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ECOSYSTEMS

Effect of frugivory on the germination of cumbaru (*Dipteryx alata*) seeds ingested by tapirs (*Tapirus terrestris*) in the area of Cerrado, Brazil

ADEMIR KLEBER M. DE OLIVEIRA, ROSEMARY MATIAS, RICHARD MATHEUS FERNANDES & JOSÉ CARLOS PINA

Abstract: Endozoochory refers to the ingestion of fruits by animals and their release through faeces as it plays an important role in maintaining ecosystems. This study aimed to evaluate the effect of cumbaru fruit ingestion by tapirs on cumbaru seed germination. In latrines, fruits ingested and excreted by the animals were collected among tapir faeces. The collected material was taken to the laboratory and placed in a germination chamber and greenhouse. Fruit ingestion and its subsequent defection propitiated a higher germination rate and germination vigor compared to non-ingested fruit grown in a germination chamber and greenhouse. The results in the greenhouse showed that the latrines (decomposing organic matter) have different thermal conditions (higher temperature), in relation to the external environment, which probably accelerated the germination processes. Evaluation of seedlings grown in a greenhouse and germination chamber indicated that growth in the greenhouse was better due to humic and fulvic acids, which are common stimulants of seed germination and seedling growth. Therefore, tapirs can be considered good fruit dispersers and inducers of cumbaru seeds germination.

Key words: Endozoochory, humic acids, seedlings growth, tapir latrines.

INTRODUCTION

Frugivory plays an important role in ecosystem maintenance. It is an interaction between animals and plants and one of the main events related to the establishment of new individuals of certain plant species, by providing processes such as seed dispersal, overcoming dormancy, acceleration of seed germination, and subsequently, seedling formation, for example (Van Der Pijl 1982, Traveset 1998).

Endozoochory is a frugivory process characterised by the complete intake of fruits by animals. The fruit travels through the entire digestive system and is released through the stool (Van Der Pijl 1982). Tropical forests have many fruit-producing plant species whose seeds are dispersed by vertebrates, mainly birds and mammals, reaching in certain regions 94% of plant species with seed dispersal by animals (Jordano 2000).

In the Cerrado biome, there are 195 species of mammals, 17 of which are on the national list of endangered species of Brazilian fauna (Rocha & Silva 2009, Paglia et al. 2012). The tapir [*Tapirus terrestris* (Linnaeus 1758)] stands out among these. It is considered the largest neotropical land mammal. It has a wide geographic distribution throughout South America, except

Chile and Uruguay and its adult weight ranges from 150 to 300 kg (Médici et al. 2012, Prado 2012).

According to Médici et al. (2012), the cause for the tapir's endangerment is deforestation, which leads to habitat fragmentation and population isolation. This situation results in low connectivity between individuals, due to agricultural activities. In addition, predatory hunting, firearms, trampling on roads, infectious diseases by contact with domestic animals, and insufficiently protected areas have also reduced the species' population. Prado (2012) writes that the tapir has a diet based on fruits, leaves, stems, buds, small branches, aquatic plants and aquatic organisms, for example. According to Bodmer (1990), the consumption of fruits corresponds to about a third of their diet, and they can ingest fruits whose seeds vary from 1 to 50 mm in diameter. Their eating behavior points to an ecological plasticity in the ingestion, according to the resources available in the environment (Prado 2012). A study by Talamoni & Assis (2009) corroborates this assertion of the species' nutritional versatility, especially in transition areas between Cerrado and the Atlantic Forest.

Cerrado has high plant biodiversity, comprising more than 11,627 vascular species, representing approximately 30% of the Brazilian flora (Mendonça et al. 2008). Many fruit plants are important for food due to their high levels of proteins, vitamins, sugars, and minerals (Avidos & Ferreira 2000). Among these species is *Dipteryx alata* Vog., popularly known as cumbaru, cumaru, or baru, among other popular names.

Belonging to the family Fabaceae, cumbaru is a common species, considered initial to late secondary in the processes of plant succession. Its reproductive phenology from October to January (flowering) and from March to August (fruit maturation). They can reach up to 20 meters in height as mature plants (Carvalho 2010). However, its distribution has been threatened due to agricultural activities. The Cerrado is the second most significantly anthropogenic altered area after the Atlantic Forest (Canuto et al. 2015).

The fruits of the species (pulp) are consumed by mammals (bats and monkeys, for example), in addition to the seeds (almonds) being used in human food due to their chemical characteristics and flavor (Carvalho 2010). Its pulp consists of carbohydrates (63%), with a predominance of starch, insoluble fibers, and sugars (Alves et al. 2010, Carvalho 2010). The almonds are composed mainly of lipids and proteins. They also contain calcium, phosphorus, manganese, and potassium (Sousa et al. 2011).

