PREPARING WINE FOR BOTTLING

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PREPARING WINE FOR BOTTLING

• Introduction
• Clarification and Fining
• Stabilization
  • Microbial Stabilization
  • Protein Stabilization
  • Color Stabilization
  • Tartrate Stabilization
• Filtration and Filterability
• Bottling Timelines
INTRODUCTION
Clarity and Stability

“Clarity is an essential quality required by consumers, especially for white wines.

Nowadays, the only normally acceptable deposit is red coloring matter in old wines. Sediment should not appear until the wine is four or five years old, and then only in small quantities, and easy to eliminate by decanting.

Wine must not only be clear at the time of bottling, but also maintain its clarity during aging and storage for an indefinite period, whatever the temperature conditions.”

Pascal Ribéreau-Gayon, Handbook of Oenology.
Key consumers insights

Consumer studies:
- CHINA: 1027 wine consumers of middle-high class,
  66% men,
  43% 30-39 years old,
  80% from Shanghai/Beijing
- USA: 2053 people regularly consuming wine,
  56% women,
  46% 35-54 years old

In China, 50% of the consumers had previously seen wines with sediments; 33% in USA.

In the US and in China, almost 50% of the consumers surveyed view any sediment in bottle in a negative way.

Although 30% of the consumers surveyed understand that sediment in red wine is a consequence of the winemaking process, 40% would not buy a wine with sediment.

In both of the consumer groups surveyed, the presence of tartrate crystals / sediment was perceived in a negative way.

Only 16% of American and 32% of Chinese consumers would buy wine with sediment, including red wine.
Market status on wine stability
Analysis of wines from super market shelves

Study carried out in France:

In 2010 with 63 bottles
(41 red, 12 white & 10 rosé)

In 2016 with 80 bottles
(43 red, 32 white & 5 rosé)

- **Microbiological Stabilization**
  - Non-compliant: pop > 1 CFU/10 mL
  - | AB | LB | Brett |
  - | 52% | 15% | 9% |

- **Protein stabilization**
  - Heat test - $\Delta$ NTU < 2
  - 17%

- **Tartaric stabilization**
  - ISTC50 ≤ 3μS or crystallization test
  - 66%

- **Coloring matter stabilization**
  - Cold test - $\Delta$ NTU < 10
  - X

- **Protein stabilization**
  - Heat test - $\Delta$ NTU < 2
  - 13%

- **Tartaric stabilization**
  - ISTC50 ≤ 3μS or crystallization test
  - 59%

- **Coloring matter stabilization**
  - Cold test - $\Delta$ NTU < 10
  - 72%
What do we prepare wines for?

To reach clarity and stability

Before bottling, the aim is to:

• **Obtain total clarity** by appropriate clarification methods.
• **Achieve stability of that clarity** by means of efficient treatments.

We must understand the consequences of each treatment.

Examples:

• **Filtration clarifies, but does not stabilize, except from a microbiological standpoint.**
• **Fining has a double effect: clarifying and stabilizing.**
• **Certain colloid additions improve and prolong the stability equilibrium but do not clarify the wine.**
Clarification

- Elimination of a current haze
- Short term

Stabilization

- Colloidal particle precipitation avoiding potential future precipitation
- Long term
- Clarity preservation

Existing haze

Existing haze

Potential haze
CLARIFICATION & FINING PRINCIPLES
"Clarification aims to eliminate wine haze, consisting of visible and/or light absorbing or deflecting particles. These are particulate suspensions of yeasts, bacteria, crystals, vegetal debris visible microscopically or to the eye, but also colloidal solutions."

Knowing and Making Wine, Jacques Blouin and Emile Peynaud, 2005
What Does Fining Achieve

- Turbidity reduction
- Phenolic compound elimination
- Organoleptic polishing
- Coloring matter stabilization
Particle size of protein fining agent

Vegecoll®

Liquid Gelatin
0.1-1 µm

Vegecoll®
< 10 nm

Albumin
< 10 nm

Pea
< 10 nm

Yeast extract
0.1-1 µm

The fining phenomenon
Wine turbidity evolution during fining

It is the compromise between flocculation capacity and sedimentation speed that optimises the clarification effectiveness.
The flocculation capacity depends on the nature and the dose of the fining agent and of the wine.

A high flocculation is not associated with a higher clarification speed.

The sedimentation speed and the clarification speed depend on the size and the weight of the flake.

It is the compromise between flocculation capacity and sedimentation speed that optimizes the clarification effectiveness.
Zêta Potential
Indicates the balance of attractive and repulsive forces of a particle in a medium

- When none of these forces is predominant, particles flocculate.
- This number helps in the prediction of fining agent reactions, because it indicates the flake formation type.
The fining phenomenon

Fining agents

Fining agents with fast sedimentation speed

Flakes are large size and heavy

Fining agents with slow sedimentation speed

Flakes are smaller size and light

A fining agent that shows a fast clarification, will automatically produce a high volume of lees.
Zeta Potential and Clarification

Classification of fining agents with respect to sedimentation rate

The physical phenomenon can be explained by the Zeta Potential

A fining agent with a high Zeta Potential (positive or negative) clarifies quickly

Red wine pH 3.6
Zeta Potential and Clarification

Classification of fining agents with respect to sedimentation rate

The Zeta Potential changes with the wine pH and the same phenomenon is observed on white wines.

Other physico-chemical parameters have an impact (particle size)

White wine pH 3,4

- The fining phenomenon
The colloidal stabilisation phenomenon

Source: Cédric Saucier, 1997, Wine tannins: Study of their colloidal stability – PhD thesis at Université de Bordeaux II
Fining trial in the lab: DIY

For a successful lab fining trial:

✓ Adjust free SO$_2$ to 30mg/L if necessary
✓ Use a 375mL bottle (minimum)
✓ Keep a control of each wine batch
✓ Keep wines at room temperature
✓ Add the fining agent(s)
✓ Always try at least 2 different fining agents at 2 different doses
✓ Taste blind after 2 to 3 days (and measure turbidity if possible)
Over-fining

What is over-fining?

Part of the fining agent remains in suspension in the wine (gelatin and other fining agents)

To avoid over-fining:

✓ Thoroughly homogenize the fining agent in the wine
✓ Do not add more fining agent than necessary
✓ Keep a treatment temperature lower than 15°C / 60°F
✓ Use silica gel prior to the fining agent

What to do in case of over-fining?

Bentonite (white) or tannins (red)
Keys for a successful fining treatment

• Validation of the absence of glucan in the wine

• Choice of the fining agent nature and dosage
The lab performs a complete analysis of the wine and does the fining trials.

• Incorporation
The fining agent must be incorporated homogenously into the entire volume of wine to be treated; using a venturi like Oenodoseur is recommended.

• Fining timing
Dependent on: volume to be treated, temperature, initial turbidity and fining agent type.

• Racking
Careful racking to eliminate entire fining lees.

Note: any enzyme treatment beforehand will improve the fining quality
Fining treatment

Incorporation:
- progressive
- homogenous

During a pump-over:
- using a Venturi system (OENODOSEUR)
<table>
<thead>
<tr>
<th>Products</th>
<th>Preparation Addition</th>
<th>Over-finishing risk</th>
<th>Contact time before racking and filtration</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Gelatins:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Gecoll Supra</strong></td>
<td>Liquid: Add directly to the wine during a pump-over.</td>
<td>Yes</td>
<td>1 to 3 weeks</td>
</tr>
<tr>
<td><strong>Gelarom</strong></td>
<td>Powder: Dissolve in warm water (40°C): Add directly to the wine during a pump-over while maintaining the temperature.</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Gel. Extra N°1</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Egg Albumin:</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Ovoclaryl</strong></td>
<td>Powder: Dissolve in 5 to 6 times its weight in water. Add directly to the wine during a pump-over.</td>
<td>Yes</td>
<td>1 to 3 weeks</td>
</tr>
<tr>
<td>Products</td>
<td>Preparation Addition</td>
<td>Over-fining risk</td>
<td>Contact time before racking and filtration</td>
</tr>
<tr>
<td>---------------------</td>
<td>---------------------------------------------------------------------------------------</td>
<td>------------------</td>
<td>--------------------------------------------</td>
</tr>
<tr>
<td><strong>OENOLEES®</strong></td>
<td>Dissolve in 5 to 6 times its weight in water. Add directly to the wine during a pump-over.</td>
<td>No</td>
<td>2 to 4 weeks</td>
</tr>
<tr>
<td><strong>ICHTYOCOLLE®</strong></td>
<td>Dissolve in 100 times its weight in water. Let swell for 2 hrs while stirring to ensure a good dispersion. Add directly to the wine during a pump-over.</td>
<td>Yes</td>
<td>2 to 4 weeks</td>
</tr>
<tr>
<td><strong>CASÉINE CASÉI +</strong></td>
<td>Dissolve in 10 times its weight in water. Add directly to the wine during a pump-over.</td>
<td>No</td>
<td>1 to 3 weeks</td>
</tr>
<tr>
<td><strong>POLYMUST® Rosé POLYMUST® PRESS</strong></td>
<td>Dissolve in 10 times its weight in water. Add directly to the wine during a pump-over.</td>
<td>No</td>
<td>5 days to 3 weeks</td>
</tr>
<tr>
<td><strong>VEGECOLL®</strong></td>
<td>Dissolve in 10 times its weight in water. Add directly to the wine during a pump-over.</td>
<td>Yes</td>
<td>5 days to 2 weeks</td>
</tr>
</tbody>
</table>

To see VEGECOLL® preparation
<table>
<thead>
<tr>
<th>Products</th>
<th>Preparation Addition</th>
<th>Over-finishing risk</th>
<th>Contact time before racking and filtration</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>PVPP (VINICLAR®)</strong></td>
<td>Dissolve in 5 to 6 times its weight in water. Add directly to the wine during a pump-over.</td>
<td>No</td>
<td>1 to 3 weeks</td>
</tr>
<tr>
<td><strong>MICROCOL® CL MICROCOL® Alpha</strong></td>
<td>Dissolve in 6 to 10 times its weight in hot water (50°C), keep stirring for 2 hrs, let it swell for 12 to 24 hrs. Add directly to the wine during a pump-over.</td>
<td>No</td>
<td>5 days to 3 weeks</td>
</tr>
<tr>
<td>Silica gel (SILIGEL®)</td>
<td>Add directly to the wine or after dilution in water or wine. Shake vigorously. Add prior to the organic fining agent</td>
<td>No</td>
<td>With the fining agent</td>
</tr>
<tr>
<td>Tannin</td>
<td>Add prior to the organic fining agent</td>
<td></td>
<td>With the fining agent</td>
</tr>
</tbody>
</table>

*To see MICROCOL® ALPHA preparation*
STABILIZATION
Questions to ask ourselves:

What are the risks linked to instability?

