English Version

Abstract

The objective of this work was to characterize and verify the effect of different phases of fruit ripening and its relationship with germination and dormancy resulted from tegument impermeability of surucucu seeds. Eight fruit collections were accomplished at Vitória da Conquista - Bahia, in the period from May to July of 2007, in intervals of seven days. Each harvest period was characterized by evaluations of water content, dimensions of the seeds and Spad index of pod and seeds. After a period of 59 days of seed collection seeds were submitted to germination test. The trial was defined by a completely randomized design with an experimental grid of 50 seeds. The greening intensity of pod and seeds, seed dimensions and seed number per pod were reduced as maturity age increasing. The germination raise associated to the periods after full fructification was indirectly related to dormancy and water content of seeds.

Key words: Harvesting time; Impermeability of seed coat; Surucucu.

Pod development and seeds maturation of *Piptadenia viridiflora* related to germination and dormancy[1]

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Introduction

Piptadenia viridiflora (Kunth) Benth is a rustic plant, fast growing, belonging to the family Mimosaceae, occurring in areas of Northeast of Brazil and in Pantanal of Mato Grosso. Despite the aggressiveness of their thorns, this is one of the most promising species for the establishment of forests for multiple uses, i.e. for the production of firewood and charcoal, because it has a high density of wood. It is used as fuel in the ceramic industry in the Southwestern region of Bahia and as charcoal to meet the steel park in Minas Gerais. The introduction of this species has been shown in studies of forest ecosystem restoration, enhancement of disturbed areas, rehabilitation of degraded areas and segments related to the energy matrix, since the species presents a management system suitable for different cutting cycles.

Piptadenia viridiflora is a species with great importance as a component in agroforestry systems, due to its high capacity related to nitrogen fixation. According to Freitas et al. (2006), species of the genus Piptadenia and Mimosa reach rates between 27 and 85% contribution as components of nitrogen fixers in bioma of caatinga in Brazil.

For the feasibility planting of this specie, it is important to adequate the management techniques for production of seedlings. Studies of propagation, mainly through the seed are essential, therefore, many legumes are restricted from spreading since the occurrence of dormancy.

According to Nascimento et al. (2009), the dormancy of legume seeds is a hereditary character, attributed to the palisade layer of cells whose walls are thick and covered externally by a waxy cuticle layer. The impermeability of the tegument can restrict the absorption of water, oxygen, and result in a physical resistance to the embryo development. Chevalier et al. (2007) found that the impermeability of the seed coat of *Colubrina glandulosa* was related to the speed restriction of imbibition. Benedict et al. (2008) noted the occurrence of dormancy for the coat impermeability in seed of *Piptadenia moniliformis* Benth, resulting in restriction of 37% germination of the seeds analyzed. The occurrence of seed coat

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dormancy has a great ecological importance because it allows the rapid colonization of areas devasted by fire and promotes high temporal and spatial dispersion (SILVEIRA FERNANDES, 2006).

Due to the scarcity of information about this species, the objective of this work was to study the relationship between the morphological and chromatic pods and seeds of the stages of fruiting and incidence of dormancy by impermeability of seed coat of *Piptadenia viridiflora* (Kunth.) Benth.

Material and methods

The experiment was conducted during May-July 2007 in Southwestern of Bahia, Vitoria da Conquista, characterized by predominantly Semi-arid climate, with temperatures averaging 20 °C and annual rainfall index of 900 mm, concentrated in a period from November to March.

It was identified and marked 25 trees of *Piptadenia viridiflora* selected by the vigor and the health condition, with heights ranging from 5 to 8 m and age about 10 years. The stage of flowering occurred in the first half of February and the fruit collection was initiated in mid-May, a time which, by visual observation, the selected trees had full pod formation.

One time in each week, pods were hand harvested and immediately after, they were packed in paper and sent to the Laboratory of Seed Analysis, State University of Southwest Bahia, Campus de Vitória da Conquista. Data about dimensions of pods and seed, weight, electrical conductivity and the seed humidity was recorded. After that, seeds together with pods were kept in paper bags.

It was also determined relative chlorophyll content using portable chlorophyll meter, SPAD units (SPAD, Minolta, Japan). Data was obtained from four samples per collection, each sample consisting of ten pods.

The germination test followed a completely randomized design scheme with four replications and 50 seeds per plot. The treatments consisted of eight stages of maturation, defined by samples taken at intervals of seven days, from full pod formation.

