Food Research International 107 (2018) 544–550

Contents lists available at ScienceDirect

Food Research International

journal homepage: www.elsevier.com/locate/foodres

A different approach for the analysis of grapes: Using the skin as sensing element

Raquel Muñoz[a,b], Celia García-Hernández[a], Cristina Medina-Plaza[a], Cristina García-Cabezón[a], J.A. Fernández-Escudero[c], Enrique Barajas[d], Germán Medrano[e], María Luz Rodriguez-Méndez[a,*]

[a] Group of Sensors UVASENS, Universidad de Valladolid, 47011 Valladolid, Spain
[b] Dept. Bioquímica, Biología Molecular y Fisiología, Universidad de Valladolid, 47011 Valladolid, Spain
[c] Estación Enológica de Castilla y León, C/Santísimo Cristo, 26, 47490 Rueda, Valladolid, Spain
[d] ITACYL Avenida de Burgos, KM.118, Finca Zamadueñas, 47071 Valladolid, Spain
[e] R&D Dept. Bodega Cooperativa de Cigales, C/Las Bodegas, s/n, 47270 Cigales, Valladolid, Spain

ARTICLE INFO

Keywords:
Phenolic maturity
Grape
Voltammetric sensor
Skin-sensor
TPI
Multisensor systems

ABSTRACT

In this work, an alternative method to monitor the phenolic maturity of grapes was developed. In this approach, the skins of grapes were used to cover the surface of carbon paste electrodes and the voltammetric signals obtained with the skin-modified sensors were used to obtain information about the phenolic content of the skins. These sensors could easily detect differences in the phenolic composition of different Spanish varieties of grapes (*Mencía*, *Prieto Picudo* and *Juan García*). Moreover, sensors were able to monitor changes in the phenolic content throughout the ripening process from *véraison* until harvest.

Using PLS-1 (Partial Least Squares), correlations were established between the voltammetric signals registered with the skin-modified sensors and the phenolic content measured by classical methods (Glories or Total Polyphenol Index). PLS-1 models provided additional information about Brix degree, density or sugar content, which usually used to establish the harvesting date. The quality of the correlations was influenced by the maturation process and the structural and mechanical skin properties. Thus the skin sensors fabricated with *Juan García* and *Prieto Picudo* grapes (that showed faster polyphenolic maturation and a higher amount of extractable polyphenols than *Mencía*), showed good correlations and therefore could be used to monitor the ripening.

1. Introduction

Harvesting grapes in the optimal point of maturity is a main concern for winemakers (Jackson, 2014). It is therefore important to develop chemical markers to define the optimal ripeness. The physicochemical and biochemical processes occurring during ripening process, produce a continuous rising of the sugars concentration and a decrease in the acid levels. For this reason, the classical parameters measured to monitor grape ripeness and to determine the harvesting date are the berry weight, the must density and the relation between sugars content and total acidity (Robredo, Junquera, González San José, & Barren, 1991) in musts.

To elaborate a high quality red wine, grapes need to reach an appropriate phenolic maturation. To elaborate a high quality red wine, grapes must reach an appropriate phenolic maturation. According to the literature, phenols accumulate in the berries during ripening. Therefore, an increase in the total phenolic content is a good indicator of the grape maturity (González-San José, Barren, & Díez, 1990; González-San José, Barren, Junquera, & Robredo, 1991; Mahmood, Anwar, Abbas, & Saari, 2012).

The phenolic maturity has been measured using different methods and TPI is one possible approach (Adams, 2006; Garrido & Borges, 2013; Kennedy, 2008; Nogales-Bueno et al., 2017).