Regarding my wine:

A. Is my wine stable?
B. What are the treatment options for stabilization?
C. How can I choose the necessary and appropriate treatments?
D. How do I check the efficacy of the treatment?
E. What are the parameters influencing stability?
F. How to best carry out the treatment in the cellar?

... for each stability!
Stabilization Roadmap

4 to 6 weeks prior to bottling

Microbiological stabilization

1 to 2 weeks prior to bottling

Protein stabilization

Bottling

Or

Color stabilization

Tartaric stabilization

Enzyme, fining agent, lysozyme, SO₂, chitosan...

Finishing tannins

Bentonite

CMC Mannoproteins

Gum Arabic (D-2)
SO₂ & ascorbic acid (D-1)
Sorbic acid (D-1)

Acidification/deacidification

NTU < 5  CI < 20
T° > 15°C (60°F)
Checking Free SO₂, CO₂, etc., Complete analysis and tasting
Stabilisation roadmap: change in mindset

4 to 6 weeks prior to bottling

1 to 2 weeks prior to bottling

Bottling

**Microbiological stabilization**

**Protein stabilisation**

Or

**Colouring matter stabilisation**

**Tartaric stabilisation**

Filterability index monitoring: turbidity < 5 and CI < 20

**NTU < 5  CI < 20  
T° > 15°C (60°F)**

Checking Free SO₂, CO₂, etc., Complete analysis and tasting

Enzyme, fining agent, lysozyme, SO₂, chitosan...

Finishing tannins

Bentonite

Metatartaric acid
CMC
Mannoproteins

Gum Arabic (D-2)
SO₂ & ascorbic acid (D-1)
Sorbic acid (D-1)

Acidification/deacidification
MICROBIOLOGICAL STABILIZATION
### Risks linked to absence of microbiological stability

<table>
<thead>
<tr>
<th>HAZE IN BOTTLE</th>
<th>Yeasts: re-fermentation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><em>Brettanomyces</em>: phenolic character</td>
</tr>
<tr>
<td></td>
<td>Acetic bacteria: volatile acidity</td>
</tr>
<tr>
<td></td>
<td><em>Lactic bacteria</em>: “maladie de la graisse”, biogenic amines</td>
</tr>
<tr>
<td></td>
<td><em>formation of a haze on the surface</em></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>TYPES OF WINE CONCERNED</th>
<th>All types of wine</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>HAZE FORMATION FAVORED BY</th>
<th>- Quality of wine preparation to bottling</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>- Molecular SO$_2$ level (i.e. take into account pH)</td>
</tr>
<tr>
<td></td>
<td>- Wine storage conditions: exposure to heat</td>
</tr>
<tr>
<td></td>
<td>- Closure quality</td>
</tr>
<tr>
<td></td>
<td>- Residual sugar level</td>
</tr>
</tbody>
</table>
Haze / Alterations due to micro-organisms:

Accidents still happen **TOO** often!

ANALYSES OF WINES SAMPLED ON SUPERMARKET SHELVES:

Non-compliant wine: pop > 1 CFU/10 mL

<table>
<thead>
<tr>
<th></th>
<th>2010</th>
<th></th>
<th>2016</th>
</tr>
</thead>
<tbody>
<tr>
<td>AB</td>
<td>52%</td>
<td>AB</td>
<td>46%</td>
</tr>
<tr>
<td>LB</td>
<td>15%</td>
<td>LB</td>
<td>11%</td>
</tr>
<tr>
<td>Brett</td>
<td>9%</td>
<td>Brett</td>
<td>28%</td>
</tr>
</tbody>
</table>

In 2010: 36%  
In 2016: 47%
A. Is my wine stable from a microbiological standpoint?

Before each bottling, perform a complete microbiological assessment on the final blend; by plate cell count on gel medium specific for:

- Yeasts
- Yeasts Brettanomyces (red wine)
- Acetic bacteria
- Lactic bacteria

Today there is no regulations regarding the micro-organism population in wine after bottling.

A test post- bottling helps to assess the “quality” of this crucial and definitive step

Thresholds are defined by some buyers:
From < 1 CFU / 10 mL to < 1 CFU / 750 mL (sweet wines)
B. Options for treatment?

Tools available to reduce microbial load:

- SO₂ addition (active SO₂)  
  Non selective action
- Enzyme addition
- Fining
- Lysozyme  
  Selective action on bacteria only
- Sorbate  
  Selective action on yeasts only

Physical treatment: pay attention to potential "collateral" damages when treatments take place early in the aging process

- Flash pasteurization  
  Impact on some aromatic compounds (esters)
- Filtration (sterile)  
  Careful regarding microbiological emptiness
- Cross flow filtration  
  Sterilizing filtration vs sterile filtration + impact on coloring matter stability
- OENObrett®  
  Selective action on Brettanomyces (+ Bacteria)
Impact of crossflow filtration on coloring matter stability (2012 filtered wine in December 2012 measures 15 days after treatment)

Impact of Flash pasteurisation on some esters (2011 wine *flashed* in January 2012. Analysis 10 days after treatment)
C. How do I check the efficacy of treatment?

Example of reduction of the microbial load through fining

WINE FINED WITH GELATIN

Wine racked, unfined

BRETT = 3.1. 10^4 CFU/ mL

20 mL/hL

BRETT = 1.8.10^2 CFU/ mL

40 mL/hL

BRETT = 1.10^2 CFU/ mL
Adding a pectinase and β-glucanase blend

<table>
<thead>
<tr>
<th></th>
<th>Unracked control wine</th>
<th>Racked wine</th>
<th>Wine + EXTRALYSE® Then racked</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brettanomyces (D+10 after operation)</td>
<td>4.4.10^4 CFU/mL</td>
<td>2.2.10^2 CFU/mL</td>
<td>&lt; 1/10 mL</td>
</tr>
<tr>
<td>4-EP + 4-EG (µg/L) (end of aging)</td>
<td>660</td>
<td>290</td>
<td>120</td>
</tr>
</tbody>
</table>

*From the work of Vincent Renouf*
Curative use of **OENO Brett**

![Graph showing the reduction of Brettanomyces population over days with and without OENO Brett.](image)
What is Chitosan?

*N*-acetyl-*D*-glucosamine & *D*-glucosamine units.

**Chitin and cellulose:**

- Most abundant polysaccharides: $10^{11}$ tonnes/year
- Origin: crustaceans (15-30 % MS), fungus (42 % in *A. niger*)

**Characterization of chitosan:**

- Deacetylation degree (DD)/acetylation (DA)
- Molecular weight (MMw or MMn)

In winemaking: Non-allergenic and natural polysaccharide, non-animal, extract from the fungus *Aspergillus niger.*
Curative use of Chitosan
Microscopic illustrations

Action of the Chitosan on Brettanomyces. 8 days after treatment with 100 mg/L.

✓ Destruction of the wall and cell membrane.

✓ Breakdown of the intracellular medium (no more cellular organization).

SEM: Scanning Electronic Microscopy

Control T0 – x 20000
Control T0 – x 40000
Treated terms – x 20000
Treated terms – x 40000

Visible organels
Curative use of Chitosan
Microscopic illustrations

In wine

Day 0 (untreated)

Day + 1 (treated)

Day + 4 (treated)

Day + 8 (treated)

Evolution of the cell structure of *Brettanomyces* within 8 days of treatment with 10 g/hL of Chitosan- SEM (x 20,000)

Compared with the model medium, the cells already seem affected by the environment, however, their walls and membranes are normal and the architecture of the cell tends to demonstrate their viability.

The day after treatment cells are already affected by Chitosan

- Intracellular breakdown.
- Wall and membrane damage.

The majority of cells do not exhibit a wall and viable membrane structure any more.

Next step in the process of cell death.
Preventative use of OENOBrett®

Concept of persistence: experimental protocol

Wine contaminated by *Brettanomyces*
Red wine from South East of France / *Brettanomyces Population:* 4.8x10^4 cell/mL - 332 µg/L of E-4-P + E-4-G

OENOBrett® treatment (10 g/hL)

Racked wine

Wine left on OENOBrett®

Uncontaminated

Re-contamination by *Brettanomyces*

- After racking: T0
- 1 month later: T+1mo
- 2 months later: T+2mo

Uncontaminated

Re-contamination by *Brettanomyces*

- 8 days after Chitosan treatment: T0
- 1 month later: T+1mo
- 2 months later: T+2mo

Ethyl phenol analysis at 4 months.

