

**REDUCING  
SULPHITE CONTENT**



Bioprotection,  
Vinification, Storage

---



# ROLES AND PREVALENCE OF SO<sub>2</sub> IN OENOLOGY



**Oxidised SO<sub>2</sub>**  
= sulphate  
(SO<sub>4</sub><sup>2-</sup>)



## ANTIMICROBIAL EFFECT

- ✓ Bacteria
- ✓ Yeasts

## ORGANOLEPTIC EFFECTS

- ✗ Hardness, dryness
- ✗ Odour of SO<sub>2</sub> masks the fruitiness
- ✓ Blocks ethanal and other aldehydes (reduces the musty smell)

• Oxygen  
• Quinones...

**Active SO<sub>2</sub>**  
(H<sub>2</sub>SO<sub>3</sub>)



## ANTIOXIDANT EFFECT

- ✓ Oxygen
- ✓ Quinones

• Low pH  
• High temp.  
• High ethanol

**SO<sub>2</sub> in salt form**  
(HSO<sub>3</sub><sup>-</sup>)

**Free SO<sub>2</sub>**

• Aldehydes  
• Ketones  
• Sugars

**Total SO<sub>2</sub>**

**Bound SO<sub>2</sub>**

## METABOLIC EFFECTS

- ✗ Yeast : SO<sub>2</sub> / ethanal / H<sub>2</sub>S production  
*Depending on temperature, turbidity, nutrition.*
- ✗ Human: toxicity, allergenicity

## PREVENTS ENZYMATIC OXIDATION

- ✓ Tyrosinase
- ✓ Laccase

## THE DIFFERENT FORMS OF SO<sub>2</sub> AND THEIR IMPORTANCE IN OENOLOGY

Sulphur dioxide has been used as a disinfectant since the days of Homer (900 BCE), and has been used in winemaking since 1487. Sulphur dioxide has since become the wine additive of choice due to its vital benefits. Its “active” molecular form (H<sub>2</sub>SO<sub>3</sub>) has antimicrobial properties and its bactericidal and fungicidal properties play a role in the microbiological stabilisation of wine. In its free form (molecular sulphur dioxide + bisulphite (HSO<sub>3</sub><sup>-</sup>)), sulphur dioxide exhibits both an antioxidant effect<sup>1</sup> – by indirectly neutralising the dissolved oxygen or quinones to form sulphates – and prevents enzymatic oxidation<sup>2</sup> by inhibiting the enzymes that cause oxidation (complete inhibition of tryptophanase originating in the grape, and partial inhibition of laccase produced by *Botrytis cinerea*). Finally, sulphur dioxide can help to neutralise any musty odours by combining with ethanal.

However, SO<sub>2</sub> has been under the spotlight in recent years due to its numerous drawbacks:

- it is poisonous to humans, and therefore poses a risk both to the consumer and the handlers in the cellar.
- it can lead to an unpleasant sulphurous, “rotten egg” smell caused by a reduced sulphur (H<sub>2</sub>S) produced during fermentation<sup>3</sup>; in addition, the H<sub>2</sub>S can be oxidised into a sulphate, which is often responsible for the sensation of dry mouth. Another potentially undesirable effect of SO<sub>2</sub> is increased ethanal formation in the yeast<sup>4</sup>.
- it has a noticeable smell and/or can mask some of the more positive aromas of the wine<sup>5</sup>.
- when combined with anthocyanins (pigments in red and rosé wines), it can cause partial, but reversible, decolouration of these pigments.

The significance of these drawbacks has meant that a lot of research has been conducted in this area, with the aim of reducing the use of SO<sub>2</sub> in oenology and finding alternatives for both its antimicrobial and antioxidant properties.

One of the major risks involved when attempting to reduce sulphite levels in the vinification of whites or rosés is oxidation. This is a key component in producing these types of wine, whereas the microbiological risks often take precedent for reds; it is this latter problem which the alternatives seem to be able to manage well.

