

## PHOTOBLASTISM AND GERMINATION OF *Campomanesia aurea* (Myrtaceae) SEEDS

### FOTOBLASTISMO E GERMINAÇÃO DE SEMENTES DE *Campomanesia aurea* (Myrtaceae)

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**ABSTRACT:** *Campomanesia aurea* (Myrtaceae) is a native species from the Pampa region. It has edible fruits, ornamental characteristics and ability to recover potential degraded areas. It is naturally propagated by seeds. However, for its most efficient use for seedling production or replanting of vegetation, a basic knowledge about the conditions required for maximum germination is required. The objective of this work is to evaluate the germination of *C. aurea* seeds and its germinative behavior under different light conditions. Seeds from three collection sites were analyzed as for photoblastism, provenance and maturation by germination test, germination speed index, mean germination time and normal seedling formation. Data were subjected to analysis of variance and compared by Tukey test at 5% probability of error. For the three collection sites, no differences were observed for germination and the MGT of seeds in the absence or presence of light, indicating that it is a neutral photoblastic species. For different collection sites, no differences were observed for germination percentage, but seeds collected in Barão do Triunfo showed higher GSI and normal seedling formation than seeds collected at Morro do Osso and Morro Santana. Such differences may be related to the degree of fruit maturation and seed moisture. Seeds of *C. aurea* are neutral photoblastic, and present a germination rate higher than 85%.

**KEYWORDS:** Guabirobinha-do-campo. Myrtaceae. Light.

## INTRODUCTION

*Campomanesia aurea* O. Berg is a native species of Myrtaceae from the Pampa Biome. It is popularly known in Brazil as "guabirobinha-do-campo" or "araçá-rasteiro" (LORENZI, 2006). It grows in the southern plateau of the South region of Brazil in areas occupied by grass or shrub fields, mainly in Rio Grande do Sul (REITZ, 1977), distributing to Paraná, which is the Brazilian northern limit of this species, where it occupies open areas in the Mixed Ombrophylous Forest dominion (LIMA et al., 2011).

It is an essentially heliophyte species, with a subterranean xylopodium, and indifferent to soil physical conditions. It is able to grow from dry soils to swamp borders (REITZ, 1977). It is perennial and may reach up to one meter in height. It presents woody and branched stems. It has an ornamental potential for use in gardens or pots due to its shrub size, irregular shape and intense and aromatic flowering (STUMPF, 2009). Flowering extends from October to January. Fruit ripening occurs from December to February (LORENZI, 2006). Leaves are used in folk medicine (CRUZ; KAPLAN, 2004).

It is naturally propagated by seeds. However, for its most efficient use for seedling production or replanting of vegetation, a basic knowledge about the conditions required for maximum germination is required. Percentage, uniformity and germination speed are influenced by intrinsic factors related to genotype, degree of maturity, dormancy, among others, and also by environmental factors related to water availability, temperature, oxygen and light. Such conditions must be satisfactory so that the process can be duly triggered (MARCOS FILHO, 2015).

Responses to light variations are a mechanism of adaptation of plants to specific niches of environment. Light perception is related to the phytochrome, which allows seeds to respond to environmental variations (GUALTIERI; FANTI, 2015). Phytochrome is a soluble chromoprotein present in embryonic stem cells. Its conversion to the active form ( $F_{VD}$ ) occurs by the exposure of the inactive form ( $F_V$ ) to radiations at the 660 nm range. The exposure of the active form at 730 nm, or in the dark, makes the phytochrome assume the inactive form. The phytochrome in the active form at sufficient concentrations initiates the germinative process through a synthesis of hormones and the

restart of genetic transcription (MARCOS FILHO, 2015).

Thus, for species with positive photoblastic seeds, the light effect is beneficial and triggers the synthesis of hormones and enzymes, respiratory control, permeabilization of the integument to oxygen and lipid metabolism. For negative photoblastic seeds, the low concentration of phytochrome in the active form, due to exposure to dark periods or fast exposure to red-distant radiation, is enough to initiate the process (MARCOS FILHO, 2015).

Due to the lack of studies on the germination of *C. aurea* seeds, in addition to the potential of use of this species as an ornamental and fruit species, and the increasing demand for the propagation of native species for the recovery of degraded areas of the Pampa biome, further studies evaluating the germinative capacity of seeds and factors that influence this process are necessary. The objective of this study was to evaluate the germination of *C. aurea* seeds and the germinative behavior under different light conditions.

