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Evaluation of phenolic extraction during fermentation of red grapes treated by a continuous pulsed electric fields process at pilot-plant scale

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ABSTRACT

The feasibility of processing red grapes by pulsed electric fields (PEF) at pilot-plant scale to improve the extraction of anthocyanins and phenols during the maceration-fermentation step of the winemaking process has been investigated. With this general purpose a colinear continuous treatment chamber was developed. The influence of field strength (2, 5 and 7 kV/cm) and grape variety (Cabernet Sauvignon, Syrah and Merlot) in the extraction kinetics was studied. Extraction curves were described by an exponential equation that permits to estimate the extraction rate (k) and the maximum extraction yield (Y_{max}). An increment of the electric field from 2 to 7 kV/cm increased the extraction rate of anthocyanins and total phenols for the three varieties investigated. The increment of Y_{max} due to the application of PEF was more remarkable in Cabernet Sauvignon than in Merlot and Syrah. The continuous PEF system presented in this work constitutes an important step for the application of PEF technology at commercial scale.

1. Introduction

Food industry requires a continuous adaptation of the production processes to manufacture the best quality products possible at low production costs. In order to achieve these requirements, the potential advantages of the application of innovative technologies in the food industry are being investigated (Knorr et al., 2004; Raso and Barbosa-Cánovas, 2003; San Martín et al., 2002; Wan et al., 2009).

Pulsed electric fields (PEF) is an emerging technology that has gained increasing interest in recent years for liquid food pasteurization and for improving mass transfer operations in the food industry (Fincan et al., 2004; Toepfl et al., 2006; Vorobiev and Lebovka, 2006; Wouters et al., 2001). The process is based on the application of external electric fields that induce the electroporation of both eukaryotic and prokaryotic cell membranes. The pore formation increases the permeability of the membranes causing microbial inactivation or enhancing the diffusion of solutes through cell membranes. The mechanism of action of PEF is based on a non-thermal effect. In contrast to other non-thermal processes, such as high hydrostatic pressure, the PEF technology requires very short processing time and the treatment can be applied in continuous flow.

Solid–liquid extraction is a unit operation extensively used in the food industry. This operation consists in a mass transfer phenomenon in which determined compounds, generally located in the cells, migrate to the liquid phase. Therefore, extraction rate of these compounds notably depends on the permeability of the cell membranes (Sensory and Sastry, 2004). The potential of PEF technology for improving extraction of different compounds by increasing the cell membrane permeability has been investigated to obtain different intracellular compounds such as sugar, coconut milk, paprika, red beetroot pigment or juice from different fruits such as apple or carrot (Ade-Omowaye et al., 2000, 2003; Bazhal et al., 2003; El-belghiti and Vorobiev, 2005; Eshtiaghi and Knorr, 2002; Fincan et al., 2004; López et al., 2009; Tiwari et al., 2009). In these studies, it has been demonstrated that the application of a PEF treatment to the food material before the extraction process increases the extraction rate and the extraction yield. It has been also reported that these treatments allow the application of milder processing conditions during extraction process. For example, decreasing the temperature or the pressure applied during the extraction of red beetroot pigment from 50 to 20 °C or from 14 to 2 kg/cm^2, respectively (López et al., 2009). However, these investigations have been conducted at laboratory scale using batch treatment chambers. Commercial application of PEF technology requires evaluating this technology in continuous treatment chambers at pilot-plant scale.

Mainly three treatment chamber designs have been suggested for PEF applications in continuous treatments: parallel plate, coaxial and colinear configurations (Van den Bosh, 2007). These treatment chamber geometries produce electric fields with different levels of field uniformity. Colinear configuration permits

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working in a wide range of conductivities and its tubular design is very appropriate in terms of processing particulate foods and of integrating the chamber in a pipeline system of industrial size equipment. A lack of uniformity in the electric field may cause an under-processing of a part of the food material (Gerlach et al., 2008). This task is of a great importance in PEF applications for microbial decontamination of foods, but it is not so significant when application of PEF is focused on improving intracellular extraction.

Wine is essentially an alcoholic beverage resulted from the fermentation by yeasts of the sugar presents in the grape juice. During the red winemaking process, phenolic compounds are extracted from the grape pomace along maceration-fermentation step and transferred to the must. Phenolic compounds desired in red wines are mainly found in the grape skin, but some of them are also present in seeds. These compounds play an important role in the colour of red wine and they also are implicated in other sensory attributes of this kind of wines such as bitterness, astringency or mouthfeel (Boulton, 2001; Noble, 1990; Vidal et al., 2004). The antiradical and antioxidant properties of phenolic compounds also determine the potential health-promoting effects associated with consumption of wine (Scalbert et al., 2005; Stoclet et al., 2004). Anthocyanins are one of the most important polyphenols presents in red wine. These compounds, in their monomeric form, are the pigments responsible of the red colour of young red wines, and they contribute to the development of red polymeric pigments during wine aging (Boulton, 2001; Budic-Leto et al., 2006; Monagas et al., 2006).

Recently, it has been demonstrated that the application of a PEF treatment to the red grape pomace before the maceration-fermentation step increases the extraction of phenolic compounds during the vinification of different grape varieties (López et al., 2008a, 2008b). Therefore, PEF technology has the potential to obtain wine with a high content of phenolic compounds or to reduce the maceration time required to obtain a given level of these compounds. Although these studies were very promising, the PEF treatment was applied in batch systems of low capacity. In order to implement this technology to the wineries it is necessary to develop and evaluate PEF systems that allow the treatments in continuous conditions.

