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Relative impact of oenological tannins in model solutions and red wine according to phenolic, antioxidant, and sensory traits

Maria Alessandra Paissoni [a], Giovanni Bitelli [a], Mar Vilanova [b], Carlo Montanini [c], Susana Río Segade [a,*], Luca Rolle [a], Simone Giacosa [a]

[a] Department of Agricultural, Forest and Food Sciences, University of Torino, Largo Braccini 2, 10095 Grugliasco, TO, Italy
[b] Instituto de Ciencias de la Vid y del Vino (ICVV-CSIC), Carretera de Burgos Km. 6, Finca La Grajera 26007, Logroño, Spain
[c] AEB S.p.A., Via Vittorio Arici 104, 25134 Brescia, Italy

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ABSTRACT

Oenological tannins (OETs) are winemaking processing aids used to facilitate stabilization and fining, to increase the antioxidant capacity, and to promote colour stability of grape juice and wine. A wide variability of pure or mix formulates are available for winemaking purposes, including hydrolysable tannins (gallotannins and ellagitannins), proanthocyanidins from grape skins and seeds (prodelphinidins and procyanidins), and from exotic wood (prorobinetinidins and profisetinidins). In this study, seventeen OETs pure and mix formulates were characterized in terms of polyphenolic content and antioxidant capacity in a model wine and in a red wine after one-month storage, as well as aroma, astringency, and bitterness sensory characteristics in water and red wine. Colour-related features were also analysed in the added red wine after one-month storage. For the first time, correlations among the obtained results in the different matrices were investigated to understand the most suitable OETs for winemaking applications. The results showed a great variability among the formulates studied in terms of phenolic content, which was strictly correlated to their antioxidant capacity. Regarding origin, hydrolysable tannins had the highest antioxidant ability, followed by exotic wood formulates. A strong and positive correlation was found in antioxidant capacity of OETs in model wine and red wine after one-month storage, in particular for ellagitannins, which confirmed also their ability to increase pigments polymerization. By contrast, quebracho tannins resulted the bitterest and most astringent when tasted in water (0.4 g/L), although in-mouth and aromatic descriptors of OETs tasted in water were not correlated with the ones of the added red wine. Therefore, the choice of OETs formulate and its optimal dose requires a characterization in terms of polyphenolic content and antioxidant capacity because these properties were well correlated with those of the added wines in a short storage period, whereas the sensory impact at oenological range doses is mainly dependent on wine features.

1. Introduction

In grapes, condensed tannins and their compositional subunits, the flavan-3-ol monomers, are naturally present in skins and seeds, from where they are extracted during the maceration of grape solids into the must-wine (Vazallo-Valleumbrocio, Medel-Marabolí, Peña-Neira, López-Solís, & Obreque-Slier, 2017). Their contribution to wine quality is related to their role in mouthfeel and longevity (Ma et al., 2014). Besides tannins naturally present in grapes, some others from grape or different botanical sources can be used during the winemaking process as processing aids and are named oenological tannins (OETs). According to the International Code of Oenological Practices and the recent resolutions OIV-OENO-612–2019 and OIV-OENO-613–2019 of the *Organisation Internationale de la Vigne et du Vin* (OIV, 2022, 2019a, 2019b) their use is authorized to facilitate the stabilization and fining of musts and wines, as well as to increase the antioxidant and antioxidasic capacity of grape juice and to promote colour stability.

OETs are manufactured with a solid–liquid extraction from vegetal material, and they can be found as formulation from a single botanical species or from a mixture of them, such as grape, oak, chestnut, quebracho, acacia, and tara (Versari, Du Toit, & Parpinello, 2013). The efficacy of tannin addition in winemaking is related to the formulation

* Corresponding author.
E-mail address: susana.riosegade@unito.it (S. Río Segade).

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botanical origin, chemical characteristics, polyphenolic content, and by the stage and dose of addition (Versari et al., 2013). Considering the chemical characteristics, OETs can be divided in two groups: condensed and hydrolysable tannins. The former, also named proanthocyanidins, is constituted by polymers that differ according to the structural monomeric units, the flavan-3-ols and the flavan-3,4-diols. In grapes, flavan-3-ols-based polymers are found, namely procyanidins and prodelphinidins. Procyanidins are composed of (+)-catechin and (-)-epicatechin with different extent of galloylation, whereas prodelphinidins are composed also of (-)-epigallocatechin and (+)-gallocatechin (Souquet, Cheynier, Brossaud, & Moutounet, 1996). Other types of condensed tannins presenting flavan-3,4-diols subunits have been characterized in the exotic woods commonly employed in oenological industry: profisetinidins from quebracho (*Schinopsis* spp.), which are principally composed of fisetinidol monomeric units (Venter, Sisa, van der Merwe, Bonnet, & van der Westhuizen, 2012a), and prorobinetinidins containing robinetidinol as subunit, which are the main components of *Mimosaceae* family (e.g., acacia) tannins together with prodelphinidins and profisetinidins at lower percentage (Venter et al., 2012b). Concerning hydrolysable tannins, they are usually classified in gallotannins and ellagitannins. Gallotannins are present in plant gallnuts and they are composed of gallic acid and D-glucose, with different extent of substitution with galloyl-moiety (Hagerman, 2011). Instead, ellagitannins are formed by D-glucose and ellagic, gallic, or hexahydroxydiphenic acids (Hagerman, 2011). They are commonly extracted from chestnut and oak, and the eight most common forms are monomers, i.e. castalagin, vescalagin, grandinin, roburin E, and dimers, i.e. roburin A, B, C, D (Puech, Feuillat, & Mosedale,1999).

During the winemaking process, OETs can be added at crushing or during the first days of maceration to protect endogenous polyphenols from the enzymatic and chemical oxidative reactions that occur in the must. In fact, research highlighted that OETs addition in some cultivars with a low content of anthocyanins or a high percentage of di-substituted forms, or in highly contaminated grapes by *Botritys cinerea* can help in preserving the extracted anthocyanins, leading to an improved final wine colour (Keulder, 2006; Venturi, Andrich, Serni, Taglieri, & Sanmartin, 2015; Vignault et al., 2019a, Paissoni et al., 2020). OETs can act as both antioxidants and copigments (Bautista-Ortìn, Martínez-Cutillas, Ros-Garcia, López-Roca, & Gómez-Plaza, 2005), as demonstrated in model-wine solution (Gombau et al., 2019a; Vignault et al., 2018; Vignault et al., 2019b). Furthermore, they can be utilized in maceration and ageing to promote the formation of more stable pigments, by anthocyanin direct polymerization with condensed tannins or indirectly because of acetaldehyde production by ellagitannin oxidation (Vivas & Glories, 1996, Picariello, Gambuti, Petracca, Rinaldi, & Moio, 2018).

Beyond their influence on wine chemical evolution, OETs can potentially modify sensory perceptions of wine, in terms of aroma, astringency, and bitterness. Tannin concentration, type of proanthocyanidins, and their structural properties, such as stereochemistry, substitution, and mean degree of polymerization, can influence astringency and bitterness of condensed tannins to different extent (Ma et al., 2014). Astringency, which is a tactile sensation, is mainly due to the interaction between tannins and salivary proteins with the consequent loss of mouth lubrication (Ma et al., 2014). Hofmann et al. (2006) compared constituents of OETs determining that proanthocyanidins owned lower molar detection thresholds than pentagalloyl glucose and castalagin, although grandinin resulted in the lowest thresholds. Depending on individual compound, they reported a different perception given by the different affinity between the molecule structure and salivary proteins. In complex OETs formulations, Gombau et al. (2019b) found ellagitannin OET formulation was more astringent than gallotannin OET, and both hydrolysable formulations showed higher intensities than seed proanthocyanidin OET. Besides intensity, different astringency sub-qualities were found, ellagitannins being perceived as smoother and more velvety than grape proanthocyanidins (Chira et al., 2015). Together with astringency, bitterness is considered relevant in wine overall quality evaluation. Bitterness is a taste and, in the wine, it is influenced mainly by phenolic compounds, in particular by low molecular weight flavan-3-ol-based tannins (Ma et al., 2014). Given the grape proanthocyanidins high abundance in wine, several studies have been performed on them confirming bitterness is influenced by polymer length, subunits, and conformation (Ma et al., 2014), whereas little is known on exotic wood tannins. However, Puech, Prida, & Isz (2007), comparing several OETs, stated that proanthocyanidins from quebracho are the most bitter with respect to ellagitannins, gallotannins, and grape proanthocyanidins when tasted at a concentration of 0.5 g/L in water.

Besides in-mouth sensation, OIV (2015) indicates that the technical use of tannins at normal usage doses must not change the olfactory properties of wines. Some scientific evidence of the modification of the aromatic profile in wines added with OETs is available, varying by type and dose, as a result of their usage during the process or after addition (Bautista-Ortín et al., 2005; Harbertson, Parpinello, Heymann, & Downey, 2012; Larcher et al., 2015; Chen et al., 2016; Li, Wei, Yu, & Cui, 2020; Corona, Bambina, De Filippi, & Cinquanta, 2021). OETs can also change the perception of wine volatile compounds, such as esters and alcohols (Mitropoulou, Hatzidimitriou, & Paraskevopoloulou, 2011; Chen et al., 2016), in light of the ability of some polyphenols to modify aroma volatility (Pittari, Moio, & Piombino, 2021).

In the recently published scientific research, it is still not certain what is the direct correlation between the formulation characteristics and the extent of their impact on final wine, depending, on one hand, on several tannin formulation specifications (such as dosage, purity, and molecular characteristics), and, on the other hand, on the wine characteristics. Therefore, the main aim of the present research is to evaluate the chemical, antioxidant, and sensory properties of seventeen selected OETs in different media, and to compare these findings in order to assess the extent of these changes across model wine solution, water, and red wine conditions. For all media considered, OETs addition was done at a standardized substantial dosage to better evaluate the modifications induced and to provide consistency in the comparisons done. In detail, after an exhaustive OETs characterization in terms of chemical and antioxidant traits in model wine solution, and sensory properties in water, their performance in these features was analysed in a finished red wine. For this purpose, the sensory properties of the wines with added tannin formulations were evaluated, as well as the colour, phenolic content, and antioxidant features after one month from the addition. Similarities and differences of the tannin effect in the two matrices were compared highlighting the importance of this information for oenologists in the choice of the most suitable formulation to obtain the desired final product.

2. Materials and methods

2.1. Chemicals

All chemicals of analytical reagent grade, gallic acid, Folin-Ciocalteu reagent, methylcellulose, bovine serum albumin (BSA), 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS), 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2,4,6-tri(2-pyridyl)-s-triazine (TPTZ), iron (III) chloride hexahydrate, neocuproine, copper (II) chloride dihydrate, and 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox) were obtained from Sigma-Aldrich (St. Louis, MO, USA). Malvidin-3-O-glucoside chloride was purchased from Extrasynthese (Genay, France). All the aqueous solutions were prepared in ultrapure water produced by a Purelab Classic system (Elga Labwater, Marlow, UK).

