Colegio de Postgraduados

Preliminary study on the reproductive phenology of *Eucalyptus urophylla* in Huimanguillo, Tabasco (Mexico)

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ABSTRACT

Objective: to describe the reproductive phenology of *Eucalyptus urophylla* in Huimanguillo, Tabasco.

Design/Methodology/Approach: this is a preliminary, descriptive, and non-experimental study of the different stages of the reproductive phenology cycle of *E. urophylla* conducted in a 16-year-old provenance-progeny trial with 63 half-sib families. The study was done during 2019, 2020 and 2021.

Results: the reproductive cycle of *E. urophylla* lasts approximately one year. There was a lag of about a month and a half for the beginning of flowering among genotypes that flowered during the three years of evaluation.

Study Limitations/Implications: the results are limited to a single site condition; so, it is recommended to make evaluations in other locations. This is, to observe plant behavior in different site conditions to evaluate the potential of the species development in southeastern Mexico.

Findings/Conclusions: the beginning of the reproductive cycle of *E. urophylla* coincided with that of other species of the genus from tropical and subtropical climate. However, the duration differed compared to other genotypes of the same species growing in other parts of the world under similar environmental conditions. This study allowed to know relevant information of the species in Huimanguillo, Tabasco, which will contribute to improve the process of domestication of this species in the southeast of Mexico.

Keywords: eucalyptus, flowering, controlled pollination.

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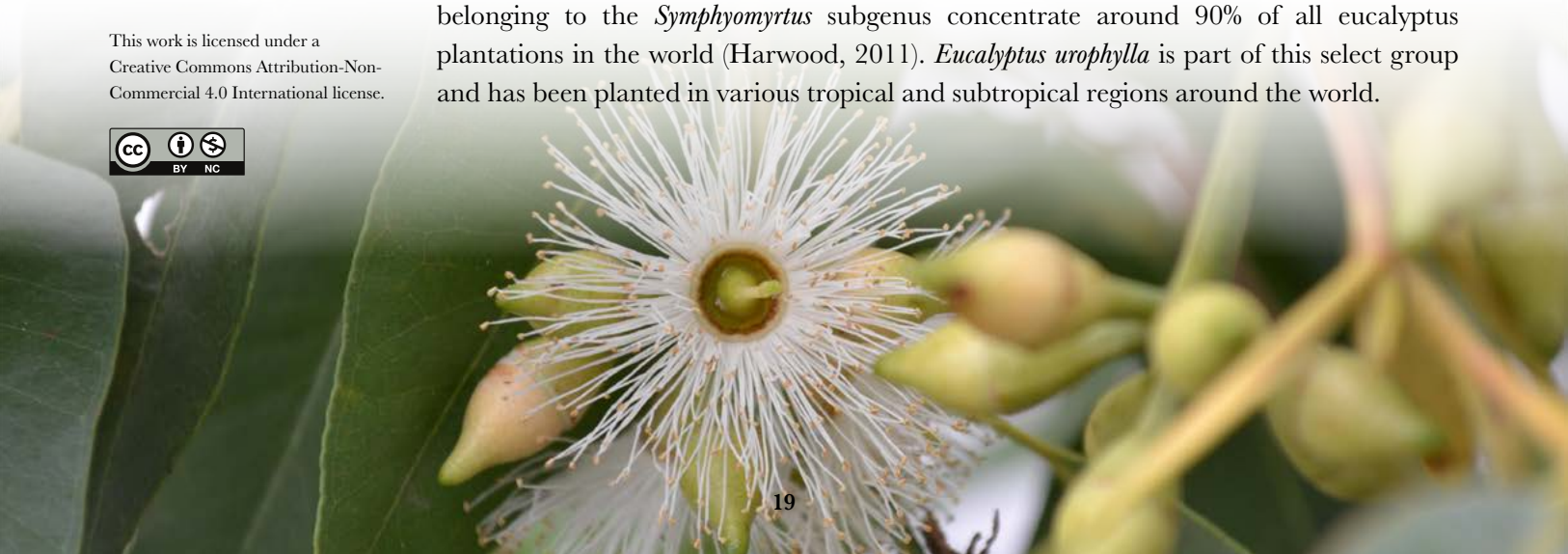
INTRODUCTION

Eucalyptus is one of the most planted genera with around 30 million hectares worldwide (Payn *et al.*, 2015). The genus comprises about 700 species, however, only nine of them belonging to the *Symphyomyrtus* subgenus concentrate around 90% of all eucalyptus plantations in the world (Harwood, 2011). *Eucalyptus urophylla* is part of this select group and has been planted in various tropical and subtropical regions around the world.



