Результаты статистической обработки данных представлены в таблице 1. Они дают возможность утверждать, что опыт достаточно точный, так как ошибка ни в одном случае не превышает 5 %. Наиболее низкую степень изменчивости имеют коэффициенты формы кроны и углы отхождения ветвей первого порядка от ствола. Это говорит о том, что как угол отхождения, так и отношение протяженности кроны к ее диаметру, являются признаками, специфичными для выделенных форм. Расчет коэффициентов достоверности различий между формами по всем изучаемым признакам (табл. 2) показывает, что все формы ели европейской, выделенные в урочище «Дубки», достоверно отличаются друг от друга по диаметру кроны. Ширококоническая-канделябровидная форма достоверно ниже всех прочих форм и имеет достоверно меньшую протяженность кроны. Узкоколонновидная форма достоверно меньше всех остальных форм по диаметру ствола. Коэффициент формы кроны и угол отхождения ветвей 1 порядка от ствола могут рассматриваться как диагностические признаки при определении формы кроны только в совокупности. Большинство выделенных форм достоверно отличаются друг от друга по коэффициенту формы кроны, но в тех случаях, когда формы имеют сходное соотношение длины и ширины кроны, они все равно достоверно отличаются по углу отхождения ветвей 1 порядка от ствола.

Выявленные нами формы кроны, за исключением колонновидной, не описаны ни в одной дендрологической классификации [2,3,6], чем вызывают к себе большой интерес. Такого разнообразия форм ели европейской, собранных в одном месте на небольшой площади, в нашей стране больше нет. В настоящий момент урочище «Дубки» силами общественности и ученых (в том числе, профессора кафедры селекции, генетики и дендрологии МГУЛ СП. Погиба и старшего научного сотрудника Института лесоведения РАН А.В. Абатурова) приобрело статус ООПТ «Памятник природы федерального значения».

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WATER STRESSES, OSMOLYTES AND TREE STEMS GROWTH

Mapelli S., Bertani A.

CNR – Institute For Agricultural Biology and Biotechnology, via Bassini 15, 20133 Milan, Italy,
+39-0223699418, mapelli@ibba.cnr.it

Abstract. The abiotic stresses, such as soil flooding as well as drought, can have negative effect on growth of plants that can give reduced or lost of yield in annual harvest. In trees the problem sometime is under considered in view that one season stress may have relative less effect on long living species. In West European countries reforestation progamms encountered wide interest and economical support either at European Union, National, and Regional levels. The request of planting material is increasing for wide range of ecological habitats, hilly or plain lands, dry or water rich area, hot or temperate climate. In this view studies to select the best genotypes for each habitat are important points to reduce environmental effect on growth rate and stem quality to obtain trees with low defect in wood at the moment of the cutting some year decades after planting. Experiments were carried out on walnut (*Juglans regia)*, well known for the high nutritive value of fruits and one of the more appreciated wood tree in Italy and South Europe for furniture production and on black locust (*Robina pseudoacacia)*, also an interesting tree for its hardwood suitable for handwork other then for the quality of its honey. Working on those two tree species the aim was to search and test physiological and or biochemical

parameters useful in the selective process for genotypes suitables for specific habitat area. In the last ten years about 20 varieties or provenances for *J. regia* as well as for *R. pseudoacacia* has been compared, in soil flooding or soil drought growing conditions, investigating radial stem growth, photosyntetic and transpistarion rates, osmotic potential, and its main components amino acids and soluble sugars, and stress marker compunds like ethylene, ethanol. The comparison of two significant genotypes of walnut and back locust are presented and compared. The studies give indication that comparing different genotypes is possible to have indication of the presence between them of a different degree of resistance to flooding or drought stressses. The use of daily course stem diameter evolution seems to be an indicator of stress status. Experiences indicate that flooding or drought could have negative effect on cambial activity and consequently on wood formation also in the successive growing seasons. Furthermore there are evidence that flooding or drought act in different ways and different intensity also in relation of tree species or genotypes. The use of physiological and biochemical markers can contribute to obtain indication useful for selection of stress resistance aptitude. In particular ethylene or ethanol presence and evolution can be markers for hypoxia resistance and osmotic potential seems relateted to drought conditions in young trees. Long time experiments are necessary to evaluate the response on elder trees and the stresses consequence on wood characteristic and quality.

