





MALOLACTIC FERMENTATION QUESTIONS ANSWERED

Malolactic fermentation is a commonly used winemaking technique, which affects both stability and quality of the final product. In this factsheet, seven of the frequently asked questions about malolactic fermentation are answered.

1) What is malolactic fermentation (MLF)?

MLF is the decomposition of malic acid into lactic acid, which is carried out by lactic acid bacteria (LAB). MLF can be conducted spontaneously by creating a suitable environment for the growth of LAB, or commercial LAB can be inoculated. The most frequently used LAB for winemaking is *Oenococcus oeni (O.oeni)*, since it adapts and performs well in the wine media. On the other hand, another LAB, *Lactobacillus plantarum (L.plantarum)*, has been entered into the wine industry for the capacity of some of its strains to tolerate high alcohol and SO₂ levels.

2) Why MLF is performed in winemaking?

The main reasons for conducting MLF in winemaking are:

- **Deacidification of the wine:** MLF is a biological deacidification method. Upon completing a full malolactic fermentation, the acidity can be reduced, depending on the amount of malic acid consumed. Theoretically, fermentation of 1 g/L malic acid will result in a decrease of 0.6 g/L TA. The pH increase varies between 0.1 and 0.45 units (generally 0.1–0.25 pH units).
- Microbial stability: Malic acid is the preferred carbon source for various LAB, including spoilage LAB, which can alter the quality of the wine. Therefore, removing malic acid from wine media and eliminating this carbon source will increase the wine's microbial stability.
- Sensory characteristics: MLF provides changes in the organoleptic profile of the wine. These changes can be described as increasing aromatic complexity, enhancement of buttery and fruity notes, reduction of vegetative aromas and providing an impact on mouthfeel with a fuller body on the palate.

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3) What are the parameters of wine which affect the continuation of MLF?

- **pH:** It has a direct effect on the course of MLF. To give a general range, most LAB shows an active development at pHs around 3.5. Lower pHs around 2.9–3.0 will slow down their growth, with higher pHs (3.7–3.8) it's faster. The lower limit which can be tolerated by LAB depends on their strains.
- **Sulfur dioxide:** To give a general number, most LAB has difficulty growing at free SO₂ concentrations higher than 10 mg/L and total SO₂ higher than 50–100 mg/L (according to different sources). These levels are directly related to the level of pH since pH and free SO₂ affect the amount of molecular SO₂, which is the active form of sulfur dioxide. Moreover, adaptation and tolerance of LAB to different levels of molecular SO₂ is strain-dependent.
- Ethanol: Most LAB is inhibited in high ethanol concentration and has difficulty performing in levels above 13–14% volume, depending on the strain of LAB.
- **Temperature:** The ideal temperature for malic acid degradation in wine is around 20°C. Temperatures higher than 25°C or lower than 15°C slow down the process.
- Others: Initial malic acid concentration, fungicide residues (if any exist), phenolic compounds, level of dissolved oxygen, interaction with yeasts and nutrient levels in the wine have an impact on the course of MLF. The latter two will be explained in detail in the following questions.

Although it's possible to give some general numbers for the mentioned parameters, the growth, adaptation, tolerance and malic acid consumption rate of LAB in different wine media are strain-dependent. Taking this into account, it's important to be aware of the characteristics of the LAB strain that will be used. When using a commercial LAB starter, this information is provided by the manufacturer. Finally, keep in mind that these parameters inter-affect each other. Therefore, it's important to have an overall review of all the parameters before beginning MLF.

4) Is it necessary to make a nutrient addition for MLF?

It may be necessary to make a nutrient addition for MLF under less favourable or difficult conditions. This can be evaluated by examining the wine parameters mentioned above and considering the characteristic of LAB starter culture in terms of adaptation. LAB nutrient additions are especially recommended after a slow or sluggish alcoholic fermentation (AF). For this process, keep in mind that LAB cannot use inorganic nitrogen sources that yeast can use as a substrate, such as diammonium phosphate. Therefore, it's essential to apply bacteria nutrients that are available specifically for this purpose. Another recommendation for increasing nutrient levels for MLF is stirring the lees. Due to its content of nutrients and minerals, which can be used by LAB, stirring the lees during MLF provides an additional boost for this process. Yeast cell wall preparations (hulls) can be used for the same purpose.

5) What are the main reasons to choose between spontaneous or inoculated MLF?