## MATERIAL AND METHODS

The experiments were conducted in the laboratories of the Department of Horticulture of Forestry, Faculty of Agronomy, Federal University of Rio Grande do Sul, RS. The collections were performed in native vegetation areas in the municipality of Barão do Triunfo, RS (30°18' S, 51°50' W), Morro do Osso Natural Park (30°07' S, 51°14' W), and Morro do Santana Conservation Unit (30°04' S, 51°08' W) in the city of Porto Alegre, RS. The collections were performed in February and March of 2016 in approximately 50 matrices per collection site.

After collection, the fruits were taken to the laboratory, where the color of the epicarp was determined using a digital colorimeter (Konica/Minolta, CR400). The measurement was performed on the opposite side of the calice of six randomly chosen fruits in a sample of 30 fruits. The determination of colors was made according to the CIE-Lab system, according to which the coordinate L\* represents the degree of luminosity of the color (variation from zero to 100, indicating, respectively, black and white), the coordinate a\* expresses the degree of variation (a\* negative = green, a\* positive = red) and the coordinate b\* represents the degree of variation between blue and yellow (b\* negative = blue, b\* positive = yellow).

Following color determination, the seeds were manually removed from fruits and washed in

tap water using a sieve to remove the pulp and then surface-dried using paper towel. Seed disinfection was performed using 70% alcohol for 1 minute and 1% sodium hypochlorite (a.i.) for 15 minutes, and then washed with autoclaved deionized water. After disinfection, the seeds were stored in Gerbox boxes containing paper moistened with water at a ratio of 2.5 times the weight of the dry paper (Brazil, 2009), and taken to an environment with a temperature of  $25 \pm 2^\circ\text{C}$  and a photoperiod of 16 hours.

The germination test was conducted for the three collection sites using 100 seeds divided into four replicates of 25 for seeds harvested in Barão do Triunfo and Morro do Osso Natural Park, and 80 seeds divided in replications of 20 for seeds collected in the Morro do Santana Conservation Unit due to the low number of fruits collected at this site.

For the evaluation of photoblastism, part of the seeds was kept in the absence of light during the whole experiment using hermetically sealed metal boxes. Periodic counts were carried out in a dark room using a fluorescent lamp wrapped in four layers of green cellophane paper (safety light). For the comparison between collection sites, the mean values of the seeds germinated in the presence of light were used.

Seeds that showed a protrusion of the radicle visible to the naked eye germinated. Simultaneously, the germination speed index (GSI) (MAGUIRE, 1962), the mean germination time (MGT) (SILVA; NAKAGAWA, 1995) and the formation of normal seedlings were evaluated using the germination test. For this, observations were made every two or three days to account for germinated seeds in each evaluation. The first count was made as soon as the first seed germinated and the final evaluation was conducted 90 days after the beginning of the test. Normal seedlings were those that presented all essential structures well-developed, complete, proportional and healthy (BRASIL, 2009).

The degree of seed moisture after fruit extraction was determined by the drying oven method at  $105^\circ\text{C}$ , according to the methodology described in the Rules of Seed Analysis (BRASIL, 2009), using triplicates with a minimum weight of 0.3 g.

Data were subjected to analysis of variance and means were compared by Tukey test at 5% probability of error. The germination data for Barão do Triunfo and Morro do Osso Natural Park, the GSI for Morro do Santana Conservation Unit for photoblastic seeds, and the germination in

comparison between sites did not fit the assumptions of analysis of variance even after data transformation. They were therefore analyzed by Kruskal Wallis non-parametric statistics for comparison.

