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## ABSTRACT

1  
2 The addition of commercial oak wood as copigments sources has been used to get high  
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4 quality red wines in the last years. During the contact time, different oak compounds  
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6 such as phenolic compounds are released from the wood to the wine, thus affecting its  
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8 organoleptic properties such as aroma, color or astringency. Wood processing for the  
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10 elaboration of barrels generates by-products which also seem to be interesting in the  
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12 oenological field due to its potential capacity of release high added value compounds  
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14 for wine.  
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19 Determining phenolic compounds of wood and its extractability to the hydroalcoholic  
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21 medium is important in the oenological industry. The method proposed in this study  
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23 cope with this issue in an *in situ*, non-destructive and fast way. For this purpose, a  
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25 number of oak by-product samples spectrally representative have been selected.  
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27 Selected spectral data have been correlated with oak wood extractable polyphenols  
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29 (extractable total phenolic content and extractable ellagitannin content) by modified  
30  
31 partial least squares regression (MPLS) using a number of spectral pre-treatments.  
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35 The obtained results are comparable with those obtained using bench top devices and  
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37 presents the advantage of its eventual friendly use out of lab. Development of applicable  
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39 models *in situ* will allow a greater versatility and efficiency for the decision-making in  
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41 the winemaking process on the adequacy and/or dosage of these byproducts according  
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43 to the requirements of the wine. The use of cooperage by-products as source of  
44  
45 copigments for wine leads to a sustainable and competitive cooperage industry, through  
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47 waste reduction and by-product valorization.  
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53 **Keywords:** red wine, oak byproduct, phenolic compounds, ellagitannin, MICRO-NIRS  
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## Introduction

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2 In grape berries, the balance between technological and phenolic parameters is a key  
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4 factor for obtaining quality red wines, especially in terms of their color. Anthocyanins  
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6 are the main responsible for the colour of red wines, and their interactions with other  
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8 phenolic compounds, called copigments, allow improving the color stabilization of aged  
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10 wines by copigmentation or stabilization reactions (Boulton 2001; Escribano-Bailón and  
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12 Santos-Buelga 2012). However, grapevine berries are commonly harvested on the base  
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14 of technological maturity parameters often without considering properly phenolic  
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16 maturity. Under warm climatic conditions, it is a usual pattern that grapes have high  
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18 sugar content but insufficient levels of phenolic compounds (pigments and copigments).  
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20 Wines made from these grapes are not able to form much copigmentation and as a  
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22 result, the color stabilization does not correctly develop and fall of color normally  
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24 occurred during winemaking maturation, especially when they are subjected to ageing  
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26 process (Boulton 2001; Ribéreau-Gayon et al. 2006).  
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34 The addition of commercial oak wood (Del Álamo et al. 2008; Nevares et al. 2008) or  
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36 wood compounds (Balík et al. 2017) have been aimed at implementing the production  
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38 of high quality red wines in the last years. During the contact time, different types of  
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40 oak compounds are released from the wood to the wine, thus affecting its organoleptic  
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42 properties such as aroma, color or astringency. Among these compounds, ellagitannins,  
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44 with several hydroxyl groups, can take part in oxidation reactions (Vivas and Glories  
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46 1996; Ignacio García-Estévez et al. 2017) that may favor the polymerization reactions  
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48 between flavanols and between flavanols and anthocyanins. Furthermore, they can  
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50 directly react with these types of compounds giving rise to flavanol-ellagitannins or  
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52 anthocyano-ellagitanins (Chassaing et al. 2010; Ignacio García-Estévez et al. 2013; I.  
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54 García-Estévez et al. 2013). Thus, ellagitannins may be involved in the changes of  
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1 colour during maturation and ageing of wine, helping to improve colour stability and  
2 also protecting it against oxidation.  
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4 Wood processing for the elaboration of barrels generates by-products that seem to be an  
5 interesting product in the oenological field due to its potential capacity of release high  
6 added-value compounds for wine. Taking this into account, the use of barrel-shoot  
7 wastes as a source of copigments for wine could become a sustainable and profitable  
8 alternative to get high quality red wine for the oenological industry and to disposal of  
9 their byproducts for the cooperage industry. In this sense, other products of the  
10 oenological industry, as the grape pomace have been characterized for food waste  
11 valorization (Páscoa et al. 2014).  
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23 Predicting phenolic compounds of wood and its extractability from the matrix to the  
24 hydroalcoholic medium could be important in the oenological industry. Accordingly,  
25 chemical techniques, such as chromatography and mass spectroscopy, have been used in  
26 several studies focused on evaluate the oak wood phenolic extraction process and how  
27 is the extractability affected by factors as oak toasted degree or the type of aging  
28 performed (Chira and Teissedre 2013; I. García-Estévez et al. 2015; Jourdes et al.  
29 2011). These studies achieve very accurate results but they used destructive and time  
30 consuming analyses that also require the use of chemical reagents. It is undesirable  
31 when the work flow is relatively fast.  
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46 Vibrational spectroscopy techniques represent emerging analytical procedure, which are  
47 enjoying increasing popularity in the industry as non-destructive, environmental  
48 friendly and rapid technique. The different fields of application of these techniques  
49 highlight its enormous versatility. In the oenological sector, the application of infrared  
50 spectroscopy is well known and extended. Classic NIR spectroscopy has been used to  
51 determine parameters in grapes such total polyphenols, extractable anthocyanins,  
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1 concentration of sugars, density (Kemps et al. 2010), total anthocyanins (Cozzolino et  
2 al. 2006) and the main families of phenolic compounds (Ferrer-Gallego et al. 2011;  
3 Ferrer-Gallego et al. 2010). In previous works carried out in our laboratory, near  
4 infrared hyperspectral imaging has been used to develop screening methods in order to  
5 measure phenols concentration in grape or grape seeds (Hernández-Hierro et al. 2013;  
6 Nogales-Bueno et al. 2015; Rodriguez-Pulido et al. 2014). In a further step,  
7 spectroscopic methods based on near-infrared spectroscopy have also shown great  
8 potential for analyses of wood for oenological purposes. Giordanengo et al. (2009) used  
9 classic NIR spectroscopy for the screening of extractable polyphenols in staves used to  
10 manufacture barrels oak wood. Its use was then extended for the selection of oak for  
11 oenology. So, Michel et al. (2013) carried out a validation of the aforesaid method by  
12 means of the determination of ellagitannin content and organoleptic properties of wines  
13 aging in wood barrels classified by NIRS. Finally NIR hyperspectral imaging has been  
14 also used for the screening of the extractable polyphenolic compounds (extractable total  
15 phenolic content) in wood cooperage byproduct by Baca-Bocanegra et al. (2018).

