

Reproductive biology of *Bowdichia virgilioides* Kunth (Fabaceae)

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ABSTRACT. The aim of this study was to investigate the reproductive biology of *Bowdichia virgilioides* in an area of Cerrado (Brazilian Savanna). The study was carried out in Chapadinha, State of Maranhão in northeastern Brazil. *Bowdichia virgilioides* has cornucopia flowering and annual pattern. Flowering occurred at the beginning of the dry season, between June and August; and fruiting in the middle of this season, between July and October. The anthesis of *B. virgilioides* is diurnal, and the main flower resource is the nectar. During the flower opening, the anthers becomes dehiscent and thus pollen grains are deposited at the stigma, promoting automatic self-pollination. The species is self-compatible and apomitic. In the study area, they were visited by nine species of bees, four butterflies, two wasps, and two hummingbirds. The visits began in early morning and persisted throughout the day, with a peak of activity between 8:00 and 9:00 hours. Species of *Centris* were the main visitors of *B. virgilioides*. The butterfly and wasp species were considered resource robbers, and other floral visitors were considered occasional visitors. Although the flowers of *B. virgilioides* are widely visited, the species shows a high rate of inbreeding because of automatic self-pollination.

Keywords: breeding system, Cerrado, pollination, reproductive success.

RESUMO. Biologia reprodutiva de *Bowdichia virgilioides* Kunth (Fabaceae). Este trabalho teve como objetivo estudar a biologia reprodutiva de *Bowdichia virgilioides* Kunth (Fabaceae) e avaliar o papel dos visitantes florais em seu sucesso reprodutivo. *Bowdichia virgilioides* apresenta padrão de floração anual e do tipo cornucópica. A floração ocorre no início da estação seca, entre os meses de junho e agosto e a frutificação no meio desta estação, entre os meses de julho e outubro. Suas flores são de antese diurna e apresentam néctar como recurso floral. Durante a abertura floral, as anteras se encontram deiscentes e por esta razão há a adesão dos grãos de pólen no estigma da própria flor, promovendo o processo de autopolinização passiva. A espécie é autocompatível e apomítica. As flores foram visitadas por nove espécies de abelhas, quatro espécies de borboletas, duas espécies de vespas, e por duas espécies de beija-flores. As visitas iniciam logo ao amanhecer e perduram por todo o dia, com um pico de atividades de todos os insetos entre 8 e 9h. Em virtude do seu comportamento intrafloral, por sua abundância e frequência, *Centris* spp. são os principais visitantes de *B. virgilioides*. As borboletas e as vespas foram consideradas pilhadoras e os outros visitantes florais foram considerados visitantes ocasionais. Embora as flores de *B. virgilioides* sejam intensamente visitadas, a espécie pode apresentar alta taxa de endogamia pela autopolinização passiva.

Palavras-chave: sistema de reprodução, Cerrado, polinização, sucesso reprodutivo.

Introduction

Bowdichia virgilioides has a wide geographical distribution, occurring naturally from the States of Rio de Janeiro to Roraima, mainly in central and northern Brazil (SILVA JÚNIOR; SANTOS, 2005). Commonly known as “sucupira”, “sucupira-roxa” or “sucupira-preta”, it is economically value because of its timber, which is used for various purposes; the medicinal properties of its stem bark; its landscaping potential; and its use in programs for management and restoration of degraded areas (LORENZI, 1992; SILVA JÚNIOR; SANTOS, 2005; SMIDERLE; SOUZA, 2003). For these

reasons, there is a need for conservation and promotion of the spread of this species. Studies on seed germination of *B. virgilioides* have been conducted for this purpose (ALBUQUERQUE; GUIMARÃES, 2007; RODRIGUES; TOZZI, 2007; SILVA et al., 2001; SMIDERLE; SOUZA, 2003).

The reproductive phenology of *B. virgilioides* differs in different places where it occurs (FUNCH et al., 2002; LOCATELLI; MACHADO, 2004). These variations may be linked to climate and soil conditions in different regions.

