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“Gentle” sustainable extraction from whole berry by using resonance waves and slight over CO_2 overpressure

Fabio Mencarelli[1], Giuseppe Floridia[2], Stefano Pettinelli[1] and Andrea Uliva[2]

[1] Wine research consultant, Via Poggio di Montemagno 19, Calci (PI), Italy
[2] Parsec srl, Sesto Fiorentino (Florence), Italy

Abstract. The traditional methods of grape extraction of enochemical compounds use very often mechanical energy by pistons such as the pigeage or mechanical energy produced by must (delestage, pumping over). Recent trend by winemakeris trying to introduce in the fermentation tank, whole berry grape to avoid even minimal oxidation. Unfortunately, the use of the traditional mechanical techniques aforementioned, very often does not guarantee the optimal extraction of polyphenols and above all does not assure an uniform temperature in the vessel. Use of resonance waves (AM =AIRMIXING MI ™) or a slight overpressure by CO_2 (ADCF™) permits to work on whole berry guaranteeing the perfect extraction and homogenize the temperature and the oxygen in all the mass. In AM, the great goal to have an uniform temperature in all the tank, permits the winemaker to decide which temperature using to ferment and to extract depending on the wine style she/he wants to reach. This presentation we will summarize data on experimental works done with these two techniques and we show, beyond the theoretical explanation, the kinetic of extraction of different phenol classes and ofvolatile organic compounds even in the production of wine without sulfite addition.

1. 1 Introduction

Polyphenols characterize body, color, and some of the main organoleptic attributes in red wines. Wine polyphenols can be extracted from grapes and wood, or they can be metabolized by yeasts [1]. Red grape juice fermentation is a critical stage in wine production because is coupled to maceration. Transfer of polyphenols from berry skin to liquid during maceration/fermentation depends on various factors,chemical and physical ones [2]. The phenomena are complex and are not limited to a regular increase in extracted substances. During a traditional fermentation onthe grape skins, solubility and instability as for anthocyanins and tannins are the limiting factors[3]. The early peak and subsequent decline in anthocyanins during fermentation is inconsistent with solubility being the limiting factor for these compounds and instead reflects their instability [2]. Other components such asproanthocyanins or tannins relate to the mouthfeel attributes, bitterness, and astringency, and they influence mouthfeel perception [4,5]. In the first step of cell degradation after crushing operation, cell wall enzymatic activity plays an important role [6] provoking the deconstruction of cell wall polysaccharide networks, permitting the other factors to proceed with the extraction during maceration and fermentation [7], but this activityis strongly dependent on the internal temperature of the tank. This represents a critical point especially in bigtanks (high one) where a stratification of temperature is easy to have and also a very thick floating cap, supported by the carbon dioxide released during fermentation, is-formed conditioning an efficient yeast fermentation and skin maceration. Therefore, this cap should be brokendown to submerge berry skins into the fermenting juice typically a few times a day. Various alternative systems and techniques, either gentle (use of gas movement) or hard (mechanical movement), have been developed inorder to reduce the time and labor cost, providing better phenolic extraction [8]. In this paper we summarize the results of the use of two innovative techniques (gentle) to manage the fermentation: AIRMIXING M.I.® (AM) and ADCF™. The first one is an air-modulated injection using the resonance wave physical law to prevent the cap formation, while the second one keeps a slight overpressure of CO_2 released by alcoholic fermentation through an accurate pressure sensor and employs this overpressure together with a sudden pressure dropping bya valve opening to disrupt the cap

2. Materials and methods

First experiment was carried out in the Famiglia Cotarella Winery, Montecchio (TR), Italy, in 2020 and 2021 on Cabernet Sauvignon (9). Here we compare the two techniques AM and ADCF with a traditional "hard" technique. The three tanks (Control, AM (AIRMIXING MI), and ADCF) used in the experimentation have the same characteristics: a cylinder with height of 5.3 m and a diameter of about 2 m, for a total volume of 208 hL. All the tanks were equipped with a cooling jacket for temperature control, a hydraulic system consisting of a pump and pipes to automate the practice of must movement (pump-over), a rotary extraction blade placed at the base of the tank, an automatic macrooxygenation system, and the computerized system SAEn5000 [Parsec srl, Sesto Fiorentino (FI), Italy)]. The control vinification followed involved the use of daily pump-over, and "delestage". Sampling was done during fermentation and at the end of fermentation. Chemical analyses were performed with a WineScan^{TM} and for specific anthocyanins through HPLC (9).