In spite of the variety of methods mentioned, the assessment of the grape maturity is difficult task because each parameter changes with ripeness in a different manner (since they depend on different biochemical pathways). Moreover, the optimal maturity values at harvest depend on the variety of grape, the type of wine being made, weather or seasonal factors, or viticultural practices among many other factors (Lohitnavy, Bastian, & Collins, 2010). Another difficulty is that the active metabolism of the skin during grape development has an essential effect on the final quality of the grape berries and wine (Negri

Abbreviations: CPE, carbon paste electrode; PCA, Principal Component Analysis; PLS-1, Partial Least Squares; TPI, Total Polyphenol Index; *S*-CPE, Skin-CPE; PC, Principal Component; PLS-DA, Partial Least Squares Discriminant Analysis

* Corresponding author at: Dpt. Inorganic Chemistry, Escuela de Ingenierías Industriales, Paseo del Cauce, 59, 47011 Valladolid, Spain.
E-mail address: mluz@eii.uva.es (M.L. Rodriguez-Méndez).

https://doi.org/10.1016/j.foodres.2018.02.060
Received 18 December 2017; Received in revised form 16 February 2018; Accepted 25 February 2018
Available online 02 March 2018
0963-9969/

Table 1
Results of the chemical analysis carried out by traditional chemical methods.

Sample	Week	Glories pH 3	Glories pH 1	TPI	°Brix	Density (g/mL)	Sugars (g/L)	Degree 16.8	Degree 17.5
Mencía	S1	0.255	0.396	11	18.71	1.0760	177.2	10.53	10.15
	S2	0.340	0.573	11	20.16	1.0822	193.1	11.47	11.05
	S3	0.454	0.579	11	21.65	1.0889	210.3	12.49	12.00
	S4	0.394	0.508	9	20.58	1.0840	197.7	11.75	11.30
	S5	0.244	0.452	17	20.99	1.0857	202.2	12.01	11.55
Prieto Picudo	S1	0.322	0.376	9	19.56	1.0795	186.3	11.07	10.65
	S2	0.474	0.586	9	20.93	1.0857	202.2	12.01	11.55
	S3	0.443	0.824	11	22.65	1.0933	221.7	13.17	12.70
	S4	0.357	0.662	9	21.05	1.0862	203.3	12.08	11.60
	S5	0.341	0.523	19	22.89	1.0943	224.1	13.31	12.80
Juan García	S1	0.361	0.396	14	17.30	1.0699	161.5	9.59	9.25
	S2	0.376	0.480	14	19.80	1.0809	189.7	11.27	10.80
	S3	0.593	0.609	18	21.59	1.0884	209.1	12.42	11.95
	S4	0.458	0.583	17	20.70	1.0849	200.0	11.88	11.45
	S5	0.316	0.418	29	22.18	1.0911	216.0	12.83	12.35

et al., 2008; Rockenbach et al., 2011; Sokolowsky, Rosenberger, & Fischer, 2015). In fact, the majority of the phenolic compounds present in wine are derived from skin which contains anthocyanins, flavonols and proanthocyanidins (tannins) (Jara-Palacios, Hernanz, Escudero-Gilete, & Heredia, 2014; Nogales-Bueno et al., 2017). So, methods are needed that analyze the phenolic composition of skins. Phenolic compounds are redox active compounds that can be analyzed by means of electrochemical methods (Blasco, González-Crevillén, González, & Escarpa, 2007; Hoyos-Arbelaez, Vazquez, & Contreras-Calderon, 2017; Kilmartin, 2013; Kirsanov, Mednova, Vietoris, Kilmartin, & Legin, 2012; Rudnitskaya et al., 2017). For instance, using simple electrodes (graphite, platinum or ITO glass) the antioxidants present in teas (Buratti, Scampicchio, Giovanelli, & Mannino, 2008; Piljac-Žegarac, Valek, Stipčević, & Martínez, 2010), natural juices (Bordonaba & Terry, 2012; Pisoschi, Cheregi, & Danet, 2009), wines (Makhotkina & Kilmartin, 2010), or onions (Zielinska, Wiczkowski, & Piskula, 2008) can be analyzed.