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36 However, despite the fast and effective proficiency of near-infrared spectroscopy to  
37 predict different parameters in wine sector, most of these studies carried out *at lab*  
38 imply sample transportation. To solve this problem and fulfill other sophisticated  
39 conditions for its application in the industry, new generation of portable/handheld NIR  
40 spectrometers have been developed within the last years. These systems incorporate the  
41 analytical precision for chemical identification and quantitation with a spectral  
42 resolution equivalent to bench-top instruments, allowing for flexibility for in-plant  
43 analysis since the unit can be easily carried and transferred. In the agro-food industry  
44 the potential of this technique has been highlighted for assessing the quality of fruits  
45 nondestructively in the field under different weather conditions (Abu Izneid et al. 2014;  
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1 Antonucci et al. 2011; Camps et al. 2012). In the viticulture sector, portable NIR  
2 devices have been implemented for assessing and monitoring grape ripeness directly in  
3 the field. Phenol ripening parameters such as the concentration of anthocyanins and  
4 polyphenols have been evaluated (González-Caballero et al. 2012; Guidetti et al. 2010;  
5 Larrain et al. 2008). However, references for the use of this technique in the analysis of  
6 oak wood for oenological purposes have not been found. The development of applicable  
7 models *in situ* will allow a greater versatility and efficiency for the real time decision-  
8 making in the vinification process on the adequacy and / or dosage of these byproducts  
9 depending on its spectral features and extractability properties.

10 The aim of this study was to develop a rapid and reliable method using a portable NIR  
11 spectrometer for the screening of extractable polyphenols (extractable total phenolic and  
12 ellagitannin contents) in cooperage byproduct. To our knowledge, this is the first time  
13 that portable micro near infrared spectroscopy technology has been applied to face the  
14 aforementioned goals.

## 15 **Material and methods**

### 16 *Samples*

17 American non-toasted oak (*Quercus alba* L.) shavings, cooperage byproduct, were used  
18 for this study. Samples were provided by Tonelería Salas S.L. (Bollullos Par del  
19 Condado, Huelva, Spain) and were collected as previously reported in Baca-Bocanegra  
20 et al. (2018). Wood staves were seasoned under natural conditions in the open air during  
21 24 months approximately before being used in the process of making barrels. In order to  
22 achieve representative sample sets, the samples were taken at 4 different points in the  
23 process of making barrels, so four samples were collected at each date (A, B, C and D).  
24 Samples belonging to A, B and C groups are generated by different sawing processes of  
25 the staves but always in the longitudinal direction of the fibers. In detail, an automatic



1 or manual processing of the staves is the only difference between the samples A and B  
2 respectively. With respect to the group D, the samples are the result of a processing of  
3 the staves in the transversal direction of the fibers. All sampling points are pre-molded  
4 and pre-toasted of the staves. Samples were collected periodically, between June of  
5 2015 and January of 2016. Two hundred samples were collected during the aforesaid  
6 period. Upon receipt, the samples were sieved and subjected to the spectral analysis.  
7 Wood shaving samples were screened using 2 mm and 10 mm mesh sieves placed in  
8 tandem. Only shaves of each sample which were retained between them were taken into  
9 account in the assay. After that, wood shavings were placed in stoppered plastics bags  
10 and stored in a dry chamber until use.  
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#### 23 *Spectral data acquisition.*

24 The spectral reflectance data were collected using MicroNIR Pro Lite 1700 device  
25 (VIAVI, Santa Rosa, California, USA), an instrument designed to measure diffuse  
26 reflectance in the NIR region of the electromagnetic spectrum. The MicroNIR owes its  
27 small size to the novel thin-film linearly variable filter (LVF) used as the dispersive  
28 element. The LVF is directly coupled to a linear detector array (128-pixel uncooled  
29 InGaAs photodiode array), covering the spectral range between 908 and 1676 nm  
30 (spectral resolution of 6.2 nm). The filter coating in the LVF is wedged in one direction  
31 and as a result of the varying film thickness; the wavelength transmitted through the  
32 filter varies linearly in the direction of the wedge. The LVF makes each pixel of the  
33 detector respond to a different wavelength. This ultra-compact spectroscopic engine is  
34 coupled with a tungsten lamps diffuse illumination system. An image of the MicroNIR  
35 spectrometer and MicroNIR's optical designed, adapted from VIAVI user manual, is  
36 provided in Online Resource 1.  
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1 Spectra were recorded using 9.3 ms as integration time and 100 as scan count using the  
2 instrument acquisition software MicroNIR™ Pro v.2.2 (VIAVI, Santa Rosa,  
3 California, USA). A two-point reflectance calibration was used. A spectralon® ceramic  
4 tile was used as a white reference (100% reflectance), whereas dark current (0%  
5 reflectance) was recorded by taking a measurement placing the device about 0.5 meters  
6 from any object.  
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10 Six replicates spectra were recorded for each sample and were exported in Log (1/R)  
11 units and subsequently the average of the replication spectra was calculated. The  
12 procedure was repeated for each sample and the obtained spectra were combined into  
13 the spectral matrix.  
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#### 24 *Chemical analysis*

