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Impact of ultrasounds on the extraction of polyphenols during winemaking of red grapes cultivars from southern Italy

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ABSTRACT

Effectiveness of ultrasounds (US) on polyphenols extraction during winemaking of three red grapes cultivars grown in southern Italy (Primitivo, Nero di Troia and Aglianico) was investigated. Innovative stainless steel horizontal rotary wine fermenters were developed for the study. The US protocol was tested on the first harvested cultivar (Primitivo), and efficacy was ascertained (+ 15% of flavonoids, + 10% of total polyphenols and + 100% of proanthocyanidins), even though the delivery system reached only 30% of maximum power. It was due to the "screen" effect caused by skins around the US transducer. In order to overcome this problem, the other two cultivars were processed after diluting the grapes with previously extracted juice (1:1 w/v). This modification allowed power to increase to 60%. Extraction of polyphenols was improved in Aglianico (+ 20% of flavonoids, + 15% of total polyphenols, + 12% antioxidant activity, + 20% colour intensity), whereas little effect was observed for Nero di Troia. It was concluded that impact of ultrasounds varies, depending on the cultivar. Under our conditions, the treatment could be useful in winemaking of Primitivo and Aglianico.

Industrial relevance: The use of ultrasounds in winemaking could increase the extraction of polyphenols from grapes, improving the sensory characteristics and health benefits of wine due to higher content of nutraceuticals. The ultrasound generator and transducer can easily be included in the traditional winemaking process as an "add on" technology, without distorting the processing lines. This innovation could reduce the environmental impact of winemaking, involving lower energy consumption and reduced processing times.

1. Introduction

Grapes are a source of phenolic compounds, which play an important role both in plant physiology and for human health. Phenols work in various reactions to protect cells against abiotic stresses like UV-light, or against biotic stresses such as attacks by predators and pathogens (Weisshaar & Jenkins, 1998; Winkel-Shirley, 2002). Moreover, many phenolic compounds, such as resveratrol, quercetin and rutin, have been reported as having biological activities, including cardio-protective, anti-inflammatory, anti-carcinogenic, antiviral and antibacterial properties, attributed mainly to their antioxidant and antiradical activity (Frankel, German, Kinsella, Parks, & Kanner, 1993; King, Bomser, & Min, 2006; Santos-Buelga & Scalbert, 2000; Teissedre, Frankel, Waterhouse, Peleg, & German, 1996).

In oenology, phenols contribute to the wine's sensory properties, such as colour, flavour, astringency and bitterness. Anthocyanins and flavan-3-ols are flavonoids and are very important for the quality of red wine. Anthocyanins are responsible for colour, and flavan-3-ols, the so-called condensed tannins or proanthocyanidins, are responsible for astringency and bitterness (Gawel, 1998; Peleg, Gacon, Schlich, & Noble, 1999), and for long-term colour stability (Somers, 1971; Vivar-Quintana, Santos-Buelga, & Rivas-Gonzalo, 2002). It is well known that anthocyanins are located in grape skins, whereas flavans-3-ols are located in skins and in seeds. They are extracted from grapes at the maceration/fermentation stage of winemaking. The phenolic composition of red wine is affected by different factors, such as the grapevine genome, winemaking technology and aging conditions (Baiano, Terracone, Gambacorta, & La Notte, 2009; Gambuti, Rinaldi, Ugliano, & Moio, 2013; Pérez-Lamela, García-Falcón, Simal-Gándara, & Orriols-Fernández, 2007). As far as winemaking technologies are concerned, in recent years ultrasound-assisted extraction has been tested to enhance the content and composition of the phenolic compounds in red wines. In particular, ultrasound treatments are used to increase the extraction of polyphenols and volatiles from grape skins

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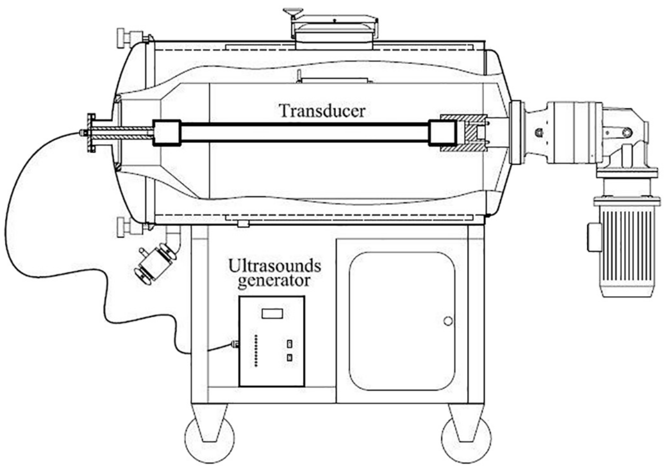

Fig. 1. Schematic drawing of the fermenter equipped with the ultrasonic delivery system.

during maceration, and to accelerate and enhance aging. Ultrasound efficacy is linked to the formation of small bubbles that then collapse, generating kinetic energy that destroys the cell walls of vegetable tissues in aqueous systems. This process is known as cavitation, and its effects are mainly mechanical at frequencies of up to 20 kHz, and chemical at higher frequencies (Mason, Paniwnyk, & Lorimer, 1996). Several studies have recently explained the effects on colour and flavour of the use of ultrasound at different stages of the winemaking process (Bates & Patist, 2010; El Darra, Grimi, Maroun, Louka, & Vorobiev, 2013; Ferraretto & Celotti, 2016). For Negroamaro an increase in anthocyanins and proanthocyanidins (Coletta et al., 2013) and for Tempranillo an increase in anthocyanins, total polyphenols and tannins (Carrera, Ruiz-Rodríguez, Palma, & Barroso, 2012) as results of US application, gave wines with better aging potential. One of the most important changes during aging is a progressive increase and stabilization of the colour due to copigment anthocyanin complexes and the formation of both tannin–tannin and anthocyanin–tannin complexes (Boulton, 2001). Masuzawa, Ohdaira, and Ide (2000), found that the polymerization of polyphenolic compounds in red wine was promoted by ultrasound at low sound pressure levels.