The reproductive biology of native species is an important parameter in maintaining the ecological balance of many biological systems, and leads to

various plant-animal interactions such as the pollination mechanism. *B. virgilioides* is adapted for bee pollination, because its nectariferous flowers show mellitophilic characteristics (SILVA JUNIOR; SANTOS, 2005). However; Rojas and Ribbon (1997) described a guild of nine species of birds, including six hummingbirds and three passeriform that visited its flowers, which indicates a variety of pollination mechanisms.

Many self-compatible species have special adaptations to prevent the automatic self-pollination, such as species herkogamous, protandrous and protogynous. However, in many other species not there is a morphological or temporal explicit barriers to prevent the transfer of pollen grains of the same flower, as observed in several species of Fabaceae (ARROYO, 1981; NOGUEIRA; ARRUDA, 2006) as well as in species of families not related (SILVA; PINHEIRO, 2009). This mechanism can favor the reproductive efficiency of a species in a fragmented environment where pollinator availability is limited (MOTTEN; ANTONOVICS, 1992).

The Self-compatibility and automatic self-pollination is a common phenomenon in many species of Fabaceae (BORGES, 2006; JACOBI et al., 2005; NOGUEIRA; ARRUDA, 2006; VIEIRA et al., 2002). Although flowers with a wide availability of resources are heavily visited, many self-compatible species have a high degree of inbreeding caused by geitonogamous pollination, apomixis, and also automatic self-pollination (BAWA, 1979). These processes can drastically decrease the genetic variability of biological populations, but may also increase the chances of producing fruits and seeds, even with a low rate of floral visits (CHARLESWORTH; CHARLESWORTH, 1995). Self- versus cross-pollination can occur concurrently in many plant species, and only specific studies for each group can infer any specific explanation for the prevalence of one or another mechanism (BAWA, 1979).

This study aimed to describe the phenological aspects, floral biology, floral visitors, reproductive system and reproductive success of *B. virgilioides* in an area of Savanna in the city of Chapadinha, Maranhão State, Brazil. In general, it is intended to answer the following question: Floral visitors are important for fruit set of *B. virgilioides*?

Material and methods

Study area

The city of Chapadinha is located in the microregion of Chapadinha (03° 44'17" S and 43° 20'29" W), also known as the microregion of "Alto

Munim" (IBAMA, 2006), in the southwestern part of the state of Maranhão, Brazil. The region is characterized by Cerrado *sensu lato*, formed by a mosaic of plant communities. *Bowdichia virgilioides* occurs in savanna, mainly in "campo sujo", grassland with scattered trees and shrubs.

The climate type of the region, according Köppen (1948) is wet tropical. Precipitation is high during the months of January to July and very low from August to November/December. The highest temperatures occur in the dry period.

Species studied

Bowdichia virgilioides Kunth (Fabaceae), Clade Genistoids (WOJCIECHOWSKI et al., 2004) is an arboreal species that is typical of the Cerrado *sensu stricto*, "Campo Sujo" (grassland with scattered trees and shrubs), and "Cerradão" (woodland with a well-developed tree canopy) (SILVA JUNIOR; SANTOS, 2005). However, it occurs predominantly in open areas such as "Campo Sujo", because the establishment and development of its seedlings is strongly influenced by shading, which is greater in areas of "Cerradão" (KANEGAE et al., 2000). According to LORENZI (1992), *B. virgilioides* occurs mainly in dry, nutrient-poor soils. However, Miranda et al., (2002) reported that this species is typical of sodium-rich soils in savannas in the state of Roraima, Brazil.

Flowering and fruiting phenology

Flowering and fruiting phenology were investigated from January 2007 until December 2008. Ten individuals previously selected, adults and healthy, were observed twice a month. During reproductive period, observations were weekly.

Floral biology

Floral biology of *B. virgilioides* was verified in natural populations. Buds and flowers were collected and analyzed in the laboratory. The stigma receptivity was tested although hydrogen peroxide (H₂O₂) 3% (KEARNS; INOUE, 1993) in flowers in natural conditions. Anthesis was observed over a period of five days in five adults and healthy focal individuals, registering time of flower opening, flower longevity, and process of anther dehiscence.

