Australian Journal of Crop Science AJCS

AJCS 17(7):563-569 (2023) ISSN:1835-2707
doi: 10.21475/ajcs.23.17.07.p3887

Phenological aspects and physiological quality of seeds of *Mimosa caesalpiniifolia* Benth.

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Abstract

Producing forest species in regions with limiting water conditions is still a challenge to be overcome. Thus, it is necessary and urgent to develop research that measure the influence of water stress on the growth, development and propagation capacity of species. Given the above, the objective of this work was to determine the influence of rainfall pulses on reproductive phenophases, production and vigor of seeds of *Mimosa caesalpiniifolia* Benth. Biweekly flowering and fruiting phenophases were recorded in selected plants in the field, and later these observations were correlated with climatic data from the region. Already the evaluation of the physiological potential was carried out through a germination test in five lots of seeds harvested from the trees of the phenophases study. Generally, it was observed that the sabiá (*M. caesalpiniifolia*) produced seeds between the first half of April and the second half of June, and in the second half of August and the first half of September 2021. The reproductive phenology of the sabiá depends on the extension and frequency of rainfall pulses. The pulse extension was favorable to the physiological potential of the seeds as the germination ranged from 85 to 96%. Therefore, *M. caesalpiniifolia* has its phenology and productive potential influenced by precipitation pulses.

Keywords: Phenophases; Seed vigor; Hydrical stress; Precipitation pulses.
Abbreviations: PL_Rain; TA_Air temperature; FL_Flowering; Fr_Fruiting; PS_Seed production; G_Germination; PC_first germination count; IVG_Germination Speed Index; TMG_mean germination time; VMG_Average germination speed; U_germination uniformity; Z_synchrony index.

Introduction

Native to the caatinga biome, the sabiá (*Mimosa caesalpiniifolia* Benth.) is a forest species belonging to the Fabaceae family and Mimosoideae subfamily (Bezerra et al., 2019). It is a common tree throughout the Northeast region, given its ability to adapt to deeper soils and its ability to survive and regenerate in xerophilic environments (Balbinot et al., 2010).
Being able to reach 10 m in height, and with high regrowth potential, this species presents characteristics that which together with the ability to adapt to dry environments, demonstrates promising potential as a species to be used in reforestation, recovery of degraded areas and income generation for farmers in the Northeast region. Since the *M. caesalpiniifolia* can be used: in the confection of live fences, production of stakes, beekeeping production and forage for ruminants (Bezerra et al., 2019; Souza, 2017), since one of the outstanding characteristics of this species is the high protein content in its leaves.
To produce any plant species, it is necessary to have an effective control of the propagation method, especially when it comes to species that develop in xerophilic environments, and with limiting conditions of water and nutrients, which can directly affect the germination potential of their seeds. To propagate the *M. caesalpiniifolia*, research is needed to measure the effects of soil and climatic conditions on seed production, seeing that in the future may compromise the entire growth and development of plants. For Carvalho and Nakagawa (2012), the temperature, luminosity and the water directly influence seed germination, Nonetheless, the water factor is the most important, since rehydration tissues active and accelerates metabolic activities, causing the supply of of energy and nutrients for embryonic growth, without tell that water also influences all other stages of plant metabolism.
The water stress suffered by the plant, compromises cell growth, reducing its development, in general, affects the physiology of the plant, passing through photosynthesis and respiration, with a negative extension in flowering and fruiting (Bezerra et al., 2019; Ataíde et al., 2014). A plant that suffers water stress, consequently produces seeds with

compromised germination potential. Investigating the influence of water stress and other climatic factors regarding the *M. caesalpiniifolia*, is important to understand the physiological aspects of the seeds, especially when it comes to establishing tolerance and adaptation limits to the species' environmental factors (Bezerra et al., 2019). Monitoring the conditions of precipitation and temperature, correlating with the phenological phases of the plant, and later submitting the seeds produced to germination and vigor tests, is an important step to guarantee the success in the propagation of the chosen species, since when the plants are cultivated in stress conditions, seed production and quality can be compromised. Given the above, the objective of this work was to determine the influence of precipitation pulses on reproductive phenophases, production and seed vigor of *Mimosa caesalpiniifolia* Benth.