Too much water (flooding or waterlogging soil) as well the low water availability (drought, deep water table) are both to be considered water stresses for plant growth. If the crops, vegetables or weeds, as consequence of unbalanced water availability, loose the growth and productivity in the year when water stresses occur, for trees a water stress may have consequence in the following years tree life, harmonical growth and wood quality. The trees may have to suffer water stresses more times during the life and as consequence the trunks show an unhomogeneous rings that can influence wood quality and economical value.

In West European countries, after the long time of tree cutting in the last decade of the 20th century, reforestation progamms encountered wide interest and economical support either at European Union, National, and Regional levels.

So, the request of planting material is increasing for wide range of ecological habitats, hilly or plain lands, dry or water rich area, hot or temperate climate. In this view it is clear that differeces inside the biodiversity of the same tree species exist and studies to select the best genotypes for each habitat are important points to reduce environmental effect on growth rate and stem quality to obtain trees with low defect in wood at the moment of the cutting some year decades after planting.

The walnut (*Juglans regia)*, well known for the high nutritive value of fruits, is one of the more appreciated wood tree in Italy and South Europe for furniture production. Sometimes the fruit production is considered only a side product. Black locust (*Robina pseudoacacia)* is also an interesting tree for its hardwood suitable for handwork other then for the quality of its honey. Working on those two tree species the aim was to search and test physiological and or biochemical parameters useful in the selective process for the best genotypes suitables for each specific habitat area. In the last ten years about 20 varieties or provenances for *J. regia* as well as for *R. pseudoacacia* has been compared, in soil flooding or soil drought growing conditions, investigating radial stem growth, photosyntetic and transpistarion rates and stomatal conducance, osmotic potential, and its main components amino acids and soluble sugars, and stress marker compunds like ethylene, ethanol and proline. Here comparison of two significant genotypes of walnut and back locust are presented and compared. For walnut two years old trees, obtained from seeds, were potted in 50 l individual cylindrical containers filled with a mixture of 50 % clay soil and 50 % peat. Blacklocust was propagated in winter by root cuttings, in same size container and soil mixture, and used from the first growing seasons. The trees were grown outdoors and irrigated with automatic system two times a day (at dawn and sunset). Early in spring trees has been selected for homogeneity considering the length of new shoot formed and number of leaves. For flooding treatment plant containers were immersed in a larger one and filled it with water to about 2 cm above soil level. For drought stress the selected tree pots do not received irrigation water and were protected from season rain under plastic shelter and maintained in open field conditions. To measure the radial growth, Linear Variable Differential Transducers (LVDT) were applied. LVDT with home build supports, were fixed to the tree stems. The complete system weighted about 40 g and the part in contact to the stems, Parafilm and glue, were permeable to water vapour. The device had a displacement of ±2,5 mm and according to the constructor data it gave an output signal of 900 mV/mm. LVDT were connected to a datalogger, with data averaged over 15 min. Datalogger resolution is at 0,1 mV in the range ±700 mV, that is

equivalent to 0,1 μm of tree stem diameter fluctuations. When necessary the LVDT can be repositioned without any disconnection from the stem and interruption of data collection.

Net photosynthesis, transpiration, conductance were simultaneously measured, during all the growing season, with a portable Infra-Red Gas Analyzer (IRGA). Periodically, in the period from June to the end of September, leaves, stem and root pieces were collected from control plants. From flooded or droughty trees, samples were collected at interval times during stresses and after stress remove. The samples are used for biochemical substances extraction and analyses that had been carried out standard and HPLC methods.

The Figure 1 shows the typical effect of stresses, flooding or drought on walnut stem trunk diameter during the growing season. The Lozeronne genotype has been identified as one of the most sensitive to soil flooding effect. The block of diameter growth effect caused by short period of flooded soil in many genotypes have consequence also in the following years, when only some lateral branches will growth and wood formation in the main trunk restart to growth only two or more years later.

