TEMPERATURE AND LIGHT UNDER THE PHYSIOLOGICAL POTENTIAL OF SEEDS OF *Handroanthus impetiginosus*

TEMPERATURA E LUZ SOB O POTENCIAL FISIOLOGICO DE SEMENTES DE Handroanthus impetiginosus

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ABSTRACT: *H. impetiginosus* belongs to the Bignoniaceae family; it has a great potential for economic exploitation and can be used in landscaping of urban areas, reforestation, recovery of degraded areas, and folk medicine. The experiment was carried out to evaluate the effect of light and temperature regimes on the germination and vigor of *Handroanthus impetiginosus* seeds at the Seed Analysis Laboratory of UFRPE/UAG. The seeds were subjected to light regimes: white, far red, red, and no light at 15°C, 20°C, 25°C, 30°C, 35°C, and 40°C, using a completely randomized experimental design in a factorial scheme (4 × 6), with four repetitions of 25 seeds. The different light regimes did not influence the seed germination of *H. impetiginosus*. The highest germination percentage (92%) and germination speed index (7.94) were obtained at temperatures 28.2°C and 29.2°C, respectively, both under red light. The longest seedling length was also obtained from the seeds subjected to red light regime at 25°C. The temperatures of 15°C and 40°C inhibited the germination of *H. impetiginosus* seeds. *H. impetiginosus* seeds are classified as neutral photoblastics, and constant temperatures of 28.2°C and 29.2°C provided maximum germination.

KEYWORDS: Forest species. Photoblastism. Ipê Rosa. Germination.

INTRODUCTION

Handroanthus impetiginosus (Mart. Ex DC) Mattos, popularly known in Brazil as "Ipê Rosa," is an arboreal species belonging to the family Bignoniaceae, found in several phytogeographic domains. This species is used for furniture manufacturing, afforestation of cities, and reforestation of degraded areas, and presents pharmacological properties (SATHIYA; MUTHUCHELIAN, 2010; PETRONE; PRETI, 2010; UGBABE et al., 2010, FERRAZ; ENGEL, 2011; LOHMANN, 2012; JOSELIN et al., 2013, JIMÉNEZ-GONZÁLEZ et al., 2013).

Is an explored species and because this, in extinction process, another reason is your dense wood and resistant to rot (SCHULZEA et al., 2008). To avoid extinction and to recover the degraded areas, there has been an increasing demand for better seed quality and quantity. Thus, knowledge regarding the conditions that aid the germination process and the production of quality and quantity seedlings for the recovery of the degraded areas and species conservation is essential (SILVA et al., 2014).

In natural environments, the seeds are found under different light and temperature conditions, which may vary according to the canopy structure and their positioning in the successional stage of the forest (PIQUERAY et al., 2013, BASTO; RAMÍREZ, 2015).

Knowledge regarding the germination process and the influence of factors such as light and temperature are paramount for the development of programs aimed to preserve and conserve the native species (GUOLLO et al., 2015). Light is an essential factor for seed germination, and respond differently in function of the species. Shen et al. (2013) verified that the germination of majority of the species present in the forest seed bank in southwest China was influenced by solar, red and extreme red radiation, and photosynthetically active radiation.

This occurs because light is responsible for the activation of phytochrome, a soluble chromoprotein that in its active form absorbs the red wavelength and turns it into an active phytochrome (TAIZ; ZEIGER, 2013).

These varying germination responses to the light quality may help in detecting the regenerative

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mechanism in areas more suitable for seed germination among several ecosystems (DOBARRO et al., 2010).

The species responding to light are divided into three distinct categories: positive photoblastics, whose seeds depend on light to promote germination; negative photoblastics, which undergo reduction or inhibition during germination in the presence of light; and non-photoblastic, which do not respond to the presence or absence of light during germination.

The temperature can break the dormancy and control seed germination. Therefore, germination occurs within a certain limit where the amplitude and absolute values depend on each species. During seed germination, the optimal temperature fluctuates slightly causing reduced seed germination. The optimum temperature is defined as the temperature at which highest seed germination is achieved in the shortest time (SOCOLOWSKI; TAKAKI, 2007).

