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Research Paper

How does seed size of *Arachis pintoi* affect establishment, top-growth and seed production?

¿Cómo afecta el tamaño de la semilla el establecimiento, la biomasa aérea y la producción de semilla de Arachis pintoi*?*

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Abstract

The adoption of *Arachis pintoi* in mixed pastures in the humid tropics remains limited to specific success cases, mainly because of high seed cost. The search for methods to reduce these costs is a key challenge towards promoting wider adoption of this legume in livestock production systems in the tropics. One possible option is to select for smaller seeds, which would allow lower sowing rates, while ensuring similar plant numbers. Alternatively, high seed production costs could be offset by utilizing forage from seed production fields for hay or silage prior to seed harvest. This study evaluated the effects of seed size on crop performance of *A. pintoi* cv. BRS Mandobi in a tropical forage + seed production system, plus the effects of harvesting forage during the growth stage on seed production. Parameters measured were: ground cover, height, pest and disease incidence, total forage and leaf yield plus seed yield and seed sizes. Smaller seeds resulted in morphologically smaller plants and lower forage mass during the initial phase of crop establishment. However, seed size had no effect on ground cover at the end of the establishment period or on seed production and quality. Harvesting forage during the growth cycle had no effect on seed production. This indicates the possibility of harvesting forage from seed crops of *A. pintoi* during growth without jeopardizing seed yields as a means of offsetting high costs of seed production. However, the study has failed to provide conclusive evidence whether variation in seed size in BRS Mandobi is mainly genetic or a response to micro-environmental conditions. Further studies with individual plants from BRS Mandobi are necessary to determine the heritability of seed size.

Keywords: Aerial biomass, forage peanut, ground cover, seed quality.

Resumen

La adopción de *Arachis pintoi* en pasturas mixtas en el trópico húmedo se limita a casos específicos de éxito, principalmente debido al alto costo de la semilla. La búsqueda de métodos para reducir esos costos es un desafío clave para promover la adopción a gran escala de esta leguminosa en sistemas de producción pecuaria en el trópico. Una posible opción es seleccionar semillas más pequeñas, lo que permitiría tasas de siembra más bajas, pero un número de plantas similar. Además, la utilización de la masa forrajera de los campos de producción de semillas para heno o ensilaje puede compensar los altos costos de producción de semillas. Este estudio se realizó en Rio Branco, Acre, Brasil con el fin de: (1) evaluar el efecto del tamaño de la semilla en el establecimiento y la producción de *A. pintoi* cv. BRS Mandobi en un sistema tropical de producción de forraje + semilla, y (2) medir los efectos de la cosecha de forraje en la producción de semilla. Los parámetros evaluados fueron: cobertura del suelo, altura de planta, incidencia de plagas y enfermedades, producción de forraje total y rendimiento foliar; además, rendimiento y tamaño de semilla. Las semillas pequeñas produjeron igualmente plantas morfológicamente pequeñas

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y con menor masa de forraje durante el establecimiento. Sin embargo, el tamaño de la semilla no tuvo ningún efecto sobre la cobertura del suelo al final del período de establecimiento o sobre el rendimiento y la calidad de la semilla producida. La cosecha de forraje durante el período de crecimiento de *A. pintoi* no afectó la producción de semillas. Esto indica que la cosecha de forraje en los cultivos de semillas durante el crecimiento puede contribuir a compensar los altos costos de la producción de semillas. Los resultados no son concluyentes sobre si la variabilidad en el tamaño de la semilla en BRS Mandobi es debida principalmente a la genética o las condiciones microambientales. Se necesitan estudios con plantas individuales para inferir sobre la heredabilidad del tamaño de la semilla.

Palabras clave: Calidad de semilla, cobertura de suelo, maní forrajero, producción de forraje.

Introduction

Despite research and development efforts during the 1990s and 2000s (Argel 1994; Ramos et al. 2010; Assis and Valentim 2013), the adoption of forage peanut (*Arachis pintoi* Krapov. & W.C. Greg.) for use in mixed pastures in the humid tropics and subtropics remains limited to specific successful cases (Shelton et al. 2005). The greatest barrier to wider adoption is limited availability and high cost of propagation material, mainly seeds. The major challenge for expanding adoption is to increase the supply and reduce the cost of seed, as already pointed out more than 2 decades ago by Ferguson (1994).