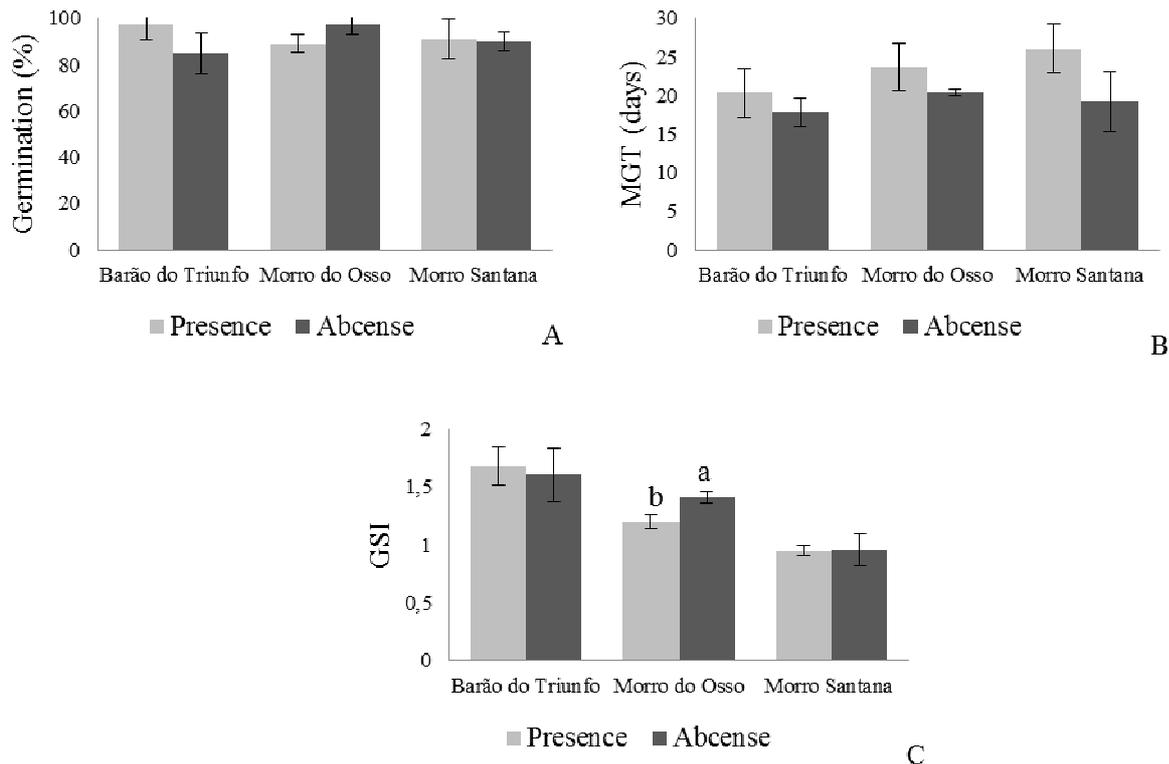
## RESULTS

For the three collection sites, no differences were observed for germination and the MGT of seeds in the absence or presence of light, indicating that it is a neutral photoblastic species. For the GSI, only the seeds collected at Morro do Osso Natural Park presented differences, with a higher average for seeds kept in the dark (1.40), that is, seeds kept in a place without light presented a faster germination (Figure 1A, 1B and 1C).

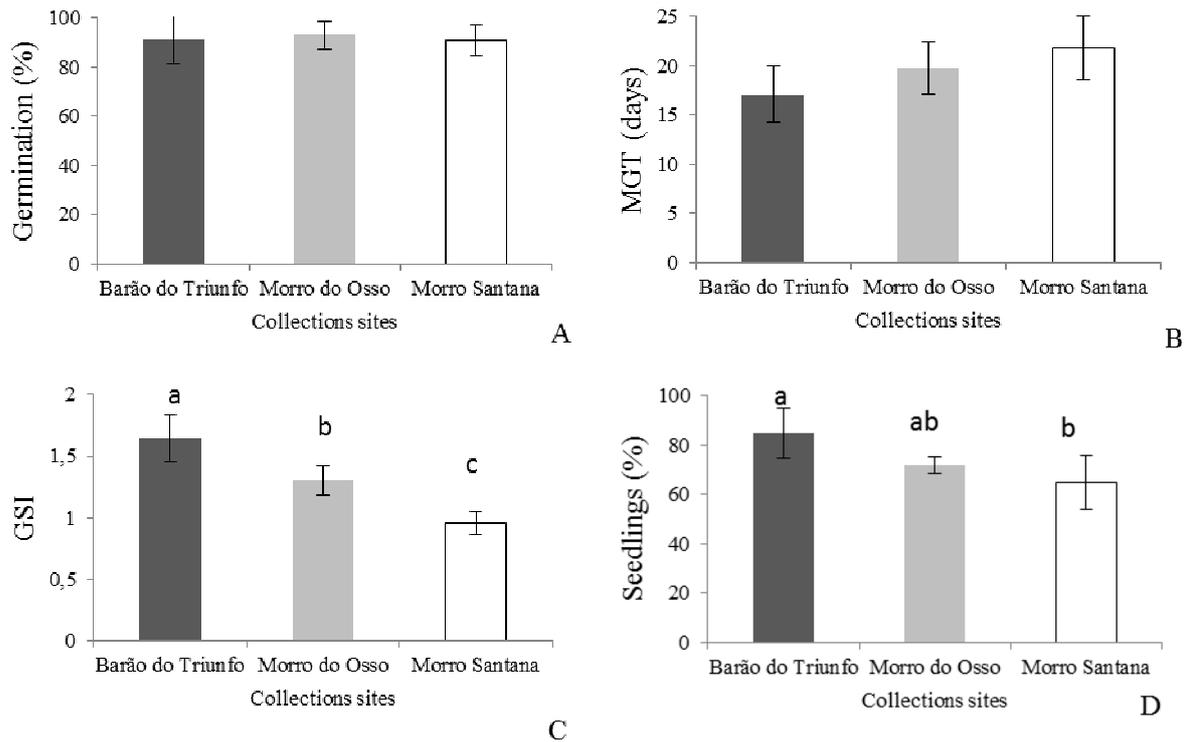
For the different collection sites, although no differences were observed for germination

