How Bovine Livestock Affects Seed Rain in Subtropical Climate Forest

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Received: April 14, 2019      Accepted: May 23, 2019      Online Published: July 15, 2019
doi:10.5539/jas.v11n10p240          URL: https://doi.org/10.5539/jas.v11n10p240

The research is financed by Coordenação de Aperfeiçoamento de Pessoal de Nivel Superior-CAPES.

Abstract
Seed rainfall may be one of the mechanisms most impacted by livestock production in forest remnants. The trampling and cattle grazing alter dynamics, structure and floristic composition of the forest. The present work characterized the seed rain in forest remnants and the possible impacts of the presence and the management regime with different cattle stocks in the Pampa biome, southern region of Brazil. We compared three areas in Seasonal Forest remnants with a management history of 43 years: cattle exclusion area (A1); area with a cattle stock of 0.5 ua ha⁻¹ (A2); and area with cattle stock of 1.0 ua ha⁻¹ (A3). The seed rain was collected quarterly in 16 collectors (1 × 1 m²) per area for 24 months. The seeds were counted and identified according to external morphological characteristics, habit and dispersion syndrome. Shannon diversity index, submitted to the Hutcheson test, Pielou evenness, expected diversity and floristic similarity were determined. The results indicated that the impacts caused by livestock to seed rain were more significant in A3 where a quantitative reduction in the number of seeds was observed, probably due to the low number of plant individuals that make up the community.

Keywords: animal stocking, cattle breeding, dispersal syndrome, ecological indexes, pampa biome

1. Introduction
The horizontal distribution of a particular species of the forest community depends on its colonization success, determined by the establishment and dispersion of its propagules (Chazdon, 2014). In this way, the relationships that plant individuals establish with the environment and with anthropic interventions can compromise the colonization and establishment of species (Hutchings, 1989; Begon et al., 2006).

Among potentially impacting economic activities in forest remnants, extensive livestock farming may hinder the establishment and development of natural regeneration (Michels et al., 2012; Brancalion et al., 2016), altering the ecological dynamics and community composition (Marchior, 2004; Eaton et al., 2016; Araújo et al., 2017).

Cattle may compromise the conservation of forest remnants by consuming flowers and seeds (Eaton et al., 2016). Cattle indirectly affect the interaction with local dispersers by modifying floral traits (Vázquez, 2002) and their diet overlaps (Eaton et al., 2016). In addition, trampling can compact the soil and hamper the establishment of natural regeneration (Pillar & Quadros, 1997; Sampaio & Guarino, 2007).

Knowledge of the floristic composition and the dispersion syndromes enables understanding the structure and successional stage of the vegetation (Chazdon, 2014), in addition to the degree of conservation or perturbation of the environment (Pinã-Rodrigues & Aoki, 2014; Capellesso et al., 2015). Therefore, characterizing seed rain in
forests is fundamental for understanding recruitment processes, abundance, spatial distribution, density and species richness (Vieira & Gandolfi, 2006).

The southern region of Brazil differs from the rest of the territory because of its subtropical climate. Its paleoclimatic history generated peculiar characteristics of phytophysiognomies in the two biomes of its range: Atlantic Forest and Pampa. There are mosaics of well-defined country and forest vegetation in both biomes. The presence of country vegetation enabled the development of cattle raising activity, especially in the Pampa biome. Its importance is not only economic, but also sociocultural.

The Pampa biome is currently the second most threatened in Brazil and there is a lack of scientific studies that can support restoration and sustainable management plans (Roesch et al., 2009).

The forest remnants are impacted by the advancing commodity productive area, mainly soybean crops, with a reduction in remnant size and connectivity between habitats (Silveira et al., 2017). In spite of this, they present fundamental ecosystem services; most of the springs and water courses of the biome are found in the forest remnants, in addition to serving as refuges for wild fauna and being maintainers of the floristic diversity, its products and services (Rovedder et al., 2016).

In view of the above, the present work aimed to characterize the seed rain in forest remnants in the Pampa biome of southern Brazil, verifying the possible impacts caused by the presence of cattle inside this forest.