2.2. Tannin formulations (OETs)

Seventeen oenological tannin formulations were used, grouped in four chemical classes depending on their composition and origin: proanthocyanidins from grape (Proc/prod), proanthocyanidins from

exotic wood (Prof/pror), hydrolysable tannins (Hydro), and mixed formulations of these classes (Mix) (Table 1). In detail, three formulations obtained from grapes, three from exotic woods (two from quebracho and one from *Mimosaceae*), three hydrolysable tannins (two from oak wood, one from *Robinia pseudoacacia* gallnut), and eight mixed formulations were used for the present study. Tannin formulations were provided by AEB S.p.A. (Brescia, Italy).

2.3. OETs polyphenolic and antioxidant capacity characterization

OETs were dissolved in triplicate in model wine solution (12 % v/v ethanol, 4 g/L tartaric acid, pH 3.5) to prepare stock solutions for analysis (1 g/L and 10 g/L for polyphenolic characterization and antioxidant capacity, respectively). Spectrophotometric analyses were performed using a spectrophotometer (UV-1800, Shimazdu, Kyoto, Japan). The total polyphenolic content (TPI) was evaluated measuring the 280 nm absorbance of OET solutions diluted 100 times in ultrapure water. The results were expressed as polyphenolic content (g gallic acid equivalents/100 g of product). External calibration curves of gallic acid (Absorbance 280 nm = 0.0350 × C (mg/L gallic acid), $R^2 = 0.9951$) and (-)-epicatechin (Absorbance 280 nm = 0.0128 × C (mg/L (-)-epicatechin), $R^2 = 0.9999$) were done and gallic acid was chosen for all formulations as standard according to OIV (2015) to facilitate the comparison. Also the values of absorbance at 230 nm and 280 nm, corrected for the dilution, were reported. These values were used as well to investigate their ability as astringency predictors (Boulet et al., 2016). The Folin-Ciocalteu method (FC) was used for the determination of polyphenol content. Previously prepared stock solutions, diluted 20 times, were used and the resulting absorbance at 750 nm was measured after 70 min. Results were expressed as g gallic acid equivalents/100 g of product through an external calibration curve (Vignault et al., 2018). For proanthocyanidin determination, the Bate-Smith assay (BS) was used from acid-catalysed depolymerisation in a warm bath (100 °C). The results were reported as g cyanidin-3-monoglucoside chloride equivalents/100 g of product (Torchio, Cagnasso, Gerbi, & Rolle, 2010). Furthermore, OETs were analysed by methylcellulose precipitation assay (MTC, Sarneckis et al., 2006), similarly to Vignault et al. (2018) using 100 μL of tannin stock solution, 1.5 mL of 0.4 % methylcellulose solution or water, 1 mL of saturated ammonium sulphate solution, and water up to a final volume of 5 mL. Results were calculated as absorbance difference at 280 nm due to precipitation (ΔA280) and expressed as g gallic acid equivalents/100 g of product. Moreover, as a further predictor of astringency, tannins reactive with bovine serum albumin (BSA) protein were evaluated following the method proposed by Boulet et al. (2016), adapted by modifying the volumes used of buffer solution (0.5 mL) and sample (1 mL). BSA index was obtained multiplying the ΔA280 by the number of dilutions.

Antioxidant capacity was investigated with four different assays: ABTS, DPPH, FRAP, and CUPRAC. For each tannin formulation, the stock solution was diluted 100 times with ultrapure water to perform the analyses. ABTS (Re et al., 1999) and DPPH (Brand-Williams, Cuvelier, & Berset, 1995) methods were carried out as proposed by Ky & Teissedre (2015) recording the absorbance at 734 nm and 515 nm, respectively. FRAP (Benzie & Strain, 1996) and CUPRAC (Apak, Güclü, Öziürek, & Celik, 2008) methods were conducted with the dose described by Vignault et al. (2018), reading absorbance at 593 nm and 450 nm wavelength, respectively. All antioxidant assays were measured after 30 min of reaction time at room temperature, and reaction blanks were performed using water. For each method, a Trolox-based calibration curve was used and results were expressed as mmol Trolox equivalents/g of product. For each tannin formulation, the *antioxidant potency* was calculated as AP = (antioxidant capacity/total phenolic content) × 1000 (De Beer, Joubert, Gelderblom, & Manley, 2003), where antioxidant capacity is the result of antioxidant test (ABTS, DPPH, FRAP, CUPRAC as mmol Trolox equivalent/g of product) and the total phenolic content is obtained with the Folin-Ciocalteu assay (as mg of gallic acid/g of product).

Table 1
Oenological tannins (OETs) formulations used in the study.

Sample	Group	Type	Description[a]
Sd1	Proc/prod	pure	Proanthocyanidins from grape seeds *Vitis vinifera* L.
Sd2	Proc/prod	pure	Proanthocyanidins from grape seeds *Vitis vinifera* L.
Sk1	Proc/prod	pure	Proanthocyanidins from white grape skins *Vitis vinifera* L.
Q1	Prof/pror	pure	Proanthocyanidins from quebracho
Q2	Prof/pror	pure	Proanthocyanidins from quebracho
Ac	Prof/pror	pure	Proanthocyanidins from *Mimosaceae*
Et1	Hydro	pure	Ellagitannins
Et2	Hydro	pure	Ellagitannins from *Quercus* spp.
Gt	Hydro	pure	Gallotannins from *Robina pseudoacacia* galls
Mx1	Mix	mix	Proanthocyanidins from grape skins and quebracho, and ellagitannins from *Quercus* spp.
Mx2	Mix	mix	Proanthocyanidins and ellagitannins
Mx3	Mix	mix	Proanthocyanidins from grape skins *Vitis vinifera* L. and quebracho
Mx4	Mix	mix	Proanthocyanidins from grape skins and seeds of *Vitis vinifera* L. and quebracho
Mx5	Mix	mix	Proanthocyanidins from grape skins and seeds of *Vitis vinifera* L. and quebracho
Mx6	Mix	mix	Ellagitannins, gallotannins, and proanthocyanidins
Mx7	Mix	mix	Ellagitannins and proanthocyanidins
Mx8	Mix	mix	Ellagitannins and proanthocyanidins

Proc/prod = procyanidins/prodelphinidins; Prof/Pror = profisetinidins/prorobinetinidins; Hydro = hydrolizable tannins, and Mix = mixed formulation.
[a]Information given by the supplier.

2.4. OETs sensory characterization

2.4.1. Samples and conditions

OETs were evaluated by sensory analysis (astringency, bitterness, and aroma profile) in water and wine. The sensory analysis was conducted in a professional-standard room at the University of Torino – Department of Agricultural, Forest and Food Sciences (DISAFA) in Asti, Italy.

2.4.2. Sensory panel

The first sensory panel included 21 volunteers' students and staff of Viticulture and Oenology (15 males and 6 females) from University of Torino. All participants had experience in wine tasting and were recruited based on their availability. Prior to the training, they were informed about the procedures, samples, and treatments, and written informed consent was obtained from all participants. After training, panelists whose ratings deviated more than one standard deviation from the group mean were not included in the final panel. Finally, two separate sensory panels were created due to individual panelist performance variability in the rating of astringency and bitterness perceptions. Thus, 8 panelists were selected for astringency, and 9 for bitterness.

2.4.3. Training

The training was carried out in eight sessions (1.5 h/session). In the first training session, 0.6 g/L of tannic acid, 1 g/L of caffeine, and 1 g/L of tartaric acid were used to identify and differentiate astringency, bitterness, and acidity sensations, respectively. The second part of training (5 sessions) was about the ability to use the unstructured scale chosen to evaluate tannins; combining ranking tests with the rating of standard solutions on a 10-cm scale. In detail, different concentrations of tannic acid (0.1, 0.2, 0.4, 0.6 g/L) and caffeine (0.25, 0.5, 0.75, 1 g/L) solutions in water were used and ranking test was performed. These concentrations were selected to establish the scale range. In the second session, the ranking of the two scales (tannic acid and caffeine) was proposed. In the third and fourth sessions the same ranking test was proposed again, introducing also repeated concentration samples. In the fifth and sixth training sessions, the panelists were asked to rate

astringency and bitterness using an unstructured 10-cm scale range, from low to high intensity. Afterwards, the training consisted in taking confidence with the tasting sheet and evaluation procedure using oenological tannins dissolved in water (2 sessions). The seventh session was performed using two OETs chosen according to their origin (proanthocyanidinic and hydrolysable) in two different concentrations each (0.2 g/L and 0.6 g/L) and a standard solution (tannic acid 0.4 g/L). Furthermore, a control tannin (0.4 g/L), i.e. a formulation not included in the products under evaluation, was used in both the training and formal tasting to evaluate panelists' repeatability and reproducibility. The eighth session was performed with the same oenological tannin formulations previously used in a concentration of 0.4 g/L (that will be used in all further tastings), the standard solution (tannic acid 0.4 g/L) in duplicate, and duplicate control samples.

In the previously described training sessions, an aroma assessment training was performed to familiarize and identify aroma descriptors of tannins. A preliminary training session was performed with tannin solutions to pick out the common aroma descriptors defining them. The aroma descriptors selected were *mushroom*, *licorice*, *caramel*, *pepper*, *vanilla*, *balsamic*, *orange*, and *wood*. The aroma training for *caramel*, *mushroom*, *pepper*, *balsamic* (pine), and *vanilla* was performed with standards from Le Nez du Vin (Jean Lenoir Ed., Cassis, France). The standards for *orange* (orange zest, 5 g/L), *wood* (medium toasted wood chips, 20 g/L), and *licorice* (licorice pure candy, 5 g/L) were singularly prepared by extraction in aqueous ethanol (20 % v/v) and subsequent dilution in water to reach the final concentration limiting the ethanol level below 5 % v/v. In the first training session, panelists smelled the standards in unlabeled vials to identify the aroma, revealing the results at the end of the session. In the training sessions 2–6, panelists were asked to identify aromas in different orders and adding repetitions. In the seventh and eighth training sessions, panelists were asked to identify the presence or absence of the aroma in the tannin solutions proposed, following a check-all-that-apply (CATA) procedure (Adams, Williams, Lancaster, & Foley, 2007; Campo, Ballester, Langlois, Dacremont, & Valentin, 2010).

2.4.4. Sensory analysis

Tannin formulations were evaluated at 0.4 g/L concentration in both mineral water (pH 7.1, dry residue 22.1 mg/L; Pontevecchio, Luserna San Giovanni, Italy) and red wine (Terre del Barolo, Castiglione Falletto, Italy), the latter presenting the following base chemical characteristics: total acidity 5.47 g/L as tartaric acid, pH 3.45, alcohol strength 12.8 % v/v. The 17 tested tannin formulations were divided into four groups, planning a separate tasting session for each group in water and the same scheme was followed for wine evaluation. Therefore, assessors evaluated 4–5 tannins per session resulting in a total of four sessions for the water and four sessions for the wine assessments. In each session an external control (OET formulation not included in the study) was used, in duplicate, to evaluate assessor's performance, with a total of 6–7 samples evaluated per session by each assessor. Each panelist rated astringency and bitterness using the unstructured 10-cm scale range, and chose the aroma descriptors for each tannin sample from the list of descriptors selected in the training sessions and here proposed as CATA evaluation sheet.

All samples were coded with a three-digit random code and placed in randomized order. A constant volume of 30 mL of each sample was evaluated at room temperature in black glasses. To minimize fatigue, panelists were asked to rinse their mouth with water, eat a piece of unsalted cracker, and then rinse again with water between samples.