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26 Total phenol and ellagitannin extractabilities were determined for oak wood samples. In  
27 order to perform this determination, wood shavings were immersed in a model wine  
28 hydroalcoholic solution (4 g L<sup>-1</sup> tartaric acid, 12.5% ethanol, adjusted at pH 3.6 with  
29 NaOH 0.5 M) for a maceration period of 72 h at room temperature and without  
30 agitation. Oak wood shavings were added to the wine-like solution in a 4 g L<sup>-1</sup> ratio.  
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39 This supernatant was used in all of the following reference analysis.  
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41 Extractable total phenolic content was determined using the Folin-Ciocalteu method  
42 (Singleton and Rossi 1965). The absorbance was recorded at 765 nm after 2 hours of  
43 reaction. This measure was performed in duplicate on an Agilent 8453 UV-visible  
44 spectrophotometer (Palo Alto, USA), equipped with diode array detection (DAD).  
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49 Gallic acid was used as a standard for construction of the calibration curve and the  
50 concentration of total phenols was expressed as gallic acid equivalent in mg g<sup>-1</sup> of  
51 wood.  
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58 Extractable ellagitannin content (castalagin, vescalagin, grandinin, and roburin E) were  
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determined using the HPLC-DAD-MS method presented by I. García-Estévez et al. (2012). HPLC analyses were performed in a Hewlett-Packard 1100 series liquid chromatograph (Agilent Technologies, Waldbronn, Germany). Detection was carried out at 250 nm as the preferred wavelength. Spectra were recorded from 220 to 600 nm. MS detection was performed in an API 3200 Qtrap (Applied Biosystems, Darmstadt, Germany) equipped with an ESI source and a triple-quadrupole linear ion trap mass analyzer that was controlled by Analyst 5.1 software (Applied Biosystems, Darmstadt, Germany). All analyses were performed in duplicate. Standards of castalagin, vescalagin, grandinin, and roburin E were extracted from *Q. petraea* oak chips and purified as it is described by García-Estévez and co-workers (I. García-Estévez et al. 2010).

#### *Data analysis*

An unsupervised pattern recognition technique, principal component analysis (PCA), was used in order to provide information about the latent structure of spectral matrix and to find spectral differences among all spectral samples. This method provides not only information related to spectral outliers and the distribution of samples in the newly-created space, but is also an important source of knowledge with which to create cross-validation groups used in the calibration process (Brereton 2003; Shenk and Westerhaus 1995). PCA was also used to select representative samples from the spectral data set. Mahalanobis distances (H) for each sample were calculated and samples were grouped according their neighborhood H values (NH).

Different spectral pretreatments were tested to eliminate effects of unwanted signals of spectra and also to enhance subtle differences between different samples. Scattering effects were removed using standard normal variate (SNV), multiplicative scatter correction (MSC) and detrending (Geladi, MacDougall, & Martens, 1985; Dhanoa,

1 Lister, & Barnes, 1995). Moreover, the effect of differentiation and variations in  
2 spectral ranges were tested in the development of the NIRS calibrations.  
3

4 Using the raw spectral data, applying spectral pretreatments and allocating the  
5 corresponding extractable total phenolic content and extractable ellagitannin content to  
6 each sample, calibrations were performed by modified partial least squares regression  
7 (MPLS). In this method, the group of calibration samples is divided into a series of  
8 subsets in order to perform cross-validation to set the number of PLS factors, reduce the  
9 possibility of over fitting (Shenk and Westerhaus 1995) and remove chemical outliers.  
10 Using the  $T \geq 2.5$  criterion, samples that presented a high residual value when they were  
11 predicted were eliminated from the set. Finally, validation errors are combined into a  
12 single figure, the standard error of cross-validation (SECV).  
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26 The software used was Win ISI<sup>®</sup> (v1.50) (Infrasoft International, LLC, Port. Matilda,  
27 PA, USA). This software allowed the data pretreatment, principal components analysis  
28 and sample selection and development of quantitative models.  
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## 34 **Results and discussion**

### 35 *Exploratory analysis of spectra*

36 Fig.1a shows the raw average spectra regarding the sampling point of oak wood  
37 shavings. It can be seen that wood samples belonging to A, B, C and D groups have  
38 different reflectance intensities along some wavelength regions, although with the same  
39 pattern.  
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48 A SNV (2,5,5,1) spectral pre-treatment was applied to the spectra of all samples in the  
49 908–1676 nm region and after that, principal component analysis was carried out in  
50 order to provide information about the latent structure of spectral matrix. PCA was also  
51 used in this study to select representative samples from the spectral data set. Sample  
52 selection was made in order to reduce the number of samples maintaining as much  
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1 spectral variety as possible. Using all spectral samples, five principal components were  
2 taken into account. More than ninety-five per cent of the spectral variability of the  
3 original spectral matrix was explained (96.23 %). In this five-dimensional space, the  
4 samples were sorted according to Mahalanobis distances (H). Then, ten spectral outliers  
5 ( $H > 3$ ) were found and removed from this matrix. The 190 remaining samples were  
6 grouped according their neighborhood H values ( $NH \leq 0.6$  criterion). So, 18 groups with  
7 different spectral characteristics were created. One sample from every group was  
8 allocated in the calibration set. Therefore, 18 samples were selected to develop a  
9 calibration process. In addition, to create the validation set, another sample from every  
10 group was selected. Only 15 samples were allocated in this set because 3 of the 18  
11 created groups had not more than one sample.

12 Fig.1b shows the scores of the wood samples in the space defined by the first and  
13 second principal components which described 53.37 % (PC1) and 30.37 % (PC2) of the  
14 spectral variability in the data. In the scores plot, the samples are represented by a color  
15 code according to the cutting direction of the staves to obtain them (longitudinal or  
16 transversal to the fibers). It can be seen that PC1 could be used to differentiate between  
17 longitudinal and transversal group. Furthermore, it also possible to appreciate a  
18 successful separation between samples according to the different staves processed to  
19 obtain them, in other words, the different sampling points (A, B, C, D) (Fig.1c).  
20 Samples belonging to A and B groups have been obtained in the same way (an  
21 automatic or manual processing of the staves is the only difference between them) and,  
22 therefore, it appears partly overlapping in this plot. Fig.1d shows the scores of the  
23 calibration and validation samples in the space defined by PC1 and PC2. This space is  
24 not entirely the same as the space used to select the calibration and validation sets  
25 (bearing in mind that 5 PCs were used). However, in this figure, it can be appreciated

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how both the calibration set and the validation set are homogeneously allocated in the space defined by PC1 and PC2 including almost all the spectral variability of samples and, therefore, representing the heterogeneity of the whole set.

