



Phenodynamics of five orchids species growing on rock outcrops in the Chapada Diamantina Mountains in northeastern Brazil

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ABSTRACT

We evaluated the vegetative and reproductive phenological patterns of the orchids *Cattleya elongata*, *Cyrtopodium aliciae*, *Epidendrum orchidiflorum*, *Epistephium lucidum*, and *Sobralia liliastrum* (Orchidaceae) growing on sandstone outcrops in the Chapada Diamantina Mountains in northeastern Brazil. Phenological events were associated with abiotic factors, leaf longevity, life form, plant growth pattern, and pollination syndrome. Phenological observations were made for 18 months and followed the phenophases of: stem/pseudobulb emission, leaf flush, leaf fall, flowering, immature fruit and mature fruit. Seasonality, synchrony, and correlations between phenophases and environmental variables were tested. The orchid species demonstrated aseasonal vegetative phenologies, except for pseudobulb emission. Reproductive events were seasonal, except for flowering in *E. orchidiflorum*. There was high flowering overlap between the species pairs *S. liliastrum* and *C. aliciae* (dry season) and *C. elongata* and *E. lucidum* (rainy season). Dispersal occurred during both the rainy and dry seasons. The vegetative phenophases exhibited low synchrony, and were related to certain morphofunctional characters (stems/pseudobulbs, CAM metabolism). The reproductive phenophases showed high synchrony consistent with the deceit pollination strategy usually associated with the group.

Keywords: *campo rupestre*, flowering overlap, photoperiod, seasonality, synchrony

Seasonality and synchrony in plant phenology are traditionally associated with environmental variables, although variations in leaf and reproductive rhythms among species occupying similar environments indicate that phenological responses may also be mediated by

intrinsic species attributes such as life form, plant growth pattern, leaf longevity, and dispersal and pollination modes (Williams-Linera & Meave 2002). The Chapada Diamantina Mountains, the northern extension of the Espinhaço range in Brazil, exhibits a vegetation mosaic with *campo rupestre*

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(open, rocky field vegetation) predominating above 900 m a.s.l. (Funch *et al.* 2009). Orchidaceae is one of the principal groups found in that habitat (Conceição *et al.* 2007a), with approximately 300 species (Brito & Cribb 2005). This diversity is associated with morphofunctional characters such as CAM metabolism and the presence of pseudobulbs that retain water resources for metabolic activities and facilitate survival in otherwise unfavorable environments (Silveira *et al.* 2009; Arévalo *et al.* 2011). There has been a notable lack of studies addressing the phenologies of the Orchidaceae family in Brazil, and the available information is generally restricted to records of their flowering periods and floral biology (Pansarin *et al.* 2012; Pansarin & Pansarin 2014). Here, we evaluated the vegetative and reproductive phenological patterns of five Orchidaceae species (*Cattleya elongata* Barb. Rodr., *Cyrtopodium aliciae* L. Linden & Rolfe, *Epidendrum orchidiflorum* Salzm. Ex Lindl., *Epistephium lucidum* Cogn., and *Sobralia liliastrum* Salzm. Ex Lindl.) in an area of *campo rupestre* vegetation (12°34'28''S x 41°24'31''W; at 614 m a.s.l.) in the Chapada Diamantina Mountains, where those species grow abundantly on rocky outcrops. Considering the regional seasonality, with marked rainy and dry periods (Funch *et al.* 2002), the relatively low availability of water in rocky mountain top microhabitats (Conceição *et al.* 2007b), and well-known aspects of the structure and function of orchids that allow them to exploit seasonally dry habitats (Benzing *et al.* 1982), we hypothesized that their vegetative phenology would exhibit pseudobulb/stem emissions and leaf flush during the rainy season, associated with life form, plant growth pattern, and leaf longevity; as their reproductive dynamics would be aggregate adjusted to the seasonality, and mode of pollination and wind dispersal of their seeds (Borba *et al.* 2003; Smidt *et al.* 2006; Spaethe *et al.* 2007; Vale *et al.* 2011).