Two samples of 15 flowers were collected in natural conditions to verify nectar production. These flowers were previously bagged one day prior to the anthesis. The first sample was collected at 8:00 hours and the second at 15:00 hours, on two consecutive days. Fifteen Flowers previously bagged were dissected to collect the nectar. Concentration

of the total volume of 15 flowers from each sample was analyzed with a portable refractometer with a scale from 0 to 32% Brix. The volume of nectar from each flower was measured with the aid of micropipettes of 0.2 mL.

Floral visitors

Floral visitors were observed, collected and identified during flowering period of five pre-selected individuals, adults and healthy. Observations were from 6:00 to 17:00 hours, during ten consecutive days, totaling about 100 hours of observation. Floral visitors were classified as effective when the behavior of visitors promotes the contact of anthers with stigma and frequency of visits is high, more than 10 visits per day, occasional when the behavior of visitors promotes the contact of anthers with stigma and frequency of visits is low, less than 10 visits per day or floral resources robbers when visitors do not touch the stigma nor the anthers.

Breeding system

The following tests were carried out to evaluate the breeding system: 1-Manual self-pollination (autogamy); 2-Cross-pollination (xenogamy). These tests were conducted with previously bagged flowers one day prior to the anthesis; 3-Control - Unbagged flowers; 4-Automatic self-pollination - calculated from the proportion of fruits formed from bagged buds.

The pollen load on the stigmas of open (naturally pollinated) and bagged (automatic self-pollinated) flowers was determined by counting the pollen grains on each stigma, using an optical microscope. The styles of some buds were removed to test for the occurrence of autonomous apomixis (RICHARDS, 1986).

Significance difference between the percentages obtained from xenogamy, autogamy and the control experiments were evaluated by comparison formula between two percentages, with a 5% significance difference level (PAGANO; GAUVREAU, 2004). The obtained values of "t" were then compared with tabulated theoretical "t" values.

Reproductive success

To estimate the fecundity rate, we followed Cruden (1972) procedure, which is the product of two ratios (seed/ovule and fruit/flower).

Results and discussion

Flowering phenology

Bowdichia virgilioides has cornucopia flowering and an annual pattern, according to the classification proposed by Gentry (1974) and Newstrom et al.

(1994), at the population level. Although, individual plants exhibit a supra-annual pattern. In 2007, flowering occurred at the beginning of the dry season, between June and August; and fruiting in the middle of this season, between July and October. In 2008, only three individuals of the population flowered between June and August, and none of the individuals that flowered in 2007 flowered in 2008. These observations agree with those of Bulhão and Figueiredo (2002) and Figueiredo (2008), studying a natural plant community in the Cerrado of Maranhão State. According to these same authors, this pattern of flowering early in the dry season is common in many species of the Cerrado.

The abscission of the leaves occurs before the beginning of flowering and increases the display of the flowers. Flowering in the dry season may be a characteristic of low latitudes, because in a Cerrado region in the state of São Paulo, the tree species flower mainly in the beginning of the rainy season (BATALHA; MANTOVANI, 2000).

The flowering of *B. virgilioides* can occur at different times in different localities, but always in the dry periods of the year. Funch et al. (2002) observed flowering between June and October in the "Chapada Diamantina", Bahia State and Locatelli and Machado (2004) observed flowering between November and January in a montane forest in Pernambuco State, Brazil. These differences may be related to climate and soil conditions in each area.

Floral biology

Bowdichia virgilioides has a paniculate inflorescence with 23.7 flowers (± 18.3 , $N = 18$). The large number of flowers per inflorescence is a common feature among the species of Fabaceae (KIILL; DRUMOND, 2001). Flowers are hermaphroditic, with 1.75 cm length (± 0.17 , $N = 20$), pedicel short, calyx campanulate with five ciliate teeth, 0.5 cm in diameter (± 0.05 , $N = 20$) and 0.8 cm in depth (± 0.1 , $N = 21$). The inner part of the calyx forms a nectariferous chamber. The color of the petals varies along the anthesis, purple at the beginning and purplish toward the end. This feature showed no relationship with pollination mechanism. The flowers are papilionacea type with vexillus subunguiculate, transverse-oblong, deeply emarginate, unguiculate wings, obovate, keels free and elliptical, as suggested by Silva Júnior and Santos (2005).

