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CELLOGUM L APPLICATION PROTOCOL FOR WHITE WINES

ADDITION BEFORE MICROFILTRATION

Preliminary Considerations for Cellogum use in White Wines

1) In order to stabilize wine solely using Cellogum L, the untreated wine must have an electric conductivity test result of $\Delta\mu S \leq 200$. If the conductibility is greater than this value, Cellogum L must be used in combination with another stabilization treatment.

2) The wine to be stabilized with Cellogum L must have been fined and be protein stable and clear (NTU <1). The stabilization process efficacy and speed is largely dependent on the initial wine clarity.

3) Instable proteins, positively charged colloids, lysozyme or protein fining agent residues present can lead to turbidity; however these can be easily eliminated by diatomaceous earth or plate filtration.

4) Once Cellogum is added, it is recommended to wait at least two days to one week before microfiltering and bottling (this waiting period depends on the Cellogum L rehydration process, homogenization into the wine, wine temperature, wine turbidity, wine filtration index). By doing so, wine stability increases and filter clogging and filterability reduction risks are minimized.

5) If the wine is completely clear (NTU < 1) at Cellogum L addition, homogenize the treated mass before microfiltration.

6) Do not use cross-flow filtration after CMC addition. Diatomaceous earth, plate and cartridge filtration are recommended.)

Practical Cellogum L Application Protocol

PHASE 1: PRELIMINARY LABORATORY TRIALS

1.1 FINDING THE BEST FINING PROTOCOL

Laboratory trials to find the best fining protocol must not only identify the fining agents that will give the best sensory results but also lead to complete protein stability. Therefore it is essential to test the clarified laboratory sample for protein stability.

1.1.1 Protein stability test Method:

a. Filter the wine sample with a 0.45 µm membrane.
b. Measure turbidity (T1) with a nephelometer/turbidimeter.
c. Heat the sample in a water bath up to 60°C and maintain this temperature for 24 hours.
d. Refrigerate for one hour.
e. Let the wine sample reach room temperature.
f. Measure turbidity (T2).
g. The wine is stable when T2-T1< 10 NTU*.

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What to do if the wine is not protein stable?

Treat the wine with a greater bentonite dose. Do not use CMC if the wine is not stable, otherwise turbidity will form in the bottle.

Note: *it is recommended to use 10-20% more bentonite than that identified in laboratory tests. In the winery, bentonite is not always used correctly (improper homogenization, insufficient contact time), therefore it is possible that after treatment the wine may be at the protein stability limit, which is a risk factor in CMC use.*

**This reference value was determined on the basis of numerous experiences and evaluations done on Italian white wines.*

1.2 FINDING THE BEST CELLOGUM L DOSAGE

Make increasing Cellogum L dosage additions on the clarified wine as describe in phase 1 in order to identify the dosage that is needed and is sufficient for tartrate stabilization.

1.2.1 Electric conductivity test (minicontact)

This test requires a specific instrument (Tartarcheck, Checkstab or other). Each instrument has specific instructions for use. The following method is the one used by Enartis with Tartarcheck. For other instruments, follow the instructions provided by the manufacturer.

The test can be done immediately after CMC addition if the wine is properly homogenized.

Method:

a. Fill the provided cuvette with a clarified and 0.45 µm membrane filtered wine sample treated with increasing Cellogum L dosages.
b. Cool the wine to 0°C and measure the electric conductivity (| S1).
c. Add 0.4 g of potassium bitartrate with the provided dispenser into the wine filled cuvette in order to supersaturate the wine.
d. Continuously agitate the wine for 30 minutes at 0°C and measure the electric conductivity again (| S2).
e. Verify the wine tartaric stability by calculating:

| S1 - | S2= | | S

ΔµS	Stability Degree
≤ 30	Very Stable
30 - 50	Stable
50- 70	At Risk
>70	Unstable

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1.2.2 Cold test

Method:

a. Place 125 mL of clarified and 0.45 µm membrane filtered wine treated with increasing Cellogum L dosages into a 125 mL clear glass bottle (see photo 1)
b. Keep the sample in refrigerator at -4°C for 6 days.
c. Observe daily the presence of crystals at the bottom of the bottle.
d. The presence of crystal is a sign of instability; on the other hand the absence of crystals indicates that the wine is stable.