Although there are no records of the dispersion of cumbaru fruits/seeds by tapirs within the Cerrado biome, studies indicate a slight preference for fruits belonging to certain botanical families, such as Anacardiaceae, Fabaceae and Myrtaceae in Athlantic Foret (Seibert 2015) and in the Cerrado biome, Poaceae, Myrtaceae, Rubiaceae, Salicaceae, Arecaceae, Fabaceae and Solanaceae (Prado 2012).

The present article aimed to test the hypothesis that the ingestion of cumbaru fruit by the species *Tapirus terrestris* affects their rates of seed germination and seedling formation.

MATERIALS AND METHODS

Collection area

The study site was located in Serra de Maracaju, situated in the central region of the state of Mato Grosso do Sul, region of Taboco, municipality of Corguinho (latitude 19°49'54" South and longitude 54°49'46" West), Brazil. Its average altitude is 320 m. According to Köppen and Geiger, the climate is tropical (Aw), with dry winters and rainy summers, with an average temperature of 24.8°C and average annual rainfall of 1444 mm (SEMAC 2011).

The collection region lies in the Chaqueno morphoclimatic domain in the subregion of Cerrado (Morrone 2001), with a predominance of cerrado (Brazilian savannah) *stricto sensu* and the presence of cerradões (xeromorphic forest) and seasonal forests, where there was the presence of tree species, such as *Caryocar brasiliense* Cambess., *Copaifera langsdorffii* Desf., *Dimorphandra mollis* Benth., *Dipteryx alata* Vogel, *Qualea grandiflora* Mart., *Terminalia argentea* Mart. & Zucc., *Vochysia haenkeana* Mart., and *Xylopia aromatica* (Lam.) Mart., among others.

In the gallery forests, there were species such as *Alibertia edulis* (Rich). A. Rich. ex DC., *Calophyllum brasiliense* Cambess., *Guarea guidonia* (L.) Sleumer., *Inga nobilis* Willd., *Maclura tinctoria* (L.) D. Don ex Steud., *Myrsine coriacea* (Sw.) R. Br. ex Roem. & Schult., *Protium heptaphyllum* (Aubl.) Marchand, *Psychotria carthagenensis* Jacq., *Tapirira guianensis* Aubl., and *Xylopia sericea* A. St.-Hill.

However, most of the region had pastures plated with *Brachiaria dictyoneura* (Figure & De Not.) Stapf dominating the flatter places.

Collection of material

The confirmation of the faeces as coming from individuals of tapir was made by means of prior identification of places used by the species and collection of fresh faeces from the individuals, observed in some places defecating, with cumbaru fruits present in the faeces. During 2017, active searches for tapir faeces took place in cerrado, cerradões, and seasonal forests (Figure 1a, b) and eight latrines were located, and a manual check was carried out, removing cumbaru fruits. According to Bachand et al. (2009), this species normally uses the same place to defecate, forming latrines (Figure 2).

In other places, fallen cumbaru fruits were collected under the canopy of mature trees (height above ± 15 m) (Figure 1c, d). All the collected material was stored in paper bags and transported for analysis.

Chemical characterisation of faeces (latrine)

The pH of the faeces was characterised in H_2O. Calcium (Ca), magnesium (Mg), and exchangeable acidity ($H^+ + Al^{3+}$ = hydrogen + aluminium) contents were obtained with KCl (1M) solution. Analyses were performed by the titrimetric method. Organic matter, phosphorus (P), and potassium (K) were extracted using Mehlich-1 solution and analysed using the colourimetric method.

The extraction, fractionation, and purification of humic acids were performed following the methodological protocol described in Dick & Burba (1999). The alkaline extract corresponding to the humic soluble substances (HSS) was acidified to pH 2.0 (HCl 0.1 mol L^{-1}). After 24 hours, the solution was centrifuged, and the fulvic acid (FA) supernatant was separated from the humic acid (HA) precipitate. The carbon contents in the acid extract of HSS and FA were also quantified by measuring the absorbance at 580 nm after oxidation of the organic matter in acid medium with $K_2C_{r2}O_7$ at 60°C (Dick & Burba 1999). The absorbance was determined by UV-Vis spectroscopy at 465 (E4) and at 665 nm (E6) for the supernatants of HSS and FA, and calculating the ratio E4/E6 (Chen et al. 1977). The scanning spectrum of the two fractions (HSS and FA) was performed at wavelengths from 200 to 800 nm, checking possible interferences of solubilized inorganic components and determining the chemical profile of the fractions. A UV/Visible spectrophotometer, Fento model 800XI, was used for this procedure.

Figure 1. Overview of the collection area in the municipality of Corguinho, Mato Grosso do Sul, Brazil (a), one of the collection points (b), cumbaru tree (c), and cumbaru fruits (d).

