



Environmental factors' action on the germination process and initial growth of weeds of Rubiaceae family



M. Gallon ^{a,*}, M.M. Trezzi ^b, F. Diesel ^b, A.A. Balbinot Junior ^c, F.D.B. Pagnoncelli Junior ^b, M.V.J. Barancelli ^b

^a Federal University of Rio Grande do Sul, Av. Bento Gonçalves 7712, Porto Alegre, RS 91540-000, Brazil

^b Federal Technological University of Paraná, Via do Conhecimento Km 1, Pato Branco, PR 85503-390, Brazil

^c Embrapa Soja, Rod. Carlos João Strass, Warta District, PO Box 231, Londrina, PR 86001-970, Brazil

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ABSTRACT

Knowledge of factors that influence the germination process and their interactions allows estimating the initial development of a species in a particular environment and optimizing the weed management decisions. The objectives of this study were to evaluate the effect of environmental factors on the germination and initial growth of the weed species *Borreria latifolia*, *Galianthe chodatiana* and *Richardia brasiliensis*. Five trials were conducted using a completely random experimental design, in factorial design, with five repetitions, each one evaluating effects of temperature, irradiance, aluminum, salinity and pH. The germination was assessed daily, and germination speed index and the length of radicle and epicotyl were determined. Temperatures between 20 and 30 °C maximized final germination and germination speed index of the three species. *B. latifolia* showed low germination with 15 °C temperature. *G. chodatiana* and *R. brasiliensis* did not germinate well at temperatures between 30 and 35 °C. *B. latifolia* and *R. brasiliensis* did not germinate in the dark condition, whereas *G. chodatiana* were indifferent to photoperiod. Seeds from *B. latifolia* and *G. chodatiana* were more tolerant to aluminum concentration in the germination than *R. brasiliensis*, while *G. chodatiana* were tolerant in germination and early growth. Low NaCl concentration inhibits the seed germination of the three species. *B. latifolia* showed higher adaptation at low pHs, while the other species at pHs close to neutrality. The responses of the species to environmental factors can be used in the planning of strategies for their management.

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1. Introduction

In Brazil, there is a trend towards simplification of cropping systems, especially in crops with large planted areas, such as soybean, which currently reaches almost 35 million hectares. This favors the establishment of an abundant community of herbicide tolerant species. The Rubiaceae weed species *Borreria latifolia* (Aubl.) K. Schum. (broadleaf buttonweed), *Richardia brasiliensis* Gomes (tropical Mexican clover) and *Galianthe chodatiana* (Standl.) E.L. Cabral have in common the tolerance to the herbicide glyphosate. The species broadleaf buttonweed and tropical Mexican clover are widespread practically everywhere in Brazil in systems of grain crops. The species *G. chodatiana* is not endemic (Cabral, 2009), but has potential for spreading to other areas, due to the difficulty of control.

The species *B. latifolia* (synonym of *Spermacoce latifolia* Aubl.), is an annual herbaceous, which reproduces by seeds. According Kissmann and Groth (2000), this species tolerates poor and acidic soils. The broadleaf buttonweed presents high germination levels at 25 °C, with tolerance

to temperatures ranging from 30 to 35 °C and dramatic germination reduction at temperatures ranging 15 to 20 °C (Parreira et al., 2011).

The species *R. brasiliensis* Gomes (tropical Mexican clover) is an infesting native from South America, which presents annual cycle and reproduction by seeds (Moreira and Bragança, 2010). The species development is stimulated in well-illuminated areas and less dense vegetation, where the plants get more aggressive (Lorenzi, 2000). It affects pastures, mainly in integrated agriculture/cattle raising systems, in which the species is considered the most important, in addition to orchards and fields where it can disturb specially in the establishment phase, because its germination is predominant in spring season (Kissmann and Groth, 2000).

The species *G. chodatiana* (Standl.) is a perennial, which can reproduce vegetatively by stolons and also by seeds. The species develops in dry or slightly moist soils, in altitudes ranging from 700 to 900 m, flowers in the fall season and fruits in January and February (Cabral, 2009).

There is still little information with regard to this species' response to environment and management. The knowledge about the factors which affect the germination and dormancy processes and their interactions would help to understand the dynamics of a species population (Vivian et al., 2008; Chauhan, 2012), supporting the development of forecast germination models. It allows the weed management decision-making

* Corresponding author at: Federal University of Rio Grande do Sul, Department of Crop Plants, Av. Bento Gonçalves 7712, Agronomia, 91540-000 Porto Alegre, RS, Brazil.
E-mail address: mtgallon90@yahoo.com.br. (M. Gallon).

process to be optimized, regarding not only the tactics to be implemented, but also the timing for taking action (Myers et al., 2004; Lim et al., 2015). The majority of the control methods do not suppress weed dormant seeds, but it gets more susceptible during the germination (Chauhan, 2012).

The weed species' seed germination and seedling emergency are influenced by many environmental factors such as oxygen, light, temperature, moisture and soil chemistry (Chauhan, 2012; Shu et al., 2015). The light activity, for example is linked to the phytochrome system, which is linked to the cell membrane functioning, which can affect the flow of several cell compounds (Vivian et al., 2008; Singh et al., 2012). Temperature relates directly to water absorption and biochemical reactions in the seeds, which regulate the metabolism involved in the germination process (Paiva et al., 2016).