High cost of seed production is due to the difficulty in harvesting the seeds, which are produced below the soil surface, like all species of *Arachis*, and seeds do not remain attached to the plant, as occurs with peanut (*Arachis hypogaea* L.) (Ferguson 1994). Seeds need to be sifted from the soil and recovered. In addition, seeds of *A. pintoi* are relatively large, when compared with other tropical forage species, and require a higher sowing rate to achieve acceptable plant numbers. A sowing rate of 12 kg of viable pure seeds/ha is recommended currently to establish mixed grass-*A. pintoi* cv. BRS Mandobi pastures (Abreu et al. 2012). Reducing the sowing rate, as long as there are adequate numbers of seeds per kilogram, would reduce sowing costs.

Variation in size of BRS Mandobi seeds does exist, as shown by Assis et al. (2013) in the Western Brazilian Amazon, and there are possibilities of intra-cultivar selection for this characteristic (Assis et al. 2016). In general, larger seeds have higher germination and seedling vigor than smaller seeds (Beckert et al. 2000), but this initial advantage disappears rapidly during the later stages of establishment (Bredemeier et al. 2001). However, how seed size affects establishment, aboveground dry matter yield and seed production of forage peanut remains unknown.

Another way for seed producers to offset the high cost of producing seed is to add value to the system, e.g. by grazing or cutting forage for hay or silage during the period of vegetative growth before harvesting a seed crop. Such a system could be considered as a forage + seed production system under integrated crop-livestock systems. Crop management is an important factor in seed production, since seed yield tends to decrease with increased frequency of harvesting forage (Bortolini et al. 2004; Awad et al. 2013). In Costa Rica, Argel (1994) found that frequent cutting intervals for forage (every 2 months) reduced seed production of forage peanut, and this effect occurred even at intervals as long as 8 months, with the response depending on the genotype.

The objectives of this study were to evaluate the influences of seed size on establishment, development and seed production of *A. pintoi* cv. BRS Mandobi in a monospecific stand, as well as performance of this legume in a dual-purpose production system (forage and seeds) in the Brazilian humid tropics.

Materials and Methods

The study extended from November 2008 to October 2009 at the Experimental Station of the Agroforestry Research Center of the Brazilian Agricultural Research Corporation – Embrapa, located in Rio Branco, AC, Brazil [10°01'34" S, 67°42'13" W (WGS 84); elevation 160 masl]. The climate is classified by Köppen as Awi (hot and humid), with maximum temperature of 31 °C and minimum of 21 °C, mean annual precipitation of 1,900 mm, mean relative humidity of 80% and a rainy season from October to May and dry season from June to September (Figure 1). The soil of the experimental area is an Oxisol, and chemical analyses at 0–20 cm depth are: organic matter, 1.3%; pH in water, 5.4; Ca, 1.6 cmol/kg; Mg, 0.4 cmol/kg; K, 0.1 cmol/kg; Al, 1.0 cmol/kg; P (Mehlich 1), 2.0 mg/kg; cation exchange capacity, 5.4 cmol/kg; and base saturation, 38.6%.

