U. decumbens*, which was attributed to the synthesis of phytohormones.

The synthesis of siderophores is another plant-growth-promoting trait of *Pseudomonas*. These low molecular weight molecules increase the mobility and availability of Fe, allowing its translocation through the plant cell membranes. Siderophores play an important role in plant growth and in the control of phytopathogens, preventing them in the rhizosphere by sequestering and thus limiting the Fe necessary for their development [99,100].

Suppression of pathogens by *Pseudomonas* can also occur based on other mechanisms, including the synthesis of antimicrobial molecules such as pyrrolenitrine, pyocyanin, and 2-4-diacetylphloroglucinol; competition for nutrients and space; the production of lytic enzymes, such as  $\beta$ -1,3-glucanases, which degrade chitin and glucans present in the cell walls of fungi; by the release of hydrogen cyanide (HCN); by degradation of toxins produced by pathogens [97]. In a study performed by Rodriguez and Pfender [101], the inoculation of *P. fluorescens* strain Pf-5 inhibited the mycelial growth of *Sclerotinia homoeocarpa* in cuts of leaves of *Agrostis* sp., while no inhibition occurred when inoculating a mutant *P. fluorescens* lacking the synthesis of antibiotics.

Phosphorus, after N, is the most limiting macronutrient for plant development participating in various metabolic processes, including cell division, photosynthesis, biosynthesis of macromolecules, energy metabolism, and signal transduction [102]. However, most of the soil P is precipitated, due to bonds with cations, immobilized in oxides and clay, adsorbed, or is in the organic form, being unavailable for plants. Considering the P present in the soil surface layer, estimated at 50 to 3000 mg P kg<sup>-1</sup> of soil, only 0.1% is available to the plants [103]. In addition to mechanisms that facilitate P absorption, a crucial feature of some *Pseudomonas* strains is their ability to solubilize inorganic phosphates to forms that can be assimilated by plants. For example, the use of *Pseudomonas plecoglossicida* along with rock phosphate in maize and wheat resulted in a higher concentration of P in shoots, roots, and grains [104]. However, it is convenient to note that many bacteria can be described as able to promote plant growth, but they may also be highly pathogenic to plants, animals, and humans. Therefore, it is mandatory to exclude these bacteria from any type of commercial use in agriculture, as is the example of *P. plecoglossicida*, first described as the causal agent of hemorrhagic in ayu fishes (*Plecoglossus altivelis*) [105], or the often-cited plant growth properties of strains of the *Burkholderia cepacia* complex, one of the most dangerous human pathogens, that are widespread in natural vegetation, such as natural ryegrass pastures in Portugal [106].

Forage grasses can benefit from inoculation with *Pseudomonas*. Under greenhouse conditions, inoculation of *U. brizantha* cv. BRS Piatã with *P. fluorescens* strain BRM-32111 increased the concentration of primary metabolites, such as carbohydrates and proteins, as well as plant biomass, N concentration, and chlorophyll concentration after 35 days of growth [107], confirming previous results on shoot and root biomass increases [108]. In another study, Begun et al. [109] inoculated *Pseudomonas grimontii* strain Bc09, *Pseudomonas veronii* strain E02, and *P. fluorescens* strain Oj24 in *Panicum* (syn. *Megathyrsus*) virgatum cv. Alamo and Cave-in-Rock (CIR) under cadmium (Cd) stress and reported increases in root dry biomass by 57%, 93%, and 96% in cv. Alamo, and by 70%, 137%, and 29% in CIR, and increases in the shoot dry biomass by 105%, 130%, and 105% in cv. Alamo and 97%, 169%, and 42% in CIR, respectively.

Inoculation with *P. fluorescens* strain CNPSo 2719 increased the rate of stem elongation and leaf expansion, and the number of basal tillers in *U. brizantha* [96]. In *M. maximus*, inoculation with the same strain of *P. fluorescens* resulted in increases in shoot and root dry weight, tiller number, and uptake of N and magnesium (Mg) [110]. In addition, inoculating the same strain into a hybrid of *Urochloa* spp. resulted in increases in shoot and root dry mass, as well as the number of tillers [111].