The chemical condition of soil such as acidity, toxic aluminum level and salinity, may affect or even inhibit the vegetative development of plants (Yaish et al., 2016). Aluminum cations are able to change the characteristics of cell membranes, favoring its degradation, increasing its permeability and promoting solute loss (Vitorello et al., 2005). In addition to the aluminum release, the soil pH has an important role on nutrient availability (Pavinato and Rosolem, 2008; Carrino-Kyker et al., 2016), however its influence upon seed germination hasn't received much attention. pH ranges lower than 3,0 and higher than 8,0 have been described as germination inhibitors (Wagner Junior et al., 2007). High levels of salts, especially sodium chloride (NaCl), may inhibit seed germination by decreasing its osmotic potential (Munns and Tester, 2008; Tavakkoli et al., 2010). Despite the fact that high salt concentrations inhibit weed germination, and some areas worldwide, specially dry lands, this condition hinders crop production (Munns and Tester, 2008).

Thus, the aims of this work were to investigate under controlled conditions the effect of temperature, light, pH, aluminum and salinity on the germination process of seeds and initial development of seedlings of the species *B. latifolia*, *G. chodatiana* and *R. brasiliensis*.

2. Material and methods

2.1. Seed collection and preparation

The studies were carried out at Federal Technological University of Paraná, campus Pato Branco, using as test materials seeds of *B. latifolia*, *R. brasiliensis* and *G. chodatiana* collected from soybean fields located at Canoinhas/SC (coordinates: -26.370833 , -50.276944), Alvorada do Sul/PR (coordinates: -22.8200 , -51.2000) and Renascença/PR (coordinates: -26.311578 , -52.910045), respectively. After collecting, the seeds were processed manually and dried in a forced air circulation oven for 24 h at 30 °C, packed in paper envelopes and stored in a refrigerator at a temperature of 10 ± 2 °C until they were used, ten months later. The bioassays were carried out between April and July 2014.

Five bioassays were carried out using a completely randomized factorial design with 5 repetitions. Each bioassay was repeated twice (the repetition was set 30 days after the installation of the first bioassay). The first bioassay was made with 10 seeds per repetition, and the second one was made with 20 seeds. The seeds were laid out in Petri dishes, over a double layer of paper towel for germination soaked with a solution used for each bioassay with a volume (in milliliters) of 2,5 times the seed weight, being sealed with plastic film. The solution was not replaced during the study.

For the species *B. latifolia*, the dormancy had to be broken laying the seeds in a stove at 60 °C for 30 min followed by an immersion in a solution of KNO₃ at 2% concentration for 3 h. The seeds of *G. chodatiana* were laid in a stove with air circulation at 60 °C for 30 min. After drying, the seeds were cover by a solution with gibberellic acid 400 ppm, straight on paper towel in the Petri disks. The seeds of the species *R. brasiliensis* didn't need to go through any dormancy braking process.

2.2. Effect of temperatures and irradiance condition on seed germination

The bioassays which evaluated the temperature effects were carried out in a factorial design 3×6 , in which the A factor was composed by the species (*B. latifolia*, *R. brasiliensis* and *G. chodatiana*) and the B factor by the temperatures (15, 20, 25, 30, 35 and 20/30 °C, being the last one characterized by 12 h at 30 °C and 12 h at 20 °C in every 24 hour cycle). The dishes containing the seeds of each species were accommodated in BOD type growth incubators with automatic temperature control, as needed for the experiment.

For the other bioassays, the dishes were kept at 25 °C, which is the best temperature rate for the bioassay. The bioassays for investigated the light effect were carried out in a factorial design 3×2 , in which the A factor was composed by the three species and the B factor by the light condition (with photoperiod of 12 h and without photoperiod). In the absence of light, the dishes were covered with flexible aluminum film, and the assessments were made in a dark room. The seeds were exposed for 15 s using only green light for safety.

2.3. Effect of pH, salinity and aluminum on seed germination

The bioassays for the pH effect investigation were carried out using a factorial design 3×6 , in which the A factor was composed by the species (*B. latifolia*, *G. chodatiana* and *R. brasiliensis*) and the B factor by pH ranges (3,0; 5,0; 7,0; 9,0 and 11,0). In order to achieve the pH ranges, distilled water solutions were prepared adding sodium hydroxide (NaOH) or hydrochloric acid (HCl), calibrated using a potentiometer. Two bioassays were carried out to evaluate the salinity effect using a factorial design 3×5 , in which the A factor was composed by the three species and the B factor by aqueous solutions of NaCl (P.M. = 58,44) 0; 25; 75; 150 and 300 mM. To the aluminum effect evaluation, two bioassays were carried out using a factorial design 3×5 , in which the A factor was composed by the three species and the B factor by aqueous solutions of aluminum sulfate ($Al_2(SO_4)_3 \cdot 18H_2O$) (P.M. = 666) 0,0; 0,5; 1,0; 1,5 and 2,0 mEq/100 mL.

2.4. Evaluations

The germination was assessed daily, for 21 days, considering as germinated only the seeds with the presence of a two-millimeter-length radicle. At the end of the assessments the length of the roots and the upper part of 3 seedlings in each repetitions chosen randomly were measured using a pachymeter. For the treatments with no germination, the grade given was zero for this assessment. In each bioassay the germination was determined by the end of the assessments as well as the germination speed index (GSI).

2.5. Statistical analyses

The data collected in the assessments was submitted to the Lilliefors test to evaluate the presupposed normality and the analysis of variance and to test the hypothesis by the F test ($p = 0,05$). The analysis was complemented by the multiple comparison test (Duncan) or regressions for the qualitative and quantitative factors respectively. The Winstat software (Machado and Conceição, 2007) and SigmaPlot 10.0 software were used to support the statistical analysis and to plot the graphics.

3. Results

3.1. Effect of temperatures and irradiance condition

In all temperature rates, except at 15 °C, the germination of *B. latifolia* was promoted, with an average above 90% in all treatments (Fig. 1A). The germination of *R. brasiliensis* was promoted at 15, 20 and 25 °C and at the alternating 20/30 °C rate achieving values superior to 80%, with no

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