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Physiological and Sanitary Characterization of Seed Lots of *Urochloa brizantha* cv. Marandú Marketed in Campo Grande, MS

Caracterização Fisiológica e Sanitária de Lotes de Sementes de Urochloa brizantha cv. Marandú Comercializadas em Campo Grande, MS

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Abstract

In recent years, Brazil has distinguished itself as the world leader in the production, consumption, and export of forage seeds. This position in the agricultural landscape has been consolidated through the use of *Urochloa brizantha* cv. Marandú seeds. Thus, the aim of this study was to characterize the sanitary and physiological quality of seed lot samples from the 2018-2019 crop of *U. brizantha*, cultivar Marandú, marketed by four forage seed companies in the city of Campo Grande, MS, obtained in 2020 for analysis and stored in paper bags under average environmental conditions of 21.9°C (temperature) and 68% (relative humidity), respectively. To evaluate the sanitary quality of the seeds, the Blotter Test was performed. Physiological evaluation included a germination test alongside biometric tests, such as seedling length, seedling dry mass, emergence in sand, length of emerged seedlings in sand, dry mass of emerged seedlings in sand, and weight of one thousand seeds. The incidence of various fungi was verified, with 89.2% in samples from company A, 89.7% from company B, 98% from company C, and 86.7% from company D; germination values ranged from 53.2% to 91.2%, and for emergence in sand, all values obtained were below 60% in the *U.*

brizantha cv. Marandú seeds analyzed. The results revealed low sanitary and physiological quality in all batches, regardless of the company, during the period when the tests were conducted.

Keywords: Forage Seeds. Fungi. Germination. Physiological Quality.

Resumo

Nos últimos anos, o Brasil tem se destacado como o líder mundial na produção, consumo e exportação de sementes forrageiras. Essa posição no cenário agrícola foi consolidada através do uso de sementes de *Urochloa brizantha* cv. Marandú. Deste modo, o objetivo deste estudo foi caracterizar a qualidade sanitária e fisiológica de amostras de lotes de sementes da safra 2018-2019 de *U. brizantha,* cultivar Marandú, comercializadas por quatro empresas de sementes forrageiras na cidade de Campo Grande, MS, obtidas em 2020 para realização das análises, neste ano, armazenadas em sacos de papel em condições ambientais de temperatura e umidade relativa do ar média de 21,9°C e 68%, respectivamente. Para avaliação da qualidade sanitária das sementes foi realizado o Blotter Test. Para avaliação fisiológica foi realizado teste de germinação, além de testes biométricos como comprimento de plântulas, massa seca de plântulas, emergência em areia, comprimento de plântulas emergidas em areia, massa seca de plântulas emergidas em areia e peso de mil sementes. Verificou-se a incidência de diversos fungos, sendo 89,2% nas amostras da empresa A, 89,7% da empresa B, 98% da empresa C e 86,7% da empresa D; para germinação, os valores variaram de 53,2% a 91,2% e para a emergência em areia, todos os valores obtidos foram abaixo de 60% nas sementes de *U. brizantha* cv. Marandú analisadas. Os resultados revelaram baixa qualidade sanitária e fisiológica, em todos os lotes, independente da empresa, para época de realização dos ensaios.

Palavras-chave: Sementes Forrageiras. Fungos. Germinação. Qualidade Fisiológica.

1 Introduction

The commercialization of forage seeds is an important economic component in the national livestock production chain. The tropical forage seed market moves around 440 million dollars annually and represents approximately 11% of the seed market in Brazil (ABIEC, 2025).

In the agricultural context, the use of seeds with high physiological and sanitary quality is essential for the success of crops, resulting in greater productivity, stand uniformity, seedling vigor, and lower incidence of seed-borne diseases (Castilhos *et al*., 2020; Chortaszko *et al*., 2019).

The presence of pathogens in forage seeds can compromise germination, vigor, yield, and even export potential, as seeds act as a primary source of inoculum for various phytopathogenic fungi and bacteria (Martins *et al*., 2017; Silva *et al*., 2019a). Thus, the identity and quality standards established by state and federal regulations require that seeds present satisfactory results in germination, tetrazolium, and physical purity tests for approval and commercialization (Brasil, 2021; Oliveira *et al*., 2021).

