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Chemical analysis and in situ dry matter disappearance of açaí seeds chemically treated with urea

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ABSTRACT. This study aimed to analyze the chemical composition and in situ dry matter disappearance of *açaí* seeds treated with urea. *Açaí* seeds were treated with urea solutions at different concentrations (0, 5 and 7.5% CH_4N_2O). Subsequently, chemical analyses and in situ tests were carried out to evaluate the disappearance of dry matter (DM) in the periods of 0, 6, 12, 24, 48, 72, and 96 hours of incubation. The increasing inclusion of urea influenced the DM, crude protein (CP), ether extract (EE), neutral detergent fiber (NDF), and acid detergent fiber (ADF), except for mineral matter (MM). The DM content decreased with increasing urea levels, the control presented 92.99%. CP increased by 23.93% as urea inclusion increased. EE content was lower in the 5% treatment, reaching 0.79%. NDF, ADF, and lignin of the *açaí* seed decreased with increasing urea, with a lower content observed in the 7.5% CH_4N_2O treatment. Urea influenced the disappearance of DM, with greater loss in the 96-hour incubation in the 7.5% CH_4N_2O treatment. *Açaí* seeds can be included in the diet of ruminants when treated with urea.

Keywords: By-product; *Euterpe oleracea* Mart; animal nutrition; alkaline treatment.

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Introduction

In Brazil, most ruminants are raised in extensive systems and receive protein and energy supplementation as part of their diet (Reis et al., 2016). However, during dry periods, there is low availability of forage mass, and the provision of energy and protein supplementation with corn and soybeans is costly, so it is necessary to use food alternatives, such as agro-industrial by-products in animal feed (Castro et al., 2023; Menezes et al., 2016).

In this sense, the by-product of *açaí*, the fruit of a palm tree (*Euterpe oleracea* Mart), can be part of the diet of ruminants as it is a native plant to the Brazilian Amazon region, of which the state of Pará is the top producer (Instituto Brasileiro de Geografia e Estatística [IBGE], 2022). However, the seed has low nutritional value, low protein content (3.78%), and high fiber content (77.2%) (Arruda et al., 2018), which makes digestion difficult, mainly due to the high lignin content. In turn, chemical treatment can improve the nutritional composition of this by-product.

Urea can break down fiber and increase feed digestibility, as it breaks the ester bonds of the cell wall, in addition to acting on phenolic acids with the partial depolymerization of lignin (Pires et al., 2004), thus increasing the availability of nutrients, especially in ruminal degradation (Silva et al., 2016) and increasing the content of non-protein nitrogen, which is important for the ruminal microbiota.

Nutrition is an important factor in the effectiveness of animal production systems (Luz et al., 2019). Assessing the nutritional value of the feed before offering it, as well as its digestion rate, is essential to know how much energy and nitrogen are available (Goes et al., 2010) so that the animal can express its full potential in productivity. Among the techniques used to assess the degradation rate, in situ incubation is a viable technique, as it is fast, inexpensive, and requires a small number of samples inside the rumen (Costa et al., 2016).

Given the above, the goal was to evaluate the effect of chemical treatment with urea on the chemical composition and the in situ dry matter disappearance of *açaí* seeds.

Material and methods

Chemical treatment and chemical analysis

The research was approved by the Ethics and Animal Welfare Committee of UFPA under protocol 2898210318.

Samples of the *açaí* by-product were collected in the municipality of Castanhal, state of Pará, obtained from a juice and pulp factory, as this showed better standardization because the pulp extraction process is fully mechanized. The material was taken to Campus II of the Federal University of Pará in Castanhal, spread on a tarpaulin in a covered and ventilated place, and homogenized for four days, obtaining the air-dry mass (ADM). At the end of this period, the material was ground using a 5 mm sieve, which was used in the chemical treatment with urea (CH_4N_2O).

