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Variation analyses of extractive contents by NIR-spectroscopy bring out the differences between agroforestry and forestry walnut (*Juglans regia* × *nigra*) trees

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Abstract: Wood characteristics of trees grown in agroforestry systems are little studied, even if growth conditions are different from conventional stands. This work aimed to determine the impact of the agroforestry system on the heartwood formation process of hybrid walnut (*Juglans regia* × *nigra*) trees, especially the resulting extractive contents. Ethanol and water extractions were successively performed on wood samples taken across the diameter of the trunk of agroforestry (AF) and forest (FC) walnut trees to get the radial distribution of the extractive contents. All the samples were analyzed by NIR-spectroscopy and NIR-hyperspectral imaging. Statistical discriminant models were developed to classify the samples from both different forestry systems, according to their chemical composition. The results indicated no significant differences between the values of extractive contents of AF and FC walnut woods, whatever the radial position. At the intra-tree scale, the quantity of extractives does not increase significantly with the radial position. However, partial least squares-discriminant analysis (PLS-DA) regression models, developed with NIRS measurements, showed that significant chemical differences exist between AF and FC trees, especially for extractives composition and lignin content. This allowed to classify wood specimens from both forestry systems. These results were confirmed by hyperspectral camera analyses.

Keywords: discrimination models; extractives; heartwood; lignin; NIR-hyperspectral imaging; NIR-spectrometry.

1 Introduction

Agroforestry systems include a wide range of solutions using trees outside of forests (Campanhola and Pandey 2019). In such systems, the production of wood and/or woody biomasses is often combined with other products or services (Nair 2005). Agroforestry systems are competitive with intensive food and industrial crops for land use. Therefore, the sum of their products and services must offset the economic value of the others alternatives (Sollen-Norrlin et al. 2020). In addition, tree species in combination with agricultural crop may increase the employment generation for the nursery growers, local labors, wood industries, and other wood merchantpersons (Shukla et al. 2018). Agroforestry has a significant potential to provide employment to rural and urban population through production, industrial application and value addition ventures. In this sense, agroforestry practices could stratified the three following long-term strategic objectives, which are stated by the UE rural development policy: (i) fostering the competitiveness of agriculture, (ii) ensuring the sustainable management of natural resources, and climate action, (iii) achieving a balanced territorial development of rural economies and

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communities including the creation and maintenance of employment (Hain 2014). Agroforestry systems are thus critical for rural development by providing determinant ecosystem services such as soil fertility enhancement, prevention of soil erosion, water, wind, and pest regulation, and pollination (Gonçalves et al. 2021).

Properly planned and managed, agroforestry systems can meet these challenges. However, adequate and comprehensive knowledge is needed to make the right decisions, from selecting the most appropriate tree species/clones for a specific site to the most efficient harvesting systems (Muschler 2015). In this sense, the quite robust walnut tree fit perfectly within an agroforestry system, not least because of the relatively low competition for light with other crops due to the more open crown, the late leafing and early leaf fall. Its tree foliage also allows to protect the inter-row plants/crops against strong UV radiations, providing moderate shade and increasing humidity level (Kulasegaram and Kathiravetpillai 1976). In addition, walnut tree leaves are good litter, speeding up nutrient cycling (Pardon et al. 2020). Moreover, walnut trees are well known to produce timber with high commercial potential, providing high-quality wood to supply the market demand in Europe, especially *Juglans nigra* × *Juglans regia* hybrid which is the preferred species for wood material uses. Such an hybrid species produces high quality timber and veneer (100 m^3/ha after 50 years). Well-accepted by farmers, it is ideally suited for use in agriculture reconversion and diversification, in Europe. In addition, new industrial options have emerged and have been well-defined in Europe on the production of homogeneous light-colored wood, exploitable trees within 25–30 years, with large growth rings and low density without heartwood (Fernández-Moya et al. 2019).

Agroforestry could allow to improve wood quality by producing a wood material with wide regular rings (suited to the needs of industry), because the trees are not subjected to cycles of competition and thinning during their growing. Walnut is also well-known for its rich source of various valuable chemical compounds, which are involved in the wood quality. Walnut heartwood is classified as durable (Scheffer and Cowling 1966; Scheffer et al. 1998). This durability has been related to the presence of phenolic compounds such as flavonoids, naphtoquinones, and hydrolysable tannins (Gupta et al. 1972). In this respect, walnut tree can be used as an excellent source of various high value active compounds (mainly with antioxidant, antimicrobial and insecticidal potentials) (Salejda et al. 2016). The amount of extractives accumulated in tree stems can vary according to the vertical and the radial position in the stem. The heartwood is generally characterized by a higher extractive content than those of the sapwood portion (Beritognolo 2001). Within the stem cross-sections containing heartwood, the extractive amount increases according to the age of the xylem growth rings, reaching its maximum in the transition zone or in the heartwood periphery (Burtin et al. 1998; Magel et al. 1994).