Figure 2. Latrines in areas of cerrado (a, c) and seasonal forests (b, d) in the municipality of Corguinho, Mato Grosso do Sul, Brazil.

Determination of total phenolic and carboxylic acidity

The total acidity was determined by the method of $Ba(OH)_2$, with titration of the excess with HCl. The carboxylic acidity was determined with $Ca(OAc)_2$ (calcium acetate) and the determination of the liberated acid was carried out by means of NaOH. The phenolic acidity was obtained through the difference of the total acidity – carboxylic, according to the method of Schnitzer & Gupta (1965).

Degree of humidity

The initial moisture degree content in the seeds was obtained using the gravimetric method with seeds dried in an oven at 105 + 3°C for 24 hours to measure the water mass removed from the seeds by drying, using the arithmetic mean of four samples (Brasil 2009).

Bioassays: laboratory and greenhouse

Laboratory

The material used (seeds) was sanitised in 1% sodium hypochlorite for three minutes and then washed in running water for one minute. Four replications of 15 seeds and/or fruits per treatment were performed. Fruits or seeds were placed in transparent plastic boxes (11x11x3.5 cm) lined with vermiculite (Figure 3c), with a volume of water equivalent to 1.5 times its dry mass. The boxes were maintained in a germination chamber with a photoperiod of 12 h of white light (± 660 lux) at a fixed temperature of 30°C.

There were four experiments: (1) Seed control, comprised of seeds taken from fruits found under the crown of cumbaru trees; (2) Fruit control, comprised of intact fruit collected under the tree canopy; (3) Fruits consumed by tapirs and found in faeces (endozoochory fruits); and (4) Seeds taken from fruits consumed by tapirs (endozoochory seeds).

Experiments 3 and 4 (germination chamber) were evaluated daily (six days). We only considered seeds that had germinated a primary root that exceeded 2 mm. Experiments 1 and 2 (greenhouse) lasted 60 days (emission of aerial part = germination).

When the seeds or fruits showed signs of contamination by fungi, the boxes were removed and immersed in a 0.1% deconil® fungicide solution for three minutes. The experimental design was completely randomised. The non-germinated seeds were subjected to the tetrazolium test, according to Brasil (2009).

The percentage of germination (visualised through the emission of the primary root, botanical procedure) and the vigor (measured indirectly by average time germination [ATG] in days) were evaluated, quantifying the germination under the kinetic point of view

Figure 3. **Experiment with faeces collected in the latrines and the emergence of cumbaru seedlings (a, b), vase with the presence of thermometer and cumbaru seedlings (c), cumbaru fruits placed in vermiculite (d), and cumbaru fruits placed in sand with fungi before disinfection (e). Specimens were collected in the municipality of Corguinho, Mato Grosso do Sul, Brazil.**

(Labouriau 1983) and through the germination speed index (GSI) (Maguire 1962). The germination data was transformed in accordance with the tests of normality and homogeneity of variances (Shapiro-Wilk test for normality of ANOVA residues and the Levene test for homogeneity between variances). The data were analyzed using the software Bioestat 5.0. A comparison was performed through Tukey test at 5% probability when there was significance. The averages presented in the tables correspond to the non-transformed data to better understand the results.

Greenhouse

Two experiments were carried out in pots: (1) Fruits taken from faeces mixed with organic matter taken from latrines (Figure 3a), and (2) Fruits collected under the canopy of mother trees placed on sandy soil (Figure 3d). The experiments (pots) were watered regularly. The temperatures inside the pots (Figure 3a) and in the environment were measured with thermometers.

The index of emergence (cumulative data) was the number of plants that emerged each day. The vigor of the seeds was measured indirectly through the calculations of the average time for emergence (ATE) in days and the emergence speed index (ESI) (Maguire 1962, Labouriau 1983).

The initial development of seedlings (Figure 3b) was evaluated after the end of the experiments. It was determined by the size of the primary root as measured by digital calliper from the seedling lap to the meristematic apex of the root system (mm) and the aerial part (mm) from the plant lap to the apical apex of the aerial part. The dry mass was calculated using the roots and shoots from the length measurement placed in paper bags and kept in a forced ventilation oven at a temperature of 80° C for 24 hours. Afterwards, they were weighed on an analytical balance, and the result was expressed in grams (average). The experimental design was completely randomised. The non-germinated seeds (Figure 4) were subjected to the tetrazolium test, according to Brasil (2009).