The adaptability of *Urochloa brizantha* cv. Marandú has enabled livestock production in various soil types, including acidic and low-fertility soils that predominate in the Cerrado biome (Carvalho *et al*., 2021 ; Delevatti *et al*., 2019). This fact explains the great interest of livestock producers in this cultivar, as it exhibits high dry matter yield, few disease problems, and good growth during most of the year, including the dry season (Ongratto *et al*., 2021).

Therefore, the objective of this study was to characterize the sanitary and physiological quality of seed lots of *U. brizantha* cv. Marandú (2018–2019 harvest) commercialized by four forage seed companies in the city of Campo Grande, Mato Grosso do Sul, in 2019.

2 Material and Methods

Seed lots of *Urochloa brizantha* cv. Marandú (2018–2019 harvest) were evaluated. The samples were obtained in 2019 from four randomly selected seed companies. The seeds were stored in paper bags under average conditions of 21 ± 2 °C and 68% relative humidity in Campo Grande, Mato Grosso do Sul, Brazil. After acquisition, the seed lots were stored under the same conditions, and the analyses were performed seven days after purchase.

To assess seed health quality, the Blotter Test was conducted as described by Neergaard (1979), using four subsamples of 100 seeds each. Procedures: the seeds were placed on a triple layer of moistened filter paper inside plastic germination boxes (Gerbox type). Incubation conditions: the boxes were maintained in an incubation chamber with a 12-hour photoperiod for seven days at 25 ± 2 °C.

Seed evaluations were carried out individually for each replication of every lot using a stereoscopic microscope. When necessary, semi-permanent slides were prepared using adhesive tape, glass slides, and Amann's lactophenol with cotton blue stain to identify reproductive and vegetative fungal structures under an optical microscope. The results were expressed as the percentage incidence of phytopathogenic fungal genera and/or species.

The thousand-seed weight was determined by measuring the mass of eight replicates of 100 pure seeds, weighed on an analytical balance. The results were calculated using the formula: Thousand-seed weight = (sample mass × 1,000) / total number of seeds (Brasil, 2009).

For the germination test, four subsamples of 100 seeds were sown on two sheets of moistened germitest paper placed in plastic Gerbox containers and incubated in a BOD (Biochemical Oxygen Demand) chamber under a 12-hour photoperiod at 25 ± 2 °C. Counts were made at seven, fourteen, and twenty-one days after sowing, considering seeds germinated when radicle protrusion occurred, according to the Rules for Seed Testing (Brasil, 2009).

Subsequently, at the final germination count (21 days after germination—DAG), the normal seedlings were evaluated for root and shoot length using a caliper, and the results were expressed in millimeters (mm).

The seedling dry mass was determined after these measurements. The material was placed in paper bags and dried in a forced-air oven at 80 ± 2 °C for 72 hours until a constant weight was reached (Nakagawa, 1999). All samples were weighed on an analytical balance, and the results were expressed in grams of dry mass.

The sand emergence test was carried out with 200 seeds in a completely randomized design, using four replicates of 50 seeds per company. The seeds were sown in plastic boxes filled with sterilized sand (autoclaved at 120 ± 2 °C for 72 hours) moistened with sterile distilled water and maintained in an incubation chamber with a 12-hour photoperiod at 25 ± 2 °C. After the first seedling emerged, daily counts were conducted for fifteen days.

At the end of this period (15th day), the length of normal seedlings (root and shoot) was measured using a caliper, with results expressed in millimeters (mm). The dry mass of these seedlings was determined after drying the material in a forced-air oven at 80 ± 2 °C for 72 hours (Nakagawa, 1999), until constant weight was achieved. The results were expressed in grams of dry mass for the lots from Companies A, B, C, and D.

The experimental design was completely randomized, with four treatments (Companies A, B, C, and D) and four replicates of 100 seeds each, totaling 400 seeds for the germination, seedling length, and seedling dry mass tests. For the sand emergence, seedling length, and dry mass tests, 200 seeds were used in four replicates of 50 seeds per company.

The mean results were tested for normality and then subjected to analysis of variance (ANOVA) and Tukey’s test for mean comparison at a 5% probability level. Statistical analyses were performed using the BioEstat 5.0 software.