2.5. Wine chemical analysis after one-month storage

After one month from the OET addition, untreated and treated wines were analysed to evaluate the impact on the polyphenolic content, the antioxidant capacity, and the polymerisation of pigments. The analyses were performed in triplicate. For phenolic content, TPI was evaluated and expressed as mg gallic acid equivalents/L. The total anthocyanin content (TA) was measured as proposed by Torchio et al. (2010) after dilution in an ethanol:water:hydrochloric acid 37 % (70:30:1 v/v) mixture. The maximum value of absorbance between 520 and 540 nm was considered and results were expressed as mg malvidin-3-glucoside chloride equivalents/L. Polymeric pigments were evaluated by Adams-Harbertson method (Harbertson, Picciotto, & Adams, 2003), based on the reactivity with BSA protein and the colour loss by SO_2, and expressed as relative abundance of monomeric anthocyanins (MON %), small polymeric pigments (SPP %), and long polymeric pigments (LPP %).

The antioxidant capacity of wine after one-month storage was evaluated using the above-mentioned DPPH and FRAP assays after a 50-times dilution in water.

2.6. Statistical analysis

Data analysis was performed with R statistical software (R Foundation for Statistical Computing, Vienna, Austria). Normality and homogeneity of the variances were tested for all the parameters with Shapiro-Wilk and Levene tests, respectively. If assumptions were respected, parametric tests were applied (ANOVA and Tukey HSD post-hoc). If populations were not distributed normally Kruskal-Wallis and Conover-Holm were used whereas if they were not homoscedastic Welch's and Games-Howell were applied. Statistical correlations were evaluated as Pearson coefficient (r) with the R software package 'corrplot' (Wei & Simko, 2021).

Correspondence analysis (CA) of aroma descriptors in water and wine was performed on citation frequencies for each sample. Multivariate technique approach was attempted to establish relationships between the OETs characteristics in model wine solution/water and the ones found in wines. For this aim, Multifactorial Analysis (MFA) was performed using investigated OETs as individuals and model wine chemical and water sensory parameters as active variables. Four groups were created for model wine analyses: polyphenolic variables as *PF-tannin*, including TPI, FC, MTC, and BS; antioxidant capacity variables as *AC-tannin* including DPPH, FRAP, ABTS, and CUPRAC; astringency intensity (*Ast-tannin*) and bitterness intensity (*Bitt-tannin*). To understand OETs behaviour in wine, wine parameters were introduced as supplementary variables and similar groups were created: *PF-wine*, including TPI, *AC-wine*, including FRAP and DPPH, astringency and bitterness intensity (*Ast-wine* and *Bitt-wine*, respectively) and *Colour-wine*, for colour parameters including TA, MON %, SPP %, and LPP %. CA and MFA analysis were performed using R software with 'FactomineR' package (Lê, Josse, & Husson, 2008).

3. Results and discussion

3.1. Characterization of OETs formulations in model solutions

3.1.1. Polyphenolic content and characterization

Polyphenolic content of OETs used for the experiment, evaluated as TPI expressed as gallic acid, is shown in Table 2. Significant differences ($p < 0.001$) were found among individual samples in model wine solution. The richest sample was Gt (128.2 expressed as % w/w), whereas the lowest concentration corresponded to Sk1 (21.9 %). Gt had a polyphenolic content higher than 100 % due to the use of gallic acid reference standard. Although being the most similar standard to its tannic composition, Gt could have a more complex chemical structure that may cause a different absorbance value at 280 nm (Vignault et al., 2018; Motta, Guaita, Cassino, & Bosso, 2020). Except for the gallotannin-based formulation, Et1, Sd1, and Q1 were the only samples with a polyphenolic concentration above the group average (40.9 g gallic acid/100 g of product), and they belong to different chemical classes, highlighting the heterogeneity of OETs formulations. Nevertheless, the chemical classes were significantly different among themselves ($p < 0.01$) with the richest family in polyphenols given by Hydro group (70.2 %), being

Table 2
Polyphenolic characterization of OETs formulations under evaluation in model-wine solution.

Sample		TPI *g gallic acid/ 100 g*	FC *g gallic acid/ 100 g*	BS *g cyanidin/ 100 g*	MTC *g gallic acid /100 g*
Proc/prod	Sd1	48.7 ± 5.7bcde	89.8 ± 4.2b	116.4 ± 5.7a	36.5 ± 1.7b
	Sd2	28.7 ± 3.8bcde	53.3 ± 2.2ef	71.3 ± 3.8b	16.7 ± 3.0efg
	Sk1	21.9 ± 1.5e	33.1 ± 1.2g	33.9 ± 1.5c	6.0 ± 0.2g
Proc/prod Average		**33.1 ± 12.5 B**	**58.7 ± 25.0**	**73.9 ± 36.1 A**	**19.7 ± 13.5**
Prof/pror	Q1	41.0 ± 0.9bc	77.0 ± 5.5c	26.9 ± 0.9d	35.7 ± 2.6bc
	Q2	36.3 ± 2.1bcd	64.4 ± 4.9d	18.6 ± 2.1g	27.8 ± 0.9bcd
	Ac	32.9 ± 1.7cd	54.4 ± 3.5def	26.2 ± 1.7de	22.0 ± 3.4def
Prof/pror Average		**36.7 ± 3.8 AB**	**65.3 ± 10.6**	**23.9 ± 4.3 B**	**28.5 ± 6.4**
Hydro	Et1	44.8 ± 2.0b	52.2 ± 2.6ef		21.0 ± 7.3def
	Et2	37.5 ± 1.1bcd	52.7 ± 4.1ef		19.1 ± 2.7def
	Gt	128.2 ± 4.6a	101.4 ± 4.1a		110.4 ± 1.2a
Hydro Average		**70.2 ± 43.7 A**	**68.8 ± 24.7**		**50.2 ± 45.4**
Mix	Mx1	39.1 ± 0.9bc	62.8 ± 4.2de	25.9 ± 0.9de	29.1 ± 1.4bcd
	Mx2	36.6 ± 1.7bcd	58.8 ± 2.9def	14.9 ± 1.7h	25.8 ± 2.9cdef
	Mx3	36.8 ± 0.5bc	59.6 ± 4.0def	23.8 ± 0.5def	23.4 ± 3.2def
	Mx4	32.9 ± 1.8cd	51.4 ± 2.0f	22.2 ± 1.8ef	15.4 ± 5.7fg
	Mx5	32.2 ± 0.6d	51.4 ± 1.0f	20.9 ± 0.6fg	16.5 ± 7.3efg
	Mx6	35.2 ± 1.5cd	52.9 ± 1.4ef	10.6 ± 1.5i	29.2 ± 0.8bcd
	Mx7	28.2 ± 2.9cde	55.8 ± 2.6def	20.8 ± 2.9fg	25.7 ± 0.6cdef
	Mx8	34.0 ± 2.0cd	51.0 ± 4.8f	5.9 ± 2.0j	26.0 ± 0.8cde
Mix Average		**34.4 ± 3.5 B**	**55.5 ± 5.0**	**18.1 ± 6.7 C**	**23.9 ± 5.9**
Sign. Samples		*** $p < 0.001$	*** $p < 0.001$	*** $p < 0.001$	*** $p < 0.001$
Sign. Groups		** $p = 0.002$	ns $p = 0.199$	*** $p < 0.001$	ns $p = 0.289$

Data are expressed as average value ± standard deviation (n = 3). Data in bold are the average values of the Groups (Proc/prod, Prof/Pror, Hydro, and Mix, corresponding to OETs procyanidins/prodelphinidins, profisetinidins/prorobinetinidins, hydrolysable, and mix group, respectively). Sign: ns, *, **, *** were used for not significant, $p < 0.05$, $p < 0.01$, $p < 0.001$, respectively, according to ANOVA, Welch's, or Kruskal-Wallis within the data of the same column. Different lowercase letters within the same column refer to the existence of a significant difference between different samples according to Tukey's test, Games-Howell, or Conover-Holm whereas the presence of differences in uppercase letters within the same column means a significant difference among groups according to Tukey, Games-Howell, or Conover-Holm.
TPI = Total phenolic index; FC = Folin-Ciocalteu method; BS = Bate-Smith method, MTC = Methylcellulose method.

significantly different from Proc/prod group (33.1 %) and Mixed formulation group (34.4 %).

Folin-Ciocalteu assay (FC) seemed to be less discriminant than the previous one since no significant differences ($p > 0.05$) were reported among the average concentration of the four chemical families. When all individual products were considered, significant differences were found ($p < 0.001$). The richest formulate was Gt (101.4 %) followed by Sd1, Q1, and Q2, whereas Sk1 was the poorest (33.1 %). Tukey test highlighted a numerous group of samples from different chemical groups (Sd2, Ac, Et1, Et2, Mx2, Mx3, Mx4, Mx5, Mx6, Mx7, and Mx8) with similar FC value (%). The average concentration of polyphenols of analysed tannins, assessed by FC, was 60.1 g of gallic acid/100 g of product, which is in line with previous results reported by Magalhaes, Ramos, Reis, & Segundo (2014) and Vignault et al. (2018).

The Bate-Smith assay (BS), specific for condensed tannins, was tested as well on hydrolysable tannin formulations, however, as expected, proanthocyanidins were not found. Significant differences were shown among samples ($p < 0.001$) and groups ($p < 0.001$). Proanthocyanidin content was significantly higher in Proc/prod (73.9 %) than in Prof/pror (23.9 %): this behaviour is explained by the resistance to acid cleavage of profisetinidins and prorobinetinidins in contrast with procyanidins and prodelphinidins (Venter et al., 2012a). Instead, the mean proanthocyanidin content of Mix was significantly lower than others (18.1 %), in accordance with the presence in the formulation of components other than procyanidins and prodelphinidins, and a high variability was found with BS values ranging from 5.9 to 25.9%.

The fourth method used for polyphenolic characterization was the methylcellulose precipitation assay (MTC) proposed and tested by Sarneckis et al. (2006) on proanthocyanidins. However, it seems to be effective also on the other types of tannins, and to be more specific for tannins than TPI and FC (Vignault et al., 2018) in agreement with the lower concentration observed for MTC in the present study. The tannin content of OETs (MTC) had an average value of 28.6 g of gallic acid/100 g of product. The individual formulations tested had statistically different tannin contents ($p < 0.001$) and, except for the very high value of Gt (110.4 %), they had values ranging from 6 % (Sk1) to 36.5 % (Sd1). Moreover, it was not detected a significant difference ($p > 0.05$) between chemical families, as already reported also for FC assay.

In general, it is possible to highlight the great variability in polyphenolic content and features among OETs formulations, also within the same botanical family, in accordance with previous studies (Vignault et al., 2018; Motta et al., 2020). Moreover, the differences in chemical families could lead to an over- or under-estimation of the formulate richness when determined using a common reference standard and/or method. The use of different methods provides a fuller understanding of the composition of pure and mixed formulations, for instance the latter based on condensed tannin presence. Concerning the standard choice, the use of (-)-epicatechin or (+)-catechin is preferred to gallic acid when proanthocyanidins-based OET formulations are analysed, given their different absorptivity coefficient (Vignault et al., 2018; Gombau et al., 2019b; Motta et al., 2020). However, this could make it difficult to interpret the results when mixed-origin formulations are analysed, and furthermore specific analysis may be required for both the determination of total content and the compositional characterization (OIV, 2015).

