

**Original Research Article**  
**Combining fully acidulated phosphates with  
phosphate-solubilizing microorganisms for  
*Urochloa brizantha* fertilization**

**ABSTRACT**

This study aimed to evaluate the productive, morphological and nutritional characteristics, in addition to the accumulation of nutrients in the aerial part of *Urochloa brizantha* cv. Marandu subjected to phosphorus chemical forms and its interaction with phosphate-solubilizing inoculant. The experiment was developed in Colorado do Oeste, RO, Brazil, being adopted a randomized block design with three replications. The treatments consisted of different phosphate fertilizer doses associated or not with phosphate-solubilizing microorganisms (PSM): 1) 0% of P; 2) 50% of P; 3) 100% of P; 4) 50% of P + 250 mL ha<sup>-1</sup> of PSM; 5) 50% P + 500 mL ha<sup>-1</sup> of PSM; 6) 50% P + 750 mL ha<sup>-1</sup> of PSM; and 7) 50% P + 1000 mL ha<sup>-1</sup> of PSM. Triple superphosphate was used as a chemical fertilizer, and fertilization with chemical fertilizer and PSM occurred on November, 19th, 2021. It was evaluated yield and morphological characteristics, forage chemical analysis and nutrients accumulation in the aerial part of Marandu grass. The obtained data were subjected to variance analysis by the F test and, when significant, the means were compared by the Tukey test on 5% of probability. For the morphological characteristics and the yields of Marandu grass the results showed that there were significant differences. The different arrangements and phosphate doses influenced the green matter yield and dry matter yield, both using the 50% P + 1000 mL ha<sup>-1</sup> of PSM presented higher yields (3,642.75 kg ha<sup>-1</sup> and 1,049.10 kg ha<sup>-1</sup>, respectively). Higher results in crop growth rate of green matter were also found when employing 50% P + 500 mL ha<sup>-1</sup> of PSM and 50% P + 1000 mL ha<sup>-1</sup> of PSM. Dry matter presented a significant effect on bromatological composition, showing higher results using 50% of P by itself (34.55%). Regarding accumulation of nutrients in the aerial part, the highest P accumulation in the aerial part of Marandu grass was obtained when utilizing 100% of P, accumulating 15.4 kg of P ha<sup>-1</sup>. Inoculation with phosphate-solubilizing microorganisms for fertilization of *U. brizantha* cv. Marandu grass does not show adverse effects regarding the increase in P availability to the plant, resulting in adequate yield and crop development associated with the area's productive potential.

*Keywords: Fertilizer doses; Marandu grass; nutrient transport; phosphorus; soil fertility.*

**1. INTRODUCTION**

In the global scenario, Brazil has showed as the second largest cattle rancher and one of the seventh top dairy farmer [1], owning an approximate 222 million herd [2] and 33.8 billion liters in a national dairy farming, by an estimated 16.35 million cattle milked herd [3].

Coming to a stock context, grassing is the most important feed income for a cattle herd, presenting a low cost compared to feeding concentrate [4]. In addition, considering feeding

cattle in tropical regions is based in some grass species monocultures, it is understood that this practice demands a whole dependency on external inputs [5], even though it shows low efficiency.

The *Urochloa* gender is one of the most grown grasses in Brazil, standing out the *U. brizantha* [6,7], which presents the fastest and intense growth intake [8] and high adapting capability to different soil and climate conditions [9,10]. Besides, considering Brazilian livestock still has low productivity and income, as a result of an unappropriated pasture management [11], *Urochloa* grass has a fundamental importance of a right path to manage this system, as well as knowing the species nutritional needs in this gender [12].

Thereby, the forage plants productivity, in front of a model mostly featured by extensive nutrient exploitation with the lack of replacement of those, needs fertilizing and soil practices aiming a better nutritional use by the plants. In face of this, phosphorus (P) shows as an essential element, since it plays a key role on the plant life cycle, being present in metabolic processes related to photosynthetic processes, cell division and energy transference [13,14].

From this perspective, P performs as a limit factor on vegetal growth, predominantly on tropical regions, demanding a huge P amount, because its high stability form, consequently low solubility [15]. It is estimated that 70% of P used by chemical fertilizers or organic matter still remains in the soil in a few accessible ways for the plants [16]. Thus, there is a need for high phosphorus fertilizers doses with the purpose of reaching the plant demand and obtaining high income [17].