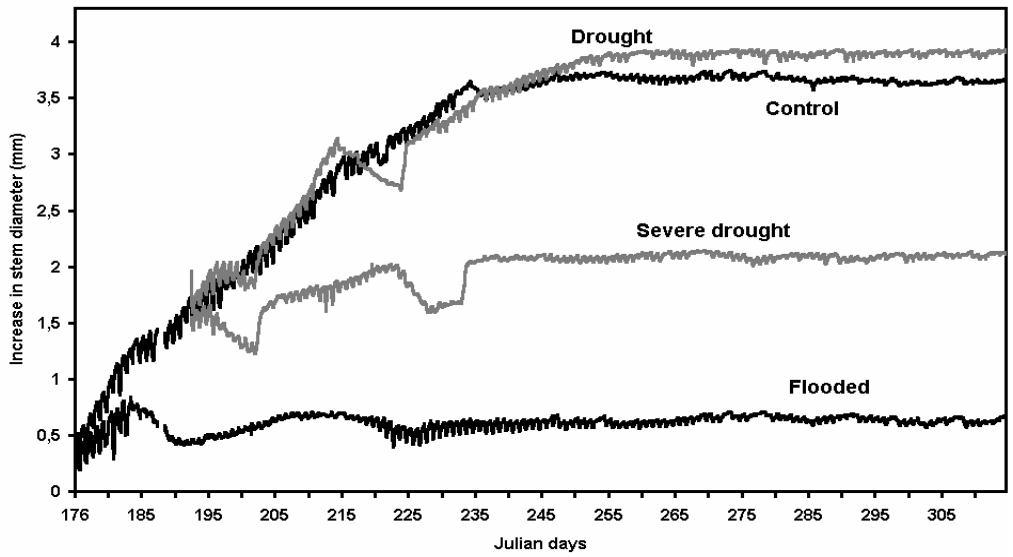

Figure 1. Lozeronne walnut stem diameter increase during growing season. Flooding was imposed from day 181 to 188. Drought was from the days 204 to 211 and again between 230 and 242. Severe drought was between days 190 and 203 and days 222 to 233.

In all *J. regia* genotypes considered, a rapid decrease of carbon dioxide assimilation was the evident consequence of the stress. In sensitive *J. regia* genotypes as v. Lozeronne, few days were enough to for wilting leaf symptoms. The wilting was extended to whole leaves and began to shed also after soil drainage. On the contrary, in the Soraloviner progeny of selected *J. regia* trees, subjected to soil anoxia, showed a decrease in CO_2 assimilation but they showed the ability to recover the photosynthetic activity when the stress was ended after 9 days (Fig. 2), without visible leaves damages. The recovered functionality of leaves, however, was not accompanied by recover of stem enlargement. In Soraloviner progeny, that is one of more resistant to soil flooding, if the stress was prolonged to 13 days leaves started to wilt and in 1–2 days all leaves desiccate and subsequently shed.

Ethylene as phytohormone is well known for its interaction in the plants growth subjected to abiotic and biotic stresses, ethanol is well know metabolite during hypoxic conditions and accumulation can became toxic. Analysis of ethylene in the leaves of Soraloviner genotypes trees (Table 1) showed increase during the stress treatment. Ethylene increase begun concomitantly to the CO_2 assimilation decrease. The ethanol, already present in leaves of control trees, also increased during root anoxia condition and it continued to rise till the 11th day, later the leaves became wilted and measures became unreliable. On the contrary in the v. Lozeronne low level of ethanol is present in control trees and relatively low increase occurred during the stress period. All those two compounds in some ways seemed related to flooding resistance in walnut considering that in our experience both were present in genotypes that showed higher resistance to flooding.