3.1.2. Antioxidant properties

Polyphenolic content and composition of OETs deeply influence their other properties (Versari et al., 2013), in particular the antioxidant capacity, which is one of the most sought actions when OETs are used in wine industry. OET formulations are proposed in this sense as processing aids to protect against oxidation and to preserve juice and wine compounds, particularly the anthocyanins responsible for wine colour from the first stage of vinification (OIV, 2019a, 2019b). Nevertheless, antioxidant activity is very complex to be fully quantified due to the numerous mechanisms involved. In fact, to have a complete view of the antioxidant capacity of a OETs formulate, or in general of a vegetal extract, several assays are recommended (Apak et al., 2007) as each method determines the antioxidant capacity under specific conditions by assessing different oxidant species. Therefore, the results of different methods are not comparable because often they are not correlated (Apak et al., 2007).

Table 3 shows the antioxidant capacity values of OETs measured with different analytical methods (ABTS, DPPH, FRAP, CUPRAC) and expressed as mmol Trolox equivalents/g of formulate (Magalhaes et al., 2014). All the chosen methods evaluate the ability of a molecule to act as antioxidant transferring electrons; ABTS and DPPH evaluate the capacity of shifting electrons to a radical, whereas FRAP and CUPRAC to a metallic ion in the oxidized state (Apak et al., 2007). Significant differences among samples ($p < 0.001$) and groups ($p < 0.001$) for antioxidant capacity were found when they were analysed with the different methods. OETs had average values of 3.91, 2.98, 3.90, and 6.14 mmol Trolox/g of product for ABTS, DPPH, FRAP, and CUPRAC, respectively. The hydrolysable tannin group (Hydro) had an antioxidant capacity significantly higher than the other chemical classes for all the analytical methods, except for DPPH where the Hydro antioxidant capacity remained higher, but it was not significantly different from exotic wood tannin group (Prof/pror), as already found by Vignault et al. (2018). Within the Hydro family, Gt was the most antioxidant formulate

according to all the assays except for CUPRAC where it had a similar value to Sd1, Q1, Et1, Mx1, and Ac. For the FRAP assay, Et1 was the most antioxidant OET (5.51 mmol Trolox/g of product). In fact, FRAP is the only method not significantly correlated with TPI and FC ($p > 0.05$, Fig. 1, **Table S1**).

Condensed tannins seemed to have less ability in radical scavenging (ABTS and DPPH) than hydrolysable ones, whereas in the reduction of metal ions some formulates seemed to behave similarly to hydrolysable tannins. Indeed, considering FRAP assay, Sd1 and Sd2 were not different ($p > 0.05$) to Et2 and Gt. As well, in CUPRAC assay, Gt showed the highest antioxidant capacity, followed by Sd1, Q1, Et1, Mx1, and Ac ($p > 0.05$). However, the proanthocyanidin tannin from the skin (Sk1) showed the lowest values of antioxidant capacity for the different methods used. Lastly, within mixed formulations, Mx1 and Mx8 had the highest ABTS values: these two were, respectively, the richest Mix formulate in polyphenols (Mx1) and the poorest Mix formulate in proanthocyanidins (Mx8; Table 2). Furthermore, within Mix group, Mx8 showed the highest antioxidant capacity according to DPPH assay whereas Mx1 and Mx8 showed the highest CUPRAC values. This different behaviour among mixed OETs may be due to the different composition in phenolics, besides from their content, and reflects the different antioxidant ability of the different types of pure tannins that compose them.

To better estimate the antioxidant capacity of different tannins, the *antioxidant potency* (*AP*) of each sample was calculated as standardized antioxidants capacity on the total phenolic concentration measured as FC. This standardization can help in understanding if certain tannin classes are more antioxidant than others. Significant differences in *AP* were present among samples ($p < 0.001$), as well as among groups ($p < 0.001$), with exception of FRAP ($p = 0.064$). Hydro family had the significantly higher *AP* values ($p < 0.001$) in all the assays except for FRAP, but samples Et1 had the highest *AP* value also in FRAP. In fact, the formulations containing ellagitannins (Et1 and Et2) had the highest *AP* values for ABTS and FRAP assays, supporting the previous findings that this class is the most antioxidant tannin family (Vivas & Glories, 1996; Vignault et al. 2018). Moreover, within the pure OETs extracted from exotic woods, Ac (from acacia) had an *AP* significantly higher than Q1 and Q2 (from quebracho).

3.2. Sensory analysis and chemical determination of astringency

Sensory assessment of OETs was performed in water and in red wine, at dose of 0.4 g/L, which can be considered a substantial dose for most tannin formulations. Water was preferred to model wine to reduce alcohol effect which may mask the perception and increase the carry-over effect (Saenz-Navajas et al., 2017). Bitterness and astringency in-mouth properties were assessed. In Table 4**,** ANOVA shows significant differences for both sensory properties assessed among samples in water, however the perception of bitterness and astringency was not different among groups ($p > 0.05$). In detail, the bitterest formulates were Q1 (4.52) and Gt (3.78), which were significantly different ($p < 0.01$) from the lowest bitterness perceived Et1 (0.44) and Sk1 (0.58). Average bitterness intensity of Prof/pror was higher than the overall mean value (2.00), although this value was not significantly different when compared to Proc/prod, Hydro, or Mix groups ($p > 0.05$). Concerning astringency intensity in water, the average value was 3.66 and a statistical difference ($p < 0.05$) was found between the most (Q1, 5.40) and the least (Sk1, 1.85) astringent tannin; the other OETs, instead, were grouped together without significant differences neither with Q1 or Sk1. Lastly, Prof/pror was the only chemical family with an astringency average value above the mean value, although it was not significantly different ($p > 0.05$) from the average astringency of the other classes. In red wine addition tests, bitterness was similar among samples and among groups ($p > 0.05$) whereas astringency was found to be different

Table 3
Antioxidant capacity and *antioxidant potency (AP)* of OETs formulations under evaluation in model-wine solution.

Sample		ABTS *mmol Trolox/g*	DPPH *mmol Trolox/g*	FRAP *mmol Trolox/g*	CUPRAC *mmol Trolox/g*	*ABTS AP*	*DPPH AP*	*FRAP AP*	*CUPRAC AP*
Proc/prod	Sd1	4.52 ± 0.02c	3.08 ± 0.04d	4.78 ± 0.02b	7.76 ± 0.29ab	5.04 ± 0.23g	3.43 ± 0.20g	5.33 ± 0.23ghi	8.66 ± 0.61de
	Sd2	3.70 ± 0.03f	2.57 ± 0.02jkln	4.55 ± 0.06b	5.85 ± 0.26cdefg	6.95 ± 0.24cde	4.83 ± 0.23bcde	8.53 ± 0.42bc	10.96 ± 0.15bcd
	Sk1	2.02 ± 0.04k	1.39 ± 0.04q	2.05 ± 0.05h	2.84 ± 0.15i	6.11 ± 0.29defg	4.19 ± 0.13defg	6.21 ± 0.17efg	8.59 ± 0.59e
Proc/prod Average		**3.41 ± 1.10 B**	**2.35 ± 0.75 BC**	**3.79 ± 1.31 B**	**5.48 ± 2.16 B**	**6.03 ± 0.86 B**	**4.15 ± 0.63 B**	**6.69 ± 1.45**	**9.41 ± 1.25 B**
Prof/pror	Q1	4.01 ± 0.04e	2.76 ± 0.06efghijkl	3.38 ± 0.13defg	7.23 ± 0.29abc	5.22 ± 0.34g	3.60 ± 0.32fg	4.39 ± 0.31i	9.41 ± 0.66cde
	Q2	3.37 ± 0.05i	2.34 ± 0.01p	3.58 ± 0.02efg	5.54 ± 0.23fghi	5.23 ± 0.38g	3.63 ± 0.26fg	5.56 ± 0.42fgh	8.64 ± 0.99e
	Ac	4.02 ± 0.06e	2.80 ± 0.04efg	3.73 ± 0.13cde	6.07 ± 0.13abcde	7.41 ± 0.45bc	5.17 ± 0.39bc	6.87 ± 0.29de	11.20 ± 0.93bc
Prof/pror Average		**3.80 ± 0.33 B**	**2.63 ± 0.23 AB**	**3.56 ± 0.18 B**	**6.28 ± 0.77 B**	**5.95 ± 1.14 B**	**4.13 ± 0.83 B**	**5.61 ± 1.11**	**9.75 ± 1.36 B**
Hydro	Et1	4.95 ± 0.01b	4.02 ± 0.02b	5.51 ± 0.02a	7.07 ± 0.09abc	9.51 ± 0.50a	7.73 ± 0.41a	10.58 ± 0.57a	13.59 ± 0.85a
	Et2	4.40 ± 0.04d	3.62 ± 0.02c	4.75 ± 0.10b	5.92 ± 0.08bcdef	8.39 ± 0.71ab	6.89 ± 0.54a	9.04 ± 0.55b	11.28 ± 0.86bc
	Gt	7.59 ± 0.02a	7.60 ± 0.01a	4.75 ± 0.03b	12.10 ± 0.06a	7.49 ± 0.3bc	7.51 ± 0.30a	4.69 ± 0.17hi	11.94 ± 0.45ab
Hydro Average		**5.65 ± 1.58 A**	**5.08 ± 1.90 A**	**5.01 ± 0.38 A**	**8.36 ± 2.84 A**	**8.46 ± 0.99 A**	**7.37 ± 0.53 A**	**8.11 ± 2.68**	**12.27 ± 1.22 A**
Mix	Mx1	3.77 ± 0.01f	2.69 ± 0.01gh	4.17 ± 0.06c	6.16 ± 0.19abcd	6.02 ± 0.42efg	4.31 ± 0.31cdefg	6.66 ± 0.49def	9.83 ± 0.42bcde
	Mx2	3.34 ± 0.02ij	2.48 ± 0.04hijklmnopq	3.59 ± 0.07def	5.43 ± 0.19ghi	5.68 ± 0.24fg	4.22 ± 0.26defg	6.11 ± 0.39efg	9.24 ± 0.30cde
	Mx3	3.35 ± 0.01ij	2.43 ± 0.03kmnpq	3.65 ± 0.14cdefg	5.60 ± 0.21efgh	5.63 ± 0.38fg	4.09 ± 0.29efg	6.14 ± 0.45efg	9.43 ± 0.76cde
	Mx4	3.50 ± 0.04gh	2.56 ± 0.04hijklmnopq	3.87 ± 0.07cde	5.21 ± 0.31hi	6.83 ± 0.35cdef	4.98 ± 0.15bcde	7.54 ± 0.44cd	10.14 ± 0.23bcde
	Mx5	3.55 ± 0.02g	2.58 ± 0.01fijkm	3.92 ± 0.04cd	5.09 ± 0.77ghi	6.91 ± 0.15cde	5.02 ± 0.08bcd	7.62 ± 0.21cd	9.91 ± 1.51bcde
	Mx6	3.25 ± 0.03j	2.47 ± 0.01mnop	3.07 ± 0.10fg	5.41 ± 0.19ghi	6.15 ± 0.14defg	4.67 ± 0.12bcde	5.80 ± 0.07efgh	10.24 ± 0.60bcde
	Mx7	3.42 ± 0.02hi	2.45 ± 0.02imnopq	3.11 ± 0.09g	5.51 ± 0.10fghi	6.14 ± 0.26defg	4.40 ± 0.20cdef	5.58 ± 0.12fgh	9.88 ± 0.57bcde
	Mx8	3.69 ± 0.02f	2.83 ± 0.02e	3.82 ± 0.08cde	5.63 ± 0.07defgh	7.28 ± 0.76bcd	5.57 ± 0.51b	7.53 ± 0.61cd	11.09 ± 1.08bc
Mix Average		**3.48 ± 0.17 B**	**2.56 ± 0.13 C**	**3.65 ± 0.38 B**	**5.50 ± 0.41 B**	**6.33 ± 0.66 B**	**4.66 ± 0.53 B**	**6.62 ± 0.87**	**9.97 ± 0.85 B**
Sign. Samples		***	***	***	***	***	***	***	***
		$p < 0.001$	$p < 0.001$	$p < 0.001$	$p < 0.001$	$p < 0.001$	$p < 0.001$	$p < 0.001$	$p < 0.001$
Sign. Groups		***	***	***	***	***	***	ns	***
		$p < 0.001$	$p < 0.001$	$p < 0.001$	$p < 0.001$	$p < 0.001$	$p < 0.001$	$p = 0.064$	$p < 0.001$