Therefore, inoculating with phosphate-solubilizing microorganisms (PSM) represents an alternative to different crops, with expressive increment on yields [18,19]. Those microorganisms are responsible to help, in a straightforward way, the plants obtain P from the soil, through promoting lateral root growth, unbalancing absorption, increasing roots superficial area throughout the whole root system along with metabolic process impulses [20,21].

It is extremely important, therefore, considering the role of PSM related to pasture growth, not just to comprehend better the mechanisms that involve the soil-plant system, as well to amplify the possibilities of the studies of this nature around soluble phosphate fertilizers frequently used.

Thus, this study aimed to evaluate the productive, morphological and nutritional characteristics, in addition to the accumulation of nutrients in the aerial part of *U. brizantha* cv. Marandu subjected to phosphorus chemical forms and its interaction with phosphate-solubilizing inoculant.

## 2. MATERIAL AND METHODS

### 2.1 Experimental Site

The experiment was carried out in a rural property in Colorado do Oeste, RO, Brazil (13° 06' 22.2" S and 60° 31' 43.4" W). This property has an area of 2.39 ha, on which 0.064 ha were used to carry out the experiment.

Soil samples showed on a 0 to 20 cm depth the following chemical characteristics: pH  $H_2O = 5.2$ ;  $P = 1.3 \text{ mg dm}^{-3}$ ;  $K = 0.07 \text{ cmol}_c \text{ dm}^{-3}$ ;  $OM = 15\%$ ;  $Al = 0.2 \text{ cmol}_c \text{ dm}^{-3}$ ;  $Ca = 1.6 \text{ cmol}_c \text{ dm}^{-3}$ ;  $Mg = 0.3 \text{ cmol}_c \text{ dm}^{-3}$ ; cation exchange capacity =  $5.4 \text{ cmol}_c \text{ dm}^{-3}$ ; and base saturation = 36%.

Classified as tropical monsoon (Am) according to Köpper-Geiger [22], the region's climate has two well-defined seasons. The average annual temperature in this region is between 24 °C and 26 °C, with maximum on its 30 °C to 35 °C, and minimum on 16 °C to 24 °C. The average annual rainfall rate varies from 1,400 to 2,600  $\text{mm year}^{-1}$  [23].

### 2.2 Experimental Design and Management

The experimental design used was a randomized block with 7 treatments and 3 replications, totaling 21 experimental units. The treatments consisted of different phosphate fertilizer doses associated or not with PSM: 1) 0% of P; 2) 50% of P; 3) 100% of P; 4) 50% of

P + 250 mL ha<sup>-1</sup> of PSM; 5) 50% P + 500 mL ha<sup>-1</sup> of PSM; 6) 50% P + 750 mL ha<sup>-1</sup> of PSM; and 7) 50% P + 1000 mL ha<sup>-1</sup> of PSM.

Triple superphosphate was used as a chemical fertilizer, with 46% P<sub>2</sub>O<sub>5</sub> and 36% of phosphorus soluble, supplied by hand on each experimental unit, and the PMS inoculant was added using a knapsack sprayer previously calibrated to introduce the established dose to each treatment on November, 19th, 2021.

*U. brizantha* cv. Marandu was used as the pasture, on which has been installed in the farm for 15 years without phosphate fertilization in this season, consequently the fertilization process for this experiment has characterized as the first one, as well as the supply of PSM. The area of 0.064 ha was previously isolated with a fence, in order to prevent access to the animals present on the property. In this area were installed 21 experimental units with 9.0 m<sup>2</sup> each, spaced 1.0 m among them.

### **2.3 Marandu grass yield and morphological characteristics evaluation**

The forage was harvested at 30, 60, 90 and 150 days after treatment implementation, where grass height was evaluated with a measuring tape attached to a wooden support surface, obtaining measures from two different random spots in each plot. Furthermore, green matter yield was evaluated using a 1.0 m<sup>2</sup> sampling frame and a digital electronic scale. After that, with a petrol grass trimmer, all plots were standard to 0.15 m height, seeking to preserve apical bud and, consequently, in order to favor the adequate reestablishment of the plant.