RESULTS

The water content of the seeds was 10.2% for seeds from the latrines and 9.6% for "Control seeds", which was not statistically different ($p = 0.1270$, $F = 2.63$, dms = 0.80). Regarding germination, there was no significant difference between "Seed control", "Endozoochory fruits",

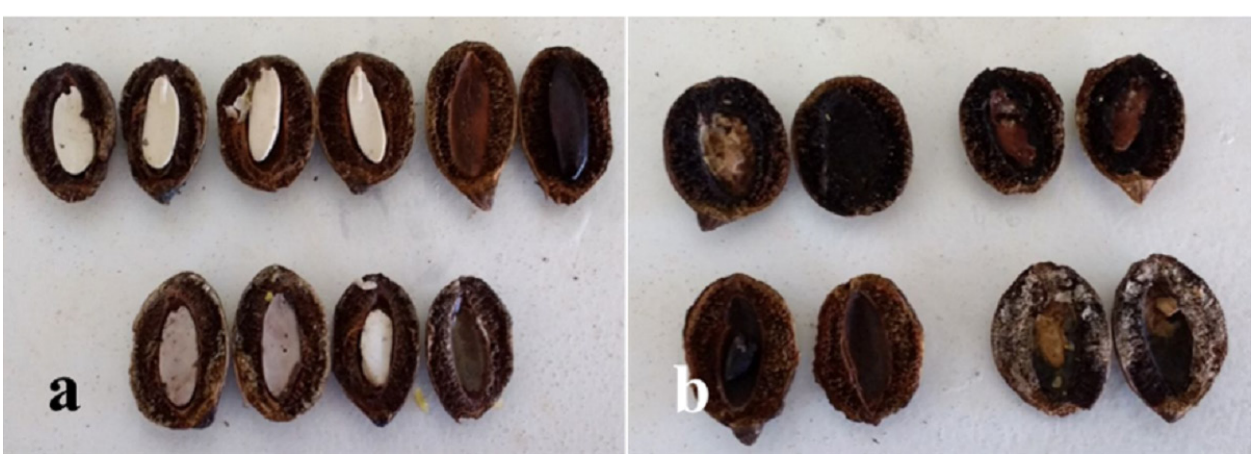

Figure 4. Open fruits with viable (a) and non-viable (b) seeds collected from the municipality of Corguinho, Mato Grosso do Sul, Brazil.

and "Endozoochory seeds" ($p < 0.0001$, $F = 5927.1$) for the second through the fifth day. The "Fruit control" using intact fruits did not show primary root protrusion (Table I).

However, the fruits (seeds) that went through the ingestion process showed greater vigor (high GSI and low ATG), compared to seeds obtained from fruits not eaten (Table I), with a significant difference between treatments (GSI: $p < 0.0001$, $F = 257.1$; ATG: $p < 0.0001$, $F = 392.0$).

At the end of the evaluation period, the opening of the seeds in the "Fruits control" (germination chamber) indicated that all the seeds had intact embryos (tetrazolium test), indicating that they were viable but dormant.

Concerning the 50% of non-germinated seeds (Fruits in the latrine - greenhouse), the tetrazolium test indicated that 24% were viable and 34% had deteriorated. Thus, if the experiment observation period had been longer, there could have been a greater number of seeds germinated. On the other hand, the seeds kept in sandy soil (Fruits on the sand) did not germinate. Even though, the seeds were viable, as determined by the tetrazolium test.

The emergence process for the fruit kept in the greenhouse began on the 19th day of the experiment and lasted until the 30th day. It only occurred in fruits from the latrines and reached 50% emergence (Table II) with a significant difference between treatments ($p < 0.0001$, $F = 1500.0$). After opening the non-geminated fruits (Fruits in the latrine), the tetrazolium test indicated that 46% of the seeds were viable, and 54% were deteriorated due to the high incidence of fungi. On the other hand, "Fruits on the sandy" soil had 100% viable seeds (tetrazolium test).

Although half of the seeds germinated, about 50% of the seedlings were predated by individuals in the larval stage belonging to the family of Bruchideos (Coleoptera), which led to their death. Other seeds did not form complete seedlings, as the larvae consumed the primary roots, killing them.

Concerning the average temperature inside the organic matter of the latrines, in the greenhouse, 28.7°C was observed. In the environment outside the greenhouse, the average temperature was 25.6°C, a difference of 3.1°C. As the experiment progressed, the greater thermal activity of the organic matter decreased, reflecting a smaller temperature range in relation to the external environment (Figure 5).

The assessments of the growth of seedlings in the greenhouse and germination chamber (Table III) indicated that the growth in the greenhouse was better regarding length and weight with a significant difference between treatments ($p < 0.0001$; weight of aerial part, F

Table I. Germination (%), germination speed index (GSI), and average time of germination (ATG) in the germination chamber.

Treatments	Germination (%)	GSI	ATG (days)
Seeds control	97 a	28.1 b	4.11 b
Fruits control	0	-	-
Endozoochory fruits	100 a	36.0 a	3.11 a
Seeds endozoochory	98 a	39.7 a	2.81 a
dms (5%)	2.69	4.70	0.37

***Averages in the same column followed by the same letter do not significantly differ by the Tukey test ($p < 0.05$). dms: value of the least significant difference for the chosen test.**

= 713.5; weight of roots, F = 115.5; length of aerial part, F = 63.2; length of roots, F = 788.9).