#### *Quantitative analysis.*

After HPLC-MS method, 4 ellagitannins were determined (castalagin, vescalagin, grandinin and roburin E) and individual extractable ellagitannin contents were obtained. The sum of all them was also expressed as total ellagitannins. From a quantitative point of view, castalagin and vescalagin are the most abundant ellagitannins in oak wood but lyxose/xylose derivatives (grandinin and roburin E) are also present. Oak wood extractable total phenolic content has also been quantified using a spectrophotometric method. Table 1 shows the main statistical descriptors for the reference parameters of the samples allocated in the calibration set.

Quantitative calibrations were developed by modified partial least squares (MPLS) regression. Extractable total phenolic content and extractable ellagitannin content were used as dependent (Y) variables and the matrix of 18 wood processed spectra was used as the independent (X). The statistical parameters of the final calibration equations are shown in Table 2 where N is the number of samples used to obtain the calibration equation after eliminating samples for chemical reasons (T criterion). The best of the different mathematical treatments, the range of application, and standard deviations are also shown.

The robustness of the selected models was tested using validation set samples, which did not belong to the calibration set, as external validation. Samples which presented reference values outside the applicability of the obtained models for each reference parameter were not used in this procedure. In the case of the total phenols parameter all sample presented reference values inside the applicability of the obtained model,

1 however, 2, 2, 5, 3 and 1 samples were not used for castalagin, vescalagin, grandinin,  
2 roburin E and total ellagitannin validations, respectively. Table 3 shows the main  
3 statistical descriptors for the reference parameters of the samples allocated in the  
4 validation set after remove from the validation set the samples that should not be  
5 predicted by the calibration models. Calibration model obtained was applied and the  
6 predicted values were compared with the reference data. As result of this external  
7 validation the standard errors of prediction (SEP) were obtained for each reference  
8 variable, these values were also included in Table 2. These values are comparatively  
9 similar or even better than the errors previously reported for total or extractable content  
10 of these compounds using near infrared spectroscopy bench top devices taking into the  
11 account the applicability range (Baca-Bocanegra et al. 2018; Giordanengo et al. 2009).

12 Fig.2 shows the loadings resulting of the MPLS model for total phenols as example and  
13 it indicates the most dominant wavelengths. The spectral region between 1100 and 1200  
14 showed important contribution to the model loadings. These could be related to  
15 combination band of O–H functional group and symmetric and anti-symmetric  
16 stretching vibration. This wavelength region is also related to C–H aromatic second  
17 overtones and C–H third overtones vibration. These can be attributed to the chemical  
18 structure of phenolic compounds (Osborne et al. 1993; Siesler et al. 2002). Combination  
19 band of C–H aromatic appears in the spectral region between 1350 and 1450 nm.  
20 Moreover, the first O–H stretching overtone contributes to spectrum at 1400 nm, hence  
21 the moisture affects expansively to this band. Definitive attribution of absorbances  
22 around 1400 nm is difficult given that absorptions around this wavelength range  
23 originate from both -OH and -CH structures.

### *Use of developed method for the screening of extractable polyphenols*

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2 The methods developed in the preceding section were used to predict the extractable  
3 content of total phenols and total ellagitannins in the whole of wood set. The results  
4 show a substantial variability in the reference parameters within each group of samples  
5 (Fig.3). The variability of oak wood extractives has been demonstrated by different  
6 authors (Doussot et al. 2002; Snackers et al. 2000). Phenols content has been found to  
7 vary even from the same provenance, the same tree and the same piece of timber  
8 (Masson et al. 1995). Zahri et al. (2008) reported that total phenols content in wood  
9 varied according to the distance from the oak tree pith. This observation confirmed  
10 previous studies which determined that the concentration of soluble tannins in oak tree  
11 decrease with age tree. A large amount of tannins and phenolic monomers in wood can  
12 form polymers and copolymers with cell-wall components with time and then their  
13 solubility would be reduced (Peng et al. 1991; Viriot et al. 1994).

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15 In addition, an inter group variability of oak wood extractives has been found. Samples  
16 generated by a processed of the staves in the longitudinal direction of the fibers hold  
17 higher extractable polyphenols content than those obtained after processing staves in the  
18 transversal direction. On the basis of these results, it can be assumed that the structure  
19 of the samples could be involved in these extractable content differences.

### **Conclusion**

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21 The MPLS model performed confirmed that spectral analysis developed from  
22 MicroNIR is a promising technique for *in situ* routine assessment of the extractable  
23 polyphenolic compounds in wood cooperage byproduct. This methodology was able to  
24 predict, *in situ*, the extractable polyphenolic content of a sample based on spectral  
25 features as the predictor variables. The obtained results are comparable with those  
26 obtained using other bench-top devices and presents the advantage of its eventual



1 friendly use out of lab. The use of a portable micro spectrometer implies very  
2 interesting features, since at a lower cost objective and instantaneous spectroscopic  
3 measurements can be carried out *in situ* with the advantage of its portability, due to its  
4 small size. Development of applicable models *in situ* will also allow a greater versatility  
5 and efficiency for the decision-making in the winemaking process on the adequacy and /  
6 or dosage of these byproducts according to the requirements of the wine.  
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14 The use of cooperage by-product as source of copigments for wine is a sustainable and  
15 profitable alternative to get high quality red wine for the oenological industry and to  
16 disposal of their byproducts for the cooperage industry.  
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21 The variability in oak wood extractives within the same provenance has been observed.  
22 Moreover, extractable polyphenols content differences between wood samples  
23 generated by longitudinal and transversal cutting of the fibers have been also found.  
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The Spanish Ministerio de Economía y Competitividad is thanked for project AGL2017-84793-C2. Universidad de Sevilla is thanked for B. Baca-Bocanegra predoctoral grant (VPPI-II.2) and J. Nogales-Bueno postdoctoral grant (VPPI-II.4). I. García-Estévez thanks FEDER-Interreg España-Portugal Programme (Project ref. 0377\_IBERPHENOL\_6\_E) for his postdoctoral grant. The authors thank the technical staff of Biology Service [Servicios Generales de Investigación (SGI), Universidad de Sevilla]. They also thank Tonelería Salas S.L. (Bollulos Par del Condado, Huelva, Spain) for supplying the cooperage byproduct samples.

