

# UHPH processing of grape must to improve wine quality

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**Abstract.** Ultra High Pressure Homogenization (UHPH) consists of continuous pumping of must at pressures above 200 MPa, usually 300 MPa, and its subsequent instantaneous depressurization to atmospheric pressure after passing through a special valve. In the valve, the intense impact forces and shear stresses, together with the temperature, lead to the death of microbial cells and also to the inactivation of oxidative enzymes. Intense mechanical stresses also result in nanofragmentation of colloidal particles increasing the release of nutritional factors such as YAN and others, thus improving colloidal stability. Molecules with sensory impact such as terpenes are not affected by the UHPH process, thus preserving the varietal character, nor can thermal markers such as furfural be detected. UHPH-processed musts show and maintain higher antioxidant activity than control musts and show less browning effects during processing and even later throughout and after fermentation. This technique also affects the extraction and stability of anthocyanins and other phenolic compounds by increasing their release from cell structures and protecting them from oxidation. The antimicrobial effect and the inactivation of oxidative enzymes allow the production of wines without or with a very low level of sulfur dioxide. The ability to inactivate enzymes by affecting their tridimensional structure may also have some effect on colloidal proteins by preventing protein haze or facilitating the use of protease enzymes.

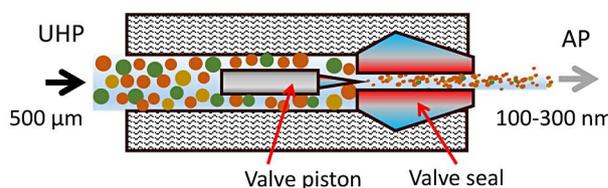
## 1 Introduction

Ultrahigh pressure homogenization (UHPH) is an ultrahigh pressure pumping (>200 MPa, usually 300 MPa) of a fluid followed by a sequential and ultrafast depressurization at atmospheric pressure or slightly higher if desired. Depressurization is done across a special ultra-resistant valve, frequently performed in tungsten carbide covered by artificial diamond or extremely hard ceramic materials. At such pressure in the capillary conduction the speed is really high reaching values around Mach 3 (3x sound speed) what make UHPH an ultrashort treatment with a high-pressure time lower than 0.2 seconds [1]. The ultra-rapid treatment produces a gentle effect on delicate liquid foods with low effects in the stability of sensitive constituents as: vitamins, pigments, aromas.

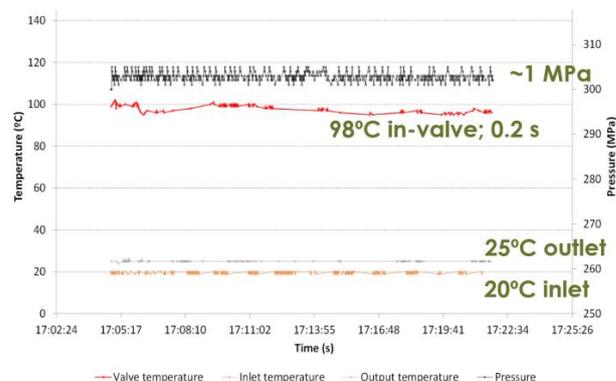
In the valve fluids suffer intense impact and extreme shear efforts, additional effects as cavitation and others can also happen. At molecular effect an intense nano-fragmentation is produced in biopolymers and cell structures. Colloidal particles in fluids must be lower than 0.5 mm to pass by the capillary pipeline and after the valve fragmentation have an average size of 400-500 nm or lower (Fig. 1). The more effective UHPH technology is currently commercialized under international Patent EP2409583 by YPSICON TECHNOLOGIES (<https://www.ypsicon.com/>).

The in-valve pass usually increase the temperature being usual to raise from 20-25 to 80-100°C easily [2]. After the valve the immediate depressurization reduce again the temperature close to initial values (Fig. 2). Additionally, it can be observed the high precision and

stability of the new ultra-high pressure pumps that are able to pump at 300 MPa with a slight variation of just 1 MPa.