For MLF, as in AF, there are different reasons to choose between spontaneous or inoculated fermentation.

- In inoculated fermentations, *O. oeni* strains, which are commercially available as starter cultures, can be used. These are specifically selected to secure completion of MLF and to produce fewer off odours. The winemaker also has the option to choose among different strains of *O. oeni* which best suits his/her wine style goals and which can help achieve the desired effects on aroma and organoleptic profile of the wine.
- In spontaneous fermentation, indigenous LAB is involved in MLF, and the winemaker has minimal control over the strains that will be performing. Therefore, it's necessary to be aware of the risks of various undesirable sensory changes such as the increase of acetic acid concentration and the possible production of higher levels of biogenic amines, especially for higher pH wines (>3.5).









6) Which parameters should be taken into account while conducting an inoculated MLF?

- Selection of LAB: It's crucial to choose a LAB strain that can perform well in the wine where it will be inoculated. Moreover, its effect on wine's sensory characteristics should be considered, as different LAB strains are shown to have different impacts.
- Compatibility with yeasts: According to the latest research, the interaction between yeasts and LAB is fundamental to ensure a successful AF and MLF. Certain yeast strains can have a stimulatory, inhibitory or neutral impact on the development of LAB. Commercial yeast manufacturers publish information that shows the level of compatibility of the yeasts with LAB. It's a good idea to review and plan this before moving forward with the fermentation processes.
- Time of inoculation: Inoculation of the LAB can be done simultaneously with the yeasts, during AF, or after AF. Although traditionally MLF was done after completing AF, recent research has demonstrated positive impacts of different scenarios of the time of inoculation, depending on the final goal.

7) How it's possible to monitor MLF?

It's possible to monitor the continuation of MLF and verify when it's completed by tracking malic and lactic acid concentration. Malic acid concentration will give a clear understanding of MLF, as it will be possible to see the amount of malic acid left unconsumed in the wine. Various methods are available to measure malic acid, including paper chromatography, enzymatic tests, FTIR analyzers or highperformance liquid chromatography (HPLC).

References and further reading

- Impact of the Timing and Temperature of Malolactic Fermentation on the Aroma Composition and Mouthfeel Properties of Chardonnay Wine. Sereni A., Phan, Q., Osborne J. and Tomasino E. Foods 2020, 9, 802; https://doi.org/10.3390/foods9060802
- Lactobacillus plantarum, a New Biological Tool to Control Malolactic Fermentation: A Review and an Outlook. Krieger-Weber S., Heras J.M., 2 and Suarez C. Beverages 2020, 6, 23; https://doi.org/10.3390/ beverages6020023

- Metabolism of Lactic Acid Bacteria and Lactic Acid Bacteria Development in Wine. In Handbook of Enology, The Microbiology of Wine and Vinifications Volume 1. Ribéreau-Gayon P., Dubourdieu D., Donèche B., & Lonvaud A. West Sussex: John Wiley & Sons, LTD. 2006.
- Malolactic Fermentation. Gil-Sanchez I., Sualdea B.A. and Moreno-Arribas M.V. In Red Wine Technology; Antonio M., Ed.; Elsevier: Amsterdam, The Netherlands,
- Malolactic Fermentation Importance of Wine Lactic Acid Bacteria in Winemaking, Lallemand. Retrieved from https://www.lallemandwine.com/wpcontent/uploads/2015/10/Lallemand-Malolactic-Fermentation.pdf
- Malolactic Fermentation A MoreManual!™ by Shea A.J. Comfort. Retrieved from https://morewinemaking. com/web files/intranet.morebeer.com/files/mlf09. pdf
- Achieving Successful Malolactic Fermentation, AWRI. Retrieved from https://www.awri.com.au/wp- content/uploads/2011/06/Malolactic-fermentation. pdf
- Evaluating the effect of using non-Saccharomyces on Oenococcus oeni and wine malolactic fermentation. Ferrando N., Araque I., Ortís A., Thornes G., Bautista-Gallego J., Bordons A., Reguant C. Food Research International 138 (2020) 109779 https://doi. org/10.1016/j.foodres.2020.109779
- A scientific study comparing the influence of three different strains of Oenococcus oeni on malolactic fermentation kinetics and chemical properties relating to the colour and flavour of Pinot Noir wine from Burgundy. Roisin Curley MW. Retrieved from: https:// www.mastersofwine.org/rp

More questions?

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