The chemical composition of wood, including extractives content, can also vary according to several factors, among which environmental, genetic, age and seasonal variations (Haupt et al. 2003; Toshiaki 2001). Recent works have demonstrated that changes in the tree's environment during growth can alter heartwood extractives content in many species (Hillis 1971; Morais and Pereira, 2012). If such variations in heartwood extractives affect wood properties, then forestry treatments and changes in how forest resources are managed may affect the valorization of future wood products (Taylor et al. 2006).

Statistical modeling based on NIR spectrometry has revealed to be an efficient tool to assess the chemical and physical properties of a wide range of wood species, with high accuracy (Schimleck et al. 2003). This technic has been efficiently used for the determination of the chemical composition of branch wood, knots and bark fractions from walnut trees issued from Agroforestry and forestry systems (Heim et al. 2022; Terrasse et al. 2021). In addition, NIR-Hyperspectral imaging can be used for the development of models that quantify the heterogenic chemical composition of the wood surface (Sandak et al. 2016). Hyperspectral imaging provides spatially information of the wood surface in the NIR range, and then selected wavelengths can be used to reveal the presence of specific chemical compounds (Myronycheva et al. 2018).

This study aims to improve the knowledge about the chemical composition, especially the radial distribution of hydrophilic and lipophilic extractives content, within agroforestry (AF) walnut trees compared to those from standard forestry control plots (FC). In addition, the radial distributions of the extractive contents of agroforestry (AF) and forest (FC) hybrid walnut trees (*J. regia* × *nigra*), were used to evaluate the level of heartwood formation process depending on the silvicultural system. All samples (before and after ethanol and water extractions) were analyzed by NIR-spectroscopy techniques, in order to evaluate the impact of silviculture method on the chemical composition of wood and to develop discriminant models allowing the classification of wood samples according to their growing conditions; agroforestry (AF) and forest control (FC) plots.

2 Materials and methods

2.1 Experimental site

The experiment was carried out at the Restinclières farm area, located in southern France (43°420 N, 3°510 E and elevation 61 m). Two plots composed by 25 years old walnuts are studied: an Agroforestry plot (AF, with 140 walnuts) and a Forestry Control plot (FC, with 235 walnuts). Initially the planting density in both plots was identical. A thinning carried out in the in 2004, have reduced the density of the AF plot to 100 trees/ha and walnut trees were spaced 4, 8, 12 or 16 m on a same planting line. Between each line, winter cereal crops were cultivated, each year, on a 12 m wide strip. In FC plot, the walnut tree density was almost 200 trees/ha and were mixed with alder (*Alnus cordata*) and ash (*Fraxinus excelsior*) trees. In both plots, tree rows were north-south oriented Figure 1a and c shows the walnut trees distribution in AF and FC plots, respectively.

2.2 Tree selection

As shown in Figure 1a, 8 hybrid walnut (*J. nigra* × *J. regia cv. NG23*) trees were sampled from each of the two AF and FC plots. All of the selected 25 year old walnut trees (AF and FC) were harvested in February 2020, limiting the seasonal impact on wood chemical composition (i.e., starch in sapwood). For each plot, 6 trees were selected at the extremity of the plot to avoid the effect of soil and water exposure variations. In addition, 2 AF and 2 FC walnut trees were taken from the other side of their respective plot, in order to assess the influence of tree location within the plot area.

2.3 Sampling

The details of sampling, focused on one studied tree, are exposed in Figure 1b. A similar samples repartition was performed for the 16 walnut trees issued from AF and FC plots, as following described. For each walnut tree, a wooden disk of 30 mm in thickness was collected at 1.30 m from the ground. Due to the fact that the heartwood formation process of the harvested trees has not or only slightly initiated, the wood sampling was based on each tree diameter. In this sense, 10 samples [6 outer samples (references A, B, C and H, I, J) and 4 inner samples (references D, E, F, G) – Figure 1] were cut out each disk, according to the radial direction (north–south axis). The section of theses samples was 1/10 × 1/5 of the tree diameter (R × T, in mm), and the thickness was 30 mm (L) (Figure 1). According to this method, the samples dimensions were 9 × 18 × 30 mm^3 (R × T × L) for the smaller tree diameter and 13.5 × 27 × 30 mm^3 (R × T × L) for the larger one. In average, the number of annual rings of each samples was ranged between 2 and 3. The total number of samples, including AF and FC samples, used for this study was 160. All of these 160 samples were then oven dried at 103 ± 2 °C for 24 h, before being grounded and passed through different sieves to obtain particles with sizes ranging between 0.2 and 0.5 mm. Before NIR-S analyses (ETH) and extraction processes, 15 g of each sample modality was stabilized at a moisture content (MC) of 12% (stabilization in an humid air at 20 °C ± 2 °C and 65% ± 5% RH).