## **Conflict of Interest**

The authors declare that they have no conflict of interest.

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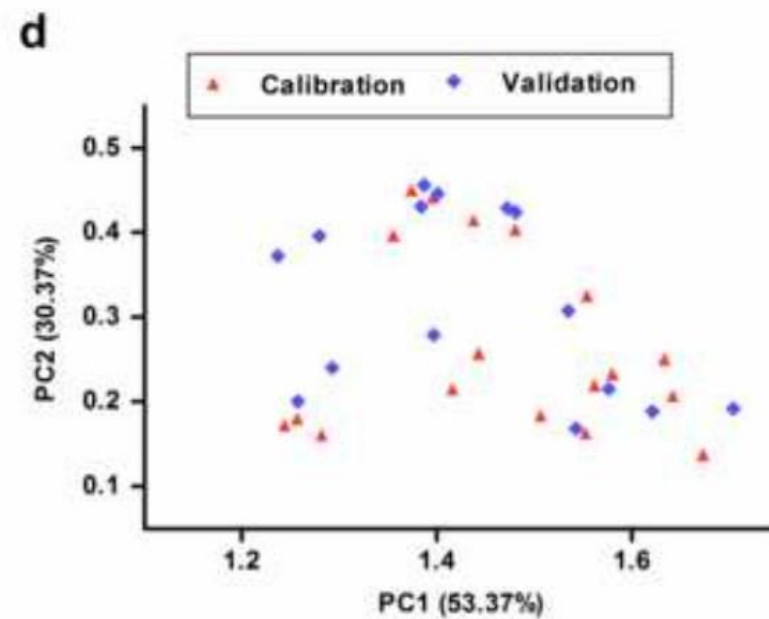
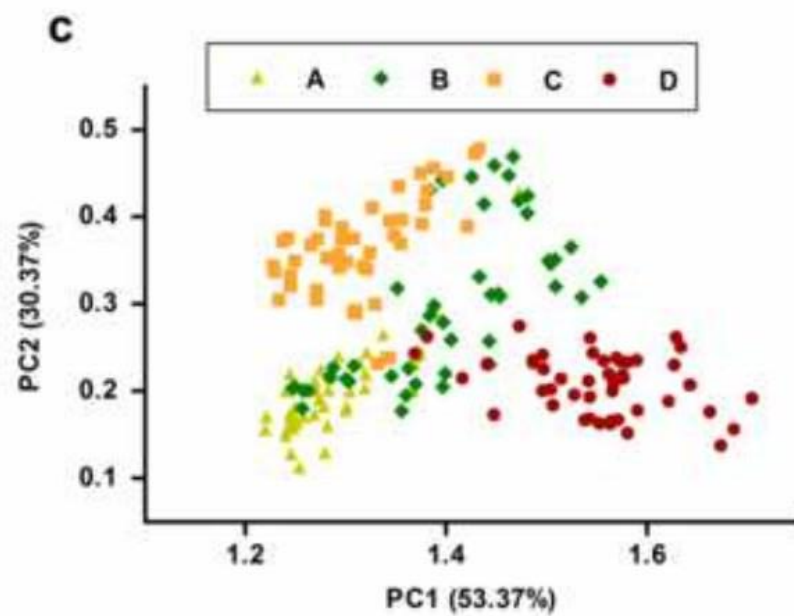
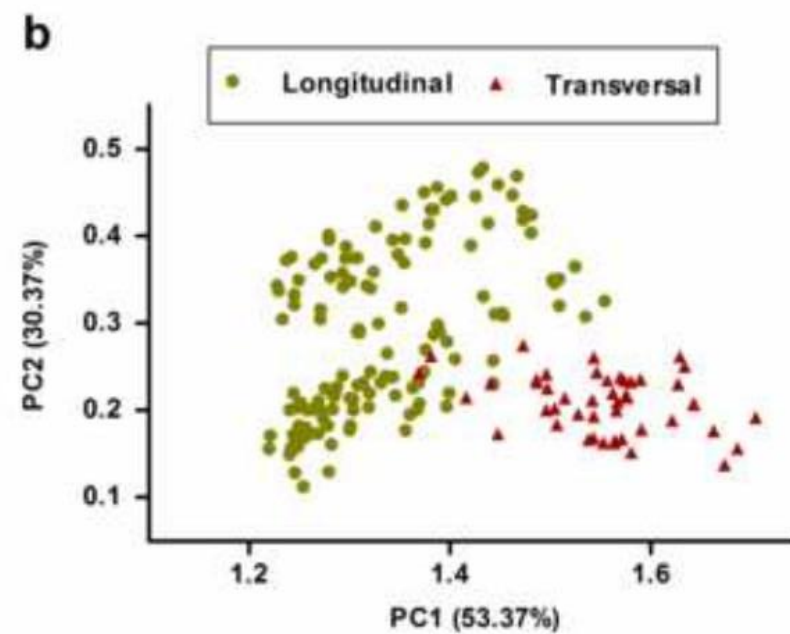
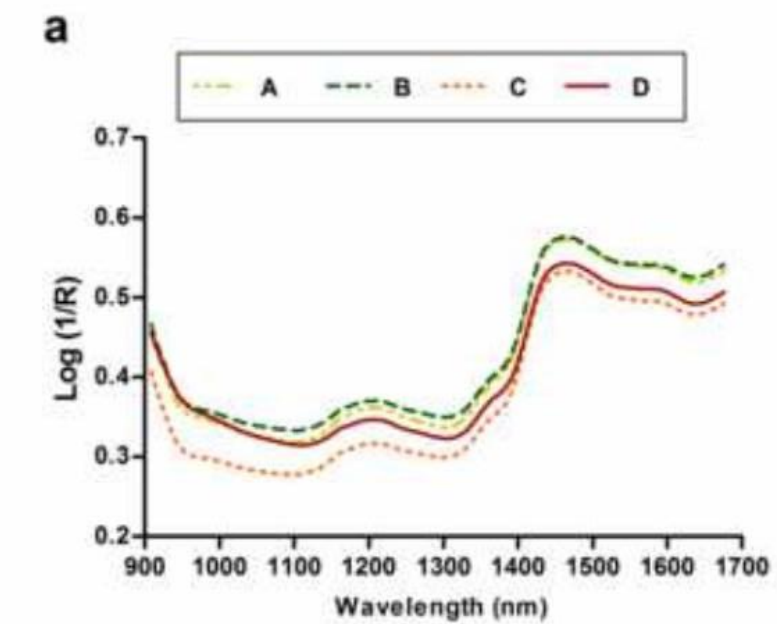
## Figure captions<sup>1</sup>