In all cases, **the pH of the must and the wine** is the key parameter that guides many decisions. At a low pH (close to 3.0), SO<sub>2</sub> is more balanced in its molecular, active form and the risk of microbial, oxidative, and enzymatic oxidative changes is reduced. At a high pH (higher than 3.5), wines are more sensitive to attacks from contaminants (e.g. *Brettanomyces* or certain harmful bacteria) and to oxidation. Managing the acidity of the wine is therefore a priority control mechanism, and musts should be treated differently according to their pH.

Similarly, **maturity** levels, **polyphenol** content in the grape varieties, the **duration** of pre-fermentation operations, the **temperature** at each step in the process and the quality of **transfers** are other key control mechanisms, which should be adapted to determine an alternative procedure.

The Institut Œnologique de Champagne has been investing in this research for several years. Drawing on our partnerships with research institutes and suppliers, IOC is now in a position to offer a vast, although not exhaustive, range of methods or tools that can be used as alternatives to sulphur dioxide. The aim is not necessarily to completely eliminate its use, rather to significantly reduce the use and content of sulphur dioxide in wines.

Of course, usage should be adapted to the individual raw materials, type of vinification, risk level, product objective and technical and economic constraints. Our oenologists will be delighted to assist you with the creation of a personalised procedure.

<sup>1</sup> Ribereau-Gayon, 1933 ; Dubernet, 1973 ; Vivas, 1999

<sup>2</sup> Kovac, 1979

<sup>3</sup> Henschke et Jiranek, 1991

<sup>4</sup> Cleroux et al, 2015

<sup>5</sup> Peynaud et Blouin, 1991

# STEP BY STEP: THE RISKS A SULPHITE ADDITION AND TH

STEP	MICROBIOLOGICAL RISK	OXIDATIVE RISK	RISK OF INCREASING SULPHITE LEVELS
Transport from grape harvest to the winery	<b>STRONG</b> depending on time, temperature, hygienic conditions, condition of the grape berry, pH etc.	<b>POSSIBLE</b> depending on the condition of the grape berry	<b>LOW</b>
Pre-fermentation cold maceration	<b>STRONG</b> depending on time, temperature, hygienic conditions, condition of the grape berry, pH etc.	<b>POSSIBLE</b> depending on the condition of the grape berry	<b>LOW</b>
Pellicular maceration (generally not advised with sulphite-free vinification)	<b>HIGH</b> depending on time, temperature, hygienic conditions, pH etc.	<b>HIGH</b> more significant polyphenol extraction	<b>LOW</b>
Pressing	<b>MEDIUM</b>	<b>HIGH</b> depending on the type of pressing and the raw material	<b>LOW</b>
Maceration of the sludge	<b>HIGH</b> depending on time, temperature, hygienic conditions, pH etc.	<b>MEDIUM</b> depending on inerting, polyphenols, time, temperature	<b>LOW</b>
Settling	<b>HIGH</b> depending on time, temperature, hygienic conditions, pH etc.	<b>MEDIUM</b> depending on inerting, polyphenols, time, temperature	<b>LOW</b>
Alcoholic fermentation	<b>HIGH</b>	<b>LOW</b>	<b>HIGH</b>
Malolactic fermentation	<b>HIGH</b>	<b>HIGH</b> (if delay before triggering MLF)	<b>LOW</b>
Ageing	<b>HIGH</b>	<b>HIGH</b>	<b>LOW</b>
Each time the wine is moved	<b>LOW</b>	<b>HIGH</b>	<b>LOW</b>
Bottling then storage	<b>MEDIUM</b>	<b>HIGH</b>	<b>LOW</b>