In parallel, two control wines are followed:
- An untreated control, to watch the evolution of *Brettanomyces* populations.
- A sterile filtered control as a physical stabilization alternative.
Preventative use of OENO Brett®

Concept of persistence

Trial n°1: Treated wine – racked after 8 days.

New contamination after 1 week

- Untreated wine
- Treated wine
- Treated wine, re-contamination after 1 week, n°1
- Treated wine, re-contamination after 1 week, n°2
- Sterilized wine and contamination after 1 week
Preventative use of *OENO* Brett®

**Concept of persistence**

**Trial n°2:** Treated wine – unracked.

**New contamination after 1 week**

- **Untreated wine:**
  - Days 0-30: Drop in contamination
  - Days 30-140: Increase in contamination to 2252 µg/L

- **Treated wine:**
  - Days 0-30: Drop in contamination
  - Days 30-140: Increase in contamination to 1637 µg/L

- **Treated wine, recontamination after 1 week:**
  - Days 140-150: Increase in contamination to 277 µg/L

- **Sterilized wine and contamination after 1 week:**
  - Days 150-160: Increase in contamination to 536 µg/L
  - Days 160-170: Increase in contamination to 316 µg/L
The concentrations of 4-ethylphenol and 4-ethyl guaiacol were determined at the end of the fourth month.

<table>
<thead>
<tr>
<th>4EP + 4EG concentration in μg/L</th>
<th>Treated wine, racked after 8 days</th>
<th>Treated wine left on Oenobrett for 3 months</th>
<th>Wine sterilized by filtration (0.45μm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Uncontaminated</td>
<td>316 - 350</td>
<td>280</td>
<td>1750</td>
</tr>
<tr>
<td>Contaminated T0</td>
<td>589 - 384</td>
<td>277 - 536</td>
<td>1637</td>
</tr>
<tr>
<td>Contaminated T+1month</td>
<td>1112 - 341</td>
<td>304 - 308</td>
<td>1657</td>
</tr>
<tr>
<td>Contaminated T+2month</td>
<td>288 - 294</td>
<td>274 - 300</td>
<td>1631</td>
</tr>
</tbody>
</table>

**A.** High production of ethyl phenol in the untreated wine!

**B.** No change in the concentration of ethyl phenols.

**C.** Spontaneous development of Brettanomyces prompted a significant production of phenols.

**D.** The wine left on Oenobrett is less sensitive to recontamination than the wine racked after 8 days.
Stabilisation roadmap: change in mindset

4 to 6 weeks prior to bottling

1 to 2 weeks prior to bottling

Bottling

Microbiological stabilisation

Protein stabilisation

Or

Colouring matter stabilisation

Tartaric stabilisation

Filterability index monitoring:
turbidity < 5 and CI < 20

Enzyme, fining agent, lysozyme, SO2, chitosan...

Finishing tannins

Bentonite

Metatartaric acid
CMC
Mannoproteins

Gum Arabic (D-2)
SO2 & ascorbic acid (D-1)
Sorbic acid (D-1)

NTU < 5    CI < 20
T° > 15°C (60°F)
Checking Free SO₂, CO₂, etc., Complete analysis and tasting
PROTEIN STABILIZATION
## Risks linked to absence of stability

<table>
<thead>
<tr>
<th>PROTEIN HAZE</th>
<th>Protein denaturation and flocculation under heat conditions in case of an accidental temperature increase</th>
</tr>
</thead>
<tbody>
<tr>
<td>TYPE OF WINE CONCERNED</td>
<td>White and rose wines: off-white flakes formation</td>
</tr>
</tbody>
</table>
| HAZE FAVORED BY | • Inefficient fining  
• Bottling filtration quality: *a stable wine before bottling that became unstable by retention of protective colloids in case of a clogging filtration*  
• Certain additives like Lysozyme at bottling  
• Poor quality natural corks: possibility of releasing *cork tannins*.  
• *Wine storage* conditions: wine exposed to heat. |

### Analyses of Wines Sampled on Supermarket Shelves:

<table>
<thead>
<tr>
<th>Year</th>
<th>Percentage</th>
<th>White/ Rosé</th>
</tr>
</thead>
<tbody>
<tr>
<td>In 2010</td>
<td>17%</td>
<td>34% white / 2% rosé</td>
</tr>
<tr>
<td>In 2016</td>
<td>13%</td>
<td>9% white / 40% rosé</td>
</tr>
</tbody>
</table>
A. Is my wine stable from a protein standpoint?

Review of protein stability tests

Over time, different lab tests have been used to evaluate protein haze risk prior to bottling.

They are based on:

• **Flocculation of proteins under different conditions:**
  - Heat
  - In presence of tannins
  - In presence of chemical reagents

• **Protein presence:**
  - Immunological test
A. Is my wine stable from a protein standpoint?

*Review of protein stability tests*

1. **Tests by chemical denaturation:**
   *Based on reactions with a chemical agent*
   
   (phosphomolybdic acid, trichloroacetic acid, etc.)

   ✓ *Do not model the natural phenomenon of protein haze formation*
   
   Reagents used are not specific of thermo-unstable proteins.

   ✓ *These tests systematically lead to an overestimation of the bentonite dose.*
### A. Is my wine stable from a protein standpoint?

*Review of protein stability tests*

2. **Immunological test:**
   
   *Based on the reaction with “specific” antibodies*
   
   - Does not model the natural phenomenon of protein haze formation
   
   Reagents used are not specific of thermo-unstable proteins.

<table>
<thead>
<tr>
<th>CONTROLS</th>
<th>Sauvignon Bordeaux 2008</th>
</tr>
</thead>
<tbody>
<tr>
<td>Immunological test</td>
<td>Heat test</td>
</tr>
<tr>
<td>Positive control</td>
<td>Immuno. test</td>
</tr>
<tr>
<td>Negative control</td>
<td>0.9 NTU</td>
</tr>
</tbody>
</table>

Wine stable
A. Is my wine stable from a protein standpoint?

Review of protein stability tests

3. Heat test in presence of tannins:
   Based on the reaction with tannins and exposure to heat

✓ Does not model the natural phenomenon of protein haze formation.

✓ These tests systematically lead to an overestimation of the bentonite dose.
A. Is my wine stable from a protein standpoint?

Review of protein stability tests

4. Heat test:

Based on exposure to heat in perfectly defined conditions (80°C 30min, room temp. 45 min)

✓ Only test modelling the natural phenomenon of protein haze formation
✓ Specific to thermo-unstable proteins
A. Is my wine stable from a protein standpoint?

*Review of protein stability tests*

**Heat test Protocol**

a. Measure the wine turbidity: if > 2 NTU, filter the wine (cellulose ester membrane, 0.65 µm) => turb1

b. Heat the wine for 30 minutes at 80°C / 176°F.

c. Let it cool for 45 minutes at room temperature.

d. Measure the wine turbidity again => turb2

*In case of a shorter cooling time (eg putting the tube under cool running water): Risk of under-estimation of the bentonite dose (minor haze).*

*In case of a longer cooling time than 45 minutes: Risk of over-estimation of the bentonite dose (formation of a haze not due to the thermo-instable protein fractions).*

The wine is UNSTABLE if Δ NTU (turb2 – turb1) > 2
Stability must be measured with an appropriate test

*Chemical denaturation or immunological protein stability tests:*

- Does not model the “natural” phenomenon of protein haze
- Precipitates all proteins, whether they are heat sensitive (thermo-unstable) or not
- Leads to *bentonite doses greater than necessary to stabilize the wine (= eliminate thermo-unstable proteins).*

⇒ *The heat test* is the only test modelling the natural phenomenon of protein haze formation
B. What are my options in terms of treatment?

The bentonite treatment is the only tool preventing protein haze to this day.

There are 3 types of bentonites:
- Calcium bentonites (Na\(^+\)/Ca\(^{2+}\)<1)
- Sodium bentonites (Na\(^+\)/Ca\(^{2+}\)>1)
- Calcium bentonites, sodium activated

Inter layer space:
- Sodium bentonite: 100A
- Calcium bentonite: 10A

Sodium bentonites swell more and adsorb proteins more effectively. Calcium bentonites precipitate and clarify more effectively.

All bentonites in the Laffort range are natural for a better stability and shelf life.
<table>
<thead>
<tr>
<th>Dominant Cation</th>
<th>Swelling rate</th>
<th>Exchange capacity</th>
<th>Lees sedimentation</th>
<th>Aromatic preservation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sodium</td>
<td>High</td>
<td>High</td>
<td>Medium Fluffy lees</td>
<td>+++</td>
</tr>
<tr>
<td>Calcium</td>
<td>Medium</td>
<td>Low</td>
<td>Rapid Compact lees</td>
<td>+++(+)</td>
</tr>
</tbody>
</table>

*Note: VEGECOLL®, or a combination of SILIGEL® and GELAROM®, at a low rate helps improve sedimentation.*
B. What are my options in terms of treatment?

Effects of bentonite treatment

An excessive bentonite treatment does not lead to over-fining. But it does, undeniably, have an impact on the wine organoleptic quality.

The necessary bentonite dose for stabilization must therefore be precisely determined.

From Moine-Ledoux, 2006
C. How do I choose the appropriate treatment?

Following the heat test results: \( \text{if } \Delta \text{NTU} > 2 \)
\[ \Rightarrow \text{Treat the wine with bentonite} \]

**Determination of the bentonite dose necessary to stabilisation**

a. Double and triple the instability value (\( \Delta \text{NTU} \)).
b. Test 2 to 3 doses of bentonite in order to frame these values.
c. 30 minutes after the bentonite incorporation (small volume), renew the stability test assessment.