3 Results and Discussion

In the sanitary analysis of naked seeds of *Urochloa brizantha* cv. Marandu, the incidence of ten fungal genera was verified among the lots from the four evaluated companies. The identified microorganisms were *Alternaria alternata*, *Aspergillus niger*, *Bipolaris maydis*, *Chaetomium* sp., *Curvularia* sp., *Fusarium oxysporum*, *Gerlachia* sp., *Penicillium* sp., *Phoma* sp., and *Rhizopus stolonifer* (Table 1).

Table 1 - Incidence of phytopathogenic fungi in naked seed lots of *U. brizantha* cv. Marandu (2018–2019 harvest) commercialized in Campo Grande, MS – 2019

Fungi	Incidence (%)	Mean
	A	B
Alternaria alternata	5.5	2.2
Aspergillus niger	23.2	10.2
Bipolaris maydis	10.2	36.2
Chaetomium sp.	–	0.5
Curvularia sp.	11.0	4.2
Fusarium oxysporum	10.7	7.2
Gerlachia sp.	10.2	4.7
Penicillium sp.	11.5	5.5
Phoma sp.	–	10.0
Rhizopus stolonifer	6.7	8.7
Total incidence*	**89.2**	**89.7**

(A) Company A; (B) Company B; (C) Company C; (D) Company D.*Total incidence per company. **Mean cumulative incidence per fungal genus or species.
Source: research data.

Seed lots from Companies B and C showed the highest fungal frequencies (89.7% and 98%, respectively), with ten fungal genera detected in both cases. Seeds from Company A exhibited 89.2% contamination by eight genera, while those from Company D presented 86.7% contamination by nine genera (Table 1).

Among the identified fungi, *Bipolaris maydis* showed the highest mean incidence (22.1%), particularly in Company B lots (36.2%). Conversely, *Alternaria alternata* exhibited the lowest mean incidence (3.7%), ranging from 2.2% to 5.5% across seed lots.

Among the detected genera, *Aspergillus* sp. and *Penicillium* sp. are classified as storage fungi, whereas the others are considered field fungi. According to Alves *et al*. (2017), storage fungi may colonize seeds during processing or storage, without necessarily originating in the field. All evaluated companies showed the presence of these fungi, with higher incidence of *A. niger* in Company A (23.2%) and *Penicillium* sp. in Company C (13%).

Saprophytic or necrotrophic microorganisms such as *Fusarium* sp., *Helminthosporium* sp., *Phoma* sp., *Aspergillus* sp., *Chaetomium* sp., *Nigrospora* sp., *Alternaria* sp., *Phomopsis* sp., *Rhizoctonia solani*, *Pythium* sp., and *Cylindrocladium* sp. are commonly found in soil. These fungi exhibit rapid mycelial growth and intense sporulation, characteristics that facilitate their penetration into healthy seeds—especially those collected by sweeping—and allow contamination during transport, processing, and storage (Martins *et al*., 2017; Machado *et al*., 2019; Cruz *et al*., 2022).

In this study, *Fusarium* sp. (7.3%), *Phoma* sp. (13.7%), *Aspergillus* sp. (14.8%), *Chaetomium* sp. (6.7%), and *A. alternata* (3.7%) were observed in the evaluated seed lots. According to

Verzignassi *et al*. (2012), among soilborne fungi with high capacity to form resistance structures, *Fusarium* has been identified as one of the main agents associated with the decline and death of Marandu pastures in the Amazon region (Pará, Acre, Maranhão, and Rondônia), along with *Pyricularia grisea*. *Fusarium* can impair seed germination, reducing pasture establishment and productivity, and is often associated with delayed harvesting or deterioration due to moisture in the field (Silva *et al*., 2019a; Silva *et al*., 2019b; Castilhos *et al*., 2020).

For physiological quality assessment, the germination test was used as the main parameter to estimate the physiological potential and vigor of seeds from the different companies (Table 2). This test represents the maximum germinative performance of a sample under ideal and controlled conditions, generally provided artificially for each species (Nakagawa, 1999; Marcos Filho, 2015; Chortaszko *et al*., 2019; Oliveira *et al*., 2021). The percentages obtained were 53.5% (Company A), 91.2% (Company B), 80.5% (Company C), and 53.2% (Company D).