Data are expressed as average value ± standard deviation (n = 3). Data in bold are the average values of the Groups (Proc/prod, Prof/Pror, Hydro, and Mix, corresponding to OETs procyanidins/prodelphinidins, profisetinidins/prorobinetinidins, hydrolisable, and mix group, respectively). Sign: ns, *, **, *** were used for not significant, $p < 0.05$, $p < 0.01$, $p < 0.001$, respectively, according to ANOVA, Welch's, or Kruskal-Wallis within the data of the same column. Different lowercase letters within the same column refer to the existence of a significant difference between different samples according to Turkey's test, Games-Howell, or Conover-Holm whereas the presence of differences in uppercase letters within the same column means a significant difference among groups according to Tukey, Games-Howell, or Conover-Holm.
ABTS, DPPH, FRAP, and DPPH AP = Antioxidant potency, calculated as antioxidant capacity of each test / total phenolic content (as FC, mg/g gallic acid)) *1000.

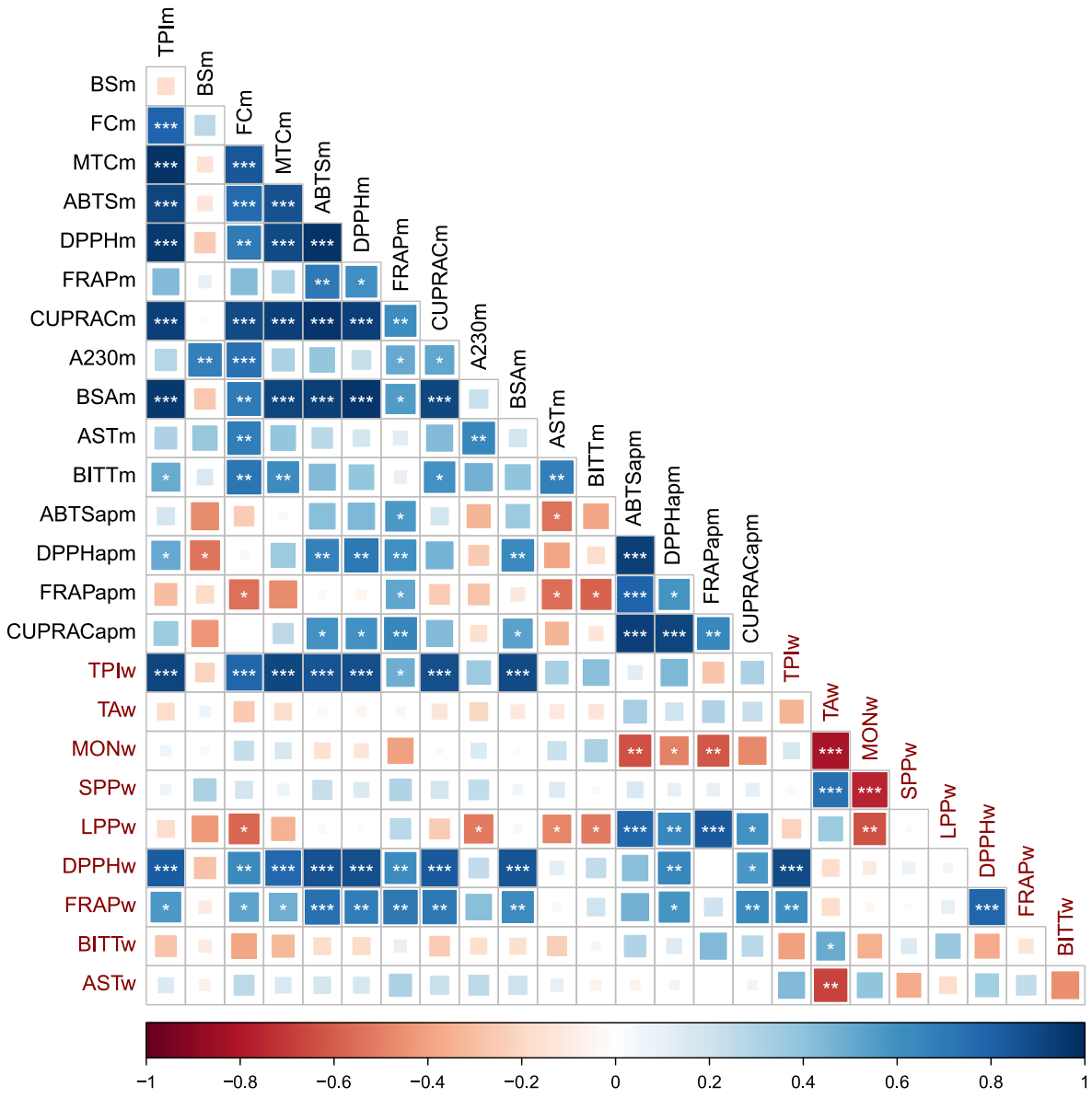

Fig. 1. Pearson's correlation (*r*) of the OETs investigated parameters in model solution (water or model wine) and wine. TPI = Total phenolic index, BS = Bate-Smith method, FC = Folin-Ciocalteu method, MTC = Methylcellulose method, AST = sensory analysis intensity of astringency, BITT = sensory analysis intensity of bitterness, TA = Total anthocyanins; MON, SPP, and LPP = percentage of monomeric forms, percentage of small polymeric pigments, and percentage of long polymeric pigments for Adams-Harbertson method, respectively. ABTSap, DPPHap, FRAPap, CUPRACap, where ap = antioxidant potency, calculated as antioxidant capacity of each test / total phenolic content (as FC, mg/g gallic acid)) *1000. Letter "m" represents parameters evaluated on formulate dissolved in model wine (for chemical analysis) or water (for sensory analysis), and letter "w" represents parameters evaluated on wine after one-month storage from formulate addition (0.4 g/L).

among samples ($p = 0.036$) and among groups ($p = 0.041$), even if the post-hoc test applied did not underline any difference. It highlights that, in a more complex matrix, the added dose (0.4 g/L of OET) was only slightly perceivable in terms of in-mouth related properties, probably due to the high content of polyphenols in red wine. In fact, tannin-added wines were usually bitter and more astringent than the control (red wine without additions), even though significant differences were not observed.

Sensory bitterness and astringency ratings in water were also standardized for phenolic content determined by Folin-Ciocalteu method (Tables 2 and 4), and the bitterness perception was in line with the unstandardised results (Table 4). Significant differences in standardised bitterness were only reported between Q1 and Et1 ($p < 0.05$), showing Q1 the highest value (5.87) and Et1 the lowest (0.84) one. The higher bitterness of quebracho tannins with respect to the other families was already reported (Puech et al., 2007), although further research may be useful regarding the effect of wine composition on the final sensation. In contrast, standardised astringency was not found significantly different, with the tannin content being the most impactful driver on the tannin types.

Alternative methods to predict astringency, avoiding panel training and tasting sessions, may help in a quicker evaluation of tannin formulations. With this aim, together with sensory analysis, a physico-chemical determination of indexes known for astringency prediction was performed. Among them, the BSA assay is a tannin precipitation method that is highly correlated with wine astringency (Boulet et al., 2016), and therefore used to investigate a possible correlation with sensory ratings. In our study, statistical correlation between BSA assay and astringency was not significant ($r = 0.188$, $p > 0.05$, Fig. 1) highlighting a poor effectiveness of this method for astringency prediction on OETs because, differently from wine that has mainly proanthocyanidin-based tannins, OETs are composed of several chemical families that own

Table 4
Chemical determination of astringency and sensory analysis of OETs formulations.