The chemical characteristics evaluated in the organic matter of the latrines indicated a pH 6.1 (pH_{H2O}), exchangeable acidity = 0.2 mg dm^{-3} of Al^{+3}, calcium = 5.0 mg dm^{-3}, magnesium = 3.6 mg dm^{-3}, phosphorus = 29 mg dm^{-3}, and potassium = 636 mg dm^{-3}. The total organic carbon content was 77.1 g dm^{-3}, humic acids = 39.1 g dm^{-3}, and fulvic acids = 27.5 g dm^{-3}. The results of the scanning spectrum (Visible UV) confirmed the separation of these two acids, with humic acid and fulvic acid showing three maximum absorption bands, 290, 464, and 513 nm (Figure 6a) and 226, 335, and 445 nm (Figure 6b), respectively.

The values of acidity were: total = 612.7 cmol kg^{-1}, carboxyliv = 544.7 cmol kg^{-1}, and phenolic = 124.7 cmol kg^{-1}. These values indicated carboxylic (COOH), phenolic (Ar-OH), or enolic groups (Ar-O-R) that are rich in oxygen and sustain the reactivity of humic substances.

DISCUSSION

The seed water content was consistent with the values usually cited for most plant species, between 5 and 20% (Bewley et al. 2013). This is one of the characteristics of orthodox seeds. The results are higher than those found by Botezelli et al. (2000), with cumbaru seeds from different origins presenting moisture variations between 6.1% and 8.2%. The higher water content in seeds of the Taboco region may be related to genetic and environmental factors that may alter plant structures for adaptation to the conditions existing in its place of origin (Bewley et al. 2013). The high germination rates in our experimental groups (Seeds control, Endozoochory fruits and Endozoochory seeds) indicated that the seeds were at their highest point of physiological maturity, and ready for germination.

Cumbaru fruits or seeds collected from latrines have germination rates similar to those found in seeds obtained from intact fruits collected under the canopy of parent plants. However, fruits (seeds) that went through the ingestion process showed greater vigor than seeds obtained from fruits not eaten. This demonstrated that ingestion, in addition to allowing dormancy break, also results in a greater number of germinated seeds in less time. During the process of digestion and excretion of fruits, putrefaction occurs through the fermentation of the pericarp and endocarp, a situation called “overcoming dormancy by fermentation” (Melo et al. 2008). This benefits the germination process. In other words, the passage of cumbaru fruits through the tapirs’ digestive system provides a competitive advantage in the germination process.

We found shorter germination periods than those mentioned by Botezelli et al. (2000), with laboratory tests (Without endozoochory process) and primary root protrusion starting between the fifth and eighth day. However, the origin region for the seeds interfered with its vigor, as previously indicated. Vigor, positively affected by endozoochory, showed that greater

Table II. **Emergence (%), emergence speed index (ESI) and average time of emergence (ATE) in the greenhouse.**

Treatments	Emergence (%)	ESI	ATE (days)
Fruits in the latrine	50 a	0.12 a	25.3 a
Fruits on the sand	0 b	-	-
dms (5%)	3.16	-	-

***Averages in the same column followed by the same letter do not significantly differ by the Tukey test ($p < 0.05$). dms: value of the least significant difference for the chosen test.**

physiological maturity was achieved after passing through the digestive system, leading to faster germination (shorter time), which is beneficial for the species, avoiding attack by pathogens, by example. Oliveira et al. (2011) confirmed that seeds that take a long time to germinate can be attacked by a greater number of fungi during the imbibition process and not germinate. Scremin-Dias et al. (2006) also described that seeds with higher germination speeds (high vigor) will suffer less influence from pathogens.

The germination results (between 88 and 95%) are consistent with those reported by Botezelli et al. (2000) and Carvalho (2010), with fruits of the same species but harvested directly from trees. Corrêa et al. (2000), evaluating the emergence of cumbaru seeds from different sources, found an overall mean value of 97%. These results demonstrate that the seeds of the species germinate in large numbers when under suitable conditions. However, passage through the digestive system positively affects the germination vigor, in relation to the seeds obtained from fruits that were not ingested by the animals.

It is necessary to remove the pulp from the fruits for germination, as it represents a mechanical (integumentary) dormancy due to the impermeability of the external integument (Carvalho 2010). This was observed for the “Seed control”, using intact fruits, where no germination occurred. The opening of the fruits indicated that 50% of the seeds were viable and 50% had apparent signs of degradation, a factor related to the high incidence of fungi, despite the fruits being disinfected. Quirino et al. (2016), working with the health of cumbaru seeds, verified the predisposition to attack by nine species of fungi. They also observed the inefficiency of seed asepsis methods. For this reason, the passage of seeds through the digestive system, in addition to providing better rates of germination and vigor, reduced the loss of seeds through the attack of fungi.

