

## Nº1 CO-INOCULATION

Co-inoculation is the practice of inoculating selected wine bacteria at the beginning of the winemaking process shortly after yeast inoculation, usually 24 to 48 hours after yeast inoculation. This technique is advantageous because not only will it secure the malolactic fermentation (MLF), but also because there are definite advantages that are recognized by winemakers and professionals.

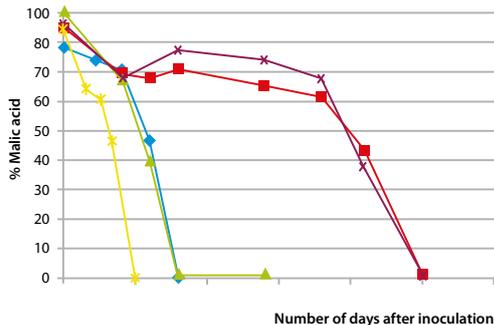
For a successful co-inoculation, some parameters are crucial for its success—choosing the right wine yeast, correctly rehydrated, good temperature management and the proper yeast nutrition strategy are keys point to integrate for any fermentations. Well-fed and healthy wine yeast and bacteria leads to complete and regular alcoholic and malolactic fermentations

Based on 20 years of experiences, and from the results of many collaborations between Lallemand and research center from France, Spain, Italie, South Africa, Argentina and Germany has shown the benefits of co-inoculation with either *Oenococcus oeni* or *Lactobacillus plantarum*.

### Fast and reliable Malolactic Fermentation (MLF)

MLF can be completed in between 3 days to 2 weeks depending on the type of musts and the bacteria used.

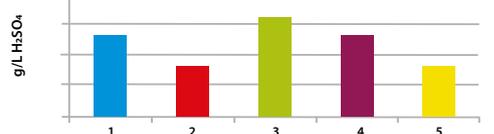
Kinetics of malic acid degradation after co-inoculation in Merlot 2014



Number of days after inoculation

- 1 Co-inoculation Alpha™ in pH 3.5 wines
- 2 Sequential inoculation Alpha™ in pH 3.5 wines
- 3 Co-inoculation Alpha™ in pH 3.9 wines
- 4 Sequential inoculation Alpha™ in 3.9 wines
- 5 Co-inoculation ML Prime™ in 3.9 wines

Volatility acidity



Malolactic Fermentation Induced by *Lactobacillus plantarum* in Malbec Wines from Argentina  
Lerena et al. S. Afr. J. Enol. Vitic., Vol. 37, No. 2, 2016

Exploitation of Simultaneous Alcoholic and Malolactic Fermentation of Incrocio Manzoni, a Traditional Italian White Wine  
Guzzon et al. S. Afr. J. Enol. Vitic., Vol. 37, No. 2, 2016

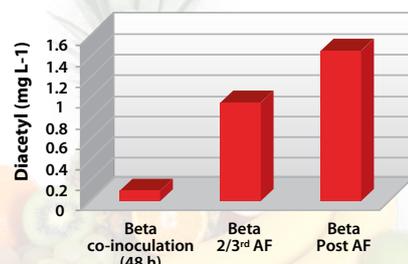
### Co-inoculation to produce fresh wines styles with low diacetyl content

Co-inoculation has a stronger impact on diacetyl production compared to the type of wine bacteria used. Our studies with institutes such as Neustadt and IFV and many winery trials have shown that whatever the wine bacteria used (and even with highest producer of diacetyl), co-inoculation always result in more fruit-driven wines styles and very low diacetyl content in wines.

Early results also show that in the case of co-inoculation the high content of sugars could repress the metabolism of the diacetyl, as opposed in post-alcoholic fermentation inoculation. And under the reductive conditions generate by the active yeast, diacetyl produced will be immediately reduced to the less active metabolites, acetoin and butanediol.

Impact of different malolactic fermentation inoculation scenarios on Riesling wine aroma  
Knoll et al. World J Microbiol Biotechnol (2012) 28:1143–1153

Impact of the timing of inoculation on diacetyl content in Chardonnay wines



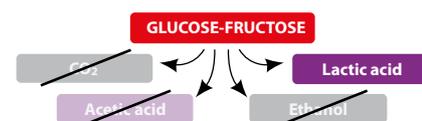
Using malolactic fermentation to modulate wine style  
AWR November 2015

### Co-inoculation : *Oenococcus oeni* or *Lactobacillus plantarum* (ML Prime™)

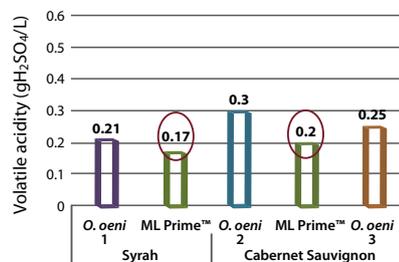
Co-inoculation is recognized as a secure and safe practice with our *Oenococcus oeni* cultures (VP41™, BETA™, ALPHA™, PN4™, OMEGA™, ...) for the different wines conditions.

As a bio-control agent for low acidity /high pH wines, ML Prime™ *Lactobacillus plantarum* is ideal as it completes MLF in 3-5 days during the AF with no risk of increased VA due to its specific metabolism. It enables early stabilization of wines, as soon as the AF is finished.

### Facultative heterofermentative metabolism (ML Prime™ metabolism)



### Volatility Acidity production in co-inoculated wine



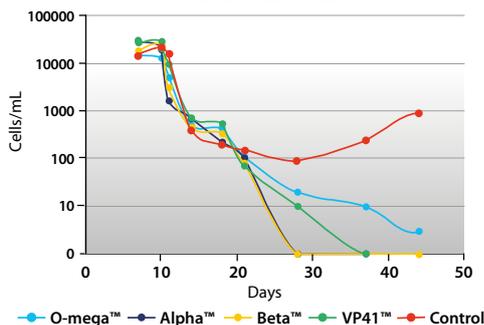
### Co-inoculation to limit the development of *Brettanomyces* and off-flavors

The increase in sugar levels, pH and sometimes lower SO<sub>2</sub> addition can influence the development of spoilage microorganisms, especially *Brettanomyces* which can produce phenolic off-odors in wines.

It is well known that the period from the end of AF to the start of MLF is particularly conducive to the development of *Brettanomyces*

Early inoculation with wine bacteria, either right after AF or in co-inoculation (24 hours after inoculation with yeast), has proven to be a simple and effective method for preventing the development of *Brettanomyces* and the production of ethyl phenols off-flavors. Our recent studies with IFV in Burgundy (Gerbaux) show that co-inoculation with our selected bacteria inhibit the growth of *Brettanomyces* (below 10 cell/mL) as opposed to the spontaneous control which is still contaminated with 500 cell/mL of *Brettanomyces* while the MLF is not completed and the wine is not stabilized.

*Brettanomyces* population in Pinot Noir in co-inoculation with different wine bacteria at the end of MLF



Influence of Inoculation with Malolactic Bacteria on Volatile Phenols in Wines  
Gerbaux et al. 2009 Am. J. Enol. Vitic. 60:2

« Co-inoculation is now recognized as a simple and safe practice for the management of malolactic fermentation for wine quality optimization. »