The regional climate is type Aw by the Köppen's climate classification (Alvares *et al.* 2014), marked by maximum rainfall in the Austral summer (November to April), and a dry winter (July to October). The temperature, rainfall and day lengths for the region were published by Souza *et al.* (2012). *Campo rupestre* is a complex vegetation type comprising numerous microhabitats on rocky mountain tops with water and soil nutrients restrictions and soil depths that severely limit plant establishment and growth (Conceição *et al.* 2007). The specimens of Orchidaceae examined here (n=89) were growing directly on rock surfaces (*C. elongata* n=24, *C. aliciae* n=17, *E. orchidiflorum* n=18, and *S. liliastrum* n=21) and in narrow cleft microhabitats with shallow soils contained by rock walls (*E. lucidum* n=9).

Phenological observations were made on a monthly basis from June/2009 to November/2010 on marked individuals. Individuals were considered to be distinct plants when separated from one another by approximately 1 m. The phenophases considered were: pseudobulb/stem emission, leaf flush, leaf fall, flowering, immature fruit and mature fruit. The phenophases were recorded on a

presence-absence basis. Four individuals of each species were closely accompanied during the phenological observations to determine leaf longevity by sketching them on paper and thus recording and numbering existing leaves and all new leaves. We also analyzed life forms (Raunkiaer 1934), plant growth patterns, pollination syndromes (Faegri & Pijl 1979), and floral and inflorescence characteristics (such as color, size, the presence of nectar or oil) by observations in the field or by consulting the literature (Chingel 2001). The Shapiro and Wilk test was used to determine the normality of the distributions of the phenological and environmental data (Zar 1996). The Spearman correlation index was calculated for data that did not have normal distributions, and the Pearson correlation index was calculated for the data that showed normal distributions, using Past 2.1.7 software. The seasonality and synchrony of the phenophases in each species and the overlap in flowering species pairs were evaluated using circular statistics (Morellato *et al.* 2010), run on Oriana software.

Four Orchidaceae species were classified as hemicyptophytes, having rhizomes flush to the ground/rock surface; *E. lucidum* was classified as a cryptophyte, as its rhizomes grow below the soil surface in narrow rock-cleft microhabitats. The orchid species all showed sympodial growth pattern, with pseudobulbs (*C. elongata* and *C. aliciae*) or stems (*E. orchidiflorum*, *S. liliastrum*, and *E. lucidum*) united by rhizomes; stems were numerous in *E. orchidiflorum*, *S. liliastrum*, while *E. lucidum* produced only a single stem. Plants with stems showed numerous leaves along the stem, with continuous leaf flush and leaf fall. Plants with pseudobulbs produced just a few apical leaves; the pseudobulbs and leaves of *C. aliciae* persisted for up to 12 months; the leaves of *C. elongata* remained attached to the older pseudobulbs for periods exceeding 18 months. The orchids examined demonstrated aseasonal and asynchronous vegetative phenology during the study period, except for pseudobulb emission in *C. elongata* and *C. aliciae* and leaf fall in *C. aliciae*, which were seasonal and synchronous (Fig. 1, Tab. 1).

Reproductive events were markedly seasonal and synchronous (Tab. 1). There was high flowering overlap in certain species pairs, with *S. liliastrum* and *C. aliciae* flowering during the dry season ($r = 0.83$), and *C. elongata* and *E. lucidum* flowering during the rainy season ($r = 0.87$); the only species that demonstrated aseasonal flowering was *E. orchidiflorum* (Fig. 1). *S. liliastrum* and *E. orchidiflorum* produced fruits during the dry season that matured during the rainy season; *C. elongata*, *C. aliciae*, and *E. lucidum* initiated fruit production during the rainy season, with maturation occurring during the dry season (Fig. 1). In general, the species demonstrated correlation of their flowering and fruiting with photoperiod and temperature (Tab. 2). Melittophily predominated, except in *E. orchidiflorum*, which was identified as sphingophilous. The species produced apical racemes, with up to 15 flowers,