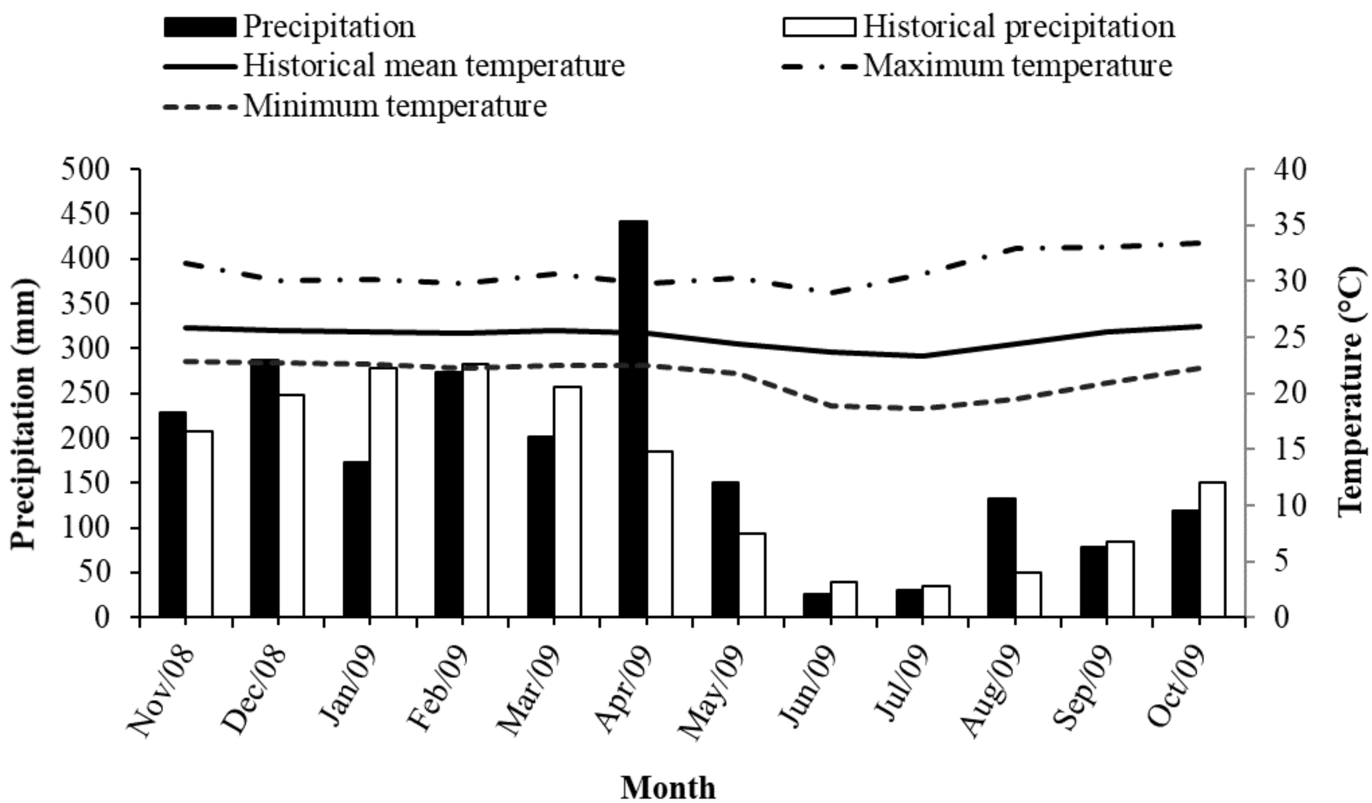

Figure 1. Temperature and precipitation during the experimental period (November 2008 to October 2009) and historical data (1969–2014) (INMET 2015).

The experimental design was a randomized complete block design, with treatment combinations of 3 seed sizes (small, medium and large) and either harvesting forage at 3 separate times or not harvesting forage before reaping the seed crop, with 4 replications. While *A. pintoi* is planted using the entire fruit, i.e. seed plus pod, we use the term 'seed' as a substitute for 'fruit', i.e. the propagation unit. The seed lot of *A. pintoi* cv. BRS Mandobi was harvested in July 2007 and stored for 17 months, a period considered long enough to overcome seed dormancy naturally without physical treatment. After this period, we measured pure seeds with a digital caliper and separated them into 3 size categories (treatments), based on the product of seed length by seed width: small, ≤61 mm^2; medium, 62–88 mm^2; and large, >88 mm^2. Seed numbers per kilogram, according to size categories, were: small, 10,070; medium, 6,250; and large, 4,700 seeds.

Seeds were sown on 13 November 2008 in 3 x 3 m plots, with 50 cm spacing between rows and 14 cm between plants within rows, in furrows at 3 cm soil depth, with 21 seeds/row for all treatments, which corresponds with sowing rates of 14, 22 and 30 kg/ha for small, medium and large seeds, respectively. Triple superphosphate (21.8 kg P/ha), potassium chloride (33.2 kg K/ha) and Fritted Trace Elements (40 kg FTE/ha) were applied at sowing.

The following aspects were examined: establishment period; leaf morphological characterization; forage and seed production (forage + seed evaluation); and seed quality (germination rate and seedling vigor), in order to evaluate the effects of seed size on forage peanut growth and its influence on morphological characteristics of plants plus seed production and quality. All statistical analyses, consisting of analysis of variance, regression analysis and comparison of means, were conducted using the Sisvar computer program (version 5.3) (Ferreira 2014).