percentages, on average higher than 85%, seeds collected in Barão do Triunfo showed higher GSI and normal seedling formation than seeds collected in Morro do Osso and Morro Santana (Figure 2A, 2B and 2C). The first germination count was performed in nine days for seeds collected in Barão do Triunfo and Morro do Osso, and in 13 days for seeds collected at Morro Santana. The counting extended for up to 73 days after sowing for the last location.

In relation to fruit color, the only difference observed was for the coordinate  $a^*$ , which provides information about the intensity of green and red (Table 1). However, seeds collected in Barão do Triunfo presented a higher moisture (57%) than seeds collected in Morro do Osso (46%) and Morro Santana, respectively.



**Figure 1.** Germination (A), Mean Germination Time (MGT) (B) and Germination Speed Index (GSI) (C) of *Campomanesia aurea* seeds for different collection sites subjected to the presence or absence of light. UFRGS, Porto Alegre, RS.



**Figure 2.** Germination (A), Mean Germination Time (MGT) (B), Germination Speed Index (GSI) (C), and seedling formation (D) of *Campomanesia aurea* seeds for different collection sites. UFRGS, Porto Alegre, RS.

**Table 1.** Means for luminosity ( $L^*$ ) and color coordinates ( $a^*$  and  $b^*$ ) of fruits and seed moisture (M%) of *Campomanesia aurea* collected at three sites and used for germination test. UFRGS, Porto Alegre, RS.

Accessions:	$L^*$	$a^*$	$b^*$	M (%)
Barão do Triunfo	$61.74 \pm 6.30$ <sup>ns</sup>	$-16.69 \pm 3.51$ a	$40.45 \pm 5.23$ <sup>ns</sup>	57.34 a
Morro do Osso	$63.29 \pm 6.65$	$-14.58 \pm 2.30$ ab	$42.30 \pm 5.22$	46.46 b
Morro do Santana	$64.25 \pm 4.16$	$-11.48 \pm 2.60$ b	$44.14 \pm 4.45$	43.53 b

Means followed by different letters in columns differ statistically by Tukey test at 5% probability of error. Mean and standard deviation. n = 24.

## DISCUSSION

The absence of light response to germination has also been verified for seeds of other Myrtaceae such as *Eugenia brasiliensis*, *E. involucrata*, *E. pyriformis* and *E. uniflora* (LAMARCA et al., 2011). Seeds of *Blepharocalyx salicifolius* (Myrtaceae) also presented a behavior indifferent to light for germination. However, in the absence of light, there was a lower MGT and a higher GSI (REGO et al., 2011), similar to that found for *C. aurea*.

Seeds of *Curitiba prismatic* showed high GSI and germination (91.2%) in the presence of light, although this also occurred (35.6%) in the dark. This species is then classified as preferential positive photoblastic. No differences were observed for MGT, which was on average 22 days (REGO et

al., 2011). Other species of the family Myrtaceae, such as *Campomanesia guazumifolia*, *Acca sellowiana*, *Myrcianthes pungens* and *Psidium cattleianum*, were also positive photoblastic (SANTOS et al. 2004). However, for *C. pubescens*, germination was higher than 90% regardless of luminosity. However, in the absence of light, greater germination and GSI were observed independent of temperature and substrate, resulting in the classification of this species as negative photoblastic (DOUSSEAU et al., 2011).