The objective of this work was to assess the feasibility of processing grapes by continuous PEF treatments at pilot-plant scale in order to improve the extraction of phenolic compounds during the maceration-fermentation step of the red winemaking process. The study has been conducted with three of the most used grape varieties for elaboration of red wine: Cabernet Sauvignon, Syrah and Merlot. The influence of the electric field strength on the extraction of anthocyanins and polyphenols during the maceration-fermentation process was studied, and the data were mathematically modelled to optimize the process.

2. Materials and methods

2.1. Samples

Grapes of *Vitis vinifera* var. Cabernet Sauvignon, Syrah and Merlot harvested in the 2007 vintage in Aragon (Northeast Spain) were used in this investigation. The three varieties were manually harvested in their optimal ripening stage and in good sanitary conditions. Physical and chemical characteristics of grapes and must of the three varieties investigated are shown in Table 1.

2.2. Pilot-plant PEF equipment

PEF equipment used in this investigation was supplied by ScandiNova (Modulator PG, ScandiNova, Uppsala, Sweden). The apparatus generates square waveform pulses of a width of 3 μs with a frequency up to 300 Hz. The maximum output voltage and current were 30 kV and 200 A, respectively. The equipment consists of a direct current power supply which converts the 3-phase line voltage to a regulated DC voltage. It charges up 6 IGBT switching modules (high-power solid-state switches) to a primary voltage around 1000 V. An external trigger pulse gates all the modules and controls its discharge to a primary pulsed signal of around 1000 V. Finally, a pulse transformer converts this primary 1000 V pulse to a high voltage pulse of desired high voltage.

Table 1
Physico-chemical characteristics of the grapes and musts at the time of harvest. Data expressed as mean values ± standard deviation.

	Cabernet Sauvignon	Syrah	Merlot
Weight of 100 berries (g)	110.2 ± 9.1	152.1 ± 10.2	128.3 ± 9.1
Moisture (g/100 g)	58.4 ± 3.4	44.9 ± 3.1	52.9 ± 4.3
Total solids (g/100 g)	41.6 ± 2.4	55.1 ± 3.9	47.1 ± 3.8
Medium berry diameter (mm)	11.3 ± 1.7	13.2 ± 1.2	10.2 ± 1.6
Sugar content (Brix)	23.5 ± 0.4	21.6 ± 0.2	23.1 ± 0.3
Potential alcohol value (% v/v)	13.8 ± 0.2	12.6 ± 0.1	13.6 ± 0.1
Density (g/L)	1099 ± 12	1092 ± 8	1098 ± 10
pH	3.3 ± 0.2	3.3 ± 0.1	3.2 ± 0.1
Titrable acidity (g/L)	5.8 ± 0.3	7.3 ± 0.2	7.1 ± 0.3
Total phenols (GAE mg/L)	492 ± 56	551 ± 35	561 ± 13
Anthocyanins (mg/L)	31.5 ± 3.4	50.3 ± 2.4	20.8 ± 1.1

The colinear treatment chamber used in this investigation (Fig. 1A) is based on a previous design of Toepfl et al. (2007). The treatment chamber consists of three cylindrical electrodes made of stainless steel, separated by two insulators of methacrylate. Whereas the central electrode is connected to the high voltage, the electrodes of the both extremes are grounded. This colinear design defines two treatment zones of 2 cm between the electrodes with an inner diameter of 2 cm. A feature of colinear treatment chambers is that the distribution of the electric field strength applied is not uniform. Therefore the peak of the field strength takes different values depending on the position inside the treatment zone. In order to know this distribution, the electric field strength in the treatment zone of the chamber was numerically simulated by the finite elements method, using the Comsol Multiphysics software (Comsol Inc., Stockholm, Sweden) (Fig. 1B). Depending on the position in the treatment zone, the electric field strength reached a determined value. In order to standardize and compare the results, the electric field strength used to characterize the PEF treatments applied correspond to the electric field strength in the center point of the treatment zone (Toepfl et al., 2007).

The actual voltage and the current intensity applied were measured with a high voltage probe (Tektronix, P6015A, Wilsonville, Oregon, USA) and a current probe respectively (Stangenes Industries Inc., Palo Alto, California, USA) connected to an oscilloscope (Tektronix, TDS 220, Wilsonville, Oregon, USA).

A progressive cavity pump (Rotor-MT, Bominox, Gerona, Spain) was used to pump the grape mass to the colinear treatment chamber. The mass flow rate was 118 kg/h. This flow corresponds with a medium residence time in the treatment zone of 0.41 s.

2.3. PEF treatments

After crushing and de-stemming grapes were PEF treated. The PEF treatments consisted in of fifty pulses at a frequency of 122 Hz and at electric field strengths of 2, 5 and 7 kV/cm. Previous experiments showed that longer treatments did not increase the extraction of phenolic compounds (López et al., 2008a). The total specific energy applied at 7 kV/cm was 6.76 kJ/kg, at 5 kV/cm was 3.67 kJ/kg and at 2 kV/cm was 0.56 kJ/kg.