Sample		Physico-Chemical astringency analysis				Sensory analysis intensities				
		BSA assay	**A280**	**A230**	**Bitterness[a]**	**Astringency[a]**	**Bitterness[b]**	**Astringency[b]**	**Bitterness[c]**	**Astringency[c]**
		ΔA280l/g	*A280 /g*	*A230l/g*	*0.4 g of OET/L in water*	*0.4 g of OET/L in water*	*standardized on g of polyphenols*	*standardized on g of polyphenols*	*0.4 g of OET/L in red wine*	*0.4 g of OET/L in red wine*
Red wine without addition									1.73 ± 0.60	4.09 ± 0.46a **A**
Proc/prod	Sd1	4.58 ± 1.00bcd	17.1 ± 2.0bcde	66.9 ± 7.1a	2.49 ± 0.71ab	5.28 ± 0.96ab	2.77 ± 0.79ab	5.88 ± 1.06	1.75 ± 0.75	6.45 ± 2.65a
	Sd2	3.89 ± 0.81cd	10.1 ± 1.3bcde	40.7 ± 4.2bc	2.74 ± 1.27ab	3.70 ± 0.60ab	5.14 ± 2.37ab	6.94 ± 1.12	3.04 ± 0.78	5.17 ± 0.63a
	Sk1	0.96 ± 0.12e	7.7 ± 0.5e	26.7 ± 1.8d	0.58 ± 0.34b	1.85 ± 0.68b	1.75 ± 1.03ab	5.59 ± 2.04	2.20 ± 0.72	3.98 ± 0.61a
Proc/prod Average		**3.14 ± 1.78 B**	**11.6 ± 4.41 B**	**44.8 ± 18.2**	**1.94 ± 1.18**	**3.61 ± 0.99**	**3.26 ± 1.72**	**6.14 ± 0.71**	**2.33 ± 0.65**	**5.20 ± 1.24 A**
Prof/pror	Q1	3.06 ± 0.25d	14.4 ± 0.3bc	46.8 ± 4.7ab	4.52 ± 1.00a	5.40 ± 0.76a	5.87 ± 1.30a	7.01 ± 0.99	1.95 ± 0.56	5.14 ± 0.38a
	Q2	4.16 ± 0.17cd	12.8 ± 0.7bcd	40.8 ± 1.0bc	2.08 ± 0.73ab	4.48 ± 0.54ab	3.82 ± 1.35ab	8.24 ± 0.99	2.15 ± 0.88	6.26 ± 0.97a
	Ac	4.13 ± 0.61cd	11.6 ± 0.6cd	40.5 ± 2.2bc	1.54 ± 0.52ab	3.24 ± 0.39ab	2.38 ± 0.80ab	5.02 ± 0.61	3.20 ± 0.99	3.57 ± 0.61a
Prof/pror Average		**3.80 ± 0.64 B**	**12.9 ± 1.3 AB**	**42.7 ± 4.1**	**2.71 ± 1.59**	**4.37 ± 0.62**	**4.02 ± 1.75**	**6.73 ± 1.62**	**2.43 ± 0.67**	**4.99 ± 1.35 A**
Hydro	Et1	6.07 ± 1.19b	15.8 ± 0.7b	40.1 ± 1.7bc	0.44 ± 0.11b	2.30 ± 0.86ab	0.84 ± 0.22b	4.41 ± 1.65	2.20 ± 1.40	7.25 ± 0.65a
	Et2	4.25 ± 0.08cd	13.2 ± 0.4bcd	36.6 ± 0.9cd	1.44 ± 0.56ab	2.93 ± 0.69ab	3.42 ± 1.08ab	5.56 ± 1.30	2.78 ± 0.55	6.00 ± 0.75a
	Gt	13.68 ± 0.88a	45.1 ± 1.6a	42.9 ± 1.7ab	3.78 ± 0.76a	4.42 ± 0.66ab	3.72 ± 0.75ab	4.36 ± 0.65	1.78 ± 0.63	6.00 ± 0.96a
Hydro Average		**8.00 ± 4.39 A**	**24.7 ± 15.4 A**	**39.9 ± 3.0**	**1.88 ± 1.71**	**3.22 ± 0.63**	**2.66 ± 1.58**	**4.77 ± 0.68**	**2.25 ± 0.50**	**6.42 ± 0.72 A**
Mix	Mx1	3.93 ± 0.32cd	13.7 ± 0.3bc	44.3 ± 1.2ab	2.03 ± 0.33ab	3.84 ± 0.48ab	3.23 ± 0.53ab	6.12 ± 0.76	2.65 ± 0.50	6.22 ± 0.69a
	Mx2	4.42 ± 0.37bcd	12.9 ± 0.6bcd	40.4 ± 1.8bc	2.45 ± 0.65ab	3.00 ± 0.34ab	4.16 ± 0.38ab	5.10 ± 0.58	2.50 ± 2.00	5.95 ± 1.35a
	Mx3	4.15 ± 0.33cd	13.0 ± 0.2bc	42.8 ± 0.5ab	1.84 ± 0.44ab	3.88 ± 0.96ab	3.09 ± 0.76ab	6.51 ± 1.62	2.25 ± 1.55	8.40 ± 1.30a
	Mx4	3.82 ± 0.50cd	11.6 ± 0.6cd	36.8 ± 1.4cd	1.12 ± 0.29ab	4.29 ± 0.44ab	2.18 ± 0.52ab	8.34 ± 0.86	2.70 ± 0.58	4.86 ± 0.41a
	Mx5	3.87 ± 0.37cd	11.3 ± 0.2d	35.5 ± 0.3d	1.43 ± 0.38ab	3.94 ± 0.93ab	2.77 ± 0.74ab	7.67 ± 1.67	1.58 ± 0.73	5.62 ± 0.65a
	Mx6	4.07 ± 0.16c	12.4 ± 0.5cd	35.4 ± 1.0d	2.26 ± 0.84ab	4.67 ± 0.67ab	4.27 ± 0.73ab	8.82 ± 1.27	2.55 ± 0.50	6.03 ± 0.51a
	Mx7	3.47 ± 0.27cd	9.9 ± 1.0cde	33.5 ± 3.6d	0.96 ± 0.23ab	2.80 ± 0.62ab	1.72 ± 0.49ab	5.02 ± 1.11	1.25 ± 0.85	7.35 ± 1.45a
	Mx8	5.06 ± 0.88bc	12.0 ± 0.7cd	35.9 ± 2.1cd	2.26 ± 1.02ab	2.45 ± 0.89ab	4.43 ± 0.98ab	4.80 ± 1.75	2.97 ± 0.62	5.34 ± 0.84a
Mix Average		**4.10 ± 0.59 B**	**12.1 ± 1.2 B**	**38.1 ± 4.0**	**1.79 ± 0.20**	**3.61 ± 0.27**	**3.23 ± 1.00**	**6.55 ± 1.57**	**2.31 ± 0.59**	**6.22 ± 1.14 A**
Sign. Samples		*** $p < 0.001$	*** $p < 0.001$	*** $p < 0.001$	** $p = 0.002$	* $p = 0.013$	* $p = 0.016$	ns $p = 0.249$	ns $p = 0.954$	* $p = 0.036$
Sign. Groups		** $p = 0.003$	** $p = 0.002$	ns $p = 0.135$	ns $p = 0.123$	ns $p = 0.317$	ns $p = 0.136$	ns $p = 0.237$	ns $p = 0.995$	* $p = 0.041$

Chemical analysis data are expressed as average value ± standard deviation (n = 3) in absorbance units for bovine serum albumin (BSA) assay, absorbance at 280 nm (A280), absorbance at 230 nm (A230) corrected for g of OETs formulations. Bitterness and astringency sensory analysis scores (0–10 scale) in water[a] and wines[c] added at 0.4 g/L of OET are expressed as average value ± errors calculated as $s/(n)^{1/2}$ (s, standard deviation; n, number of panelists), where groups are represented as average value ± standard deviation (n = samples). Bitterness[b] and Astringency[b] are calculated in water standardized on phenolic content determined with Folin-Ciocalteu method.

Data in bold are the average values of the Groups (Proc/prod, Prof/Pror, Hydro, and Mix, corresponding to OETs procyanidins/prodelphinidins, profisetinidins/prorobinetinidins, hydrolysable, and mix group, respectively). Sign: ns, *, **, *** were used for not significant, $p < 0.05$, $p < 0.01$, $p < 0.001$, respectively, according to ANOVA, Welch's, or Kruskal-Wallis within the data of the same column. Different lowercase letters within the same column refer to the existence of a significant difference between different samples according to Tukey's test, Games-Howell, or Conover-Holm whereas the presence of differences in uppercase letters within the same column means a significant difference among groups according to Tukey, Games-Howell, or Conover-Holm.

different affinity to BSA (Hofmann et al., 2006). Also, absorbance at 280 nm (A280) and 230 nm (A230) were attempted to be correlated to sensory data. Significant correlations between *A230-Astringency* ($p < 0.01$, $r = 0.645$) and *TPI-Bitterness* ($p < 0.05$, $r = 0.499$) were found, but TPI (as A280) was not significantly correlated with astringency. Therefore, TPI (as A280) is not a good predictor of astringency for OETs, mainly because it is influenced also by other polyphenolic compounds that are not strictly involved in the astringency perception (Boulet et al., 2016). On the contrary, A230 could have potentialities also in the prediction of OETs astringency, given the significant correlation with the

sensory assessment.

Concerning aroma descriptors of OETs in water, assessed by CATA method, the ones perceived by tasters (with a frequency higher than 15 %) in more than one sample were *wood*, *caramel*, *licorice*, and *balsamic*. Generally, *wood* was the most common descriptor, and the most intense samples were Mx4, Et2, and Mx3 with a perceived frequency of 55.6 %, 50.0 %, and 46.7 %, respectively (**Table S2**). The Hydro group showed the highest *wood* average frequency (33.4 %). Regarding the other aroma descriptors, *licorice* was detected only in Mix group, whereas *caramel* was reported in samples from Mix and Prof/pror groups reaching an average frequency of 20.6 % and 20.1 %, respectively. A correspondence analysis (CA) of the perceived aromas in water and wine is represented in Fig. 2, separated by the media used for sensory analysis; for water (Fig. 2**A**), Dimension 1 (Dim 1) represented the 66.64 % of total variance and it was positively correlated with *balsamic* descriptor and negatively with *licorice* and *caramel*, whereas Dimension 2 (Dim 2) was positively correlated with *licorice* aroma. The most common descriptor found, namely *wood*, was located at the center of the plot, and therefore it was not correlated with any of the two dimensions obtained. Tannin groups (e.g., Hydro, Prof/pror, Proc/prod, and Mix) were separated from Dimension 1, being Hydro group different from the Prof/pror and Mix groups. This confirms the former is closely related to the *balsamic* descriptor, whereas the latter was more characterized by *licorice* and *caramel* notes. In fact, *balsamic* aroma descriptor was detected only in Sd1, Et2, and Gt. Two samples, Et1 and Ac, did not evoke any aroma in water and therefore they were not included in the CA. Fig. 2**B** shows the CA of wine aroma: Dimension 1 accounted for the 36.31 % of variance, whereas the Dimension 2 reached 25.58 %. *Orange* and *pepper* descriptors particularly influenced the positive side of Dimension 1, and *mushroom* and *caramel* did the negative one. Dimension 2 was correlated with *mushroom* aroma. Interestingly, *balsamic* descriptor was not cited in the assessment of wines added with OETs, instead the frequency of *wood* descriptor was the highest also in wine, with the higher average perception for Hydro group, in particular for Gt (66.7%). The Hydro group had also high *vanilla* frequency citations, reaching 60.0 % and 20.0 % for ellagitannins Et1 and Et2, respectively. Interestingly, when the aroma association between the two matrices (water and wine) was tested through the *RV* coefficient (Escoufier, 1973), OETs did not share similar position ($RV = 0.128$, $p > 0.05$), thus the addition of OETs in red wine at the tested dose did not influence its final aroma perception.

3.3. Analysis of wines added with OETs after one-month storage

After OETs addition (0.4 g/L), wines were stored for one month at room temperature in sealed dark bottles, and then analysed to assess polyphenolic and antioxidant characteristics, including colour as total anthocyanins (TA) and their polymerization, as well as the modifications with respect to the red wine subjected to the same storage period without OETs addition (Table 5). After the storage period TPI values in wine were significantly correlated with the results of the same analysis in model wine solution ($p < 0.001$; $r = 0.915$) (Fig. 1). The control wine had the lowest polyphenolic content as TPI (1455 mg gallic acid/L) and Gt, being the richest formulate, resulted in a 30 % increase over the control ($p < 0.001$), whereas Sk1 only increased just about 1 %. All chemical groups had an average TPI value significantly higher than the control ($p < 0.001$) except for Proc/prod formulates that showed a non-significant increase.

