GERMINATION AND VIGOR OF Dimorphandra mollis *BENTH. SEEDS UNDER DIFFERENT TEMPERATURES AND SUBSTRATES*[1]

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ABSTRACT – The Dimorphandra mollis Benth. – Caesalpiniaceae is a native forest species coming from the Cerrado and Caatinga due to its economical and ecological use, which justifies the studies on seed germination. In this work, germinative performance of D. mollis seeds were studied in different conditions of temperature regime and substrate. The experimental delineation used was completely randomized in factorial 4 x 4 (4 substrates –sand, coconut fiber, vermiculite and paper towel; and 4 temperatures: 25, 30, 35 and 20-30ºC), with four replications of 25 seeds each. The following parameters were evaluated: seed moisture content, final germination, first germination count, germination speed index, length and dry matter weight. The best germination and vigor is obtained at 30 and 35ºC. The substrates paper towel and vermiculite allow satisfactory germinative performance of seeds, being suitable to evaluate the physiological quality of D. mollis seeds.

Keywords: Forest seeds, Physiological quality and Fava d'anta.

GERMINAÇÃO E VIGOR DE SEMENTES DE *Dimorphandra mollis* BENTH. SOB DIFERENTES TEMPERATURAS E SUBSTRATOS[1]

RESUMO – A fava d'anta (*Dimorphandra mollis* Benth. - Caesalpiniaceae) é uma espécie florestal nativa, encontrada nos biomas Cerrado e Caatinga, de grande importância em função de sua utilidade econômica e ecológica, o que justifica estudos sobre a germinação das sementes. Este trabalho avaliou o desempenho germinativo de sementes de *D. mollis* em diferentes substratos e regimes de temperatura. O delineamento experimental adotado foi inteiramente casualizado, em arranjo fatorial 4 x 4 (quatro substratos: entre areia, pó de coco, vermiculita e papel-toalha; quatro temperaturas: 25, 30, 35, 20-30 ^{o}C), com quatro repetições de 25 sementes cada. Foram avaliados os seguintes parâmetros: teor de água, germinação, primeira contagem da germinação, índice de velocidade de germinação, comprimento e massa seca da plântula. As temperaturas de 30 e 35 ^{o}C proporcionaram às sementes os melhores resultados de germinação e vigor. Os substratos papel e entre vermiculita permitiram bom desempenho germinativo, mostrando-se adequados para a avaliação da qualidade fisiológica de dementes de *D. mollis*.

Palavras-chave: Sementes florestais, Qualidade fisiológica e Fava d'anta.

1. INTRODUCTION

The "fava d'anta" or "faveira" tree (*Dimorphandra mollis* Benth., a member of the Caesalpiniaceae family) is widely distributed in the *Cerrado* (savanna) (LORENZI, 2002) and *Caatinga* (dryland) (ADEODATO, 2008) biomes in the states of Mato Grosso, Goiás, Minas Gerais, Pará, São Paulo, Mato Grosso do Sul (LORENZI, 2008) and Ceará (ADEODATO, 2008) in Brazil.

In addition to the great utility of "fava d'anta" wood for making boxes, plywood, ceilings, paneling, firewood and charcoal (LORENZI, 2008), other products from this tree have found niches in international markets. Rutin, a bioflavonoid carrier of vitamin P extracted from the fruits of this tree, acts with vitamin C to slow aging by normalizing the permeability of capillaries and increasing their strength, thus aiding blood circulation (SOUSA et al., 1991; FAPESP, 2008).

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The seed endosperm is rich in galactomannans (polysaccharides that are chemically identical to guar gum) that have wide industrial use in Brazil for making thickeners for yogurt and ice cream, medicine capsules, lubricants for oil drilling bits, and wrappers for dynamite, although almost all of this material is imported from species native to India and Pakistan (FAPESP, 2008).

As this species is not cultivated, “fava d’anta” fruits are harvested by unregulated extractivist techniques in the Cerrado and Caatinga regions and are bought by middlemen who sell it to production laboratories (FRANÇA, 2008).

These forest species are not reproduced using standardized methodologies as are most agricultural crops, and much research is still needed to define optimal conditions for seed germination and to determine the influence of temperature and the planting substrate.