Photoblastism differs among species of a same family, within the same species and even in the same plant. This results in ecological advantages because at least some seeds germinated independently from the light conditions of the environment in which they were found. Other factors, such as temperature, drying, storage and

conditions under which the plant generated seeds, may be responsible for the different germination responses to light (KLEIN; FELIPPE, 1991).

Species requirements for light for germination are related to the ecological groups to which they belong. Thus, in a shade condition, where there is predominance of red-distant radiation, seeds of pioneer species do not germinate, as it is considered an environment not propitious to the development of seedlings due to a low blue and red radiation, which is important for photosynthesis. The effects of light filtering by vegetation are also exerted by litter (TAKAKI, 2015). In this sense, although the distribution of *C. aurea* is concentrated in open areas and it is a heliophile species (REITZ, 1977; LIMA et al., 2011), we expected that the species would show a positive photoblastic behavior because of the characteristics of the biome where it is found. However, in areas of Pampa native vegetation, there is a strong thickening of undergrowth vegetation, preventing the incidence of light on the soil surface, as it occurs in forest areas. Thus, a behavior indifferent to light as verified for the species may be a strategy to occupy areas with this type of vegetation.

Regarding collection sites, we verified that *C. aurea* seeds have a high viability. However, we noticed, by the data of MGT and GSI, that germination does not occur quickly and uniformly. MGT was 18 days for seeds collected in Barão do Triunfo and 22 days for Morro do Osso and Morro Santana, respectively.

Differences in GSI between collection sites may be due to environmental variations, genetic variability of the parent plant and genetic segregation between individuals of each progeny, which may cause differences in germination and emergence (FIOR et al., 2010). A behavior similar to that found in this study for these variables was observed for different seed lots of *Myrcianthes pungens*, which present a germination rate above 80% between the eighth and the fifteenth day after sowing, with a mean germination time of up to 30.1 days (FIOR et al., 2010). This also occurs with other species of the family Myrtaceae, such as *Blepharocalyx salicifolius*, in which germination begins between the sixth and the eighth day after sowing, and stabilizes from the 36<sup>th</sup> day, and *Curitiba prismatica*, which begins its germination only on the 18<sup>th</sup> day extending to the 65<sup>th</sup> day (REGO et al., 2009; REGO et al., 2011).

Seed germination of non-domesticated species occurs in a non-simultaneous manner, resulting in the distribution of germination over time. This reduces the exposure of seeds of a

population to environmental adversities that could compromise the survival of the species (MARCOS FILHO, 2015).

Such differences observed for GSI may also be related to the degree of fruit maturation and seed moisture. In many fruits, the coloring of the epicarp is indicative of its maturation. However, for *C. aurea*, the only difference in fruit color was observed for the coordinate  $a^*$ , which provides information regarding the intensity of green and red the less negative this value for intensity of green color is. However, when the values for the coordinates  $a^*$  and  $b^*$  are crossed in the CIE-lab color chart, there is a very tenuous variation between the different collection sites, which makes the use of colors a poor parameter for the separation of mature fruits of *C. aurea*, unlike other species of the family Myrtaceae, for which the coloring of fruits is a good indication of maturation for seed collection. An example of this is *Eugenia pyriformis* and *E. involucrata*, species which the coloring of the epicarp is efficient regarding the determination of fruit collection point for seed extraction. It may be used as an indication for the physiological maturity of seeds, reflecting in higher percentages of germination and seedling development (ORO et al., 2012). Fruit coloring also proved to be a practical and efficient parameter to identify the physiological maturity point of *E. uniflora* seeds because it is related to moisture, accumulation of dry mass and germination (AVILA et al., 2009).

Some seeds have the ability to germinate a few days after egg fertilization. However, the formation of normal seedlings at this stage is compromised since histological differentiation and reserve accumulation are not complete. Thus, the percentage of seeds able to germinate and produce normal seedlings increases during maturation, reaching a maximum level in the period close to the stoppage of dry mass flow from the plant to the seed (MARCOS FILHO, 2015), this was observed in this study. Although there were no differences in the percentage of germination for the different sites, we observed a high formation of normal seedlings for seeds collected in Barão do Triunfo in relation to Morro Santana (Figure. 2D).