The moisture content was determined by the method of the greenhouse at 105 ± 3 °C, for 24 hours (BRASIL, 1992). After the drying period, samples were placed in desiccator and then weighted in analytic scale with precision of 0.001 g. At the time of test installation, the moisture of all samples was maintained between 12 to 13%. For the germination test the seeds were placed in rolls, consisting of three sheets of paper germitest moistened with deionized water at a ratio of two times the dry weight of paper, according to standard procedures. The rolls were placed in plastic bags in an incubator, set at a temperature of 25 °C and a photoperiod of eight hours of light. The germination percentage and the incidence of hard coat seed were evaluated on the tenth day after sowing, and the data subjected to analysis of variance.

All these characteristics were related to the periods of seed harvest and the mathematical models were defined by F test of regression and by T test of coefficients, biological significance and value of the coefficient of correlation. For the analysis of germination and hard coat seeds, the data were transformed into arcsine $\sqrt{(x/100)}$. For all analysis of data it was used SAEG 9.1 software (RIBEIRO JÚNIOR, 2001).

Results and discussion

It was not possible to establish a model for the relation between the SPAD index and pod collection (Figure 1A), due to the null values of this index verified in latest samplings. For the first three collections, it was verified the homogeneity of the SPAD index for the pods, whose color was characterized as reddish green (Figure 1a). From the fourth harvest (21 days after full fruiting), when the pods had a grayish green color, there was a drastic reduction in the SPAD index. With the progress of the maturation process, from the fifth collection, the pods have suffered a dramatic darkening and then became black. Thus, it was not possible to obtain readings from SPAD index, due to the degradation of chlorophyll in the pods.

Pod color is an important feature to determine seed physiological maturity (NAKAGAWA et al. 2007c, GUIMARAES and BARBOSA, 2007, AGUIAR et al, 2007). Evaluating the morphological characteristics of fruits of *Sesbania virgata* (Cav.) Pers, Araujo (2004) found that the pod maturation

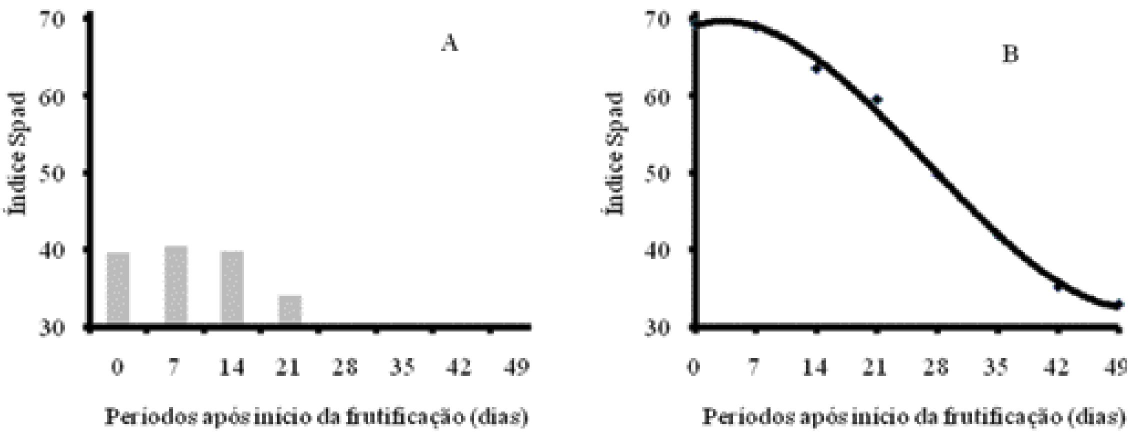


Figure 1. Spad Index of pods (A) and seeds (B) of *Piptadenia viridiflora* evaluated in a harvest time at different stages from beginning of fructification.

of this species was related to color, distinguishing green when young, opaque brown when ripe. For the alterations of SPAD index of seed it was set the quadratic model (Figure 1b), occurring an indirect relation between age of pods and intensity of green color of the seeds. Unlike the pods, the green color of the seeds was maintained during the whole period of evaluation, ranging from 69.4 to 32.9.