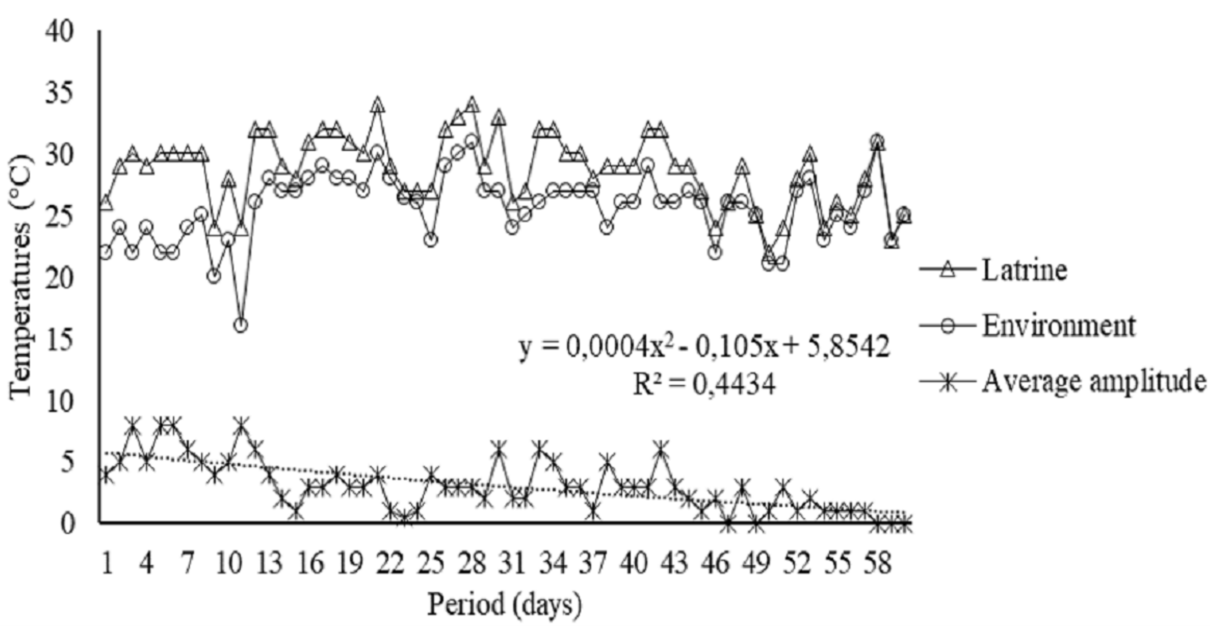

Figure 5. Average temperature of the external environment, the interior of organic matter, and temperature range (°C) at the municipality of Campo Grande, Mato Grosso do Sul, Brazil.

The emergence experiment carried out in a greenhouse showed that 50% of the fruits in the latrine treatment showed seedling emergence, a percentage lower than that obtained in a germination chamber, which could be expected. Under controlled conditions (germination chamber), germination tends to be higher, when compared to what occurs in field conditions (greenhouse), where environmental factors (temperature, for example) are variable. In addition to this factor, the presence of predators and pathogens are a threat to the integrity of the seeds, which can lead to a lower germination rate, as observed in our experiment.

Among those that were not germinated, 46% were viable. If the experiment continued to develop, a portion of the viable seeds would probably form new seedlings, demonstrating that the seeds present in the latrines take longer than 60 days to emerge. However, according to Carvalho (2010), the germination of the species occurs between 13 to 60 days after sowing.

The results obtained by this work indicate that the time for germination to occur can vary, depending on the origin of the seeds and the methodologies used. Under field conditions, the germination process can extend beyond those outlined above, demonstrating the species' adaptation to existing environmental conditions. In contrast, fruits kept on sandy soil (Fruits on the sand) did not produce seedlings despite seed viability. The tetrazolium test later confirmed this, a related factor to the tegumentary dormancy present in the species.

Table III. Length (mm) and weight (g) of the root and aerial parts of seedlings maintained in a germination chamber or greenhouse, 30 days after germination.