Establishment period

The establishment period was from November 2008 to March 2009 and involved evaluation of the following parameters: incidence of pests and diseases; plant vigor; and flowering intensity. Incidence of pests and diseases was evaluated visually using the scale: 0 = no damage; 1 = slight damage on ≤50% of plants; 2 = slight damage on 51–100% of plants; 3 = moderate damage on ≤50% of plants; 4 = moderate damage on 51–100% of plants; and 5 = severe damage. Therefore, the score allocated was a combination of the extent of damage, in terms of the affected area within the plot, and the degree of its severity. Plant vigor assessment was based on a 1–10 scale, with: 1 = very poor vigor; and 10 = excellent vigor. Plant flowering intensity was estimated visually based on the percentage of plants with flowers: 0 = no flowering; 1 = 1–20% of plants flowering; 2 = 21–40%; 3 = 41–60%; 4 = 61–80%; 5 = 81–100% flowering. During the establishment period, ground cover was assessed using a 1 x 1 m wooden frame subdivided into smaller squares of 10 x 10 cm. Plant height was also determined, based on measurements at 3 different points in each plot, in order to obtain the average height of the stand.

Leaf morphological characterization

Morphological characterization of leaves occurred in a single evaluation, before the first forage harvest, in March 2009. The following measurements were made on 10 plants (one leaflet of each type per plant) in each replication of each treatment: basal leaflet length and width (BLL and BLW); and apical leaflet length and width (ALL and ALW). Petiole length (PL) was determined as the mean of measurements on 5 leaves. All variables were measured in millimeters (mm) with a digital caliper.

Forage + seed evaluation

To evaluate the potential for a forage peanut crop to yield both forage and seed, we harvested forage from half of the plots before harvesting the seed crop and compared this with the remaining plots where no forage was harvested during the production of the seed crop. Forage from the dual-purpose plots was harvested from a 1 m^2 area, at 2 cm above ground level on 3 occasions: 30 March (rainy season), 13 June (early dry season) and 19 October 2009 (dry season). Harvested forage was sorted into leaf and stem before drying in a forced-ventilation oven at 65 °C for 72 hours. Response variables measured were: leaf: stem ratio; forage dry mass (FDM); and leaf dry mass (LDM). For statistical analyses, experimental treatments were seed size (small, medium and large) and different harvesting times (rainy, early dry and dry seasons), in a split-plot arrangement, considering seed size as main plots, and presence or absence of harvesting and evaluations along the seasons as subplots. Ground cover values were square-root transformed to satisfy the analysis of variance assumptions.

Seed production was evaluated in October 2009, at the beginning of the rainy season, after the evaluation of forage production in all plots by retrieving seed from the top 10 cm of soil on 1 m^2 of area. Seeds were separated from the soil by hand-sieving followed by washing under running water. Subsequently, they were air-dried by natural ventilation.

Seed quality

Harvested seeds were sorted according to seed size, and their proportions in the total seed lot were determined in the plots that were not subjected to previous harvesting of forage. Limited resources prevented repeating these measurements on seed from plots where forage was harvested before seed harvesting. Seed quality was determined on samples of 300 seeds per treatment based on the emergence test, according to the Seed Testing Guidelines established by Brazilian law (MAPA 2009; 2013) after placing seeds in a forced-ventilation oven at 40 ºC for 14 days (Ferguson 1994) to break dormancy. Seed germination was assessed in trays with sterilized sand, with daily irrigation, under uncontrolled temperature and humidity conditions. Average environmental local temperature and relative humidity of the air during the emergence test were 27 °C and 85%, respectively. Germination rate was determined based on seedling counts at 7, 14 and 28 days after sowing. Additionally, the lengths of seedling shoots and roots were measured (cm) with a graduated scale, 7 days after emergence. The experimental design was a randomized complete block with 4 replications.

Results

Establishment period

There were no significant interactions between seed size and time after sowing on parameters except for ground cover. Pest and disease incidence, plant vigor, flowering intensity and plant height varied significantly with time after sowing (P<0.05) (Table 1), while seed size had significant effects on disease incidence and plant height overall and ground cover at some observations.

Table 1. Effects of seed size and time after sowing on pest and disease incidence, plant vigor, flowering intensity, ground cover and plant height during the establishment period of *Arachis pintoi* cv. BRS Mandobi.

