Trees (2016) 30:1331–1339
DOI 10.1007/s00468-016-1369-6

ORIGINAL ARTICLE

Cambial dormancy lasts 9 months in a tropical evergreen species

Natália Oliveira Totti de Lara[1] · **Carmen Regina Marcati**[2]

Received: 13 September 2015 / Accepted: 15 February 2016 / Published online: 1 March 2016

Abstract

Key message **Cambial dormancy in a tropical evergreen species is long lasting, and the initiation of cambial activity is related to day length.**

Abstract Studies on cambial activity allow us to understand the growth dynamics of plants. In this study, we investigate cambial activity and the conducting phloem in a population of an evergreen shrubby species from a semi-deciduous tropical forest with distinct wet and dry seasons. Stem samples from *Cordiera concolor* were collected bimonthly in 2010 and prepared according to the standard methods for plant anatomy. The relationship between cambial activity and climate factors as well as phenology was investigated using Spearman's correlation. The cambium is dormant during the rainy season and dormancy lasted up to 9 months. Cambial activity was positively related to day length, and although it occurred in the rainy season, the period of its onset and termination was not concurrent with the beginning and end of the rainy season. The conducting phloem corresponded most to the non-collapsed phloem and was present year-round, ranging in width (in transverse section) from 62 µm in the rainy season to 112 µm in the dry season. Together, these results illustrate that cambial activity in an evergreen species of a seasonal tropical forest is seasonal with long-lasting cambial dormancy, and that at least some conducting phloem is present year-round.

Keywords Cambial activity · Cambial dormancy · *Cordiera concolor* · Evergreen species · Secondary phloem · Tropical species

Introduction

Studies on cambial activity allow us to understand the dynamics of forest growth (Jacoby 1989; Worbes 1995), and how this growth is influenced by environmental factors such as temperature, rainfall, and day length (Begum et al. 2013). The activity of the vascular cambium in tropical species has been studied over the years; however, cambial responses in relation to environmental conditions still remain unexplored for most of these species.

Rainfall seasonality has been considered a determinant for cambial activity in tropical species (Dave and Rao 1982a; Rajput and Rao 2000, 2001a, b; Marcati et al. 2006; Venugopal and Liangkuwang 2007; Pumijumnong and Buajan 2012). Worbes (1995) stated that an annual dry season with a length of 2–3 months with less than 60 mm monthly precipitation induces cambial dormancy. Although the inhibition of plant growth by water stress is well recognized, responses to seasonal drought vary widely among individuals, and their annual development is not well synchronized with climatic seasonality (Borchert 1999).

Longer periods of cambial activity have been reported for evergreen species, with an active cambium throughout the year (Aljaro et al. 1972; Dave and Rao 1982b; Rajput and Rao 2002) or during most of the year (Paliwal and Paliwal 1990; Rajput and Rao 2000, 2001). Here, we study

Communicated by Y. Sano.

✉ Natália Oliveira Totti de Lara
natotti@gmail.com

[1] Programa de Pós-Graduação em Ciências Biológicas (Botânica), Instituto de Biociências de Botucatu, Univ Estadual Paulista (UNESP), 18618-970 Botucatu, SP, Brazil

[2] Dep. Ciência Florestal, Faculdade de Ciências Agronômicas de Botucatu, Univ Estadual Paulista (UNESP), 18610-307 Botucatu, SP, Brazil

the cambial activity of an evergreen species in a semi-deciduous seasonal forest raising the question which factors affect radial growth.

During radial growth, secondary phloem is formed, and its conducting region translocates the products of photosynthesis and regulates carbon stocks in the plant (Taiz and Zeiger 2004). The yearly increment of the conducting phloem produced in one season varies with species and growth conditions (Evert 2006), and indicates plant functionality.

In this study, we address several of the points mentioned above for an evergreen species in a seasonal tropical forest: (1) Are the onset and termination of cambial activity related to beginning and end of the rainy season? (2) How long does cambial dormancy last? (3) Which phenological events and climatic variables are related to cambial activity? (4) Is the conducting phloem present year-round? (5) Which time of year does the larger and narrower conducting phloem occur?

Materials and methods

Study area

The study was carried out in a fragment of semi-deciduous seasonal forest (Fig. 1a) in a reserve area of the Botanical Garden of the Univ. Estadual Paulista (UNESP) (22°53′152″S e 48°29′938″W) in Botucatu, state of São Paulo, Brazil. The soil is acidic and sandy (Lara 2012).

The average annual rainfall is about 1300 mm, with a mean annual temperature of 20 °C. July is the driest and coldest month with a mean temperature of 15 °C, while January is the wettest and warmest month with a mean temperature of 25 °C. The dry season typically lasts from May to September.

The species and sampling

We studied all specimens of the evergreen shrub *Cordiera concolor* (Cham.) Kuntze (Rubiaceae) (Fig. 1b) available in the reserve area (population size $n = 5$). *Cordiera concolor* was chosen for this study because it is an evergreen species and it has been mentioned in the literature to have growth markers in its wood (marginal bands of axial parenchyma) (see Jansen et al. 2002; Sonsin et al. 2014) which indicate seasonal activity of cambium.