In Mexico, *E. urophylla* plantations have great potential for the development of the forestry industry due to their rapid growth and the quality of their wood for various purposes. Currently, most of the production of this species is used for the elaboration of medium density fiberboard (MDF). Previous research in our country has made progress in the genetic improvement of *E. urophylla*. Studies have been carried out on wood density (Ortega-Ramírez *et al.*, 2020), pollen management (Ortega-Ramírez *et al.*, 2021) and estimation of genetic parameters to model growth (Sánchez-Vargas *et al.*, 2004; Ignacio-Sánchez *et al.*, 2005).

However, aspects of the sexual reproductive cycle are still unknown in order to carry out controlled crosses, which is an activity necessary for the continuity of genetic improvement programs. With a better understanding of the stages of the reproductive cycle, it will be possible to program more efficient crosses, which increase the quantity, and genetic and physiological quality of the seeds (Mora and Ferreira, 1978).

For this reason, the objective of this study was to describe the phenological stages of the sexual reproductive cycle of *E. urophylla* in populations of Huimanguillo, Tabasco.

MATERIALS AND METHODS

For the study, 114 (16-year-old) trees were observed for three consecutive years; selected from 63 families in a provenance-progeny trial of the company "Forestaciones Operativas de México SA de CV" (FOMEX) in Huimanguillo, Tabasco.

Trees were evaluated for 10 years and selected due to proven records of good growth and cylindrical stem. These trees will be part of a second stage of improvement, so it was essential to obtain seeds and pollen to assess their genetic superiority. In addition, scions were obtained to graft them in a controlled pollination orchard of the company. As part of the breeding process, these trees will be crossed to obtain progenies of high genetic quality.

During the first stages of flower development, observations were made to the trees weekly or once every 15 days. Since the appearance of the flower bud until the anthesis stage, monitoring was done at ground level with binoculars. Climbing was made on some chosen trees to obtain buds and flowers for the photographic record. In those cases, climbing started once the flower opened to collect pollen and seeds.

After the period of natural pollination, record collections were prolonged, in order to monitor the development of the fruit. To describe the different stages, the approach of Loewe *et al.* (1996) for *Eucalyptus* species. In addition, virtual herbarium collections worldwide that had records of flowers and fruits of *E. urophylla* were reviewed to compare with the phenology observed at the study site.

Despite the fact that the collections are quite extensive, few herbaria have precise information on the phenology of *E. urophylla*. The virtual collections of the Global Biodiversity Information Facility —GBIF (GBIF, 2022); of the herbaria Missouri Botanical Garden (MO, 2022), The New York Botanical Garden (NYBG, 2022), Museum National d'Histoire Naturelle (MNHP, 2022), REFLORA (REFLORA, 2020) and University of South Florida Herbarium (USF, 2022) supplied information on the presence of flowers and fruits. As well as the date of collection to classify the possible phenology state in which the tree was found at the time of collection.

During the years of evaluation, records of precipitation, relative humidity and solar radiation were kept with a portable weather station of Pessl Instrument Ges® located approximately 30 km from the selected trees to see if those variables could explain the reproductive phenology of *E. urophylla* during the evaluation years.

RESULTS AND DISCUSSION

The inflorescence of *Eucalyptus urophylla* is axillary, it mainly developed in odd number umbels of three to seven flower buds; the most abundant were groups of five to seven. Groups of buttons were also found in pairs of four or six, though these are rarer. Flowers do not have typical petals, but petals merge to form the operculum (Loewe *et al.*, 1996) characteristic of the species of the *Symphyomyrtus* subgenus (Junior and Garcia, 2021). The operculum protects the male (stamens) and female (stigma) reproductive system.