The results of DPPH assay in wine and model wine solution were also significantly correlated ($p < 0.001$, $r = 0.870$) (Fig. 1). Gt was the most antioxidant formulation in wine and it was significantly different from the control ($p < 0.01$). Hydro was the only family significantly higher than the control, highlighting that hydrolysable tannins are the most efficient formulates in radical scavenging, also in wine. FRAP assay confirmed Hydro group as the most antioxidant, with a remarkable value in Et1 (10.50 mmol Trolox/L), as already noted in model wine solution (Table 3); the variation for Prof/pror formulates was statistically similar to that corresponding to Hydro one, and Mix group was also significantly higher than the control ($p < 0.001$). Previously, Vazallo-

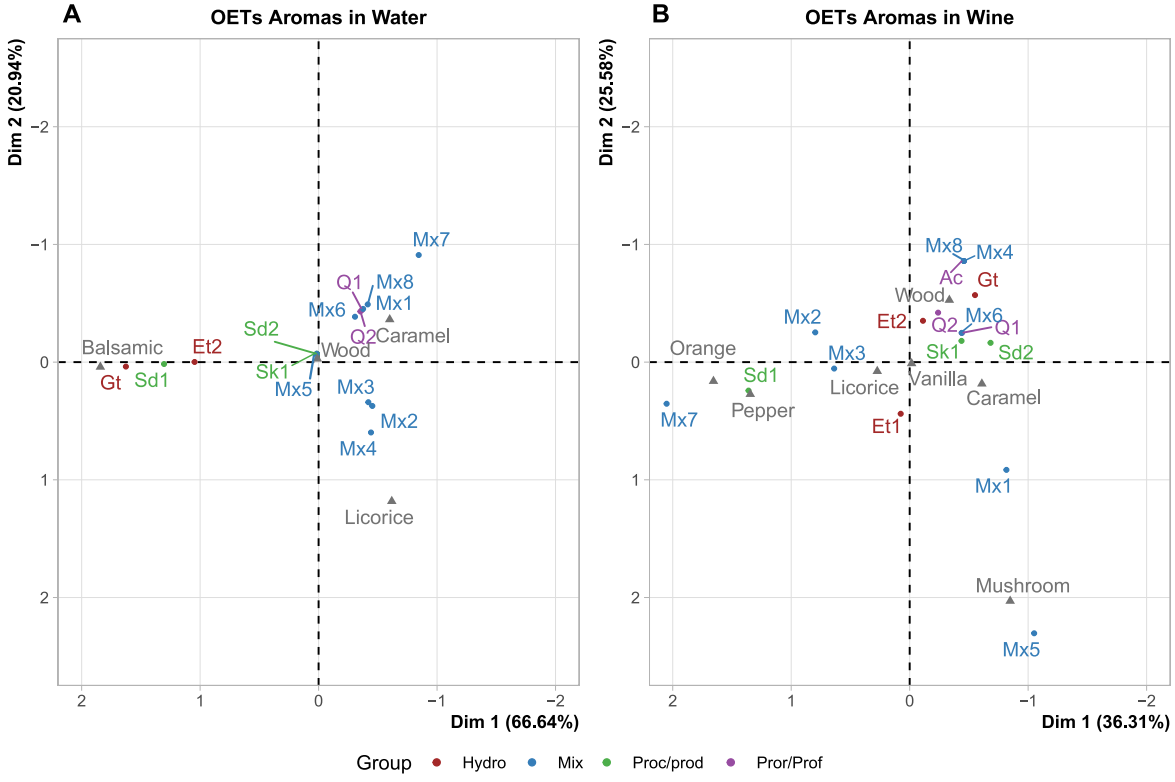

Fig. 2. Correspondence Analysis of OETs aroma in water (A) and wine (B). Hydro = hydrolysable tannins; Mix = mixed formulation; Proc/prod = procyanidins/prodelphinidins; Prof/pror = prorobinetinidins/profisenitinids. The aroma descriptors are plotted with gray triangles and gray text labels.

Valleumbrocio et al. (2017) noted evident increases of antioxidant capacity in wines added with oenological tannins after 5 days, however after 45 days of storage the differences were flattened, and no significant ones were found after 90 days between control and added wines. Therefore, after one month it is probable that the antioxidant effect of the added OETs is still appreciable, in line with higher phenolic contents of added wines.

Red wine colour-involved compounds were evaluated, such as the content of total anthocyanins and their polymerization ratio (Harbertson et al., 2003). Although no significant differences in the concentration of total anthocyanins (TA) between individual OETs-added wines and the control were found after one month from the addition ($p = 0.073$), the Mix group reported significantly lower average value of TA than the untreated wine ($p < 0.05$). In this last case, OETs seem not to have prevented TA losses, on the contrary leading to a decrease with respect to the control, as previously found after finished wine storage for 45 and 90 days (Vazallo-Valleumbrocio et al., 2017). Considering pigment polymerization, OETs addition affected these parameters to a different extent. Mixed formulations had, on average, a percentage of monomeric pigments (MON %) significantly higher than Hydro group ($p < 0.05$), in particular samples Mx3, Mx2, and Mx7 showing the highest percentages (41.9, 41.3, and 38.7 %, respectively) even with respect to control. Also Q1 preserved the monomeric forms (39.3 %), which may be given by known antioxidant activity of quebracho tannins (Vignault et al., 2018). By contrast, the lowest monomeric forms ratio was found in Et2 (33.4 %), in line with the capacity of ellagitannins to favour

Table 5
Chemical characterization of control wine and wine samples added with OETs formulation under evaluation after one month.

Sample		TPI *mg gallic acid/L*	DPPH *Mmol Trolox/L*	FRAP *Mmol Trolox/L*	TA *mg malvidin-3-O-glucoside chloride/L*	MON% *(Adams-Harbertson assay)*	SPP% *(Adams-Harbertson assay)*	LPP% *(Adams-Harbertson assay)*
Red wine without addition		1455 ± 25e **B**	11.32 ± 0.13bcd **B**	7.95 ± 0.40e **C**	194 ± 4 **A**	35.9 ± 1.8efg **AB**	29.5 ± 1.5a **A**	34.6 ± 1.9abc
Proc/prod	Sd1	1644 ± 41b	11.84 ± 0.19abc	9.38 ± 0.32bc	185 ± 6	37.1 ± 0.1abcdef	29.3 ± 0.4a	33.6 ± 0.4c
	Sd2	1499 ± 46cde	10.97 ± 0.28cd	8.61 ± 0.25cde	190 ± 9	35.7 ± 1.4abcdefg	28.3 ± 0.1abc	35.9 ± 1.4abc
	Sk1	1468 ± 51de	10.68 ± 0.09d	7.91 ± 0.25e	186 ± 4	37.2 ± 2.0abcdefg	26.6 ± 0.5cd	36.2 ± 1.7abc
Proc/prod Average		**1537 ± 91 AB**	**11.16 ± 0.55 B**	**8.63 ± 0.68 BC**	**187 ± 6 AB**	**36.7 ± 1.4 AB**	**28.1 ± 1.3 AB**	**35.2 ± 1.7**
Prof/pror	Q1	1584 ± 48bcd	11.63 ± 0.43abcd	9.27 ± 0.20bc	182 ± 1	39.3 ± 0.3bc	26.5 ± 0.2cd	34.2 ± 0.3bc
	Q2	1603 ± 41bc	11.72 ± 0.32abcd	8.40 ± 0.22cde	185 ± 8	36.6 ± 3.6abcdefg	28.9 ± 1.0ab	34.5 ± 4.6abc
	Ac	1498 ± 27cde	11.04 ± 0.64bcd	8.92 ± 0.30cd	192 ± 2	35.8 ± 1.0cdefg	29.2 ± 1.9ab	35.0 ± 1.9abc
Prof/pror Average		**1561 ± 59 A**	**11.43 ± 0.54 B**	**8.86 ± 0.43 AB**	**186 ± 6 AB**	**37.2 ± 2.5 AB**	**28.2 ± 1.7 A**	**34.6 ± 2.5**
Hydro	Et1	1663 ± 27b	13.16 ± 0.22ab	10.50 ± 0.45a	182 ± 3	35.4 ± 2.5abcdefg	26.4 ± 0.7cd	38.5 ± 2.7abc
	Et2	1572 ± 25bcd	11.45 ± 0.16abcd	8.91 ± 0.19cd	191 ± 7	33.4 ± 0.3g	29.2 ± 0.1a	37.5 ± 0.3a
	Gt	1894 ± 58a	13.82 ± 0.03a	9.92 ± 0.36ab	183 ± 3	37.7 ± 0.5abcdef	27.5 ± 0.4abcd	34.8 ± 0.7abc
Hydro Average		**1710 ± 148 A**	**12.81 ± 1.07 A**	**9.77 ± 0.76 A**	**185 ± 6 AB**	**35.5 ± 2.3 B**	**27.7 ± 1.3 AB**	**36.9 ± 2.1**
Mix	Mx1	1644 ± 49b	11.77 ± 0.55abcd	8.55 ± 0.38cde	185 ± 5	36.8 ± 1.9abcdefg	28.5 ± 1.5abc	34.7 ± 3.2abc
	Mx2	1619 ± 13bc	11.47 ± 0.25abcd	9.24 ± 0.36bc	179 ± 1	41.3 ± 1.1abcd	24.3 ± 0.8d	34.4 ± 1.2abc
	Mx3	1590 ± 29bcd	10.98 ± 0.27cd	8.51 ± 0.31cde	171 ± 5	41.9 ± 1.2ab	23.4 ± 0.7d	34.7 ± 1.2abc
	Mx4	1550 ± 77bcde	11.34 ± 0.09abcd	8.96 ± 0.18bcd	189 ± 5	36.0 ± 1.5abcdefg	26.4 ± 0.2cd	37.6 ± 1.5abc
	Mx5	1573 ± 74bcd	11.48 ± 0.55abcd	8.23 ± 0.22de	184 ± 8	36.2 ± 1.2abcdefg	27.3 ± 1.0abcd	36.6 ± 0.5ab
	Mx6	1604 ± 53bc	11.39 ± 0.30abcd	8.24 ± 0.38de	188 ± 10	37.6 ± 2.0abcdefg	26.6 ± 0.9bcd	35.8 ± 2.0abc
	Mx7	1587 ± 34bcd	11.68 ± 0.28abcd	8.78 ± 0.48cd	184 ± 6	38.7 ± 0.1bcd	27.3 ± 0.5abcd	34.1 ± 0.6bc
	Mx8	1566 ± 29bcd	11.68 ± 0.55abcd	8.82 ± 0.23cd	190 ± 9	35.5 ± 1.2cdefg	27.5 ± 0.4abcd	37.1 ± 1.4abc
Mix Average		**1592 ± 51 A**	**11.47 ± 0.40 B**	**8.70 ± 0.43 B**	**184 ± 7 B**	**38.0 ± 2.6 A**	**26.4 ± 1.8 B**	**35.6 ± 1.9**
Sign. Samples		*** $p < 0.001$	** $p = 0.004$	*** $p < 0.001$	ns $p = 0.073$	*** $p < 0.001$	*** $p < 0.001$	*** $p < 0.001$
Sign. Groups		*** $p < 0.001$	** $p = 0.008$	*** $p < 0.001$	* $p = 0.032$	* $p = 0.038$	*** $p < 0.001$	ns $p = 0.102$

Data are expressed as average value ± standard deviation (n = 3). Data in bold are the average values of the Groups (Proc/prod, Prof/Pror, Hydro, and Mix, corresponding to OETs procyanidins/prodelphinidins, profisetinidins/prorobinetinidins, hydrolysable, and mix group, respectively). Sign: ns, *, **, *** were used for not significant, $p < 0.05$, $p < 0.01$, $p < 0.001$, respectively, according to ANOVA, Welch's, or Kruskal-Wallis within the data of the same column. Different lowercase letters within the same column refer to the existence of a significant difference between different Samples according to Tukey's test, Games-Howell, or Conover-Holm whereas the presence of differences in uppercase letters within the same column means a significant difference among groups according to Tukey, Games-Howell, or Conover-Holm.