Parameter	Seed size	Weeks after sowing					
		4	8	12	16	20	Mean
Pest incidence	Small	1.50	2.75	1.75	2.75	2.00	2.15
	Medium	2.00	2.75	1.50	2.75	2.00	2.20
	Large	2.50	3.75	2.00	3.00	2.50	2.75
	Mean	2.00	3.08	1.75	2.83	2.17	2.37
Disease incidence	Small	1.00	2.25	3.25	2.25	4.00	2.55 b[1]
	Medium	1.00	2.75	3.25	3.75	3.00	2.75 ab
	Large	1.25	4.00	3.50	3.75	4.00	3.30 a
	Mean	1.08	3.00	3.33	3.25	3.67	2.87
Plant vigor	Small	3.75	4.00	4.50	7.00	9.50	5.75
	Medium	4.50	5.00	5.25	8.00	10.00	6.55
	Large	4.75	5.00	5.50	8.00	10.00	6.65
	Mean	4.33	4.67	5.08	7.67	9.83	6.32
Flowering intensity	Small	0.25	1.00	1.00	2.00	3.25	1.50
	Medium	0.25	1.00	1.00	2.75	2.75	1.55
	Large	0.50	1.00	1.25	2.25	2.00	1.40
	Mean	0.33	1.00	1.08	2.33	2.67	1.48
Ground cover (%)[2]	Small	1.82 a	4.4 a	21.0 b	67.8 b	94.9 a	38.0
	Medium	1.82 a	19.6 a	52.1 a	92.5 a	100.0 a	53.2
	Large	7.29 a	23.7 a	55.1 a	92.5 a	100.0 a	55.7
	Mean	3.6	16.0	42.7	84.2	98.3	36.6
Plant height (cm)	Small	3.25	8.67	13.08	5.08	7.58	7.53 b
	Medium	3.83	8.92	15.33	7.17	11.83	9.42 ab
	Large	4.58	11.25	14.25	8.58	14.75	10.68 a
	Mean	3.89	9.61	14.22	6.94	11.39	9.21

[1]Means within columns followed by the same letter are not significantly different (P>0.05) by Tukey's test. [2]Data analyzed with transformation $x^{(1/2)}$. Pest and disease incidence and flowering: rating scale (0–5); vigor: rating scale (1–10) with higher values indicating higher levels.

Plant vigor and flowering intensity of forage peanut increased with time after sowing, with the coefficient of determination above 90% (Figure 2A). However, pest incidence was constant during the establishment period, so no model was fitted. Disease incidence tended to increase over time, becoming stable towards the end of the study, with coefficient of determination near 90%. Plant height, with coefficient of determination above 70%, showed a cubic model fit, with highest values at the third evaluation. The fitted models were: $-0.24x^2 + 2.01x - 0.47$, $R^2 = 0.90$ for disease; $0.42x^2 - 1.1x + 5.03$, $R^2 = 0.99$ for plant vigor; $0.6x - 0.32$, $R^2 = 0.94$ for flowering intensity; and $1.07x^3 - 10.66x^2 + 32.66x - 19.68$, $R^2 = 0.73$ for plant height.

Ground cover increased linearly during the establishment period, with coefficient of determination above 90% for all seed sizes. The fitted models were: $2.29x - 1.68$, $R^2 = 0.96$ for small seeds; $2.25x - 0.23$, $R^2 = 0.95$ for medium seeds; and $1.94x + 1.12$, $R^2 = 0.96$ for large seeds (Figure 2B).

Leaf morphological characterization

Seed size had significant effects (P<0.05) on all leaf morphological characteristics among established plants. Plants originating from small seeds presented smaller morphological characteristics than plants originating from large seeds (Table 2).