Note: *the presented method is the standard one. If the wine is unstable, crystal precipitation can occur in a shorter amount of time, and therefore it is not necessary to wait until the sixth day. If the wine will be sold in very cold countries and very cold transport and/or storage temperatures are foreseen, it is possible to reduce the cold test temperature to -6°C/-7°C.*

Do not use a freeze test: *the resultant instability of the freeze test results from tartrate and potassium ion concentration following water freezing. Even a stable wine will be considered unstable by a freeze test.*

What to do if the wine is not tartrate stable?

Increase the Cellogum L dose (while respecting legal limits) or apply a short physical stabilization treatment (cold stabilization, electrodialysis, cation exchange resins) and complete the stabilization by adding Cellogum L.

Note: *even in this case it is a good idea to use a 10-15% greater Cellogum L dose in the winery than that identified in laboratory trials since CMC added before microfiltration cartridges is partly retained by the membranes.*

1.2 COLLOID STABILITY EVALUATION

Once the Cellogum L dosage necessary for clarified wine stabilization is identified as outlined in phase 1.2, it is possible to evaluate colloidal stability.

Colloidal stability test

Method:

a. Place 125 mL (see photo 1) of clarified and 0.45 µm membrane filtered wine treated with Cellogum L as identified in phase 1.2, in an oven at 40°C for 72 hours.
b. After 72 hours remove the sample from the oven and let it cool to room temperature.
c. Evaluate the colloidal stability by measuring the turbidity

NTU < 1.5 the wine is stable
1.5 <NTU < 3 the wine risks instability
NTU > 3 the wine is unstable

What to do if the wine is not colloid stable?

Return to the initial fining practices of phase 1: increase bentonite dose in order to eliminate positively charged colloids. Do not use CMC if the wine is unstable, failure to do so could lead to turbidity in bottle.

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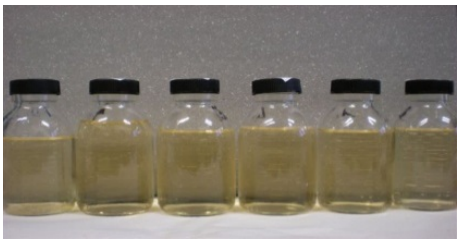
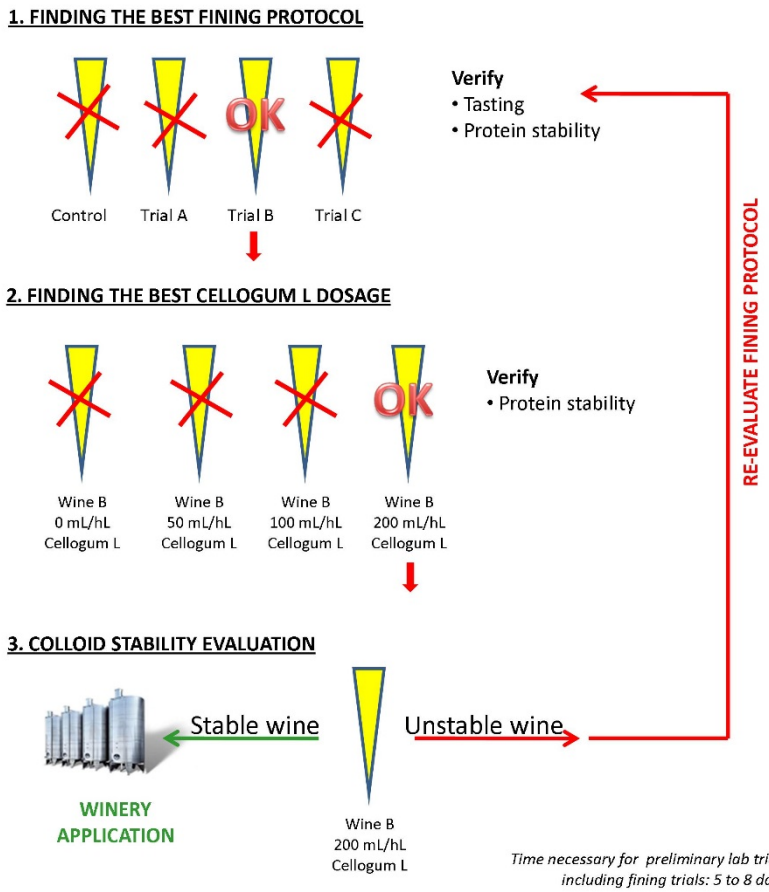