**Figure 1.** UHPH valve and nanofragmentation effect, extracted from [1].



**Figure 2.** Pressures (MPa) and temperatures (°C) in a typical UHPH process [2].

The high temperature and pressure are enough to fully eliminate grape microorganisms even in juices with high microbial load. We have eliminated by UHPH

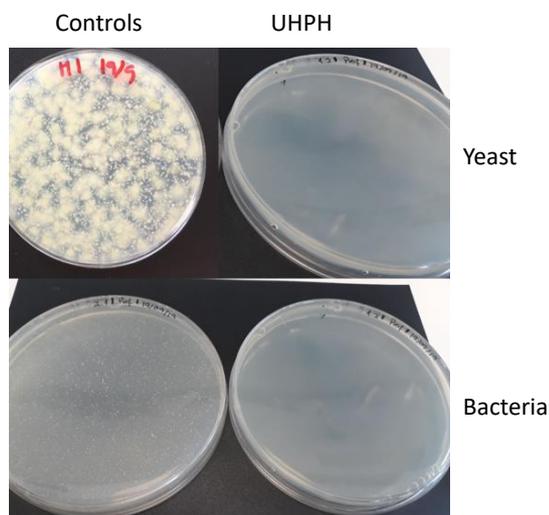
populations of 6-log CFU/mL of yeast and 4-log of bacteria [2, 3] in white wines (Fig. 3). If the in-valve temperature is higher enough even sporulate microorganisms can be eliminated. The effect is a must sterilization but with a really gentle management of the compounds with sensory impact as volatiles and pigments. UHPH musts kept in closed vials remain unfermented during several months [3]. When red grape juices are processed in similar conditions also a full elimination of grape microorganisms is observed [4].

This technique has been recently approved by OIV RESOLUTION OIV-OENO 594B-2020 (<https://www.oiv.int/public/medias/7587/oiv-oeno-594b-2020-en.pdf>).

## 2 Effect in juice quality

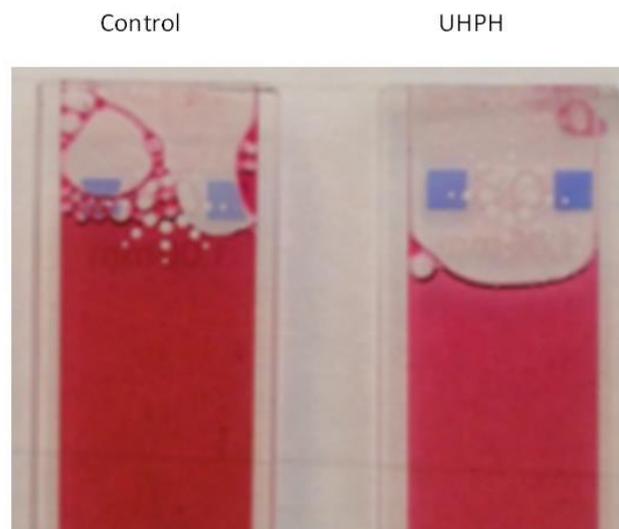
The intense nanofragmentation effect produced by UHPH on the colloidal particles of the grape juice can release nutrients from them. If the settling of the initial must is not too intense the UHPH processing can promote the release of Yeast Assimilable Nitrogen (YAN) compounds from the plant cells what can increase the nutritional properties of the must. It has been observed that the higher release of YAN promotes the formation of higher levels of fruity esters during fermentation [2].

Additionally, delicate aroma compounds such as terpenes are protected by UHPH. Non-significant differences (p set at 5%) were found in the contents of linalool, terpinen-4-ol, epoxy linalool,  $\beta$ -citronellol, geraniol and  $\alpha$ -terpineol among UHPH musts and controls when this technique has been used in muscat grape juice [3].



**Figure 3.** Microbial counts (yeasts and bacteria) by plating before and after an UHPH processing of grape juice.

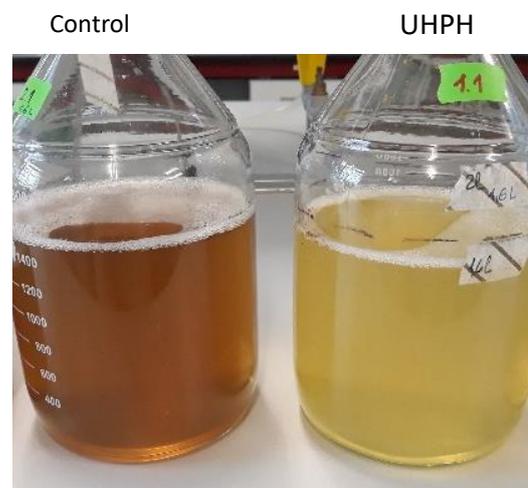
A protective effect can be observed also in anthocyanins with similar contents of 3-O-glucosides and what is more important higher contents of acylated anthocyanins [4] with better repercussion in red bluish color because of their higher wavelength absorptions (Fig. 4).