This was a completely randomized experimental design, with three treatments according to the CH_4N_2O level. The experimental groups were: Control - treated only with water; 5% CH_4N_2O - treated with 5% CH_4N_2O; 7.5% CH_4N_2O - treated with 7.5% CH_4N_2O, with the urea percentages based on the ground ADM. In each treatment, the solute was diluted in 1.5 L of water in a plastic bucket to which the respective ADM was added and homogenized, leaving it to stand for 24 h. The samples were then taken to an open area and remained there for 96 h. During this period, they were left in the shade for the chemical reaction of the product with the ground *açaí* seed and were mixed with a shovel twice a day to facilitate drying. During this period, the temperature and relative humidity averaged 25.3°C and 86.4%. At the end, the samples from each treatment were individually bagged for chemical analysis.

Initially, pre-drying (method G-001/1) and grinding were carried out in a knife mill with a 1 mm sieve, subsequently, the dry matter (DM; method G-003/1), mineral matter (MM; method M-001/1), crude protein (CP; method N-001/1), ether extract (EE; method G-005/1), neutral detergent insoluble fiber (NDF; method F-002/1), acid detergent insoluble fiber (ADF; method F-004/1), and lignin (Lig; method F-005/1) were determined according to Detmann et al. (2012). To characterize the *açaí* seed (Table 1), ADM was collected and analyzed according to the procedures and methods mentioned.

Table 1. Chemical characterization of açaí seeds.

Analysis	%
Dry matter	91.37
Mineral matter[1]	1.74
Crude protein[1]	5.70
Ether extract[1]	1.30
Neutral detergent insoluble fiber[1]	90.20
Acid detergent insoluble fiber[1]	86.62
Lignin[1]	16.63

[1]% DM.

In situ Incubation

Two cows, approximately 48 months of age and with an average body weight of 480 kg, rumen fistulated, fed elephant grass and a concentrate based on wheat bran, soybean meal, limestone, urea, and mineral salt, were used for incubation in the rumen. For incubation in the rumen, 4 g samples were weighed in nylon bags, and 3 bags were introduced at each respective time (6, 12, 24, 48, 72, and 96 hours) of incubation, considering each treatment, which totaled 54 bags incubated per cow. Reverse incubation was used, so that all bags were removed at the same time, immersed in ice water to stop microbial activity and fermentation, washed in running water, and samples collected for DM analysis.

Statistical analysis

The results were tested by analysis of variance and when differences were detected, the means were compared using the Student-Newman-Keuls test at 5% probability.

Results and discussion

The chemical composition of the *açaí* seed treated with urea is listed in Table 2. The contents of DM, CP, EE, NDF, ADF, and lignin were influenced ($p < 0.05$) by the inclusion levels of CH_4N_2O.

Table 2. Chemical composition of *açaí* seed chemically treated with urea.

Variable	Treatments			p-value
	Control	5% CH_4N_2O	7.5 % CH_4N_2O	
DM	92.99[a]	92.87[b]	91.83[c]	< 0.05
MM[1]	1.52[a]	1.59[a]	1.41[a]	0.952
CP[1]	5.70[c]	20.07[b]	23.93[a]	< 0.05
EE[1]	1.30[a]	0.79[b]	1.26[a]	< 0.05
NDF[1]	90.16[a]	87.73[b]	80.44[c]	< 0.05
ADF[1]	87.57[a]	85.16[b]	77.67[c]	< 0.05
Lig[1]	17.56[a]	13.99[b]	9.60[c]	< 0.05

[1]% of DM; DM: dry matter; CP: crude protein; EE: ether extract; NDF: neutral detergent insoluble fiber: ADF: acid detergent insoluble fiber; Lig: lignin. Mean values followed by a different letter in the same row are significantly different by the SNK Test at 5% probability.

The DM content decreased with increasing urea levels, differing significantly ($p < 0.05$) in each treatment; the control showed the highest DM content. As the urea inclusion level increased, greater water retention occurred; therefore, these treatments lost more water when they underwent the drying process than the control. Urea has high hygroscopicity, that is, high affinity with water; therefore, ammoniated materials have a lower dry matter content than untreated materials (Pires et al., 2010).

