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Alellopathic effects of aqueous extracts from *Bambusa vulgaris* Schrad. Ex J. C. Wendl. on seed germination and vigor from *Lactuca sativa*

Danielle Roballo de Moura[1], Emmanoella Costa Guaraná Araujo[2], César Henrique Alves Borges[3*], Luan Henrique Barbosa Araújo[3], Tarcísio Viana de Lima[4], Thiago Cardoso Silva[2] and Camila Costa da Nóbrega[5]

[1]Engenharia Florestal, Universidade Federal Rural de Pernambuco, R. Manuel de Medeiros, s/n - Dois Irmãos, Brazil.
[2]Programa de Pós-graduação em Ciências Florestais, Universidade Federal Rural de Pernambuco, R. Manuel de Medeiros, s/n - Dois Irmãos, Brazil.
[3]Programa de Pós-graduação em Ciências Florestais, Universidade Federal Rural de Pernambuco, R. Manuel de Medeiros, s/n - Dois Irmãos, Brazil.
[4]Departamento Engenharia Florestal, Universidade Federal Rural de Pernambuco, R. Manuel de Medeiros, s/n - Dois Irmãos, Brazil.
[5]Programa de Pós-Graduação em Ciência do Solo, Cidade Universitária, s/n - Castelo Branco III, João Pessoa - PB, 58051-085, Brazil

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The aim of this work was to evaluate allelopathic effects of aqueous extracts from *Bambusa vulgaris* leaves and culm on *Lactuca sativa* L. germination. Plant material was collected at Dois Irmãos State Park in Recife, Brazil, and analysis was performed at Laboratory of Forest Seeds Analysis at Rural Federal University of Pernambuco. Experiments were conducted under natural conditions using leaf and culm extracts at concentrations of 100, 75, 50 and 25%, using distilled water as control. Seed germination was evaluated for seven days. Percentage of germination (%G), speed index of germination (SIG), mean time of germination (MTG), and mean speed of germination (v) were determined. Aqueous extract of *B. vulgaris* leaves negatively influenced percentage of germination and speed of germination index of *L. sativa* seeds. Control treatment showed the best means, being statistically superior than other treatments that did not differ among each other. Considering *L. sativa* as a bioindicator, it is possible that allelopathic effects might significantly interfere on other species establishment and germination. This is due to substances present on plant residues deposited or incorporated to the soil annually. Finally, the inhibitory allelopathic behaviour of the germination process of *L. sativa* seeds in response to aqueous extracts of *B. vulgaris* was observed in all parts and concentrations tested.

Key words: Allelopathy, allelochemicals, culm, bioindicator, *Lactuca sativa, Bambusa vulgaris.*

INTRODUCTION

In the last years, with traffic development, increase in human activity and strengthening of international business, communication between biological species from different habitats has become more frequent. Some species were strongly adaptable to the environment and could grow and spread rapidly in the new environment, which affected local economy, ecology and society. Nowadays, allelopathy has been considered the main factor influencing invasion and dissemination of exotic plants (Chengxu et al., 2011).

Dois Irmãos State Park (PEDI) is a Full Protection Conservation Unit located in Recife/PE, Brazil. In the park, there are some species outside their natural habitat fixed on an Atlantic forest fragment. Among these, *Bambusa vulgaris* stands out. It is originated from Asia (Francis, 1993), popularly known as bamboo. The reasons why this species occur at PEDI are not certain, but according to Dechoum, their introduction happened years ago for ornamental and reforestation purposes and also by visitors and residents around the park.

Resistance or tolerance to secondary metabolites is a specific characteristic of this species, with presence of sensitive species referred as indicators of allelopathic activity such as *Lactuca sativa* L. (lettuce), *Lycopersicon esculentum* Miller (tomato) and *Cucumis sativus* L. (cucumber). To be indicated as a test plant, a species should present quick and uniform germination and sensitivity that allows expression of results under low concentration of allelopathic substances (Souza et al., 2007).

B. vulgaris Schrad. ex J.C. Wendl. belongs to the Poaceae family. It is a woody perennial plant with cylindrical stem, alternate leaves, parallel vernation and ligule between the lamina and the pulvinus (Lorenzi and Souza, 2005). Also, it is widely used in furniture, construction material, utensils, handcrafting, water pipes, etc. From an ecological point of view, it presents great efficiency in capturing CO_2 and thus can be used in recovery of degraded areas, erosion control and aggradation of water courses (Ribeiro, 2008).

Bamboo is quick and aggressive during colonization, impairing natural regeneration of other species (Ferreira, 2014). Impacts can be explained by possible allelopathic effects produced during secondary metabolism, which can be beneficial or prejudicial, direct or indirect, causing interference of one species on another (Fernandes et al., 2007; Souza et al., 2006).