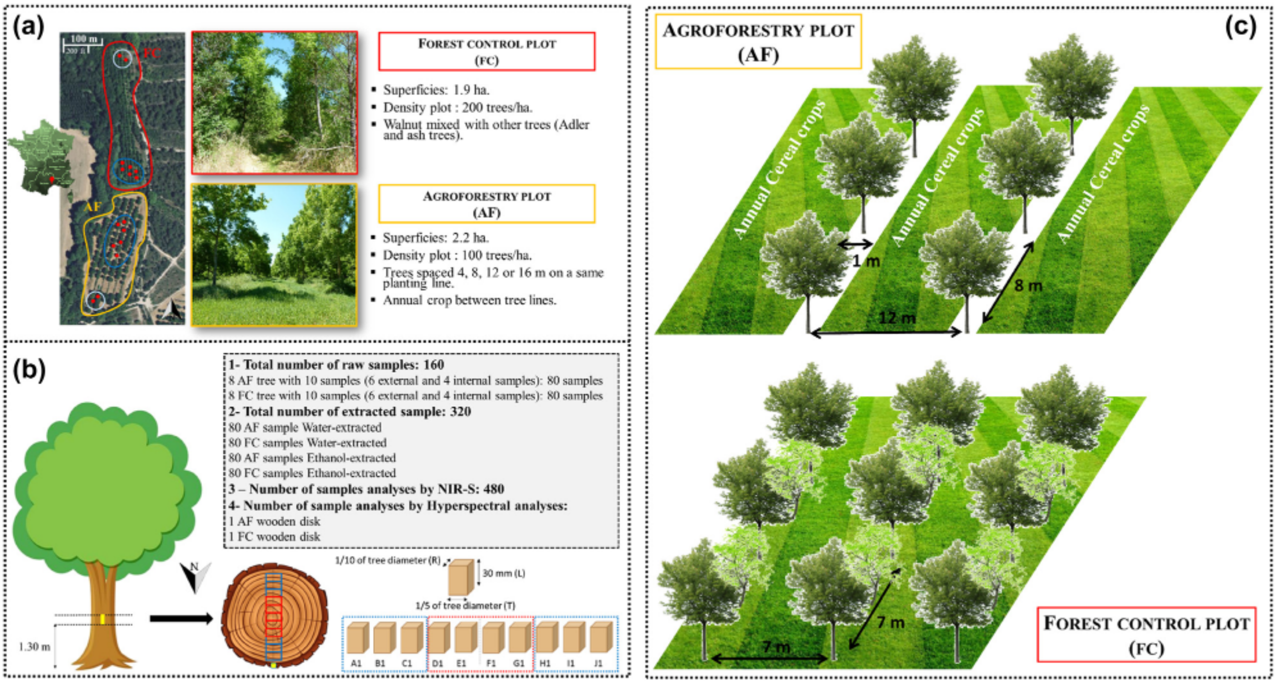


Figure 1: (a) Walnut trees selection in the agroforestry (AF) and forestry control (FC) plots at the Restinclières farm in southern France. (b) Sampling and selection of wood samples for extraction process, NIRS and LC-MS analyses. (c) Graphical drawing plan of walnut trees distribution in AF and FC plots.

2.4 Extraction processes

All experimental procedures used in the determination of the extractive content were adapted, with minor modifications, from procedures found in the scientific literature (Rowell et al. 2005).

Each grounded wood sample was oven-dried at 103 ± 2 °C in order to determined its initial anhydrous mass (m_0) and then 15 g (in dry basis) was extracted in a Soxhlet apparatus with ethanol [HoneyWell, Germany, 32201-M] during 6 h. After this first extraction step, the sample was oven-dried 103 ± 2 °C until mass stabilization (m_1), before being submitted to a second extraction using hot distilled water for 8 h, and then dried at 103 ± 2 °C to obtain the anhydrous mass (m_2).

Extractive contents were determine according the following equations Eqs. (1) and (2):

$$E_{\mathrm{eth}}\,(\%DW) = [(m_0 - m_1) \div m_0] \times 100 \tag{1}$$

$$E_{\mathrm{water}}\,(\%DW) = [(m_1 - m_2) \div m_0] \times 100 \tag{2}$$

where E_{eth} and E_{water} are the sample extractives content using ethanol and water, respectively in percentage of dry weight (DW), m_0 is the theoretical anhydrous mass of the test piece before extraction, m_1 and m_2 are the anhydrous mass after the ethanol and water extractions, respectively.

A total of 320 powder samples were characterized for their hydrophilic (water extraction) and lipophilic extraction (ethanol extraction) rate (Figure 1b). The measurement error on the extractive contents determination was estimated at ±0.04 (% *DW*).

2.5 NIR-spectrometry measurements

2.5.1 Hyperspectral camera analyses: Firstly, hyperspectral measurements were performed on the wooden discs, previously sanded to obtain a surface without irregularities avoiding scattering effect during NIR-S measurements (Mancini et al. 2019). The camera was a Specim FX17 (Specim, Spectral Imaging Ltd.) with a spectral range of 933–1721 nm and a 3.5 nm increments. Each NIR spectrum was digitized in 224 wavelengths. The spatial resolution was set to 0.27 mm/pixel. For an image, spectral data were collected as a 3D matrix of 640 × 1002 × 224 values. The dimensions 640 × 1002 were associated with the spatial dimensions.