The luminosity directly influences the seed development. Inside the forest, the cycle of species substitution governed by the tolerance to light, where the primary species, intolerant to shade, begin to climax with species that tolerate shade (CHAZDON, 2008; POMPELLI et al. 2012).

In general, the pioneer species required light to germinate, and present rapid and vigorous plant growth, short life cycle, low diversity, and high population density. In contrast, climax plants have antagonistic characteristics, less seed production, slow growth and germination, and shade development, with long life cycle, greater species diversity, and less population density. The secondary or intermediate species present transient characteristics between the two groups (RODRIGUES; BRANCATION; ISERNHAGEN, 2009).

The objective of this study was to evaluate the effect of light and temperature regimes on the germination and seed vigor of *H. impetiginsous*.

MATERIAL AND METHODS

The research was conducted in the Seed Analysis Laboratory, Federal Rural University of Pernambuco/Garanhuns Academic Unit (UFRPE/UAG), in Garanhuns,-PE, Brazil. The pods were collected from trees in the municipality of Garanhuns-PE (08°53'25"S 36°29'34"W, altitude of 896 m), and the fruits of *H. impetiginosus* were separated with a harvest trimmer. The postharvest seeds were segregated manually by placing the good quality seeds in plastic trays and discarding the bad ones.

Initially, the water content of the seeds was determined by heating them in an oven at 105 ± 3°C and their weight of 1000 seeds was calculated using 8 subsamples of 100 seeds, according to the Rules for Seed Analysis (BRASIL, 2009).

For the germination test, germitest paper was used as a substrate, which was previously sterilized in an oven at 105°C for 5 h and moistened with distilled water with an amount equivalent to 2.5 times the dry weight, according to Brasil (2009). The seeds were disinfected using a 3% sodium hypochlorite solution for 5 min in a dark environment (NASCIMENTO et al., 2007). After disinfetion, the seeds were placed on two sheets of germitest paper, organized in a roll form. For each treatment, 100 seeds, divided into 4 replicates of 25, were used.

The rolls were stored in transparent bags in order to maintain the moisture of the substrate and to simulate the light regime, after that confectioned the envelopes in which placed the rollers within their respective envelopes. We obtained the red light, extreme red light, and dark conditions via a filter comprising an envelope with four layers of red cellophane paper, two layers of red cellophane paper and two layers of blue cellophane paper (YAMASHITA et al., 2011), and four layers of plastic bag in black color, respectively. Provided the light by 20 w fluorescent lamps (white light) located inside the germinators; and the treatments referring to red light, distant red and absence of light, test assembly and done the germination counts in a dark room under green light.

Thereafter, the samples were placed in germination chambers with biochemical oxygen demand (BOD), regulated at temperatures of 15°C, 20°C, 25°C, 30°C, 35°C, and 40°C for a period of 14 days (FONSECA et al., 2005).

For the germination test, the percentage of normal seedlings was evaluated (BRASIL, 2009) from 7th to 14th day. Along with the germination test, we calculated the germination speed index according to the formula proposed by Maguire (1962).

At the end of the germination test, we measured the normal seedlings of each replicate with a graduated ruler in centimeters, and the results were expressed in cm pl^{-1}. To determine the dry mass, the seedlings were packed in paper bags and heated in the oven at 105°C for 24 h, followed by weighing in an analytical balance with an accuracy of 0.001 g, and the results were expressed in g pl^{1} (NAKAGAWA, 1999).

The completely randomized experimental design was used, in a factorial scheme of 4 (light regimes) × 6 (temperatures), with 4 replicates of 25 seeds each. Thereafter, the data of measured variables was subjected to analysis of variance by the test F. Next, the data was subjected to the regression analysis, by applying the quadratic model.

RESULTS AND DISCUSSION

The water content of the *H. impetiginosus* seeds during the experiment commencement was 9.25% and the weight of 1000 seeds, on average, was 9.23 g. The water content acts as an important determinant for the test performance, considering that the uniformity of the water content of the seeds is essential for standardizing the evaluations and obtaining consistent results; moreover, high water content can enhance the seed performance (MARCOS FILHO, 2005). The weight of 1000 seeds and water content will predict the seed count per kilograms and that associated with the germination percentage, we have the quantity of seeds used in the production of seedlings.