**Bentonite preparation in the lab:**

*Use the same bentonite as the one used in the winery!!*

- Pick the bentonite dose that reaches \( \Delta \text{NTU} (\text{turb2} - \text{turb1}) < 2 \)
\[ \Rightarrow \text{Add 10 g/hL (100ppm) to this bentonite dose value for the cellar treatment.} \]
C. How do I choose the appropriate treatment?

Following the heat test results: if $\Delta NTU > 2$

⇒ Treat the wine with bentonite

Bentonite preparation and treatment in the cellar:

Use the same bentonite in the cellar as the one used in the lab!!

- Prepare a 5 % solution in water.
- Keep stirring for 2 hours, let it swell for 12 to 24 hours, stir vigorously before use.
- Incorporate the bentonite into the wine to be treated with a venturi.
- To accelerate sedimentation, add silica gel and gelatin 24 hours later.
- Check the treatment efficacy.

See the bentonite preparation
D. What are the parameters influencing stability?

1. The maturity level influences the protein concentration
   - The concentration in thermo-unstable proteins increases during ripening
   - The higher the pH, the more bentonite required to stabilize the wine

![Protein charge according to pH](image)

- Charge
  - Positive
  - Negative
- pH
- pI = 4
D. What are the parameters influencing stability?

1. The maturity level influences the protein concentration
   ✓ The concentration in thermo-unstable proteins increases during ripening
   ✓ The higher the pH, the more bentonite required to stabilize the wine
D. What are the parameters influencing stability?

*pH effect on bentonite action:*

Bentonite type, wine *pH*, but also bentonite preparation influence protein stabilization!
D. What are the parameters influencing stability?

2. Pre-fermentation operations impact stabilization

✓ In the case of a ripe Sauvignon or Semillon, skin contact will double the instability!
✓ SO₂ addition on grapes during maceration enhances protein extraction.
✓ Press juices are more unstable than free run juices.

3. Lees aging improves protein stability

<table>
<thead>
<tr>
<th>Results (ΔNTU)</th>
<th>Wine post fermentation</th>
<th>Wine aged 4 months on lees</th>
<th>Wine aged 10 months on lees</th>
</tr>
</thead>
<tbody>
<tr>
<td>HEAT TEST</td>
<td>45</td>
<td>34</td>
<td>17</td>
</tr>
</tbody>
</table>

Moine-Ledoux, 2006
D. What are the parameters influencing stability?

4. A LYSOZYM® treatment increases wine instability

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>Wine treated with LYSOZYM®</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heat test results (in Δ NTU)</td>
<td>12</td>
<td>57</td>
</tr>
<tr>
<td>Bentonite dose required to stabilise (mg/L)</td>
<td>400</td>
<td>1200</td>
</tr>
</tbody>
</table>
D. What are the parameters influencing stability?

5. Grape varietal, terroir and vintage also have an influence on the protein content of must and wines.

6. A clogging filtration can make the wine unstable.

<table>
<thead>
<tr>
<th>CLOGGING FILTRATION</th>
<th>Beginning of bottling</th>
<th>End of bottling</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heat test results (in Δ NTU)</td>
<td>0.5</td>
<td>4</td>
</tr>
</tbody>
</table>

The clogging filtration made the wine unstable by retaining protective colloids.
E. How do I carry out the treatment in the cellar?

When to treat: on must? on wine?

On must
✓ For early release wine (no barrel aging, no lees aging).
✓ For *Botrytis* affected fruit.

✓ Select a calcium bentonite: more clarifying than stabilizing.
✓ Quickly rack the wine after fermentation.
✓ *No lees aging possible.*

On wine
✓ For all other wines.
✓ Select a sodium bentonite: high protein removal power, respectful of the wine aromas.

In all cases, the bentonite treatment effectiveness is directly linked to *its preparation conditions.*
The Laffort bentonite range

MICROCOL® ALPHA
Natural sodium bentonite, microgranulated, with high protein removal power. For stabilization and clarification of must and wine over a large pH spectrum.

MICROCOL® CL
Natural calcium bentonite, powder. For clarification and stabilization of must and wine.

MICROCOL® FT
Calcium sodium bentonite. For stabilization during cross flow filtration.
Bentonite specificities

Wine pH effect on adsorption power

Bentonite dose (g/hL)
Bentonite specificities

Clarifying power of different bentonites

% of lees

- Microcol
- Microcol alpha
- Microcol-CL

20 g/hL
40 g/hL
60 g/hL
80 g/hL
Bentonite specificities

Effect on aromatic compounds

3MH (ng/L)

- Microcol
- MicrocolALPHA
- Microcol-CL
- Bentonite Z

Concentrations: 0, 50 g/hL, 100 g/hL, 150 g/hL
Bentonite specificities

Effect on rose color protection

Optical Density

- Microcol
- Microcol alpha
- Microcol-CL

- 0
- 25 g/hL
- 50 g/hL
- 100 g/hL
Stabilisation roadmap: change in mindset

4 to 6 weeks prior to bottling

- Microbiological stabilisation
- Protein stabilisation
- Color stabilization

1 to 2 weeks prior to bottling

- Tartaric stabilisation

Bottling

Filterability index monitoring: turbidity < 5 and CI < 20

Enzyme, fining agent, lysozyme, SO2, chitosan...

Finishing tannins

Bentonite

Metatartaric acid
CMC
Mannoproteins

Gum Arabic (D-2)
SO2 & ascorbic acid (D-1)
Sorbic acid (D-1)

NTU < 5  Cl < 20
T° > 15°C (60°F)
Checking Free SO2, CO2, etc., Complete analysis and tasting
**Risks linked to absence of stability**

<table>
<thead>
<tr>
<th>COLORING MATTER PRECIPITATION</th>
<th>Part of the colouring material in red wines is in a colloidal state; this fraction can potentially precipitate.</th>
</tr>
</thead>
<tbody>
<tr>
<td>TYPES OF WINE CONCERNED</td>
<td><strong>Red wines:</strong> <em>red coloured aggregate often associated with crystals</em></td>
</tr>
<tr>
<td>PHENOMENON FAVORED BY:</td>
<td>- Bottling filtration quality: wine stable before bottling becoming unstable through <em>retention of protective colloids in the case of a clogging filtration</em>&lt;br&gt;- Tartaric instability&lt;br&gt;- Wine storage conditions: <em>exposure to cold</em></td>
</tr>
</tbody>
</table>
A. Is my wine stable in terms of coloring matter?

COLD TEST:

Stability is estimated by measuring the turbidity before and after cold storage in the following conditions:

✓ Filter 30 mL of wine on a 0.65 µm membrane (+ prefilter).
✓ Measure the turbidity of the sample: $\text{NTU}_{\text{before cold}}$.
✓ Place the sample at $4^\circ\text{C for 48 hours.}$
✓ Take out of the cold and, after 15 min at room temperature, measure the turbidity $\text{NTU}_{\text{after cold}}$.

$\triangle\text{ NTU} = \text{NTU after cold} - \text{NTU before cold}$
A. Is my wine stable in terms of coloring matter?

Cold test:
Stability is estimated by measuring the turbidity before and after cold storage.

<table>
<thead>
<tr>
<th>Δ turb (NTU)</th>
<th>&lt; 5 NTU</th>
<th>Stable</th>
</tr>
</thead>
<tbody>
<tr>
<td>Δ turb (NTU)</td>
<td>5-10 NTU</td>
<td>Very slight instability</td>
</tr>
<tr>
<td>Δ turb (NTU)</td>
<td>10-20 NTU</td>
<td>Slight instability</td>
</tr>
<tr>
<td>Δ turb (NTU)</td>
<td>20-50 NTU</td>
<td>Usual Medium instability</td>
</tr>
<tr>
<td>Δ turb (NTU)</td>
<td>&gt; 50 NTU</td>
<td>Strong instability</td>
</tr>
</tbody>
</table>
Reduction of fining dosage mainly driven by organoleptic objective

Early bottling and cellar thermoregulation preventing stabilization of the colouring matter in a natural way.

New winemaking practices (thermovinification and flash pasteurisation) which extract more unstable compounds.

Illustration of the rise of the colouring matter instability
Which macromolecules (fractions) from *Acacia senegal gum* are responsible of stability properties?

Which molecular mechanisms play a role in stabilisation reaction?

- Identification of coloring matter precipitate
  - *Project MATCOL*
  - Institute of Chemistry and Biology of Membranes and Nano-objects
  - Chemical composition of coloring matter precipitation.
  - Identify differences among wines from different varieties.
  - Influence of temperature (+4 and -4 °C) on coloring matter formation

- Stabilization mechanism by Gum Arabic
  - *Project VINARABIC*
  - INRA SPO Montpellier
  - Which macromolecules (fractions) from *Acacia senegal gum* are responsible of stability properties?
  - Which molecular mechanisms play a role in stabilisation reaction?
Phenolic compounds by themselves do not precipitate but they are sensitive to be adsorbed by a colloidal support (polysaccharide or protein).

Glories Y., 1979

Cold storage of a limpid red wine (filtered or centrifuged) could produce a red amorphous precipitate after two days. Glories Y., 1979

Colouring matter could be eliminated by dialysis however it reassembles consequently, more or less fast, according to wine conservation.