Flowering began in some genotypes in May, and it extended throughout June. This floral asynchrony of *E. urophylla* has already been reported in other species of the genus such as *Eucalyptus dunnii* (Sousa and Higa, 1991) and *Eucalyptus grandis* (Aguiar and Kageyama, 1987; Chaix *et al.*, 2007). According to Chaix *et al.* (2007) this characteristic may be linked to the origin of the parents. Species with a wide natural distribution have been subject to different site-specific evolutionary processes, which differentiate the adaptation processes of the species.

Spencer *et al.* (2020) found that the provenance effect also modified the number of flowers per tree in *Eucalyptus loxophleba*. Floral asynchrony plays a role for and against in eucalyptus improvement programs. In open-pollination orchards, a much extended floral asynchrony can be a problem, since it would reduce panmixia. Since mating would take place just between the few genotypes that were blooming together, so decreasing the genetic variability of the progenies.

In orchards with controlled pollination, foliar asynchrony can be managed differently. The pollen of those genotypes that bloom early is stored to apply it to the genotypes that bloom later. In a similar way, it is recommended to store the pollen of the late-blooming genotypes for use it in the following season with the genotypes that flower first (Molina *et al.*, 2014); thereby ensuring an increase in the genetic variation of the progenies in breeding programs.

In accordance with the approach of Loewe *et al.* (1996), nine stages of reproductive phenology were observed in eucalyptus at Huimanguillo, Tabasco (Table 1). The beginning of flowering in the first week was visible by the emergence of the flower bud covered by the bract (Figure 1a). Starting at the third week (Figure 1b) the buds lost the outer operculum, they remained as such, until their complete development (Figure 1e).

During the three years of the study, the beginning of flowering coincided with the end of the dry season (May and June) and the longest days of the year in the Northern hemisphere (Figure 2).

The first flowers matured at the end of July and the ripening period extended by a month up to September, what gave an average of 90 days from the start of flowering until the flower is fully mature. The flower of *E. urophylla* is hermaphroditic with a staminophore that supports numerous white stamens and the stigma. On top of the stamens are the anthers

Table 1. Reproductive phenology stages of *Eucalyptus urophylla* in Huimanguillo, Tabasco.

Phenophase	Week	Description	J	F	M	A	M	J	J	A	S	O	N	D
0	0	The floral bud emerges in the leaf axils of the plant, only the bract that protects the flower is visible (Figure 1a)					X	X						
1	1	The green bract darkens, opens, dries up and falls, the flower bud emerges.					X	X						
2	3	The flower bud grows, the outer operculum detaches (Figure 1b)					X	X						
3	4-12	Full development of the flower bud					X	X	X	X				
4	12	The operculum changes to a yellow color and detaches, leaving the stamens and stigma visible (Figure 1f).							X	X				
5	13-14	The flower is in anthesis, the anthers release pollen, four to six days after the flower opens, the stigma slightly thickens and releases a crystalline nectar with a sweet scent							X	X	X			
6	14-15	The stamens change to dark brown color and detach, the stigma loses turgidity and undergoes gradual color changes, from green to brown. When it is completely brown, it detaches from the flower (Figure 1g).									X	X		
7	16-50	Fruit growth, the green color is characteristic of this phenophase (Figure 1h).	X	X	X	X	X			X	X	X	X	X
8	45-55	The ripe fruit releases the seed, the capsule turns dark brown and acquires a woody appearance and texture						X	X					
9	>55	The empty capsules fall, some remain on the tree for a longer time						X	X	X	X	X		

that store pollen, the anthers are pale yellow (Figure 1F). The flowers are protandrous, this means that the male sexual organs mature before the female ones. This behavior has been observed in other *Eucalyptus* species (Sousa and Higa, 1991; Rojas, 2014).