Chemically modified electrodes can also be used for the detection of antioxidants. In such sensors, the electrode surface is covered with a sensing material that facilitates the charge-transfer reactions between the electrode and the solution (Barroso, Santos-Alvarez, Delerue-Matos, & Oliveira, 2011; Durst, Baumner, Murray, Buck, & Andrieux, 1997; Ziyatdinova, Kozlova, & Budnikov, 2016). Many sensing materials have been used as chemical modifiers, for instance, carbon nanotubes, graphene, nanoparticles, porphyrins, phthalocyanines or conducting polymers among others (García-Hernández, García-Cabezón, Martín-Pedrosa, de Saja, & Rodríguez-Méndez, 2016).

Electrochemical sensors can be combined to form Electronic tongues (ET) According to the IUPAC, an electronic tongue is a multisensor system, which consists of a number of low-selective sensors and cross-sensitivity to different species in solution, and an appropriate method of pattern recognition and/or multivariate calibration for data processing (Rodriguez-Méndez, 2016). Such systems provide global information about complex samples. For instance, the electrochemical responses of the multisensor system, contain information not only of the electroactive components of the sample (i.e. phenols) but also, about other components such as protons, ions, etc. The responses of the sensors are analyzed using a pattern recognition software that allows discriminating samples with different characteristics. In addition, after an appropriate training, mathematical models can be built to correlate the measures carried out using the multisensor system and measures carried out by traditional chemical analysis.

Inspired by the concept of ETs, he aim of this work is to develop a new method to monitor the phenolic maturity of grapes based on Carbon Paste Electrodes (CPEs) that were modified with a piece of the skin of the corresponding grape. Three different varieties of *Vitis vinifera* grapes were included in this study (*Mencía*, *Prieto Picudo* and *Juan García*). Grape samples were collected weekly along their ripeness process, starting one week after *véraison* and until complete maturity. The electrochemical response of the skin-sensor (*S*-CPE) was registered in phosphate buffer (pH 7.0) using cyclic voltammetry. The discrimination capability of the electrodes and the ability to monitor the phenolic ripening, were analyzed by means of Principal Component Analysis (PCA) and Partial Least Squares (PLS-1). In addition, the electrochemical response of the electrodes was analyzed and compared with chemical data measured by classical chemical methods.

2. Materials and methods

2.1. Reagents and solutions

All reagents were of high purity and purchased from Sigma–Aldrich. Solutions were prepared using deionized water (Milli-Q, Millipore). Phosphate buffer solution 0.01 M (pH 7.0) was prepared from potassium monobasic and dibasic phosphate salts.

2.2. Grape and must samples: selection and chemical analysis

Samples of three different varieties of red grapes: *Mencía*, *Prieto Picudo* and *Juan García* were harvested in September–October 2013 in the vineyards of the Bodega Cooperativa de Cigales and of the Instituto Tecnológico Agrario de Castilla y León, both located in the Castilla y León region (Spain). 200 berries of each variety were collected on a weekly basis, starting one week after *véraison* until harvest (12, 20 and 26 of September, and 4, 8 of October 2013, samples numbered S1 to S5 respectively).

The polyphenolic content of the grapes was estimated using Total Polyphenol Index (TPI) measured as the Absorbance at 280 nm and the Glories method (Glories & Augustin, 1993). Brix, Density, sugar content, Degree (16.8) and Degree (17.5) were analyzed following the international regulations (OIV, 2013). All chemical parameters are shown in Table 1.

2.3. Skin-carbon paste electrodes (S-CPE)

Carbon paste electrodes were prepared by mixing graphite powder (High purity Ultracarbon®, Ultra F purity. Bay City, MI, USA) with the mineral oil Nujol (Fluka), in 1.5:1 (w/w) ratio. The obtained paste was packed in a 1 mL PVC (polyvinyl chloride) syringe (Yang, Denno, Pyakurel, & Venton, 2015). A copper wire, inserted into the syringe, was used as the electrical contact. The voltammetric behavior of the carbon paste electrodes was analyzed in 0.01 M phosphate buffer