# ASSOCIATED WITH REDUCING E POTENTIAL ALTERNATIVES

RISK OF COMBINING SO <sub>2</sub>	MICROBIOLOGICAL CONTROL SOLUTION	OXIDATION CONTROL SOLUTION
LOW	<ul style="list-style-type: none"> <li>MICROBIOLOGICAL BIOPROTECTION with GAÏA™</li> </ul>	<ul style="list-style-type: none"> <li>GLUTAROM EXTRA (reducing power)</li> <li>+ potentially ascorbic acid.</li> </ul>
LOW	<ul style="list-style-type: none"> <li>Conduct at low temperature and with GAÏA™.</li> <li>Enzyme addition with EXTRAZYME MPF to catalyse changes.</li> </ul>	<ul style="list-style-type: none"> <li>GLUTAROM EXTRA (reducing power)</li> <li>+ potentially ascorbic acid.</li> </ul>
LOW	<ul style="list-style-type: none"> <li>Enzyme addition with EXTRAZYME TERROIR for selective extraction and to begin the breakdown of pectin.</li> </ul>	<ul style="list-style-type: none"> <li>GLUTAROM EXTRA (reducing power)</li> <li>+ potentially ascorbic acid</li> <li>For certain product types: controlled oxygenation.</li> </ul>
LOW	<ul style="list-style-type: none"> <li>Conduct at low temperature and with GAÏA™.</li> <li>Enzyme addition with EXTRAZYME MPF to catalyse changes.</li> </ul>	
LOW	<ul style="list-style-type: none"> <li>FLOTATION with Qi'Up and INOZYME TERROIR recommended.</li> <li>For static settling: GAÏA™ and INOZYME TERROIR.</li> </ul>	<ul style="list-style-type: none"> <li>Flotation settling recommended for deoxygenation: QI'UP</li> <li>Bonding of polyphenols and oxidation catalysts: QI NOOX (non-animal, non-allergenic antiradical).</li> </ul>
HIGH	<ul style="list-style-type: none"> <li>Yeast as soon as possible with a <i>S. cerevisiae</i> flora that either does not produce SO<sub>2</sub> or ethanal, or produces them in small quantities (IOC BE yeasts).</li> </ul>	<ul style="list-style-type: none"> <li>GLUTAROM EXTRA after yeasting to enrich the wine with glutathione and to increase future resistance.</li> <li>Organic nutrition with thiamine (ACTIVIT O) to restrict the formation of compounds that combine with SO<sub>2</sub>.</li> </ul>
MEDIUM	<ul style="list-style-type: none"> <li>Co-inoculation of selected wine bacteria recommended to restrict bacterial contamination and to respect the varietal characteristics of the wines.</li> </ul>	<ul style="list-style-type: none"> <li>Co-inoculation or early inoculation at 2/3 of AF with MAXIFLORE SATINE or INOFLORE to remove the ethanal.</li> </ul>
HIGH (oxidation of ethanol into ethanal)	<ul style="list-style-type: none"> <li>pH management (acidification) where necessary.</li> </ul>	<ul style="list-style-type: none"> <li>Ageing on selected lees to protect the wines against oxidation.</li> <li>Tannins adapted to restructure the wines if the polyphenols have been oxidised.</li> </ul>
MEDIUM	<ul style="list-style-type: none"> <li>Hygiene, pumps, pipes, vats etc.</li> </ul>	<ul style="list-style-type: none"> <li>DESOXYGENATION.</li> </ul>
HIGH	<ul style="list-style-type: none"> <li>Appropriate filtration.</li> </ul>	<ul style="list-style-type: none"> <li>Choice of stoppers</li> <li>Care with filtration and bottling in order to minimise oxygen addition.</li> <li>Ascorbic acid: in certain circumstances only</li> <li>It may still be necessary to add SO<sub>2</sub>.</li> </ul>

