The Wine EXPERT
Practical Winemaking Information
LALLEMAND

H_2S PRODUCTION BY WINE YEAST DURING ALCOHOLIC FERMENTATION

What is hydrogen sulfide?

Hydrogen sulfide, also known as H_2S (Figure 1), is a powerful volatile compound. Its odor is associated with rotten egg and/or sewage-like. It is produced in different concentrations by wine yeasts depending on a variety of factors both environmental and genetics.

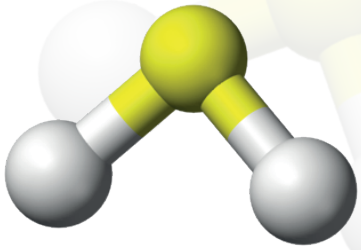

Figure 1: Molecular structure of hydrogen sulfide (H_2S)

Why do I want to avoid this compound in my wine?

Hydrogen sulfide (H_2S) is an important contributor to the so-called 'reductive' off-flavor present in some wines, with an odor threshold of only 0.9-1.1 µg/L for white wine and 1.6 µg/L for red wine. Even at a level that can't be detected by a human nose, H_2S can impact the wine aroma negatively, masking the fruit, and giving a 'closed' sensation. Moreover, the chemical reactivity of H_2S can lead to the formation of other negative compounds such as sulphides and mercaptans especially during aging.

How is it produced?

Research has shown that all yeasts, indigenous or selected produce H_2S and the capacity of the yeast to do so is genetically determined. The wine yeast will also respond differently to different environmental factors, for example, nutritional status of the must, which can impact H_2S formation. There are various mechanisms through which hydrogen sulphide may be produced by *Saccharamyces cerevisiae*. H_2S may be generated through the degradation of sulphur containing amino acids, the reduction of elemental sulphur, or the reduction of sulphite or sulphate (Karagiannis and Lanaridis 1999,).

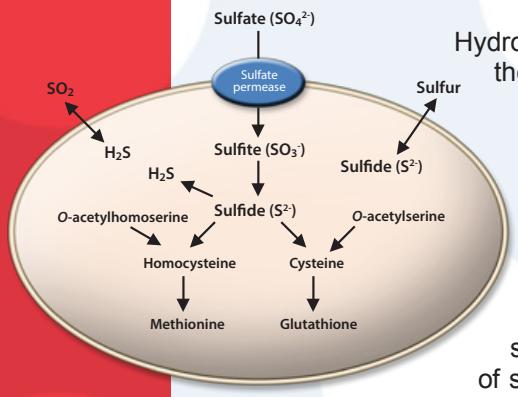

Figure 2: Mechanism of early production of H_2S

Hydrogen sulphide can be formed during primary fermentation, at the beginning when biomass production is at its peak, or late in the final stages of sugar consumption (Spiropoulos *et al.* 2000). Early formation of H_2S is thought to be due to sulphate reduction and the release of reduced sulphide prior to incorporation into the organic carbon acceptor, O-acetyl –L-homoserine, in the biosynthesis of methionine, cysteine and their derivatives (Figure 2). Inefficiency of incorporation of the reduced sulphur into the precursors of these amino acids coupled with release of the reduced sulphide from the active site of sulphite reductase has been proposed to result in leakage of sulphide from the pathway and the formation of H_2S (Eschenbruch et al. 1978, Jiranek *et al.* 1992, Rankine 1963, Spiropoulos *et al.* 2000). Late formation of sulphide may be due to turnover of S-containing components in the cell, such as methionine, cysteine or glutathione, or may be a consequence of the role of the sulphate reduction pathway in stress tolerance. When other preferred nitrogen sources are depleted, *Saccharomyces* can degrade sulphur-containing amino acids, even its own amino acids to utilize the nitrogen, resulting in the release of H_2S or other volatile sulphur compounds as by-products.

How the level of negative sulfure compunds (especially H_2S) are impacted by the yeast and its environment?

Extensive research has provided evidence that yeasts, therefore genetic background, is an important variable in H_2S production, and that yeasts respond differently to physiological and environmental factors in the production of reduced sulphide (Spiropoulos and Bisson 2000, Spiropoulos *et al*. 2000). Therefore, it is crucial to know the potential of a specific yeast to produce H_2S.