Results and discussion

***Reproductive phenology and seed production in** M. caesalpiniifolia*

The flowering and fruiting phenophases of sabiá (*M. caesalpiniifolia)* intensified in April, May, June, July and August 2021, showing Fournier intensity peaks of 80.0 and 90.0, 72.5 and 92.5, 60.0 and 77.5, 57 .5 and 75.0, and 82.5 and 97.5%, respectively, in the monthly intervals of highest rainfall, 331.7, 281.7, 268.7, 195.8 and 206.2 mm, respectively (Fig 1). In this case, these phenophases presented higher percentage index as the precipitation pulses increased in the rainy period. Probably, the physiological and structural responses in the *M. caesalpiniifolia* plants were influenced by the temporal distribution of rainfall. (Lima et al., 2018; Noy-Meir, 1973).

In April, May, June and August 2021, the percentage of individuals of *M. caesalpiniifolia* in flower and fruit activity was 90 and 100 (331.7 mm), 100 and 100 (281.7 mm), 80 and 90 (268.7 mm), and 90 and 90% (206.2 mm), respectively (Fig 2). In turn, the synchronism and magnitude of precipitation pulses were indispensable for the phenological activity of trees during the rainy season, possibly, because these phenophases would be favored by the availability of water and microbiological activity in the soil (Souza et al., 2014; Andrade et al., 2006).

Imposed on temporal variation, and interacting with it, is the spatial variation of rainfall, persistent and random. Random spatial variation can be expressed by the correlation of daily, monthly or annual rains between two seasons. This frequently high spatial variation, both persistent and random, has obvious implications for the interpolation of rainfall records and for input into hydrological models. This variation can hardly be ignored in ecological modeling in arid zones (Parente et al., 2012; Noy-Meir, 1973).

The flowering and fruiting indices were correlated significantly with the rainfall factor, showing that the variations in each phenophase were controlled by the temporal variability the rains in the experimental area (Table 1). Certainly, these results also point to the influence of precipitation interpulses in the reproductive phases, regulating the intensity of production of these phenophases by *M. caesalpiniifolia*.

Opposite to the results found in this work, Nunes et al. (2008) found that the species *Myracrodruon urundeuva* presented the reproductive phenophases influenced differentially by the environmental variables of precipitation and temperature, in the dry season flowering and fruiting occurred. Lima et al. (2018) found that flowering in *Poincianella pyramidalis* occurs in sync with the temporal distribution of rainfall pulses in the area it your occurrence. These last authors reinforce, that the intensity and duration of developmental phenophases in *P. pyramidalis* depend on the amplitude and frequency of precipitation pulses throughout the rainy season.

Souza et al. (2014), studying the phenology of six woody species from the caatinga (*Dipteryx odorata*, *Cordia oncocalyx*, *P. pyramidalis*, *Manihot pseudoglaziovii*, *Handroanthus heptaphyllus* and *Pseudobombax* cf. *marginatum*), in addition to the relationship between phenophases and environmental variables, they found that only D. odorata did not flower. Also according to these authors, the other species flowered in the mid of the rainy season or at the end of it, whereas fruiting usually occurred in mid of the rainy period and at the beginning of the dry season. Precipitation was the factor that most influenced the phenophases of these six species.

For the *M. caesalpiniifolia* activity index, in the same way, the percentage of individuals manifesting the phenophases was significantly correlated with the temporal distribution of rainfall in the experimental area (Table 2). These results confirmed that there is a dependence of *M. caesalpiniifolia* on rain to flower.

Flowering induction represents the beginning of the preparatory stages for seed development. The diversity of the floral structure and the acting of its parts are paramount for vegetable reproduction and activity of the cells responsible for the events that lead to the formation of new individuals. In thit way, the flower contains tissues that contribute to the formation or that will be part of the constitution of the fruit or seed (Marcos-Filho, 2015).

The spatial variation of precipitation (in addition to the redistribution of runoff and edaphic diversity) is one of the causes of irregularity in desert environments, affecting both species diversity and the adaptive behavior of organisms (Andrade et al., 2006; Noy-Meir, 1973).

Parente et al. (2012), evaluating the effect of precipitation regarding the phenology of *Croton sonderianus*, *Caesalpinia pyramidalis*, *Malva sylvestris* and *Aspidosperma pyrifolium* in the caatinga area, they noticed that the precipitation pulses were enough to trigger the phenological events in the species. According to these authors, rainfall interfered with the phenological behavior of the species, triggering flowering and fruiting, which took place in the middle of the rainy period.

Generally, it was observed that *M. caesalpiniifolia* produced seeds between the first half of April and the second half of June, and in the second half of August and the first half of September 2021 (Table 3). It is noted that only biweekly rainfall affected fruit maturation, that is, they were events influenced by the distribution of rains in the experimental area.