The good performance of *P. fluorescens* strain CNPSo 2719 in Brazil has resulted in its use in commercial inoculants for *Urochloa* since 2021. Inoculation of the strain via seeds or leaf spray increased shoot biomass by 15.2 and 14.2%, respectively. In addition, seed and leaf spray inoculation resulted in average increases in leaf concentration of 33.3 and 36.6% for P, and 10.6 and 13.6% for K, respectively [27].

It is also worth mentioning that *P. fluorescens* is considered a "mycorrhizal helper bacterium", capable of stimulating an increase in the rate of root colonization by arbuscular mycorrhizal fungi (AMF) [112], symbionts that increase the area of soil explored by roots and are essential for grasses to survive, grow, and reproduce in degraded tropical soils [113–115].

# 4.3. Bacillus spp.

In 1835, Christian Gottfried Ehrenberg cited the genus *Bacillus* for the first time. They are Gram-positive, facultative anaerobic, and endospore-forming bacteria. Because of their ability to sporulate, *Bacillus* can survive under extreme pH, temperature, and salinity [116]. These properties made them very attractive to the industry, as spore-based bioproducts may have a long shelf life, making easier the logistics of storage and distribution [117].

Many studies have demonstrated the ability of *Bacillus* to promote plant growth [116]. For example, in a study carried out with *U. brizantha*, Araujo et al. [118] reported that seed inoculation with *Bacillus* sp., previously isolated from the rhizosphere of this grass promoted, an increase in tillering and leaf expansion. Studies have also shown that *Bacillus* can improve the development, nutrient uptake, and yield of other grasses such as maize [119], wheat [79], and millet [*Pennisetum glaucum* (L.) R.Br.] [120]

Here it is worth mentioning that the taxonomy of *Bacillus* is very complex and has gone through several modifications. The last one proposed that the genus *Bacillus* should be limited to the species members of the *B. subtilis* and *B. cereus* clades, resulting in the description of the new genus *Priestia* [121]. However, for practical purposes, we will refer to the generic genus *Bacillus*, as the proper taxonomic description of several strains used in the

studies is not always clear. For updated information on the taxonomy of *Bacillus*, the website (https://lpsn.dsmz.de/genus/bacillus; accessed on 11 July 2022) should be consulted.

Several *Bacillus* are known to have the ability to produce siderophores. In addition, they can improve solubilization or facilitate the mobilization of nutrients in the soil, allowing plant absorption [122]. In some species, such as *Bacillus cereus*, *Bacillus chitinolyticus* (now *Paenibacillus chitinolyticus*), *Bacillus circulans* (now *Niallia circulans*), *Bacillus coagulans* (now *Weizmannia coagulans*), *Bacillus fusiformis* (now *Lysinibacillus fusiformis*), *Bacillus megaterium* (now *Priestia megaterium*), *Bacillus mycoides*, *Bacillus polymyxa* (now *Paenibacillus pumilus*, and *Bacillus subtilis*, the ability to solubilize phosphates has been demonstrated [123].

Hussain et al. [124] reported that 14 *Bacillus* isolates obtained from the maize rhizosphere could solubilize zinc (Zn), and Basak and Biswas [125] observed increased K absorption in sorghum (*Sorghum vulgare* Pers.) inoculated with *Bacillus mucilaginosus* (now *Paenibacillus mucilaginosus*). In addition, *Bacillus* can make P available to plants by other mechanisms such as the production of enzymes, including phosphatases and phytases, which act as catalysts in the conversion of organic to inorganic P, that is, in the mineralization of P [126].

*Bacillus* species can also produce and modulate phytohormone levels in plants, thus regulating plant growth and stress responses [127]. Gas chromatography coupled to mass spectrometry analyses indicated the synthesis of auxins, cytokinins, abscisic acid, and gibberellins in *Bacillus aryabhattai* SRB02 (now *Priestia aryabhattai*) [128]. In another study, an isolate of *Bacillus* sp. producer of auxins led to greater tillering in *U. brizantha* [118]. In addition, some *Bacillus* expresses the enzyme ACC-deaminase, which helps plant growth by reducing the levels of ethylene [129].