Images from hyperspectral camera analyses corresponding to absorbance spectra were collected at the wavelength of 1450 nm, attributed to first overtone O–H stretching vibration of lignin/extractives compounds. Then, these images were analyzed with Image J 1.53k software in order to determine the values of pixels for each colors in the RGB referential.

2.5.2 NIR-S analyses on powder samples: Near infrared spectra were obtained on raw and extracted powder samples previously stabilized at a moisture content (MC) of 12% (stabilization in an humid air at 20 °C ± 2 °C and 65% ± 5% RH). A MicroNIR OnSite-W (VIAVI Solutions Inc.) spectrometer was used in reflectance mode. Data were measured for wavelengths between 908 and 1676 nm in 6 nm increments. The spectra were thus composed by 125 wavelengths of absorbance values.

2.6 Statistical analysis

Statistical analyze were led with RStudio Desktop software (v 1.2, 2019, RStudio Inc.). The comparison of extractives contents obtained after ethanol and water extraction between both AF and FC plots was performed with a Wilcoxon test. To understand the radial repartition of the extractive contents measured within walnut trunk from AF and FC walnut trees, Kruskall–Wallis tests have been applied on each potential explicative factor and response variable. For each test indicating an effect of a factor on a studied variable, Wilcoxon tests were used to investigate the nature of this effect and the significance of the differences between the groups created by the modalities of the descriptive variables.

NIRS spectra (from Specim FX17 and MicroNIR) were first transformed (Naes et al. 2004) with a standard normal variate (SNV) correction to reduce the effect of irregularities on surface and the intra spectrum variability (correction of the light dispersion). The second derivative was then computed using the algorithm of Savitzky Golay with a smoothing range of 11 data points and a third degree polynomial (Savitzky and Golay 1964). This derivative allowed separating peaks that overlap and correcting the baseline deviation of spectra. The mathematical corrections were performed using the package "prospectr" (v 0.1.3, 2013).

NIRS models were developed with the partial least squares-discriminant analysis regression method (PLS-DA, package "mdatools", v 0.11.5, 2021) to discriminate the samples as AF or FC (highlighting differences in chemical composition). For this purpose, raw samples (ETH), samples after ethanol extraction (AVE) and after water extraction (APR) were used separately.

3 Results and discussion

3.1 Radial distribution of extractive content within AF and FC trees

As shown in Figure 2a, the global water extractive contents of AF and FC samples were slightly higher than those from extraction using ethanol, highlighting that studied walnut trees contain more hydrophilic extractives than lipophilic extractive compounds (Kebbi-Benkeder et al. 2015a; Vek et al. 2014). For both solvents, the extractive contents between the two plots were not statistically different (p-value = 0.3958 for ethanol and p-value = 0.0649 for water). The relatively low extractive contents recorded for AF (1.99% with water and 1.92% with ethanol) and FC (2.42% with water and 1.92% with ethanol) walnut trees, compared to results from other past scientific study [6.3% in sapwood and 13.3% in heartwood of 41 years old common walnut, by successive solvent extractions (Kebbi-Benkeder 2015b), indicate that the trees studied are relatively young (25 years) and that the wood is still in the early stages of its heartwood formation process and is mainly composed of sapwood. Even if Figure 2b shows that ethanol-extracts level trends to increase from bark to the pith of the AF and FC walnut trees. This tendency is statistically significant (p-value < 0.001), samples located in the center of the trunk having a higher rate of extractives than those in the periphery. Concerning the radial distribution of water-extractives within AF and FC walnut trees, water-extractive contents are quite