Figure 1 presents the germination percentage as a function of temperature and light regimes. The quadratic polynomial regression model derives values with maximum estimated points at temperatures of 28.5°C, 27.9°C, 28.3°C, and 27.6°C and maximum germination of 80%, 83%, 92%, and 79% in the bands of white, dark, red, and extreme red light, respectively.

Notably, no statistical differences were observed between red, distant red, and dark light regimes at 20 °C. At 25°C, maximum germination was observed when the seeds were subjected to red and extreme red light regimes (72% and 65%, respectively). No statistical difference was observed between the light regimes when the seeds were subjected to a temperature of 30°C, whereas at 35°C, similar behavior was observed for the red and white light regimes. The seeds subjected to 15°C and 40°C did not germinate, and were deteriorated at the end of the test (Figure 1).

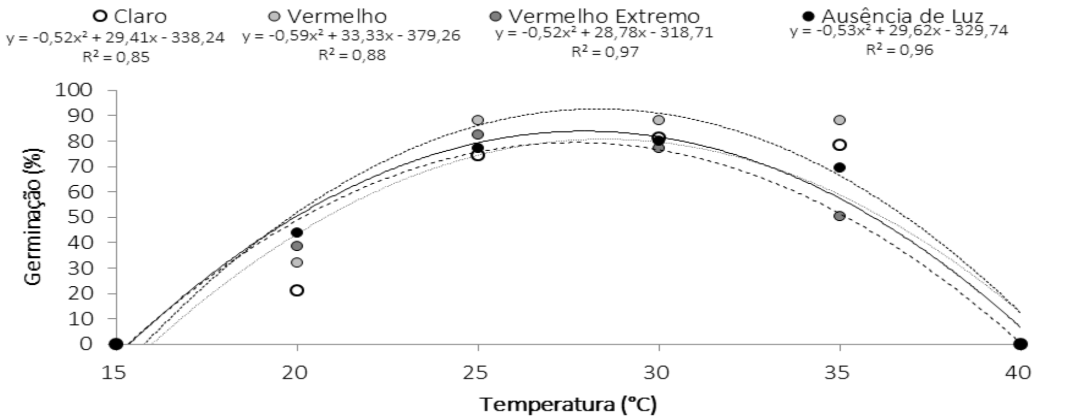

Figure 1. Germination of *Handroanthus impetiginosus* seeds in function of temperatures and light regimes (Clear, Red, Extreme Red and Absence of Light).

During germination at low (5–10°C) or extremely high (>45°C) temperatures, the metabolism is not appropriately activated, and the reserves deployed in phase I are necessary for the growth and development of the embryo in phase III and are not carried to the meristematic regions (BEWLEY et al., 2014). Thus, the germination remains incomplete, and the seeds do not pass beyond phase II. The temperature interferes with the biochemical reactions that determine the germination process, thus influencing the capacity and speed of germination. Hence, seeds have the ability to germinate within a certain temperature range, characteristic for each species; however, the time required to reach the maximum percentage of germination is temperature dependent (BEWLEY et al., 2014).

Initial absorption rate and temperature can drastically alter the germination and seed quality, wherein some seeds are damaged by rapid absorption at low temperatures and this process occurs due to damage to the membrane system, resulting in leaching of cellular content and negatively affecting germination. In addition, low temperatures reduce the enzymatic activities in seed metabolism, to an extent where the essential pathways for the onset of germination stop functioning (CASTRO et al., 2004; MATHEUS; LOPES, 2009; MARINI et al., 2012). At high temperatures, a reduced supply of free amino acids and protein synthesis (SANTOS, SUGAHARA; TAKAKI, 2005), is observed among the seeds of species that occur in various regions of Brazil, and few germinate at temperatures above 40°C (NOGUEIRA et al., 2014).

Seeds belonging to various species of the same botanical family, for example, *Tabebuia serratifolia, T. chrysotricha and T. roseo-alba*, revealed germination behavior indifferent to light, with the optimum temperature range between 20°C and 30°C (SANTOS, SUGAHARA; TAKAKI, 2005). Basto and Ramirez (2015), who investigated the effect of light quality on the germination of *T. rosea* at 30°C, concluded that the seeds of *T. rosea* responded to all treatments of light quality; however, the lengths of short and medium waves increases the germination speed and that the absence of light has a negative effect.