Vouchers were deposited at the Herbarium "Irina Delanova de Gemtchujinicov" (BOTU) under the register numbers: Lara, N. O. T. BOTU 027342 until 027346. The wood samples were deposited in the Wood Collection "Profa. Dra. Maria Aparecida Mourão Brasil" (BOTUw), under the register numbers: BOTUw 2224 until 2228.

We collected the most developed branches from the same side of the crown every other month during 2010 (Feb 09; Apr 10; Jun 10; Aug 12; Oct 12; Dec 09). Each collection was made from the same individuals. We collected branches because (1) the species is short with a height between 3 and 5 m, (2) the specimens are ramified from the base and the stem diameter at breast height ranges

Fig. 1 View of the study area, a fragment of a semi-deciduous seasonal forest located in a Botanical Garden of the Univ. Estadual Paulista (UNESP) at Botucatu, state of São Paulo, Brazil (**a**), and one of the specimens studied of *Cordiera concolor* (**b**). *Arrows* indicate the stems of the shrub

between 16 and 20 cm, and (3) of restrictions on the use of destructive methods in an environmental reserve area.

Light microscopy of cambial zone and secondary phloem

Stem portions with approximately 2 cm diameter and 6 cm length were collected above the basal part, using a hacksaw. These portions were immediately fixed in CRAF III (Sass 1958), where they remained for about half a month, and thereafter stored in 70 % ethanol. The entire circumference of the branches were cut using a sliding microtome. Small samples (1 cm^3) containing the cambial zone and secondary tissues, chosen from the entire circumference, were trimmed and dehydrated in ethanolic sequence, embedded in plastic resin according to Bennett et al. (1976), and transversally and longitudinally sectioned using a semi-automated rotary microtome at 5 µm. The sections were stained with toluidine blue (TB), in acetate buffer, pH 4.7, producing a metachromatic stain (O'Brien et al. 1964). Important results were photo-documented under a light microscope equipped with a digital camera.

Transmission electron microscopy (TEM)

To illustrate the active cambial zone, samples collected in February and December were fixed in 2.5 % glutaraldehyde (0.1 M sodium phosphate buffer, at pH 7.3, for 24 h at 5 °C), and postfixed with 1 % osmium tetroxide (OsO_4) in the same buffer for 2 h at room temperature. After washing in distilled water, the material was dehydrated in an acetone series and embedded in Araldite resin. Ultrathin sections were cut with an ultramicrotome and contrasted with uranyl acetate and lead citrate (Reynolds 1963) and then examined with a Tecnai Spirit TEM (FEI) at 80 kV.

Phenological observations and climate data

Phenology status (0 = absent, 1 = present) followed Morellato et al. (2000), but modified: (1) swollen buds characterized by expanded buds before leaf primordia; (2) budding, the appearance of the light-green colored leaf primordia; (3) new green leaves characterized by expanding leaf blades; (4) mature leaves characterized by fully expanded leaves; (5) leaf abscission, when the leaf drops easily from the branch or we observed the presence of leaf scars at the apex of the branch; (6) flower buds, reproductive bud before flowering; (7) flowering characterized by opened flowers; (8) fruiting, green or mature. The phenophases were monitored at 2-week intervals during the vegetative and reproductive periods in 2010.

Climate data (temperature and precipitation) were obtained from the Estação Meteorológica of the Faculdade de Ciências Agronômicas, UNESP, Botucatu, São Paulo, which is located about 7 km from the study site. For standardization, we calculated temperature and day length means for the 30-day period that preceded the collection date, as well as total precipitation for the same period. Day length data were obtained from Time and Date (2014).

Statistical analyses

Since the cambial activity data did not present normal distribution (Shapiro–Wilk normality test), the Spearman rank correlation coefficients were calculated to check whether the cambial activity for each specimen was related to climate factors, and plant phenology. We rated the significant ($P < 0.05$) correlation coefficients as suggested by Cann (2002): values up to 0.33 are considered to indicate weak relationships; between 0.34 and 0.66 indicate medium strength relationships, and over 0.67 indicate strong relationships.

Terminology

We used the term "cambial zone" to describe the radial rows of rectangular fusiform cells and ray initials, which are initial cells plus their derivatives between differentiating/differentiated xylem and phloem cells. Cambial activity was identified by the presence of very thin, recently formed periclinal cell walls, and mitotic figures. Cambial dormancy was identified by the absence of very thin, recently formed periclinal cell walls, and mitotic figures, and also by thickness of radial walls which have a beaded appearance of fusiform initial and mother cells as viewed in tangential sections (Evert 2006). The cambial activity (1) or cambial dormancy (0) were evaluated by each specimen. Judgment of active/dormant cambium was made based on the entire circumference of the basal part of each sampled branch per specimen. This analysis was possible because we cut cross sections of the entire circumference of the branches using a sliding microtome as we already mentioned in the item light microscopy of cambial zone and secondary phloem.

The conducting phloem was characterized by open sieve-area pores of mature sieve elements, while the non-conducting phloem was covered by a callose mass (definitive callose) as defined by Evert (2006).