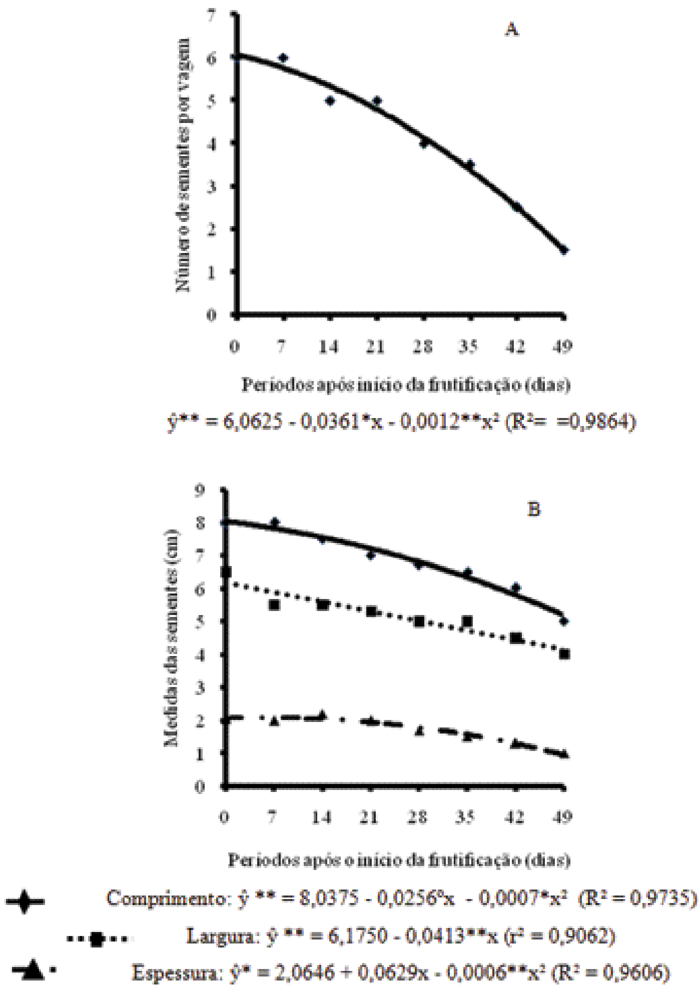
For the number of seeds physically intact per pod it was verified a quadratic adjustment of the model in relation to sampling periods (Figure 2a). The greatest number of seeds (six seeds) was observed in the first sampling, occurring gradual decreases in subsequent stages of fruiting.

In the last picking, with a period of forty-nine days after the onset of fruiting only 1.5 seeds per pod were recorded. Two factors have contributed effectively to reduce the number of seeds in the last collection: susceptibility to insects and microorganisms, and dehiscence of the seed. *Piptadenia viridiflora* is a species considered as cyanotic, with elevated levels of these compounds accumulating in leaves and fruit. According Tokarrnia et al. (1994) during early fruiting, the cyanogenic compound is high, resulting in a lower incidence of attack by insects and microorganisms, thus constituting an important ecological characteristic for dissemination. Menezes et al. (2010) found that damage to pods of *Mimosa bimucronata* trees could affect the remaining seeds when predation occurs during the fruit ripening. This effect was related to the lower Nitrogen content in seeds from pods predated.

In this study, due to the opening of pods, the natural dehiscence of seeds occurred from the fifth collection. From this it was verified an accentuated decrease of seed number in pods. For seeds of *Mimosa caesalpiniifolia* Benth , Alves et al. (2005) found that the period of seed collection was limited by the high loss of seed dispersal in the last stages of development, resulting from the natural opening of the pods.

A quadratic model was adjusted for seed length and thickness and a linear model for seed width (Figure 2b). There was a decreasing in length, width and thickness of seeds in relation to the collection periods, with variation of values from 8 to 5.5 mm in length and 6.2 to 4.5 mm for the width and also a reduction from 2.0 to 1.0 mm to thickness.

For the initial samplings there was a strong effect of seed water content to anatomic features such as increased length, width and thickness. The dimensions of the seeds reached the maximum values when the water content was above 70%. Although the reducing of the water content occurred concomitantly with the decrease of seed dimensions it was observed the maintenance of seed dry matter in the sampling dates (data not shown). Thus, changing the size of



***, *, ° Significant at 1%, 5% and 10% probability, respectively, according to the f test to the model and for the second t-test for the coefficients.*

Figure 2. Number of seeds per pod (A) and seed dimensions (B) from *Piptadenia viridiflora* valued at collection, with different stages of fruiting.