Feuillets Œnologiques, 1979

2016 new scientific insights:

Identification of colouring matter precipitate

Project MATCOL
Institute of Chemistry and Biology of Membranes and Nano-objects

- Chemical composition of coloring matter precipitation: 81% of the precipitate has been identified
- Identify differences among wines from different varieties.
- Influence of temperature (+4 and -4 °C) on colouring matter formation

> Analyse by Nuclear Magnetic Resonance (NMR)
Identification of compounds in coloring matter precipitate

Analysis of coloring matter precipitate obtained after 2 days at 4°C by solid $^{13}$C NMR

Barrel aging does not modify the involved compounds in the color matter precipitate.

Precipitates profiles from Cabernet Sauvignon and Merlot show the same family compounds.

R&D PROJECT: MATCOL
Shipra Prakash, Axelle Grelard & Erick Dufourc (CBMN)
Main goals of this project:

- Identification 81% of precipitate.
- Identification of minerals, mainly potassium, calcium and iron.

Polyphenol fraction (procyanidins and anthocyanins) is in higher amount in the coloring matter precipitate from Cabernet Sauvignon than in Merlot.

R&D PROJECT: MATCOL
Shipra Prakash, Axelle Grelard & Erick Dufourc (CBMN)
Gum Arabic and properties

- Natural exudate from trees Acacia senegal & Acacia seyal
- Functional properties: interfacial, stability agent, surface agent.
- In wine, Acacia senegal gum is responsible of colouring matter stabilisation and Acacia seyal gum of organoleptic quality enhancement.
- Complex hetero-polyoside, charged and hyper branched (AGP family).
- Macromolecules continuum
  - Molecular masse, hydrophobicity (protein concentration) and charge.
- PROJECT VINARABIC : Acacia senegal gum

R&D PROJECT VINARABIC
Michaël Nigen, Thierry Doco & Christian Sanchez (INRA SPO Montpellier)
B. What are my options in terms of treatment?

Fining and cold treatment lead to coloring matter precipitation.

- Different effectiveness on stabilization depending of fining agents and wine.
- It reassembles in colloidal state during aging.

Stabilization of coloring matter.

- Limited effectiveness over time.

Stabilization mechanism by Arabic gum

Project VINARABIC
INRA SPO Montpellier

- Which macromolecules (fractions) from Senegal gum are responsible of stability properties?
- Which molecular mechanisms play a role in stabilization reaction?
B. What are my options in terms of treatment?

- **Fining**
  - Fining and cold treatment lead to coloring matter precipitation.
  - Different effectiveness on stabilization depending on fining agents and wine.
  - It reassembles in colloidal state during aging.

- **Cold treatment**

- **Arabic gum**
  - Stabilization of coloring matter.
  - Limited effectiveness over time.

- **Mannoproteins**
  - Stabilization of coloring matter.
B. What are my options in terms of treatment?

Observations since 2009: general improvement of red wine stability treated with mannoproteins \(\rightarrow\) combined effect on tartaric and coloring matter instability

- **St Estèphe 2007**
  - Treated 15 g/hL
  - Not treated

- **St Julien 2007**
  - Treated 15 g/hL
  - Not treated
B. What are my options in terms of treatment?

- Methodology for Gum Arabic efficacy evaluation according to the Oenological Codex applied to mannoprotein products

Stabilization Index

Efficacy Test reference for gum Arabic accordingly to OIV COEI-1-GOMARA:2000
Methodology for Gum Arabic efficacy evaluation according to the Oenological Codex applied to mannoprotein products

B. What are my options in terms of treatment?

Stabilisation of a mineral solution with mannoprotein products at 7.5 g/hL, 15 g/hL and 30 g/hL.

Quantification of the stabilizing power (value 9 is equivalent to the stabilizing power of a Verek or Senegal gum)
Stabilisation roadmap: change in mindset

4 to 6 weeks prior to bottling

Microbiological stabilisation

1 to 2 weeks prior to bottling

Protein stabilisation

Bottling

Or

Colouring matter stabilisation

Tartaric stabilization

Filterability index monitoring: turbidity < 5 and CI < 20

Enzyme, fining agent, lysozyme, SO2, chitosan...

Finishing tannins

Bentonite

Metatartaric acid
CMC
Mannoproteins

Gum Arabic (D-2)
SO2 & ascorbic acid (D-1)
Sorbic acid (D-1)

Acidification/deacidification

NTU < 5  CI< 20
T° > 15°C (60°F)
Checking Free SO2, CO2, etc., Complete analysis and tasting
Tartrate Stabilization
### Risks linked to absence of stability

<table>
<thead>
<tr>
<th>TARTRATE PRECIPITATIONS</th>
<th>At a specific temperature, tartaric acid salts become super-saturated: their concentration is higher than the quantity theoretically soluble. Under cooler conditions this state leads to the formation of crystals.</th>
</tr>
</thead>
<tbody>
<tr>
<td>TYPES OF WINE CONCERNED</td>
<td>All types of wines: crystal formation</td>
</tr>
</tbody>
</table>
| PRECIPITATION FAVORED BY | - Bottling filtration quality: wine stable before bottling becoming unstable by *retention of protective colloids due to clogging.*  
- Coloring matter instability (blending with younger vintages).  
- De-acidification treatments before bottling.  
- Wine storage conditions: *wine exposure to cold.* |

### ANALYSES OF WINES SAMPLED ON SUPERMARKET SHELVES:

<table>
<thead>
<tr>
<th></th>
<th>In 2010</th>
<th>In 2016</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Red</strong></td>
<td>56%</td>
<td>72%</td>
</tr>
<tr>
<td><strong>White</strong></td>
<td>72%</td>
<td>41%</td>
</tr>
<tr>
<td><strong>Rosé</strong></td>
<td>90%</td>
<td>60%</td>
</tr>
</tbody>
</table>

In 2010

Red 56% / white 72% / rosé 90%

In 2016

Red 72% / white 41% / rosé 60%
Tartaric acid:

✓ Not very commonly found in nature.
✓ Acid the most important in grapes, must and wines.

\[
\begin{align*}
&\text{COOH} \\
&\text{OH} \quad \text{H} \\
&\text{H} \quad \text{OH} \\
&\text{COOH}
\end{align*}
\]

\[
pK_{a1} = 2.97 \\
pK_{a2} = 4.05
\]

<table>
<thead>
<tr>
<th>Concentration of tartaric acid</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Verjus</strong></td>
</tr>
<tr>
<td>Must from cooler areas</td>
</tr>
<tr>
<td>15 g/L</td>
</tr>
<tr>
<td>&gt; 6 g/L</td>
</tr>
<tr>
<td><strong>Must from warmer areas</strong></td>
</tr>
<tr>
<td>2 to 3 g/L</td>
</tr>
</tbody>
</table>
Tartaric acid and its salts

At wine pH and given the presence of potassium and calcium cations, tartaric acid finds itself mainly in a salt state under 5 forms:

- Potassium hydrogen tartrate (KHT) (referred to in below table as Potassium bitartrate)
- Neutral potassium tartrate (K2T)
- Neutral calcium tartrate (CaT)
- Potassium and calcium tartrate double salt
- Mixed salt potassium and calcium tartromalate

<table>
<thead>
<tr>
<th>Solubility of some tartaric acid salts</th>
<th>In water at 20°C / 68°F</th>
<th>In 10% alcohol at 20°C / 68°F</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tartaric acid</td>
<td>4.9 g/L</td>
<td>-</td>
</tr>
<tr>
<td>Potassium bitartrate KHT</td>
<td>5.7 g/L</td>
<td>2.9 g/L</td>
</tr>
<tr>
<td>Neutral calcium tartrate CaT</td>
<td>0.53 g/L</td>
<td>-</td>
</tr>
</tbody>
</table>
Tartrate precipitation and pH modification

Free forms of tartaric acid, bitartrate and tartrate percentage, depending on pH:

H₂T + K <-> KHT <-> K₂T

Remarkable pH (Négre, 1953):
- 3.59 for wines with 12% v/v alcohol
- 3.53 for wines with 10% v/v alcohol

pH 3.6: KHT precipitation fast and abundant
pH < 3.6: KHT precipitation lowers pH
pH > 3.6: KHT precipitation increases pH
Potassium bitartrate states in wine
Tartaric salt supersaturation range

Protection effect of some molecules towards tartaric acid salts crystallisation
(after Maujean et al., 1985)

<table>
<thead>
<tr>
<th>Super-saturation window</th>
<th>Saturation Temp</th>
<th>Supersaturation Temp</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>20.80</td>
<td></td>
</tr>
<tr>
<td>Bentonite (30 g/hL)</td>
<td>18.20</td>
<td></td>
</tr>
<tr>
<td>Decolorizing carbon (30 g/hL)</td>
<td>19.75</td>
<td></td>
</tr>
<tr>
<td>Gum arabic (10 g/hL)</td>
<td>20.60</td>
<td></td>
</tr>
<tr>
<td>Metatartaric acid (6.6g/hL)</td>
<td>&gt;23.00</td>
<td></td>
</tr>
<tr>
<td>Membrane filtration 10000 Da</td>
<td>14.05</td>
<td></td>
</tr>
</tbody>
</table>
A. What are my options in terms of treatment?

**Subtractive methods**: physical techniques

Removing constituents responsible for precipitation

- **Cold treatment** (*with or without seeding)*:
  Preventive KHT precipitation

- **Electrodialysis**: selective removal of K+ ions

**Inhibition methods**

Inhibition of KHT crystal nucleation and/or growth phase

**Additives:**

- ✓ Sodium Carboxymethylcellulose (CMC): **CELSTAB®**

**Ingredient naturally occurring in wine:**

- ✓ Yeast mannoproteins- **MANNOSTAB®**
B. Is my wine stable from a tartaric standpoint?