CP differed significantly ($p < 0.05$) with a higher content in the 7.5% CH_4N_2O treatment; this is because the chemical treatment with urea enabled an increase in nitrogen levels in the *açaí* seed, as it is the addition of non-protein nitrogen that results in an increase in total nitrogen, computed as CP (Deminicis et al., 2015). By increasing the availability of nitrogen in the feed, it enables its use as a protein supplement in the diet of ruminants. Typically, bulky feeds have low CP content, and nutrient correction is almost always necessary to improve animal performance since CP deficiency in the diet can limit microbial digestion and contribute to the physical limitation of intake (Morais et al., 2020).

The EE content was lower in the 5% CH_4N_2O treatment and differed significantly ($p < 0.05$) from the other treatments. The values found are considered low, coinciding with those reported by Deminicis et al. (2015). The EE content should not exceed 7% in the total diet of ruminants as it may impair digestibility, and intake, and cause deleterious effects (Van Soest, 1994).

The NDF, ADF, and lignin of the *açaí* seed decreased with increasing levels of urea inclusion, with the lowest values in the 7.5% CH_4N_2O treatment, and differing significantly ($p < 0.05$) from the other treatments. One of the effects of the action of urea on bulky materials is the breakdown of the complex formed by the components of the cell wall, usually more expressive on the NDF fraction, due to the partial solubilization of hemicellulose, causing the disruption of bonds between constituents of the cell wall and phenolic acids, thus promoting the partial depolymerization of lignin (Cruz & Silva, 2016; Souza Junior et al., 2011). When evaluating the effect of increasing urea levels on the nutritional value of Tanzania grass hay (*Panicum maximum*, cv. Tanzania), some authors found that the chemical treatment of this material with urea caused a reduction in the content of NDF, ADF, and hemicellulose (Zanine et al. 2007).

Moreover, NDF content influences animal intake and it is very important to know its levels since when they are above 55-60% in dry matter, they are negatively correlated with forage intake (Van Soest, 1994). High levels of ADF in the feed can impair digestibility, since the indigestible fiber fraction, lignin, represents the largest proportion of ADF (Figueiredo et al., 2019). Therefore, it is important to chemically treat the *açaí* seed for supply in ruminant diets.

The incubation period and the urea inclusion level significantly influenced ($p < 0.05$) the *in situ* DM disappearance (Table 3). The disappearance of DM was more pronounced in the 96h period with an inclusion level of 7.5% urea, corroborating Silva et al. (2016) who reported the disappearance of DM when there was a greater inclusion of chemical products in the food.

Table 3. In situ dry matter disappearance for different incubation periods of *açaí* seeds treated with different levels of urea.

Treatment	Incubation period (hours)						p-value
	6	12	24	48	72	96	
Control	13.86 Ad	14.07 Ad	17.64 Ad	38.52 Ac	38.52 Ab	39.91 ABa	< 0.0001
5% CH_4N_2O	11.29 Ac	10.54 Ac	23.42 Ab	27.58 Ab	34.91 Aa	39.79 Ba	< 0.0001
7.5% CH_4N_2O	10.74 Ac	10.97 Ac	17.01 Ac	27.02 Ab	32.68 Ab	41.75 Aa	< 0.0001

Means followed by distinct lowercase letters in the same row and distinct uppercase letters in the same column are significantly different by the SNK test at 5% probability.

The addition of urea to the *açaí* seed improved the nutritional value of the food, but the high NDF and LIG values prevented greater efficiency in the disappearance of the food in the rumen since foods with high NDF content require more time to be degraded (Sousa Júnior et al., 2011; Romão et al., 2014). Another factor may be the increase in urea in the treatments, which increases the degradation of urea itself in the ruminal microbiota by almost 100% (Sniffen, 1974).

Conclusion

Açaí seeds chemically treated with urea promoted a reduction in the content of dry matter, neutral and acid detergent insoluble fiber and lignin, and an increase in crude protein. However, due to the lignin content, it is considered that this material should not be used as the sole bulky feed.

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