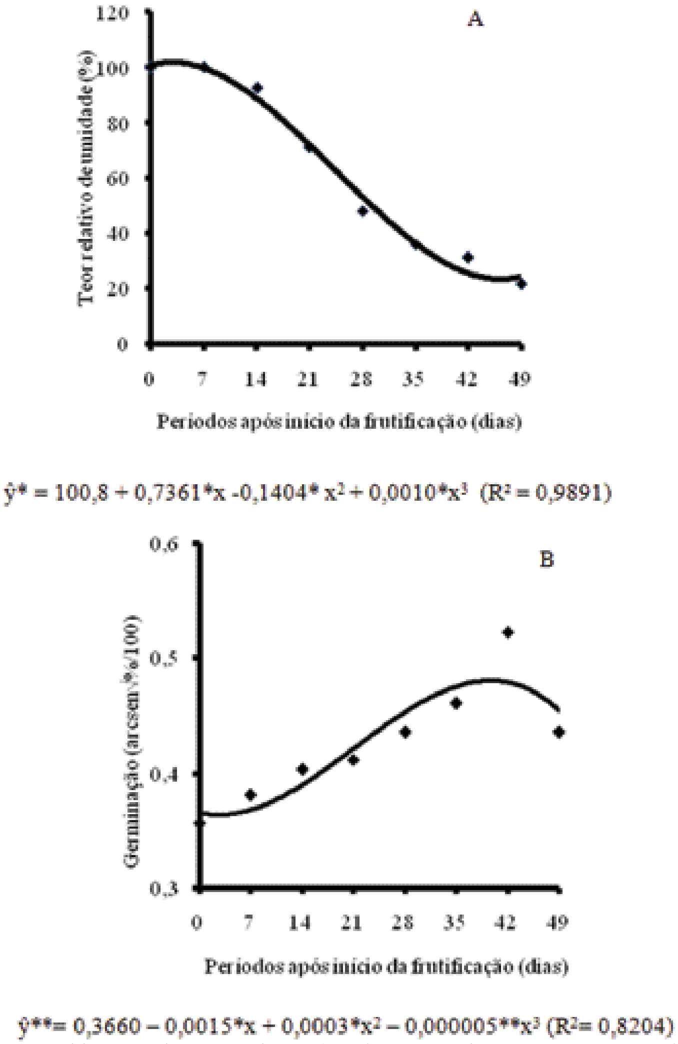
the seeds due to volume reduction enable greater dispersal ability, without being associated to a reduction in storage material restricting the "seedling size effect" (SSE), described by Laboreau et al. (2006).

For the relation between the different periods after fruit set and reducing the water content it was adjusted a cubic model (Figure 3a), defined by a decreasing behavior in relation to periods of fruiting. The highest values were recorded in the first three samples (70.0%, 69.6% and 65.4%) respectively. It was found that the water content of seeds remained stable in the first two collections, decreased thereafter until the seventh collection, and from this point, had some stability. The initial high moisture content found in seeds of first collections and the subsequent decline was related to the importance of water in the processes of seed formation and maturation. As it can be seen in Figure 3 a and 3 b, the water content in relation to sampling dates showed an inverse behavior compared to percentage of seed germination.

According to Carvalho and Nakagawa (2000), to make suitable the photosynthates of leaves to the seed formation as structural material and later as storage, initially it is necessary to maintain a high degree of humidity, which occurs even the dry weight reaches its maximum value, when it begins a rapid dehydration. In the final stages of maturation of the pods of velvet bean seeds, Nakagawa et al. (2007b) verified that reduction the water content of velvet bean seeds was concomitant with the increasing of dormancy by coating impermeable.

Percentage of seed germination and incidence of hard coat dormancy were influenced by stage of pod development which was defined by the time of collections, as shown in Table 1.

The percentage of germination evaluated at 10 days was adjusted to a cubic model (Figure 3b). The highest value of the germination (17.98%) was observed at 37 days after first collection date, later occurring a decreasing, reaching a minimum of 13% for seeds harvested at 49 days after fruiting. The high incidence of seed coat impermeability provided a low germination rate of seeds, mainly in the initial harvesting time of pods (Figure 4). For



***, * Significant at 1% and 5% probability, respectively, according to F test for the regression model, and the t-test for the coefficients.*

Figure 3. Relative moisture content evaluated at time of pods collection (a) and germination of *Piptadenia viridiflora* at 10 days after beginning the experiment (b) for different fruiting stages.