The second experiment to produce wine without sulfite addition using AIRMIX was performed on Sangiovese grape upon the Cantina Tuscania (Bargino, Florence, Italy). The destemmed grape mass (7 hl each tank) was pumped in 6 stainless steel tanks, 10 hL each, (Parsec srl, Sesto Fiorentino, Florence, Italy), equipped with cooling jacket, three used for the Control and three with the AM system (10). In the Control wine process, sulfites were added (8 g/hL in total) while in AM process no sulfites were added. Chemical analyses were performed with WineScan^{TM}, HPLC and GC-MS (10).

3. Results and discussion

3.1. Theoretical principle

AM technique is based on resonance wave physical principal. Resonance frequency consists in a natural vibration frequency (f) of an object and is indicated with frequency = propagation speed/wavelength ($f = v/\lambda$). Resonance wave occurs when an external force amplifies the motion of the system to the point of maximizing the height of the wave at a given frequency. The technique developed by Parsec srl based on this principle, hypothesized that, by modulating the gas supply to a liquid/solid mass such as pressed grapes, contained in a tank, by using a sequential modulated injection with a fixed pressure, it is possible to avoid the cap formation, uniform the temperature, and managing the fermentation rate and the extraction (11). ADCF technique is based on the principle of the Henry law that states that the volume of dissolved gas in a liquid is directly proportional to its partial pressure above the liquid and its solubility. Thus increasing slightly the pressure of the CO_2 produced during fermentation, it will allow for the increase of CO_2, but also all the VOCs (volatile organic compounds) produced with fermentation, in the liquid mass increasing its pressure. When this mass under over pressure will be subjected to a sudden pressure drop, the liquid will expand and the solid particle, expanding, will releaseinner components in the liquid, with a gentle transferfrom solid to liquid.

In both techniques, AM and ADCF, the basic principle is the pressure change of the mass, in one case (AM) supplying gas and in the other (ADCF) blocking the gas from fermentation to escape thus increasing the pressure, suddenly reducing it.

Beyond the aspect of physical effect of solid particles, wemust take into account the partition coefficient of VOCs . All of these pressure changes will affect the partition coefficient of VOCs . Indeed, the concentrations of volatile molecules, as well as the overall composition of the fermenting must, are continuously changing during alcoholic fermentations, and depending on their solubility and their partial pressure, they can leave or solubilize. Another way of VOCs loss is due t*o* the production and release of CO_2, bubbles of which increase the transfer from the liquid to the gas, by stripping. Analyses ofconstant rate fermentations demonstrated that thepartitioning was not influenced by the CO_2 productionrate and was a function of only the must composition andthe temperature.

3.2. AM and ADCF and phenol extraction

As you can see in Table 1, the two "gentle" techniques in two years of experimental work on the same grape vineyard, allowed for a reduction of volatile acidity and an increase of YAN. This higher content of volatile acidity was due to the cap presence, in the Control, on thesurface of the must/wine in fermentation, which stands between one pump-over and the other. As regards YAN, the significant increase is probably due to greaterextraction with the innovative technique as confirmed by the slight increase of polyphenols and anthocyanins in ADCF system.

Table 1. Percentage changes (as a mean of two years) of ADCF and AM wine characteristics over the Control technique used on Cabernet Sauvignon. The data represents the percentage changein two tanks each test, sampling done after wine mixing in the tank. The reported values are the most significant in the two years 2020 and 2021.

	Control	ADCF	AM
Alcohol (%v/v)	14.77-14.75	0	0
pH	3.84-3.55	0	0
Titratable acidity (g/L tartaric acid)	5.72-7.02	-9	-9
Volatile acidity (g/l acetic acid)	0.40	-40	-25
YAN (mg/L)	63-73	+26	+22
Total anthocyanins (mg/L)	475-851	+7	0
Total polyphenols (mg/L)	2388-2977	+13	0