For statistical analysis, we considered the average height and yield of the Marandu grass harvesting seasons for each treatment. For crop growth rate (CGR) determination it was considered the total matter input related to the last harvesting season according to Rajput A et al. [24].

Only at 60 days (second harvesting season) were the morphological components collected. For this purpose, grass was harvested from the areas of the sampling frame (1.0 m<sup>2</sup>) and proceeded with the morphological separation (leaf, stem and dead material), weighing the portions on precision scales and later converting the values to determine the proportions of each component.

### **2.4 Forage chemical analysis**

At the time of the second grass harvesting season (at 60 days), forage samples were taken and wrapped in paper bags for chemical analysis. The forage samples pre-dry were milled in a Willey mill with a 1.0 mm screen to provide dry matter (DM), crude protein (CP), neutral detergent fiber (NDF) and acid detergent fiber (ADF) contents according to Silva JD and Queiroz AC [25].

### **2.5 Nutrients accumulation in the aerial part of Marandu grass**

The concentrations of phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), sulfur (S), boron (B), copper (Cu), iron (Fe), manganese (Mn) and zinc (Zn) were determined, using the methodology recommended by Claessen MEC et al. [26]. With the dry mass and nutrient concentration, we calculated the accumulation of nutrients in the aboveground part of Marandu grass, in kg ha<sup>-1</sup> for macronutrients and in g ha<sup>-1</sup> for micronutrients.

### **2.6 Statistical Analysis**

The obtained data were subjected to analysis of variance by the F test and, when significant, the means were compared by the Tukey test on 5% of probability, on an Experimental Designs package of R Software, version 3.5.3 [27].

### 3. RESULTS

#### 3.1 Yield and morphological characteristics of Marandu grass

For the morphological characteristics and the yields of Marandu grass subjected to the chemical sources of phosphorus associated with PSM, the results showed that there were significant differences ( $P = .05$ ) for the variables green and dry matter yield, crop growth rate of green matter, height, leaf proportion, stem proportion, and dry matter content (Table 1).

**Table 1. Summary of the analysis of variance for morphological characteristics, yields and dry matter content of Marandu grass (*Urochloa brizantha*) subjected to increasing doses of phosphate fertilization and inoculation with phosphate-solubilizing microorganisms.**

Variable	Effect				
	Mean	Treat.	Block	CV (%)	P (value)
GMY (kg ha <sup>-1</sup> )	3,290.35	*	ns	7.69	0.0165
DMY (kg ha <sup>-1</sup> )	958.33	*	ns	7.71	0.039
GMCGR (kg GM day <sup>-1</sup> )	93.81	*	ns	8.52	0.0212
DMCGR (kg DM day <sup>-1</sup> )	27.48	ns	ns	9.10	0.0663
Height (cm)	28.45	*	ns	4.61	0.0061
Leaf (%)	85.94	*	ns	2.24	0.008
Stem (%)	12.58	*	ns	14.43	0.0047
DMP (%)	1.07	*	ns	24.20	0.0086
LSR	7.36	*	ns	20.88	0.0146

GMP: green matter yield. DMP: dry matter yield. GMCGR: green matter crop growth rate. DMCGR: dry matter crop growth rate. GM: green matter. DM: dry matter. DMP: dead material proportion. LSR: leaf/stem ratio. \*Significant at 5% probability of error by the F test. ns: not significant. CV: coefficient of variation.

As presented in Table 2, the different arrangements and phosphate doses (chemical and PSM) influenced the green matter yield ( $P = .0165$ ) and dry matter yield ( $P = .039$ ). On both, using the 50% phosphorus dose recommended associated with 1000 mL ha<sup>-1</sup> of PSM presented higher yields than the other treatments.

In addition, the crop growth rate of green matter was also influenced ( $P = .0212$ ) by using chemical phosphorus sources and the PSM, and when using 50% of the recommended phosphorus dose associated with 500 mL ha<sup>-1</sup> of PSM and 50% of the recommended phosphorus dose associated with 1000 mL ha<sup>-1</sup> of PSM, a higher growth rate was obtained. The lowest growth rate was obtained when operating with 50% of the recommended phosphorus dose associated with 750 mL ha<sup>-1</sup> of PSM.