Temperature is known to have a great influence on the germination percentage and the vigor of seedlings as it affects water absorption by the seed and essentially all plant metabolic processes (BEWLEY and BLACK, 1994).

The optimal temperatures for seedling development are quite variable among forest species, and most tropical species require temperature ranges from 20 to 30ºC (BORGES and RENA, 1993), depending on the normal temperature ranges in their home regions. Some species such as *Dinizia excelsa* Ducke (VARELA et al., 2005), *Caesalpinia ferrea* Mart. ex Tul. (LIMA et al., 2006), *Myracrodruon urundeuva* Fr. All. (PACHECO et al., 2006), *Apeiba tibourbou* Aubl. (PACHECO et al., 2007a), *Tapirira guianensis* Aubl. (CESARINO et al., 2007), and *Hylocereus setaceus* Salm-Dick ex DC. (SIMÃO et al., 2007) demonstrate good development under constant temperatures; while seeds of plants such as *Sebastiania commersoniana* (Baill.) Smith & Downs prefer diurnal variations in temperature (SANTOS and AGUIAR, 2000); and others, such as *Hypericum perforatum* L. and *H. brasiliense* Choisy (FARON et al., 2004) appear to be largely insensitive to the temperature regime they are exposed to. Accordingly, a seed’s response to different temperature regimes is directly associated to the conditions it is normally exposed to in its natural habitat (ALBUQUERQUE et al., 2003).

Another important environmental factor that must be considered in germination studies is the planting substrate. The substrate has the function of supplying the seeds with moisture conditions adequate for germination and seedlings development (FIGLIOLIA et al., 1993). As such, the substrate must strike an adequate balance between water availability and aeration while avoiding the formation of a water coating on the seed that impedes oxygen diffusion (POPINIGIS, 1985) and contributes to pathogen proliferation.

In choosing a planting substrate, a number of factors must therefore be considered, including: the size of the seed and its moisture and light requirements, the ease of substrate use, and the ease with which the medium facilitates seed counting and monitoring (BRASIL, 1992). The standardization presented in the *Regras para Análise de Sementes* (RAS - Seed Analysis Rules) (BRASIL, 1992) recommends substrates such as paper (towels, filter paper and blotting paper), sand and soil. There are few firm recommendations for forest species, however, and other substrates have been tested and used, such as Plantmax® with *Matayba guianensis* Aubl. seeds (OLIVEIRA et al., 2003), vermiculite with *Mimosa caesalpiniaefolia* Benth. seeds (ALVES et al., 2002), and coconut fiber with *Myracrodruon urundeuva* Fr. All. (PACHECO et al., 2006), *Apeiba tibourbou* Aubl. (PACHECO et al., 2007a) and *Platypodium elegans* Vog. (PACHECO et al., 2007b).

The present study evaluated the effects of different temperatures and substrates on the germination and seedling vigor of “fava d’anta” seeds.

2. MATERIALS AND METHODS

“Fava d’anta” seeds were obtained from Sementes Caiçara LTDA in August/2006 from the municipality of Araçatuba, State of São Paulo, Brazil, and tested in the Laboratório de Sementes do Departamento de Agronomia da Universidade Federal Rural de Pernambuco, State of Pernambuco, Brazil.

Before initiating the germination experiments, the seeds were treated with a 2.0% solution of the fungicide Thiran® to avoid the fungal infestations that were frequently observed during preliminary germination tests.

The germination tests were performed in *Biochemical Oxygen Demand* (B.O.D.) germinators with an eight hour photoperiod (supplied by four 20w day-light fluorescent lamps) at fixed temperatures of 25, 30, 35°C and alternating temperatures of 20-30ºC. The seeds exposed to alternating temperatures were held at the highest temperature during the eight hour light period.

Preliminary tests of seed dormancy indicated that mechanical scarification was necessary for successful germination, and this was performed manually by sanding the seed extremity opposite from the micropyle with nº 50 emery-paper until the cotyledons became visible. The seeds were sown into substrates of sand, coconut fiber, vermiculite and paper towels that had all been previously autoclaved at 120ºC for two hours and subsequently humidified with a 0.2% solution of Nistatina to prevent infestation by pathogens. The first three substrates were moistened to 60% of their water retention capacity in gerboxes (11 x 11 x 3cm) with lids, and the seeds were then covered with a 1.0cm thick layer of the same substrate. The paper towels were formed into rolls and moistened with a quantity of water equivalent to 2.5 times their dry weight (BRASIL, 1992).