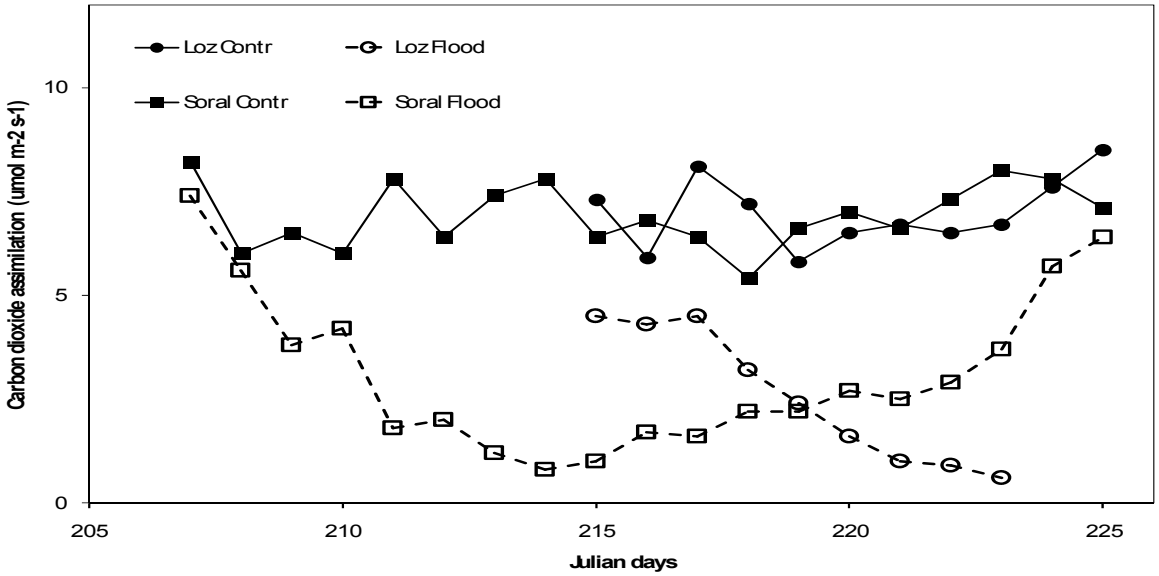

Figure 2. Net photosynthesis in control or in soil flooded walnut trees. The treatment began at day 207 for Soraloviner and ended 12 days later. For Lozeronne was from day 215 till day 222. At this day leaves were wilted.

Table 1. **Changes of ethylene and ethanol content in leaves of tolerant or untolerant *J. regia* tree varieties to soil anoxia treatement**

	Ethylene ng/g leaf fresh weight		Ethanol µg/g leaf fresh weight	
		Soraloviner		
	6 days	11 days	6 days	11 days
drained soil	Trace	Trace	15,23 ± 1,81	18,39 ± 1,92
flooded soil	31,21 ± 2,66	23,36 ± 2,73	21,65 ± 1,73	33,67 ± 2,14
		v. Lozeronne		
	2 days	4 days	2 days	4 days
drained soil	Trace	Trace	1,91 ± 0,09	1,27 ± 0,11
flooded soil	1,95 ± 0,06	3,56 ± 0,13	2,76 ± 0,84	5,53 ± 1,07

When osmotic potential is evaluated of wide interest is the free aminoacids component or solutes. In leaves of control plants free aminoacids ranged between 2–5 nmoles/mg F W. In genotypes resistant to soil flooding the concentration rised up to 25–30 nmoles/mg F W. The increase was characteristic of flooding stress. When the same walnut genotypes are subjected to drought the the total osmotic potential increase but not more of 25–30 % comparing the control trees.

Over then the high increase seemed of interest the aminocids composition. The glutamine increased to be about 70 % of the total aminocid quantities. The post flooding period showed the increase of the citrulline. The quantities of those two amino acids contribute to support the hypothesis of role as compounds for nitrogen storage and remobilization during hypoxia and recovery periods.

The black locust trees were mainly investigated for responses to drought stress, large part of experiments had been carried out on Nyirsegi and Ulloi varieties constituted several decades ago in Hungary with indication that Ulloi is more sensitive to drought then the Nyirsegi.

The Figure 3 show the stem radial growth for the Nyirsegi when the drought has been imposed at two different level of replacing the water transpired by leaves, the effect on radial stem increase is evident with different intensity. The severe drought blocked the stem growth in few days. When soil had been rewatered the stem of droughty trees showed a recovery of different amplitude in relation to the drought degree previously applied to the trees. The Ulloi gave similar response in stem radial growth; the two varieties did not evidenced differences in the rate of growth and sensitivity to the drought and this seems in contrast to previous field observation made during long period. Together all the *R. speudoacacia* accessions tested can be dividied in three groups on the basis of stem diameter responses to drought stress. Control and high stressed trees of Ulloi

and Nyirsegi were monitored during winter and next spring season. The stem radial growth started to increase at least 15 days later nd with slow rate on Ulloi tree subjected to drought in the previous year.