Photo 1

CELLOGUM L USE IN WHITE WINES

PRELIMINARY LABORATORY TRIALS



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PHASE 2: CELLOGUM WINERY APPLICATION

2.1 WINE PREPARATION

a. Clarify the wine as identified in the best fining protocol laboratory trials.
b. Filter (cross-flow, plate or diatomaceous earth).
c. Control the wine physical parameters.

2.2 WINE PHYSICAL PARAMETER CONTROLS BEFORE CELLOGUM ADDITION

2.2.1 TURBIDITY MEASUREMENT

If before CMC addition the wine is turbid, the turbidity as well as the filterability index will increase in the first hours after treatment. After 3 or more days (according to wine temperature) the turbidity decreases. In order to reduce this phenomenon and to avoid cartridge clogging, it is advisable to have an initial turbidity of less than 1 NTU.

What to do if the wine turbidity after filtration is greater than 1 NTU?

Filter again if possible.

If the turbidity is between 1 and 15 NTU and one wants to proceed with CMC addition in any case, it is important to know that part of the CMC will react with the turbidity and flakes will form- these will precipitate in 3-4 days, according to wine temperature. In this case it is necessary to overdose with CMC in order to compensate for these losses. Furthermore, before microfiltering, it is essential to:

- complete an intermediate filtration with diatomaceous earth or plates to avoid any clogging risks that the turbid particles can cause to microfiltration cartridges;
- control the wine tartrate stability again.

2.2.2 TEMPERATURE CONTROL

Verify that the wine temperature is greater than 12°C, ideally between 12 and 20°C.

What to do if the temperature is below 12°C?

If the wine temperature is below 12°C, CMC addition can cause a turbidity increase even in wines with an initial turbidity below 1 NTU; homogenizing the product in the treated mass is slower and the filterability index increases and stays high for a long time. It is therefore essential to delay bottling until the filterability index returns to an acceptable value.

2.2.3 CONTROLLING WINE FILTERABILITY

Modified filterability index (IF_m)

Method:

a. Filter around 700 mL of wine through a 25 mm diameter membrane with a 0.45 µm porosity at a constant 2.2 bar pressure.
b. Measure the time T, in seconds, necessary to filter 200, 400 and 600 mL of wine.
c. Calculate the filterability index

$$IF_m = \{ (T_{600}-T_{200}) -2(T_{400}-T_{200}) \}$$

A white wine is consider filterable if $IF_m < 10$.

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Note: *if IF_m < 10, the CMC addition does not drastically increase the filterability index and therefore the wine can be microfiltered and bottled after a few hours or days have passed since the treatment, according to wine temperature.*

What to do if IF_m is greater than 10?

If IF_m >10 and one wants to proceed with CMC addition, it is important to know that after treatment, an intermediate filtration with diatomaceous earth or plates will be necessary before cartridge microfiltration. In this case, part of the CMC will be lost during filtration, therefore it is necessary to overdose with CMC and control the wine tartrate stability again before bottling.

2.3 CELLOGUM PREPARATION AND ADDITION

2.3.1 CELLOGUM L PREPARATION

a. Calculate the Cellogum L amount necessary for the volume of wine to be treated.
b. "Rehydrate" Cellogum L in at least 3-4 times its volume of wine (for example, add 1 liter of Cellogum L to 3-4 liters of wine).
c. Agitate well and for a long time to assure perfect homogenization of Cellogum L in wine.
d. Let sit for at least 2-3 hours.
e. Mix the Cellogum L and wine solution and add to the mass to be treated, if possible using a Venturi system and being careful to homogenize it well throughout the mass.
f. Wait at least 48 hours before microfiltering and bottling the wine.