**Table 2. Green and dry matter yield and crop growth rate of Marandu grass (*Urochloa brizantha*) subjected to increasing doses of phosphate fertilization and inoculation with phosphate-solubilizing microorganisms.**

Treatment	Yield (kg ha <sup>-1</sup> )	CGR (kg GM day <sup>-1</sup> )
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	GM	DM	
Control	3,003.08 AB	887.37 AB	84.62 AB
50% P	3,153.83 AB	950.36 AB	90.12 AB
100% P	3,492.75 AB	1,020.89 AB	98.81 AB
50% P + 250 mL ha <sup>-1</sup> PSM	3,380.83 AB	990.86 AB	96.56 AB
50% P + 500 mL ha <sup>-1</sup> PSM	3,525.75 AB	980.52 AB	102.89 A
50% P + 750 mL ha <sup>-1</sup> PSM	2,833.42 B	829.22 B	80.01 B
50% P + 1000 mL ha <sup>-1</sup> PSM	3,642.75 A	1,049.10 A	103.63 A
CV (%)	7.69	7.71	8.52

50% P: 50% of the recommended phosphorus dose. 100% P: 100% of the recommended phosphorus dose. 50% P + 250 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 250 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. 50% P + 500 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 500 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. 50% P + 750 mL ha<sup>-1</sup> PSM: 50% recommended phosphorus dose + 750 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. 50% P + 1000 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 1000 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. CGR: crop growth rate. CV: coefficient of variation. GM: green matter. DM: dry matter. Means followed by the same letter in the column do not differ by Tukey test (P = .05).

In the morphological characteristics of Marandu grass subjected to the chemical sources of phosphorus associated or not with PSM, there was a significant effect on the proportions of leaf (P = .008), stem (P = .0047) and dead material (P = .0086), leaf/stem ratio (P = .0146) and plant height (P = .0061) (Table 3).

**Table 3. Morphological characteristics of Marandu grass (*Urochloa brizantha*) subjected to increasing doses of phosphate fertilization and inoculation with phosphate-solubilizing microorganisms.**

Treatment	Height (cm)	Leaf (%)	Stem (%)	DMP (%)	LSR
Control	27.33 AB	90.15 A	9.10 C	0.43 B	10.52 A
50% P	30.00 A	86.89 AB	11.81 ABC	1.89 A	7.43 AB
100% P	25.55 B	85.36 AB	13.24 ABC	0.83 AB	6.47 AB
50% P + 250 mL ha <sup>-1</sup> PSM	27.70 AB	82.98 B	15.42 AB	1.56 A	5.52 B
50% P + 500 mL ha <sup>-1</sup> PSM	30.43 A	83.13 B	16.17 A	1.02 AB	5.22 B
50% P + 750 mL ha <sup>-1</sup> PSM	28.22 AB	85.59 AB	10.76 BC	0.47 B	8.77 AB
50% P + 1000 mL ha <sup>-1</sup> PSM	29.93 A	87.44 AB	11.57 ABC	1.31 AB	7.61 AB
CV (%)	4.61	2.24	14.43	24.20	20.88

50% P: 50% of the recommended phosphorus dose. 100% P: 100% of the recommended phosphorus dose. 50% P + 250 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 250 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. 50% P + 500 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 500 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. 50% P + 750 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 750 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. 50% P + 1000 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 1000 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. DMP: dead material proportion. LCR: leaf/stem ratio. CV: coefficient of variation. Means followed by the same letter in the column do not differ by the Tukey test (P = .05).

The leaf proportion and leaf/stem ratio demonstrated superiority among the other treatments when it was not being subjected to phosphate fertilizer. On the other hand, making use of the 50% phosphorus dose recommended associated with 250 mL ha<sup>-1</sup> PSM

and 50% of the phosphorus dose recommended associated with 500 mL ha<sup>-1</sup> PSM were the lowest on both variables.