If we observe the kinetics of polyphenol extraction, we can note how fast is the extraction of polyphenols with the two "gentle" techniques, thus it means that the cap immersion in ADCF and the no cap formation in AM favor the extraction (Figure 1). ADCF is always the fastest in the extraction and also the greatest extraction at the end while AM , after a rapid extraction the content of polyphenols remain still. The reason of this effect is because in AM the winemaker needs to have in mind that temperature is uniform in all the tank. Thus the winemaker must be decide the temperature to work. While with the traditional technique the cap temperatureis higher than in the liquid, in AM the temperatures are the same. In the experiment which we show data here of, the temperature in AM was managed as in traditional technique; this is the reason not to have rising of extraction with the time, after the initial strong extraction.The berry peel at the end of AM process is completely light pink and in, almost, perfect shape, showing the gentle system adopted for the extraction. Fine lees due to the pulp degradation can be in higher content than in the Control, because the gas homogenization of the mass, the modulated oxygen, and the uniform temperature favor the activity of cell wall enzymes. Thus it is important to rack soon the final wine after the end of fermentation.

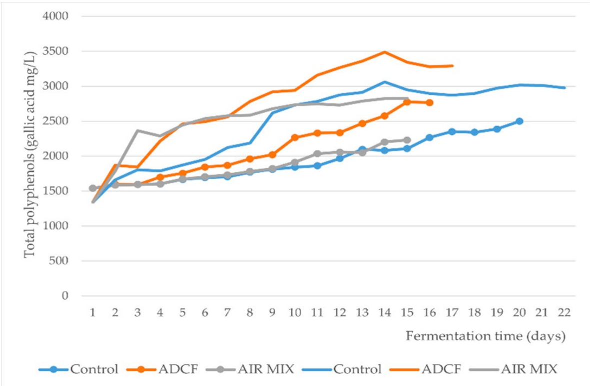

Figure 1. Kinetics of total polyphenols extraction in 208 hLtank, 2020 and 2021 years (from 9)

The reason of the end of polyphenol lines on day 15-17, 4-5 days earlier than the control is due to the more rapid fermentation due to the oxygen management and uniform temperature maintained in the tank especially with the AM technique.

3.3. AM and no added sulfites

The other experimental work was performed to see how AM could facilitate the production of red wine without sulfite addition. There is a rising interest on the reduction of sulfite concentration by the wine consumer. In this case we concentrated especially on VOCs (volatile organic compounds) and specific polyphenols ofproduced wines. Fermentation ended 2-3 days earlier thanthe Control in the AM tank. Chemical analyses revealed significant differences for volatile acidity, higher in the Control, in titratable acidity higher in AM as well as total anthocyanins while total polyphenols was higher in the Control (10).

Table 2. Compounds with significant differences. The values ofAM are in percentage of increase or decrease over the Control value reported in mg/L.

	AM (%)	Control
Quercetin-3-O-glucoside	-35	2.61
Quercetin	+50	1.60
Trans-resveratrol	+55	4.36
Hydroxycinnamic acids	-36	3.81

Interesting the behaviour of quercetin and its glucoside:in AM without sulfites was higher as quercetin while in the Control as glucoside. Probably the way of

Table 3. Compounds with significant differences. The values ofAM are in percentage of increase or decrease over the Control value reported in ug/L.

	AM (%)	Control (ug/L)
Decanoic, dodecanoic, hexanoic acids	30	2700
Isoamylic and phenethyl alcohol	50	71000
Isoamyl acetate	400	221
Ethyl octanoate	150	142
γ-Butyrolactone	26	3080

The higher content of VOCs in AM wine could be due as to extraction but also to amore regular fermentation rate due to oxygen management. But if we look the compounds reported in Table 3 they are fatty acids,esters of branched alcohols, and one lactone. This means that the maintenance of gentle maceration with oxygen management permits an anabolism and catabolism of yeasts more accentuated (this explains the rapid end of fermentation) but it is also due to the partition coefficient of these compounds. The slight, transient, overpressure due to the oxygen injection in AM favors the partition of volatile compounds in the liquid, avoiding the loss thatwe have when we use more energic maceration technique such as pigegage, delestage or pumping over.

4. Conclusions

Gentle maceration techniques such as the use of resonance waves (AIRMIXING MI^{TM}) or a slight overpressure by CO_2 (ADCF^{TM}) have solid scientific basis and they do not “play” only on mechanical force of pressure on grape berries for the disruption. Resonance waves avoiding cap formation and assuring an uniform temperature in all the tank, increase the extraction of phenolics and VOCs as well as the use of a slight overpressure with cap immersion does. These techniques required knowledge by the winemaker on the characteristics of specific compounds in order to modulate their extraction providing different wines using the same technique.

5. References

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