# ALTERNATIVE PROCEDURE FOR REDUCING SULPHITE ADDITION AND SO<sub>2</sub> CONTENT

## LOW SO<sub>2</sub> SOLUTIONS : INNOVATIVE AND TAILOR-MADE TOOLS FOR REDUCING SULPHITE CONTENT

### PRE-FERMENTATION PHASES

- Grape transport
- Pre-fermentation maceration
- Settling

Microbiological risk

**BIOPROTECTION WITH GAÏA™**



### ALCOHOLIC FERMENTATION

Avoid production / combining of SO<sub>2</sub>

Optimise antioxidant content in wines

**LEVURES IOC BE**



**IOC BE**



**GLUTAROM EXTRA**



LOW SO<sub>2</sub> SOLUTIONS AND COMPLEMENTARY TECHNIQUES: TOWARDS REDUCING SULPHITE CONTENT IN WINES



Conscious of the fact that reducing the concentration of SO<sub>2</sub> in wines cannot be achieved through traditional procedures alone, IOC has developed the Low SO<sub>2</sub> traditional range: a set of complementary products and techniques that have been specially designed for this purpose.

The decision whether to use these tools or not is made following a detailed study of the existing procedure and its constraints and risks (microbiological and oxidation), whilst keeping the objective of reducing sulphite levels at the forefront of the process.

## GRAPE HARVEST AND PRE-FERMENTATION STEPS USING LIFE TO CONTROL LIFE: GAÏA™

From harvesting to the vat or press, the microorganisms responsible for producing acetic acid (such as *Kloeckera apiculata*) could multiply at breakneck speed. This risk is amplified once pre-fermentation maceration begins, especially if the temperature is relatively low or if the maceration duration is particularly long.

The French Wine and Vine Institute selected Gaïa™, a *Metschnikowia fructicola* yeast with no fermentation ability

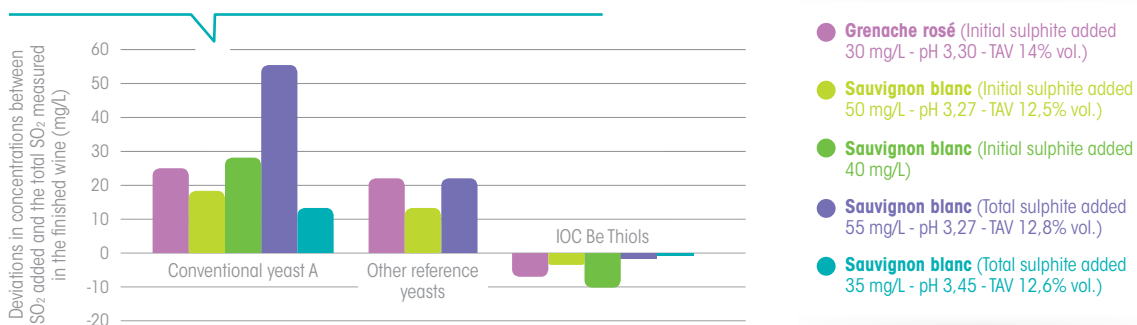
to counteract this harmful flora. It can therefore occupy the ecological niche by reducing both deviations and the risk of starting alcoholic fermentation too early. In this respect, it is quite natural that Gaïa™ is a significant tool for reducing pre-fermentation sulphite addition, both when used in the vatting process and in earlier phases (harvest bins). It also facilitates the implantation of the *S. cerevisiae* that have been selected and inoculated to drive fermentation.

## ALCOHOLIC FERMENTATION - AVOIDING PRODUCTION OF SO<sub>2</sub> AND ITS COMPOUNDS: IOC BE YEASTS

Conventional yeasts, especially certain wild yeasts, are likely to produce varying quantities, and often extremely large quantities (from 40 mg/L to upwards of 100 mg/L) of SO<sub>2</sub>. These yeasts generally produce significant amounts of ethanal, the most powerful element in combining sulphites. This ability to produce acetaldehyde depends on temperature, turbidity and the nutrition applied to the must.

