PROTOCOL P

Protocol for curative malolactic fermentation & difficult conditions

LACTOENOS® B7 DIRECT *Œnococcus œni* Bacterium.

Various parameters make it difficult to perform the MLF and their effect can be cumulative:

- Toxicity of the wine, through the presence of inhibiting compounds (pH, ethanol, SO_2, medium chain fatty acids).
- Deficiency in nutrients necessary for the proper growth of the lactic acid bacteria.

KEY POINTS:

1 - Detoxify the medium:

The addition of yeast hulls during pumping over in a closed circuit is effective in eliminating some bacteria-inhibiting compounds.

2 - Use a robust bacterium:

Not all bacteria have the same propensity to withstand the most restrictive conditions. **LACTOENOS® B7 DIRECT** is one of the most robust strains on the market in difficult conditions.

3 - Provide a medium suited to the growth of lactic acid bacteria:

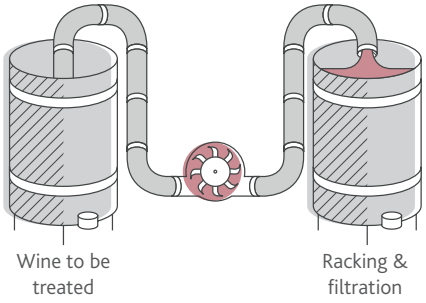
The use of **MALOBOOST®** during acclimatisation of **LACTOENOS® B7 DIRECT** corrects the nutritional deficiencies of the medium by providing the nutrients (nitrogen compounds, vitamins and minerals) essential for its proper growth.

CURATIVE MLF PROTOCOL FOR INOCULATION OF 100 hL OF WINE

A PRELIMINARY OPERATIONS IN TANK

1. If there is *Brettanomyces* contamination:

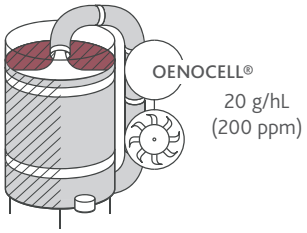
For populations greater than 10^2 cell/mL, plan to rack or even filter (1 µm) to eliminate this unwanted population.



2. Detoxify the medium:

This operation should take place 24 to 48 hours before bacterial inoculation to ensure an optimum survival rate.

- Add 20 g/hL (200 ppm) **OENOCELL®**.
- Mix thoroughly in a closed circuit.



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LACTOENOS® B7 DIRECT *Œnococcus œni* Bacterium.

B INOCULATE THE TANK TO BE TREATED - 100 hL

STEP 1 : 20 L reactivation medium, 0.2% of the final volume to be treated

- Mix 10 L water (non-chlorinated) with 10 L of the wine to be treated.
- Add 500 g **MALOBOOST®** (i.e. 5 g/hL based on the final volume of the tank).
- Mix thoroughly.
- Add 100 g **LACTOENOS® B7 Direct** (i.e. 1 g/hL based on the final volume of the tank).
- Mix thoroughly.
- Maintain the temperature at 20°C-22°C (68°F - 72°F).

Option 1: If the malic acid content of the wine is greater than 2 g/L:

- Monitor the breakdown of malic acid every day.
- When 2/3 of the malic acid is consumed, move on to step 2.

Option 2: If the malic acid content of the wine is less than 2 g/L:

- Leave the medium to stand for 24 hours, then move on to step 2.

STEP 2 : 5 hL starter - 5% of the final volume to be treated

- Take 5 hL of wine from the final batch to be inoculated.
- Add 200 g **MALOBOOST®** (i.e. 2 g/hL based on the final volume of the tank).
- Mix thoroughly.
- Add the entire reactivation medium from step 1.
- Maintain at 20°C - 22°C (68°F - 72°F).
- Determine the initial malic acid content then monitor it every 2 days.
- When 2/3 of the malic acid is consumed, move on to step 3.

Note: For wine pH <3.4 it is recommended to de-acidify the wine by adding potassium bicarbonate to raise the pH by 0.1 to 0.2 pH units.

STEP 3 : 100 hL tank

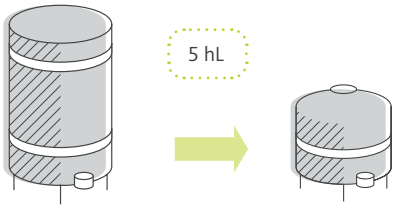
- Add 2 kg **MALOBOOST®** (i.e. 20 g/hL), previously rehydrated in 10 times its weight of wine.
- Thoroughly mix the starter before adding to the tank to be treated.
- Thoroughly mix the tank after adding the **MALOBOOST®** and the starter from step 2.

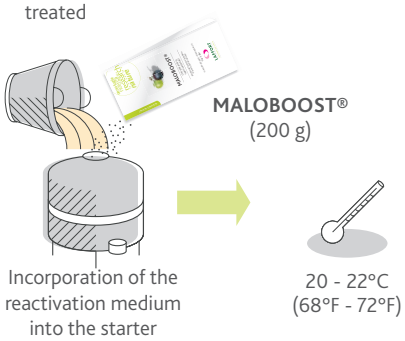
Important: maintain a stable temperature of between 20°C and 22°C (68°F - 72°F) during all these steps and until the end of MLF.

STEP1

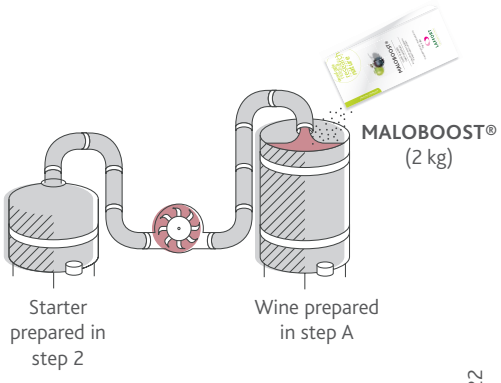
H_2O (10 L) + Wine (10 L) **+ MALOBOOST®** (500 g) **+ LACTOENOS® B7 DIRECT** (100 g)

STEP 2





STEP 3



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JB - AS - 01.12.22