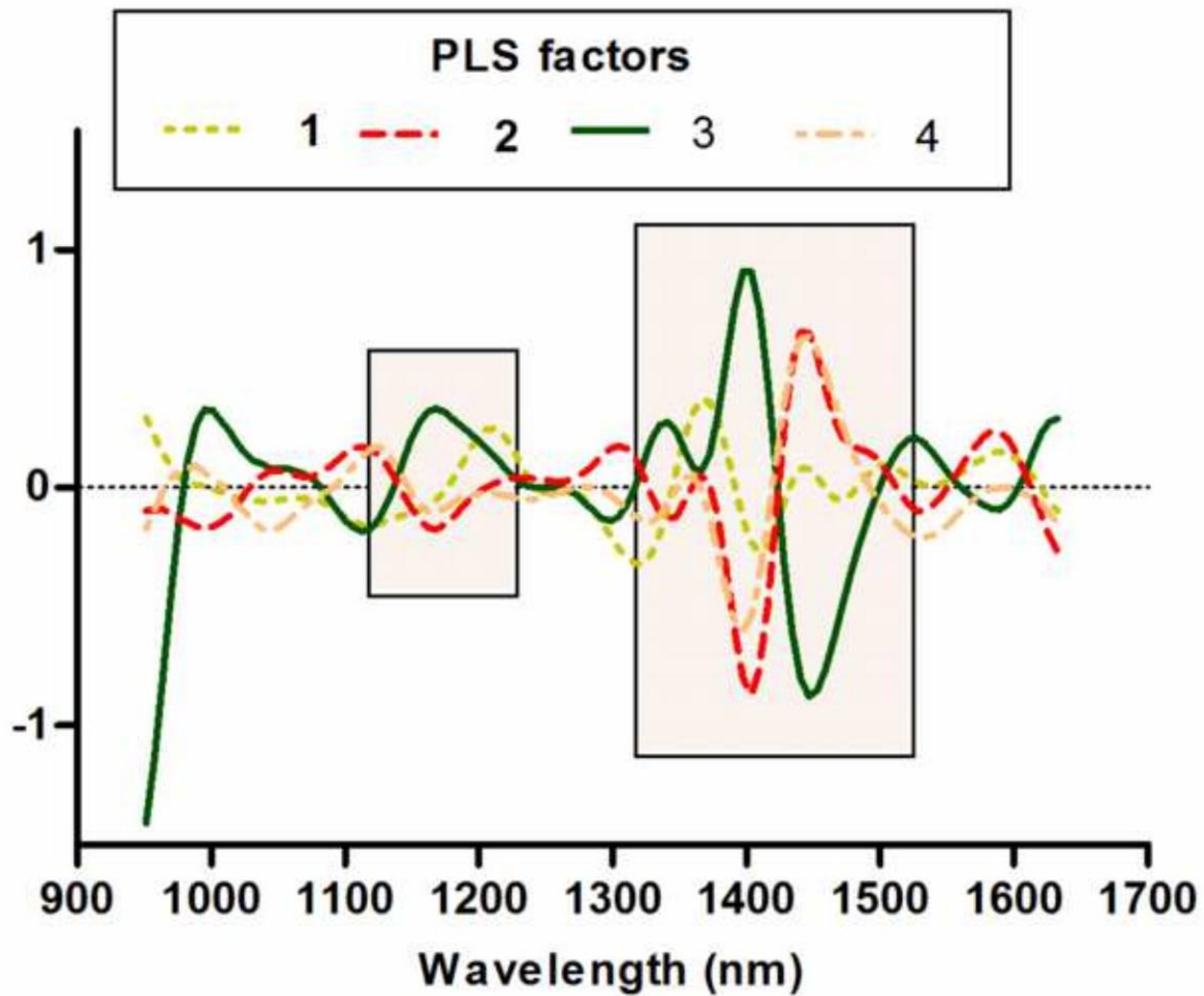
**Fig. 1** (a) Raw average spectra of wood samples according to sampling point. (b) Score plot of wood samples codified as longitudinal and transversal samples in the space defined by PC1 and PC2 (c) codified according to sampling point (A, B, C and D) (d) Score plot of wood samples codified as calibration and validation sets in the space defined by PC1 and PC2