Also, nitrogen deficiency can result in excessive production of hydrogen sulfide associated with reductive off-odors and flavors (Mestres, Busto, & Guasch, 2000; Wang, Bohlscheid, & Edwards, 2003). As a general guideline, grape musts to be fermented are considered nitrogen deficient when concentrations of YAN are below 140–150 mg N/L. Yeast strains utilize YAN with differing efficiencies and are influenced by the forms of nitrogen-containing compounds available (Julien *et al*. 2000).

A good management of yeast nutrition and oxygen addition, but also the presence of micronutrients, is one of the key tools to limit the apparition of negative sulfur compounds as they contribute to reduce the yeast stress and provide enough nitrogen and oxygen to allow a better yeast metabolism.

But despite those preventive key points of a good alcoholic fermentation management are respected, *S. cerevisiae*, whether selected or spontaneous, is susceptible to produce H_2S, if it is a naturally, and genetically a high producer.

THE RESULTS

Some results

Saccharomyces cerevisiae wine yeast, whether selected or spontaneous, will produce H_2S, but as mentioned above, it will depend on both the yeast and the environmental factors. This compound is problematic because of the low thresholds of detection. Not only its rotten egg smell is highly undesirable, its chemical reactivity can lead to the formation of more delete¬rious compounds (sulphides and mercaptans) during further wine aging.

In a study done in our research laboratory and with the work of Park (2004) at UC Davis (USA) showed that wine yeasts produced different levels of H_2S during fermentation and were categorized as low, medium and high producers (Figure 3).

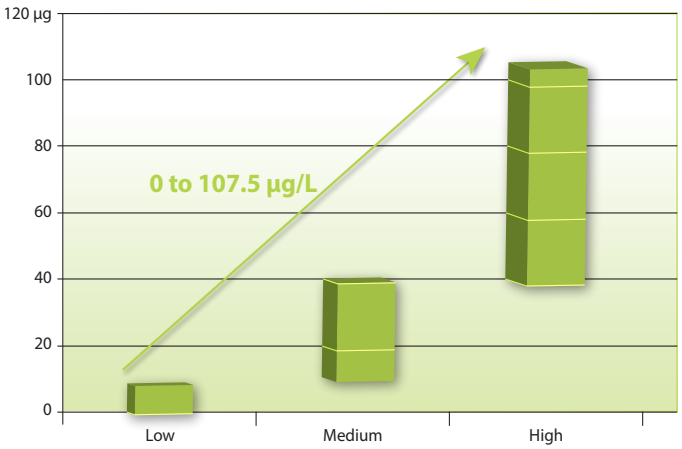

Figure 3: Wine yeast grouping based on their H_2S production (Park, 2004)

When there is a nitrogen deficiency, (low Yeast Assimilable Nitrogen – YAN), H_2S production will increase as the yeast will most likely use its own amino acids (containing sulphur molecule) as a source of nitrogen, and release in the medium (must) the remaining -HS group, that will form sulphur compounds.

The best strategy to avoid such situation is to select a wine yeast that will produce very little H_2S to know if your selected yeast has a high demand for nitrogen during fermentation and to properly manage alcoholic fermentation.

A new wine yeast

In our research program, we studied the possibility to select a yeast which would never produce H_2S in any conditions, especially in white must which are often over-clarified and lacking essential YAN. The yeast Lalvin Sensy™ was selected through natural crossing of yeast to achieve this goal.

Figure 4: Selection of the Lalvin Sensy™ compared to a yeast producing H_2S (control).

THE RESULTS

During several trials over the years, it was shown that the production of H_2S remain close to null in many conditions and the resulting wine shows varietal and desirable fermentative aromas from quality white grapes, and not overshadowed by H_2S rotten egg fault. The SO_2 and acetaldehyde production also remained low with the use of this yeast (figure 5)

