

Full Length Research Paper

Growth rings of African timber described by an approach using Gis tools: Case of *Milicia excelsa*

Cédric Ilunga^{1*}, Prosper Sabongo², Joseph Komba³, Idriss Ayaya⁴ and Leopold Ndjele²

¹University of Kisangani, Faculty of renewable resources management, B. 2012 Kisangani, Democratic Republic of Congo, ²University of Kisangani, Faculty of Sciences, B. 2012 Kisangani, Democratic Republic of Congo,

³Compagnie Forestière et de Transformation (CFT), Kisangani, Democratic Republic of Congo, ⁴Northeast Forestry University, College of Wildlife resources, B.150040.

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One of the challenges in tree-ring analysis in the tropical regions of Africa is the lack of accessible methods for researchers working in this field. Recently, some innovative methods using Geographic Information System (GIS) tools and digital images have been developed and their use proved to be inexpensive, high accurate and efficient approach to tree-ring analysis. In this study, one of these methods (Dendro-Gis) adapted to large wood samples was tested in describing tree-rings of *Milicia excelsa*. The aim was to explore whether the use of GIS tools would provide additional data compared to classic methods (microscope, magnifying glass or the naked eye). For this purpose, high-resolution digital images from sanded cross-section stem disc of *Milicia excelsa* were processed using ArcMap software enhanced with other image processing softwares. The approach used made it possible to digitize ring boundaries over the entire circumference of the disc. Tree-ring analysis provided numerous data: ring width series, occurrence and location of ring anomalies (wedging rings particularly) and other geometric information stored in the geodatabases that offer possibilities of many retrospective analysis. This new knowledge is useful for future dendrochronological studies of this species in the tropical rainforests.

Keywords: Tree-ring analysis, Gis tools, digital images, *Milicia excelsa*, ring boundaries, wedging rings, wood description, Democratic Republic of Congo.

INTRODUCTION

Tree-ring analyses have been conducted on tropical trees for more than a century (Worbes, 2002). Though reliable tree-ring chronologies have been constructed and studied in several tropical sites around the globe (Worbes, 2001; Worbes et al., 2003; Brien and Zuidema, 2006a; Brien and Zuidema, 2006b; Mbow et al., 2013), there is still a dire need for new tree-ring analyzes in the tropics on a variety of tree species growing in diverse ecosystems. Tree rings not only provide detailed information on tree age and growth, but are also linked to forest management (Gebrekirstos et

al., 2014) and changes in climatic conditions (Schongart et al., 2006; Trouet et al., 2006, Couralet et al., 2010; De Ridder et al., 2014). Unfortunately, several factors hinder the studies of tropical tree-rings, especially in Central Africa. Successful dating of tropical trees remains difficult because of the lack of marked seasonality in large areas of the tropics. On the other hand, the anatomy of tropical wood itself obscures tree-ring detection. Indistinct rings, false and double rings, wedging rings and intra-annual growth variations are among phenomena that make tree-ring delineation difficult (Worbes, 1995; Hayden, 2008). In addition, tree-analysis requires the use of specialized tools and expensive equipment, which may hinder researchers working under limited budgets (Castro et al., 2015). Despite these challenges, research efforts in the tropic