**Figure 4.** Browning effect in control (left) and protection of paleness in UHPH must (right) by the inactivation of PPOs.

## 3 Inactivation of PPOs and antioxidant activity

Other interesting effect of UHPH is the inactivation of oxidative enzymes by the intense shear efforts and the high impact that is produced when the fluid pass through the valve. It has been observed an intense inactivation of Polyphenol oxidase (PPOs) enzymes that can be higher than 90% [2]. PPOs produce the oxidation of orthodiphenols transforming them in browned compounds. The effect in absence of SO<sub>2</sub> and in with air content is very intense and easily degrade the color quality of white wines, also affecting red wines. In UHPH musts the intense PPO inactivation allow the preservation of pale colors during several days in absence of SO<sub>2</sub> (Fig. 5) [3]. The high impact of UHPH in the inactivation of microorganisms but also in the control of oxidation makes this technique a good alternative to the use of SO<sub>2</sub> [1, 3].



**Figure 5.** Browning effect in control (left) and protection of paleness in UHPH must (right) by the inactivation of PPOs.

The gentle effect on phenolic compounds and the inactivation of PPOs also helps to keep a high antioxidant activity in juices. We have measured a 156% higher antioxidant activity in a muscat must processed by UHPH compared with the controls in absence of SO<sub>2</sub> [3]. In red wines by the protective effect of proanthocyanidins the antioxidant value is less important but also clearly observed with a value of 106% [4].

#### 4 UHPH and new biotechnologies

The elimination of wild microorganisms from grape juice by UHPH in absence of SO<sub>2</sub> let us to apply emerging biotechnologies such as the use of non-*Saccharomyces* yeast. These yeasts are extensively used now to improve wine quality and to increase wine differentiation [5], however, they have a difficult application at industrial scale because of their low fermentative power and competitiveness compared with *S. cerevisiae*. The inoculation in musts without wild grape yeasts increase a lot the implantation improving the expression of non-*Saccharomyces* metabolome and enzymatic activities. Additionally, most of the non-*Saccharomyces* species are more sensitive to SO<sub>2</sub> than *S. cerevisiae*, so the reduction or elimination of this additive in the fermentation increases their viability and implantation.

Yeast-bacteria coinoculations are also used now extensively to reduce fermentation time and to improve winemaking process. The application of this biotechnology can be altered by undesired spoilage microorganisms that can affect the fermentation producing higher volatile acidity or other off-flavours. The elimination of these microorganisms by UHPH helps to produce high quality coinoculations.

#### 5 UHPH versus colloidal stability and protein haze

The intense nanofragmentation of colloidal particles produce a higher stability of the colloidal fragments (see turbidity in Fig. 5) that remains even after fermentation but can be easily removed by conventional clarification. However, preliminary trials apparently shows that UHPH wines are less prone to suffer colloidal instabilities such as protein haze. Furthermore, it is possible that the high impacts and intense shear efforts in valve can produce the unfold of colloidal proteins facilitating the use of proteases without thermal treatments.

#### 6 Conclusions

UHPH is a powerful technology to improve and protect wine quality and to facilitate the use of emerging biotechnologies. It is a really interesting tool to eliminate or to reduce the use of SO<sub>2</sub>. Delicate flavor compounds as terpenes, thiols, anthocyanins and proanthocyanins are protected by UHPH ensuring wine quality. Depending on the settling level the use of UHPH can increase the contents of YAN affecting the formation of fruity esters during fermentation. Additionally, UHPH as an ultrashort technique do not produce thermal markers of degradation

as HMF. The nanofragmentation reduces the instability against protein haze and probably facilitate the use of proteases. So UHPH opens a lot of new possibilities in winemaking.



**Figure 6.** Final aspect of an UHPH wine after clarification and filtration with a very good transparency and paleness in absence of SO<sub>2</sub>.

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