The ovary is unicarpellate, unilocular, with 8.2 ovules (± 1.32 , $N = 35$), the stigma is wet and papillose and remains receptive during the first day

of anthesis. The androecium is formed by ten stamens with orbicular anthers with rimose dehiscence.

The floral resource is the nectar that is produced inside the nectariferous chamber, in the base of the floral receptacle around the ovary. The flowers produce nectar only the first day of anthesis. In two samples, the first at 8:00 hours and the second at 15:00 hours, the volume of nectar per flower was about 0.1 mL and the concentration of nectar was 13% (Brix scale). Nectar with a low concentration of sugar cannot satiate the floral visitor on the first visit, and only prolongs their foraging to collect more food. In many species of Fabaceae, the concentration of nectar does not exceed 28% sugar (ETCHEVERRY; ALEMÁN, 2005; KUDO; HARDER, 2005; MENDONÇA; ANJOS, 2006).

The anthesis was diurnal and began soon after sunrise. The flowers remained open only on the first day of anthesis, on the second day the petals wilted, became whitish, and the anthers had no

more pollen grains. At the moment of floral opening, only part of the style and stigma receptive were exposed (Figure 1A-1); during the day, the petals reflexed and exposed the anthers (Figure 1A-2). This mechanism can act as herkogamic, temporally separating the receptive female and male structures (FAEGRI; VAN DER PIJL, 1979). However, before opening the anthers are already dehiscent, and therefore the pollen grains adhere on the stigma, causing automatic self-pollination.

The self-pollination mechanism is common in other species of Fabaceae (KILL; DRUMOND, 2001; NOGUEIRA; ARRUDA, 2006; BORGES, 2006) and may to favor a significant increase in the rate of inbreeding in self-compatible species (BAWA; WEBB, 1984). The automatic self-pollination can hinder cross-pollination pollen tube growth, owing to competition with the autogamic pollen tubes, thus reducing xenogamy rate (MAHORO, 2003; SILVA; PINHEIRO, 2009).