According to Mora and Ferreira (1978) this characteristic is a strategy to avoid self-fertilization and encourage panmixia in natural populations. Spencer *et al.* (2020) found that the highest sum of hot temperature in one year advanced the flowering in *E. loxophleba*, which shows that flowering, in addition to genetic control, is also affected by environmental conditions and their interaction. Mora and Ferreira (1978) studied the reproductive phenology of *E. urophylla* in Brazil, those authors found different results from those observed in Huimanguillo, especially in fruit development time.

In the study by Mora and Ferreira (1978) the flowering season began in the months of November and December; the fruit matured in May, six months after the start of the reproductive cycle. Such a mentioned period was shorter than that observed in Huimanguillo; however, the beginning of flowering coincided with the end of spring in

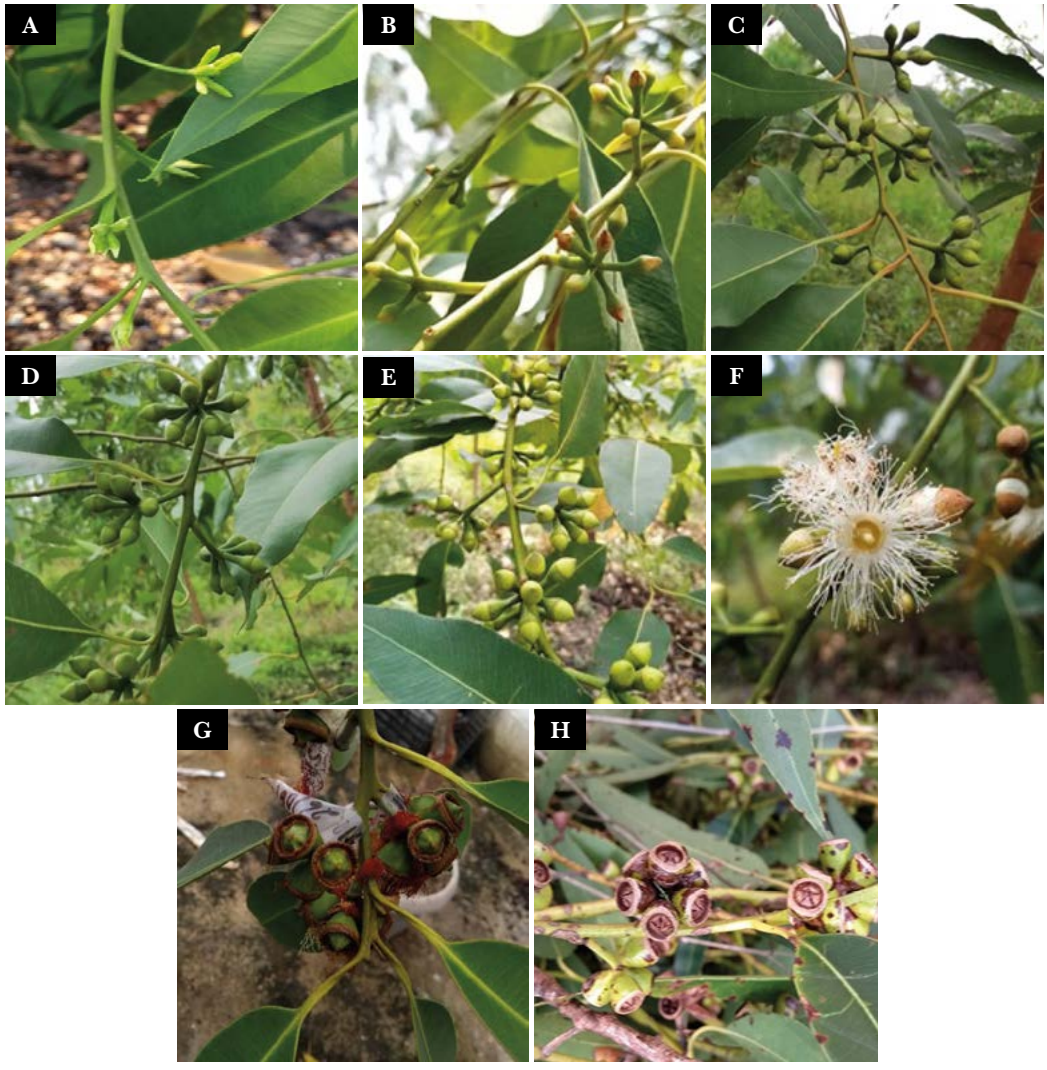


Figure 1. Reproductive phenology stages of *Eucalyptus urophylla* in Huimanguillo, Tabasco (Mexico). A: week 1, B: week 3, C: week 7, D: week 10, E: week 12, F: week 13, G: week 15, and H: week 50.

both Mexico (May-June) and Brazil (November-December). Flowering in both places coincided with the beginning of the rainy season (Figure 2); also did with the longest days and those that receive the greatest amount of light in the year (Figure 2). These are the main environmental factors that promote flowering (Molina *et al.*, 2014).

The release of pollen occurred between weeks 14 and 15, in the months of August and September (Table 1). Between four and six days later the stigma became receptive. A typical feature to distinguish receptivity is the slight thickening and the release of crystalline nectar at the tip of the stigma. Between week 16 and 17, once the flower has been fertilized, the stamens change from white to a reddish color, the stigma loses turgidity and begins to change from green to dark brown to later detach (Figure 1G).

The fruit matured between weeks 45 and 50, approximately, this stage is easy to identify because the fruit turns brown, with a woody structure (Figure 1H). The reproductive cycle of *E. urophylla* in Huimanguillo was shorter than that observed in other species such as