TPI = Total phenolic index, TA = Total anthocyanins; MON %, SPP %, and LPP % = percentage of monomeric forms, percentage of small polymeric pigments, and percentage of long polymeric pigments for Adams-Harbertson method, respectively.

acetaldehyde formation (Vivas & Glories, 1996) that may speed up anthocyanin's polymerization reaction. The control and Prof/pror-added wines had a higher average percentage of short polymeric pigments (SPP %) with respect to Mix formulations ($p < 0.001$). Observing these results, it is difficult to hypothesize a clear effect of a specific class of OETs on colour stabilisation. This seems to be in accordance with Rinaldi & Moio (2018) who underlined that the dosage (10 g/hL and 20 g/hL) was more significant than the type of tannin in the increase of LPP % after 12 months of bottle storage.

3.4. Multivariate analysis of the results from different matrices

Multifactorial Analysis (MFA) was approached to better understand the possible relationship between tannin characteristics and wine features (excluding Gt tannin since it was outlier). MFA explained the 79.75 % of total variance using OETs chemical parameters in model wine solution for polyphenolic and antioxidant characterization, and in-mouth intensity of astringency and bitterness in water (Fig. 3). The first dimension (Dim 1) accounted for the 56.28 % of the total variance explained and it was mainly composed of *PF-tannin* group (polyphenolic variables), followed by astringency and bitterness intensity (Fig. 3**C**). Within *PF-tannin* group, FC and MTC were highly correlated with Dim 1 (+0.934 and +0.857, $p < 0.001$), as well as astringency intensity (+0.857, $p < 0.001$, Fig. 3**B**). Instead, dimension 2 (Dim 2) explained 23.47% of the total variance and it was mainly given by *AC-tannin* group (antioxidant capacity variables). Indeed, Dim 2 was significantly correlated with all the antioxidant capacity parameters: +0.902, +0.840, and +0.817 for DPPH, FRAP, and ABTS, respectively (all $p < 0.001$), and +0.531 for CUPRAC ($p < 0.05$).

To the analysis of individual formulation in model wine solution and water, matrix and tannin family (*Group*) were added as qualitative variables. It is interesting to notice that tannin family was positively correlated with Dim 2 (+0.664, $p < 0.01$, Fig. 3**C**), mainly explained by antioxidant capacity, with Hydro group well distinguished in the positive side of the graph ($p < 0.05$, Fig. 3**A**).

Concerning wine parameters (considered as supplementary variables), firstly, the similarity of model solution and wine AC parameters is evident. Wine FRAP and DPPH were both positively correlated with Dim 2 (+0.763 and +0.750, respectively, both $p < 0.001$) highlighting that the antioxidant capacity of tannin formulations is preserved also in the added wines after one month (Fig. 3**B**). This is in line also with the TPI correlation with Dim 1 for both matrices (+0.746, $p < 0.001$ for model solution and +0.500, $p < 0.05$ for wine). This confirms that the increase of polyphenols given by OETs caused linear increases in antioxidant capacity. Regarding sensory properties (astringency and bitterness intensities), the correlations between matrices were weaker, if not absent; in fact, these two parameters in wine were in the opposite quadrant with respect to water (Fig. 3**B**). Therefore, the final effect of OETs addition is difficult to predict because it is strongly affected by the wine characteristics (Rinaldi, Gambuti, Moine-Ledoux, & Moio 2010) together with tannin concentration. As regards colour properties, TA seemed to be not explained by the investigated variables. More interestingly, long polymeric pigments (LPP %) were negatively correlated with Dim 1 (−0.509, $p < 0.05$), and they were opposite to condensed tannin concentration evaluated with Bate-Smith assay (*BS* variables). As previously mentioned, this may be explained as an increase in polymerization through acetaldehyde formation given by ellagitannins (Vivas & Glories, 1996), in which condensed tannins are not present (Picariello et al., 2018).

To summarize, Fig. 3**C** shows the good correlation between the antioxidant capacity (*AC-tannin*, *AC-wine*) and the tannin type (*Group*), as well as the similar behaviour found in the two matrices evaluated. By contrast, although polyphenolic concentration is closely related to the astringency and bitterness of tannins in water, it is evident that the same parameters are distant in water and wines after one-month storage. This was confirmed by the *RV* coefficient between the two matrices, which increased excluding the sensory properties from 0.558 ($p < 0.01$, with IPT, DPPH, FRAP, Astringency, and Bitterness) to 0.673 ($p < 0.001$, with IPT, DPPH, and FRAP alone). It is possible to hypothesize that wine endogenous tannins are the major contributor to the in-mouth sensations, whereas OETs addition slightly influences the final perception when doses in the oenological range are employed, such as 0.4 g/L in the present study or as previously observed with a dosage of 0.2 g/L (Vazallo-Valleumbrocio et al., 2017).

4. Conclusion

The evaluated OET formulations were very heterogeneous and this was particularly evident in polyphenolic concentration, which strongly influences their chemical and sensory properties. Antioxidant capacity was particularly high for hydrolysable tannins, ellagitannins owing the highest antioxidant potency. Also, ellagitannins were confirmed as the most effective in the formation of long polymeric pigments in a red wine after one-month of storage. Aromatic descriptors were not so highly discriminant in the characterization of OET formulations whereas in-mouth perceptions were more influenced by the total polyphenolic content of the formulates than their origin and chemical characteristics. In fact, very few differences were highlighted in terms of astringency and bitterness when tannins were compared by group, even though quebracho was characterized by the highest intensity in the two in-mouth properties studied. A significant and positive correlation of bitterness with the polyphenolic contents of OET formulates was evidenced, as well as between the perceived astringency in water and the spectrophotometric measurement of A230, which may be helpful as astringency index for OET characterization. Nevertheless, astringency and bitterness perceived in water were not correlated with the intensities found in a one-month storage red wine after OETs addition. This may be due to the high complexity of wine matrix, and the final in-mouth perception may relieve in wine polyphenols and aromatic characteristics, hindering so the effect of tannin addition on sensory parameters. This study highlighted the importance of knowing the characteristics of oenological tannins before the addition of a given formulate and its dosage, taking into account that the OETs polyphenolic content together to antioxidant properties were well correlated with those of the added wines in a short storage period whereas the sensory properties were dependent on wine features and composition. Future studies on different types of wine, and the relationship between matrix and certain OETs will be useful for better understanding their influence on the modification of chemical and sensory parameters.

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CRediT authorship contribution statement

Maria Alessandra Paissoni: Investigation, Formal analysis, Methodology, Data curation, Visualization, Writing – review & editing. **Giovanni Bitelli:** Investigation, Formal analysis, Data curation, Visualization, Writing – original draft. **Mar Vilanova:** Investigation, Formal analysis, Methodology, Data curation, Writing – review & editing. **Carlo Montanini:** Conceptualization, Resources. **Susana Río Segade:** Conceptualization, Methodology, Investigation, Writing – review & editing. **Luca Rolle:** Conceptualization, Supervision, Funding acquisition, Writing – review & editing. **Simone Giacosa:** Conceptualization, Supervision, Investigation, Methodology, Visualization, Writing – review & editing.

Declaration of Competing Interest

The authors declare that they have no known competing financial

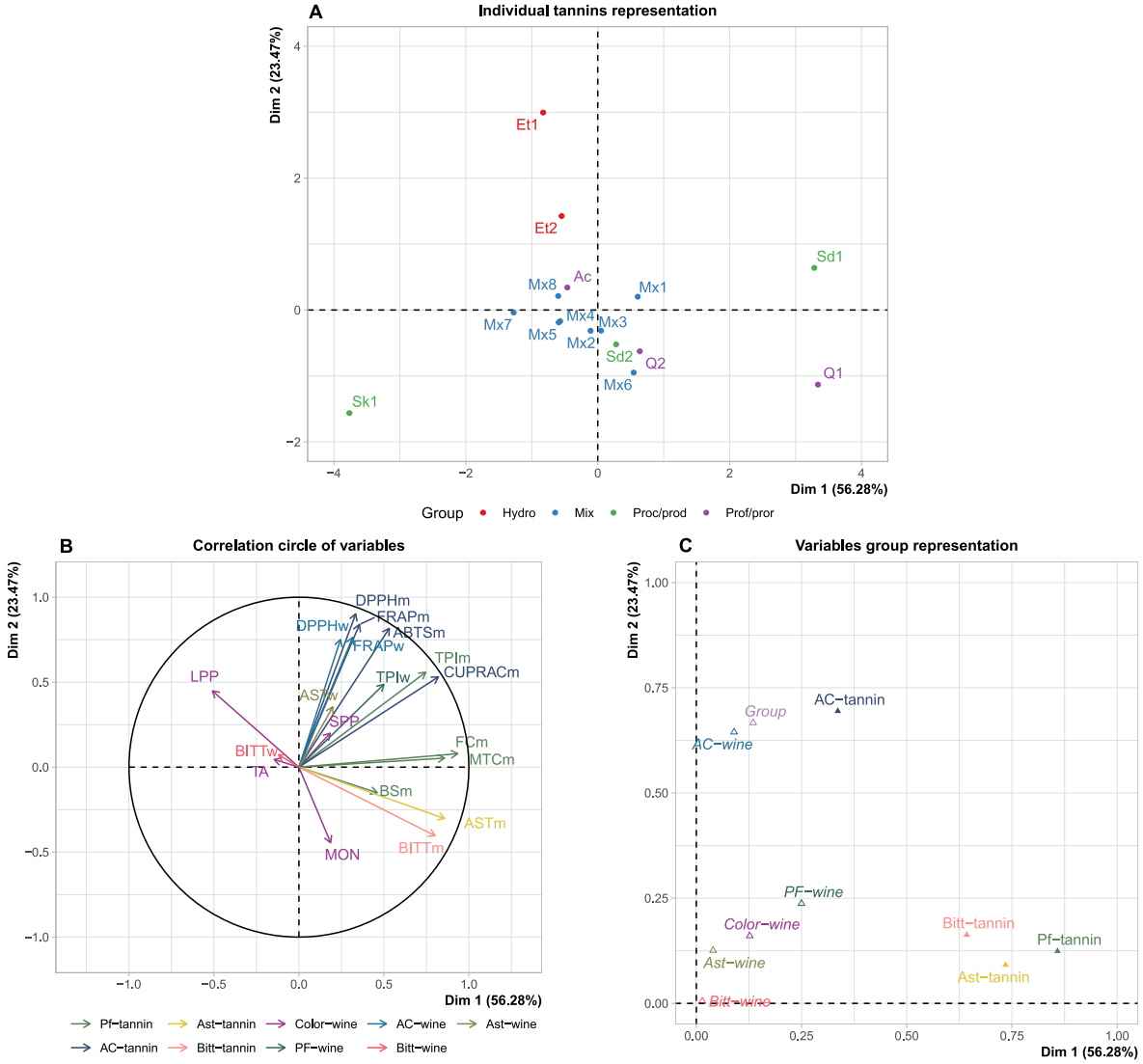

Fig. 3. Multifactorial Analysis of OETs in water and red wine with individual tannins representation (A), correlation circle of active and supplementary variables (B), and arbitrary group representation of variables (C). Individual Group: Hydro = hydrolysable tannins; Mix = mixed formulation; Proc/prod = procyanidins/prodelphinidins; Prof/pror = prorobinetinidins/profisenitinids. Variables Group: AC = antioxidant capacity variables; PF = polyphenolic variables, Ast = sensory astringency; Bitt = sensory bitterness. The suffix *-tannin* refers to model solution analysis (active variables), whereas the suffix *-wine* refers to OETs added wines analysis (supplementary variables). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.foodres.2022.111203.

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