Results

Phenology and climate data

Figure 2 summarizes phenological events and climate data. Although the species is evergreen, it replaced leaves

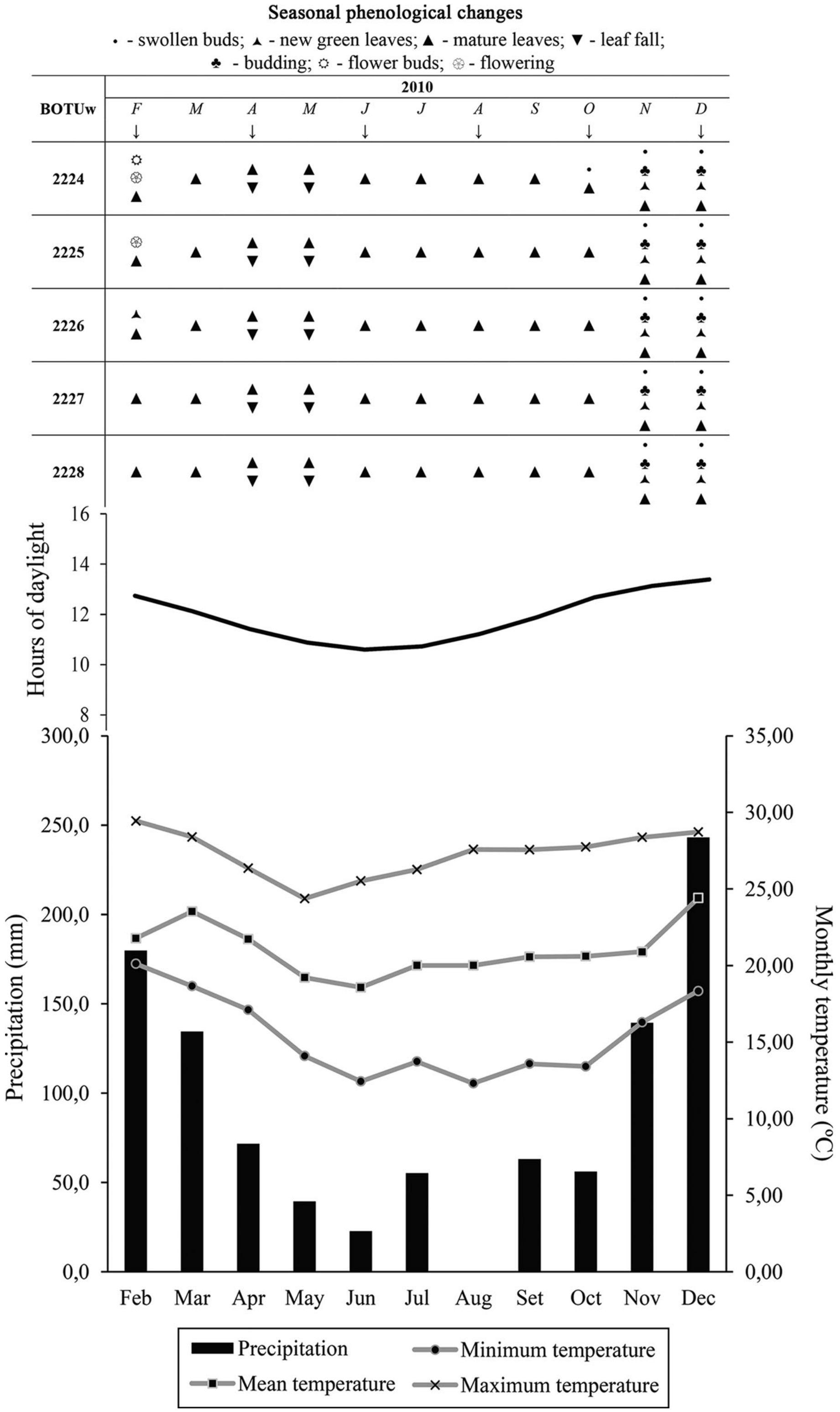

Fig. 2 Seasonal phenological changes of specimens, and day length, temperature and precipitation of the study area

seasonally. We observed swollen buds, budding and sprouting of new leaves in November (139 mm of rain), and to a lesser extent in December (243 mm of rain, 13 h of daylight), when the plants already had mature leaves.

The first rain occurred in September (63 mm) when the plants had mature leaves. From May (39.5 mm, 10 h of daylight) to August (0 mm, 11 h of daylight) plants had mature leaves and leaves were shed.

Flowering buds and flowers were present in February, the warmest month (29.4 °C) with 179.9 mm of rain, while August was the coldest (12.3 °C) with no rainfall recorded.

Characteristics of the cambial zone

Two or three active or dormant initial cells arranged radially in the cambial zone (Fig. 3a, b) were observed during all studied periods. Fusiform initial cells were not arranged in horizontal tiers as the cambial zone is non-storied.

Seasonal fluctuation of cambial activity

Initiation of cambial activity was noted in December when very thin, recently formed, periclinal cell walls were observed in the cambial zone (Fig. 3a, b). December was a rainy and warm month with 13.3 h of daylight (Fig. 2), when the plants had swollen buds and new leaves, and budding was observed (Fig. 2).

A dormant cambial zone (Fig. 3c, d) was observed in April, June, August and October in all specimens. A beaded appearance of radial walls in cambial cells (Fig. 3d) was observed in tangential sections. Cambial activity ended in February.

We found a strong positive correlation between cambial activity and day length (Table 1).