The numbers of germinating seeds were evaluated daily, considering germination to have occurred with the emergence of the hypocotyl. The following parameters were evaluated: moisture content – determined by drying at 105±3 ºC for 24 hr, using two samples of 25 seeds per experiment (BRASIL, 1992); final germination – percentage of seeds that had germinated by the end of the experiment (13 days after sowing); first germination count - the percentage of seeds that had germinated by the 7th day; germination speed index (GSI) – according to the formula proposed by Maguire (1962); length of the aerial part and of the primary root – the aerial part of the primary root of normal seedlings were measured in millimeters at the end of the experiment; dry weight matter of the aerial part and of the root system – at the end of each experiment, the normal seedlings (after removing the cotyledons) were placed in paper sacks and dried under forced ventilation at 80ºC, for 24 hours. The dry seedlings were subsequently weighed using an analytical balance (0.001mg precision) and the results expressed as mg/ seedling, following Nakagawa (1999).

The experiments tested all four temperatures with all four substrates in four repetitions using 25 seeds in each temperature/substrate combination. The percentage values were arc-sine transformed (arc-sine $\sqrt{\%/100}$), and data analysis was performed using ESTAT statistical software (FCAV/UNESP), version 2.0/2001. Averages were compared using the Tukey test at a 5% probability level.

3. RESULTS AND DISCUSSION

The "fava d'anta" seeds had a water content of 11.8% at the beginning of the tests. The evaluation of seed germination percentages (Table 1) 13 days after sowing demonstrated favorable results with both constant and alternating temperatures. The highest germination rates were seen with the sand and paper towel substrates under all temperature regimes; with seeds in coconut fiber germinating best at 25ºC; and in vermiculite at 30 and 35ºC.

Table 1 – Final germination (%) and first germination count (%) of *Dimorphandra mollis* Benth. seeds submitted at different temperatures and substrates.
Tabela 1 – *Germinação final (%) e primeira contagem da germinação (%) de sementes de Dimorphandra mollis Benth. submetidas a diferentes temperaturas e substratos.*

Substrates	Temperatures (ºC)			
	25	30	35	20-30
	Final germination (%)			
Sand	88 Aa	80 ABa	75 ABa	84 Aa
Coconut fiber	86 Aa	65 Bab	62 Bb	52 Bb
Vermiculite	54 Bc	91 Aa	86 Aab	74 Abc
Paper	89 Aa	78 ABa	86 Aa	87 Aa
	First germination count (%)			
Sand	55 Aa	69 BCa	60 Ba	69 Aa
Coconut fiber	61 Aa	54 Ca	54 Ba	0 Bb
Vermiculite	0 Cb	90 Aa	86 Aa	0 Bb
Paper	24 Bc	78 Ba	86 Aa	65 Ab

Average followed by the same letter, capital in the column and small in the line, did not differ by Tukey test at 5% probability.

The first evaluation of germination (Table 1) on the 7th day after sowing demonstrated similar germination rates as those seen after 13 days with equivalent substrate and temperature regimes. Germination in sand was highest at both 25°C and the 20-30ºC alternating regime; at 25ºC in coconut fiber; at 30 and 35ºC in vermiculite; and at 35ºC on paper towels. These results confirm the report by Borges and Rena (1993) that most tropical forest species seeds germinate best at temperatures between 20 to 30°C.

Similar results were reported with the seeds of others species from the Cerrado and Caatinga biomes, such as *Myracrodruon urundeuva* Fr. All (PACHECO et al., 2006) and *Apeiba tibourbou* Aubl. (PACHECO et al., 2007a), whose optimal germination temperatures were in the range of 20 to 30°C.