According to Alencar et al. (2015), germination tests are the most commonly used analysis to evaluate the allelochemical potential of plant species and their effects on the community.

It is possible that the allelopathic effect of bamboo secondary metabolites might significantly interfere on germination and establishment of lettuce seedlings, but few information is available on this type of study.

Schulz et al. (2010) point out in their study that some species of bamboo are easily spreadable and act as homogenizer of the environments where they establish, demonstrating some allelopathic features. Based on these, their work focused on possible allelochemicals on aqueous extract from bamboo leaves on lettuce, which is a bioindicator.

Therefore, the present study aimed to evaluate allelopathic effects through germination tests of *L. sativa* seeds in aqueous extracts from *B. vulgaris* leaves and culm occurring in Dois Irmãos State Park, in Recife/PE, using different concentrations.

MATERIALS AND METHODS

This work was performed at Laboratory of Forest Seeds Analysis (LASF) from Department of Forest Science at Federal Rural University of Pernambuco (UFRPE). Plant material was collected at Dois Irmãos State Park (PEDI), located in the urban perimeter of the city of Recife/PE at Dois Irmãos neighbourhood, Brazil. PEDI is inserted in one of the largest Atlantic forest fragments of Pernambuco State, and it is a Full Protection Conservation Unit established in 1998 by State Law number 11.622/1998.

Lettuce seeds (*L. sativa* var. *crespa cinderela*) were bought from a local store, with germination rate of 98%.

Experiments were conducted under natural conditions (temperature 25°C and relative humidity 72%). *L. sativa* seeds were laid in gerbox boxes using germitest paper as substrate. Seeds were placed on the paper in five rows with ten seeds each, each row representing an experimental unit. 5 mL of the extract was added to the five experimental units using a pipette to equally distribute it. Distilled water was used instead of extract on the control group.

For extract preparation, material from *B. vulgaris* was separated in leaf and culm. 250 g of fresh plant material was weighted in a semi-analytical balance and ground in a blender with 1000 mL of distilled water, according to methodology described by Cruz et al. (2000). From this extract with concentration of 100%, dilutions were performed to make extracts with concentration of 75, 50 and 25%. Distilled water was used to represent concentration of 0%.

Individuals presented 5 mm of root projection used as criteria to identify germination. Germinated seeds were counted on a daily basis for seven days at the same time. For each aqueous treatment, four repetitions were used and the experimental design was entirely casual.

Analysed variables were:

(1) Germination percentage (%G), according to Standards for Seed Analysis (BRASIL, 2009), based on the following equation:

$$\%G = \frac{NG}{NT} \times 100$$

where NG = number of germinated seeds and NT = total number of seeds.

(2) Speed of Germination Index (SGI) was calculated using daily measurements occurring since the first day of germination until the

*Corresponding author. E-mail: cesarhenrique27@yahoo.com.br.

Table 1. Effect of different concentrations of aqueous extract from *Bambusa vulgaris* leaves and culm on germination, speed of germination index, mean germination time and mean speed of germination of *Lactuca sativa* seeds.

Treatment (%)	Mean values							
	Aqueous extract from leaves				Aqueous extract from culm			
	G (%)	SGI	MGT (days)	v (day^{-1})	G (%)	SGI	MGT (days)	v (day^{-1})
T1-0	67.50^{a}	37.46^{a}	4.53^{a}	0.22^{a}	67.50^{a}	37.46^{a}	4.53^{a}	0.22^{a}
T2-25	39.00^{b}	18.45^{b}	4.97^{a}	0.20^{a}	27.00^{b}	14.73^{b}	4.50^{a}	0.22^{a}
T3-50	39.00^{b}	21.70^{b}	4.68^{a}	0.21^{a}	14.00^{b}	7.31^{b}	4.00^{a}	0.25^{a}
T4-75	23.50^{b}	11.56^{b}	4.62^{a}	0.22^{a}	24.50^{b}	19.57^{b}	3.88^{a}	0.26^{a}
T5-100	36.50^{b}	18.83^{b}	4.53^{a}	0.22^{a}	32.00^{b}	22.80^{ab}	3.84^{a}	0.26^{a}
CV	39.17	44.52	3.89	4.18	61.74	54.93	8.16	8.47

Mean values for germination (G%), speed of germination index (SGI), mean germination time (MGT) and mean speed of germination (v). Different lowercase letters indicate significant differences (Tukey test, $p > 0.05$).

last day of counting. Index was calculated with the equation described by Maguire (1962):

$$SGI = \frac{G_1}{N_1} + \frac{G_2}{N_2} + \ldots + \frac{G_n}{N_n}$$

where G_1, G_2, G_n = number of germinated seeds on daily counts and N_1, N_2, N_n = number of counting days.