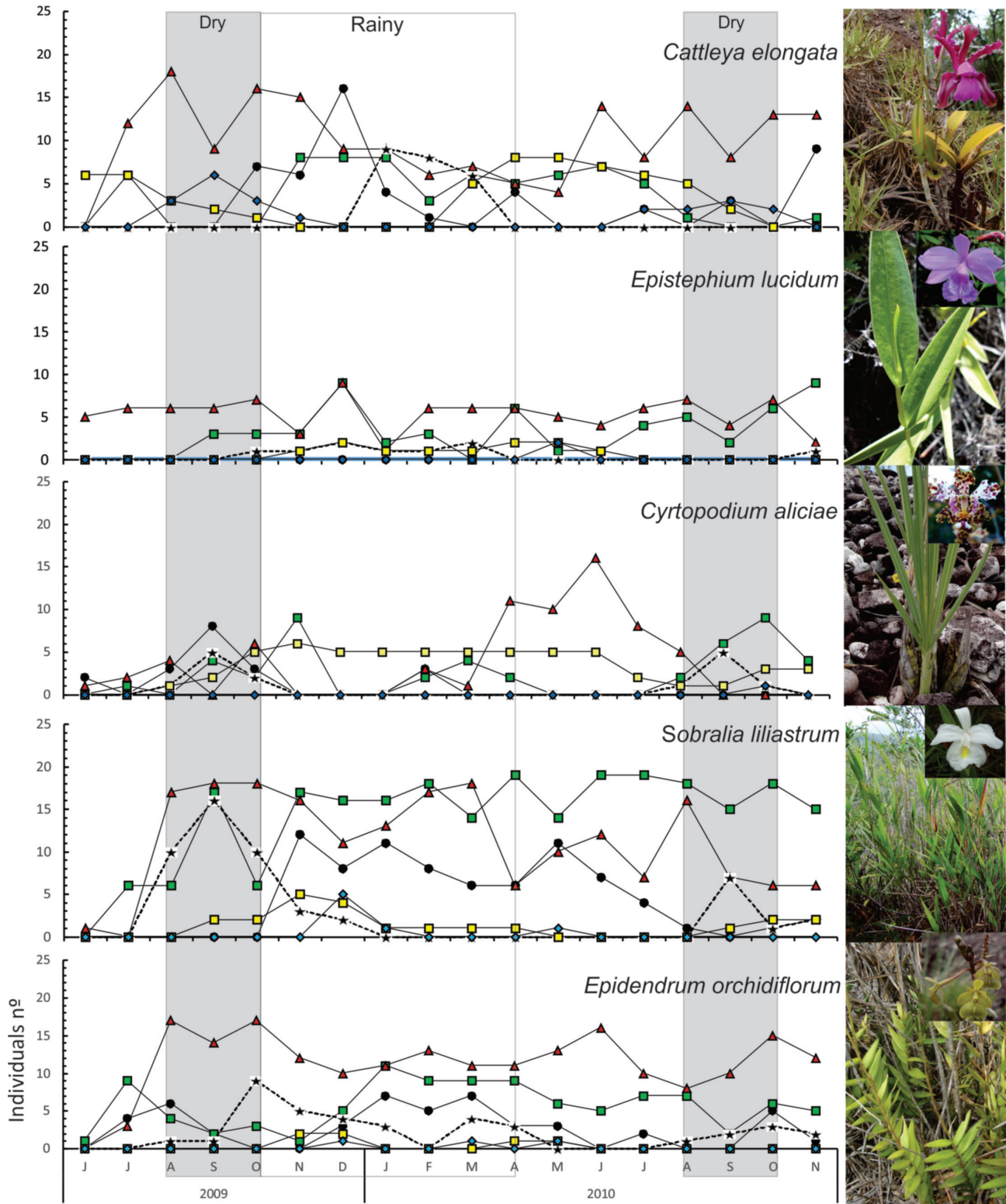


Figure 1. Habit, flower and phenology of orchids species from Chapada Diamantina, Northeast Brazil. The shaded area indicates the rainy season. Continuous lines with black circles are pseudobulbo; continuous lines with green square are leaf flushing; continuous lines with red triangles are leaf fall; dotted lines with stars are flower continuous lines with yellow square are immature fruit; continuous lines with blue rhombus are mature fruit.