Table 2 - Germination (G), seedling length (SL), seedling dry mass (SDM), sand emergence (SE), seedling length in sand (SLS), seedling dry mass in sand (SDMS), and thousand-seed weight (TSW) in naked seed lots of *U. brizantha* cv. Marandu (2018–2019 harvest) commercialized in Campo Grande, MS – 2019

Company	G (%)	SL (cm)	SDM (g)	SE (%)	SLS (cm)	SDMS (g)	TSW (g)
A	53.5b	6.0a	0.32a	46.6a	7.8b	0.3a	0.87a
B	91.2a	3.3b	0.11c	44.2a	8.8b	0.2a	0.78b
C	80.5a	5.8a	0.24b	45.2a	5.8c	0.4a	0.78b
D	53.2b	6.2a	0.25ab	50.3a	9.7a	0.2a	0.81ab

(A) Company A; (B) Company B; (C) Company C; (D) Company D. Means Followed By The Same Letter Do Not Differ Statistically By Tukey's Test At A 5% Probability Level. *Ns = Not Significant.
Source: research data.

Mean values for sand emergence and seedling dry mass in sand did not differ significantly among lots, indicating similar physiological performance among samples for these variables. However, for Company B—which had the highest germination rate—seedling length (SL) and seedling length in sand (SLS) were not proportional. This discrepancy may be explained by the fact that germination reflects only the seed's initial capacity to produce essential structures under optimal conditions, without necessarily indicating vigor. Thus, even with high germination rates, seedlings may exhibit reduced growth, suggesting that the seeds, though viable, have low energy reserves, physiological damage, or reduced metabolic efficiency during early development.

Additionally, storage conditions, physiological maturity, and seed lot characteristics may have negatively affected seedling growth, resulting in nonuniform development and shorter mean length (Chortaszko *et al*., 2019; Cook *et al*., 2020; Oliveira *et al*., 2021).

For sand emergence, values ranged from 44.2% to 50.3%: 46.6% (Company A), 44.2% (B), 45.2% (C), and 50.3% (D). Since all averages were below 60%, the evaluated lots would not meet the official quality standards for the commercialization of forage grass seeds, as established by the Brazilian Ministry of Agriculture's Normative Instruction No. 45 (Brasil, 2021; Oliveira *et al.*, 2021).

The thousand-seed weight (TSW) varied among companies: 0.87 g (A), 0.78 g (B and C), and 0.81 g (D). This parameter is essential for comparing physical seed quality, estimating crop yield, calculating sowing density, determining seed number per package, and defining sample size for purity analysis when not established by the Rules for Seed Testing (Brasil, 2009). Moreover, TSW is related to seed maturity and health status, providing an indirect indicator of developmental stage (Silva *et al.*, 2019a; Silva *et al.*, 2019b). The observed values suggest the possible presence of impurities or malformed seeds in some lots, potentially causing seedling nonuniformity and requiring higher sowing rates per hectare to achieve adequate stand density.

The presence of pathogens after physiological maturity or during storage poses a serious threat to seed quality. High infestation and infection levels are associated with reduced germination capacity and poor seedling development (Chortaszko *et al.*, 2019). According to Marcos-Filho (2015), contaminated seeds may serve as a vector for pathogen transmission to aerial and root tissues, reducing vigor and causing irregular field stands.

Seed quality is a key determinant of the establishment and persistence of tropical pastures. However, it is common for low-quality lots—physiologically, genetically, and sanitarily—to be used, often containing plant debris, soil particles, or weed seeds. Factors contributing to this situation include the lack of strict production and commercialization standards, insufficient inspection, and the acquisition criteria based solely on seed price per kilogram, which discourages quality control practices (Alves *et al.*, 2017; Cruz *et al.*, 2020).

Regarding sanitary quality, there is still a shortage of studies on pathogens associated with tropical forage seeds produced and commercialized in Brazil, which may explain the increasing incidence of diseases in pastures (Martins *et al.*, 2017). Among the main agents detected are phytopathogenic fungi, whose presence has significant impacts on the forage sector. These pathogens can reduce germination and vigor, hinder stand establishment, decrease seed production, and shorten pasture longevity—negatively affecting meat and milk production. Moreover, seed contamination by pathogens also restricts the export of Brazilian forage seeds.

4 Conclusion

Regardless of the company analyzed, the seed samples showed the occurrence of fungi from different genera, which may influence physiological quality and, consequently, contribute to the low germination percentage and seedling emergence.

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