At our knowledge there is poor information about application of US to Italian red wines, and no investigation has been carried out on some important cultivars of Southern Italy such as Aglianico, Nero di Troia and Primitivo. The aim of the present study was to evaluate the effectiveness of ultrasound treatment on these three wine grape cultivars, which have different ripening times.

2. Materials and methods

2.1. Grape sampling

The research was conducted in September–October 2014 on Primitivo (early ripening), Nero di Troia (medium-late ripening) and Aglianico (late ripening) grape cultivars from three different vineyards in southern Italy. Primitivo from the Gioia del Colle area (Puglia Region) was harvested on 19th September, Nero di Troia from the Corato area (Puglia Region) was harvested on 1st October, and Aglianico from the Avellino area (Campania Region) was harvested on 4th November. Approximately 1000 kg of grapes were hand-picked for each cultivar, packed in 20 kg perforated plastic boxes and transferred to Agricole Pietraventosa winery at Gioia del Colle for the winemaking trials.

2.2. Winemaking

The grapes were processed using four “Gioiello” pilot plants, consisting of 200-L stainless steel horizontal rotary wine fermenters with a submerged cap (Industrie Fracchiolla, Adelfia, Italy). Two of the fermenters, were equipped when necessary with an ultrasonic delivery system consisting of a Sonic Digital LC 1500 SD 25-P ultrasonic generator (25 kHz frequency, 1500 W power output) and Sonopush HD Double Twin 1500 titanium transducer (WEAL, Milan, Italy). The schematic drawing of the fermenter fitted with the ultrasonic delivery system is shown in Fig. 1. The power indicated on the display of the ultrasound generator was 100% when the fermenter was loaded with water.

Primitivo was the first cultivar to be vinified, and was also used for setting up the ultrasound parameters and screening effectiveness of this technique in comparison with traditional winemaking. Grapes were destemmed and divided into three aliquots (approximately 250 kg each), then vinified by traditional maceration (P-C, control), pre-fermentative ultrasound maceration (P-UBF) and post-fermentative ultrasound maceration (P-UAF), as described in Table 1. For the winemaking trials with Nero di Troia and Aglianico cultivars, the experimental protocol was modified as follows: i) US was applied only in pre-fermentation; ii) grapes were diluted with juice of the same cultivar in the ratio of 1:1 (w:v) in order to increase the power of US treatment. Specifically, approximately 85 kg of de-stemmed grapes were diluted with 85 L of juice (obtained with a manual wine-press), and vinified by traditional (NT-C, Nero di Troia control; A-C, Aglianico control) and pre-fermentation ultrasound maceration (NT-U, Nero di Troia ultrasound; A-U, Aglianico ultrasound), as described in Table 1. The trials were done in duplicate using the four pilot plants described above (2 controls + 2 ultrasounds for each cultivar). At the end of maceration (7 days), free-run wine was

Table 1
Winemaking technologies tested using the four pilot plants.

Winemaking technology	Action
P-C	Seven days maceration at 25 °C. Addition of potassium metabisulphite (20 g/100 kg); yeast (*Saccharomyces cerevisiae* var. *Bayanus*, Mycoferm CRU 05, 20 g/100 kg, Everintec, Pramaggiore, Italy); yeast activator (preparation based on ammonium sulphate, diammonium phosphate, chemically inert filter and as dispersing agent, Vitamin B1, Enovit, 20 g/100 kg, AEB); O_2, 10 mg/L/day after 2 days from the beginning of fermentation; yeast activator, 20 g/100 kg after 3 days from the beginning of fermentation; without any further oenological treatment.
P-UBF	As P-C, but with 2 h of ultrasound treatment before the start of fermentation as the drum rotated.
P-UAF	As P-C, but with 2 h of ultrasound treatment at the end of fermentation as the drum rotated.
NT-DC[a]	As P-C, but using about 85 kg of de-stemmed grapes + 85 L of juice.
NT-DU[a]	As NT-DC, but with 2 h of ultrasound treatment before the start of fermentation as the drum rotated.
A-DC[a]	As NT-DC.
A-DU[a]	As A-DC, but with 2 h of ultrasound treatment before the start of fermentation as the drum rotated.

P-C, Primitivo control; P-UBF, Primitivo ultrasound before fermentation; P-UAF, Primitivo ultrasound after fermentation; NT-DC, Nero di Troia diluted control; NT-DU, Nero di Troia diluted ultrasound; A-DC, Aglianico diluted control; A-DU, Aglianico diluted ultrasound.
[a] Two winemaking replicates.