Exopolysaccharides (EPS) produced by some *Bacillus* have shown effects on various soil properties and plant productivity. EPS can stabilize soil aggregates and regulate the movement of nutrients and water to plant roots by biofilm formation. In addition, they help in nutrient uptake, stimulating plant growth [130]. Because biofilms are negatively charged, they are effective sequesters of charged heavy metals. Therefore, the production of biofilms is relevant to salt stress as they bind to sodium (Na<sup>+</sup>), leading to a decrease in the accumulation of this ion in plants, conferring increased salt tolerance [130,131].

Due to the production of antimicrobial molecules, lytic enzymes, and other organic molecules, *Bacillus* are the most commonly used bacteria in the biological control of plant pathogens. Concerning antimicrobial compounds, iturin synthesized by *B. subtilis* showed activity against *Aspergillus carbonarius* by affecting the fungal membrane permeability [132], and it was also very effective against several soybean seed fungi [133]. In a study carried out by Hanif et al. [134], fengicin produced by *Bacillus velezensis* strain FZB42 decreased the pathogenicity of *Fusarium graminearum* in wheat, altering the membrane permeability and fungal cell structure.

Lytic enzymes produced by some *Bacillus* are also important for the suppression of plant pathogenic microorganisms, especially fungi. They are capable of destabilizing the cell envelope, creating pores in the cell membrane, and inactivating the ribosomes of certain fungi [135]. Saxena et al. [126] showed that *B. subtilis, B. velezensis, B. cereus, B. thuringiensis, B. licheniformis,* and *B. pumilus* are producers of certain enzymes such as chitinases, glucanases, proteases, and lipases. Agarwal et al. [136] reported the antifungal activity of *B. pumilus* strain MSUA3 due to the production of chitinases and surfactin, while El-Bendary et al. [137] observed that *Bacillus* isolates 8Es, 25Sp, and 27Sp, producers of chitinases, proteases, and glucanases were able to inhibit 70–88% of the mycelial growth of the fungus *Macrophomina phaseolina* in vitro.

Apparently, bacteria belonging to the genus *Bacillus* (and their subdivisions) are the richest in plant-growth-promoting mechanisms. However, we should again call attention to their careful use in agriculture, as many species are highly pathogenic to animals and humans, such as *B. cereus* and *B. anthracis*. Therefore, the choice of proper species and

strains based on the correct taxonomic classification is mandatory for their successful use as inoculants, biofertilizers, or biopesticides.

#### 4.4. Rhizobium spp., Bradyrhizobium spp., and Other "Rhizobia"

In 1888, the Dutch microbiologist Martinus Willem Beijerinck was the first to isolate and cultivate a microorganism present in legume nodules, later called *Rhizobium* [138]. Rhizobia is a generic term for bacteria that are symbiotic and are classified into several genera, including *Rhizobium*, *Bradyrhizobium*, *Sinorhizobium*, *Mesorhizobium*, among others. Aerobic rhizobia live saprophytically in soil, have an optimal growth temperature between 25 and 30 °C and process several mechanisms that can promote plant growth [139].

Besides the leading role of nitrogen fixation in symbiosis with legumes, and sometimes under free-living conditions [25], other mechanisms to promote non-legume growth include the solubilization of phosphates and the synthesis of phytohormones. Rhizobia are capable of producing and releasing carboxylic acids into the soil, such as citric, gluconic, oxalic, lactic, tartaric, and acetic acids, which decrease the pH of the medium and promote the release of phosphate anions from Fe, aluminum (Al), and calcium (Ca) complexes [140]. Silva et al. [141] observed an increase in shoot phosphorus concentration when inoculating maize and oat (*Avena sativa* L.) with *Bradyrhizobium* spp. isolates from *Desmodium incanum* (Sw.) DC.

The main phytohormones produced by rhizobia are auxins, cytokinins, and gibberellins [139], detected and quantified using HPLC and/or GC-MS [142,143]. Among the auxins, IAA is the most reported phytohormone. In a greenhouse experiment inoculation with auxin-producing rhizobia SEMIA 816, UFRGS Lc134, UFRGS Lc323, UFRGS Lc348, UFRGS Lc510, and UFRGS Lc524 resulted in increased shoot and root dry mass and root volume in *M. maximus, Paspalum saurae* (Parodi) Parodi, *U. decumbens*, and *Lolium multiflorum* Lam [144]. The effects of gibberellins were observed when inoculating canola (*Brassica napus* L.) and lettuce (*Lactuca sativa* L.) seeds with the mutant strain 127E14 of *Rhizobium leguminosarum* for the production of adenosine, the precursor molecule for gibberellin biosynthesis. In this case, there was no early root growth compared with the inoculation with the wild-type strain [145].