*Corresponding author. Email: ilungawak@gmail.com

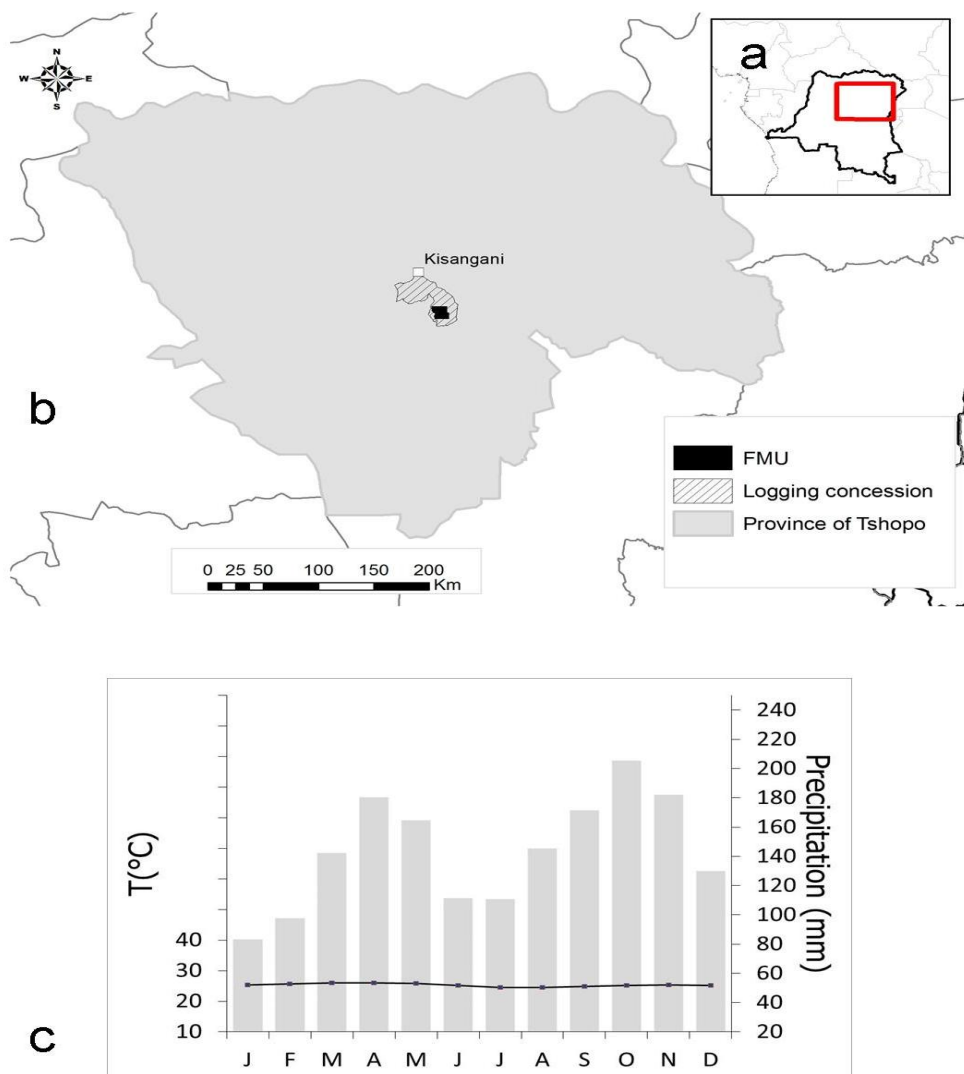


Figure 1: Geographic location of the study area in the Democratic Republic of Congo (a), in the surroundings of the city of Kisangani (b) together climate diagram. The area of CFT logging concession is indicated by the polygon (diagonal stripe) with the Forest Management Unit-FMU (black polygon) where samples originate. Mean monthly temperature (Black lines, °C) and precipitation (grey histogram, mm) were calculated for the period 1967-2005 based on the data of Kisangani airport (Kahindo, 2011).

continue in order to increase knowledge on tree growth and more specifically, tree-ring characteristics and chronology-building. The studies are currently concentrated on most commercial species, especially species facing serious threat of extinction (Included in CITES or recorded as “Endangered on IUCN Red List”) (Groenendijk et al., 2014). Research projects give particular priority to the species known to produce clear and annual tree rings. Therefore, good knowledge of anatomical wood structure is necessary to a better understanding of the delineation of tree rings in order to create the development of quantitative tree-ring studies or dating correctly ring-width chronologies.

Although pioneering studies in Sub-Saharan Africa (Mariaux, 1967; Catinot, 1970; Détienne, 1976; Détienne and Mariaux, 1997; Détienne et al., 1998)

provided basic knowledge about the nature and periodicity of tree rings of some species, the current challenge is to improve this knowledge or verify it in certain aspects. Improvements in this knowledge are possible through the use of new observation methods and measurements of ring widths. Most of these methods have evolved under the impetus of new technologies in computing, particularly imaging systems (Von Arx and Carrer, 2014). Some studies have resorted to the Geographic Information System (GIS) software and digital images for tree-ring analysis. To our Knowledge, an innovative method using Diva-Gis freeware to analyze scanned core images was described and tested against the widely-used software LINTAB-TSAPWin and WinDENDRO (Castro et al., 2015).

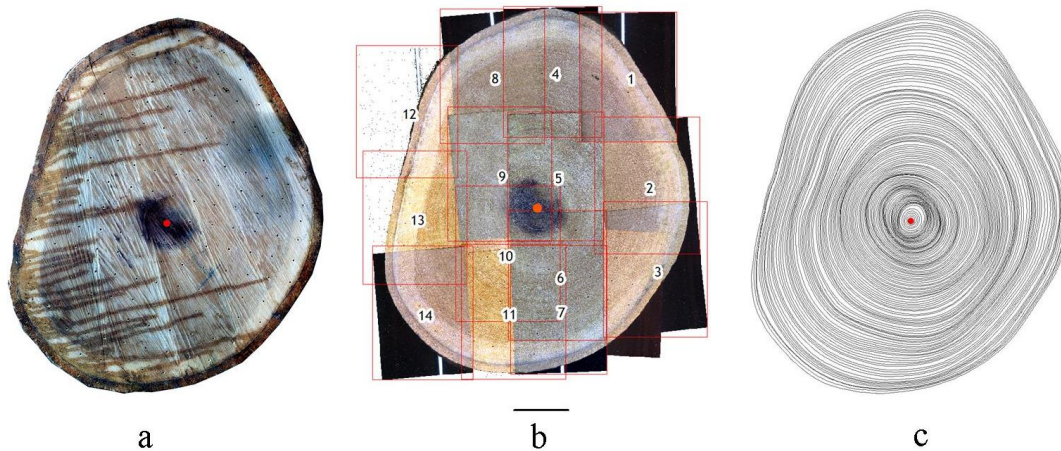


Figure 2. Main phases of wood preparation (sample 3). (a) : Green wood disc ortho-image generated by the photos using Photoscan – Red point represents pitch. (b): Scans mosaic (tree 3). -All scans georeferenced in order to generate dry wood stem disc are identified by the number (1,2,3,...). The holes used as control point to process shrinkage-free images are also visible in the dry wood stem disc. (c): Full digitalized disc- The grey lines correspond to the tree-rings digitalized over the complete area of stem disc.

More recently, Latte et al. (2015) gave an outlined description of a novel procedure to measure shrinkage-free tree-rings from a very large wood sample, highlighted its potential for tropical dendrochronological studies. This interesting approach offers opportunities of carrying out tree-ring analysis over a complete stem disc. In this sense, it seems suitable for tree-rings description and measuring of tree species from tropical regions.

Inspired by the above mentioned studies, this study tries to explore the potential of “Dendro-Gis” tools in describing the anatomical characteristics of tree rings of *Milicia excelsa* (A.Chev.) C.C. Berg from tropical rainforests of the Democratic Republic of Congo. Based on five wood samples, we tested if dendro-Gis tools could provide additional information in terms of descriptions. The output of the approach used will be compared with the previous studies focused on the same species.

MATERIAL AND METHODS

Study area

The study site is around the city of Kisangani, in the province of Tshopo (Northeast of DRC). Stem discs were collected from 46/11 logging concession sites assigned to the CFT logging company (Compagnie Forestière de Transformation). The extreme borders encompass 0°30'N 0°10'S and 25°00' E and 25°35'. The climate of the site corresponds to the Af-type according to the Köppen classification system (1936).