Secondary phloem

In *Cordiera concolor*, the secondary phloem is composed of tangential bands of fibers alternating with zones containing sieve elements, companion cells and parenchyma cells (Fig. 4a). One companion cell might be associated with more than one sieve-tube element (Fig. 4b).

The conducting phloem contains sieve-tube elements with sieve plate pores opened (Fig. 4c).

In the nonconducting phloem, the sieve plate pores are covered by definitive callose (Fig. 4d). Thus, the sieve elements in this region, although noncollapsed, are nonconducting (Fig. 4d). As the sieve-tube elements and their companion cells collapse, the parenchyma cells expand (Fig. 4a).

The conducting phloem varied between periods (Table 2). In June we observed a larger amount of conducting phloem (Fig. 5a) and in December a smaller amount (Fig. 5b).

Discussion

In this study, we found that *Cordiera concolor*, an evergreen shrub from semi-deciduous seasonal tropical forest, has a long period of cambial dormancy, lasting up to

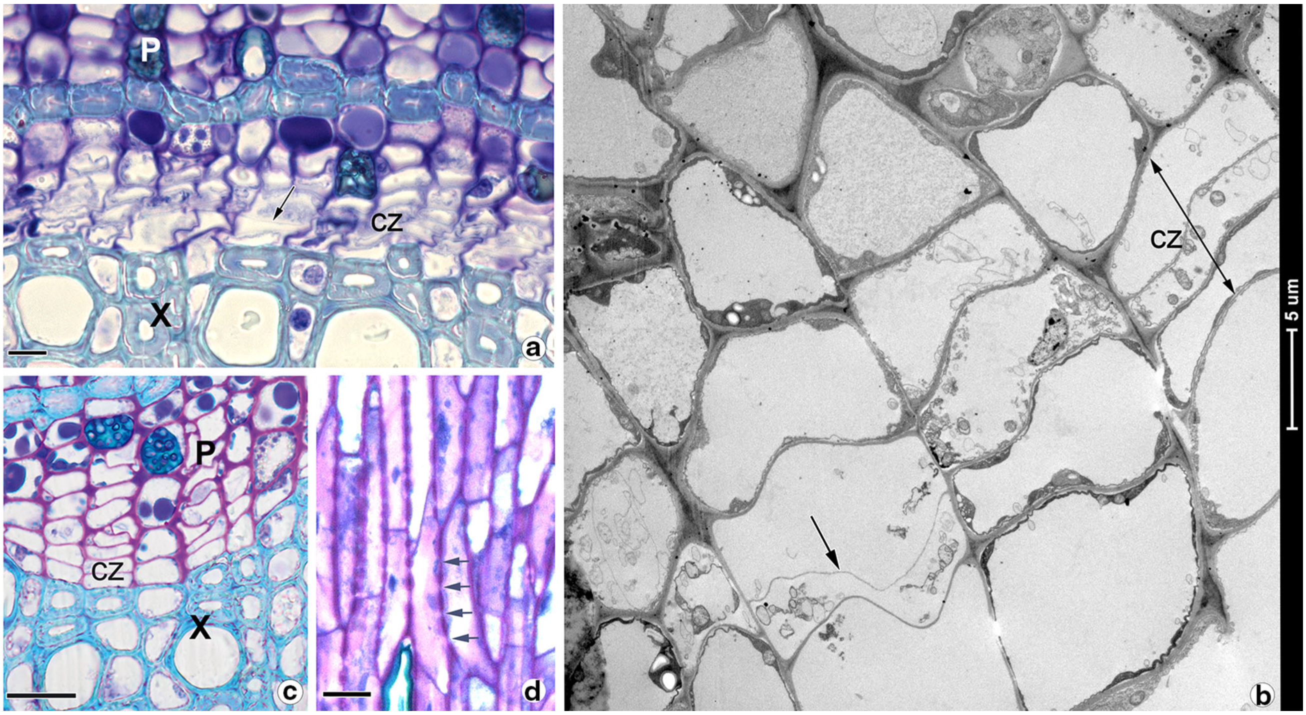

Fig. 3 Seasonal cambial activity of *Cordiera concolor*. **a**, **b** Onset of cambial activity in December. Thin, recently formed, periclinal cell walls (*arrows*) in the cambial zone in light microscopy (**a**) and TEM (**b**). **c**, **d** Dormant cambial zone with thick cell walls in October. **c** Transverse section. **d** Tangential section showing beaded appearance of the radial walls due to the presence of deeply depressed primary pit-fields (*arrows*). *Cz* cambial zone. *Scale bars* **a**, **d** 20 µm. **c** 50 µm

Table 1 Spearman rank correlation coefficients between cambial activity and climate factors (temperature, precipitation and day length), and phenology (swollen buds, budding, new green leaves, mature leaves, leaf fall, flower buds, flowering) for *Cordiera concolor*

	Spearman's correlation coefficients
Climatic conditions	
Mean temperature (°C)	0.777
Minimum temperature (°C)	0.777
Maximum temperature (°C)	0.777
Precipitation (mm)	0.777
Day length (h)	0.845*
Phenological events	
Swollen buds	0.500
Budding	0.774
New green leaves	0.602
Mature leaves	–
Leaf fall	–
Flower buds	–
Flowering	0.464

* $P < 0.05$

9 months. In contrast, no cambial dormancy (Dave and Rao 1982b; Rajput and Rao 2001) or almost no cambial dormancy (Aljaro et al. 1972; Avila et al. 1975) has been reported for several evergreen species growing in seasonal dry regions. In other species, cambial dormancy can last from 5 to 7 months (Aljaro et al. 1972; Ghouse and Hashmi 1983; Rajput and Rao 2001; Yañez-Espinosa et al. 2010), reflecting large interspecific differences for cambial dormancy periods. For *C. concolor,* the observed long cambial dormancy may be related to the species being shrubby, rarely higher than 5 m, and growing in acid sandy soil with low water retention capacity.