Other plant-growth-promoting molecules synthesized by rhizobia are lipochitooligosaccharides, also known as Nod factors. According to Souleimanov et al. [146], these molecules are involved in the cell cycle, leading to stimulation of mitotic divisions in protoplasm cultures of legumes and non-legumes. In grasses such as maize, plant growth- promotion by the addition of exogenous Nod factors synthesized by *Rhizobium tropici* and *Bradyrhizobium diazoefficiens* has been reported [147,148]. Lumichrome is another compound from rhizobia produced by the photochemical degradation of riboflavin. This molecule can induce the expression of genes responsible for cell growth and mitotic division and increase CO<sub>2</sub> concentrations in the rhizosphere [149]. Lumichrome has been reported as being able to promote the growth of legumes and grasses [150]. For example, the application of lumichrome to maize plants increased the photosynthetic rate [151].

Most rhizobia e.g., *Rhizobium tropici* strain CIAT 899, *Sinorhizobium* (syn. *Ensifer*) *meliloti* strain SU-47, and *R. leguminosarum* bv. trifolii strain TA-1, are well-known for their capacity of synthesizing EPS [152]. The benefits of EPS for plant growth were detailed in on the item 4.3 of *Bacillus*.

Some rhizobial species such as *R. leguminosarum*, *S. meliloti*, and *Bradyrhizobium japonicum* can suppress pathogens by competing for nutrients, or by producing antibiotics and enzymes such as chitinases and glucanases, or siderophores [139]. This capacity has been demonstrated in studies in which these bacteria were able to inhibit phytopathogens such as *M. phaseolina*, *Fusarium oxysporum*, *Fusarium solani*, *Sclerotinia sclerotiorum*, and *Rhizoctonia solani* [153,154]. In addition, some rhizobia can decrease the level of ethylene in plants by production and release of the enzyme ACC-deaminase (1-aminocycopropane-1-carboxylic acid) [139], modulating plant growth.

#### 5. Plant Growth-Promoting Fungi

## Arbuscular Mycorrhizal Fungi (AMF)

More than 80% of terrestrial plant species have their roots colonized by arbuscular mycorrhizal fungi (AMF), forming the symbiosis known as arbuscular mycorrhiza (or endomycorrhiza) [155]. In most cases, the association with AMF provides benefits to plants, such as increased resistance to pathogens [156], reduction of water stress effects [157,158], and mitigation of phytotoxicity caused by heavy metals [159]. However, the main benefits are the increase in soil exploitation and, consequently, the efficiency of nutrient absorption [160–162]. According to some studies, mycorrhizal plants absorb more efficiently N, K, Ca, Mg, Zn, copper (Cu), manganese (Mn), and especially P [160,163–167]. In addition, AMF can increase the quality and sustainability of natural ecosystems and agroecosystems as it improves soil physical, chemical, and biological attributes [168–171], and also reduces nutrient leaching and greenhouse gas emissions [167].

AMF are obligate biotrophs with coenocytic mycelium, asexual sporulation and ubiquitous distribution [155], and comprise a unique phylum, Glomeromycota [172], with 341 described species (http://www.amf-phylogeny.com/; accessed on 13 July 2022). When associated with roots, AMF presents internal hyphae, which grow in the apoplast of cortical cells, and external hyphae, which extend through the soil [162]. AMF symbiosis increases the area of soil exploited by roots because the external hyphae are longer and have a smaller diameter (between 2  $\mu$ m and 10  $\mu$ m) than the fine roots and the root hairs, which makes them able to explore small pores and soil regions not reached by the root system, increasing the absorption of nutrients per surface unit [162]. AMF absorbs water and nutrients from the soil through the external hyphae, transferring them to the internal hyphae and, in the arbuscles, exchanging these resources by photosynthates [155,173]. The formation and maintenance of the symbiosis requires carbon (C), so estimates point out that 4 to 20% of the total C fixed by photosynthesis can be transferred to the fungi [173,174].