Average annual precipitation varies between 1500 and 2000 mm and there is no dry season (monthly rainfall < 60 mm) (Worbes, 1995) although drier conditions occur in January-February and, to a lesser extent, from June to July (Fig. 1(c)). Mean annual temperature is 25°C with only low intra-annual variation. The annual average air humidity varies between 90% and 95% (Boyemba, 2011).

The study site is almost flat (slopes < 10%) and the elevation varies around 400m above sea level. The soils are composed of a mixture of sand and clay which present good physical properties but very limited chemical fertility because of low cation exchange capacity (2 and 8 mEq/100g), low acidity (pH 3.5-5.5) and high retention of phosphorus on iron oxides (De Ridder et al., 2014).

The vegetation is of a lowland mixed-moist tropical rainforest of the Guineo-Congolian Region and is categorized together with the moist Central African type (Lebrun and Gilbert, 1954). The dominant species observed in the CFT concession are *Gilbertiodendron dewevrei* (De Wild.) J. Léonard (limbali) in mature and almost mono-dominant stands, associated with high abundance of *Brachystegia laurentii* (De Wild.) Louis ex Hoyle (bomanga) and species such as *Khaya anthotheca* (Welw.) C. DC. (mahogany), *Entandrophragma cylindricum* (Sprague) Sprague (sapelli), *Pericopsis elata* (Harms) Meeuwen (afroomsia/assamela) or *Milicia excelsa* (iroko) in others forests.

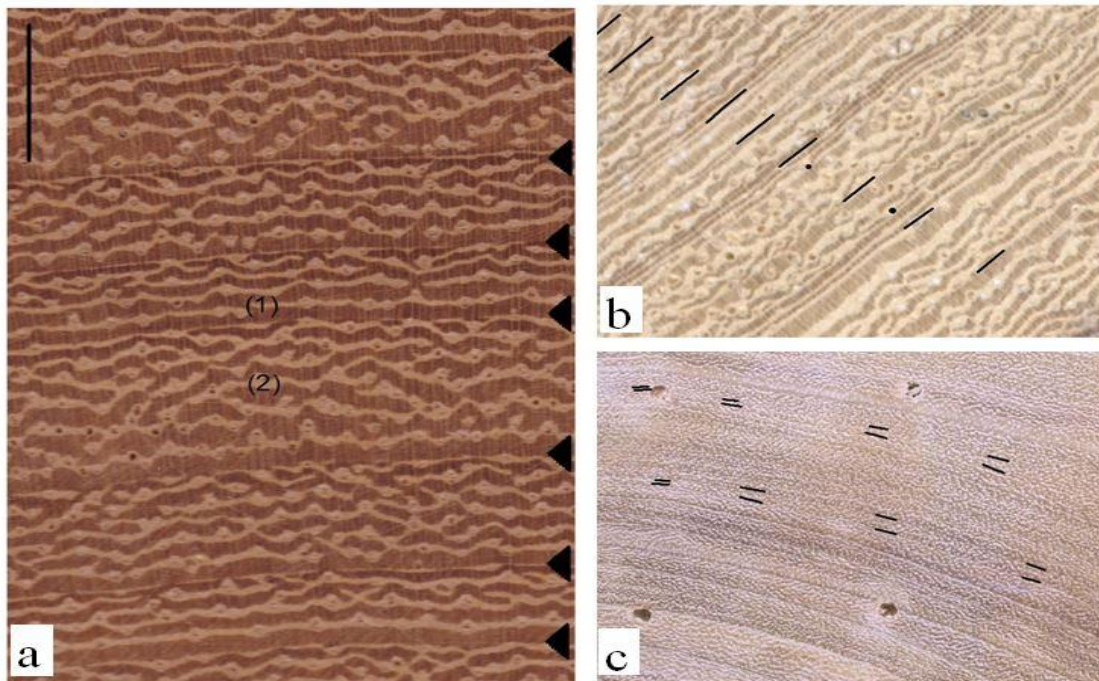


Figure 3. Images of transverse surface of wood samples. (a): High-resolution (2500 dpi) scan on sanded stem disc (sample 3). *M. excelsa* showed distinct tree-ring boundaries, consisting of marginal discontinuous parenchyma bands. Growth direction from bark (upside) to pitch (downside image); Scale bar=5 cm; filled black triangle = annual tree-ring boundaries. (b): Identification of false rings over portion of stem disc (sample 3). False rings are indicated by black points and true rings correspond to the short lines (traits). (c): Phenomena of anastomoses, leading to the formation of wedging rings. Lines show the rings boundaries that merge and form wedges. The hole corresponds to the control point used to process shrinkage-free images.

Studied species –*Milicia excelsa*

Commercially known as iroko, *M. excelsa* is a hardwood tree of great socio-economic and cultural importance in its complete distribution area in West, Central and East Africa (Durrieu de Madron et al., 2000; Ouinsavi and Sokpon, 2010; Dainou et al., 2012). As a pioneer and light-demanding species, *M. excelsa* is dioecious small-seeded and animal-dispersed (Fayolle et al., 2015). This species can exceed 130 cm in diameter and can grow taller than 40 m in height (Letouzey, 1982; Lejoly et al., 2012). Because of the highly attractive technological properties of its wood and its multipurpose use, this species is among the five most exploited species in Central Africa and is recorded among the threatened species on the IUCN Red List (De Wasseige et al., 2013). The nature and periodicity of *M. excelsa*'s rings were studied deeply by Detienne (1976).