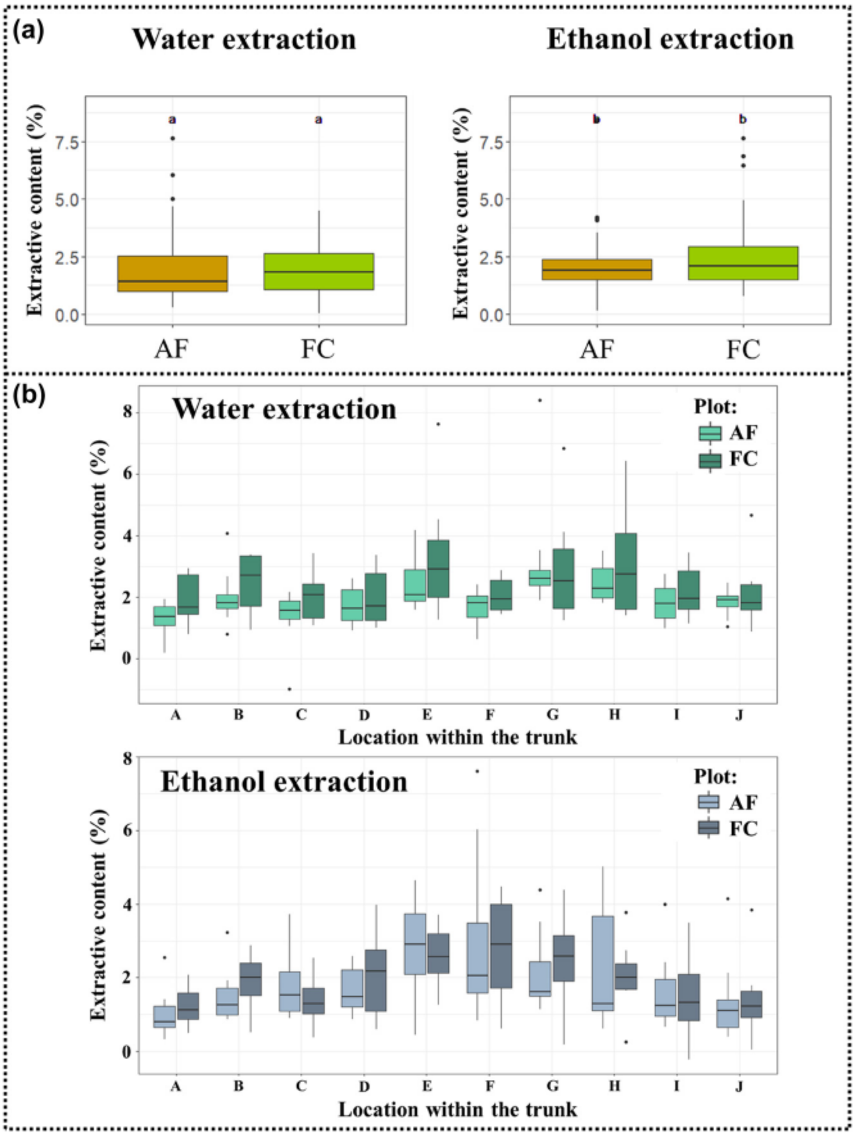


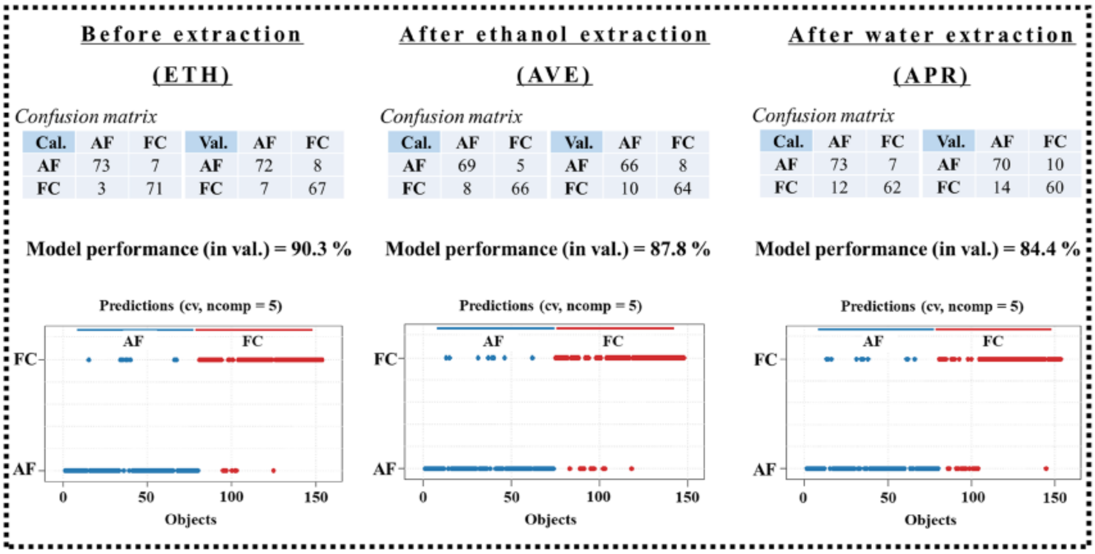
Figure 2: (a) Comparison between mean extractive contents from agroforestry (AF) and forestry control (FC). (b) Radial distribution of mean extractive contents of walnut wood from AF and FC systems.

similar whatever the radial position of the wood samples (Figure 2b) (*p*-value = 0.1385). These results were confirmed by Wilcoxon tests, regrouping the samples from A-B-C-H-I-J and D-E-F-G.

3.2 Differences in extractives contents between AF and FC trees

PLS-DA (with cross validation "leave-one-out") were carried out for developing models for classifying wood samples according to the forestry system. As shown in Figure 3, the models presented a large percentage of correct classifications (accuracy) for FC and AF raw (ETH), ethanol-extracted (AVE) and water-extracted (APR) wood specimens. The comparison between calibration matrix (Cal.) and validation matrix (Val.), presented in Figure 3 shows that the models were stable whatever the extraction modality (ETH, AVE, APR). Even if the performance of the models (accuracy in validation) decreased according to the successive extraction processes, from 90.3% (ETH), to 87.8% (AVE) and to 84.4% (APR), the AF/FC samples discrimination remained possible with 84.4% of the samples well classified at the end of the extraction processes.