Grasses, in general, have a high level of association with AMF, which becomes clear when the rate of mycorrhizal colonization in their fine roots is quantified. Under greenhouse conditions, Zangaro et al. [11] observed mycorrhizal colonization rates above 90% in the roots of the forage grasses *M. maximus*, *U. brizantha*, and *Paspalum notatum* grew in the soils of Brazilian pastures located in the Atlantic Forest biome. In the Cerrado biome, rates of colonization of 44% in *U. decumbens*, 51% in *U. brizantha*, and 52% in *U. humidicola* colonized by native AMF species (notably *Gigaspora* spp., *Scutellospora* spp., *Glomus* spp., *Acaulospora* spp., and *Entrophospora* spp.) have been reported [175]. Cavagnaro et al. [176] found mycorrhizal colonization rates of 60% for *U. brizantha* and 80% for *Paspalum dilatatum* Poir. inoculated with a mixture of three AMF species [*Rhizophagus irregularis* (syn. *Glomus intraradices*), *Simiglomus hoi* (syn. *Glomus hoi*), and *Funneliformis mosseae* (syn. *Glomus mosseae*)]. In another study, Rondina et al. [165] assessed the rate of mycorrhizal root colonization of *Sorghum* sp. and *Pennisetum purpureum* Schumach, grown in soil from the Atlantic Forest biome and reported rates of about 80%.

High mycorrhizal colonization rates are also typical in forage grassroots in the field, although the values are generally lower than those observed under greenhouse conditions. In a study carried out in Palmira, Colombia, 1000 m of altitude, with the hybrid of *Urochloa* 'Mulato' and with *U. humidicola*, the root mycorrhizal colonization was 48% and 72%, respectively [177]. However, high rates of mycorrhizal colonization, above 75% were found in pastures composed of *P. notatum*, *Cynodon* sp., and *Urochloa* spp. in the Atlantic Forest [115,178–180], and in the Pantanal [178] Brazilian biomes. In field studies conducted in the Brazilian Cerrado, Ramos et al. [181] found mycorrhizal colonization rates between 51% and 70% for *M. maximus* and 60% for *U. humidicola*. Moraes et al. [182] observed values between 25% and 48% for *M. maximus*, and 41% and 80% for *U. brizantha*. In general, in soils under forage grasses the densities of AMF spores and external mycelium are also high and correlate positively with the rate of root mycorrhizal colonization [179,180,184,185].

Together, these data indicate a high investment of C by the host plant in the symbiosis and a high multiplication capacity of AMF by grasses [115,179].

The rate of mycorrhizal colonization in forage grassroots can be positively affected by interspecific interactions between grasses and legumes [182,183]. In a study on the crop-livestock integration system in the Cerrado, with pasture and legume intercropping in the off-season [e.g., *U. brizantha* or *M. maximus* + *Vigna unguiculata* (L.) Walp. or *Cajanus cajan* (L.) Millsp.], Pires et al. [183] found that the rate of mycorrhizal colonization in grassroots increased by up to 90% when intercropped with legumes, compared with single grass cultivation. As a result, the productivity of the soybean cropped following the intercropping was up to 16% higher, showing a significant positive relationship with the increase in the rate of mycorrhizal colonization previously verified in the pasture. Similar results were obtained by Moraes et al. [182] but using *U. ruziziensis* instead of *M. maximus* intercropped with legumes. Also, edaphic and climatic characteristics, such as pH, moisture, organic matter, and soil P availability [186,187], photoperiod, temperature, and rainfall [179,180] can affect the interaction between grasses and AMF, causing variations in the rates of root mycorrhizal colonization in the field.

Commonly, the soils occupied by pastures in Brazil, natural or planted, are acidic and have low availability of nutrients, especially P [188–191]. Although forage grasses such as *Urochloa, Megathyrsus, Paspalum*, and *Cynodon* have fine roots with typical morphological traits to exploit a large volume of soil, with total and specific root lengths, small diameter, and high incidence of long root hairs [115,178,180], studies have shown that arbuscular mycorrhizae are of great importance for the acquisition of nutrients and, thus, for the establishment, growth, and regrowth of these plants in low fertility soils [114,165,192]. Indeed, there are reports that *M. maximus*, *U. brizantha*, and *P. notatum* grown in soil with low P availability ( $\approx$ 1.0 mg dm<sup>-3</sup>) were unable to survive 20 days after emergence without mycorrhiza [114]. Similar results were obtained for *Sorghum* sp. And *P. purpureum* in soil with low available P (0.89 mg dm<sup>-3</sup>) in the absence of AMF [165].