Seeds produced by fleshy fruits, such as *C. aurea*, present a lower degree of dehydration in relation to dry fruits seeds at the end of maturation (MARCOS FILHO, 2015). The seed moisture of *C. aurea* observed in this experiment after fruit extraction, above 43%, is compatible with that found for other species of the genus, such as *C. pubescens* (42%) (DOUSSEAU et al., 2011) and *C. adamantium* (57%) (DRESCH et al., 2012).

The high degree of seed moisture of the species at the time of collection is characteristic of seeds of other species in the Myrtaceae family which do not undergo a severe desiccation at the end of maturation, and are dispersed with a high degree of moisture. Similar values for initial water content as observed for *C. aurea* after seed treatment were verified for other species of Myrtaceae, such as *Eugenia brasiliensis* (48.9%), *Myrcia glabra* and *Myrcia palustris* (higher than 40%), *Myrcianthes pungens* (higher than 40%), *Eugenia pyriformis* (45%), and *Acca sellowiana* (36%) (KOHAMA et al., 2006; LEONHARDT et al., 2010; FIOR et al., 2010; SCALON et al., 2012; GOMES et al., 2013).

Seeds collected in Barão do Triunfo have a higher moisture content (57%) than seeds collected in Morro do Osso (46%) and Morro Santana (43%). The highest water content in the seeds of Barão do Triunfo coincided with higher values for GSI and normal seedling formation. This may have occurred due to a high volume of seed water possibly coinciding with the maximum accumulation of dry matter, which generally leads to a greater physiological potential, resulting in a faster seed germination speed and a higher seedling vigor (MARCOS FILHO, 2015).

The water content in seeds needs to be high during the period of intense transfer of reserves from the plant to the seed, since the presence of water is indispensable to allow the movement of photoassimilates and minerals. The percentage of seeds able to germinate and produce normal seedlings increases during maturation, reaching a maximum level in the period close to the stoppage

of dry mass flow from the plant to the seed, considered as the physiological maturity. Precisely at this stage, the physiological potential of the seed is high, if not maximum. From that point on, the physiological potential can only be maintained or decrease by seed deterioration (MARCOS FILHO, 2015).

A similar behavior was observed for seeds of *E. involucrata*, which showed a decrease in the degree of moisture with the advance of maturation. Fruits with a green epicarp showed the highest values for seed moisture (75.5%), which gradually decreased. The lowest values were observed for fruits on the ground (59.3%), but the highest percentage of germination occurred in light red fruits, when seeds had a 72% moisture (ORO et al., 2012). Thus, the moisture content of *C. aurea* seeds can also be an efficient indicator to determine fruit collection point for seed extraction.

## CONCLUSION

The seeds of *C. aurea* are neutral photoblastic, presenting a germination higher than 85% extending for periods longer than 70 days.

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**RESUMO:** *Campomanesia aurea* (Myrtaceae) é uma espécie nativa do Pampa. Possui frutos comestível, potencial ornamental e para recuperação de áreas degradadas. Propaga-se naturalmente por sementes, contudo, para sua utilização com maior eficiência para a produção de mudas ou recomposição da vegetação, são necessários conhecimentos básicos das condições necessárias para se obter máxima germinação. O objetivo do trabalho foi avaliar a germinação de sementes de *C. aurea* e o comportamento germinativo em diferentes condições de luz. Sementes procedentes de três locais de coleta foram analisadas quanto ao fotoblastismo, procedência e maturação pelos testes de germinação, índice de velocidade de germinação, tempo médio de germinação e formação de plântulas normais. Os dados foram submetidos à análise de variância e comparados pelo teste de Tukey a 5% de probabilidade de erro. Para os três locais de coleta não foram observadas diferenças na germinação e no TMG das sementes em ausência ou presença de luz, indicando que trata-se de uma espécie fotoblástica neutra. Para os diferentes locais de coleta, não foi verificada diferenças na porcentagem de germinação, porém sementes coletadas em Barão do Triunfo apresentaram maior IVG e formação de plântulas normais do que sementes coletadas no Morro do Osso e Morro Santana. Essas diferenças podem estar associadas ao grau de maturação dos frutos e a umidade das sementes. Sementes de *C. aurea* são fotoblásticas neutras, apresentando germinação superior a 85%.

**PALAVRAS-CHAVES:** Guabirobinha-do-campo. Myrtaceae. Luz.

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