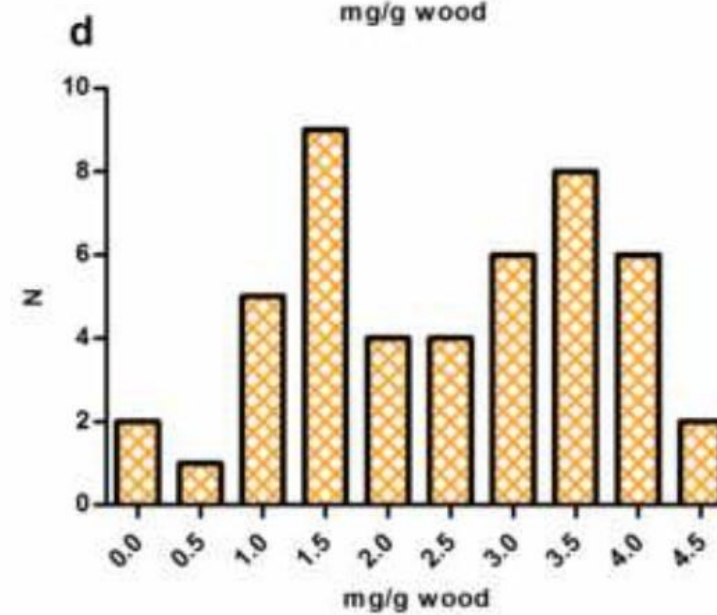
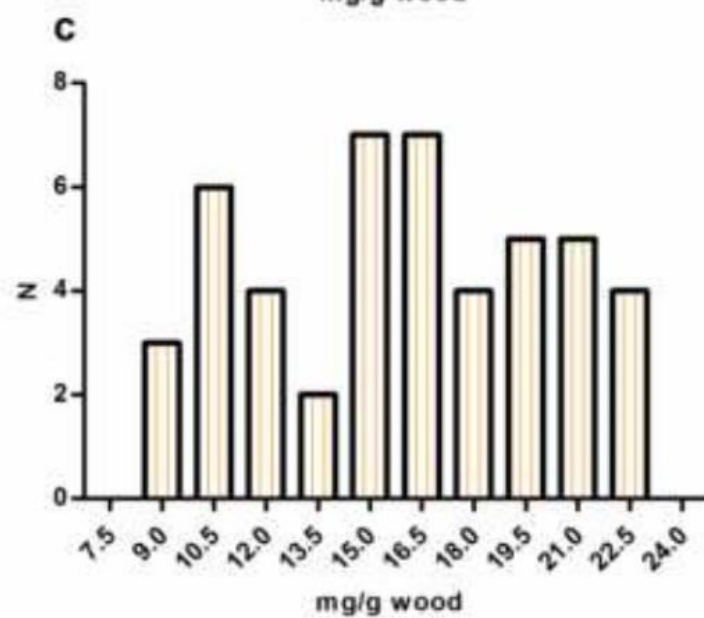
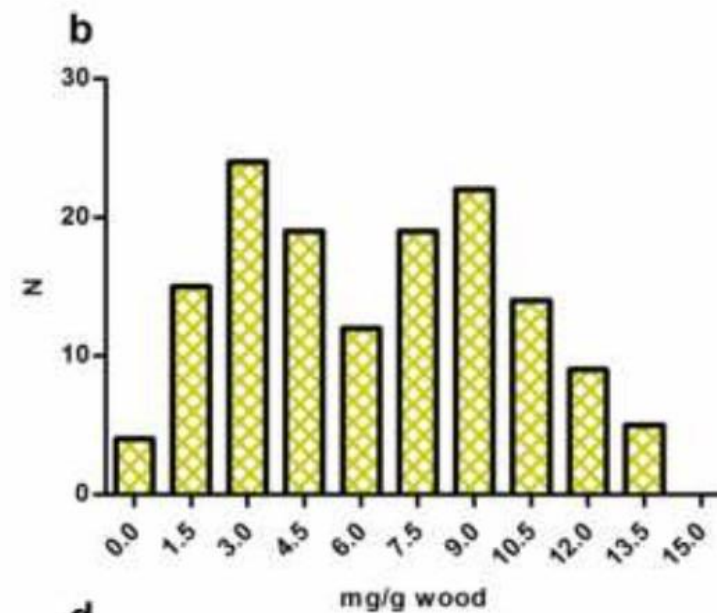
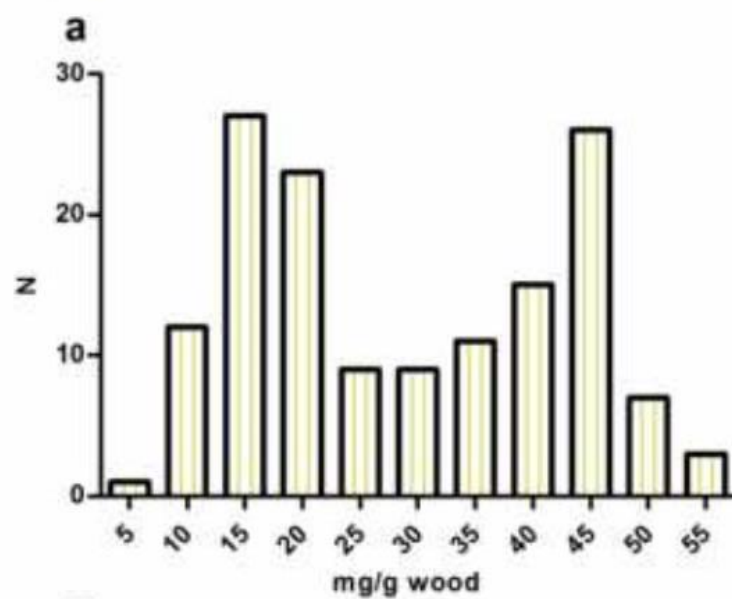
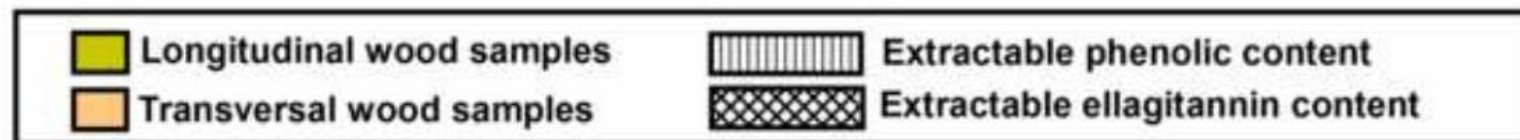
**Fig. 2** Loadings plot for the PLS factors of the regression model for extractable total phenolic content prediction