Review of tartaric stability tests

1. Reference test: Crystallization test

✓ Filter 250 mL of wine on a 0.65µm membrane.
✓ Place the wine at -4°C / 25°F for 6 days.
✓ Visual reading after 6 days:
  - **White wine:**
    • Absence of crystals (stable wine)
    • Presence of crystals: perform chemical identification tests
  - **Red wine:**
    • Filter the wine and look for the presence of crystals and/or coloring matter

✓ The crystallization test models the natural phenomenon of tartaric precipitations in bottle (takes into account the matrix and the presence of protective colloids). Instability factors: temperature, filtration.
B. Is my wine stable from a tartaric standpoint?

Review of tartaric stability tests

2. Saturation temperature (Tsat)

*Temperature at which KHT can be dissolved in wine*
- White or rosé wine: stable if \( T_{sat} < 15^\circ C \)
- Red wine: stable if \( T_{sat} < 21^\circ C \)

✓ Measures the wine state and its potential tartaric stability level
✓ Not appropriate to validate a treatment with crystallization inhibition techniques

---

Crystallization inhibition treatments do not increase KHT solubility.
B. Is my wine stable from a tartaric standpoint?

Quantify the tartaric instability: *conductivity test*

3. DIT measure: Degree of Tartaric Instability (DIT%) equivalent to a mini-contact test at 4hrs, -4°C / 25°F, + 4 g/L KHT

Definition of a de-ionisation rate to ensure stability (0 to 30%)
(Measurement device STABILAB® - Eurodia/INRA Patent)
B. Is my wine stable from a tartaric standpoint?

Review of tartaric stability tests

3. DIT (Degree of Tartaric Instability): STABILAB® Eurodia/Inra Patent

Measure of the conductivity drop to infinity, after addition of cream of tartar in excess

Test -4°C/ 25°F, 4g/L cream of tartar, 4 hours

✓ Measures the wine state and its potential tartaric stability level
✓ **Detects the presence of crystallization inhibitors**
✓ **BUT cannot validate** an inhibition treatment, especially if instability is high.

![Graph showing DIT values for different treatments](image)

**Stability threshold:**

DIT < 5%
B. Is my wine stable from a tartaric standpoint?  
*Review of tartaric stability tests*

4. **ISTC50 (Critical Tartaric Stability Index):** *STABILAB®* Eurodia/Inra Patent

Measurement of the conductivity variation on a wine considered stable:
- Dissolve 0.5g/L ultra-purified cream of tartar at 37°C/99°F in the wine
- Measure conductivity at -4°C/25°F,
- Then, measure conductivity over 4 hours
- (2 critical stabilization factors: saturation of cream of tartar and temperature)

- Validates a crystallization inhibition treatment for white and rosé wines
- Equivalent to crystallization test 6 days / -4°C (white wine)

![Graph showing ISTC (µS) for different treatments](image)

**Stability threshold:** ISTC50 ≤ 3µS in white and rosé

In red wines: the crystallization is the appropriate test due to the possible interaction between coloring matter stability and tartaric stability.
C. How do I choose the appropriate treatment?

Tartaric instability level & type of wine

**Potential tartaric stability state: DIT (%)**

*Stability threshold (white, rosé, red wine): < 5 %*  
(under our lab measurement conditions)

<table>
<thead>
<tr>
<th>DIT Value (%)</th>
<th>&gt; 20</th>
<th>&gt; 20</th>
<th>&lt; 20</th>
</tr>
</thead>
<tbody>
<tr>
<td>Category of wine</td>
<td>Basic / premium Early consumption</td>
<td>Basic / premium Early consumption</td>
<td>Super Premium – ageing wines (6 months minimum)</td>
</tr>
<tr>
<td>Recommended treatment</td>
<td>POLYTARTRYL®</td>
<td>CELSTAB®</td>
<td>MANNOSTAB®</td>
</tr>
<tr>
<td>Maximum legal dosage (g/hL)</td>
<td>10</td>
<td>10</td>
<td>-</td>
</tr>
<tr>
<td>Treatment dosage (g/hL)</td>
<td>10</td>
<td>10</td>
<td>10 - 30</td>
</tr>
<tr>
<td>White wines</td>
<td>Direct treatment</td>
<td>Direct treatment</td>
<td>Natural stabilisation of white, rosé and red wines</td>
</tr>
<tr>
<td>Red &amp; rosés wines</td>
<td>Direct treatment</td>
<td>Risk of interaction with colouring matter: haze and/or crystal formation</td>
<td></td>
</tr>
</tbody>
</table>
### D. How do I check the efficacy of the treatment?

**Did the elected treatment stabilize my wine?**

<table>
<thead>
<tr>
<th>DIT Value (%)</th>
<th>&gt; 20</th>
<th>&gt; 20</th>
<th>&lt; 20</th>
</tr>
</thead>
<tbody>
<tr>
<td>Category of wines</td>
<td>Basic / premium Early consumption</td>
<td>Basic / premium Early consumption</td>
<td>Super Premium – ageing wines</td>
</tr>
<tr>
<td>Treatment to validate</td>
<td><strong>POLYTARTRYL</strong>®</td>
<td><strong>CELSTAB</strong>®</td>
<td><strong>MANNOSTAB</strong>®</td>
</tr>
<tr>
<td>White wines</td>
<td>ISTC50 / CHECKSTAB</td>
<td>ISTC50 / CHECKSTAB</td>
<td>ISTC50 / CHECKSTAB</td>
</tr>
<tr>
<td>Rose wines</td>
<td>ISTC50 / CHECKSTAB</td>
<td>Crystallisation test*</td>
<td>ISTC50 / CHECKSTAB</td>
</tr>
<tr>
<td>Red wines</td>
<td>Crystallisation test*</td>
<td>Not recommended</td>
<td>Crystallisation test*</td>
</tr>
</tbody>
</table>

**CHECKSTAB validation**: we have observed an overestimation of the mannoprotein dose necessary for the stabilization

*Tartaric stability is linked to colouring matter stability. The crystallisation test is the only test taking into account the potential interaction between both stabilities.*
E. What are the parameters influencing tartaric stability?

In the case of non temperature controlled cellars, the winter cold leads to spontaneous tartaric precipitations reducing therefore the instability potential of aging wines.

Evolution of the tartaric instability potential during aging

- Natural precipitation in the cold
- DIT = 15-20%
- DIT = 10-15%
E. What are the parameters influencing tartaric stability?

Ageing on lees improves tartaric stability

<table>
<thead>
<tr>
<th>Graves white wine</th>
<th>Before treatment</th>
<th>After cold storage</th>
<th>After 10 months on lees</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tsat</td>
<td>21</td>
<td>16</td>
<td>18</td>
</tr>
<tr>
<td>Crystallization (visually)</td>
<td>**</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>mini contact</td>
<td>120</td>
<td>30</td>
<td>20</td>
</tr>
<tr>
<td>Tcs</td>
<td>-6</td>
<td>-18</td>
<td>-24</td>
</tr>
</tbody>
</table>

After Moine-Ledoux, 1996
E. What are the parameters influencing tartaric stability?

**Coloring material stability (red wines)**

If the wine shows a very high coloring matter instability (young wines, wine blended with younger vintages, inappropriate fining), there is a risk of tartaric precipitation, caused by the precipitation of colloidal coloring matter.

<table>
<thead>
<tr>
<th>Wine</th>
<th>2008 fined wine</th>
<th>2008 wine + 10 g/hL MICROCOL® ALPHA bentonite</th>
</tr>
</thead>
<tbody>
<tr>
<td>DIT %</td>
<td>19.1 %</td>
<td>9.7%</td>
</tr>
<tr>
<td>MANNOSTAB® Dose</td>
<td>No stabilization</td>
<td>15 g/hL</td>
</tr>
</tbody>
</table>
Metatartaric acid: POLYTARTRYL®

- Metatartaric acid has been *authorised since 1956*.
- At this time, the LAFFORT® company has developed its production in collaboration with Emile Peynaud.
- This tartaric acid polyester is the strongest inhibitor of potassium bitartrate.
- It hydrolyses over time; the higher the temperature, the faster the hydrolysis.

![Diagram showing DIT % comparison between untreated wine and treated wines with Polytartryl®, Celstab®, and Mannostab®.](image-url)
E466 Cellulose gums (CMC): CELSTAB®

Coupled with Na⁺ ion: “Sodium CMC”

Characterised by:

- **DS** = substitution degree (carboxymethyl groups)  ➔ **EFFICACY**
- **DP** = polymerisation degree (glucose units)  ➔ **VISCOSITY**

pKa = 4
E466 CMC HPLC profile
One single peak confirms the product’s purity
CMC + GA HPLC profile
2 peaks = 2 products

P1 > 48 Kda
P2 = 39 Kda
Benchmark – CMC market product HPLC profiles

Pur product profile

Mix CMC + GA profile

Product with a possible hydrolysis or blend of products

HPLC peaks molecular weights

- > 48 kda
- 39.1 Kda
- 38.4 Kda
F. How do I perform the treatment in the cellar?