After studying the effects of light quality on the germination of five tree species (*Larix kaempferi, Phellodendron amurense, Acer monkey, Fraxinus mandshurica, and Pinus koraiensis*) found in the Chinese forest, Zhang, Zhu and Yan (2012) concluded that the quality of light significantly affected the germination of large seeds.

Although germination is observed in all light regimes, it is higher under the red light spectrum, indicating that germination is faster when it occurs under an enclosed canopy compared to complete day light, that is, the seeds of *H. impetiginosus* prefer forest conditions under which smaller thermal amplitudes predominate. Similar characteristics were found in seeds of *Mimosa caesalpiniifolia*, in which there is a greater capacity of seed germination and consequent establishment of seedlings in the field, making them able to resist the adverse environmental conditions (HOLANDA et al., 2015).

According to the classification proposed by Takaki (2005), the seeds of *H. impetiginosus* must possess phytochrome of the type A (phyA) controlling the germination through the very low fluency response. However, categorizing a seed as sensitive or insensitive to light depends on the maturation conditions, storage, imbibition temperature, test conduction and osmotic treatment (AMARO et al., 2006).

The seeds of *H. impetiginosus* germinated both in the presence and in absence of light, and were classified as neutral photoblasts. According to Holanda et al. (2015), if the light does not influence seed germination, they can germinate in areas with different successional stages.

According to Leão et al. (2015), the best temperature for seeds of *H. serratifolius* was 30°C in white light conditions, whereas for *H. impetiginosus* seeds, the average temperature of 28 °C favors germination, also classified as neutral photoblastic.

Figure 2 presents the values of germination speed index of *H. impetiginosus*, subjected to different temperatures and light regimes. At 20°C, the highest germination rate index was 1.68 in the absence of light, and that of the red light at temperatures of 25, 30 and 35°C, was 7.29, 7.28 and 10.27 respectively. At 30 °C, the highest index was 9.24, as observed in the white light range.

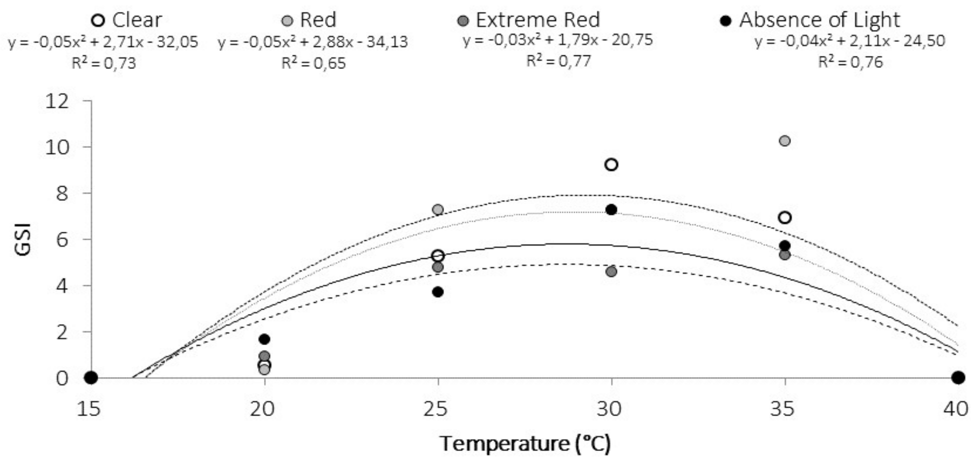

Figure 2. Germination Speed Index (GSI) of *Handroanthus impetiginosus* seeds in function of temperatures and light regimes (Clear, Red, Extreme Red and Absence of Light).

The variation in the germination velocity index (IVG) considering the function of estimated temperature, derivatives to the second degree polynomial regression, presents maximum values at 28.9, 28.7, 29.2 and 28.6°C, thus revealing the following indices of germination speed with values of 7.23, 5.83, 7.94 and 4.89 for the white, dark red, and extreme red light regimes respectively.

These results are in accordance with those of Bewley et al. (2014), where the germination rates are highly sensitive for temperature, generally increasing till they attain ideal temperature and then decreasing drastically. Although the total percentage of germination reveals a broad spectrum of maximum temperature, it is possible to use the germination rates to accurately identify the ideal germination temperature. Nogueira et al. (2014) reported that the greater influence in germination speed in *Dalbergia cearensis* seeds is due to temperature rather than by light and dark conditions.