Cambial activity of *C. concolor* occurred after budding. Leaf replacement is linked to cambial activity, mainly in seasonal environments (Ghouse and Hashmi 1983; Rajput and Rao 2000; Yañez-Espinosa et al. 2010). The sprouting of new leaves plays an important role in the initiation of cambial activity due to the necessary synthesis of phytohormones (Little and Savidge 1987). In this study, we found that the reactivation of cambial activity in *C. concolor* began in December, one month after budding. This finding is in accordance with the idea that leaf emergence and young leaves are responsible for auxin supplies and stimulation of the cambial zone. In the studied case, after a long period of dormancy, the vascular cambium requires a stimulus that is provided by the auxin produced in young leaves, leading to an increased hormonal level and metabolic rates. An increase in the total amount of endogenous auxin is related to the initiation of cambial activity, which suggests that auxin is related to cambial growth (Funada et al. 2002). In addition, cambial divisions are correlated with auxin concentrations that correspond to the highest stimulation of cambial activity (Lachaud et al. 1999). Although an evergreen, *C. concolor* showed a conspicuous timing of budding and initiation of new leaves, which occurred during the warm season of the year (December), and was associated with the reactivation of cambial activity.

Apart from phenological events, cambial activity is usually related to the rainy season in seasonal tropical environments (Aljaro et al. 1972; Avila et al. 1975; Worbes 1995; Rajput and Rao 2000, 2001; Pumijumnong and Buajan 2012). In our study, onset and termination of cambial activity were not concurrent with the beginning and end of the rainy season. Worbes' statement that an annual dry season with a length of 2–3 months, with less than 60 mm monthly precipitation, induces cambial dormancy was not observed for *C. concolor*, since the cambium was dormant during the rainy season.

A strong positive correlation was found between cambial activity and day length for *C. concolor* in this study. In tropical species, a relationship between cambial activity and day length was reported by Yañez-Espinosa et al. (2006) for branches from five species in subtropical forests with seasonal droughts in Mexico. From an ecological perspective, seasonal day length variation determines the timing of bud break in species from Costa Rica (Rivera et al. 2002). According to the authors, an increase of 30 min or less in day length induced sprouting, and the critical day length for the induction of bud break ranges from 11.5 to 12.5 h in the subtropics. Under this assumption, the critical day length for cambial activity in *C. concolor* seems to be below 12.5 h of light.

The number of initial cells in the cambial zone did not fluctuate in our study: two to three initial cells were observed throughout the analyzed period. When we compared dormant and active periods, the number of initial cells did not provide information about cambial activity. This was also reported for *Cordia trichotoma* from Brazil (Amano 2002), and for ten species of Cerrado (C.R. Marcati, personal communication). Various studies on cambial growth (Gričar et al. 2006; Pumijumnong and Wanyaphet 2006; Rajput et al. 2008; Pumijumnong and Buajan 2012; Gričar 2013; Morel et al. 2015) described the beginning of xylem production by recording the number of cells in radial files in the cambial zone to determine cambial activity. However, cambial activity might be initiated before the already produced cells are visible. According to Esau (1977), cambial activity should be recognized by the presence of mitotic figures or very thin, recently formed periclinal cell walls. Also, the cambium might already be dormant while xylem cells are still undergoing