Other studies confirm the importance of colonization of pasture grasses with AMF. Cavagnaro et al. [176], in an experiment conducted in pots with sterile sand receiving nutrient solution with low nutrient content, with or without AMF inoculation, found that *P. dilatatum* and *U. brizantha* presented twice as much, and the triple production of both shoot and root biomass, respectively, compared with the non-inoculated controls. In addition, leaf P concentration and the number of tillers increased by 16% and 95%, respectively, in mycorrhizal U. brizantha plants [176]. In a study performed with *U. brizantha*, *U. decumbens*, *U. humidicola*, and *M. maximus* grown in pots containing sterilized soil and available P between 2.0 and 2.5 mg kg<sup>-1</sup>, with or without inoculation with a pool of AMF species native to the Brazilian Cerrado, inoculation resulted in average increases of 67% in shoot and of 32% in root biomass [175]. Furthermore, on average, P uptake doubled in inoculated plants compared with uninoculated controls [175]. In another study to evaluate the ability of grasses growing in substrates with low P to resprout after 60% defoliation to simulate grazing, AMF inoculation promoted greater and faster regrowth of *U. brizantha* and *Agropyron elongatum* (Host) P.Beauv [192].

In low fertility soils, the nutrient requirement of grasses, in addition to their rapid growth rates, can lead to the formation of a nutrient depletion zone in the rhizosphere [115], as nutrient absorption often occurs faster than their movements in the soil and their rates of mineralization from the soil organic matter [115,193]. In this context, the symbiosis with AMF ensures that forage grasses obtain nutrients beyond the depletion zone, which helps to explain the maintenance of high rates of mycorrhizal colonization by these plant roots [115,179,180], and the high responsiveness to mycorrhization, in terms of biomass production [165,175,176,192]. Furthermore, AMF greatly influences the spread of grasses, as they can anticipate and/or increase flowering when associated with AMF [165,194]. However, when P availability is high, the AMF symbiosis can decrease the grass biomass production [114,192], a phenomenon known as "growth depression" [195]. When cultivated in sterile soil containing 32 mg kg<sup>-1</sup> of P, non-inoculated *U. brizantha, P. nota*-

*tum*, and *M. maximus* showed between two and three times more shoot biomass than AMF-inoculated plants after 90 days of growth [114]. In the same soil, non-inoculated *P. purpureum* exhibited, on average, 11% more shoot biomass than inoculated individuals after 100 days of growth [165]. Cavagnaro et al. [192] observed that when cultivating *A. elongatum* in a sterile, inert substrate with a high supply of P via nutrient solution (1.0 mM), non-inoculated plants produced 47% more shoot biomass and 32% more tillers than those inoculated with AMF, 22 days after defoliation, simulating grazing. "Growth depression" events occur due to the C cost imposed on the host plant by the AMF in a condition where the fungi would not be required to obtain adequate amounts of P for the host growth [165,192,195]. As a result, when mycorrhizal colonization in the roots is high (which is frequent in grasses), the fungal demand for photosynthates can be high, and the greater allocation of C to maintain large fungal structures in the roots and soil may contribute to a lower accumulation of biomass by the mycorrhizal plant [165,196,197].

Some AMF species may be more effective than others, differing in root colonization rates, nutrient uptake, and the response levels they induce in the host plant [168,198]. The most effective species have been used, singly or in a consortium, to develop inoculants based on AMF [199,200]. Several companies worldwide have developed and marketed AMF-based inoculants for agricultural, turf, nursery, and phytoremediation applications (e.g., Rootella<sup>®</sup>, MycoApply<sup>®</sup>, Privi Mycoxol<sup>®</sup>, Mobilizer<sup>®</sup>, Myco Gold<sup>®</sup>, Root Plus <sup>®</sup>, RISEHoP<sup>®</sup>, MYKE<sup>®</sup> Pro) [199–201]. In Brazil, Rootella<sup>®</sup> was the first commercial inoculant based on AMF (*Rhizophagus intraradices*) [202] and resulted in average increases in grain yield by 54% and 25%, for maize and soybean, respectively, as well as a reduction in the amount of P-fertilizer required to achieve high yields [203,204]. Currently, other inoculants containing AMF are under development and in the registration process in Brazil [202,205,206]. However, as far as we know, despite the potential of AMF to increase productivity and decrease the degree of soil degradation, no study on the application of commercial inoculants containing AMF in tropical forage grasses has been developed in the country to date.