In addition, the stem proportion showed higher mean when subjected to the 50% phosphorus dose recommended associated with 500 mL ha<sup>-1</sup> PSM. The non-fertilization with phosphate provided a lower average for this variable. As for the dead material proportion, higher results were found employing only 50% phosphorus dose recommended and 50% phosphorus dose recommended with 250 mL ha<sup>-1</sup> of PSM. The inverse materialized when there was no phosphate fertilization and also when 50% phosphorus dose recommended associated to 750 mL ha<sup>-1</sup> PSM were supplied.

### 3.2 Bromatological composition of Marandu grass forage

There was a significant effect of chemical phosphorus sources associated with PSM only for dry matter content (P = .0048) (Table 4).

**Table 4. Summary of the analysis of variance for the bromatological composition of Marandu grass (*Urochloa brizantha*) subjected to increasing doses of phosphate fertilization and inoculation with phosphate-solubilizing microorganisms.**

Variable	Mean	Effect		
		Treat.	Blocks	P (value)
DM (%)	31.97	*	ns	0.0048
CP (%)	8.86	ns	ns	>0.05
NDF (%)	62.97	ns	ns	>0.05
ADF (%)	29.84	ns	ns	>0.05

DM: dry matter. CP: crude protein. NDF: neutral detergent fiber. ADF: acid detergent fiber. \*Significant at 5% probability of error by the F test. ns: not significant.

The higher dry matter content obtained using only the 50% phosphorus dose recommended by itself. Conversely, the use of 100% of the recommended phosphorus dose, 50% of the recommended phosphorus dose with 500 mL ha<sup>-1</sup> of PSM, 50% of the recommended phosphorus dose with 750 mL ha<sup>-1</sup> of PSM, and 50% of the recommended phosphorus dose with 1000 mL ha<sup>-1</sup> of PSM provided lower dry matter contents (Table 5).

**Table 5. Dry matter content of Marandu grass (*Urochloa brizantha*) subjected to increasing doses of phosphate fertilization and inoculation with phosphate-solubilizing microorganisms.**

Treatment	DM (%)
Control	32.74 AB
50% P	34.55 A
100% P	30.53 B
50% P + 250 mL ha <sup>-1</sup> PSM	32.40 AB
50% P + 500 mL ha <sup>-1</sup> PSM	30.92 B
50% P + 750 mL ha <sup>-1</sup> PSM	30.32 B
50% P + 1000 mL ha <sup>-1</sup> PSM	32.31 B
CV (%)	3.36

50% P: 50% of the recommended phosphorus dose. 100% P: 100% of the recommended phosphorus dose. 50% P + 250 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 250 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. 50% P + 500 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus

dose + 500 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. 50% P + 750 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 750 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. 50% P + 1000 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 1000 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. DM: dry matter. CV: coefficient of variation. Means followed by the same letter in the column do not differ by the Tukey test (P = .05).

### 3.3 Accumulation of nutrients in the aerial part of Marandu grass

There was a significant effect of chemical sources of phosphorus associated with PSM on all macronutrients and micronutrients (Table 6).

**Table 6. Summary of the analysis of variance for the accumulation of nutrients in the aerial part of Marandu grass (*Urochloa brizantha*) subjected to increasing doses of phosphate fertilization and inoculation with phosphate-solubilizing microorganisms.**

Variable	Mean	Effect		
		Treat.	Blocks	P (value)
P (kg ha <sup>-1</sup> )	11.30	*	ns	<0.00001
K (kg ha <sup>-1</sup> )	96.10	*	ns	0.0004
Ca (kg ha <sup>-1</sup> )	11.90	*	ns	0.000058
Mg (kg ha <sup>-1</sup> )	8.40	*	ns	0.0004
S (kg ha <sup>-1</sup> )	4.20	*	ns	<0.00001
B (g ha <sup>-1</sup> )	30.30	*	ns	<0.0001
Cu (g ha <sup>-1</sup> )	16.70	*	ns	<0.00001
Fe (g ha <sup>-1</sup> )	386.30	*	ns	<0.00001
Mn (g ha <sup>-1</sup> )	557.90	*	ns	0.0121
Zn (g ha <sup>-1</sup> )	96.10	*	ns	0.021

P: phosphorus. K: potassium. Ca: calcium. Mg: magnesium. S: sulfur. B: boron. Cu: copper. Fe: iron. Mn: manganese. Zn: zinc. \*Significant at 5% probability of error by the F test. ns: not significant.