The aim of distinguishing wood samples from AF and FC walnut trees was promising. The results obtained from PLS-DA showed that chemical differences existed between AF and FC samples, and these differences in composition (even low) was sufficient to distinguish wood specimen from both forestry systems. Previous study conducted on various tree organs (bark, knots and branch woods) of



Before extraction (ETH)

Cal.	AF	FC	Val.	AF	FC
AF	73	7	AF	72	8
FC	3	71	FC	7	67

After ethanol extraction (AVE)

Cal.	AF	FC	Val.	AF	FC
AF	69	5	AF	66	8
FC	8	66	FC	10	64

After water extraction (APR)

Cal.	AF	FC	Val.	AF	FC
AF	73	7	AF	70	10
FC	12	62	FC	14	60

Figure 3: Discriminant analyses (PLS-DA) classifying samples from AF and FC trees, before extraction (ETH), after ethanol extraction (AVE) and after water extraction (APR), with their respective model's performances.

Walnut (*J. regia* × *nigra*) highlighted that PLS-DA models based on treated NIR signatures are robust (with accuracy ranging between 75 and 91.4%) for distinguish wood, knots and bark specimens from different forestry systems (agroforestry and traditional forest control) (Heim et al. 2022). Other past works, based on NIRS combined to PLS-DA models, were conducted to classify efficiently (with accuracy ranging from 86 to 100%) *Eucalyptus* wood specimens from natural and planted forest (Ramalho et al. 2018), and to classify and identify twelve native wood species from homogeneous plantations, with good accuracy (93.2%) (Pace et al. 2019).

3.3 Chemical differences between AF and FC samples by NIR-spectrometry analyses

The above results from PLS-DA models highlighted that ethanol and water extractive compounds only contributed to approximately 5.9% [90.3% (ETH)–84.4% (APR)] in the AF and FC samples discrimination. In this sense, other extractive compounds (by using other solvents, or components highly bound to the cell wall) or the composition in macromolecules constituting the wood cell wall play an important role in the differences in chemical compositions between AF and FC walnut woods. This statement is in agreement with the results obtained by NIR-spectroscopy analyses, presented in Figure 4 and Table 1.

Figure 4a and b show the regression coefficients of the 1st component form PLS-DA analyses and the difference in average NIR spectra, respectively, of ETH, AVE and APR walnut wood samples from forestry (FC) and agroforestry (AF). Table 1 indicates the NIRS absorption bands normally associated with the main chemical components contained in the wood specimens.

It appeared clearly that the difference in chemical composition between raw (ETH), ethanol extracted (AVE) and water extracted (APR) from AF and FC systems depends not only on extractives compounds (peaks 1, 7 and 11) but also on hemicelluloses (peaks 5 and 6), cellulose (peaks 4, 8, 9 and 10) and lignin (peaks 2, 3,7). Figure 4b highlights that the main differences in chemical composition of AF and FC wood could be also explained by the contents in lignin (peaks 3 and 7) and hemicelluloses (peaks 5 and 6). According to the literature, lignin content is affected in wood formed under gravitropic stimulation or mechanical stress (Novaes et al. 2010; known as reaction wood). In addition, significant effects of eucalyptus plant spacing in insoluble lignin (increases with lower stand density) and hollocellulose (decrease with lower stand density) content in wood were observed by Rocha et al. (2016), while extractives and soluble lignin contents were not significantly affected. The hollocellulose variation in wood is mainly due to the hemicelluloses content than those of cellulose, this latter being quite constant whatever the hardwood stand population density (Nasser 2008). The competition among plants for light is much more intense in smaller spacing due to the need of the trees to expand their leaf surface for carbon assimilation. Thus, the general trend of increasing insoluble lignin in wood can be explained by greater increase in height of trees cultivated at larger spacing with expected greater percentage of