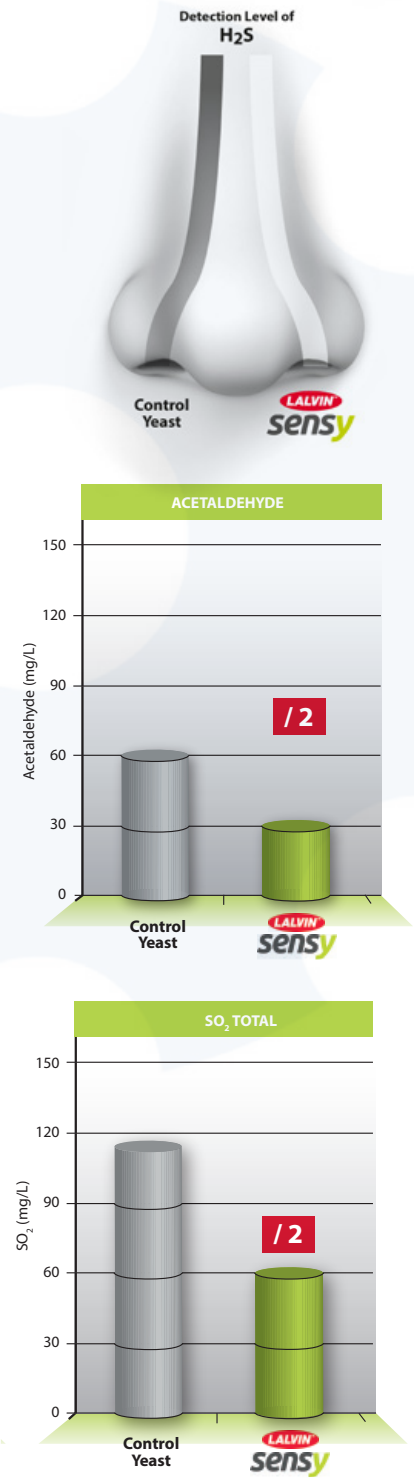

Figure 5: H_2S, SO_2 and acetaldehyde production by Lalvin Sensy™

The low capacity to produce H_2S is a great advantage to fully leave expression of aroma from quality white grape. For example, in a Sauvignon blanc wine (Figure 6), the sensory profile was described as with higher intensity in retro olfaction, more fruit, more tropical fruit less "mercaptan".

However, even if Lalvin Sensy™ is not able to produce H_2S whatever the conditions are, it does not mean that we don't have to pay attention to nutrition and oxygen management. Yeast nutrition doesn't have only an effect on the negative sulfur compounds but on the whole yeast metabolism, from its fermentative performance to its aromas metabolism.

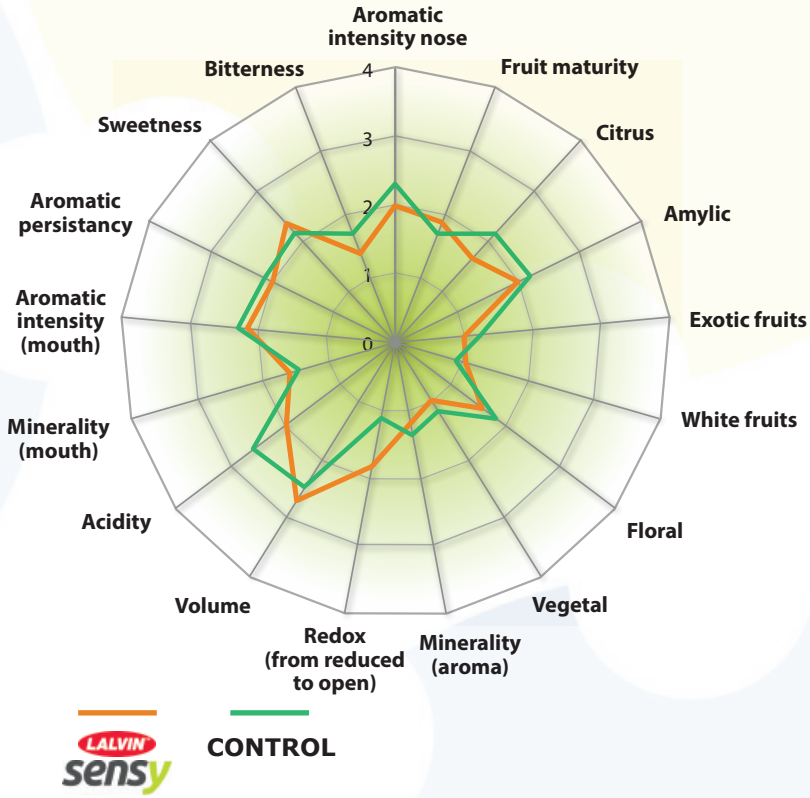

Figure 6: Sensory analysis of Sauvignon blanc (2014 – France) wine fermented with Lalvin Sensy™ compared to a control.