However, some rare yeasts do not have this ability to produce SO<sub>2</sub>, regardless of the fermentation environment. Recent innovative selection methods have made the procurement of these yeasts possible for oenology, the fruit of which is IOC BE range. The first of these yeasts, IOC BE THIOLS and IOC BE FRUITS, transcend the conventional range of low SO<sub>2</sub> /ethanal producing yeasts on offer (IOC TwICE, IOC R 9008, IOC PrimRouge etc.).

Production SO<sub>2</sub> - Deviations between SO<sub>2</sub> added and total SO<sub>2</sub> measured



## MUST AND ALCOHOLIC FERMENTATION – ANTICIPATING ENRICHING THE WINE WITH GLUTATHIONE: GLUTAROM EXTRA

Glutathione (GSH) is a tripeptide that indirectly exhibits a strong antioxidant effect. It reacts with quinones to prevent their agglomeration (responsible for browning musts and for oxidised wines) and the oxidation of aromatic compounds. Although GSH is found naturally in grapes, its concentration is too weak to effectively protect the wine.

GLUTAROM EXTRA is the result of the latest selection and production techniques of inactivated yeasts with very high levels of GSH. If it is added at the start of the fermentation process, it is possible to obtain a wine with a higher

concentration of GSH at the end of the process, provided that appropriate amounts of organic nitrogen are added to the yeast.

In cases of low sulphite levels, the positive impact created by this richness in GSH is distinct on the aroma of the wine, including in reds.

It has also been demonstrated that adding an inactivated yeast that is rich in GSH could be more effective for the aromatic content than by adding pure glutathione. This is probably due to synergies with the other yeast compounds.

# MORE INFORMATION

You can find more information in the articles published by IOC available on our website: [www.ioc.eu.com](http://www.ioc.eu.com)

## Stratégies de limitation des sulfites dans les vins - Quelles alternatives? Partie 1/3 : L'axe microbiologique, bioprotection et étapes préfermentaires

Olivier Pillet\*, François Davaux\*, Philippe Gabillet\*, Stéphane Peyrot\*, Anthony Silvano\*, Bertrand Robillot\*  
 \* Institut Oenologique de Champagne - France  
 \* Institut Français de la Vigne et du Vin - Lons-le-Saunier - France  
 \* Chambre d'Agriculture Inter-Communes - Champagne-Cote des Bars - France  
 \* Equale - Lyon - France  
 \* Lafont SAS - Blagnac - France

**Introduction**  
 Le dioxyde de soufre est un auxiliaire technologique devenu incontournable dans les vins, en raison de ses nombreux bénéfices. Cependant, son utilisation est soumise à des réglementations strictes, ce qui pousse à la recherche d'alternatives. Cette partie explore l'axe microbiologique, la bioprotection et les étapes préfermentaires.

**Le SO<sub>2</sub> est cependant pointé du doigt depuis quelques années, en raison de ses nombreux inconvénients :**  
 - toxique pour l'organisme humain, il présente un danger pour le consommateur de vin en cas de manipulation en cave ;  
 - précurseur possible d'allergènes sulfite réactifs et de réactions allergiques lors des fermentations (Renaud et al., 2019) ;  
 - son odeur peut être envahissante, il peut également nuire à la formation de pigments importants pour la couleur du vin (Phylinaud et Robillot, 2017) ;  
 - en se combinant aux anthracènes, pigments des vins rouges et noirs, il provoque leur décoloration partielle, voire réversible. Pour ces raisons, de nombreuses recherches visent à réduire son utilisation en oenologie et à lui trouver des alternatives. Dans cette première partie, nous nous attachons à évaluer quelques

### Levures fractionnées de *S. cerevisiae* en macération préfermentaire de raisins noirs

Un premier stratégie pour limiter ces risques est d'inoculer au plus tôt la levure *S. cerevisiae* choisie pour réaliser la FA. Cependant, en situation - fréquente - de macération préfermentaire à une température supérieure à 18°C, des risques trop précoces en fermentation peuvent survenir en cas d'un levantage à peine dose (20-30 mg/l) à l'aveugle, nuant au travail d'oxidation en phase espérée renforcé par le vieillissement dans ce type de processus. Une alternative possible