The GSI results were very similar to those observed for the germination percentage (Table 2), with the most rapid germination being seen at a constant temperature of 25ºC in all of the substrates, except vermiculite. High germination velocity rates were also observed at 30ºC in sand, vermiculite, and on paper towels (but not with coconut fibers); at 35ºC in vermiculite and on paper towels; and with alternating temperatures of 20-30ºC with sand and paper towels. Environmental temperatures thus influence the percentage and the velocity of germination, the water uptake by the seed as well as the metabolic and biochemical reactions that regulate these processes (BEWLEY and BLACK, 1994).

In light of the fact that the temperature optimums for seed germination can be directly associated with the ecological characteristic of each species (PROBERT, 1992), different requirements would also be expected for seeds of pioneer species versus non-pioneer or climax species. The former generally germinate in open areas that receive direct solar radiation for at least part of the day (SWAINE and WHITMORE, 1988) and are presumably adapted to developing under environmental temperatures that fluctuate during the day. The seeds of non-pioneer or climax species, on the other hand, usually germinate under the fully shaded or partial shaded conditions of the forest understory (SWAINE and WHITMORE, 1988) and would be expected to be adapted to only reduced thermal oscillations.

Table 2 – Germination speed index (GSI) of *Dimorphandra mollis* Benth. seeds submitted at different temperatures and substrates.
Tabela 2 – *Índice de velocidade de germinação de sementes de Dimorphandra mollis Benth. submetidas a diferentes temperaturas e substratos.*

Substrates	Temperatures (°C)			
	25	30	35	20-30
	GSI			
Sand	2,87 Aa	2,74 ABa	2,56 Ba	2,85 Aa
Coconut fiber	2,86 Aa	2,48 Bab	2,13 Bb	1,01 Cc
Vermiculite	1,30 Bc	3,21 Aa	3,10 Aa	2,20 Bb
Paper	2,75 Aa	2,78 ABa	3,10 Aa	2,93 Aa

Average followed by the same letter, capital in the column and small in the line, did not differ by Tukey test at 5% probability.

Within this context, the data in Tables 1 and 2 indicate that the high germination percentage and germination speed index of the “fava d’anta” seeds seen in essentially all of the temperature regimes utilized in the present study indicates the ability of these seeds to become established in nature under a wide range of environmental conditions.

While the substrates and temperatures used in the present study all resulted in high germination percentages (up to 91%) (Table 1), the concept of an optimal temperature must also consider other aspects related to seedling vigor (LABOURIAU, 1983).

The best over-all substrate and temperature combinations in terms of the length of the aerial part of the seedlings (Table 3) were found with paper towels and temperatures of 30 and 35ºC. Additionally, seedlings grown in the sand and in coconut fiber substrates at these same temperatures also demonstrated their greatest average aerial part lengths; as was also seen with vermiculite at 35°C and paper towels at 25ºC. The greatest seedling vigor, as evaluated by the length of the primary root (Table 3), was seen at constant temperatures 30 and 35ºC with all of the different substrates. Significant seedling primary root development was also observed with the paper towel substrate at 25ºC.

In terms of the average dry weight matter of the aerial part of “fava d’anta” seedlings (Table 4), the best results were seen under conditions of constant temperatures at 30 and 35ºC with the paper substrate, followed by 25ºC conditions with the coconut fiber and paper towel substrates. Similar results were observed in terms of seedling vigor as measured by the length of the primary root (Table 3) and by

Table 3 – Length (cm/seedling) of the aerial part and primary root of *Dimorphandra mollis* Benth. seedlings originated from seeds submitted at different temperatures and substrates.
Tabela 3 – *Comprimento (cm/plântula) da parte aérea e da raiz primária de plântulas de Dimorphandra mollis Benth., originadas de sementes submetidas a diferentes temperaturas e substratos.*

Substrates	Temperatures (ºC)			
	25	30	35	20-30
	Lenght (cm/seedling) of the aerial part			
Sand	2,0 Bb	4,3 Ca	4,6 Ba	1,1 Ab
Coconut fiber	2,7 Bb	5,6 Ba	5,4 Ba	1,2 Ac
Vermiculite	1,9 Bc	3,8 Cb	5,2 Ba	0,8 Ad
Paper	4,2 Ab	7,1 Aa	7,6 Aa	1,4 Ac
	Lenght (cm/seedling) of the primary root			
Sand	8,1 Ab	7,9 Aa	7,6 Aa	3,9 Ab
Coconut fiber	3,0 Bb	9,7 Aa	8,3 Aa	2,5 Ab
Vermiculite	3,3 Bb	8,3 Aa	8,5 Aa	3,0 Ab
Paper	8,1 Aa	8,2 Aa	9,4 Aa	3,7 Ab

Average followed by the same letter, capital in the column and small in the line, did not differ by Tukey test at 5% probability.