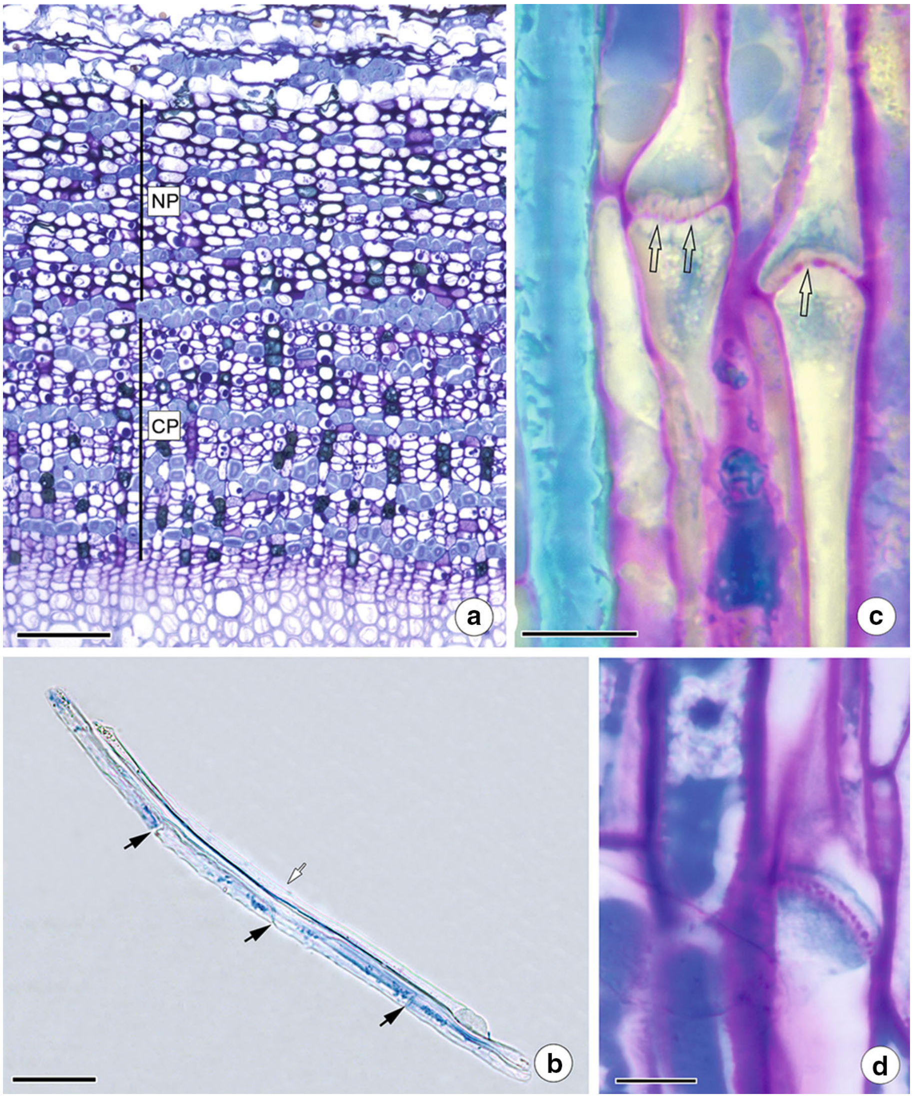

Fig. 4 Secondary phloem of *Cordiera concolor*. **a** Collapsed (CP) and noncollapsed (NP) phloem zones in June 2010. Bars indicate the width of each zone. **b** One companion cell (*white arrow*) associated with more than one sieve-tube element (*black arrows* point to sieve plates) in dissociated cells. **c** Sieve plate pores opened (*arrows*) in conducting phloem in December 2010. Note the sieve plate pores surrounded by callose and P-protein near the sieve plates. **d** Definitive callose covering the sieve plate pores in nonconducting phloem in December 2010. *Scale bars* **a** 100 µm; **b** 50 µm; **c**, **d** 20 µm

Table 2 Width of the conducting phloem (mean values) for all periods in *Cordiera concolor*

Months	February	April	June	August	October	December
Conducting phloem in micrometers	78.7	101.8	112.0	95.6	69.1	61.7

differentiation. Thus, cambial activity might be overestimated in such studies.

In general, the number of radial rows of fusiform cells in the cambial zone varies among species. Comparing the number of the fusiform cells in the cambial zone of *C. concolor* in our study to other tropical species from different studies (Dave and Rao 1982a; Siddiqi 1991; Marcati et al. 2008; Begum et al. 2010; Prislan et al. 2011), we found that the cambial zone of *C. concolor* is narrow, possibly related to the shrubby nature of *C. concolor*.

The analysis of the secondary phloem in *C. concolor* indicated a reduction of the conducting phloem during the year. A narrower conducting phloem was observed in December with the onset of cambial activity. This was also observed for *Schizolobium parahyba* (Leguminosae), a deciduous legume tree, by Marcati et al. (2008). For *C. concolor*, the maximum width of the conducting phloem was observed in June when the cambium was dormant. Marcati et al. (2008) observed the same for *S. parahyba*. A larger conducting region may be an important factor for the

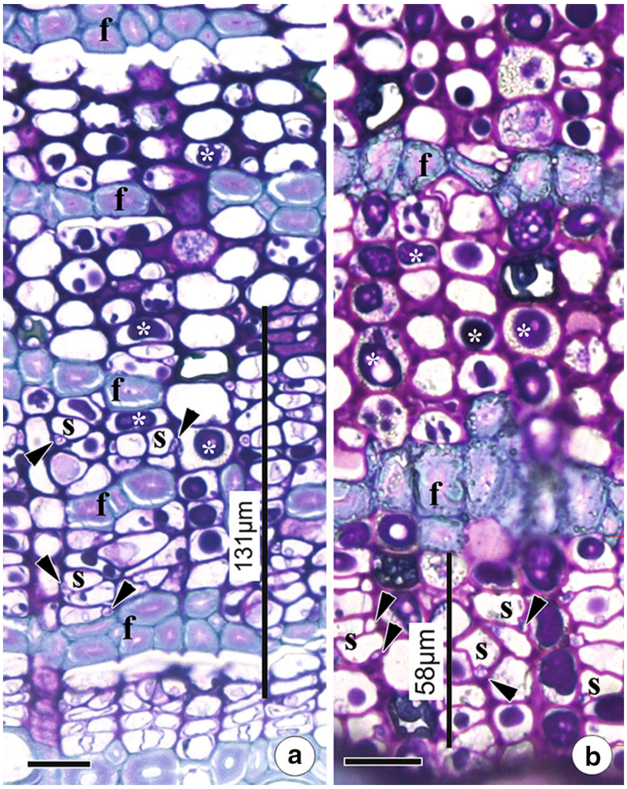

Fig. 5 Conducting phloem in cross sections in June (**a**), and December (**b**). *Bars* indicate the width of the conducting phloem. Some cell types were indicated: *f* fibers; *s* sieve tubes; *asterisks* parenchyma cells; *arrowhead* companion cell. *Scale bars* **a**–**b** 20 μm

efficiency of translocation of photoassimilates and maintenance of full foliage year-round for evergreen species.