2. Material and Methods

The study was carried out in a Seasonal Forest remnant in the Pampa biome, Rio Grande do Sul state, in the southern region of Brazil. The region presents Oxisols in association with Regosols and Gleisols (Streck et al., 2018; EMBRAPA, 2013). The altitudes range between 50 and 200 masl. The climate is “Cfa” according to the Köppen classification, subtropical humid, without a dry season, with an average annual temperature of 22 °C. The average annual precipitation varies between 1300 and 1800 mm year⁻¹, with higher values recorded in winter (Alvares et al., 2013).

The vegetation cover is composed of fields and seasonal forests (Marchiori, 2004). The socioeconomic profile is predominantly characterized by small and medium-sized rural properties, based on the commercial cultivation of soybean, corn and extensive livestock farming (Instituto Brasileiro de Geografia e Estatística [IBGE], 2014).

2.1 Characterization and Usage History of the Study Area

The property has belonged to the same family for 49 years, which enabled gathering the management history of the place. The remnant forest is located at the coordinates 28°16′42.14″ S; 55°13′14.14″ W and has 57 ha, with eight springs and water courses that flow into the Piratini River, a tributary of the Uruguay River. In 1975, the remainder was divided into three areas (Figure 1), which in the present study configured into the following treatments:

Area 1 (A1): part of the forest remnant surrounded and isolated from the entrance of cattle 43 years ago to preserve springs. In the present study we have considered it as a reference area.

Area 2 (A2): part of the forest remnant managed with an animal stock of 0.5 ua ha⁻¹ for 43 years.

Area 3 (A3): part of the forest remnant managed with an animal stock of 1 ua ha⁻¹ for 43 years.
2.2 Sampling
A grid of 25 m² (5 × 5 m) was superimposed on the remnant. We established transects accompanying the toposequencing for each area, in which 16 sample plots were drawn in each area. We georeferenced the central points of each plot and installed a seed rain catch per plot, totaling 16 collectors/area. Each collector (1 m²) was fixed to 20 cm of soil height. The collectors located in areas A2 and A3 were surrounded with barbed wire to prevent livestock access.

The material was collected quarterly for 24 months, from January 2015 to December 2016. Seed and fruit sorting was carried out at the Núcleo de Estudos e Pesquisas em Recuperação de Areas Degradadas (NEPRADE) from Universidade Federal de Santa Maria, Brazil.

We performed luminosity readings with a HCVET HT-8318 digital luximeter at ground level and quarterly, considering the extremities and center of the sample plot, adding five readings per plot (Table 1).

Table 1. Luminosity rate in subtropical forest area excluding livestock (A1); (A2) and stock at 1.0 ua ha⁻¹ (A3), south of Brazil

| Area | A1   | A2   | A3   |
|------|------|------|------|
| Lum (lux) | 249.60 a* | 389.04 b | 328.1 b |

* Values not followed by the same letter in the line differed significantly by Student’s t test (P < 0.05).

2.3 Data Analysis
We counted and identified the collected seeds according to external morphological characteristics and taxonomic classification at the family, gender and species levels. When identification was not possible, the propagules were grouped into morphospecies (Venzke et al., 2014; Piña-Rodrigues & Aoki, 2014; Scociti et al., 2016). We used the term morphospecies during the course of the work for the sum of identified and unidentified species in order to standardize the nomenclature.

The dispersion syndromes were classified according to Van Der Pijl (1982) as: zoochoric for those that presented dispersion characteristics by the frugivorous fauna, especially drupaceous fruits, berries and seeds with aril; anemochoric, when they presented structures or wings that enabled the propagules to glide; or autochoric, when the structure of the fruit presented a mechanical dispersion characteristic such as an “explosion”.

The Shannon diversity index and Pielou evenness were calculated (Magurran, 1988). The Shannon diversity values (logarithmic basis) were compared by the Hutcheson test, at 5% probability of error (Hutcheson, 1970).
In relation to the total number of seeds, the assumptions of normality and the distribution of errors were verified by the Shapiro-Wilk and Bartlett test.

When the assumptions were not met, the non-parametric Kruskal-Wallis test (p < 0.05) was performed using the Action software (Equipe Estacamp, 2014). The rarefaction curve was elaborated with a 95% probability of confidence (Gotelli & Colwell, 2001) and the similarity between areas, we used cluster analysis with Jaccard similarity measures (Müller-Dombois & Ellenberg, 1974) both analysis were performed in PAST software, version 1.79 (Hammer et al., 2001).