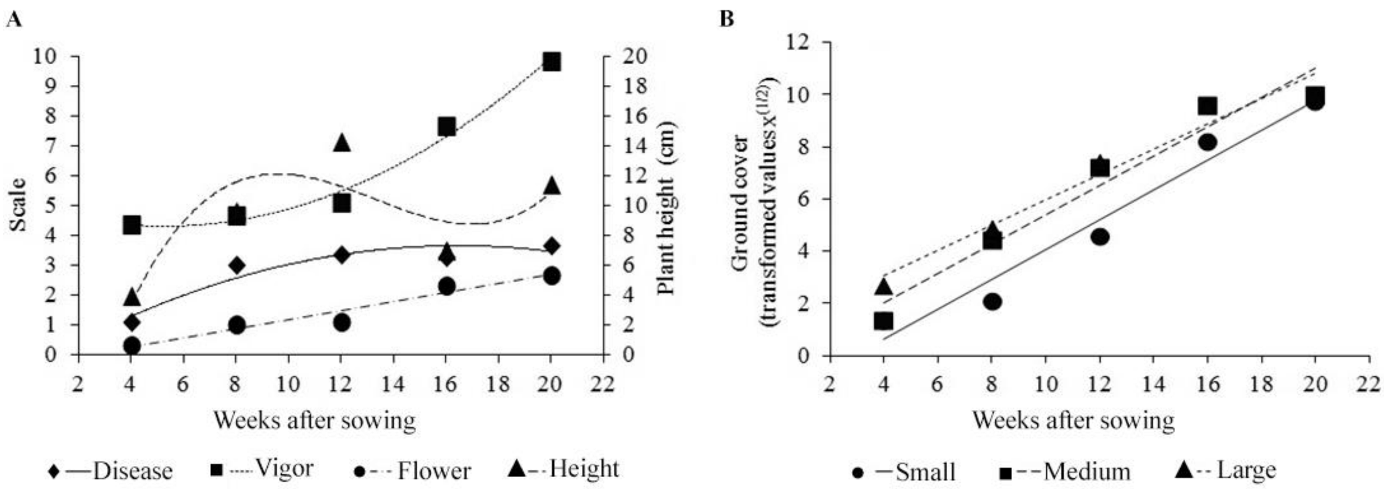

Figure 2. Effects of: **A**) time following sowing on disease incidence, plant vigor, flowering intensity and plant height of *Arachis pintoi* cv. BRS Mandobi (disease incidence and flowering intensity vary on a scale between 0 and 5, and plant vigor varies between 1 and 10); and **B**) time following sowing and seed size on ground cover.

Table 2. Effects of seed size on length and width of basal and apical leaflets and length of petiole of *Arachis pintoi* cv. BRS Mandobi at 120 days after sowing.

Parameter	Treatment	Length (mm)	Width (mm)
Basal leaflet	Small	31.0 b[1]	16.7 b
	Medium	34.1 ab	17.6 ab
	Large	37.5 a	19.2 a
P value		0.011*	0.028*
Apical leaflet	Small	33.8 b	20.3 b
	Medium	37.7 ab	21.3 ab
	Large	42.2 a	23.4 a
P value		0.005**	0.017*
Petiole	Small	39.7 b	
	Medium	44.0 ab	
	Large	47.4 a	
P value		0.022*	

[1]Values within columns and parameters followed by the same letter are not significantly different (P>0.05) by Tukey's test.

Forage + seed evaluation

Total forage dry mass (FDM) and leaf dry mass (LDM) production were significantly affected (P<0.05) by an interaction between seed size and season (Table 3). Stands established with small seeds produced lower amounts of both FDM and LDM than stands established with large seeds, during all seasons (rainy, early dry and dry). On the other hand, differences in FDM and LDM of stands established with small and medium seeds occurred only during the rainy season. Leaf:stem ratio differed only between harvesting seasons, with significant (P<0.05) reductions at successive harvests from the rainy to early dry and dry seasons (Table 3).

Table 3. Effects of seed size and season on total forage (FDM) and leaf (LDM) dry mass production and leaf:stem ratio (L:S) of *Arachis pintoi* cv. BRS Mandobi in pure stands.

Seed size	Season of the year			
	Rainy	Early dry	Dry	
	FDM (kg/ha)			Total
Small	2,663 Bb[1]	3,353 Ba	2,400 Bb	8,416
Medium	3,784 Aa	3,418 Ba	2,579 Bb	9,781
Large	3,729 Aa	4,290 Aa	3,636 Aa	11,655
Mean	3,392	3,687	2,872	9,951
	LDM (kg/ha)			Total
Small	1,695 Ba	1,523 Ba	908 Bb	4,126
Medium	2,451 Aa	1,714 ABb	859 Bc	5,024
Large	2,473 Aa	2,099 Aa	1,353 Ab	5,925
Mean	2,207	1,778	1,040	5,025
	L:S ratio			Mean
Small	1.81	0.88	0.69	1.13
Medium	1.89	1.09	0.59	1.19
Large	2.02	1.01	0.72	1.25
Mean	1.91 a	0.99 b	0.67 c	1.19

[1]Values within columns and parameters followed by the same upper-case letters and within rows followed by the same lower-case letters are not significantly different (P>0.05) by Tukey's test.