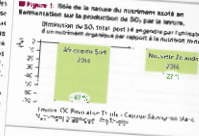
## Stratégies de limitation des sulfites dans les vins - Quelles alternatives? Partie 2/3 : L'axe microbiologique - les fermentations

Olivier Pillet\*, Jean-Marie Marie-Monier\*, Bertrand Robillot\*  
 \* Institut Oenologique de Champagne - France  
 \* Chamber d'Agriculture Inter-Communes - Champagne-Cote des Bars - France

**Introduction**  
 Lors de la fermentation alcoolique, le dioxyde de soufre est consommé par les levures. Cette consommation est influencée par plusieurs facteurs, notamment la température et la concentration en sucre. Cette partie explore les stratégies de limitation des sulfites pendant les fermentations.

**Le SO<sub>2</sub> est consommé par les levures, ce qui pousse à la recherche d'alternatives.** Cette consommation est influencée par plusieurs facteurs, notamment la température et la concentration en sucre. Cette partie explore les stratégies de limitation des sulfites pendant les fermentations.

**Le SO<sub>2</sub> est consommé par les levures, ce qui pousse à la recherche d'alternatives.** Cette consommation est influencée par plusieurs facteurs, notamment la température et la concentration en sucre. Cette partie explore les stratégies de limitation des sulfites pendant les fermentations.



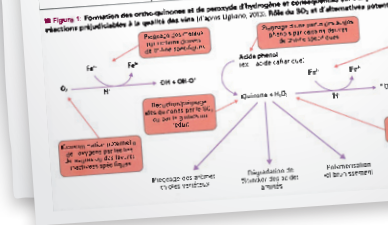
**Facteurs aggravaants**  
 La production de SO<sub>2</sub> est influencée par plusieurs facteurs, notamment la température et la concentration en sucre. Cette partie explore les facteurs aggravaants de la production de SO<sub>2</sub>.

## Stratégies de limitation des sulfites dans les vins - Quelles alternatives? Partie 3/3 : L'axe antioxydant - Anticipation et conservation

Olivier Pillet\*, François Davaux\*, Bertrand Robillot\*  
 \* Institut Oenologique de Champagne - France  
 \* Institut Français de la Vigne et du Vin - Lons-le-Saunier - France

**Introduction**  
 L'axe antioxydant est crucial pour la conservation des vins, en particulier en ce qui concerne la limitation des sulfites. Cette partie explore les stratégies de limitation des sulfites dans les vins, en particulier l'axe antioxydant.

**Le SO<sub>2</sub> est consommé par les levures, ce qui pousse à la recherche d'alternatives.** Cette consommation est influencée par plusieurs facteurs, notamment la température et la concentration en sucre. Cette partie explore les stratégies de limitation des sulfites pendant les fermentations.

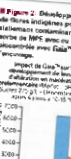


## Limitation des tenueurs en sulfites par le contrôle de la flore microbienne et de son comportement avant et pendant fermentation alcoolique

Olivier Pillet\*  
 \* Institut Oenologique de Champagne - France

**Introduction**  
 Le contrôle de la flore microbienne est essentiel pour la limitation des tenueurs en sulfites pendant la fermentation alcoolique. Cette partie explore les stratégies de limitation des sulfites par le contrôle de la flore microbienne.

**Le SO<sub>2</sub> est consommé par les levures, ce qui pousse à la recherche d'alternatives.** Cette consommation est influencée par plusieurs facteurs, notamment la température et la concentration en sucre. Cette partie explore les stratégies de limitation des sulfites pendant les fermentations.



**Conclusion**  
 La limitation des tenueurs en sulfites par le contrôle de la flore microbienne est une stratégie efficace pour réduire l'utilisation de sulfites dans les vins. Cette partie explore les conclusions de cette étude.