Important evidence regarding the P accumulation, since this nutrient was subject to variation as it was part of the treatments in this study. The highest P accumulation in the aerial part of Marandu grass (Table 7) was obtained when utilizing 100% of the recommended phosphorus dose. Only the control treatment showed low P accumulation, justified by the fact that there was no supply of phosphorus.

The other treatments, when there was an association of phosphorus sources and different doses of PSM, showed intermediate values of P accumulation.

**Table 7. Accumulation of nutrients in the aerial part of Marandu grass (*Urochloa brizantha*) subjected to increasing doses of phosphate fertilizers and inoculation with phosphate-solubilizing microorganisms.**

	Treatment						
	Control	50% P	100% P	50% P + 250 mL ha <sup>-1</sup> PSM	50% P + 500 mL ha <sup>-1</sup> PSM	50% P + 750 mL ha <sup>-1</sup> PSM	50%P + 1000 mL ha <sup>-1</sup> PSM
P (kg ha <sup>-1</sup> )	7.1 D	10.4 C	15.4 A	11.00 C	12.1 B	10.2 C	12.6 B
K (kg ha <sup>-1</sup> )	83.3 B	87.0 B	106.1 A	90.20 B	107.3 A	81.3 B	117.5 A
Ca (kg ha <sup>-1</sup> )	9.8 B	12.1 B	13.2 A	11.80 B	11.7 B	10.9 B	13.5 A
Mg (kg ha <sup>-1</sup> )	6.7 C	8.3 B	9.7 A	8.50 B	8.4 B	7.0 C	9.8 A
S (kg ha <sup>-1</sup> )	3.4 B	3.1 B	5.3 A	2.9 B	5.2 A	3.8 B	5.6 A
B (g ha <sup>-1</sup> )	30.6 B	27.7 B	35.2 A	29.4 B	24.1 B	28.0 B	37.3 A
Cu (g ha <sup>-1</sup> )	16.9 A	13.9 B	17.6 A	14.7 B	16.1 B	14.0 B	23.3 A
Fe (g ha <sup>-1</sup> )	421.4 B	447.1 B	48.4 D	353.4 C	603.0 A	364.4 C	466.3 B
Mn (g ha <sup>-1</sup> )	570.9 A	516.5 B	638.1 A	541.2 B	558.8 A	469.5 B	610.8 A
Zn (g ha <sup>-1</sup> )	81.6 B	90.1 B	114.4 A	99.4 A	100.5 A	84.1 B	102.6 A

50% P: 50% of the recommended phosphorus dose. 100% P: 100% of the recommended phosphorus dose. 50% P + 250 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 250 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. 50% P + 500 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 500 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. 50% P + 750 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 750 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. 50% P + 1000 mL ha<sup>-1</sup> PSM: 50% of the recommended phosphorus dose + 1000 mL ha<sup>-1</sup> of phosphate-solubilizing inoculant. P: phosphorus. K: potassium. Ca: calcium. Mg: magnesium. S: sulfur. B: boron. Cu: copper. Fe: iron. Mn: manganese. Zn: zinc. Means followed by the same letter in the column do not differ by the Tukey test (P = .05).

Regarding the K and S accumulation in the aerial part of Marandu grass, greater accumulations were observed when 100% of the recommended phosphorus dose, 50% of the recommended phosphorus dose with 500 mL ha<sup>-1</sup> of PSM and 50% of the recommended dose of phosphorus with 1000 mL ha<sup>-1</sup> of PSM.

For the Ca, Mg, B and Cu accumulation in the aerial part of Marandu grass subjected to the use of chemical sources of phosphorus and MSP, superiority was verified when 100% of the recommended phosphorus dose was provided and also for the supply of 50% recommended dose of phosphorus with 1000 mL ha<sup>-1</sup> of PSM.

Regarding the Fe accumulation, statistical superiority was observed only when there was the supply of 50% of the recommended phosphorus dose associated with 500 mL ha<sup>-1</sup> of PSM. In contrast, concerning Mn accumulation, greater accumulation was observed with the supply of 100% of the recommended phosphorus dose, 50% of the recommended phosphorus dose with 500 mL ha<sup>-1</sup> of PSM and 50% of the recommended phosphorus dose with 1000 mL ha<sup>-1</sup> of PSM.