**CELSTAB**

**Celstab® Treatment:**
Before final bottling filtration on a wine fined and clarified
(Clogging Index < 20, Turbidity < 5 NTU)

**Dosage:** 1 mL/L (CELSTAB® is a 10% solution)

**Implementation:**
Dilute the solution in twice its volume of wine.

- **Still wines:** incorporation using a dosage pump or an Oenodoseur 48 hours before bottling.
- **Sparkling wines:** incorporation at tirage

**Enological conditions:**
- Use CMC on protein stable wines.
- CMC forms a haze on wines treated with LYSOZYM®.
- CMC forms a haze with tannins.
Impact of the addition of tannins on a wine treated with bentonite regarding protein and tartaric stabilities

**Protein Stability**

**Tartaric Stability**

 ISTC50 (µS)

Addition of CMC = 1 mL/L
Impact of the addition of CMC or Mannoproteins on wines treated with tannins during tartaric stabilization

Late ageing of the wine in new barrels or late addition of finishing tannins

Potential Tartaric Instability

Colloidal instability

PROTEIN precipitation

No protein precipitation observed & The efficiency of the treatment with mannoproteins is not impacted
Impact of CMC implementation on clogging index and coloring matter
# Stabilization

**Wine 1:**
Mesterrieux Coop (rosé)

**Wine 2:**
Lafon (rosé)

**Wine 3:**
Sauveterre Coop (rosé)

## CELSTAB®: Results on rose wines

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>CELSTAB® 10 cL/hL</th>
<th>CELSTAB® 10 cL/hL STABIVIN® 2.5 cL/hL</th>
<th>CELSTAB® 10 cL/hL STABIVIN® 5 cL/hL</th>
<th>CELSTAB® 10 cL/hL STABIVIN® 10 cL/hL</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Turbidity</strong></td>
<td>0.3</td>
<td>0.3</td>
<td>0.3</td>
<td>0.3</td>
<td>0.3</td>
</tr>
<tr>
<td><strong>Clogging index</strong></td>
<td>7</td>
<td>7</td>
<td>8</td>
<td>8</td>
<td>8</td>
</tr>
<tr>
<td><strong>DIT</strong></td>
<td>18.50%</td>
<td>3.70%</td>
<td>3.70%</td>
<td>3.30%</td>
<td>2.90%</td>
</tr>
<tr>
<td><strong>Crystallisation</strong></td>
<td>+ (mc -)</td>
<td>- (mc-)</td>
<td>- (mc-)</td>
<td>- (mc-)</td>
<td>- (mc-)</td>
</tr>
<tr>
<td><strong>Protein stability</strong></td>
<td>Made stable (45g/hl)</td>
<td>Stable</td>
<td>Stable</td>
<td>Stable</td>
<td>Stable</td>
</tr>
<tr>
<td><strong>DO280</strong></td>
<td>10.62</td>
<td>10.56</td>
<td>10.42</td>
<td>10.64</td>
<td>10.54</td>
</tr>
<tr>
<td><strong>ICM</strong></td>
<td>0.667</td>
<td>0.760</td>
<td>0.764</td>
<td>0.734</td>
<td>0.804</td>
</tr>
</tbody>
</table>

|                  | 1.6     | 1.6               | 1.6                                  | 5.7                                 | 5.7                                  |
| **Clogging index** | 27      | 27                | 32                                   | Clogging                            | Clogging                            |
| **DIT**          | 21.80%  | 3.10%             | 2.60%                                | 1.40%                               | 0.80%                                |
| **Crystallisation** | + (mc -) | - (mc-)           | - (mc-)                              | - (haze)                            | - (haze)                            |
| **Protein stability** | Stable            | Stable            | Stable                               | Stable                              | Stable                               |
| **DO280**        | 13.42   | 13.04             | 12.74                                | 13.44                               | 13.00                                |
| **ICM**          | 0.604   | 0.622             | 0.606                                | 0.795                               | 0.775                                |

|                  | 0.2     | 0.5               | 0.5                                  | 0.6                                 | 0.7                                  |
| **Clogging index** | 7       | 7                 | 7                                    | 7                                   | 7                                    |
| **DIT**          | 20.40%  | 5.30%             | 5.20%                                | 4.20%                               | 4.10%                                |
| **Crystallisation** | + (mc -) | - (haze)          | - (haze)                            | - (haze)                             | - (haze)                            |
| **Protein stability** | Stable            | Stable            | Stable                               | Stable                              | Stable                               |
| **DO280**        | 13.62   | 13.08             | 13.02                                | 13.02                               | 13.00                                |
| **ICM**          | 0.628   | 0.746             | 0.728                                | 0.729                               | 0.721                                |
CELSTAB®

Results on rosé wines

Identification of haze:

Crystals of a very particular shape, brown colour.

*Presence of K, Ca and tannins.*
CELSTAB® on rosé wines

On 15 rosé wines from different origins:
✓ 7 can be stabilized with CMC (46.6%).
✓ 8 cannot be stabilized with CMC (53.3%).
✓ The wines not advised for CMC treatment show a high polyphenol index (IPT) or a weak clogging index.
✓ After treatment they all show a good DIT or ISTC50, but they all have the same haze with the crystallisation test (6 days at -4°C).
✓ The addition of stabilizing arabic gum has no effect on this haze.
✓ No increase of protein instability is noticed.
✓ When the clogging index is good, it remains good after addition of CELSTAB® and STABIVIN®.

Results of these trials confirm that a feasibility test in the lab is necessary on rosé wine before a CELSTAB® treatment.
Trials on red wines

Coloring matter interaction risk

Crystallization test results (6 days at -4°C):

*Bordeaux Sup Rouge 2008 + 10g/hL CMC A, B and C*

Reading on pre-filters after 6 days:

- Absence of crystals: stable wine from KHT point of view
- But: significant sediment of coloring matter – reaction under cold conditions

Reading room temperature controls on pre-filter
Wine treated with 10 g/hL CMC
Absence of coloring matter sediment

Reading on pre-filter / test 6d -4°C
wine treated with 10g/hL CMC
Highlights the reaction CMC – colouring matter in the cold (-4°C)
Cold temperatures catalyse haze formation “CMC – coloring matter”

Trials beforehand are necessary to evaluate the risk of interaction with coloring matter, prior to any CMC treatment on red and rose wines.
F. How do I perform the treatment in the cellar?

MANNOSTAB® TREATMENT: Before final bottling filtration on a wine fined and clarified (Clogging Index < 20, Turbidity < 5 NTU)

Dosage: 100 – 300 mg/L according to a test, or according to the aging period

Implementation: (check out the video!)
✓ Dissolve MANNOSTAB® in 10 times its weight in warm water (10% solution)
✓ Let it rest a few minutes and add during a racking or a pump-over
✓ Homogenize with a pump-over for at least 1.5 times the tank volume

Enological conditions:
✓ Treatment of a wine aged for a minimum of 6 months
✓ Any clogging filtration can lead to a loss of colloids and/or MANNOSTAB® and therefore renders the treatment partially or completely ineffective
✓ Homogenise with a pump-over for at least 1.5 times the volume of the tank
✓ Avoid any thermal shock > 5°C in the 72h following bottling
MANNOSTAB® treatment in red wines

Observations since 2009: general improvement of red wine stability → combined effect on tartaric and coloring matter instability
Neutral calcium tartrate is a low solubility salt, 10 times less than KHT. In case of super-saturation (high level in Ca + high pH), there is a risk of CaT precipitation.

**TYPES OF WINE CONCERNED**

All types of wine: crystal formation

**PRECIPITATION FAVORED BY:**

- *Wine calcium content*
- Bottling filtration quality: wine stable prior to bottling becoming unstable through the *retention of protective colloids in the case of a clogging filtration.*
Calcium Stabilisation

**White, rosé and red wines: risk if \([\text{Ca}] > 60 \text{ mg/L}\)**

We recommend testing juice as early as possible, during fermentation if necessary.

Wine de-acidification with calcium carbonate can elevate calcium levels above 60 mg/L ppm, test treated wines.

**Elevated calcium levels can cause calcium tartrate precipitation, and inhibition methods are efficient in preventing only KHT precipitation.**

**Treatment options:**
- **Cold:** although the CaT solubility is little sensitive to temperature, it seems part of CaT may be precipitated when the wine is cooled down?
- **CMC?**
- **Treatment with calcium racemate:** not easy to implement; addition at the juice phase is preferential
Stabilisation roadmap: change in mindset

4 to 6 weeks prior to bottling

- Microbiological stabilisation
- Protein stabilisation
- Colouring matter stabilisation

1 to 2 weeks prior to bottling

- Filterability index monitoring: turbidity < 5 and CI < 20

Bottling

- NTU < 5 CI < 20
  - $T^\circ > 15\,^\circC$ ($60\,^\circF$)
- Checking Free SO$_2$, CO$_2$, etc., Complete analysis and tasting

Ingredients:
- Enzyme, fining agent, lysozyme, SO$_2$, chitosan...
- Finishing tannins
- Bentonite
- Metatartaric acid
- CMC
- Mannoproteins
- Gum Arabic (D-2)
- SO$_2$ & ascorbic acid (D-1)
- Sorbic acid (D-1)

Acidification/deacidification
Filtration & Filterability
Effect of colloids on filterability

If wines are well prepared ($CI < 50$): no effect of colloid addition
Below 15°C / 60°F, wines is more susceptible to clogging!
Effect of membrane nature on filterability

Millipore: cellulose acetate / nitrate

Sartorius: cellulose acetate

Pall: Nylon

CI very different depending on membranes!
Preparing the wines for filtration

Wine filterability is therefore paramount, depending (amongst others) on:

- temperature
- membrane on which the clogging index is tested,

There is no correlation between filterability and wine turbidity; a clear wine can clog filters! It is essential to assess both parameters when preparing the wine for filtration.
☑️ POSITIVE FACTORS TO IMPROVE FILTERABILITY

- **ENZYME Addition**
  - action on filterability
  - Ensures pectin and/or glucan chains breakdown, to improve settling (racking)
    - PECTINASES
    - β. GLUCANASES

- **FINING**
  - decreases the load
  - Ensures settling of particles in suspension (colloids) present in the wine

- **RACKING**
  - decreases the load
  - Lees removal

- **DEGASSING**
  - Reduction of the CO2 load ensures minimal degradation of the cake during DE filtration.