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Table 1. Circular statistical analysis of the frequency of seasonal phenological phases (vegetative and reproductive) of five species of Orchidaceae, Chapada Diamantina, Northeast, Bahia, Brazil. E. pseudobulb = emission pseudobulb; E. stem = emission stem; L. flush = leaf flush; L. fall = leaf fall.

Especies	Phenophases	N° Obs.	Vector mean (μ)	Date mean	Length vector r	Standard deviation	Test de Rayleigh (p)
<i>Cattleya elongata</i>	E. pseudobulb	39	339.839°	09/dec.	0.626	55.485°	6.31e ⁻⁰⁸
	L. flush	50	82.911°	24/mar.	0.328	85.543°	0.005
	L. fall	110	265.985°	25/sep.	0.177	106.69°	0.032
	Flowering	23	45°	15/feb.	0.925	22.548°	< 7.31e ⁻⁰⁹
	Immature fruit	41	150.56°	01/jun.	0.653	52.916°	0.50e ⁻⁰⁹
	Mature fruit	9	241.936°	01/sep.	0.861	31.324°	2.21e ⁻⁰⁴
<i>Cyrtopodium aliciae</i>	EP	16	237.626°	28/aug.	0.578	74.765°	0.003
	L. flush	29	289.687°	20/oct.	0.427	74.765°	0.004
	L. fall	50	150.579°	01/jun.	0.737	44.794°	< 1e ⁻¹²
	Flowering	7	255°	15/sep.	0.973	13.457°	< 1e ⁻¹²
	Immature fruit	45	65.935°	06/mar.	0.266	93.214°	0.04
	Mature fruit	**	*****	*****	****	*****	*****
<i>Epidendrum orchidiflorum</i>	E. pseudobulb	36	40.216°	10/feb.	0.448	72.598°	5.4e ⁻⁰⁴
	L. flush	82	70086°	11/mar.	0.177	106.552°	0.00054
	L. fall	140	102.626°	13/apr.	0.023	157.041°	0.926
	Flowering	22	356.787°	26/dec.	0.368	81.041°	0.049
	Immature fruit	4	50.104°	20/feb.	0.381	79.615°	0.591
	Mature fruit	3	69.896°	10/mar.	0.508	66.706°	0.503
<i>Epistephium lucidum</i>	E. stem	**	*****	*****	*****	*****	*****
	L. flush	48	309.399°	09/nov.	0.316	86.958°	0.008
	L. fall	63	185.789°	06/jul.	0.033	149.58°	0.933
	Flowering	8	19.107°	20/jan.	0.739	44.604°	0.016
	Immature fruit	8	99.896°	10/apr.	0.711	47.358°	0.013
	Mature fruit	4	60°	01/mar.	0.262	93.803°	0.8
<i>Sobralia liliastrum</i>	E. stem	65	74.371°	16/mar.	0.38	79.747°	0.855e ⁻⁰⁴
	L. flush	201	187.189°	08/jul.	0.028	152.887°	0.85
	L. fall	129	70.822°	11/mar.	0.133	115.175°	0.103
	Flowering	12	280.517°	10/oct.	0.828	35.189°	3.1e ⁻⁰⁵
	Immature fruit	13	341.235°	11/dec.	0.592	58.624	0.008
	Mature fruit	7	356.31°	26/dec.	0.737	44.78°	0.016

Table 2. Correlation between phenophases and environmental variables of five species of Orchidaceae, Chapada Diamantina, Bahia, Northeast, Brazil.