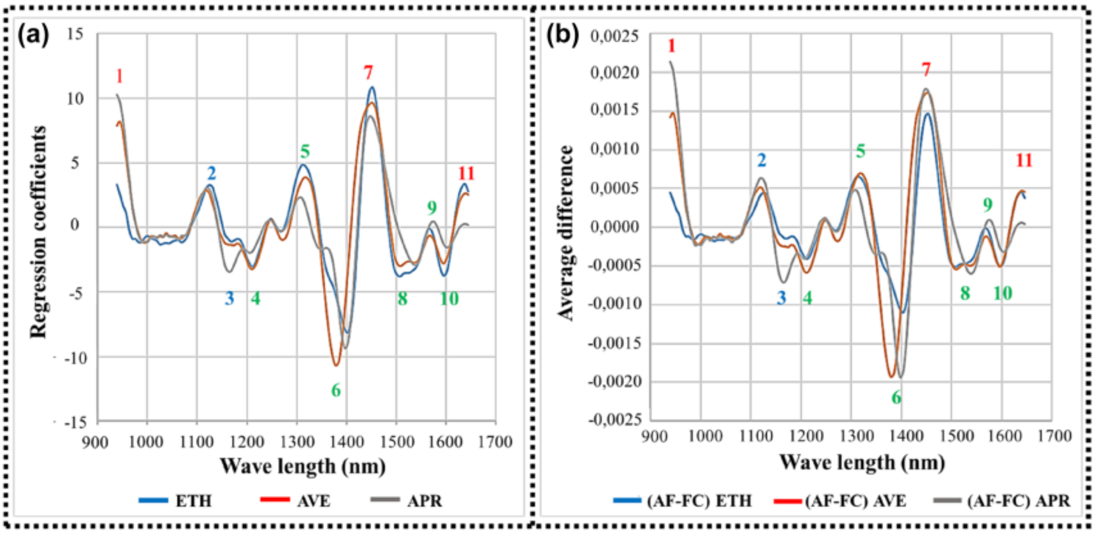


Figure 4: (a) Regression coefficients of the 1st component from PLS-DA analyses and (b) difference in average spectrum of walnut wood samples from forestry (FC) and agroforestry (AF) systems before extraction (ETH), after ethanol extraction (AVE) and after water extraction (APR).

Table 1: NIRS absorption bands normally associated with the main wood components (cellulose, hemicelluloses, lignin and extractives) contained in the wood specimens. Index numbers (in colour) relate to the specific band in Figure 4.

Index	Wavenumber bands (nm)	Bond vibration	Structure	Remarks	References
1	900–980		Lignin extractives	The major vibrations include the yellow–brown color of the wood that are primarily due to the presence of lignin and extractives	Kelley et al. (2004), Yi et al. (2017)
2	1100–1150	Second overtone C–H stretching of CH_3 groups	Lignin	CH_3 groups and aromatic moieties	Workman and Weyer (2007), Schwanninger et al. (2011)
3	1150–1200	Second overtone asym. C–H, HC=CH stretchings	Lignin		Kelley et al. (2004), Schwanninger et al. (2011)
4	1200–1220	Second overtone C–H stretching	Cellulose	Two to three bands t.a. CH and CH_2 groups, cellulose	Schwanninger et al. (2011)
5	1290–1330	First overtone C–H stretching + C–H deformation	Hemicelluloses all	Tentative assignment to CH_3 groups in acetyl ester groups in hemicelluloses and lignin and all wood components after acetylation	Schwanninger et al. (2011)
6	1350–1400	First overtone C–H stretching and C–H deformation	Hemicelluloses all	Tentative assignment to CH_3 groups in acetyl ester groups in hemicelluloses (normal wood) and all wood components after acetylation	Schwanninger et al. (2011)
7	1420–1460	First overtone O–H stretching	Lignin extractives	Vibration of phenolic hydroxyl groups	Schwanninger et al. (2011)
8	1500–1550	First overtone O-H stretching	Cellulose	Semi-crystalline region of cellulose	Fujimoto et al. (2007), Schwanninger et al. (2011)
9	1570–1600	First overtone O–H stretching	Cellulose	Crystalline region of cellulose in C_1 and C_2	Tsuchikawa and Siesler (2003), Schwanninger et al. (2011)
10	1600–1610	First overtone O–H stretching	Cellulose	Strongly H-bonded O–H group in cellulose	Schwanninger et al. (2011)
11	1610–1650	First overtone C–H stretching	Extractives		Schwanninger et al. (2011)

Index numbers (in colour) relate to the specific band.