Thus, it can be said that the reproductive phenology and seed production in *M. caesalpiniifolia* was interrupted or at low intensity in the months of lower rainfall, such as January to February 2021, 63.8 and 21.4 mm, and October 2021 to January 2022, 36.8, 4.8, 96.0 and 97.0 mm, respectively.

These results corroborate the fact that the species *M. caesalpiniifolia*, possibly, presented growth and development dynamics adjusted to the rain events, regulating the flowering and fruiting seasons for the most favorable conditions for its reproduction (Lima et al., 2018; Souza et al., 2014; Parente et al., 2012).

Table 1. Spearman correlation between the phenological intensity of *M. caesalpiniifolia* and the climatic factors in the period 2021 and January/2022, in the experimental area of CECA.

Phenophase	Climate factor	Spearman Correlation	Probability
Flowering	Rainfall	-0.570	0.000*
	Temperature	0.146	0.293
Fruiting	Rainfall	-0.589	0.000*
	Temperature	0.140	0.095

*Significant at 5% probability.

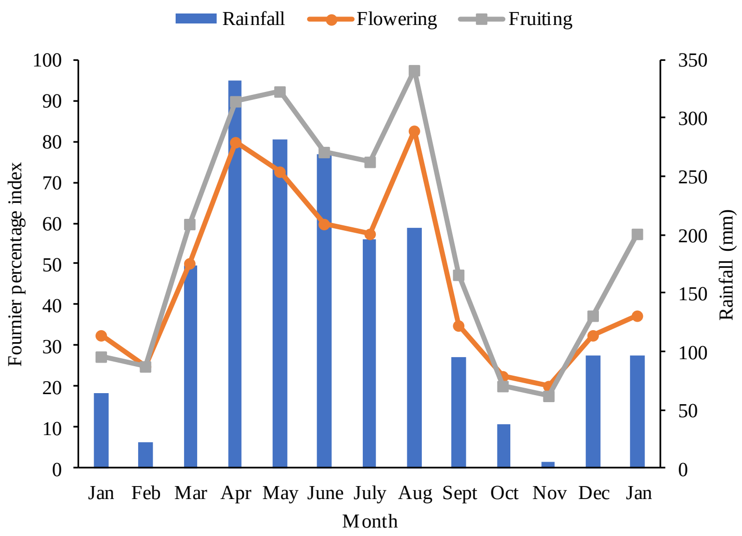

Fig 1. Phenological intensity presented by *M. caesalpiniifolia* and monthly rainfall, in the experimental area of CECA (2021-January/2022).

Table 2. Spearman correlation between the phenological activity of *M. caesalpiniifolia* and climatic factors in the period 2021 and January/2022, in the experimental area of CECA.

Phenophase	Climate factor	Spearman Correlation	Probability
Flowering	Rainfall	-0.507	0.000*
	Temperature	0.173	0.182
Fruiting	Rainfall	-0.626	0.000*
	Temperature	0.232	0.132

*Significant at 5% probability.

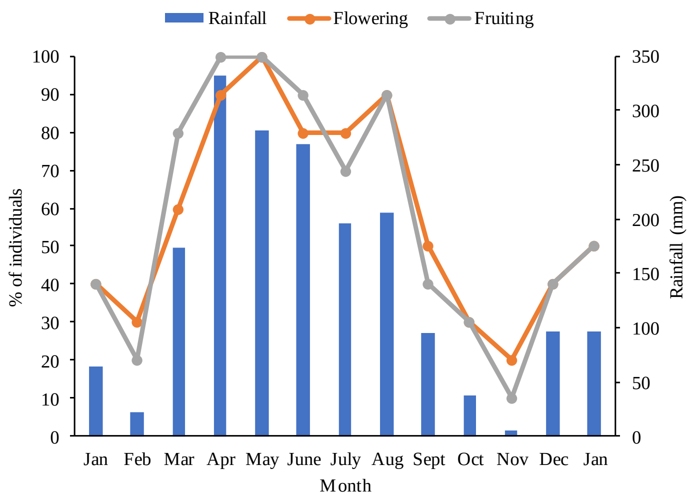

Fig 2. Phenological activity presented by *M. caesalpiniifolia* and monthly rainfall, in the experimental area of CECA (2021-January/2022).

Table 3. Influence of rainfall pulses on reproductive phenophases in *M. caesalpiniifolia*, from January 2021 to January 2022.