3. Results

3.1 Morphospecies Distribution

20,661 propagules were sampled from January to December 2015, distributed in 126 morphospecies (identified and unidentified species), considering the three study areas (A1, A2 and A3) (Figure 2). We collected 18,012 propagules of 136 morphospecies in 2016, considering the three study areas (Figure 2).

![Figure 2. Total number of seed rain propagules during 24 months (2015 and 2016) in a Subtropical Forest, southern Brazil](image)

We counted 59 morphospecies in A1 in 2015. *Ilex paraguariensis*, *Cordia trichotoma*, *Ficus luschnathiana* and *Apuleia leiocarpa* accounted for 38.12% of the total collected seeds. A2 presented 53 morphospecies. *Helietta apiculata*, *C. trichotoma*, *I. paraguariensis* and *Jacaranda micrantha* added 41.77% of seeds collected. A3 presented 60 morphospecies, with *C. trichotoma*, *A. leiocarpa* and *F. luschnathiana*, accounting for 45.93% of the total. In 2015, the number of seeds collected in the three study areas did not statistically differ by the Kruskal-Wallis test (p < 0.05) (Table 2).

Table 2. Seed rain (2015 and 2016) in seasonal forest with livestock excluded (A1); 0.5 uha⁻¹ of livestock capacity (A2) and 1.0 uha⁻¹ of livestock capacity (A3) in South Brazil

| Area  | Number of seeds | Number of seeds |
|-------|----------------|----------------|
|       | 2015           | 2016           |
| A1    | 6896a*         | 8266a          |
| A2    | 8790a          | 5579ab         |
| A3    | 4975a          | 4157b          |

Note. *Differences of letters in the column indicate that there is significant difference at the level of 5% probability of error by the Kruskal-Wallis test.

In 2016, A1 presented 51 morphospecies. The species with the highest number of seeds collected were *C. trichotoma*, *Balfourodendron riedelianum*, *A. leiocarpa* and *F. luschnathiana*, accounting for 39.04% of the total. A2 presented 57 morphospecies. The species with the highest number of seeds were *C. trichotoma*, *Cedrela fissilis*, *Cordia americana*, *Luehea divaricata* and *F. luschnathiana*, accounting for 33.77% of the total. A3 presented 50 morphospecies, among which *C. trichotoma*, *Brunfelsia australis* and *C. fissilis* added 32.64% of the total collected seeds. In 2016, the number of seeds sampled at A1 differed significantly from A3 by the Kruskal-Wallis test (p < 0.05) (Table 2).
It should be noted that *C. trichotoma* presented 3145 seeds in 2015, and 2636 seeds in 2016, being the most abundant species in the seed rain in the three areas for the two years of the study.

Lifestyle and Dispersion Syndrome: Seeds of arboreal tree species predominated in the two years of evaluation in all areas (45% in 2015 and 47% in 2016). The predominant dispersion syndrome was zoochoric in both evaluation years in the three study areas (Figure 3).

![Figure 3. Number of morphospecies with zoochoric (ZOO), anemochoric (ANE) and autochoric (AUT) morphospecies present in the seed rain in a subtropical forest area with cattle exclusion (A1); stock rates of 0.5 ua ha⁻¹ (A2) and 1.0 ua ha⁻¹ (A3), southern Brazil.](image)

### 3.2 Ecological Indices

The Shannon diversity index did not show significant differences between the three study areas in each period (2015 and 2016), by the Hutcheson test at 5% probability of error (Table 3). Figure 4 shows the expected seed rainfall richness for each study area.

| Indexes          | 2015          | 2016          |       |
|------------------|---------------|---------------|-------|
|                  | A1            | A2            | A3    | A1            | A2            | A3    |
| Shannon (H)      | 3.263a*       | 3.068a        | 3.000a| 3.193a        | 3.356a        | 3.231a|
| Equability (J)   | 0.8002        | 0.7727        | 0.7327| 0.8122        | 0.8302        | 0.8259|

*Note.* *Equal letters on the same line indicate that there is no significant difference at the 5% probability level of error by the Hutcheson test.*

![Figure 4. Expected richness in seed rain in 2015 and 2016, in subtropical forest area excluding livestock (A1); cattle stock of 0.5 ua ha⁻¹ (A2) and cattle stock of 1.0 ua ha⁻¹ (A3), south of Brazil.](image)
3.3 Floristic Similarity

The composition of seed rain for the year 2015 presented high similarity between the areas according to the Jaccard index (Figure 5). A1 and A2 were more similar in comparison to A3. In 2016 (Figure 5), A1 and A3 areas presented greater morphospecies similarity in seed rain, although with quite different quantitative contributions from A2.