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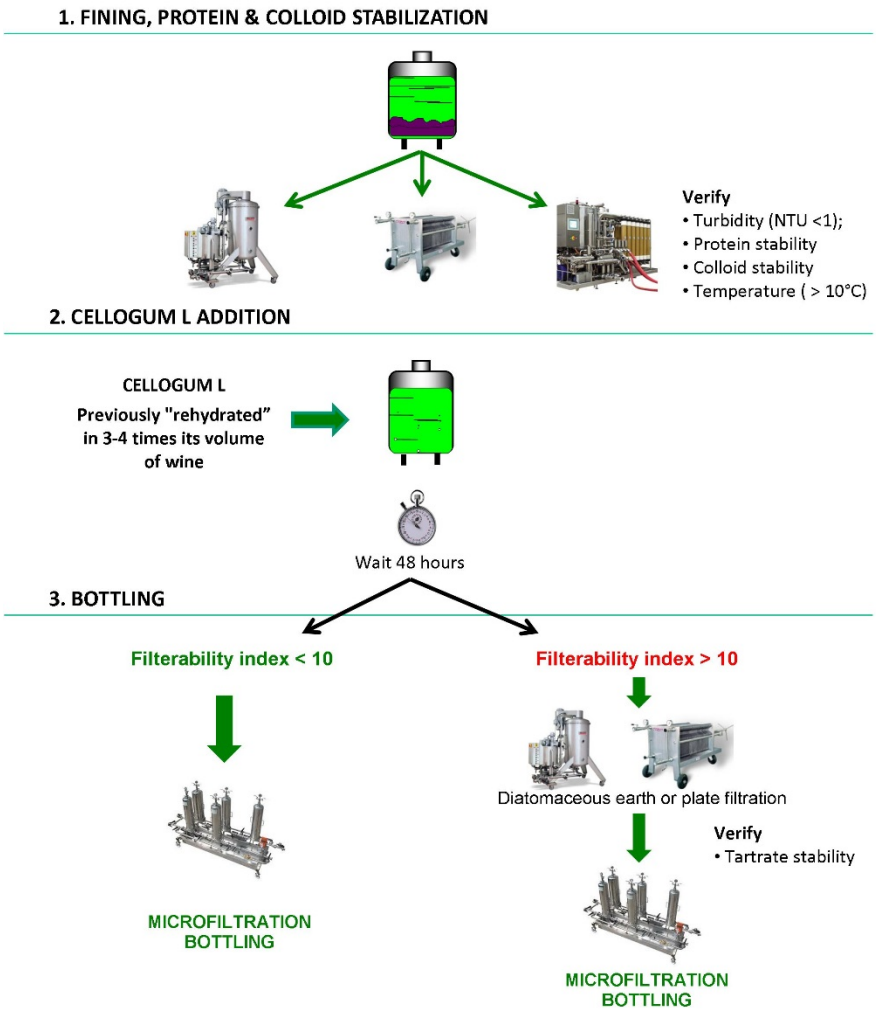
CELLOGUM APPLICATION IN WHITE WINES

PRE-BOTTLING APPLICATION



CELLOGUM APPLICATION FOR WHITE WINES

Pre-Bottling Application

1. Fining, Protein and Colloid Stabilization
    a. Verify – Turbidity (<1 NTU), Protein Stability, Colloid Stability, Wine Temperature
2. Cellogum L Addition
    a. Previously "rehydrated" Cellogum L in 3-4 times its volume of wine- Waiting Time 48 hours
3. Bottling
    a. Filterability Index <10
        i. Microfiltration and Bottling
    b. Filterability Index > 10
        i. Diatomaceous earth or plate filtration
        ii. Verify tartrate stability

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iii. Microfiltration and Bottling

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