Seed production was independent of harvesting management and seed size, with a mean of 2,449 kg/ha. The percentages of small, medium and large seeds produced by plots sown with seeds of different sizes were: 20, 77 and 3% for small; 11, 77 and 12% for medium; and 17, 68 and 15% for large seeds, respectively. However, it was not possible to perform statistical analyses on these data because the seeds belonging to each treatment were mixed, losing individual data for different replicates.

Seed germination rate and seedling vigor

There was no significant (P>0.05) effect of seed size on seed germination rate and seedling vigor, based on lengths of the shoots and roots at 7 days after emergence. The mean lengths of shoots of seedlings originating from small, medium and large seeds were 9.79, 10.16 and 10.25 cm, and the mean lengths of roots were 5.50, 5.27 and 6.04 cm, respectively. Seed size at planting did not affect viability of seed produced (P>0.05), with germination percentages of 13.7, 16.6 and 18.1% for seed originating from small, medium and large seeds, respectively.

Discussion

The failure of harvesting management to affect seed production of *A. pintoi* was an important finding as it justifies harvesting forage from a seed crop prior to collecting seed. This would provide an additional source of high quality forage to supplement livestock in periods with restriction of forage supply (Ladeira et al. 2002; Awad et al. 2013) or an additional source of income through sale of the forage.

Brum et al. (2009) and Demétrio et al. (2012) suggest that management practices can increase biomass production of forage species, especially legumes in intercropping systems with two or more harvests, by promoting the appearance of new shoots, which results in higher regrowth rates. In contrast, production of seeds and grains may decrease, mainly because regrowth interval after the final harvest is too short to allow for complete seed development. Therefore, forage + seed production systems require different management strategies for production of seed and forage simultaneously for most species (Bortolini et al. 2004; Dávila et al. 2011). This is particularly important in the case of forage peanut BRS Mandobi, which has a cycle of 128 days between flower setting and seed maturity. Additionally, depending on the *A. pintoi* genotype, seed production may be influenced by crop management strategies, mainly related to harvest intensity and frequency (Argel 1994; Ferguson 1994; Dávila et al. 2011). It was interesting that forage harvesting times we chose varied from March to October, but there was no effect on the seed yields obtained.

The fact that plants established with large seeds had higher incidence of diseases is possibly due to their taller and more closed canopy conditions, providing favorable environmental conditions for survival and multiplication of plant pathogens, with higher humidity between the canopy and the soil and dead organic matter from crop residues (Assis et al. 2011). Increases in plant height during the early part of the establishment period were expected as plants were growing rapidly, while the declines after 12 weeks resulted from partial lodging due to increase in size and mass.

As all treatments had similar plant numbers, different seed sizes resulted in different seeding rates (kg/ha), which may have resulted in the lower ground cover observed during the establishment when small seeds were sown. If the same seeding rates had been employed for all treatments, this would have resulted in higher plant densities in treatments with medium and small seeds and areas established with these seeds would be expected to achieve higher levels of ground cover in the same period after planting than areas planted with large seeds. Since seed size did not affect ground cover at the end of the establishment period, around 120 days after sowing, this suggests that the lower seeding rates employed with smaller seed could be quite satisfactory. However, even with satisfactory ground cover and plant vigor at the end of the establishment period, the production of forage in areas established with small seeds was lower during all seasons, when compared with areas established with large seeds.

The smaller leaves on plants originating from small seeds had a negative effect on initial speed of plant growth and establishment of ground cover and increased the opportunity for emergence of weeds that compete directly for light, water and nutrients. Larger leaves tend to have higher energy expenditure for growth than smaller leaves, thus demanding greater availability of resources, such as light and nutrients (Milla and Reich 2007). Even though the use of smaller seeds for crop establishment may result in plants with smaller leaves, the adjustment of seeding rate with higher plant densities could compensate for this initial disadvantage (Andrade et al. 1997).

The failure of seed size to affect leaf:stem ratio showed that leaf emission is independent of seed size, but greatly affected by climatic conditions. During the approximately 100-day regrowth period evaluated in the dry season, leaf mass was 50% lower than during the rainy season, while total forage mass was only 15% lower, indicating greater presence of stem tissue. This pattern of crop growth is a response to climate variation during the year. Leaf:stem ratio is important in studies with forage species, since it directly affects sunlight interception and plant metabolism, thus influencing seed and forage production. It also affects liveweight gain of grazing animals, because higher leaf percentage improves pasture nutritive value and increases digestibility and forage dry matter intake (Rocha et al. 2007; Ribeiro et al. 2012; Awad et al. 2013).