Table 4 – Dry weight matter (mg/seedling) of the aerial part and root system of *Dimorphandra mollis* Benth. seedlings originated from seeds submitted at different temperatures and substrates.
Tabela 4 – *Massa seca (mg/plântula) da parte aérea e do sistema radicular de plântulas de Dimorphandra mollis Benth., originadas de sementes submetidas a diferentes temperaturas e substratos.*

Substrates	Temperatures (ºC)			
	25	30	35	20-30
	Dry weight matter (mg/seedling) of the aerial part			
Sand	22,8 Ba	25,5 Ba	25,6 Ba	14,4 ABb
Coconut fiber	26,0 ABa	28,2 Ba	26,8 Ba	10,8 BCb
Vermiculite	17,8 Cb	24,4 Ba	26,4 Ba	9,2 Cc
Paper	28,8 Ac	39,1 Aa	33,5 Ab	15,3 Ad
	Dry weight matter (mg/seedling) of the root system			
Sand	12,5 Ba	11,3 Aa	10,6 Aa	8,3 Ab
Coconut fiber	5,2 Db	9,5 Aa	9,8 Aa	5,6 Bb
Vermiculite	8,7 Cab	10,6 Aa	9,8 Aab	7,7 Ab
Paper	14,9 Aa	9,2 Ab	11,1 Ab	9,7 Ab

Average followed by the same letter, capital in the column and small in the line, did not differ by Tukey test at 5% probability.

the dry weight matter of the root system (Table 4), where the best combinations were seen with the paper substrate at 25ºC, and by the sand, coconut fiber, and vermiculite substrates at 30 and 35ºC. These results are congruent with the report by Nakagawa (1999) that the best method for evaluating seedlings growth is by determining their dry weight matter.

Seedling development in the vermiculite substrate at low temperatures was statistically inferior to all of the other combinations evaluated, while improved results were seen at high temperatures, mainly at 35ºC. These results may be related to the fact that vermiculite (in its expanded form) has low thermal conductivity (UGARTE et al., 2005) that may have influenced water uptake by the seeds and the grow rate of the seedlings.

The results presented here demonstrate that the seeds of “fava d’anta” can germinate well over a wide range of temperatures, but that the initial development of the seedlings is temperature sensitive and seedlings exposed to temperatures of 30 and 35ºC yield the highest length and dry weight values (Table 3 and 4 respectively). Additionally, in terms of the germination and vigor tests, it could be seen that temperatures of 30 and 35ºC gave better results for all of the parameters evaluated.

Thus, in spite of the fact that temperatures between 20 and 30ºC have been found to be ideal for promoting seed germination in most tropical forest species (BORGES and RENA, 1993), *D. mollis* (found in the Cerrado and Caatinga biomes) responded better at higher temperatures in the present work, indicating that this "ideal" temperature range should be amplified. Similar results were also seen with the seeds of *Dinizia excelsa* Ducke (VARELA et al., 2005), *Adenanthera pavonina* L. (SOUZA et al., 2007), and *Dyckia tuberosa* (Vell.) Beer (VIEIRA et al., 2007).

Paper towels were found here to provide a favorable germination and growth environment for all of the parameters evaluated with "fava d'anta" seeds; this substrate has likewise been recommended for germinating forest species seeds such as *Crataeva tapia* L. (SILVA et al., 2007) and *Prunus selowii* Koehne (RODRIGUES et al., 2008). It is possible that the greater spacing possible between the seeds on paper towels contributes to their greater germination velocity (Table 2) and the greater lengths of the aerial part and primary roots (Table 3).