![Figure 5. Jaccard floristic similarity for seed rain in 2015 (a) and 2016 (b) in subtropical forest area excluding livestock (A1); cattle stock of 0.5 ua ha⁻¹ (A2) and cattle stock of 1.0 ua ha⁻¹ (A3), south of Brazil](image)

4. Discussion

4.1 Distribution of Morphospecies

The highest seed deposition occurred in the months corresponding to the growing season in southern Brazil (September to March) (Figure 2), when the temperature averages do not exceed 22 °C in spring and are higher than 30 °C in summer. There is no rain restriction in this period, since the a Cfa climate does not present a dry season (Alvares et al., 2013). The seasonality in the Brazilian subtropics occurs by temperature variation without a dry season, which differs from the seasonality in tropical Brazil (Budke et al., 2005; Venzke et al., 2014; Capellessso et al., 2015).

Regarding the number of seeds per species, we highlighted *I. paraguariensis* in A1 and A2 areas in 2015. In analyzing the same area, Stefanello (2016) observed low individual sampling of the species in natural regeneration. Considering this information, it is suggested that despite presenting a large number of seeds in the collectors, the species does not present the same abundance of individuals when analyzed in the regeneration of the three study areas. The *I. paraguariensis* propagules in the seed rain probably come from nearby (allochthonous), since the species presents/displays zoochoric dispersion.

The input and output seed flow between remnants is of great importance, since it is through this flow that we can predict the potential of establishing populations in a certain place (Capellessso et al., 2015; Lopes et al., 2010), especially in areas of natural vegetation that suffer some kind of anthropic interference, as in the case of the present study.

In relation to the temporal distribution in the year 2016 (Figure 2), seed deposition in the winter/spring (July to September) and spring/summer (October to December) periods in the three areas was influenced by local climatic atypicality. According to data from the Instituto Nacional de Meteorologia (INMET), the year 2016 presented an atypical rainfall regime for the region, with precipitation above average in a few months (February, March, April and October, recorded precipitations between 190 and 320 mm) and precipitations well below average for other months (January, May, June, July and August). This condition may have contributed to the reduction in the number of collected seedlings and to the period inversion of seed deposition peaks.

There is a record on the influence of the rainfall regime on the phenology of subtropical forest species in southern Brazil (Felippi et al., 2012; Scocri et al., 2016). In addition, high precipitation can damage to flowers and fruits, also impair the activity of pollinators and dispersers (Jackson, 1978; Kageyama & Piña-Rodrigues,
Furthermore, prolonged droughts compromise the reproductive cycle and production of flowers and fruits (Piña-Rodrigues & Aoki, 2014).

The significant difference between A1 and A3 for the number of seeds in 2016 may be a consequence of an interaction between the climatic atypicity of that year and the intensity of management in A3 (Table 3). In the present study, we observed a decrease in the number of seeds and the number of seeds per hectare, respectively. In addition, the presence of livestock may affect the presence of dispersers due to the diet overlap, modifying floral traits of the existing species (Vázquez, 2002; Eaton et al., 2016). Thus, the higher animal stock for 43 years in A3 contributed to affecting the regeneration mechanisms and altered the vertical structure of the forest, reducing sub-forest strata. This can be verified by the light incidence (Table 1) due to the lower number of plant individuals in the community. The area consequently presents lower resilience and greater susceptibility to environmental variations, such as extreme weather events.

The highest amounts of seeds of arboreal species found in all areas in the two years of evaluation can be considered as a positive indicator. According to Martini and Santos (2007), species of arboreal habitats attract and favor the permanence of pollinating and dispersing animals in the local promoting greater ecological balance in the community.