Finally, when considering the Zn accumulation, the treatments that showed the highest accumulations were the supply of 100% of the recommended phosphorus dose, 50% of the recommended phosphorus dose with 250 mL ha<sup>-1</sup> of PSM, 50% of the recommended phosphorus dose with 500 mL ha<sup>-1</sup> of PSM, and 50% of the recommended phosphorus dose with 1000 mL ha<sup>-1</sup> of PSM.

## 4. DISCUSSION

### 4.1 Yield and morphological characteristics of Marandu grass

From the evaluation of the productive and morphological characteristics of Marandu grass subjected to chemical sources of phosphorus and the interaction with phosphate-solubilizing inoculant, this study reveals that the aspects of production and plant configuration in its form, is closely related to the maintenance of nutrients supplied to it. Such finding reinforces the benefits of using inoculants for forage grasses, exploiting microorganisms already used in commercial inoculants [28,29,30].

When evaluating the yield of Marandu grass subjected to increasing doses of phosphorus, Porto EMV et al. [31] also found increases in dry matter production when increasing the doses of P, reaffirming the importance of this element in forage yield in soils under pasture, naturally deficient in phosphorus [32,33].

Though majority forage grasses, including *Urochloa* gender, present typical morphological characteristics to an immense soil volume exploitation, studies have demonstrated the importance of using microorganisms to optimize the phosphorus supply to the plant [34,35,36], directly contributing to establish, growth and regrowth of those plants in low fertility soils.

A study related to chemical fertilizing doses combined with co-inoculated microorganisms to promote growth and nutrients efficiency in Marandu grass, showed inoculating with microorganisms along to 50% of P, increased grass growth [37], as evidenced in the present study. This could be an important resolution to a problem Brazilian livestock has been facing relating to low productivity and profitability when related to inappropriate managing pasture systems [11].

Highlighting the possibility of using pasture recovery methods in this context, Souza R et al. [38] report growing forage grasses require improvement on soil fertility to retain adequate pasture growth. That allied to Marandu grass morphological characteristics, it is strengthened by the higher proportions of plant parts when using chemical phosphorus sources associated with PSM, except leaf portion, which could relate with requirement limitations on other nutrients, as nitrogen, since this nutrient promote new cell generation, allowing leaf elongation rate [39].

#### **4.2 Bromatological composition of Marandu grass forage**

In relation to Marandu grass subjected to the use of chemical sources of phosphorus and its interaction with the phosphate solubilizer inoculant, it was found that only the dry matter content showed statistical difference among the treatments, about chemical analysis. This consolidates the importance of composition chemical analysis, being an important factor in evaluating forage grasses quantifying process [38], since its determination is fundamental to diet formulation, evaluation of animal intake and performance.

It is worth emphasizing that feed nutritional balance is done based on organic and chemical nutrients [40] and both are contained in the dry matter [41]. Thus, such a result supports the found differences as nutrients intake on Marandu grass aerial part, even though it has not happened differences among the treatments in the relation to the bromatological composition of the evaluated material.

Although CP, NDF and ADF contents did not show significant differences among the treatments, those variables presented adequate contents on which refers to the grass bromatological composition, according to the Brazilian Cattle Feed Composition Table [7], with 8.86%, 62.97% and 29.84%, respectively.

The mean CP content was higher than 7.0% (desirable minimum for the proper functioning of the rumen microbiota). Sampaio FAR et al. [43] mention that CP content below 7.0% in the dry matter of tropical grasses leads to reduced digestibility due to inadequate levels of some nutrients in the plant.

The mean NDF content is in agreement with the values found by Guimarães AKV et al. [44] who reported that Marandu grass dry matter under phosphate fertilization at 56 days had 63.72% of NDF content. According to Van Soest PJ [45], values higher than 60% of NDF in dry matter limit forage intake. The mean content found is closely connected to those established by this author (62.97%).