Table 1. Mean squares of the variance analysis of germination percentage at 10 days after beginning of experiment and percentage of hard coat seeds of *Piptadenia viridiflora*, from pods collected to each eight stages of maturation.

	DF	Germination	Hard coat seed
Harvesting time	7	0,008041*	0,03523**
Error	24	0,004536	0,004404
Total	31		
CV(%)°		13,982	9,172

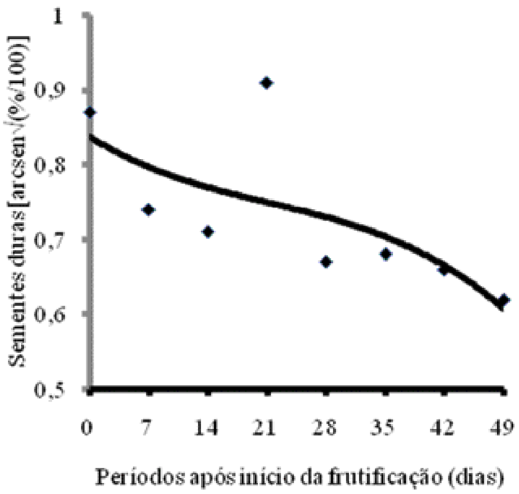
***, * **, * Significant at 1% and 5% probability, respectively, according to F test*

° Coefficient of variation

the relation between sampling dates after fruiting and percentage of hard seeds it was defined the cubic model. Cavalheiro et al. (2007) verified the restrictive effect of coat impermeability in seeds of *Colubrina glandulosa*. In the referred study, even after overcomming dormancy the time index of germination remained high.

The highest percentage of hard seed occurred in the first collection with a percentage of 55.71%, and the lowest at 49 days after the onset of fruiting (31.92%). Therefore, in last collections, because the seeds reach an advanced stage of maturation, the mechanism of dormancy related to hard coat was expressed with less intensity. A similar phenomenon was described by Nakagawa et al. (2007c), for seeds of mucuna preta; in that study the drying of immature seeds inside the pods resulted in higher incidence of impermeable tegment compared to the seeds from mature pods.

In contrast to that observed in this study, Alves et al. (2004) found for seeds of *Mimosa caesalpiniifolia* Benth. that the occurrence of impermeability of seed coat for germination was restrictive only at 154 days after the beginning of anthesis. For seeds in formation, in the lowest stage of seed coat development there is a greater permeability, which



$$\hat{y}^{**} = 0,8426 - 0,007540^{*}x + 0,0002^{*}x^2 - 0,000003^{**}x^3 \ (R^2 = 0,5211)$$

°Original data transformed into arcsine √(x/100) and data with zero value √(x + 0.5).

***, * Significant at 1 and 5% probability by test F for the regression model and, according to t test for the coefficients.*

Figure 4. Percentage of hard coat seeds° of *Piptadenia viridiflora* in different stages of pod maturation after collection from the full fruiting.

reduces the incidence of hard seeds. In this study, the collections began when the pods were already fully formed and therefore the structure of the seed coat was in an advanced stage, differently of a circunstance described by Alves et al. (2004).

Factors such as rising temperatures and reduction on water availability were related to stress induction resulting in high occurrence of hard coat seeds in later periods of pods maturation. According to Nakagawa et al. (2007a), who carried out a study on velvet bean seeds, the percentage and intensity of the hard coating depend on seed maturation stage when the drying starts. The harvesting and drying of immature pods can restrict the deposition of lignin in the outer integument of the seed and, according to Cavariani et al. (2009) may restrict the speed of seed water absorption.

Conclusion

The portable chlorophyllmeter is an appropriate tool to characterize the chromatic variations of pod and seeds of *Piptadenia viridiflora*.

The germination of *Piptadenia viridiflora* seeds was enhanced by sampling in more advanced stages of pods maturity when it had a dark color.

The opposite behavior occurred for the hard seed, with the highest percentage taking in seeds of pods harvested in the early stages, characterized by green color.

There is a requirement a fine-tuning of this study, related to a major period of observations especially in the early stages of pod formation and to evaluating of germination after outcomming dormancy.

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References

AGUIAR, F. F. A. et al. Maturação de frutos de *Caesalpinia echinata* Lam., pau-brasil. **Revista Árvore,** Viçosa, v. 31, n.1, p. 1-6, 2007.