Sampling and wood preparation

A total of five trees of *M. excelsa* were logged in 2014, from June to December in the CFT concession region (Figure 1). Stem disks of 10-15 cm thickness were

collected at the bottom of the logged trees (1-1.8 m at stump height) ranging from 83 to 103 cm in diameter, equal or higher than the Minimum Cutting Diameter (MCD) of 80 cm (DIAF/MECNT, 2009). All wood samples were prepared following the standard procedure of Latte et al. (2015) that combines photogrammetry, high-resolution image processing and GIS tools.

The first step in the preparation consisted in taking photos of the surface of stem discs with an off-the-shelf camera. Photos were then assembled in a green wood ortho-image (Figure 2(a)) with the Photoscan software v1.0 (Agisoft; Carshalton, UK). The stem discs were then air-dried (during 72 hours, ambient air temperature around 25 °C) and sanded, using different grits (40, 80, 100, 120, 240, 400 and 1000).

The complete stem discs were scanned at a resolution of 1200-1600 dpi using a flatbed A4 scanner (A4-Epson Perfection V500). Between 15 and 20 scans were necessary to cover a disc of 90 centimeter in diameter.

The scans were assembled to form a high-resolution image mosaic (Figure 2(b)) called the dry wood image shrinkage-free images (Latte et al., 2015). Using the scan mosaic as background, tree-ring boundaries were manually digitalized as polylines over the complete cross-section of the disc (Figure 2(c)).

Table I. Frequency of tree-ring anomalies in *Milicia excelsa* for five wood samples (tree 1 to 5): Diameter at Brest Height (DBH), total number of rings along each radius (Nr), Wedging rings (Wr) and False rings (Fr).

Sample	DBH cm	Radius									Mean (%)	
		1			2			3			Wr	Fr
		Nr	Wr	Fr	Nr	Wr	Fr	Nr	Wr	Fr		
Tree 1	103	113	7	1	94	14	1	110	5	1	8	1
Tree 2	98	143	19	5	137	15	7	162	24	0	19	4
Tree 3	81	125	10	1	131	16	2	132	14	0	14	1
Tree 4	83	130	46	5	94	26	2	96	27	6	33	4
Tree 5	90	109	3	0	142	15	0	145	12	0	10	0
											17	2

Tree-ring data analysis

Description of wood structures and identification of ring boundaries

The ring boundaries were identified on the cross-section of each sample from the high-resolution images. The structure of the ring boundary was described according to the general classification (Wagenführ (1999) cited in Nzogang (2009)) which distinguishes four types of criteria: (1) intra-annual wood density variation, (2) boundaries marked by a marginal parenchyma, (3) repeated pattern of alternating fibre and parenchyma bands and (4) variation in vessels distribution and/or vessels size.

After detection of ring boundaries, tree-ring boundaries were manually digitalized with ArcMap over the cross-section stem disc (Figure 2(c)). Some elements of the ring structure visible on high-resolution images have been described and their size and distribution have been estimated as much as possible. Special attention was given to the detection of tree-ring anomalies: false rings and wedging rings. Generally, false rings result from intra-annual variation caused by a short drought during the growing season (Détienne, 1989). We considered as wedging rings ('partially missing rings'), rings that do not cover the whole circumference of tree or are locally absent (Détienne, 1976; Trouet et al., 2006). False rings and wedging were detected by the differences in the formation of the ring boundary and by checking the continuity of the rings over the entire disc respectively. Using the topology tools available in ArcMap, Wedging rings were isolated by identifying polylines (Figure 2(c)) that do not form a polygon. This means concretely that after the digitalization of all ring boundaries (Figure 2(c)), the complete rings have been converted to polygons and the others (called "dangles") have been identified as wedging rings. We counted the number of wedging rings along the radii (transects) used for the measurements of ring widths (three radii par tree).

Ring measurement

Although the rings were digitized throughout the circumference of discs, we considered three radii per wood sample for the calculation of ring widths. On each radius considered, all identified rings were marked and counted automatically from the pitch to the bark under ArcMap. Using the function "point distance" available in ArcMap's analysis tools, the ring width measurements were done to the nearest 10^{-5} mm and in a direction perpendicular to the ring boundaries. All ring width series obtained were saved in the geo-database.

Geometric, spatial and statistical analysis

The use of scan mosaic (Figure 2(a)) enables researchers to distinguish the sapwood from the heartwood, two parts that are visually distinct in color. The heartwood was digitized as polygons and quantified in terms of basal area. The surface of sapwood was derived from subtracting basal area of the stem disc from the delimited heartwood area. These geometric information (polygons) stored in the geo-database were analyzed in relation with ring width series, hence it was possible to deduce the variability of ring width within the sapwood and the heartwood.

On the other hand, the polygons characterizing ring layers (see polylines in Figure 2(c)) were used in calculating the parameter of eccentricity. We used one parameter (D) expressed by the ratio between basal area of the actual stem disc and the area calculated from the perimeter of a circular disc (Ngomanda et al., 2012):

$$D = 1 - B / (1/4\pi * L^2) \text{ (Equation 1)}$$

Where B is the basal area of the stem disc and L corresponds to the perimeter of the same disc. D value varies between 0 and 1 ($0 \leq D < 1$): D=0 for a circular disc and D tends to 1 for irregular form (e.g. buttresses). We first estimated the parameter D for each entire sample, then for individual ring layers for possible analyses of eccentricity in relation to the occurrence of the wedging rings.

