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**Vanilla wild relatives naturally occurring in Atlantic rainforest–central biodiversity
corridor in southern Bahia – Brazil**

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Abstract

The genus *Vanilla* Mill., due to being a source of vanillin, an aromatic organic compound with high commercial value, is one of the best-known of the Orchidaceae family. The extracted oil has been used in the food, pharmaceutical, and cosmetics industries. Considering the complex activity of growing vanilla for oil extraction, studying species of this genus is of huge interest for conservation and breeding programs. The current study was conducted in the extreme south of Bahia, in northeastern Brazil. A region characterized by the presence of many traditional communities and small farmers, and a human development index below the OECD average. The study area covered four different provenances, including the conservation unit “Descobrimento National Park” and its surrounding communities, and aimed to identify the naturally occurring *Vanilla* species. Expeditions were carried out in the forest to find and register specimens using geo-location for further monitoring and data collection on the morphology, habitat, floral visitors, and floral phenology. The observation of the reproductive organs occurred weekly/daily for a period of 60 days, from the flower bud stage until the first flower has fallen naturally. Fifteen specimens were marked all over the studied area, three of them blossoming provided data for this study. Through the analysis of the morphological and phenological characteristics of the observed flowers, it was possible to identify with certainty two species, *Vanilla phaeantha* Rchb.f. and *Vanilla chamissonis* Klotzsch. These results contribute to determining and registering vanilla wild relatives' occurrence in the extreme south of Bahia. In addition, relevant information was generated on phenological, and morphological aspects of the species found in this environment.

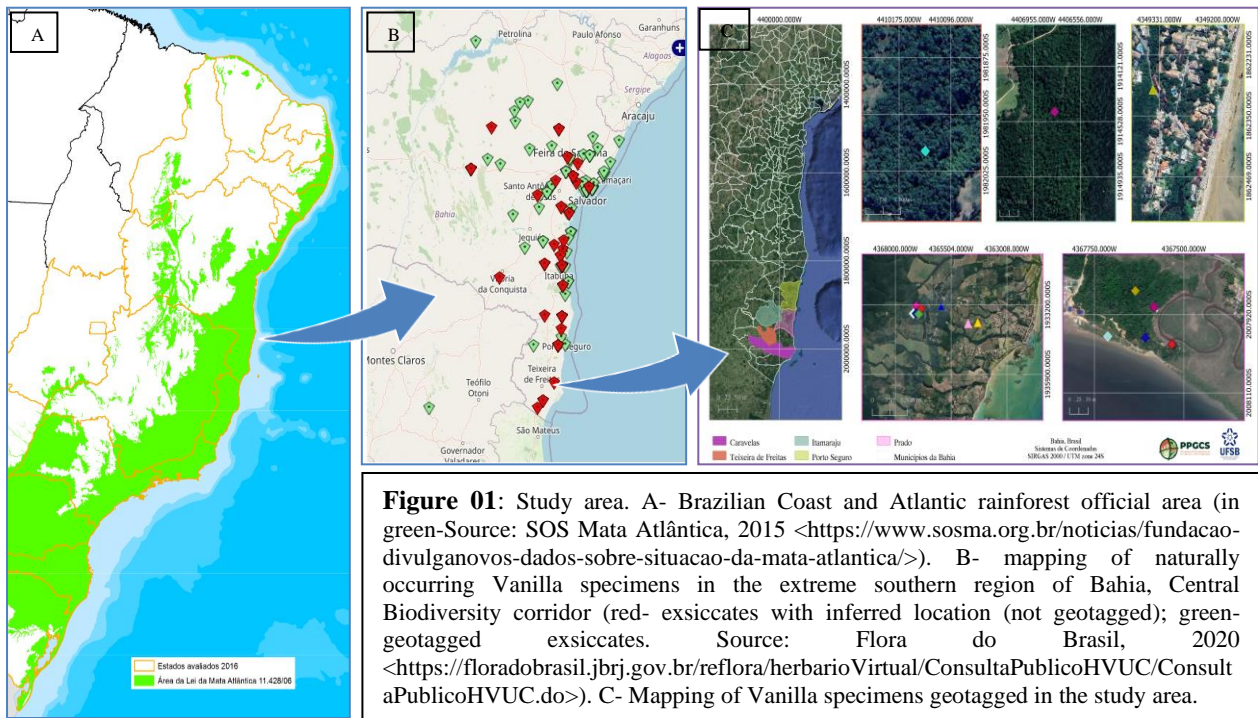
Keywords: Floral biology, Orchidaceae, vanilla, wild relatives

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Introduction

For most of the population, it is not of knowledge that *Vanilla* belongs to a genus of the same name in the Orchidaceae family together with others so far estimated 899 genera and 27,801 species (THE PLANT LIST, 2018). In Brazil, 20 of the 38 registered *Vanilla* Mill. species are endemic, and distributed in the Amazon, Caatinga, Cerrado, and Atlantic Rainforest biomes (Flora do Brasil, 2020). The plant develops best in the range of 21-32°C and around 2,500mm of rain per year, requiring water stress for flowering (Reis, Brondani and Almeida, 2011). Considering the complex activity of growing vanilla for oil extraction, conserving the diversity guarantees the potential for resilient new species and cultivars to be grown (Tompa, 2012).

Environmental changes are leading to genetic erosion and extinction risk of cultivated species (*V. planifolia* and *V. pamplona*) and wild relatives demand efforts for studying and collecting genetic resources (Castañeda-Álvarez, et al., 2016). In Southern Bahia one of the most biodiverse spots of the Atlantic Rainforest is located, this central biodiversity corridor is also called “Hileia Baiana”. The region lacks scientific information about Vanilla’s natural occurrence, the data available at the REFLORA Virtual Herbarium website comes from old records with genus level determination only and without geotagging, raising doubt about its accuracy (**Figure 01**). However, local people affirm the traditional use of vanilla plants. It led to the objective of this study, to find specimens of the Vanilla genus in the region and describe the floral biology aiming to identify the species to confirm its occurrence.