## 6. Final Remarks

We have shown that several microorganisms, including bacteria and fungi, can highly promote plant growth, and in this review, we focused on the opportunity of improving quantitatively and qualitatively the production of forages with grasses. As grasslands occupy far more areas than crops, their improvement may not only enhance livestock production but also release land for other crops without the need to advance into deforestation. Improving pasture productivity and reducing degradation also have important social impacts, with higher economic inputs to the farmers. Environmental benefits arise from the possibility of partially replacing synthetic fertilizers and pesticides, helping to mitigate the emission of greenhouse gases in addition to decreasing pollution of water reservoirs by chemicals.

Certainly, some microorganisms may contribute to more than one mechanism, namely multifunctionality. This review points out the importance of selecting elite strains with superior performance to be applied as inoculants or biopesticides. The commercialization of microbial inoculants has significantly increased in the past few years; however, their use is still modest on pastures, which have the crops most lacking in the use of inputs to increase their quality and sustainability. One example is Brazil, where over 100 million doses of inoculants have been commercialized annually, but less than 0.1% are destined for pastures, despite outstanding results confirming the benefits of inoculation, as shown in Table 1. Therefore, efforts towards increasing the use of PGPM in pastures with grasses can have profound positive global, environmental, economic, and social impacts.

**Shoot Biomass Root Biomass** Microorganism Nitrogen **Forage Species** Strain Increase Reference Increase (kg ha<sup>-1</sup>) **Genus/Species** (g) (g) Urochloa Azospirillum brizantha cv. Ab-V5 and Ab-V6 40 24.66 [26] brasilense Marandu Urochloa Azospirillum brizantha cv. Ab-V5 and Ab-V6 50 13.04 [207] brasilense Marandu Urochloa brizantha Azospirillum Ab-V5 and Ab-V6 25 81.1 [95] cv. Marandu brasilense Urochloa brizantha cv. Bacillus subtilis AP-3 27.41 13.59 [208] Marandu Urochloa brizantha Acaulospora 353 [209] cv. Marandu muricata Urochloa brizantha Azospirillum Ab-V5 and Ab-V6 50 [94] 41.25 \_ cv. Paiaguás brasilense Azospirillum Urochloa ruzizienses Ab-V5 and Ab-V6 40 23.26 [26] brasilense Pseudomonas 0 Urochloa ruziziensis **CNPSo 2719** 60 [96] 43 fluorescens Pseudomonas 0 Urochloa ruziziensis AMG521 44 70 [96] ananatis Azospirillum Urochloa ruziziensis Ab-V5 and Ab-V6 80 29.3 \_ [27] brasilense Pseudomonas Urochloa ruziziensis CNPSo 2719 80 27.6 [27] \_ fluorescens Azospirillum Urochloa brizantha Ab-V5 and Ab-V6 60 66.5 [27] brasilense Pseudomonas Urochloa brizantha CNPSo 2719 60 97.5 [27] fluorescens Claroideoglomus etunicatum and Urochloa decumbens 31 [210] Acaulospora morrowiae Megathyrsus Azospirillum Ab-V5 and Ab-V6 50 17.02 17 [211] maximus cv. Zuri brasilense Megathyrsus Pseudomonas maximus cv. BRS CNPSo 2719 100 7 [212] fluorescens Zuri Megathyrsus 50 maximus cv. Mesorhizobium sp. SEMIA 816 12,5 [144]Tanzânia Paspalum saurae SEMIA 816 50 11.7 [144] Mesorhizobium sp. \_ Acaulospora Andropogon gayanus \_ 107 \_ [213] cv. Planaltina muricata

**Table 1.** Recent studies reporting the benefits of inoculation with plant growth-promoting microor-<br/>ganisms in forage grasses in Brazil.

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