RESULTS

Tree ring formation and ring anomalies

Ring boundaries were mostly marked by marginal parenchyma band on high-resolution image of sanded

cross-sections. However, they appeared discontinuous; it was easy to distinguish them from other bands of parenchyma (confluent or aliform parenchyma). In summary, tree rings were of type 2, with ring boundaries formed by marginal parenchyma, according to the classification of Wagenführ (1999) (Cf. § 2.4.1)

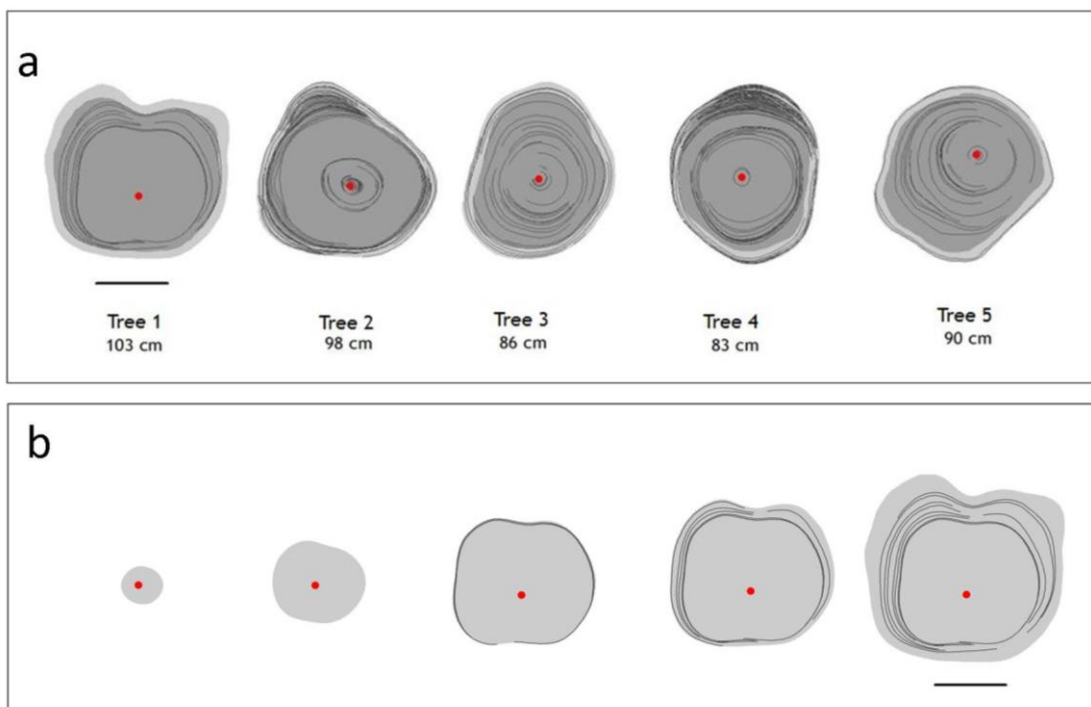


Figure 4. (a): Mapping wedging rings, using the form of the stem discs and the following descriptors: pith (red point), sapwood (light grey), heartwood (dark grey), wedging rings (black lines). (b): Illustration of the possibility of retrospective analysis relating to the evolution of the form of stem disc (Tree 1).

Vessels are present with a density of 1-2/mm². We have also distinguished the growth zone with fewer vessels and aliform parenchyma (1) and the zone with more vessels and confluent parenchyma (2) (Figure 3(a)).

As we worked on disc samples, tree ring anomalies were easily detected, particularly false and wedging rings. Table I presents the statistics on ring anomalies.

For all the 5 trees sampled, the frequency of false rings was estimated to be 2% on average. That means 2 out of 100 rings observed corresponded to false rings. The frequency of the wedging rings observed was very high (average of 17%) compared to the false rings and there were high differences between the tree samples. In this sense, this could cause serious difficulties for dating ring width series for individual trees. Visual analysis (Figure 4(a)) showed different patterns. Trees 2 and 4 had similar over-distribution of wedging rings near the bark while Trees 3 and 5 had continuous distribution from the pith to bark. Tree 1 seemed less sensitive to the phenomenon of wedging rings and had a particular pattern in terms of the distribution. Globally, it appeared that the number of anomalies increased with number of rings (cambial age) or radius length (diameter) as shown in Figure 4(a).

Ring widths variability

Identifying rings was easier and quicker to perform on high-resolution digital images. Due to the presence of wedging rings, the counting of rings by radius showed differences. The measurements of ring widths were therefore carried out depending on the rings identified and marked along the radius. Figures 5 (a) and 5 (b) show the radius length per tree (three/tree), as well as the mean ring widths from the measurement. Ring-width derived from all samples (three radii per tree) gave an average of 3.63±2.41 mm. The comparison of ring-width series between trees showed some difference (p-value = 0 by analysis of variance).

The use of ArcMap software allowed making analyses between the series of ring widths and other geometric information. This resulted in the variability of ring widths within the sapwood and Heartwood (Table II). With the exception of two samples (trees 4 and 5), the average ring widths in the sapwood (4.03 ± 0.32mm) was greater than in the heartwood (3.72 ± 0.12 mm). For all samples, the comparison between sap and heartwood in terms of ring-width showed non-significant difference (p-value=0.517).