- **APPROPRIATE ADDITION OF ENOLOGICAL PRODUCTS**
  - Every enological product has its own solubilization properties. A solubilization in water OR in wine prior to the final addition to the wine will facilitate a better filterability of treated wines
### Autolees preparation and its impact on filterability

Turbidity and clogging index of a red wine treated directly with AUTOLEES at 150 and 300 mg/L are much more important than those of the red wine treated with a prior dissolution in water and this even after 72h of contact.

<table>
<thead>
<tr>
<th></th>
<th>Control (red wine)</th>
<th>15 g/hl</th>
<th>30 g/hl</th>
<th>15 g/hl</th>
<th>30 g/hl</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Turbidity 1h</strong></td>
<td></td>
<td>0,8</td>
<td>1,8</td>
<td>3</td>
<td>6,9</td>
</tr>
<tr>
<td><strong>Clogging Index (PALL) 1h</strong></td>
<td>42</td>
<td>53</td>
<td>66</td>
<td>93</td>
<td>227</td>
</tr>
<tr>
<td><strong>Turbidity 72h</strong></td>
<td></td>
<td>0,8</td>
<td>1,3</td>
<td>2</td>
<td>4,6</td>
</tr>
<tr>
<td><strong>Clogging Index (PALL) 72h</strong></td>
<td>42</td>
<td>51</td>
<td>66</td>
<td>95</td>
<td>189</td>
</tr>
</tbody>
</table>

Autolees must first be dissolved in **WATER** (at 10%) before its addition to the wine for a better filterability of treated wines.
Tannins preparation and its impact on filterability

Tannins must be dissolved directly in **WINE**, and not in water, for a better filterability of treated wines. They can be prepared from 1% up to 10%.

The turbidity of tannins or wine solutions is not correlated to the filterability of the treated wine.

<table>
<thead>
<tr>
<th>Solubilisation in water (30 min)</th>
<th>Solubilisation in wine (30 min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>wine + 10 g/hl tannin</td>
</tr>
<tr>
<td>Turbidity</td>
<td>0,4</td>
</tr>
<tr>
<td>Clogging Index 1h</td>
<td>32</td>
</tr>
</tbody>
</table>
NEGATIVE FACTORS FOR FILTERABILITY

- **TEMPERATURE**
  The lower the temperature, the poorer the filterability (viscosity increases). Greater propensity for oxygen to dissolve.

  $1^\circ C = 2\%$ flow rate

- **CHARGE**

  Evaluate wine filterability by controlling the clogging index (minimum one week before bottling).
Cross-flow filtration / pad or DE filtration

Representation of filtered volumes kinetics in cross-flow (tangential) filtration and frontal filtration

Flow rate vs. Functioning time

- Tangential filtration
- Frontal filtration
Cross-flow filtration and enological treatments

Enological products compatible with cross-flow filtration:

**Organic fining agents:**
Gelatine, isinglass, casein, albumin

**Vegetal fining agents:**
Pea and potato proteins (VEGECOLL®) – *rinse with cold water!*

**Mineral fining agents:**
Bentonite (MICROCOL® FT)
Silica gel and Carbon – *PROHIBITED, too abrasive*

**Synthetic fining agents:**
PVPP – do the treatment 7 days prior to cross-flow
Cross-flow filtration and enological treatments

Oenological products compatible with cross-flow filtration: (i.e. Bucher Vaslin recommendations)

**Tartaric stabilisation:**
Mannoproteins, CMC (5 days ahead)

**Other products:**
Tannins, Concentrated must, SO\(_2\) – prior to filtration
Gum arabic, N\(_2\)/CO\(_2\) – post filtration

Note: each cross-flow supplier may have different recommendations
Cross-flow filtration and enological treatments

In line additions compatible with Cross-Flow Filtration (Bucher Vaslin):

- Rectified concentrated must
- Specific bentonite (Microcol FT)
- Vegecoll
- SO₂ solution
- N₂/CO₂ (to adjust CO₂ levels)
BOTTLING TIMELINE

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l'œnologie par nature
Our recommendations at bottling:

- **SO₂**: 24 hours prior to bottling
- **Ascorbic acid**: 24 hours prior to bottling
- **Sorbic acid**: 24 hours prior to bottling
  *(with a SO₂ addition)*
- **Gum Arabic**: 48 hours prior to bottling
- **Cellulose gum**: 48 hours prior to bottling
- **Mannoproteins**: 48 hours prior to bottling

*The addition of these different products must be combined with an effective homogenisation to avoid changes in quality at bottling.*
Last additions prior to bottling

Incompatibilities:

- CMC is incompatible with Lysozyme.
WHITE WINE CHECK-LIST

We recommend completing the final blend prior to initiating any stabilisation process.

#1 MICROBIOLOGICAL STABILITY
- Microbial load determination
  - Complete assessment (Yeast, Acetic Bacteria, Lactic Bacteria)
- Treatment
  - Treatment options to reduce the microbial load:
    - SO₂
    - Enzyme addition
    - LYSOZYME
    - Fining
    - Physical treatments

#2 PROTEIN STABILITY
- Protein instability determination
  - Heat test 30 min. at 80°C (Refer to the detailed protocol)
  - If ∆NTU < 2: stable wine, if ∆NTU > 2: unstable wine
- Treatment
  - Bentonite dose determination
  - Bentonite treatment: same bentonite in lab and in cellar
  - Importance of product preparation and implementation

#3 TARTARIC STABILITY
- Tartaric instability determination
  - Crystallisation test 6 days at -4°C
  - DIT Test 4 hrs at -4°C
    - +4 g/L of cream of tartar
    - If no crystals or DIT < 5% = stable wine
    - If presence of crystals or DIT > 5% = unstable wine
- Treatment
  - LAFORT® expert's advice
    - We recommend removing Carbon Dioxide in the wine prior to any tartaric stabilisation treatment
    - Physical processes
    - Inhibitory methods
    - POLYTRAETYL®
    - CELSTAB®
    - MANNOSTAB®
  - Fillerability index monitoring: turbidity < 5 and CI < 20
  - Importance of mixing the tank
  - Double check stabilisation
    - ISTC50 measure:
      - (test 4hrs at -4°C + 0.5g/L cream of tartar)
      - ISTC50 = 3µS = stable wine

#4 BOTTLING
- Complete wine analysis and tasting (D+4)

THE KEYS TO A SUCCESSFUL BOTTLING
- 1 stable batch + 1 stable batch = 1 stable blend.
- Use CMC on protein stable wines.
- CMC and metatartaric acid form a haze on wines treated with lysozyme.
- CMC forms a haze with tannins.
- Metatartaric acid used on a cold wine creates a reversible haze.

DIT: Degree of Tartaric Instability
CI: Clogging Index

LAFORT
l'oenologie par nature

The quality of the filtration is essential to prevent the retention of protective colloids which might cause a new instability in the wine. Below 15°C / 60°F, wine flow decreases / - 1°C = 2% flow rate loss.
**RED WINE CHECK-LIST**

**Microbiological Stability**
- Microbial load determination
- Complete assessment (Yeast, Acetic Bacteria, Lactic Bacteria)
- Treatment options to reduce the microbial load: $SO_2$, addition, Enzyme addition, LYSOZYME
  - Fining
  - OENOBRETT®
  - Physical treatments

**Colouring Matter Stability**
- Instability determination
  - Cold test ($4^\circ C$ for 48h)
<table>
<thead>
<tr>
<th>Aturb (NTU)</th>
<th>Instability</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 5 NTU</td>
<td>Stable</td>
</tr>
<tr>
<td>5-10 NTU</td>
<td>Slight Instability</td>
</tr>
<tr>
<td>10-20 NTU</td>
<td>Medium Instability</td>
</tr>
<tr>
<td>20-50 NTU</td>
<td>Typical Instability</td>
</tr>
<tr>
<td>&gt; 50 NTU</td>
<td>Strong Instability</td>
</tr>
</tbody>
</table>

**Tartaric Stability**
- Instability determination
- Crystallisation test 6 days at $-4^\circ C$
  - If no crystals or $DIT < 5\% = stable wine$
  - If presence of crystals or $DIT > 5\% = unstable wine$
- DIT Measure test 4hrs at $-4^\circ C$ + 4g/L cream of tartar

**Fining Trials**
- At least 2 fining agents at 2 different doses

**Treatment**
- Gelatin
- Fining: OENOLEES® VEDECOL®
- Albumin

- Physical processes
- Inhibitory methods: POLYTRARYL® MANNOSTAB®

**Bottling**
- Complete wine analysis and tasting ($D+4$)

**The Keys to a Successful Bottling**
- 1 stable batch + 1 stable batch + 1 stable blend.
- Metatartaric acid may form a haze in wines treated with lysozyme.
- Metatartaric acid used on a cold wine creates a reversible haze.
- The use of arabic gum will improve the colouring matter stability.

**LAFORT® expert’s advice**
- BENTONITE: a 5 to 10g/L dose following the addition of the proteic fining agent will enhance the coloring matter stability and optimise the use of the fining agent
- ENZYME (EXTRAZYSE®) used in the end of vinification or ageing will favor the stabilisation of colouring matter.

- Filterability index monitoring: turbidity < 5 and $CI < 20$

- Double check stabilisation => crystallisation test

---

**Important of Homogenisation**
- $15^\circ C / 60^\circ F$, wine flow decreases $\times -1^\circ C = 2\%$ flow rate loss
Preparing your wines for your next bottling?

Let’s talk...

Thank you for your attention!