(3) Mean germination time (MGT) is based on the equation by Ferreira and Borghetti (2004):

$$MGT = \sum_{i=1}^{k} n_i . t_i \ / \sum_{i=1}^{k} n_i$$

where n_i: number of germinated seeds in a certain time interval (t_i).

(4) Mean speed of germination (v): It is the opposite of mean germination time, which is another way to quantify germination from a kinetic point of view. The lower its value, the more vigorous the seeds (Ferreira and Borghetti, 2004).

$$v = \frac{1}{t} \text{ or } \sum_{i=1}^{k} n_i \ / \sum_{i=1}^{k} n_i . t_i$$

Data was transformed to $\text{arc sen} \sqrt{x/100}$ and submitted to ANOVA. Means were compared using Tukey test at 5% probability using BioEstat 5.0 programme.

RESULTS AND DISCUSSION

Variance analysis showed that aqueous extract from *B. vulgaris* leaves and culm presented significant negative effect on germination percentage and speed of germination index (SGI), but no significant effect on the remaining variables when compared with control (Table 1).

Aqueous extract from *B. vulgaris* leaves negatively influenced germination percentage and speed of germination index of *L. sativa* seeds, where control treatment showed the best means with statistically higher values. Other values were not different statistically. Based on regression analysis (Figure 1), concentrations of 70.22 and 69.36% promote maximum detriment of highlighted variables, respectively.

Schulz et al. (2010) analysed allelopathic effects of *Dendrocalamus giganteus*. They verified that aqueous extract of this species in various concentrations did not negatively influenced lettuce germination. However, Novais et al. (2017) analysed allelopathic effects of aqueous extracts from *Luetzelburgia auriculata* leaves on lettuce germination and found that this species had its germinative performance affected negatively while submitted to the extract in various concentrations. These effects were more evident as concentration increased.

Similar results were found in a study performed by Alencar et al. (2015) where they evaluated effects of aqueous extracts of *B. vulgaris* leaves on corn and beans seeds. The authors found that different concentration of the extract negatively affected both germination and SGI of the mentioned species. According to the authors, metabolites found on phytochemical analysis of *B. vulgaris* such as tannin, phenols, flavonoids, flavones, xanthones and catechins are probably related to the observed allelopathic action.

According to Pires et al. (2001), secondary metabolites responsible for negative allelopathic effect on different plant species are highly present on leaves.

Negative effects in germination percentage and speed of germination index were also observed when the effect of aqueous extract of *B. vulgaris* on the variables mentioned earlier. Control treatment showed the best means, all superior to the treatments with extract at concentrations of 25, 50 and 75% but not differing from treatment T5.

From regression analysis, maximum detriment of germination percentage and speed of germination index

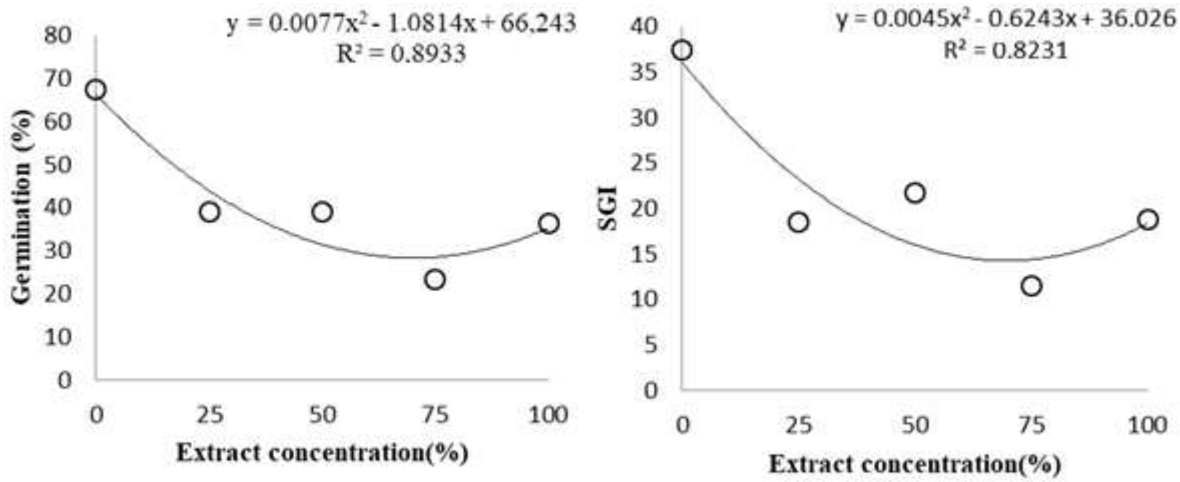

Figure 1. Effect of extract from *Bambusa vulgaris* leaves on germination and speed of germination index of *Lactuca sativa*