Month	climate factor		Fournier percentage index		% of individuals		PS
	PL (mm)	TA (ºC)	Fl	Fr	Fl	Fr	
Jan	63.8	25.3	32.5	27.5	40	40	
Feb	21.4	28.3	25.0	25.0	30	20	
Mar	174.2	25.8	50.0	60.0	60	80	
Apr	331.7	25.2	80.0	90.0	90	100	Lot 1
May	281.7	24.3	72.5	92.5	100	100	Lot 2
June	268.7	23.3	60.0	77.5	80	90	Lot 3
July	195.8	22.9	57.5	75.0	80	70	
Aug	206.2	22.6	82.5	97.5	90	90	Lot 4
Sept	95.5	23.9	35.0	47.5	50	40	Lot 5
Oct	36.8	24.9	22.5	20.0	30	30	
Nov	4.8	26.3	20.0	17.5	20	10	
Dec	96.0	26.2	32.5	37.5	40	40	
Jan	97.0	25.9	37.5	57.5	50	50	

PL: PL: Rain; TA: Air temperature; FL: Flowering; Fr: Fruiting; PS: Seed production.

Table 4. Germination (G), first germination count (PC), Germination Speed Index (IVG) and mean germination time (TMG) of *M. caesalpiniifolia* seeds harvested in the 2021 and January/2022 seasons.

lots of seeds	G (%)	PC (%)	IVG	TMG (days)
Lot 1	88 bc	78 b	2.443 c	5.1 bc
Lot 2	85 c	76 b	1.380 c	7.3 d
Lot 3	96 a	87 a	7.199 a	3.5 a
Lot 4	90 b	79 b	1.491 c	6.2 cd
Lot 5	91 b	85 a	4.915 b	4.1 ab
Average	90	81	3.485	5.2
CV (%)	5.73	12.35	5.74	3.17

The means followed by the same letter in the column do not differ statistically from each other by the Tukey test at the 5% significance level.

Table 5. Average germination speed (VMG), germination uniformity (U) and synchrony index (Z) of *M. caesalpiniifolia* seeds harvested in the 2021 and January/2022.

lots of seeds	**VMG (dias^{-1})**	**U (*bit*)**	**Z**
Lot 1	0.20 c	1.8133 b	0.2383 bc
Lot 2	0.14 d	2.3629 c	0.1107 c
Lot 3	0.28 a	1.1481 a	0.4328 a
Lot 4	0.16 d	2.1583 c	0.1853 bc
Lot 5	0.24 b	1.5974 b	0.3030 ab
Average	0.20	1.8160	0.2540
CV (%)	2.20	2.65	10.24

The means followed by the same letter in the column do not differ statistically from each other by the Tukey test at the 5% significance level.

On the other hand, the reproductive phenology and fruit production outside the rainy season could be an advantage for the establishment of this species, as the seeds would produce plants that would find ideal conditions for their development at the end of the dry season and beginning of the rainy season (Oliveira et al., 2017).

Even with a small diversity of species in the experimental area, it was possible to identify different phenological patterns in the same community, mainly with regard to the flowering and fruiting phenology of *M. caesalpiniifolia*. The detailed study and the volume of knowledge accumulated on the subject provide basic information for the determination of physiological maturity and the most suitable time for harvesting *M. caesalpiniifolia*. Generally, spatio-temporal variations in seed production were determined by the distribution of rains.

Germination and vigor of** M. caesalpiniifolia **seeds

The seed lots of *M. caesalpiniifolia* produced about the influence of different precipitation pulses, showed significant differences among themselves, for almost all parameters evaluated. The seeds from Lot 3 (collected on June 15, 2021) showed greater physiological potential, with germination of 96%, first germination count of 87%, Germination Speed Index of 7.199 and average germination time of 3.5 days. On the other hand, the seeds of Lot 2 (collected on May 15, 2021) had lower physiological potential, with germination of 85%, first germination count of 76%, Germination Speed Index of 1,380 and average germination time of 7 .3 days (Table 4).

It was also found that the seeds of Lot 3 had an average germination speed of 0.28 days^{-1}, it was also found that the seeds of Lot 3 had an average germination speed of 0.28 days–1, germination uniformity of 1.1481 *bit* and a synchrony index of 0.4328, whereas Lot 2 and Lot 4 (collected on August 15, 2021) presented in lowest average germination speed and the lowest germination uniformity, Lot 2 still had the lowest synchrony index (Table 5).