A WORD FROM OUR EXPERT

Sam Harrop MW

Sam Harrop MW, has spent more than 20 years working in the international wine industry, with a diverse career that bridges the gap between the technical and complex world of wine making and the consumer's appreciation of the end product. Sam holds a Bachelor of Commerce and a post-graduate diploma in Oenology. He is also a Master of Wine, having passed on his first attempt in 2002, and the recipient of the Tim Derouet Award for outstanding performance. Sam regularly appears in both the trade and consumer press in the UK and around the world as a key wine industry commentator, and in 2011, he co-authored the book 'Authentic Wine' with Jamie Goode, focusing on concepts of naturalness and sustainability in winemaking.

In 2012 the International Wine and Spirit Competition presented Sam with 'The Julian Brind Memorial Trophy for Outstanding Achievement in the Wine Industry'. In December 2013 The Drinks Business, one of the key trade publications in the UK, selected Sam as the 10th most influential wine consultant in the world

From creating profitable wine ranges for Marks and Spencer (UK), to consulting for small vineyards in Europe; judging and co-chairing the International Wine Challenge in London and establishing the competition's flourishing Sake cate-gory, Sam is one of the most influential wine making consultants in the world.

Because of its distinctive 'rotten egg' character, H_2S in a finished wine indicates a fault.

H_2S readily reacts and combines with other wine components and sulphides to form longer chain mercaptans and disulphides, which have different sensory expressions. These can include cabbage, onion, garlic, struck match, rubber, canned corn and truffles.

For many wine professionals, the presence of some, if not all of these characteristics render a wine faulty, as volatile sulphur compounds tend to dominate the pure, less intense primary, secondary and tertiary aromas, and impact wine quality in a negative way.

Sometimes, in the right context and at balanced very low levels – like seasoning is to cooking – some of these sulphides can enhance wine complexity: Think 'struck match' in Puligny-Montrachet, 'gunflint' in Pouilly Fumé', and 'truffles' in Cornas.

However, trying to achieve the right expression and balance of these 'complexed sulphide' characters in a wine is a risky business. While there are many premium wines using these characters to enhance quality, there are many, many more that can justifiably be considered faulty because the balance is wrong.

It must be noted that the wines displaying the positive and balanced sulphide expression are exclusively in the fine wine segment. In my opinion, there is no place for these earthy aromas in aromatic styles and / or any style in the fighting price points where customers prize fruit above all else.

Throughout my career as a winemaker, retail buyer, wine judge and consultant I have tasted tens of thousands of wines. I am very confident in saying that faults linked to sulphides (derived from H_2S throughout fermentation) are a bigger issue than cork taint, *Brettanyomyces* and, potentially even oxidation. The majority of wine consumers want vibrant, pure, intense fruit expression in a wine and, as such, any winemaker not working to minimize H_2S production during fermentation is doing themselves and their customers a disservice.

A QUICK SUMMARY

Hydrogen sulphide (rotten egg) aroma is a serious fault in wine and will result in quality loss in the final product. Its production is mainly found during alcoholic fermentation. H_2S production by wine yeast varies based on the yeast used as well as on the environmental factors, especially the assimilable nitrogen concentration and micronutriens present. Ranging from low to high producers, this characteristic is important to take into account, as well as the nutritional status of the must, when deciding on a fermentation strategy.

The best way to avoid formation of H_2S during winemaking are to choose a low H_2S yeast producer and apply good fermentation practices, and also use wine yeast such as Lalvin Sensy™ which have been especially selected to avoid the production of this compound while keeping in mind a good nutrition and oxygen addition strategy. It is adapted to white winemaking fermentation where the conditions (such as low NTU, low temperature, low YAN) could affect H_2S formation, and the Lalvin Sensy™ with its low capacity to produce H_2S, as well as SO_2 and acetaldehyde, will let the varietal aromas be fully expressed.