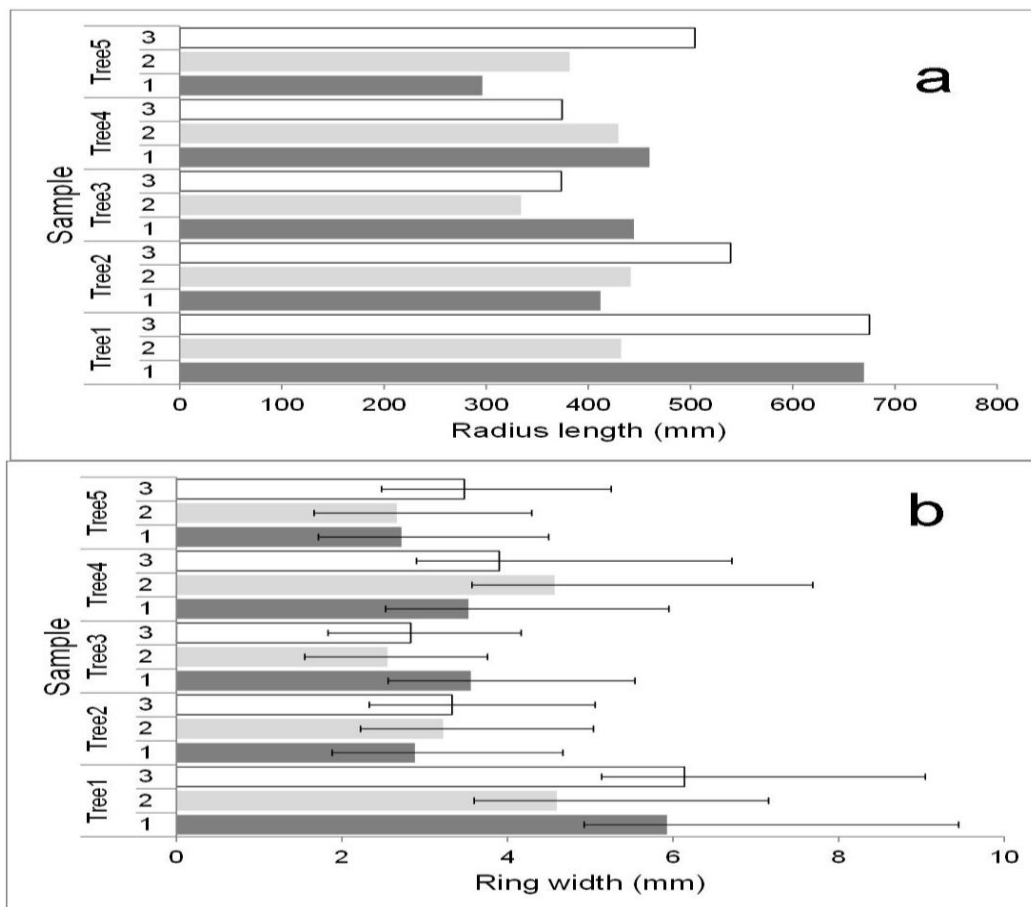


Figure 5. (a): Radius length (mm) measured (tree) and (b): mean ring-width (with standard deviation). The radii considered by tree are presented in different color (dark grey, light grey and white)

Other parameters characterizing stem disc

Other parameters relating to wood disc, such as the eccentricity (D) and the proportions of sap- and heartwood were also estimated (table II). Following the approach Dendro-Gis used, the parameter of eccentricity D have been computed for a large number of ring layers (polygons) for each sample, but only the data corresponding to the final form of the discs of each sample are presented in table II below. For the five trees the value of D varies between 0.15 and 0.26, meaning that the discs sampled had less irregular shapes (no buttresses). It should be noted that the eccentricity parameters have several variations over time, as shown in Figure 4 (b): changes in form reflect variations in eccentricity parameters.

On the other hand, the proportions of the sapwood and heartwood are presented in terms of surface in the table (see figure 4(a) for visual examination). The result showed that the proportion of sapwood varied from 19 to 25% and heartwood covers 75-80% in terms of the basal area, this corresponds to an average sapwood/heartwood ratio of 1/4. In addition, the results showed that the sapwood/heartwood ratio appears to

decrease with increasing tree diameter. These trends may well be verified in more samples.

DISCUSSION

Tree-rings of *M. excelsa*: additional information provided by the use of Gis tools

To our knowledge, studies on growth rings of *M. excelsa* (iroko) are not exhaustive despite the economic value of the wood of this species. However, since the pioneer studies in tropical Africa (Detienne, 1976; Mariaux, 1967; Normand and Paquis, 1976), iroko is acknowledged as capable of producing growth rings and the description has been uniform in all the areas that the tree is grown. The wood anatomical description of *M. excelsa* was done in further detail by Détienne (1976) and partially by other authors (Nzozang, 2009; Durrieu de Mandron, 2003; Catinot, 1970). Under climatic conditions specific to tropical rainforest of Guineo-Congolian region, marginal parenchyma has been identified as the main element that forms the ring boundaries of this species. The limits between the late wood of the previous ring and the early wood of the next ring are marked by the

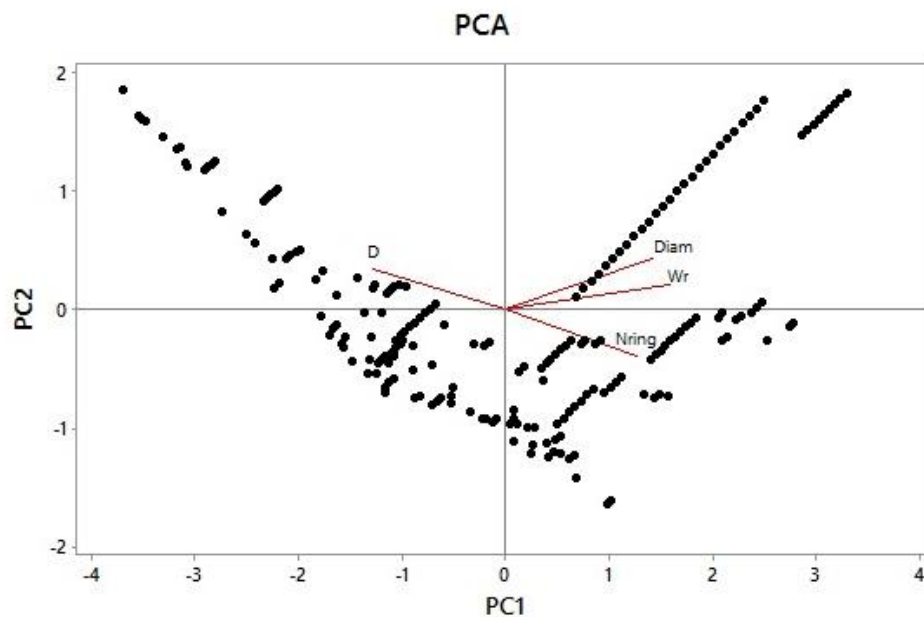


Figure 6. PCA of main characteristics of five individuals full digitalized. Corrections between 4 factors identified: Wr (Number of wedging rings observed), Nring (Total number of rings), Diam (Cumulative diameter) and D (parameter of eccentricity).