As can be seen in Figure 1, the paper towel and vermiculite substrates promote a very homogeneous development of the seedlings, which is a very important characteristic when uniform populations are required. Vermiculite has been used with positive results for germinating a number of different forest species seeds (FIGLIOLIA et al., 1993; ALVES et al., 2002; PACHECO et al., 2006). Vermiculite is a low-density substrate, which facilitates its use with gerbox in the germinators, and its high water absorption capacity means that it does not need to be refreshed on a daily basis, and this substrate has therefore been found to be very useful for germinating "fava d'anta" seeds.

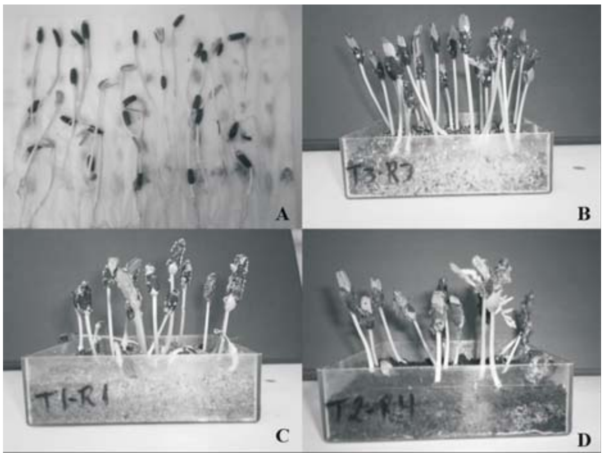

Figure 1 – *Dimorphandra mollis* Benth. seedlings proceeding from seeds subjected at 35ºC, in substrates: paper (A), vermiculite (B), sand (C) and coconut fiber (D), 13 days after sowing.

***Figura 1 –** Plântulas de Dimorphandra mollis Benth., originadas de sementes submetidas a 35ºC, entre os substratos papel (A), vermiculita (B), areia (C) e pó de coco (D), aos 13 dias após a semeadura.*

D. mollis seeds also germinated at high percentages in the sand substrate but, as was reported by Pacheco et al. (2006) with seeds from *Myracrodruon urundeuva* Fr. All., it is sometimes difficult to maintain high humidity levels in this substrate (which may have contributed to the lack of uniformity observed in the length of the aerial part of the seedlings, as seen in Figure 1). Additionally, the water often drains from the top layer of the sand substrate leaving it much drier, and sand is also quite heavy which makes manipulating germination trays more difficult (FIGLIOLIA et al. 1993). The RAS regulations (BRASIL, 1992) recommend that germination substrates should not be watered again after sowing, as this often results in more variable germination and growth rates.

Coconut fiber has physical properties that make it a very satisfactory substrate in many respects, such as high porosity (95.6%), optimal water retention capacity (538 mL/L) and good aeration (45.5%) (CARRIJO et al., 2002). These same authors note that coconut fiber is essentially inert, that it does not contain nutrients essential to seedling development, can be acquired at low cost, and is easily available. While neither coconut fiber or vermiculite are mentioned in the RAS, they have been used with success in many germination tests of forest species (MELO et al., 2005; PACHECO et al., 2006; LIMA et al., 2007; PACHECO et al., 2007a; PACHECO et al., 2007b; SILVA et al., 2007). However, good germination percentage at first counting (Table 1) as well as germination velocity (Table 2) of seeds sown in coconut fiber was only observed at 25ºC in the present work. The lower observed seed germination at higher temperatures (Table 1) may be associated with the fact that this substrate becomes quite dense when moist, and cotyledon emergence through this covering fiber layer appears to be more difficult.

Accordingly, the density of the coconut fiber substrate layer covering the seed and the resulting slowing of germination speed (Table 2) may explain the lack of uniformity in terms of seedling development in the substrate (Figure 1) as well as the low values of the aerial part length (Table 3). Although the high density of the coconut fiber impeded the development of the aerial part of the seedlings, the roots were observed to develop normally when the seeds were germinated and grown at temperatures of 30 and 35ºC (Table 3).

4. CONCLUSIONS

"Fava d'anta" seeds demonstrate superior germination and vigor when germinated on paper towel or vermiculite substrates at constant temperatures of 30 and 35ºC.

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