The statistical differences presented between the evaluated seed lots, may be related to the influence of the different precipitation pulses, in which the mother plant was submitted in the field. According to Marcos Filho, (2015), both the size and the physiological potential of the seeds are affected by the environmental conditions in which the mother plant was exposed during the productive phase. For Gomes et al. (2017), it is possible that there are variations in the size of the seeds of the same mother plant in the same harvest or different harvest. These last authors verified that the size of the seeds of *Anadenanthera colubrina* (Vell.) Brenan produced in the caatinga was mainly influenced by precipitation and temperature, in which the mother plant was subjected. And as Marcos Filho, (2015) reinforces, seeds with distinct sizes may have different germination and vigor. According to Carvalho and Nakagawa, (2012), larger seeds tend to be more vigorous, as they have a greater amount of reserve substances, and consequently originate more vigorous plants and seedlings. The percentage of germination found in Lot 3 was the same as that verified by Passos et al. (2007) in seeds of *M. caesalpiniifolia*. However the dormancy breaking used in this research was the integumentary, already these authors submitted the evaluated seeds to chemical treatments to break dormancy. The aforementioned authors also verified that as the water restriction increased in the substrate, there was a reduction in the germination of *M. caesalpiniifolia*. This can also be observed in the field, as when the plant is subjected to shorter precipitation pulses, seed formation can be compromised, and this can also reflect on the physiological potential of the seed.

The germination and vigor of a seed lot depends on several factors, the most important of which are related to the plant, such as physiological maturation and the dormancy, and those related to environmental conditions, temperature, water and available light. However, water plays a fundamental role in all the metabolic processes involved in the maturation of orthodox seeds, insofar as thee seeds pass from a metabolically active state (development) to an inactive state after maturity (thanks to the effect of desiccation), returning later to the metabolically active state during germination (Marcos-Filho, 2015). The differences between seed lots, may also be associated with the fact that the flowers of the same plant or the same inflorescence are not pollinated or fertilized simultaneously, so that the complete uniformity of development of the seeds formed in an individual should never be expected; this situation is accentuated when considering a population of plants (Marcos-Filho, 2015). Maturation and drying also influence the germination potential of the seed, Nogueira et al. (2013) found that when the seeds were dried while still in the plant, the germination potential reached 95%, a value equal to that found in Lot 3. The seeds analyzed in this research were also dried under the same conditions as in the aforementioned study. It is very important to know the factors that affect the germination of forest seeds, only in this way it will be possible to control them or even to conduct them to improve the performance in the production of seedlings. However, in Brazil there is still a shortage of research and materials, that standardize germination tests, especially when it comes to a country with such a variety of forest species.

Material and methods

Place of conducting the experiments

The experiments were carried out in the field and at the Laboratory of Phytotechnics, to the Campus of Engineering and Agrarian Sciences (CECA), from the Federal University of Alagoas (UFAL), both located in the municipality of Rio Largo, AL, Brazil. The climatological data (January/2021 to January/2022) used in the research were provided by the Laboratory of Irrigation and Agrometeorology (LIA), through the Automatic Agrometeorological Station, CECA/UFAL, Rio Largo-AL. Latitude: 9º28'29.1''S; Longitude: 35º49'43.6''W; Altitude: 127.0 m.

Record of phenophases of *M. caesalpiniifolia*

The trees selected for phenological side dish stay located in an experimental area of CECA/UFAL, Rio Largo, AL. That area has the following geographic coordinates: Latitude: 9º27'57''S; Longitude: 34º50'1''W; Altitude: 127.0 m. According to the Köppen climate classification, the climate of the region is type As, humid coastal tropical (Cardim, 2003). In total, ten plants were selected, with approximately the same height, with rectilinear trunks, abundant crowns and good phytosanitary conditions (apparent absence of diseases and parasite infestations), as suggested by Nunes et al. (2008).

Phenological data were recorded between the first half of January 2021 and the second half of January 2022. At fortnightly intervals, the presence and absence of flowering and fruiting phenophases were recorded. To identify the beginning and end of flowering and fruiting, the criteria defined by Lima et al. (2018).

To determine the Fournier intensity index (1974), were obtained phenological data in the field, through a semiquantitative interval scale of five categories (0 to 4), with an interval of 25% between each category. Fortnightly, the values obtained from all individuals were added and divided by the maximum possible value (number of individuals multiplied by four). That value corresponds to a proportion was then multiplied by 100, to transform it into a percentage value (D´Eça-Neves and Morellato, 2004).