The number of C. trichotoma seeds in the two years of evaluation may be related to the species phenology, since it presents abundant flowering between February and June (Reitz et al., 1988; Felippi et al., 2012). Nevertheless, the species’ number of seeds was affected by the atypicality of the precipitation regime in the year 2016. As C. trichotoma is a species that occupies the upper strata of the forest formations and presents anemochoric dispersion, its flowers, fruits and seeds are subject to the effects of excessive rainfall. According to Felippi et al. (2012), the species flowers show greater sensitivity in the rainy season due to the impact of raindrops which damage the floral buds, in turn damaging the entire reproductive process.

A. leiocarpa is one of the species with the greatest abundance in the seed rain in 2015 and 2016, and in addition to I. paraguariensis did not present expressivity in the development of the natural regeneration of areas with cattle (A2 and A3), according to a floristic survey carried out in the same areas by Stefanello (2016). It is assumed that these species are more susceptible to the impacts of livestock management, since the development of a species in the regenerating stratum is related to seed rain and is determined by the seed rainfall density and of the seedling density (Simpson, 1989).

### 4.2 Scattering Syndrome

The predominance of zoochory in seasonal forest in southern Brazil (Figure 3), has been reported by several authors (Scipioni et al., 2011; Giehl et al., 2007; Scipioni et al., 2013; Sccoti et al., 2016). This also seems to be a typical pattern of forests in tropical Brazil. According to Fenner (1985) and Piña-Rodrigues and Aoki (2014), the high amount of zoochoric species is indicative of plant communities in more advanced succession stages.

However, finding values between 31 and 37% of anemochoria in formations with a more advanced succession stage, as in the case of areas A2 and A3, can be a response to the disturbances caused to the vegetation by the cattle management during the four decades. The presence of anemochoria can be considered as an indication of environmental disturbance in already established forest formations, since wind dispersion benefits these conditions (Piña-Rodrigues & Aoki, 2014). The luminosity values for the respective areas (Table 1) show that the forest cover is more spaced in these places, which would justify the anemochoria values found in this forest formation.

### 4.3 Diversity Indices

There were no significant differences for Shannon diversity in the two years. However, the distribution of morphospecies in A1 for the year 2015 showed to be more heterogeneous in relation to areas with livestock, based on the Pielou evenness index. For the year 2016, A2 showed to be the area with more heterogeneous distribution in relation to the others.

The expected species richness (Figure 4) demonstrates a curve-stabilizing behavior in all areas (Magurran, 2004; Kageyama & Piña-Rodrigues, 1993; Felfili et al., 2011). In addition, there is an inversion behavior trend of the seed rain from the year 2015 to the year 2016 for the evaluated areas. The seed rain in A2 suffered greater oscillations in relation to the atypical precipitation, probably due to the presence of native field patches in the interior of the sentence fragment (Figure 1). This discontinuity of the forest area in A2 contributes to the action of wind (Giehl et al., 2007) and rain (Vieira & Gandolfi, 2006; Felippi et al., 2012; Piña-Rodrigues & Aoki, 2014) in the interior of the forest, unlike A1 and A3, which have a continuous forest formation, considering only the external physiognomy.
4.4 Floristic Similarity

Changes in floristic similarity between the analyzed areas in a short time may be related to several factors; among them climatic factors, such as the case of the atypical precipitation in 2016. Extreme conditions such as excessive rainfall or prolonged droughts with cleared gaps are disturbances that influence the dynamics of the forest community (Giehl et al., 2007) in terms of quantity and quality of flowering and fruiting of species and seed dispersion (Piña-Rodrigues & Aoki, 2015). These disturbances may directly influence the dissimilarity of areas that may have been closer in floristic terms (Giehl et al., 2007).

In the location under study, we observed that the oscillations in the rainfall regime caused changes in cattle behavior and diet, causing the native field areas surrounding the A2 and A3 areas to decrease their biomass, resulting in a lower supply of food for cattle. This factor may have influenced the animals to look for food inside the forest, intensifying the herbivory and trampling in the local. It also may have contributed to the change in similarity in the evaluation period. The management history and the in loco observations carried out in different years with unequal climatic situations, as in the case of the present study, provide such inferences.

5. Conclusion

The presence of cattle within the forest remnant acts effectively as a filter for the dynamics and composition of seed rain.

The higher animal stocking directly impacts the number of seed rain propagules.

Increased stocking reduces ecosystem resilience, making the forest community more susceptible to other disturbances, such as extreme weather events.

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