In summary, we conclude that *Cordiera concolor* undergoes cambial seasonality with a long lasting cambial dormancy of 9 months. Cambial activity is related to day length and occurred after budding. A conducting phloem region is present year-round. A larger conducting region is observed when the cambium is dormant, while a narrower conducting region can be observed at the onset of cambial activity.

Author contribution statement N. O. T. Lara conducted the analyses and wrote the manuscript. N. O. T. Lara and C. R. Marcati designed the study and wrote the manuscript. C. R. Marcati supervised the research, helped in data interpretation and manuscript evaluation. Both authors approved the final version of this manuscript.

Acknowledgments This study was funded by the Fundação de Amparo à Pesquisa do Estado de São Paulo (No. 2009/17778-9). N. O. T. Lara received Grants from Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) for MSc thesis, and from Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP, No.2013/05700-0) for PhD thesis. The authors gratefully acknowledge the staff of the Electron Microscopy Center of UNESP for helping in the sample preparation; Clemente José Campos for helping in the field work; Diego Sotto Podadera for assistance in statistic analysis; Liliane Catarina Pereira for assistance in the laboratory; and the anonymous reviewers for invaluable comments on earlier versions of this manuscript.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

References

Aljaro ME, Avila G, Hoffmann A, Kummerow J (1972) Annual rhythm of cambial activity in woody species of Chilean matorral. Am J Bot 59:879–885

Amano E (2002) Sazonalidade da atividade cambial em *Cordia trichotoma* (Vell.) Arrab ex. Steud Boraginaceae. Departamento de Botânica, vol Tese. Universidade de São Paulo, São Paulo, p 87

Avila G, Aljaro ME, Araya S, Montenegro G, Kummerow J (1975) Seasonal cambium activity of Chilean and Californian shrubs. Am J Bot 62:473–478

Begum S, Nakaba S, Oribe Y, Kubo T, Funada R (2010) Cambial sensitivity to rising temperatures by natural condition and artificial heating from late winter to early spring in the evergreen conifer *Cryptomeria japonica*. Trees Struct Funct 24:43–52

Begum S, Nakaba S, Yamagishi Y, Oribe Y, Funada R (2013) Regulation of cambial activity in relation to environmental conditions: understanding the role of temperature in wood formation of trees. Physiol Plant 147:46–54

Bennett HS, Wyrick AD, Lee SW, Mcneil JH (1976) Science and art in preparing tissues embedded in plastic for light-microscopy, with special reference to glycol methacrylate, glass knives and simple stains. Stain Technol 51:71–97

Borchert R (1999) Climatic periodicity, phenology and cambial activity in tropical dry forest trees. IAWA J 20:239–247

Cann AJ (2002) Maths from scratch for Biologists. Wiley, Chichester

Dave YS, Rao KS (1982a) Seasonal activity of the vascular cambium in Gmelina arborea Roxb. IAWA Bull 3:59–65

Dave YS, Rao KS (1982b) Cambial activity in Mangifera indicaL. Acta Bot Hung 28:73–79

Esau K (1977) Anatomy of seed plants, New York

Evert RF (2006) Esau's Plant Anatomy: meristems, cells and tissues of the plant body—their structure, function and development. Wiley, New Jersey

Funada R, Kubo T, Sugiyama T, Fushitani M (2002) Changes in levels of endogenous plant hormones in cambial regions of stems of *Larix kaempferi* at the onset of cambial activity in springtime. J Wood Sci 48:75–80

Ghouse AKM, Hashmi S (1983) Periodicity of cambium and the formation of xylem and phloem in *Mimusops elengi* L, an evergreen member of tropical India. Flora 173:479–487

Gričar J (2013) Influence of temperature on cambial activity and cell differentiation in *Quercus sessiliflora* and *Acer pseudoplatanus* of different ages. Drvna Ind 64:95–105

Gričar J, Zupancic M, Cufar K, Koch G, Schmitt U, Oven P (2006) Effect of local heating and cooling on cambial activity and cell differentiation in the stem of Norway spruce (*Picea abies*). Ann Bot 97:943–951

Jacoby GC (1989) Overview of tree-ring analysis in tropical regions. IAWA Bull 10:99–108

Jansen S, Robbrecht E, Beeckman H, Smets E (2002) A survey of the systematic wood anatomy of the Rubiaceae. IAWA J 23:1–67

Lachaud S, Catesson AM, Bonnemain JL (1999) Structure and functions of the vascular cambium. Cr Acad Sci III-Vie 322:633–650