To determine the presence or absence of phenophase and to estimate synchrony between individuals in the population, the activity Index was used (Morellato et al., 1990). Every 15 days, all individuals exhibit phenophase were summed; this number was multiplied by 100 and divided by the maximum possible value (number of individuals), and the result was obtained in percentage values.

The phenological data of intensity and activity of each phenophase (flowering and fruiting) were correlated with the precipitation and temperature average fortnightly through the Spearman correlation (Zar, 1996).

Evaluation of the physiological potential of seeds of *M. caesalpiniifolia*

The seeds of *M. caesalpiniifolia* used in that experiment, were harvested from the trees where the phenophases were recorded. During that record, the trees of sabiá presented five fruiting periods, the seeds produced were harvested and separated by lots (lot 1, lot 2... lot 5), each lot corresponds to a production period. The fruits were harvested with the aid of aerial scissors with an extension handle, during the ripening period, characterized by their opaque brown color with a slightly rough surface (Freitas et al., 2013). The seeds were manually extracted and processed, discarding the badly formed ones, attacked by insects or fungi, and then, packed in kraft paper bags and stored in a dry chamber under temperature of ± 20 ºC and relative humidity between 50 and 55 %, for approximately 60 days until the germination test was performed (Pereira et al., 2020).

For the evaluation of the physiological potential, a germination test take place out in the laboratory, where the experimental design used was the completely randomized design (DIC), in total five lots of seeds of *M. caesalpiniifolia* were evaluated, each lot with four replications, and each repetition was composed of 25 seeds as determined by Brasil, (2013).

The seed asepsis, was performed by immersion in a detergent solution (5 drops for every 100 mL of distilled water) for 5 minutes, followed by washing in running water (Brasil, 2013). Before the germination test, the break of the integumentary dormancy of the seed was carried out, making the integument cut with scissors in the region opposite the hilum (Avelino et al., 2018; Brasil, 2013).

After dormancy breaking, the seeds were willing between two sheets of germitest paper moistened with a volume of water in an amount equivalent to 2.5 times the weight of the substrate, then these sheets were rolled, placed in plastic bags and then in the *Biochemical Oxygen Demand* chamber, regulated at 25 °C and photoperiod of 12 hours for nine days (Brasil, 2013).

Were realized germination counts daily (G%), after the beginning until when did it happen the stabilization of the number of germinated seeds (Brasil, 2009). The first count test (PC%) was conducted together with the germination test, computing the percentage of normal seedlings at five days after sowing (Brasil, 2013). The Germination Speed Index (IVG) was also calculated for each treatment according to the formula proposed by Maguire, (1962).

The mean germination time (AMR) was obtained by the formula $t = \Sigma ki$=1(niti)/Σ ki=1ni, where ti: time from start of experiment to i nth observation (days or hours); ni: number of seeds germinated in time i (number corresponding to the i nth observation); k: last day of germination (Czabator, 1962). The mean speed of germination (VMG) was calculated through the formula $v = 1/t$, with t being the mean germination time (Ranal and Santana, 2006).

To obtain the germination uniformity (U), the formula U = −ΣFrlog2Fr was used, being Fr is the germination frequency and log2 – logarithm at base 2 (Labouriau and Valadares, 1976; Labouriau, 1983). For the synchrony index (Z), the formula Z = ΣCn1,2/N ≍ Cn1,2 = ni(ni–1)/2 adopted; N = Σni(Σni–1)/2, where Cn1,2 the combination of the seeds germinated at the i nth time and ni the number of seeds germinated at the time i (Primack, 1980).

Statistical analysis

The data referring to the evaluation of the physiological potential of the seeds of *M. caesalpiniifolia*, were submitted to Analysis of Variance and the means were compared by the Tukey test at 5% probability, using the Sisvar software (Ferreira, 2014).

Conclusion

Reproductive phenology in *M. caesalpiniifolia* depends on the length and frequency of rainfall pulses. The Fournier percentage index and the activity index for the flowering and fruiting phenophases of *M. caesalpiniifolia* are synchronized with the temporal distribution of rain pulses in the experimental area. *M. caesalpiniifolia* presents intricate ecological relationships with thisof seasonality accented habitat, and this affects the physiological aspects of the seeds produced. The extension of the precipitation pulses, is favorable to the physiological potential of the seeds of *M. caesalpiniifolia*.

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