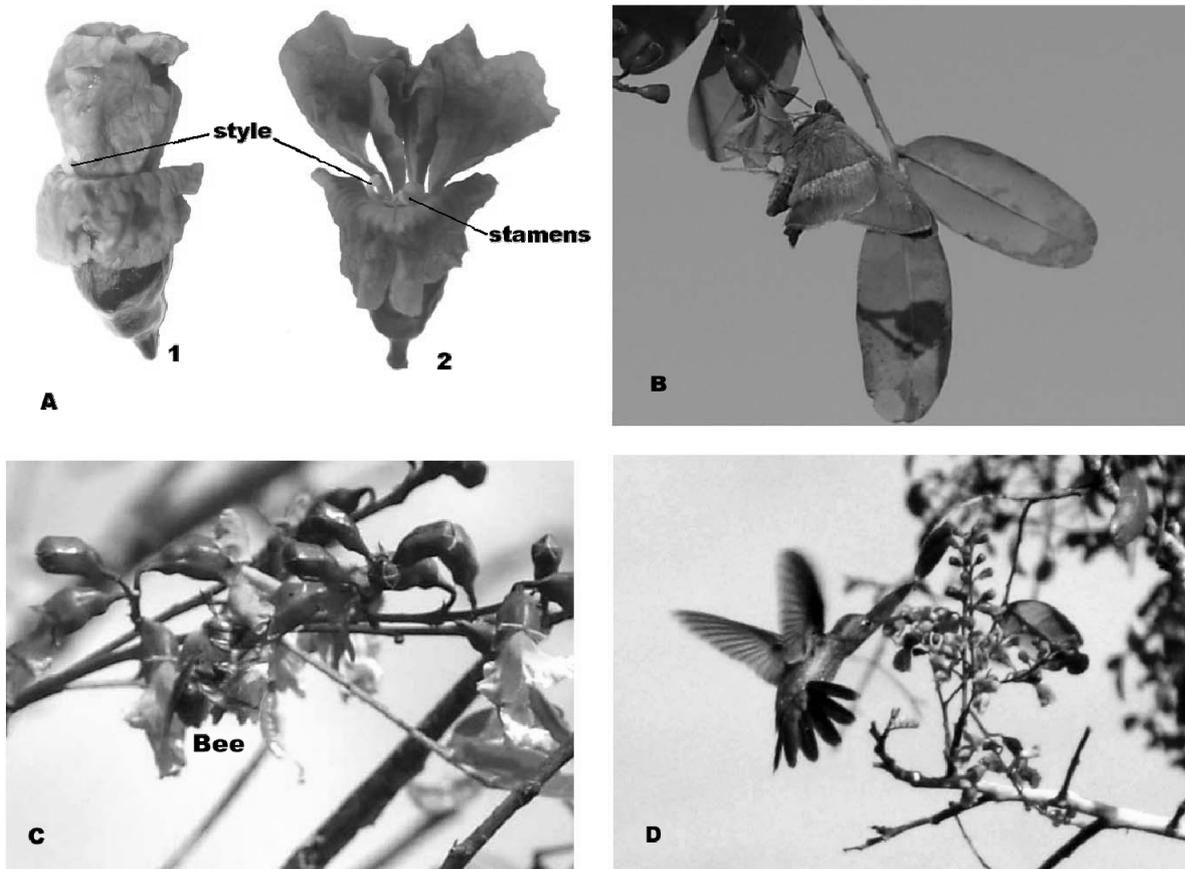


Figure 1. A- Flower of *Bowdichia virgilioides* Kunth (Fabaceae): A1- Flower at the beginning of anthesis, showing part of estyle; A2- Flower with stamen and style exposed; B- Visit of Lepidoptera; C- Visit of *Centris* sp.; D- Visit of *Chlorostilbon mellisugus*.

Floral visitors

Flowers of *B. virgilioides* were visited by nine species of bees (Apidae), four species of butterflies (Hesperiidae); two species of wasps (one of Braconidae and the other Vespidae) and two species of hummingbirds (Trochilidae) (Table 1).

Table 1. Floral visitors of *Bowdichia virgilioides* Kunth (Fabaceae) in an area of Cerrado in Chapadinha, Maranhão, Brazil: mv- main visitors; ov- occasional visitors, ro- robber.

Order/Family	Species	Pollination efficiency
Himenoptera		
Apidae	<i>Centris (Centris) aenea</i> Lepeletier	mv
	<i>Centris (Trachina) grupo</i> <i>fuscata</i> Lepeletier	mv
	<i>Xylocopa</i> sp.	ov
	<i>Apis mellifera</i> L.	ov
	<i>Trigona</i> sp. 1	ov
	<i>Trigona</i> sp. 2	ov
	<i>Partamona</i> sp.	ov
	<i>Geotrigona</i> sp.	ov
	sp. 1	ro
	sp. 2	ro
Vespedae		
Braconidae		
Lepidoptera		
Hesperiidae	<i>Urbanus dorantes dorantes</i>	ro
	<i>Chioides catillus catillus</i>	ro
	sp. 1	ro
	sp. 2	ro
Trochiliformes		
Trochilidae	<i>Chlorostilbon mellisugus</i>	ov
	<i>Amazilia fimbriata</i>	ov

The bees visited the flowers throughout the day, especially from 7:30 to 8:30 hours. The bees foraged more intensely intra-plant because of the abundance of flowers on one individual, which guarantees an unlimited supply of resources throughout the day. The species of Apidae were the most frequent and abundant visitors, and of these, *Centris (Centris) aenea* Lepeletier, 1841 and *Centris (Trachina) grupo fuscata* Lepeletier, 1841 were the most common species. Species of Apidae are the main pollinators of other species of Fabaceae (VIEIRA et al., 2002; GUEDES et al., 2009), and bees are the main pollinators in the Cerrado biome (MARTINS; BATALHA, 2007).

During foraging, each bee remained around 10 seconds on each flower. They were positioned in the flower, seizing the wings and keels, and introduced the proboscis inside the nectariferous chamber to collect nectar (Figure 1C). All visiting bees showed the same intra-floral behavior. Large amounts of pollen grains were found, mainly in Corbicula and on the feet of these bees. Because of their intra-floral behavior, its abundance and frequency, species of *Centris* are the main visitors of *B. virgilioides*.