marginal parenchyma band (Figure 3(a)). Referring to the classification of Wagenführ (1999) cited in Nzogang (2009), ring formation is associated with type 2. Our observations on high-resolution image (1200-1600 dpi) of cross-sections of the stem disc (sanded sections) came to a similar conclusion. Distinctiveness of rings on *M. excelsa* was always sufficiently clear on images (sanded sections) or by the naked eyes. The marginal parenchyma band is often combined with fibre cells at the beginning of growth rings but the fibre characteristics are hardly observable on a well-sanded surface even at high-resolution digital images used in this study. Recognition of fibre characteristics was possible through the difference in wood coloration of tissues (darker tissues). In this study some additional information about ring formations of *M. excelsa* has been provided. The main outputs in terms of wood description concern the full digitalization of tree rings (see in Supplementary material), ring-width series, the estimation of ring anomalies (especially the location of wedging ring), the quantification of basal area and the variability of ring widths associated to the sap- and heartwood and numerous geometric information (e.g. parameter of eccentricity) that could be used for several retrospective analysis (Figure 4(b)).

The anomalous rings were detected either by the differences in the formation of the ring boundary (False rings) or by checking the continuity of the rings over the entire stem disc (Wedging rings). As mentioned (§2.4.1), false rings result from intra-annual variation, caused by a short drought during the growing season. Indeed in tropical regions true ring boundaries corresponding to seasonal dry periods, generally span the entire circumference of the tree (Worbes, 2002). So false ring boundaries are often produced in the middle

of the season of ring formation because of episodic drought (Kozłowski et al., 1991, cited Mbow et al. (2013)). Wedging rings (rings that do not cover the whole circumference of trees or are locally absent) were detected by a semi-automatic procedure (topology rules) followed by a visual control. According to Worbes (2002), wedging rings occur with changing light saturation in a tree's life when competition from neighbours changes its direction, so that rings tend to wedge only in some part within one stem disc. We only observe rings on one side of the stem while no rings have formed on other. Worbes (2002) also believes that climate fluctuations within seasons can cause formation of false or wedging rings.

Contrary to conclusions of Détiénne (1976), our research showed that ring anomalies were sufficiently frequent on the selected species, *M. excelsa*. The percentage of false rings varied from 1 to 4% and those of wedging rings gave an average of 17% (for all five trees). There are various explanations for the observed incidence of ring anomalies, especially for wedging rings. Firstly, Détiénne (1976) examined ring boundaries microscopically (with a magnifying glass) or by the naked eye. Due to this procedure wedging rings and other ring anomalies were not detected clearly as the number of errors in ring marking was high. A second explanation might be the quality of sample: the diameter of the trees collected in this study (diameter ≥ 80 cm) were very large than sampled trees used by Détiénne (diameter ≤ 40 cm). We observed that the frequency of wedging appeared to increase with diameter (Figure 4(b)).

Finally, the incidence of wedging rings also seems to be potentially related to the eccentricity of stem discs (Worbes, 2002).

Table II. Characteristics of five tree prepared on wood disk (stem disc): ID of wood samples (Tree n), Diameter at Breast Height (DBH), Parameter of eccentricity (D), Basal area of transverse section (Ba) and proportion of area and ring width (rw) associated to Heartwood and Sapwood.

Sample	DBH	D	Ba	Heartwood		Sapwood		Total
	Cm			cm ²	% Ba	Rw	% Ba	
Tree 1	103	0.261	8492.9	80.1	5.42±2.95	19.9	7.44±4.05	6.43±3.5
Tree 2	98	0.138	5544.6	78.8	3.12±1.78	21.2	3.35±1.78	3.26±1.17
Tree 3	81	0.121	4166.9	76.8	2.89±1.57	23.2	3.63±1.62	3.26±1.59
Tree 4	83	0.125	4888.8	74.9	4.13±2.97	25.1	3.14±1.48	3.64±2.23
Tree 5	90	0.128	4839.2	80.4	3.02±1.82	19.6	2.61±1.02	2.81±1.42
Mean(±sd)				78.2±2.3	3.72±0.12	21.8±2.3	4.03±0.32	3.63±2.41

Table III. Matrix of correlation between factors explaining probably wedgings rings. Wr= Wedging ring observed, D= Parameter of eccentricity, DBH= Diameter at Breast Height. The presence of wedging rings (Wr) is mainly correlated to the eccentricity and the age (number of rings).

	PC1	PC2	PC3	PC4
Wr	0.568	0.298	0.141	-0.753
D	0.455	-0.572	0.639	0.236
DBH	0.511	0.599	0.054	0.613
Nring	-0.456	0.472	0.753	-0.015

Unfortunately, it is difficult to make a comparison on the disc aspects due to lack of information on the form of samples used by Détienne. The Gis tools used in this study offer researchers possibilities of retrospective analysis of the wedging rings in relation to the characteristic of stem disc (diameter, eccentricity, age), this potential will be exploited in future studies (Figure 4(b)).

For verifying some assumptions, we have undertaken some exploratory analyses. Particularly, we have at least attempted to analyze the incidence of wedging rings observed on our tree samples with the various parameters that were investigated such as cumulative diameter (Diam), total number of rings as cambial age (Nring) and the index "D" as factor eccentricity (Equation 1) and that characterize stem disc. The PCA (Figure 6) showed that the frequency of wedging rings is mainly correlated to the cambial age and diameter.