**Fig. 3** Distribution of longitudinal and transversal wood samples in different extractable phenolic content (a and c) and extractable ellagitannin content (b and d)

1 NOTE: All figures should be in color on the Web and in black-and-white in print.







# Screening of wine extractable total phenolic and ellagitannin contents in revalorized cooperage byproducts: evaluation by MICRO-NIRS technology

## Food and Bioprocess Technology

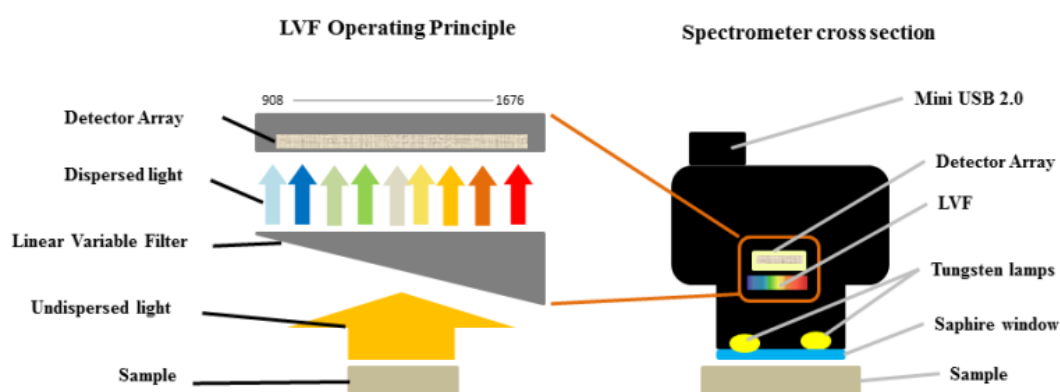
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**ESM\_1.** Illustration of the optical design and cross section of the MicroNIR operated in diffuse reflection mode adapted from VIAVI user manual.

**Table 1.** Main statistical descriptors for reference parameters in calibration set.

Set	Reference Parameters	N <sup>a</sup>	Maximum	Mean	Minimum	SD <sup>b</sup>
Calibration	EPC <sup>c</sup>	18	59.56	23.79	9.41	15.67
	EEC <sup>d</sup>	18	16.15	5.07	1.49	4.66
	Castalagin	18	6.62	2.03	0.52	1.99
	Vescalagin	18	6.94	2.01	0.60	1.86
	Gradinin	18	1.74	0.71	0.18	0.58
	Roburin E	18	0.85	0.31	0.08	0.26

<sup>a</sup>N: number of samples; <sup>b</sup>SD: Standard deviation; <sup>c</sup>EPC: extractable total phenolic content (mg g<sup>-1</sup> of wood, expressed as gallic acid equivalents); <sup>d</sup>EEC: extractable total ellagitannin content (mg g<sup>-1</sup> of wood, expressed as sum of individual ellagitannins);

**Table 2.** Calibration statistical descriptors for the models developed in the NIR zone close to 908-1676 nm.

Reference Parameter	Spectral pretreatment	T outliers	PLS factors	N <sup>a</sup>	Est. Min	SD <sup>b</sup>	Est. Max	SEC <sup>c</sup>	RSQ <sup>d</sup>	SECV <sup>e</sup>	SEP <sup>f</sup>
					(mg g <sup>-1</sup> wood)				(mg g <sup>-1</sup> wood)		
EPC <sup>g</sup>	SNV 2,5,5,1	0	4	18	0	14.92	68.23	2.07	0.98	3.83	5.86
EEC <sup>h</sup>	Standard MSC 1,5,5,1	0	2	18	0	4.66	19.04	1.30	0.92	1.76	2.07
Castalagin	Standard MSC 1,5,5,1	0	2	18	0	1.99	8.00	0.66	0.90	0.85	0.90
Vescalagin	None 1,5,5,1	1	2	17	0	1.44	6.04	0.36	0.94	0.53	0.71
Gradinin	Detrend 2,5,5,1	2	3	16	0	0.55	2.34	0.09	0.98	0.13	0.16
Roburin E	Standard MSC 1,5,5,1	0	2	18	0	0.30	1.16	0.08	0.91	0.11	0.13

<sup>a</sup>N: number of samples (calibration set); <sup>b</sup>SD: standard deviation; <sup>c</sup>SEC: standard error of calibration; <sup>d</sup>RSQ: coefficient of determination (calibration set); <sup>e</sup>SECV: standard error of cross-validation (7 cross-validation groups); <sup>f</sup>SEP: standard error of prediction (external validation); <sup>g</sup>EPC: extractable total phenolic content (mg g<sup>-1</sup> of wood, expressed as gallic acid equivalents); <sup>h</sup>EEC: extractable total ellagitannin content (mg g<sup>-1</sup> of wood, expressed as sum of individual ellagitannins)



**Table 3.** Main statistical descriptors for reference parameters in validation set.

Set	Reference Parameters	N <sup>a</sup>	Maximum	Mean	Minimum	SD <sup>b</sup>
Validation <sup>e</sup>	EPC <sup>c</sup>	15	56.54	32.03	11.26	17.73
	EEC <sup>d</sup>	14	14.44	7.50	1.40	4.49
	Castalagin	13	5.50	2.91	0.51	1.73
	Vescalagin	13	4.85	2.82	0.67	1.53
	Gradinin	10	1.57	0.83	0.25	0.48
	Roburin E	12	0.78	0.43	0.09	0.26

<sup>a</sup>N: number of samples; <sup>b</sup>SD: Standard deviation; <sup>c</sup>EPC: extractable total phenolic content (mg g<sup>-1</sup> of wood, expressed as gallic acid equivalents); <sup>d</sup>EEC: extractable total ellagitannin content (mg g<sup>-1</sup> of wood, expressed as sum of individual ellagitannins); <sup>e</sup>Validation: statistical descriptors for the reference parameters of the samples allocated in the validation set after remove the samples that presented reference values outside the applicability of the calibration models.