Lara NOT (2012) Sistema vascular e atividade cambial em *Alibertia concolor* (Cham). K. Schum (Rubiaceae) Botany, vol Master. Univ Estadual Paulista, Botucatu, p 139

Little CHA, Savidge RA (1987) The role of plant growth regulators in forest tree cambial growth. Plant Growth Regul 6:137–169

Marcati CR, Angyalossy V, Evert RF (2006) Seasonal variation in wood formation of *Cedrela fissilis* (Meliaceae). IAWA J 27:199–211

Marcati CR, Milanez CRD, Machado SR (2008) Seasonal development of secondary xylem and phloem in *Schizolobium parahyba* (Vell.) Blake (Leguminosae: caesalpinioideae). Trees 22:3–12

Morel H, Mangenet T, Beauchene J, Ruelle J, Nicolini E, Heuret P, Thibaut B (2015) Seasonal variations in phenological traits: leaf shedding and cambial activity in *Parkia nitida* Miq. and *Parkia velutina* Benoist (Fabaceae) in tropical rainforest. Trees 29:973–984

Morellato LPC, Talora DC, Takahasi A, Bencke CC, Romera EC, Zipparro VB (2000) Phenology of Atlantic rain forest trees: a comparative study. Biotropica 32:811–823

O'Brien TP, Feder N, Mccully ME (1964) Polychromatic staining of plant cell walls by toluidine blue O. Protoplasma 59:368–373

Paliwal SP, Paliwal GS (1990) Influence of climatic variations on the seasonal behaviour of the vascular cambium in some Himalayan trees. III. Rhododendron arboreum Smith. Phytomorphology 40:257–271

Prislan P, Schmitt U, Koch G, Gricar J, Cufar K (2011) Seasonal ultrastructural changes in the cambial zone of beech (*Fagus sylvatica*) grown at two different altitudes. IAWA J 32:443–459

Pumijumnong N, Buajan S (2012) Seasonal cambial activity of five tropical tree species in central Thailand. Trees 27:409–417

Pumijumnong N, Wanyaphet T (2006) Seasonal cambial activity and tree-ring formation of *Pinus merkusii* and *Pinus kesiya* in Northern Thailand in dependence on climate. For Ecol Manage 226:279–289

Rajput KS, Rao KS (2000) Cambial activity and development of wood in *Acacia nilotica* (L.) DEL. growing in different forests of Gujarat State. Flora 195:165–171

Rajput KS, Rao KS (2001) Cambial activity and development of xylem in *Tamarindus indica* L. growing in different forests of Gujarat State. Acta Botanica Hungarica 43:379–390

Rajput KS, Rao KS (2002) Cambial anatomy and annual rhythm of secondary xylem development in the twigs of *Azadirachta indica* A. Juss. (Meliaceae) growing in different forests of Gujarat state. J Sustain For 14:115–127

Rajput KS, Rao KS, Kim YS (2008) Cambial activity and wood anatomy in *Prosopis spicigera* (Mimosaceae) affected by combined air pollutants. IAWA J 29:209–219

Rao KS, Rajput KS (2001a) Relationship between seasonal cambial activity, development of xylem and phenology in *Azadirachta indica* growing in different forests of Gujarat State. Ann For Sci 58:691–698

Rao KS, Rajput KS (2001b) Xylem structure and annual rhythm of development in the twigs of *Acacia nilotica* (L.) DEL. growing in different forests of Gujarat state (India). Phyton-Ann Rei Bot A 41:1–12

Reynolds ES (1963) The use of lead citrate at high pH as an electron-opaque stain in electron microscopy. J Cell Biol 17:208–212

Rivera G, Elliott S, Caldas LS, Nicolossi G, Coradin VTR, Borchert R (2002) Increasing day-length induces spring flushing of tropical dry forest trees in the absence of rain. Trees 16:445–456

Sass JE (1958) Botanical microtechnique. Iowa State University Press, Iowa

Siddiqi TO (1991) Impact of seasonal variation on the structure and activity of vascular cambium in *Ficus religiosa*. IAWA Bull 12:177–185

Sonsin JO, Marcati CR, Gasson P, Machado SR, Caum C (2014) Atlas da diversidade de madeiras do cerrado paulista/Atlas of wood diversity of Cerrado of São Paulo. FEPAF, Botucatu

Taiz L, Zeiger E (2004) Fisiologia vegetal. Artmed Editora, Porto Alegre

Time and Date (2014) Time and Date. http://www.timeanddate.com/sun/brazil/sao-paulo. Retrieved in 24 Mar

Venugopal N, Liangkuwang MG (2007) Cambial activity and annual rhythm of xylem production of elephant apple tree (*Dillenia indica* Linn.) in relation to phenology and climatic factor growing in sub-tropical wet forest of northeast India. Trees 21:101–110

Worbes M (1995) How to measure growth dynamics in tropical trees—a review. IAWA J 16:337–351

Yañez-Espinosa L, Terrazas T, Lopez-Mata L (2006) Integrated analysis of tropical trees growth: a multivariate approach. Ann Bot 98:637–645

Yañez-Espinosa L, Terrazas T, Lopez-Mata L (2010) Phenology and radial stem growth periodicity in evergreen subtropical rainforest trees. IAWA J 31:293–307