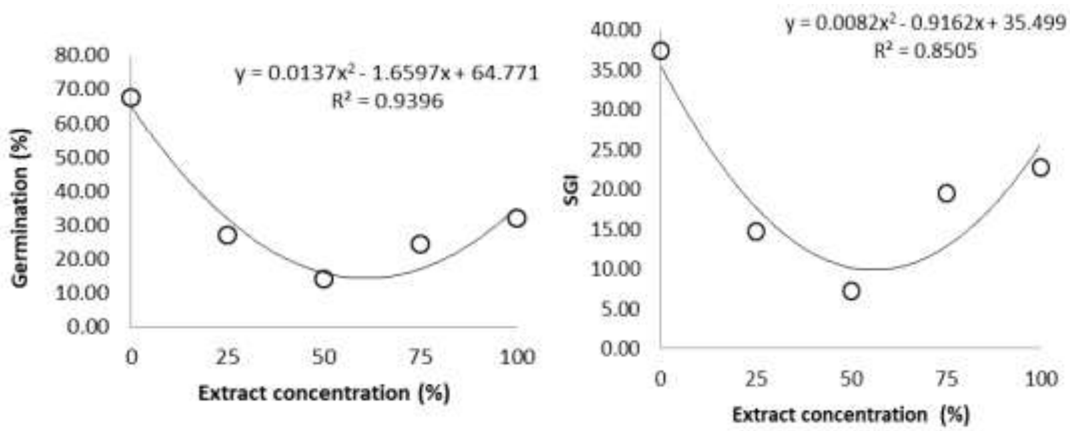

Figure 2. Effect of aqueous extract from *Bambusa vulgaris* culm on germination and speed of germination index of *Lactuca sativa* seeds.

occurred at estimated concentrations of 60.67 and 56.55% of culm extract, respectively (Figure 2).

Inhibition of seed germination can be one of the first physiological effects caused by allelopathic interactions of secondary metabolites produced by plants, being a secondary response of primary effects that happen in plant metabolism (Pedrol et al., 2006).

Works associated with allelopathic effects of aqueous extract from *B. vulgaris* culm are existing in the literature. However, the present study showed that this extract has the same allelopathic effects on lettuce, releasing phytotoxins capable of inhibiting germination and SGI.

The present study corroborates with Faria and Grombone-Guaratini (2011). The authors showed substances capable of inhibiting development and growth of other taxa by analysing aqueous and ethanolic extracts from a native bamboo from Atlantic Forest, *Merostachys pluriflora*, leaves, culm and rhizome. According to Putnan and Tang (1986), allelochemicals are present in all plant tissues, including rhizomes, roots, leaves, stems, fruits and seeds.

At the same time, these results differ from the findings of Sanquetta et al. (2013), that evaluate allelopathic potential of a native species of bamboo from Atlantic Forest *Merostachys skvortzovii*. No significant effects of aqueous extracts from leaves were observed on germination and SGI of *Mimosa scabrella*. Fernandes et al. (2007) also observed no significant effects of aqueous extract from *Merostachys multiramea* leaves on *Araucaria angustifólia* seeds germination or germination speed.

Regarding mean germination time and mean speed of germination, Silveira et al. (2014) tested allelopathic activity of *A. angustifólia* on *L. sativa*. They found that aqueous extract from the Brazilian pine has inhibiting properties, thus showing allelopathic potential of this species.

Silva et al. (2010) demonstrated that, according to mean time and speed of germination of two forest species, there is a direct interference of their extract, therefore presenting its allelopathic potential on *L. sativa* germination independently of the concentration used.

The present study evidenced that aqueous extract of *B. vulgaris* leaves and culm has allelopathic effects on lettuce, releasing phytotoxins capable of inhibiting germination and SGI.

Considering that lettuce is a bioindicator, it is possible that allelopathic effects can significantly interfere on other species establishment and germination due to substances present on plant residues deposited on the surface or incorporated in the soil annually.

In this sense, new works evaluating allelopathic effects of aqueous extracts on species germination associated on *B. vulgaris* in natural environment are suggested. From such results, it is possible to propose management and conservation strategies for native species in natural vegetation fragments.

Conclusion

Based on the analysed results, the inhibitory allelopathic behaviour of *B. vulgaris* extract on *L. sativa* seed germination was found in all plant parts and concentrations tested.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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