While there is a dearth of information regarding the inheritance and mode of action of seed size in *A. pintoi* in the literature, *A. hypogaea* shows high heritability for seed size, with potential for indirect selection for multiple characters positively and negatively related with this trait (Kotzamanidis et al. 2006; Sikinarum et al. 2007). This has been observed for grain species, suggesting the action of a low number of genes with additive effects, which allows selection for this character in the first segregating generations (Lopes et al. 2003; Hakim et al. 2014). The results of our study showed that size of seeds at planting had little effect on sizes of seeds produced. However, there was some variation, which may have genetic and/or micro-environmental causes. While the proportion of variation resulting from each of these effects could not be determined in this study, other studies with forage peanut have shown that there is intra-cultivar variation for BRS Mandobi. Assis et al. (2016) demonstrated that there is genetic variation for seed size within BRS Mandobi by evaluating individual plants. Later, Azevedo (2017) confirmed the existence of genetic variability between progenies selected from individual plants of BRS Mandobi, and estimated average heritability for weight of 100 seeds of 0.89. However, additional studies are needed to better understand the mechanisms of seed size inheritance in *A. pintoi*.

Quality of seed produced was not affected by seed size at planting, although Beckert et al. (2000) and Perin et al. (2002) suggested that larger seeds tend to produce more developed and vigorous seedlings. According to Bredemeier et al. (2001), the ability of large seeds to produce larger seedlings than small seeds is more pronounced under stress conditions and in early stages of seedling development, and this effect tends to disappear by the end of the establishment period, with no effect on the resulting seed yield. We observed a similar phenomenon in this study among areas of *A. pintoi* planted with different seed sizes, which showed no differences in ground cover at the end of the establishment period, leaf:stem ratio and seed production and quality. As seed size did not influence germination rate, using a standard planting rate would result in sowing 61% more small seeds per unit area than with medium seeds, resulting in higher planting densities, thus speeding the establishment process and reducing weeding costs. Alternatively using lower sowing rates with small and medium seeds would reduce seed costs, while achieving similar planting densities as compared with the use of large seeds. This is an important consideration because availability of seeds in the world market is low and the unit cost is very high (around US$ 40/kg), which limits wider adoption of this technology.

Germination rates below 20% obtained in the present study were due to the seed dormancy mechanism present in *A. pintoi*, which possibly has both physiological and physical components (Ferguson 1994; Assis et al. 2015). Although low germination rates were obtained, they were not related to seed size from which the plant originated and appropriate adjustments in the seed dormancy-breaking method would most likely result in substantially higher seed germination rates. We tested the ungerminated seeds from the germination tests using tetrazolium and verified that viability of all seed size lots was the same ($P>0.05$), with an average of 90.3%.

Forage + seed production systems are of great interest, because they enable farmers to diversify and increase their sources of income with the production of high quality forage, while maintaining their capacity to produce seeds from the same area. In fact, similar systems are common in Southern Brazil, mainly for production of grain as well as forage for feeding animals, especially during the dry season (Fontaneli et al. 2009; Meinerz et al. 2012). Forage + seed production systems are recommended in tropical and semi-arid regions of Central and South American countries, Southern Africa, Asia and Australia (Peters et al. 2001; Whitbread and Pengelly 2004). These systems can also become a viable alternative for high quality forage and seed production in tropical areas with adequate environmental conditions for growth and persistence of *A. pintoi*. The dry matter yields of forage would present a significant resource for sale or use as forage on the farm, and should either increase returns or enable seed to be marketed at a cheaper price.

Conclusions

While seed size at planting had no effect on most parameters, including seed yield in a mono-specific pasture, these findings might not relate to results from a grass-legume pasture where competition from the grass might be strong. The lower initial crop establishment and forage mass in areas cultivated with small seeds support this possibility. Further studies on the effects of different seed sizes and seeding rates on forage peanut establishment and forage production in mixed stands seem warranted to determine if seed number and not seeding rate/ha is the critical factor in establishment.

Harvesting of forage as a by-product during the growth cycle prior to harvesting seed seems not to affect seed yields, so this strategy could be a mechanism to offset the high costs of seed production in *A. pintoi.*

Results from this study have failed to provide conclusive evidence whether variation in seed size of BRS Mandobi is mainly genetic or a response to micro-environmental conditions. Further studies with individual plants are necessary to attempt to clarify the factors relating to heritability of seed size.

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