Material and Methods

The expeditions took place in the municipalities of Teixeira de Freitas, Caravelas, Prado (in the district of Cumuruxatiba), and Itamaraju, in the State of Bahia, Northeast of Brazil. The approached area was conserved Atlantic Rainforest fragments, in private reserves and public conservation units, such as the Descobrimento National Park (PND) and the Marobá trail, which belongs to the Visitor Center of the Abrolhos Marine Park (PARNAM). It took place between December 2021 and April 2023. The approached areas were reconnoitred together with local officials, using the walking method (FILGUEIRAS et. al 1994), which consisted of walking through the entire study area, identifying the specimens, geotagging, and collecting, when possible, plant material. The specimens considered for the research were those in their fertile period. Monitoring notes and photographic recordings were made to compare the development and possible identification between species. The plants selected to be monitored are located in the Indigenous Village of Tibá, belonging to the Descobrimento National Park, in Cumuruxatiba, Prado, Bahia. Fertile plants were monitored on habitat, morphology, phenology, and floral visitors for a period of 60 days, from the flower bud stage until the first flower senescence. Analyses were carried out throughout the daylight. The average temperature was max 25.1°C/ min 24.4°C and the humidity (RH%) of the site was max 78.9/ min 68.4.

Results and Discussion

Wild species of the genus *Vanilla* are mostly described in the literature as occurring in the north coast portion of the State of Bahia, with only a few records of occurrence without determination at the species level in the extreme southern Bahia region (REFLORA, 2020). Sixteen specimens of the genus *Vanilla* were found in four different environments, on the roadside, in the conserved forest, on sandbanks (restingas), and river side. Three of them were blossoming and provided data for this study. Based on the organography of Vidal, Vidal, and De Paula (2021), it was possible to characterize the found plants as having axillary, multi-flowered, indefinite, and spike-like inflorescences. The flower is epigynous. The phenological and morphological characteristics are presented in **Tables 02** and **03**. The data were compatible with those described for the species *Vanilla chamissonis* Klotzsch (Hoehne, 1945) and *Vanilla phaeantha* Rchb.f. (Karremans et al., 2020). A study by Lopes et al. (2019) confirms that the pods of *Vanilla phaeantha* Hoehne appear to be an alternative source of vanilla flavor since they have been shown to express some of the most important enzymes in the biosynthesis of vanilla flavor compounds.

Table 02- Phenological aspects of the observed specimens of *Vanilla* spp.

Specimens	Beginning Inflorescence development	Beginning of flowering	From bud to senescence	From inflorescence start to first flower fall	Open flower time	From flower open till fall	Odor
<i>V. chamissonis</i>	September	November	~ 43 days	~ 49 days	4 days	6 days	Cinnamon
<i>V. chamissonis</i>	September	November	~ 41 days	~ 44 days	2 days	3 days	Sweet, but not intense
<i>V. phaeantha</i>	October	November	~ 34 days	~ 36 days	~ 24hrs	2 days	Barely Noticeable

Table 03- Floral morphological metrics of *Vanilla* spp. observed specimens.

Specimens	Sepals (cm)	Petals (cm)	Lip (cm)	Ovary (cm)	Column (cm)
<i>V. chamissonis</i>	5,4	5,2	5,5	3,7	4,0
<i>V. chamissonis</i>	8,3	8,2	9,6	6,2	6,5
<i>V. phaeantha</i>	8,7	8,5	8,3	3,7	6,4

V. chamissonis specimens presented smaller floral verticils ($\pm 5,5$ cm- **Figure 03**). During the observation periods, it was possible to smell a pleasant, sweet scent, notes of cinnamon exhaling from young flowers, which intensified as the corolla closed and lost its intensity when completely closed. The complete phenological cycle from the start of inflorescence development to the fall of the second flower totaled 49 days. The floral verticils of *V. phaeantha* are larger ($\pm 8,5$ cm- **Figure 04**). The flowers complete their senescence within a day and have no characteristic smell. This description is compatible to that made by Anjos, Barberena and Pigozzo (2017). The complete cycle involving the start of inflorescence development, flower bud development and the fall of the first flower took an average of 28 days. As also reported by Reis, Brondani, and Almeida (2011), ants were found at all stages of flower bud development in both specimens, concentrated at the base of the perianth, especially at the start of growth and during flower opening. Beetles were also seen walking along the lip and inside the flowers. Their presence did not change the flower structure or fertilize them. They are therefore not pollinators of the species.



Figure 03- Morphological and phenological characteristics of the identified species *Vanilla chamissonis* Klotzsch. A- Flower frontal; B- Petal Lateral; C- Sagittal section of the flower; D- Sepal Lateral.

Figure 04- Morphological and phenological characteristics of the identified species *Vanilla phaeantha* Rchb.f. A- Petal Lateral; B- Adaxial side, Column; C- Sagittal section of the flower; D- Abaxial side, Lip.

Conclusions and Outlook

The occurrence of wild *Vanilla* relatives in Southern Bahia was determined and registered. Genetic resources were successfully collected and relevant information on the species' phenological and morphological description in that environment was obtained. The support of the Tibá indigenous village community was extremely important in identifying the species. Studies on *Vanilla* species are few and often differ for the same species, making it difficult to recognize and possibly confirm the specific epithet. The presence of vanilla wild relatives in the region is an opportunity for its sustainable use. The fragmentation of forests is a concern for the conservation of the species demanding strategies of appropriate management.

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