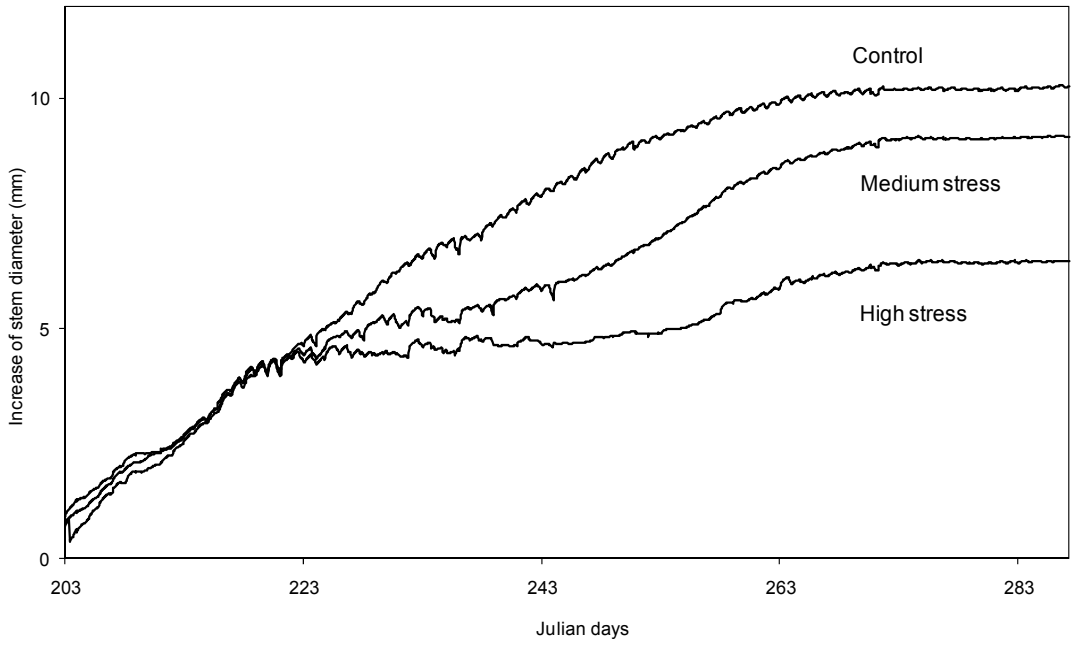

Figure 3. Comparison of stem radial growth in growing season of *R. pseudoacacia* cv Nyirsegi trees grown in well watered condition and drought condition started at day 216 and terminated at day 244.

Within the physiolgical and biochemical parameters measured the osmotic potential seemded the more interesting. The osmotic potential of Ulloi and Nyirsegi samples are shown in Figure 4. The Ulloi leaves and root do not show differences in osmotic potential comparing control to medium or high stressed trees. The Nyirsegi samples show an increase in osmotic potential with increasing stress intensity. To note that the maximum potential measured in Nyirsegi stressed leaves was comparable to the osmotic value measured in control leaves of Ulloi. So, the absence of total osmotical increase in stressed Ulloi trees could be due either to the absence of adaptive mechanism or the high osmotic potential in control is already too high to permit a further increased by drought condition. In general, considering all the black locust genotypes studied the high osmotic potential in trees growing in well watered soil can be related to their lower adaptiveness to drought.

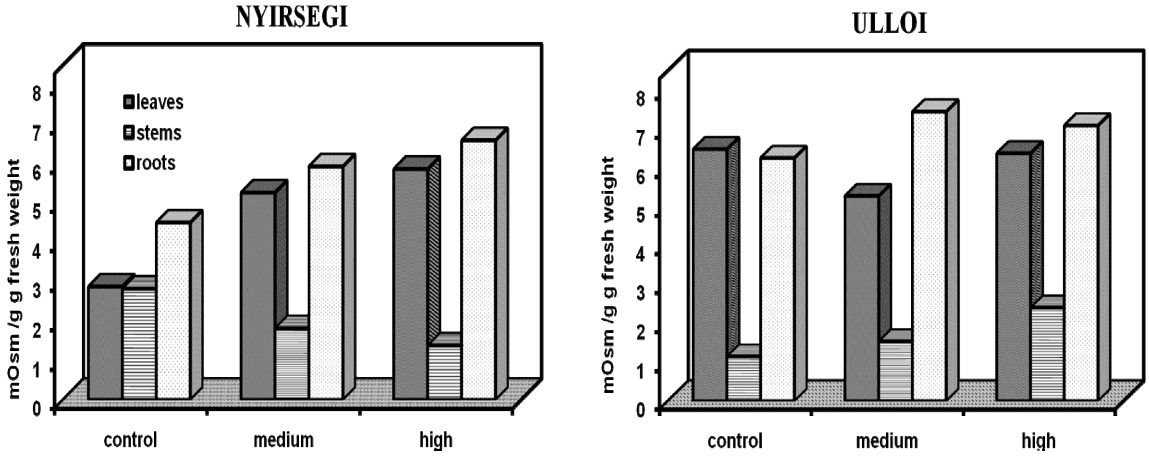

Figure 4. Comparison of osmotic potential in control and drought stress robinia trees in tree components measured after 28 days of controlled stress conditions.