Species	Phenophases	Precipitation	Temperature	Photoperiod
<i>Cattleya elongata</i>	Flowering	-	-	-0.87
	Immature Fruit	-0.47	-0.47	
<i>Cyrtopodium aliciae</i>	Immature Fruit	-	0.71	0.55
<i>Epidendrum orchidiflorum</i>	Flowering	-	0.39	0.73
<i>Epistephium lucidum</i>	Flowering	-	0.66	0.79
<i>Sobralia lucidum</i>	Immature Fruit	-	-	0.84

except *C. aliciae* which produces panicles with up to 300 flowers; *C. elongata*, *E. lucidum*, and *S. liliastrum* produce large flowers (5-8 cm diam.) and *C. aliciae* and *E. orchidiflorum* produce small flowers (1.5-3.5 cm diam.). *C. elongata* and *E. lucidum* produced white flowers while *C. aliciae* and *S. liliastrum* produce rose-colored blooms, and *E. orchidiflorum* greenish flowers (Fig. 1). Only *E. orchidiflorum* and *E. lucidum* offered nectar rewards to pollinators.

Precipitation has been recognized as the principal factor influencing plant phenological patterns in tropical

environments (Brearley *et al.* 2007). The absence of any correlations between the phenophases and precipitation among the orchid species evaluated in the present study was probably related to the fact that they all use water resources very efficiently and demonstrate CAM metabolism (Silveira *et al.* 2009). The pseudobulbs/stems store water for metabolic activities and facilitate their survival in otherwise unfavorable environments (Silva & Milaneze-Gutierrez 2004); the seasonality and synchrony observed for pseudobulb emission with few apical leaves can be understood in terms



of their construction costs, in contrast to aseasonal stem emissions with continuous leaf production and leaf fall.

The species studied demonstrated reproductive phenophases correlated with seasonal changes in photoperiod and temperature – the most predictable environmental parameters in this region. Photoperiodicity has been found to be associated with breaking and/or induction of bud dormancy, making it the trigger for growth and flowering initiation in many plant species (Stevenson *et al.* 2008). The high observed overlap of flowering between pairs of species with similar flower colors during the dry and rainy seasons (including *E. lucidum* which produces nectar) appears to be a viable reproductive strategy as most orchids mimic melittophilous flowers and, as they offer no floral rewards, are pollinated using deceit strategies (Pansarin & Pansarin 2014). The flowers of species utilizing deceit strategies will be visited less frequently than the flowers of species that offer some type of reward (as insects have the capacity to learn) - and will consequently show less fruiting (Borba & Semir 2001); as such, the high synchrony and overlapping between them presumably favors pollination processes by accentuating floral displays. *E. orchidiflorum* was the only species showing aseasonal flowering, which appears to reflect nectar production by their flowers (with characteristics of sphingophily).

A low fruiting rate is considered an adaptation among orchids, especially among epiphytic and rupicolous species, with limited access to resources; their fruits generally result from cross-pollination events and their seeds show high genetic viability – so that the production of even small numbers of fruits will allow the maintenance of viable populations (Ackerman & Zimmerman 1994). The maturation of dry fruits is favored by dry periods with severe water restrictions, and the low relative humidity of the air contributes to their desiccation and to the dispersal of anemochoric seeds (Pezzini *et al.* 2008). As Orchidaceae seeds are very small and essentially without nutritional reserves their dispersal during dry periods might be expected to hinder germination and establishment, but they demonstrate dormancy due to the presence of lipidic reserves on their surfaces (which will also provide for their initial sustenance) (Baskin & Baskin 2001), allowing dispersal in both the dry and rainy seasons.

The low intraspecific synchrony and aseasonality demonstrated by the vegetative phenophases of the orchid species examined in the present study appeared to be a reflection of the influences of the morphofunctional characters (pseudobulbs/stems and CAM metabolism), and the high synchrony and seasonality of their reproductive events is likewise related to the pollination mechanisms of the group, with overlapping of flowering among species with similar flower colors and the production of nectar by at least one of them.

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