Weight (g)	Aerial part	Roots
Germination chamber	0.94 b	0.57 b
Greenhouse	2.52 a	1.36 a
dms (5%)	0.12	0.15
Length (mm)	Aerial part	Roots
Germination chamber	56.17 b	95.04 b
Greenhouse	76.59 a	253.62 a
dms (5%)	5.27	11.57

***Averages in the same column followed by the same letter do not significantly differ by the Tukey test ($p < 0.05$). dms: value of the least significant difference for the chosen test.**

It is important to emphasize that the emergence, ESI, and mean time of seedling emergence in a greenhouse were underestimated. These parameters were affected by Coleoptera, whose eggs and larvae were transported along with the latrines' organic matter. When insects were found, damage to the plants had already occurred. Studies involving seeds belonging to the family Fabaceae reported the same predation behaviour by Bruchidae (for example, Sari et al. 2002, predation on seeds of *Lonchocarpus muehlbergianus* Hassl. and Arruda et al. 2015, seeds of *Acacia polyphylla* DC. and *Bauhinia cheilantha* (Bong.) Steud.), which was observed in the experiment.

Despite the attack by insects, the organic matter of latrines propitiated seed germination at good rates, demonstrating the beneficial action of ingestion and deposition. Fermentation and increased temperatures in the middle of the faeces, due to the activity of decomposing microorganisms, formed a differentiated thermal environment with higher temperatures. This situation probably affected the germination and initial formation of the cumbaru seedlings in a beneficial way. However, as the days went by, the temperature of the organic matter decreased, reflecting a smaller thermal amplitude than the external environment. The average temperature inside the organic matter was 28.7°C, well within the optimal temperature range standards for the germination of species from tropical regions of 20 to 35°C (Larcher 2004).

The decomposition of organic matter through soil microbiota has an important ecological function in the plant development processes (Ferreira et al. 2017, Moraes et al. 2018). The increase in temperature of the organic matter and the release of organic compounds can enhance with the germination processes (degradation of the integument and breaking of tegumentary dormancy), leading to greater vigor in germination and positively affecting the formation and initial growth of seedlings, as observed in our experiment.

For these reasons, the ingestion of fruits by the species *Tapirus terrestres* is relevant. According to Fragoso & Huffman (2000), tapirs are the last representatives of the Pleistocene neotropical megafauna in the tropical ecosystems of South and Central America, providing an efficient dispersal service for medium, large, and very large seeds not dispersed by other animals. The authors reported that the ingestion of fruits by this species results in high germination rates of seeds, above 65%, depending on the plant species. For this reason, the presence of this mammal is essential for the maintenance of certain plant species, such as cumbaru.

The positive effect of the seed ingestion process by mammals has already been reported by Oliveira & Leme (2013). The ingestion by *Didelphis albiventris* Lund, 1840 significantly increased the germination and average time germination of *Rapanea ferruginea* (Ruiz et Pav.) Mez. seeds. On the other hand, Cáceres & Monteiro Filho (2000), studying *D. albiventris* and *D. aurita* and 14 plant species, achieved significant results only for *Rubus rosifolius* SM., whose seeds collected from faeces had higher germination rates than control seeds. Bizerril et al. (2005) did not find significant differences between germinated seeds of fresh fruits after passing through the digestive system after evaluating the fruits of *Dimorphandra mollis* consumed by *Tapirus terrestris*. These results indicate that the effects of ingestion and digestion of fruits and seeds on breaking dormancy and the germination processes occur differently, depending on the species. Positive interactions are more relevant for certain groups, as observed in this study.

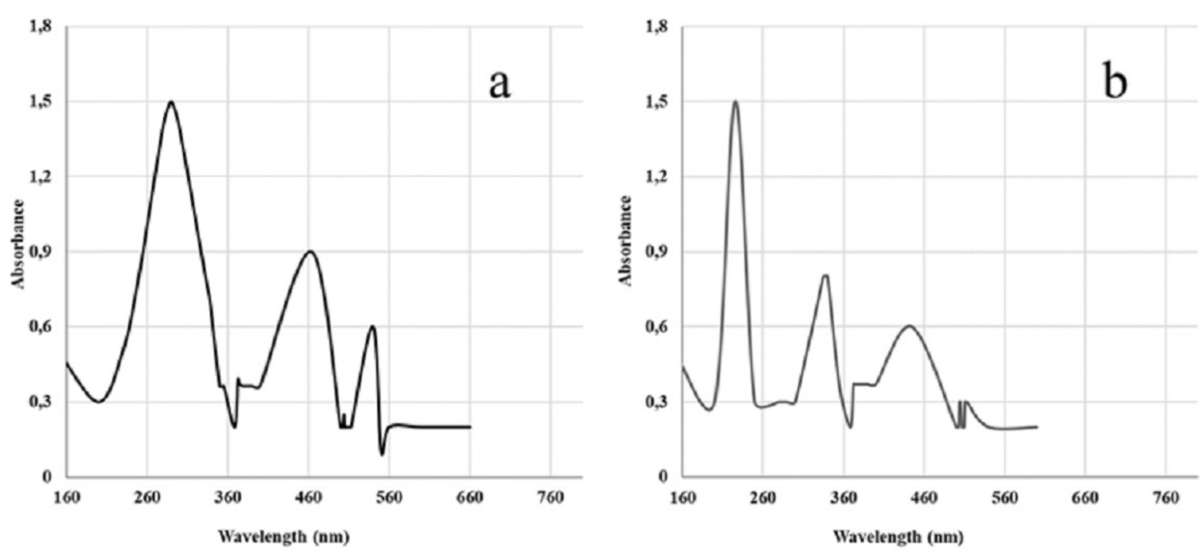

Figure 6. **UV/Vis spectra for humates extracted from the faeces of *Tapirus terrestres.* (a) Humic acids, absorption bands at 290, 464, and 540 nm. (b) Fulvic acids, absorption peaks at 225, 335, and 445 nm.**