The analyses of some components of the substances that are part of osmotical components has been carried out. In both clones determination of total amino acids showed increased concentration due to drought. Furthermore, the proline (widely considered as amino acid stress marker) content increases also in both clones during drought condition. Those data indicate that both clones react to stress increasing amino acids concentration. The determination of total soluble sugars (sucrose plus reducing sugars) indicate no general significant increase during stress period in both clones comparing the level in the control samples The analyses carried out did not explain what are the components of high osmotic potential measured in control Ulloi. Further analysis will be developed considering other carbohydrates (fructan, threalose), organic acids as possible osmolytes.

Considering the wide differences in osmotic potential measured between Ulloi and Nyirsegi young control trees used for experiment, leaves from large number of trees, 2 and 35 years old has been collected in black locust plantations. This in order to subject them to osmotic measures and evaluate the statistical significance between the two clones. The Table 2 reports data from which it is possible to see that the two varieties have significant osmotic component concentration.

Table 2. **Osmotic potential measured in leaves of 35 year old and 2 year old Ulloi and Nyirsegi trees (mOsm / g F W)**

	35 years old trees (n 21)	2 years old trees (n 36)
Nyirsegi	1,016 ± 0,04542	1,358 ± 0,03239
Ulloi	3,149 ± 0,03584**	3,774 ± 0,05942**

** significant different at $P < 0,05$

The studies give indication that comparing different tree genotypes is possible to have indication of the presence between them of a different degree of resistance to flooding or drought stressses. The use of daily course stem diameter evolution seems to be an indicator of stress status. Experiences indicate that flooding or drought could have negative effect on cambial activity and consequently on wood formation. The negative effects may have consequence also in the successive growing seasons. Furthermore there are evidence that flooding or drought act in different ways and different intensity also in relation of tree species or genotypes. The use of physiological and biochemical markers can contribute to obtain indication useful for selection of stress resistance aptitude. In particular ethylene or ethanol presence and evolution can be markers for hypoxia resistance and osmotic potential seems relateted to drought conditions in young trees. Long time experiments are necessary to evaluate the response on older trees and the stresses consequence on wood characteristic and quality.

CAMBIAL ACTIVITY IN CERRADO SPECIES OF SOUTHEASTERN BRAZIL

Marcati C. R. [1]***, Wiedenhoeft A. C.*** [2]

[1] Unesp – Universidade Estadual Paulista FCA, Departamento de Recursos Naturais. Fazenda Lageado - Rua José Barbosa de Barros, nº 1780, CP 237, 18603-970, Botucatu, SP, Brazil, +55(14)38117162, carmen@fca.unesp.br;
[2] Center for Wood Anatomy Research, Forest Products Laboratory, One Gifford Pinchot Dr., Madison, WI, 53726-2398, USA.

Studies on seasonal activity of the vascular cambium and how that activity is influenced by environmental factors (temperature, rainfall, soil moisture, daylength), phenology, and plant habitat provide critical data for understanding the growth dynamics of trees. It is generally assumed that, in temperate trees, cambial activity is influenced primarily by temperature and daylength, and that in tropical trees, rainfall and available water are more important factors in the control of cambial activity.

Distinctly seasonal cambial activity occurs in many tropical and subtropical regions that experience severe annual dry seasons, including regions of Brazil [31, 32, 4, 19, 6, 10, 20, 21], but most of the detailed studies from such regions have been conducted on Indian taxa [13, 14, 15, 8, 9, 1, 18, 29, 23, 24, 25, 26, 30]. The period of cambial activity in tropical trees typically is longer than that of temperate trees – in some tropical species extending throughout the year [11, 17].

Because of the paucity of detailed information on cambial activity in Brazilian species, the present work was aimed at providing information on cambial activity in relation to water availability in 1, 2, and 3