Apis mellifera showed low frequency of visits to flowers of *B. virgilioides*; however, Carvalho and Marchini (1999) reported that this species is frequent in the flowers of *B. virgilioides* in the municipality of

Castro Alves, Bahia State, Brazil. Although species of *Xylocopa* are considered efficient pollinators in species of Fabaceae (KIILL; DRUMOND, 2001; BORGES 2006; GUEDES et al., 2009), they were infrequent in this study. Two species of *Trigona* observed visiting the flowers of *B. virgilioides* showed intra-floral behavior similar to other bees, although they were also uncommon, as were *Geotrigona* sp. and *Partamona* sp. Because of the low frequencies of these species of bees, they were considered occasional visitors.

The butterflies visited the flowers intensely throughout the day, and because of the wide availability of flowers in one individual, they exhibited a foraging behavior like bees, visiting the flowers of the same plant for long periods. They land on the flower and introduce their proboscis into the nectariferous chamber to collect the nectar, which according to Proctor and Yeo (1972) is the principal food of Lepidoptera adults. During the visit, only the proboscis touches the floral reproductive structures (Figura 1B), and few or no pollen grains from this structure were observed in specimens collected. For this reason, the butterflies were considered only as robbers of nectar. These insects also act only as nectar robbers from other species of Fabaceae (KIILL; DRUMOND, 2001; NOGUEIRA; ARRUDA, 2006), and also from species of other families (FAEGRI; VAN DER PIJL, 1979; MARTINS; BATALHA, 2007). According to Arroyo (1981), the morphology of flowers of Fabaceae does not correspond to the Lepidoptera pollination syndrome.

Wasps were rarely observed, and it was not possible to determine whether their visits were to seek for pollen grains or nectar. Because of the low frequency of visits and because they did not enter the full flower reproductive structures, these insects were considered as robbers of floral resource. In *Centrosema pubescens* Benth. (Fabaceae), wasps can act as pollinators as much as pillagers (BORGES, 2006), which shows the great diversity of pollination systems among species of Fabaceae (ARROYO, 1981).

Hummingbirds were also rarely observed visiting flowers of *B. virgilioides*. On each visit to a plant, a bird visited up to five flowers and left soon after. The purpose of the intra-floral visits is to search for nectar. Because the flower tube is short, only part of the beak of this animal touches the flower (Figure 1D). These animals were observed more frequently around 6:30 and 8:30 hours. Because of their low frequency, these birds were considered occasional visitors. However, Rojas and Ribbon (1997) observed that in a Cerrado remnant in Minas Gerais State, Brazil, *B. virgilioides* was heavily visited by nine species of birds, including three species of

passariforms and six species of hummingbirds (Trochilidae), which visited the flowers to look for nectar and small insects. Hummingbirds are considered effective pollinators of many species of Fabaceae (GALETTO et al., 2000; ETCHEVERRY; ALEMÁN, 2005).

Breeding system

Bowdichia virgilioides is self-compatible, because it showed a rate of fruit production of 36% (N = 25) in an experiment of manual self-pollination (Table 2). The xenogamy test showed an index of 36% (N = 25), and the fruit/flower ratio under natural conditions was 13.2% (N = 114). Comparing the results of the experiments of autogamy and xenogamy, which were identical to each other, to the result for the control, found that there was a significant difference between them ($t_{obs} = 32.85 < t_{teor} = 2.06$). These data indicate that there is a significant increase in the production of fruit when there is deposition of grains in the stigma, which may represent inefficiency in the mechanism of natural pollination. The low production of fruit in natural conditions is a common phenomenon in many species of Fabaceae, and may be related to several factors, such as the low rate of pollination, herbivores, and the effect of climate, among other factors (GALETTO et al., 2000; KIILL; DRUMOND, 2001; ETCHEVERRY; ALEMÁN, 2005). However, mass flowering of *B. virgilioides* can counterbalance the low fruit/flower ratio.