Three elements contribute together to the first axe: number of rings or cambial age (Nring), cumulative growth or diameter (Diam) and total number of wedges observed (see correlation in tables II). The variation of parameter of eccentricity (D) appears independent from the previous elements. The parameter D used express the ratio between basal area of the actual stem disc and the area calculated from the perimeter of a circular disc. As part of basal area measurements, the parameter D corresponds to the fractional basal area deficit as one minus the ratio of the basal area of the tree over the area of the circular disc with the same perimeter as the tree (Ngomanda et al., 2012). In this study, we used it

to express eccentricity of ring layers (polygons) because it depends on the form of stem disc. Nevertheless, it is possible to find another way of expressing the eccentricity.

In addition to ring irregularity described, the use of GIS tools enhanced the quantification of sap- and heartwood by digitalization around the entire cross-section. The sapwood and heartwood distinction was made possible by the contrast of colors. The results showed that the proportion of sapwood correspondson average, to a quarter of the heartwood (Table II) and the sapwood/heartwood ratio appears to decrease with increasing tree diameter. This information seems useful to quantify the proportion of non-durable wood and the proportions of wood by color (Richard, 2012). In this sense, it is important for future dendrochronological studies. The presence of sapwood is of key importance in the interpretation of tree-ring dates (Hillam, 1985). As the date of the outer ring is also the date of felling, our study gave information (average ring widths in sapwood) that can be considered if sapwood is not complete or clear. For all samples, the ring widths varied from very narrow to very wide, with an average of 3.1 mm. It is supposed that some of the trees must have grown under conditions that were limiting perhaps emanating from other trees. Globally, the obtained mean ring width appears normal under natural conditions with all the disadvantages (competition or dominance of neighbors) (Détienne, 1998; Nzogang, 2009). However, with the lack of sufficient data on environmental condition, it was

not possible to explain the variability of mean ring widths between trees sampled.

Future directions for research: annual periodicity and cross-dating

The cambial activity works within the constraints imposed by the environment, allowing for periodic growth and the formation of tree ring formation in regions with almost constant favorable growth (Hayden, 2008; Worbes, 2002). It has been demonstrated that many tropical forest trees form distinct or clear rings (Catinet, 1970; Mariaux, 1967; Stahle, 1999), but one of the limiting factors for dendrochronological work in tropical tree species is the lack of research on growth ring periodicity. The rhythm of growth is unknown for most of tropical tree species, especially in wet tropical forest. Even if a large fraction of tropical species possess distinct tree rings, growth rings may be formed annually, biannually or irregularly. Therefore, studies such as those recently done by Fétéké et al. (2016) and Tarelkin et al. (2016) are necessary to better the understanding of the periodicity of growth rings and the development of quantitative tree-ring studies.

Various methods are usually used to monitor growth rhythms in tropical trees: phenological investigations (Fétéké et al., 2016), wood anatomy, ring counting in trees of known age (De Ridder et al., 2014), periodic or cambial wounding (Détienne, 1976), radiocarbon dating (Worbes, 1989) and ring-width analysis or cross-dating (Nzogang, 2009). To our knowledge, cambial wounding has been used in some works (Détienne, 1976; Détienne et al., 1998) to analyze the periodicity of growth rings of *M. excelsa*. Détienne (1976) was the first to prove the annuality on tree rings of *M. excelsa*. Besides, the periodicity of formation of the tree rings of *M. excelsa* with four other deciduous tropical trees has also been confirmed by cross-dating (ring-width series vs rainfall series) in the tropical rainforest of Cameroun by Nzogang (2009).

In our study, the wood samples were taken in tropical semi-deciduous rainforest of region of Kisangani (Republic Democratic of Congo), where environmental conditions are similar to those of the mentioned research. We did not build chronologies of ring width series in this study, although *M. excelsa* seems to satisfy many conditions for successful dendrochronological studies (deciduousness, long-lived pioneer species, presence of marginal parenchyma band, cross-dating possibilities, etc.) (De Ridder et al., 2014). We expected that ring width series obtained through the Dendro-Gis tools could be successfully cross-dated. In the same region (semi-deciduous forest at Yangambi), De Ridder et al. (2014) have examined plant trees of known age and proved that *Pericopsis elata* forms tree rings annually despite the relatively short dry period.

For future dendrochronological studies, the annual periodicity of *M. excelsa* may be confirmed in the

context of study area by the methods such as the phenological investigations, examination of trees of known age and radiocarbon dating. The description of wood disc samples provided more information and helped to identify some problems related to the anatomy of wood structures of *M. excelsa* (false rings, wedging ring and missing rings) that could make cross-dating difficult (personal observation). The incidence of false and wedging that we observed emphasizes the necessity to use stem disc (wood disc) instead of core or bar (Latte et al., 2015) for dendrochronological studies. So, the use of discs seems more suitable than bars or cores for dendrochronology studies (Worbes, 1995; Trouet et al., 2006; De Ridder et al., 2014). Whichever wood sample are chosen, correct cross-dating requires good preparation of samples (great care with sanding methods), as much as it is possible, in order to increase the visibility of tree rings. The use of a large number of wood samples for good results is also recommended.

CONCLUSIONS AND PERSPECTIVES

This work proved that the use of Dendro-Gis tools can provide important information about ring description of tropical tree species, especially for *Milicia excelsa*. Despite tree-ring anomalies that we observed, this species could be used for tree-ring dating because its rings are clear and distinct, and it has been reported that its growth rings are annual. The use of stem disc (wood disc) instead of core or bar for dendrochronological studies is also indicated. The geometric information obtained by the use of Gis tools also offer interesting perspectives for many retrospective analyses. Deep analysis of additional data from this study and experimentation of the same approach on other tropical tree species are recommended.

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Supplementary Material

Overview of tree rings digitalized using scans for the five samples (Legend: ID of sample and diameter).