Noteworthy, the higher temperatures lead to greater water absorption process, accelerate enzymatic activities, and enhance metabolic activity, in order to accelerate and standardize the germination process (MATHEUS & LOPES, 2009). According to the results of Carvalho & Nakagawa (2012) the higher the temperature, the faster and more uniform the germination, so that the metabolic processes are not compromised by protein denaturation due to excessive temperatures where germination is annulled.

In Figure 3, we present the values related to the total length of seedlings in function of temperatures and light regimes. The values derivatives for the polynomial regression model with maximum estimated points at temperatures of 27.5; 27.8; 27.6 and 27°C in the white, dark, red and distant red light regimes, respectively.

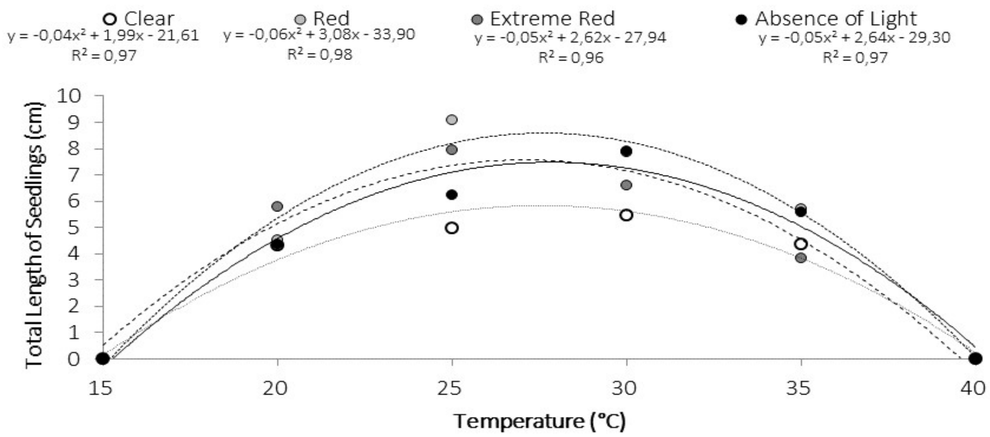

Figure 3. Total Length of Seedlings of *Handroanthus impetiginosus* in function of temperatures and light regimes (Clear, Red, Extreme Red and Absence of Light).

The temperatures above the optimum range harm the seed growth with a consequent decrease in the rate of photosynthesis and the concentration of the rubisco enzyme and chlorophyl (YAMASAKI et al., 2002). The *H. impetiginosus* seedlings are able to develop in a wide range of environmental conditions, which is essential for the natural regeneration of the species. The species under study reveals characteristics of germination and initial growth in, preferably, the sub-forest environment, in which smaller thermal amplitudes predominate.

The values for the dry mass of *H. impetiginosus* seedlings in relation to temperature and light regimes are presented in Figure 4. The values were adjusted to the quadratic regression model with maximum points at the temperatures of 28.28, 27.41, 28.81 and 28.15°C for the regimes of white, dark, red, and extreme red light, with maximum values of 0.22, 0.34, 0.39, and 0.35 g, respectively.

The highest dry mass content obtained in the mentioned treatments explained by the provision of the conditions necessary for the germination because the seeds originate seedlings with higher growth rate, due to the greater capacity of transformation and supply of reserves of the storage tissues and greater incorporation of these embryonic axes (NAKAGAWA, 1999). Sunlight when filtered by green leaves has its spectral distribution altered due to selective absorption of leaves, specifically by chlorophylls (SMITH, 2000).

The effect of high temperatures on the reduction of seed vigor may be explained by possible enzymatic alterations, by the physiological condition of the seed or the insolubility of the oxygen under these conditions, thus increasing its demand and accelerating the respiratory rate of the seeds (MARCOS FILHO, 2005). In addition, low temperatures may lead to decreased metabolic rates to the point where processes essential for germination negated (GUAN et al., 2009).