Flowers previously bagged in bud had approximately 29 pollen grains on the stigma (N = 25), whereas unbagged control flowers had 36 (N = 25); these means are not significantly different ($t_{obs} = 1.04 < t_{teor} = 2.06$). Natural biotic pollination did not significantly increase the amount of pollen grains on the stigma, and therefore automatic self-pollination is possible in this species. The fruit set in an experiment of automatic self-pollination was 20.4% successful (N = 201). This index, although higher than the fruit set in natural conditions, was not significantly different from it ($t_{obs} = 1.75 < t_{teor} = 2.06$). Automatic self-pollination is also common in other species of Fabaceae (BORGES, 2006; NOGUEIRA; ARRUDA, 2006), as well as in species of unrelated families, and can promote fruit set if there is a decrease in rates of natural pollination (BAWA; WEBB, 1984; CHARLESWORTH; CHARLESWORTH, 1995; FAEGRI; VAN DER PIJL, 1979; SILVA; PINHEIRO, 2009).

Self-compatibility may be responsible for a higher production of fruit in natural conditions caused by the process of automatic self-pollination in *B. virgilioides*. The reproductive system is

diversified in Fabaceae, which includes both self-compatible (ARROYO, 1981; VIEIRA et al., 2002; KUDO; HARDER, 2005; NOGUEIRA; ARRUDA, 2006; BORGES, 2006) and self-incompatible species (ETCHEVERRY; ALEMÁN, 2005; GALETTO et al., 2000; GUEDES et al., 2009; KIILL; DRUMOND, 2001).

The rate of apomixis was 8% (N = 25), significantly different from that found in natural pollination, 13.2% ($t_{obs} = 10.0 < t_{teor} = 2.06$). Self-compatible species of Fabaceae usually have some incidence of apomixis (BORGES, 2006; NOGUEIRA; ARRUDA, 2006), whereas in self-incompatible species this phenomenon does not occur (ETCHEVERRY; ALEMÁN, 2005; GALETTO et al., 2000; KIILL; DRUMOND, 2001). As with automatic self-pollination, apomixis can also promote fruit set when there is a low rate of natural pollination (BAWA; WEBB, 1984; FAEGRI; VAN DER PIJL, 1979).

Table 2. Breeding system of *Bowdichia virgilioides* Kunth (Fabaceae) in an area of Cerrado in Chapadinha, Maranhão State, Brazil.

	Flowers	Fruits
Control	114	15 (13%)
Xenogamy	25	9 (36%)
Self-pollination	25	9 (36%)
Automatic self-pollination	201	41 (20.4%)
Apomixis	25	2 (8%)

Reproductive success

In natural conditions the number of seeds per fruit was 3.5 (± 1.63 , N = 26), the seed/ovule was 0.43, and the resulting fecundity rate (CRUDEN, 1972) was 0.056 (Table 3), a value considered low for self-compatible species (WIENS et al., 1987). In an automatic self-pollination experiment with bagged flowers in bud, the number of seeds was 3.9 (± 1.96 ; N = 25), the seed/ovule was 0.47, and the resulting fecundity rate was 0.096 (Table 3). These results show that *B. virgilioides* is primarily autogamous, which is a common feature among many species of Fabaceae (BORGES, 2006; NOGUEIRA; ARRUDA, 2006).

Table 3. Reproductive success of *Bowdichia virgilioides* Kunth (Fabaceae) in an area of Cerrado in Chapadinha, Maranhão State, Brazil, in natural conditions.

	Control	Bagged flowers (automatic self-pollination)
Ovule/flower	8.2 \pm 1.32 N = 25	
Pollen grains/stigma	36.0 N = 25	28.4 N = 25
Seed/fruit	3.5 \pm 1.63 N = 26	3.9 \pm 1.96 N = 25
Seed/ovule	0.43	0.47
Fruit/Flower	0.13 N = 114	0.2 N = 201
Fecundity rate	0.056	0.096

Conclusion

Although the flowers of *B. virgilioides* are visited by a variety of animals with a great potential to pollinate, the dynamics of flower's opening allows the transfer of pollen grains to the stigma through of the automatic self-pollination. This process promotes the fruit set, since this species is self compatibility. Natural biotic pollination did not significantly increase the amount of pollen grains on the stigma in relation to the amount of pollen deposited by automatic self-pollination. All these data show that *B. virgilioides* is independent of pollinators for fruit set. However, only studies on the genetic diversity in natural populations will aid in understanding the balance between self- and cross-pollination and its consequent genetic structure.

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