ALVES, E. U. et al. Dormancy and development in "sabiá" (*Mimosa caesalpiniifolia* Benth.) seeds. **Revista Árvore**, Viçosa, v.8, n.5, p.655-662, 2004.

ALVES, E. U. et al. Maturação fisiológica de sementes de sabiá. **Revista Brasileira de Sementes**, Brasília, v. 27, n.1, p.01-08, 2005.

ARAUJO, E. C. de et al. Caracterização morfológica de frutos, sementes, e plântulas de *Sesbania virgata* (CAV.) Pers. **Revista Brasileira de Sementes**, Brasília, DF, v.26, n.1, p.105-110, 2004.

BENEDITO, C. P. et al. Superação da dormência de sementes de catanduva (*Piptadenia moniliformis* Benth.). **Revista de Ciência Agronômica**, Fortaleza, v.39, n.01, p.90-93, 2008.

BRASIL. Ministério da Agricultura e da Reforma Agrária. **Regras para análise de sementes.** Brasília: SNDA/DNDV/CLAV, 1992. 365p.

CARVALHO, N. M.; NAKAGAWA, J. **Sementes: ciência, tecnologia e produção.** 4ª. Edição. Jaboticabal: Funep, 2000. 588p.

CAVARIANI, C., et al. Velocidade de hidratação em função de características do tegumento de sementes de soja de diferentes cultivares e localidades. **Revista brasileira de sementes**, Brasília, v.31, n.1, p.30-39, 2009.

CAVALHEIRO, A. L.; PIMENTA, G.A.;TOREZAN, J. M. D. Effect of some physical and chemical treatments on germination of *Colubrina glandulosa* seeds. **Seed Science and Techonology**, v.36, n.3, 744-748, 2007.

FREITAS, A. D. S. et al. Biological nitrogen fixation in tree legumes of the Brazilian semi-arid caatinga, **Journal of Arid Environments,** v.74, n.3,p.344-349, 2010.

GUIMARÃES, D. M., BARBOSA, J. M. Coloração dos frutos como índice de maturação para sementes de *Machaerium brasiliense* Vogel (Leguminosae – Fabaceae). **Revista Brasileira de Biociências**, Porto Alegre, v.5, supl.2, p.567-569, 2007

LAHOREAU, G. et al. Positive effect of seed size on seedling survival in fire-prone savannas of Austrália, Brazil and West Africa. **Journal of Tropical Ecology**, v.22, n.6, p.719-722, 2006.

LORENZI, H. **Árvores brasileiras:** manual de identificação e cultivo de plantas arbóreas nativas do Brasil. 3^0. ed. Nova Odessa: Instituto Plantarum, 2000. v.1. 351p.

MENEZES, L. C. R., KELIN, J., KESTRING, D., ROSSI, M. N. Bottom-up and top-down effects in a pre dispersal seed predations system: are non-predated seeds damaged? **Basic and applied ecology**, v.11, p. 126-134, 2010.

NAKAGAWA, J. *et al.* Dormancy degree during velvet bean seeds maturation. **Revista Brasileira de Sementes**, Brasília, v. 29, n. 1, p. 165-170, 2007a

NAKAGAWA, J. et al. Maturação de sementes de mucuna-preta. **Bioscience Journal,** Uberlândia, v. 23, n.1, p.41-47. 2007b

NAKAGAWA, J. et al. Viabilidade de sementes de mucuna-preta em função do tamanho, da maturação e da secagem. **Acta Scientiarum Agronomy**, Maringá, v.29, n.1, p.107-112, 2007c.

NASCIMENTO, I. L. et al. Superação da dormência em sementes de faveira (*Parkia platycephala* Benth). **Revista Árvore,** Viçosa, v.33, n.1, p.33-45, 2009.

RIBEIRO JÚNIOR, J. I. **Análises estatísticas no SAEG**. Viçosa: UFV, 2001. 301p.

SILVEIRA, F. A. O., FERNANDES. G. W. Effect of light, temperature and scarification on the germination of *Mimosa foliolosa* (Leguminosae) seeds. **Seed Science and Technology**, v.34, p.306-614, 2006.

TOKARRNIA C. H., DÖBEREINER J. E PEIXOTO P. V. Aspectos clínico-patológicos complementares da intoxicação por algumas plantas tóxicas brasileiras. **Pesquisa Veterinária Brasileira**, Rio de Janeiro, v.14, n.4, p.111-122, 1994.