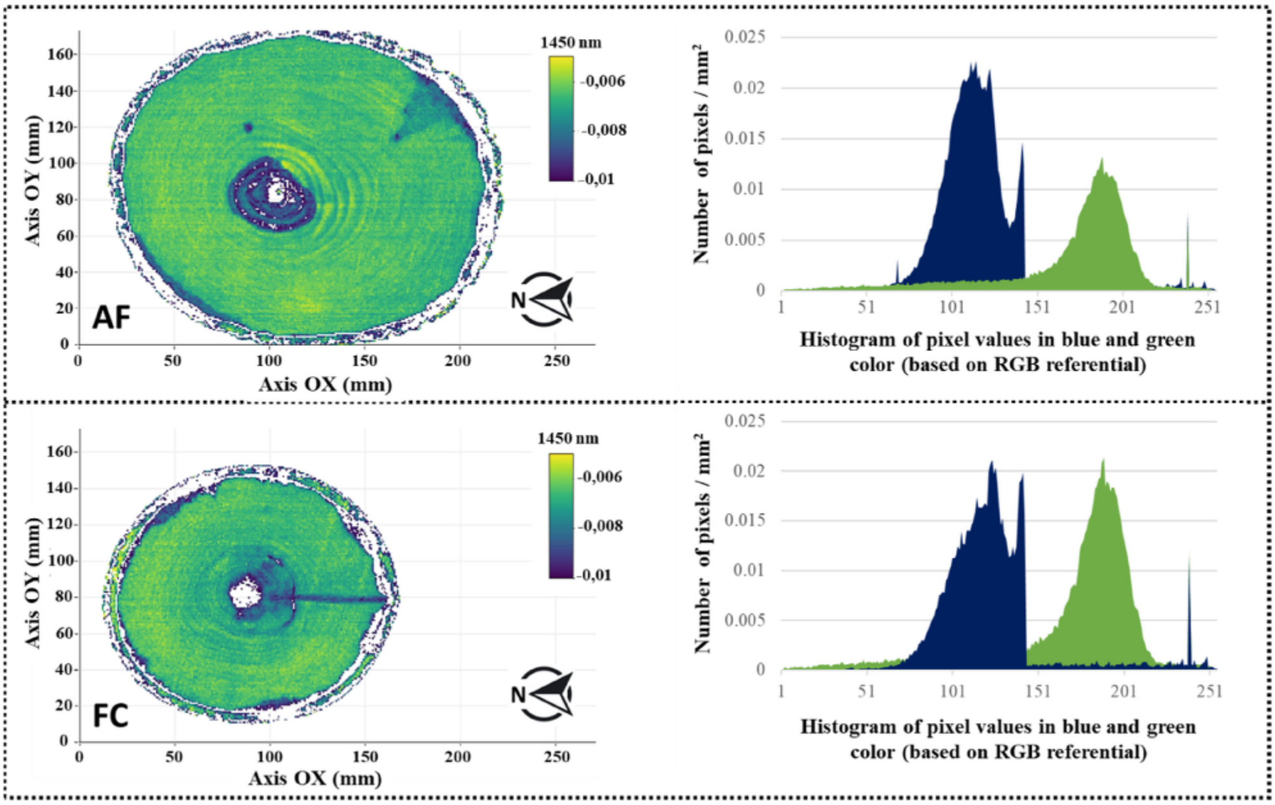


Figure 5: Illustrations of NIR absorbance after pretreatment (SNV correction – 2nd derivative) measured at the wavelength of 1450 nm. These images were acquired from agroforestry (AF) and forestry control (FC) slice of air-dried wooden disks, and they ware associated to their histograms of pixels values in blue and grey color (based on RGB referential). On the left, the color scale ranged from yellow to dark blue represents the intensity of NIR absorbance at the wavelength of 1450 nm, attributed to first overtone O-H stretching vibration of lignin/extractives compounds (Table 1). On the right, the histograms were issued from the analyses of the wooden disc pictures from hyperspectral camera, using Image J software in order to determine the level of blue pixels representative of lignin/extractives contents.

juvenile wood in its trunk compared with trees at smaller spacing. Juvenile wood has higher lignin content compared with mature wood (Zobel and Van Buijtenen 1989). These chemical composition difference between AF and FC woods could also be explained by the thinning treatments performed in the AF plots. Such silvicultural treatments improve tree growths and delay wood transition from juvenile to mature. Juvenile wood has thin cell wall, shorter fiber length and higher lignin content (Uner et al. 2009). Kramer and Kozlowski (1979) highlighted that wood extractives were products of metabolic tree growth. Hence, forestry treatments, which increase tree growth and vigor, could increase the extractive content in wood or modified their chemical composition (Heim et al. 2022).

3.4 Hyperspectral camera analyses

Images from hyperspectral camera analyses, presented on Figure 5, refer to the absorbance spectra collected at the wavelength of 1450 nm, which is attributed to first overtone O-H stretching vibration of lignin/extractives compounds. This wavelength was chosen due to its most discriminant behavior between AF and FC samples, among all the NIR-S analyses carried out previously on wood powder. The hyperspectral images associated to their respective blue and green color intensities, presented in Figure 5, clearly highlight that AF disk samples contained more lignin/extractives components (high intensity in blue color) than those of FC samples. These results are in agreement with the previous statements about the difference in chemical composition between AF and FC woods.

4 Conclusions

This study provides pioneer knowledge on woods formed in agroforestry systems, which are still very under-studied by now. The results highlight the radial evolution in extractives content within a tree and the variability of the chemical characteristics of wood from agroforestry (AF) hybrid walnut compared to the same species grown in forestry systems (FC). Extractives contents of AF and FC wood specimens are not significantly different. The ethanol and water extractives contents are distributed in a constant way in the radial cross section of the tree, for both AF and CF. However, PLS-DA models developed with NIRS measurements showed that chemical differences exist between AF and FC samples, before and after the two steps extractions processes. These chemical differences are due to the extractive contents but also to the lignin and hemicelluloses distributions in AF and FC wood samples. Such chemical variation (even low) is sufficient to distinguish wood specimens from the two different forestry systems. Hyperspectral imaging allows confirming these tendencies and that AF samples contained more lignin/extractives compounds than FC samples. The methods and results of this work provide a new angle to understand the wood properties of walnut tree growing under different growing conditions allowing to classify the wood quality.

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