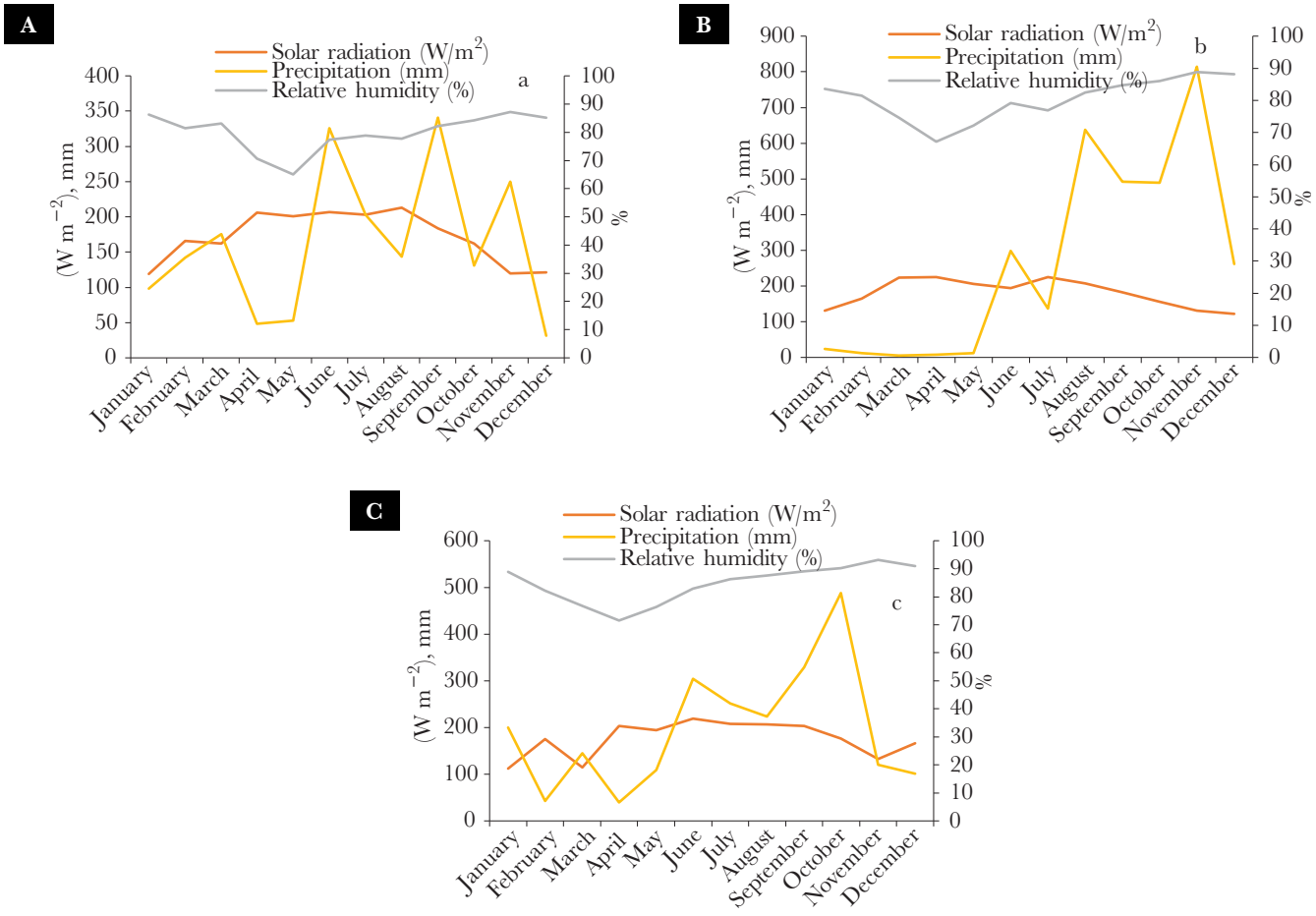


Figure 2. Environmental variables in the study site in A: 2019, B: 2020 and C: 2021.

Table 2. Collections of *Eucalyptus urophylla* reviewed virtually in herbaria located worldwide, with data of their reproductive phenology stage.

Herbarium	Collection site	Collection date	Phenophase
REFLORA	Itatinga, Sao Paulo, Brazil	28/02/2007	6
REFLORA	Itatinga, Sao Paulo, Brazil	28/02/2009	5
REFLORA	Rio Grande do Sul, Brazil	04/03/2010	3
REFLORA	Piracicaba, Sao Paulo, Brazil	03/06/2010	8, 5, 4, 6
The New York Botanical Garden	Central Mountain Range, Dominican Republic	04/07/1986	9
Herbarium of the Paris Museum	Timor Island, Indonesia	16/06/1968	9
Herbarium of the Paris Museum	Kourou, French Guiana	7/03/1987	3
Herbarium of the Paris Museum	Flores Island, Indonesia	20/10/1936	3
Herbarium of the University of South Florida	Paramaribo, Suriname	24/03/1968	3
Herbarium of the University of South Florida	Lake Yojoa, Honduras	21/04/1985	8
Herbarium of the University of South Florida	Miami, Florida, USA	22/02/1945	8

Eucalyptus dunnii, which has a reproductive period of almost two years from the moment the flower bud emerges until the seed is released. It was much shorter even than that observed in *Eucalyptus delegatensis* or *Eucalyptus fastigata*, which have a reproductive cycle of up to three years (Sousa and Higa, 1991). Table 2 shows different collections of *E. urophylla* in the herbaria reviewed virtually. Based on the photographs obtained, those were assigned a tentative reproductive phenology stage for comparison with the phenology observed in Huimanguillo, Tabasco.

The preliminary results observed in this study are different from those observed in the collections of the herbaria reviewed. The season of initiation and maturation of the flowers found in the herborized collections extended from January to July, with the release of seeds and fruit drop in the months of November and December. Facts that coincide very little with those observed in Huimanguillo.

CONCLUSIONS

Although the study is preliminary, it allows us to know important information regarding the reproductive phenology of *Eucalyptus urophylla* in Huimanguillo, Tabasco. This is an important aspect for the conservation and reproduction of the species given its economic importance in the forestry industry in Mexico.

The phenology did not change strongly during the three years of evaluation. Facing an environment of climate change these stages of reproductive phenology might be altered within longer evaluation periods. Therefore, it is recommended to continue implementing phenological studies in order to specify the information regarding *E. urophylla* for conservation and management in Southeastern Mexico.

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