Regarding seedling development, seedlings develop better in a greenhouse than in a germination chamber. This situation would be expected, as seedlings grown on organic matter have greater access to nutrients, while plants grown on vermiculite, an inert substrate, only have cotyledons as reserve structures. However, our analysis of the obtained results is complicated due to the insect attack, which consumed parts of the seedlings kept in a greenhouse, and the distinct environments in terms of substrates.

However, the chemical characteristics of the organic matter in faeces do not indicate a substrate rich in nutrients, compared to vermicompost (compost), which have higher ion concentrations (Edwards et al. 2011). Although the nutritional content of faeces does not stand out, the soils of the Cerrado in the region of our collection are composed in part by Quartzaremic Neosols, which have low natural fertility (Frazão et al. 2008). Thus, the addition of organic matter would increase nutrients and provide an adequate pH for the germination and initial growth of seedlings.

The germination of seeds of tree species from the Cerrado in different substrates can normally occur, depending on the species. For example, Bocchese et al. (2008), evaluating the germination of *Tabebuia heptaphylla* (Vell.) Toledo seeds in different substrates, found significant differences between treatments, with sandy soil having a lower percentage of germination due to its physical structure, indicating the need for the addition of organic matter. However, the necessary requirements for germination and establishment are vary depending on the ecological group (pioneers, secondary, or climaxes). Each species may have different behaviors.

An important component for the germination and development of seedlings in faecal organic matter, is the quantification of certain compounds, important for these processes. Humic and fulvic acids are stimulants of seed germination and seedling growth (Canellas et al. 2015). They have a hormonal action similar to auxin, which increases the activity of H^+-ATPase and, consequently, increases protons extrusion. They affect the electrochemical gradient of protons through cell membranes and favour a greater capacity for nutrient absorption by the root system, resulting in a better establishment in the field (Zandonadi et al. 2013). This was observed in the greenhouse experiment with better germination rates and seedling formation in faecal substrates. In contrast, seeds kept in a sandy substrate (greenhouse) did not germinate or showed less development (vermiculite substrate, germination chamber), indicating the beneficial action of acids present in decomposing faecal matter.

According to Eyheraguibel et al. (2008), humic substances derived from organic residues had a beneficial effect on seed germination and the formation of seedlings because of increased plant nutrition, resulting in increased root growth. The results reported by these authors agree with our study. The presence of humic substances in the substrate formed by decomposing faecal matter favoured seed germination and the initial development of cumbaru seedlings.

CONCLUSION

The results obtained indicated the seeds did not suffer damage when passing through the tapir's digestive system, which carried out the process of breaking the tegumentary dormancy by fermentation. The ingestion and subsequent defection provided a higher germination rate than non-eaten fruits, indicating that tapir can be considered a good fruit disperser and germination inducer of cumbaru seeds. The decomposing faecal matter also had beneficial effects on the early development of the species seedlings.

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ADEMIR KLEBER M. DE OLIVEIRA[1]
https://orcid.org/0000-0001-9373-9573

ROSEMARY MATIAS[1]
https://orcid.org/0000-0002-0154-1015

RICHARD MATHEUS FERNANDES[2]
https://orcid.org/0000-0001-8914-6708

JOSÉ CARLOS PINA[1]
https://orcid.org/0000-0001-5414-9386

[1]Programa de Pós-Graduação em Meio Ambiente e Desenvolvimento Regional, Universidade Anhanguera-Uniderp, Rua Ceará, 333, 79003-010 Campo Grande, MS, Brazil

[2]Curso de Ciências Biológicas, Universidade Anhanguera-Uniderp, Rua Ceará, 333, 79003-010 Campo Grande, MS, Brazil

Corresponcence to: **Ademir Kleber Morbeck de Oliveira**
E-mail: akmorbeckoliveira@gmail.com

Author contributions

Ademir Kleber Morbeck de Oliveira: concept, methodology, first writing, statistical analysis of results, and text editing. Rosemary Matias: chemical analysis of organic matter and second draft. Richard Matheus Fernandes: material collection and assembly and monitoring of experiments. José Carlos Pina: material collection and assembly, monitoring of experiments, and statistical analysis of results.