While evaluating the dry mass of the seedlings of *Platymiscium floribudum* at 25°C, the light regimes employed were not satisfactory for this variable; however, at 30°C, the highest dry mass contents were obtained in the red and extreme red light regimes (ALVES et al, 2016). Seeds of *Quercos liaotungensis* subjected to a high level of radiation, without shading, gave rise to seedlings with a larger basal diameter of the stem, root system growth, and dry mass accumulation, but reduced the height of the seedlings (YAN, WANG & ZHOU, 2011). In *H. impetiginosus*, the white light, at 25°C and 30°C, provided lower yields of seedlings in both size and dry mass.

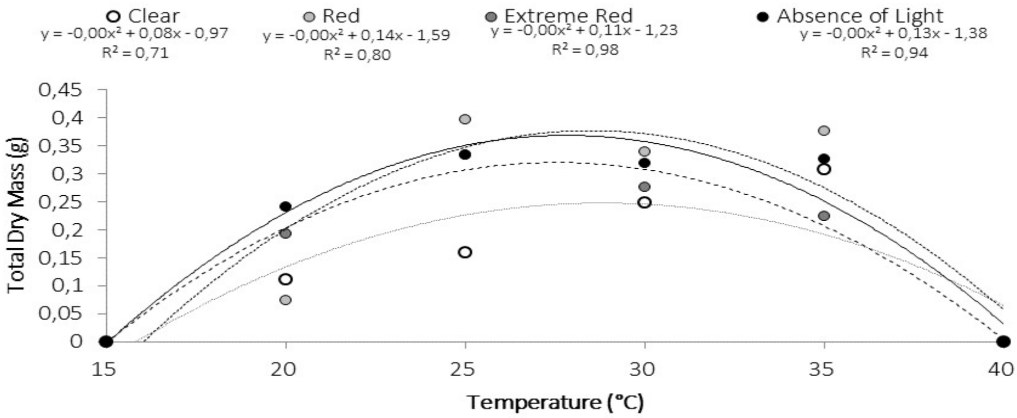

Figure 4. Total Dry Mass of seedlings of *Handroanthus impetiginosus* in function of temperatures and light regimes (Clear, Red, Extreme Red and Absence of Light).

In seeds of *Parkia platycephala* and *Dalbergia nigra*, at constant temperatures of 25°C and 30°C, the germination and vigor tests presented the highest dry mass values (GONÇALVES et al., 2015, MATOS et al., 2015). These interactions are essential for understanding the ecophysiological status of *H. impetiginosus* seeds, since the ideal temperature, water retention capacity, and the quantity of light that the substrate allows to reach the seed can be responsible for different germination responses (FIGLIOLIA et al., 1993).

The seeds of *Handroanthus impetiginosus* are neutral photoblasts and the germination test of these seeds may be carried out at constant temperatures of 28.2°C and 29.2°C.

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RESUMO: *H. impetiginosus*, pertencente à família Bignoniaceae, apresenta grande potencial para exploração econômica, podendo ser utilizado no paisagismo de áreas urbanas, reflorestamentos, recuperação de áreas degradadas e na medicina popular. O experimento foi conduzido no Laboratório de Análise de Sementes da UFRPE/UAG com o objetivo de avaliar o efeito dos regimes de luz e temperatura na germinação e vigor de sementes de *Handroanthus impetiginosus*. As sementes foram submetidas aos regimes de luz: branca, vermelho distante, vermelho e ausência de luz sob as temperaturas de 15, 20, 25, 30, 35 e 40°C, sendo utilizado o delineamento experimental inteiramente ao acaso, em esquema fatorial (4x6), com quatro repetições de 25 sementes. Os diferentes regimes de luz não influenciaram na germinação de sementes de *H. impetiginosus*. A maior porcentagem de germinação (92%) e índice de velocidade de germinação (7,94) foram obtidos nas temperaturas 28,2 e 29,2°C, respectivamente, ambos no regime de luz vermelha. O maior comprimento de plântula também foi obtido de sementes submetidas ao regime de luz vermelha na temperatura de 25ºC. As temperaturas de 15ºC e 40ºC inibiram a germinação das sementes de *H. impetiginosus*. As sementes de *H. impetiginosus* são classificadas como fotoblásticas neutras e as temperaturas constantes de 28,2 e 29,2°C proporcionaram máxima germinação.

PALAVRAS CHAVE: Espécie Florestal. Fotoblastismo. Ipê rosa. Germinação.

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