The ADF mean content in this study has been lower than which was found by Magalhães AF et al. [46] working on crescent phosphorus doses in *U. decumbens*. The average content of ADF in Marandu grass was adequate (29.64%) because according to Nussio LG et al. [47], forages with ADF contents around 40%, or above, showed lower intake and digestibility.

### 4.3 Accumulation of nutrients in the aerial part of Marandu grass

When evaluating the accumulation of nutrients in the aerial part of Marandu grass subjected to the use of chemical sources of phosphorus with an association of a phosphate-solubilizing inoculant, the results found in this study were fundamental.

The macro and micronutrient accumulations in the leaves of Marandu grass varied according to the treatments, making it possible to confirm that there were changes in the nutritional state of the plant. Such effect was also found in a study by Andrade RA et al. [48] who observed adequate macronutrient accumulation in the leaves of Tamani grass (*Megathyrsus maximus*), fertilized with natural phosphate and inoculated by *Azospirillum brasilense*.

This effect, independent of the specie studied, can be associated to biochemical changes and phytohormones produced by the plant, superficial root part increasing, which maximize the nutrient extraction from the soil, nitrogen fixation, in addition to poorly solute compounds solubilization, with better phosphorus use implemented through fertilization, or its inorganic form in the soil [49,50,51].

Forage grasses exhibit low cation exchange capacity in the root system, and soils adsorb elements with higher valence, such as calcium and magnesium, more effectively. Therefore, forage plants that are more efficient in extracting the uptake of monovalent cations, such as potassium, may limit the uptake of other nutrients, such as calcium [48].

This fact becomes evident when observing how the relations between the minerals present in the aerial part of Marandu grass occurred, being possible to verify that as the dose and the source of phosphorus for the plant increases, the other nutrients are affected in a positive or negative way. However, deficiency levels were not observed for any of the evaluated nutrients, since their contents are within the ranges considered adequate for forage plants of the genus *Urochloa* [52].

Andrade RA et al. [48] found that the interaction between phosphorus sources and rhizobacteria improved the efficiency of phosphate fertilization regardless of the source used, with increasing phosphorus, potassium, magnesium, and sulfur contents in leaf tissues, and to influence in a positive way the plant growth parameters. According to Somavilla A et al. [53], reactive natural phosphates require more time to become available in the soil, but they have a positive residual effect, especially for perennial crops such as forage plants.

Therefore, it is possible to understand why the commercialization of microbial inoculants has increased significantly over the past few years. However, their adherence is still discrete in pastures. On the other hand, through this research, it is possible to note that microorganisms can contribute in a positive way to the soil-plant environment in pasture animal production systems. It also promotes progress in solving the problem of low productivity and financial performance, mainly due to the inappropriate management of pasture systems that Brazilian cattle raising still faces.

Providing 100% phosphorus recommended dose isolated or 50% phosphorus dose recommended associated to 500 mL ha<sup>-1</sup> phosphate-solubilizing microorganisms, or even, 50% phosphorus dose recommended associated to 1000 mL ha<sup>-1</sup> phosphate-solubilizing microorganisms, appears to be linked to greater and balanced availability of other nutrients to the plant through some synergistic interaction, which can result in improving Marandu grass growth and development.

Considering the positive plant responses regarding the use of phosphate-solubilizing microorganisms as a partial replacement for chemical P sources, it becomes important to conduct studies on the economic feasibility and assessment of environmental impacts

through this practice. This will provide ranchers with a comprehensive understanding to proceed on making decisions.

It is also recommended that further studies be conducted to analyze the response of other tropical forage grass species associated with the identification and characterization of specific microorganisms involved in phosphorus solubilization in tropical forage grasses, in addition to the definition of optimal conditions for the activity of the inoculants.

## 5. CONCLUSION

Inoculation with phosphate-solubilizing microorganisms for fertilization of *Urochloa brizantha* cv. Marandu grass does not show adverse effects regarding the increase in P availability to the plant, resulting in adequate yield and crop development associated with the area's productive potential.

The use of 50% phosphorus dose recommended associated with 500 mL ha<sup>-1</sup> phosphate-solubilizing microorganisms and 50% phosphorus dose recommended associated with 1000 mL ha<sup>-1</sup> phosphate-solubilizing microorganisms, appears to be linked with higher yields and higher rates of expansion of Marandu grass.

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