INSTRUCTIONS: IODINE METHOD FOR S02 (SULFUR DIOXIDE)

Sulfur dioxide is added to musts and wines as an antioxidant and antiseptic, often in the form of sulfiting chemicals (such as potassium metabisulfite as liquid S02). Though fermentation without adding S02 at harvest is becoming popular, most grapes are sulfited before fermentation. A very small amount of S02 is made by yeasts, but not enough to protect the wine. If S02 were not added during cellaring, wines would acquire a sherry-like nose and brown color, and would be very susceptible to spoilage.

Test S02 level at least several times while making the wine: after yeast fermentation (or after ML fermentation), before the first summer, before the second winter if not bottling sooner, and when bottling. Wineries often have a regular testing schedule that includes monitoring S02.

Some of the total amount of S02 in a wine exists in combination with acetaldehyde and other wine components. This part is called "bound" as opposed to the portion called "free," which is more loosely bound. Free S02 plus bound S02 equal total S02. The most active part, called "molecular S02," is a very small amount (under 1 ppm) of actual dissolved S02. Molecular S02 is a small percentage of the free S02, and is calculated from pH and the measured free S02.

Since the percentage of free S02 present as molecular S02 depends on pH (a wine with pH 3.0 has 10 times as much molecular S02 at the same free S02 level as does a wine at pH 4.0), we speak in terms of molecular S02 when giving S02 recommendations, not just free S02. (see "interpretation" section).

Winemakers try to maintain a free S02 level sufficient to prevent the wine from spoiling (calculate in terms of molecular S02), without adding so much that the total S02 becomes offensive. Whenever possible, keep total S02 under 100 ppm. Too much molecular S02 is detected as a burning or pungent sensation in the nose and throat; many consumers can also detect high total S02 (over 125-150) ppm as a lingering slipperiness or a chemical taste. Note that the H2S (rotten-egg smell) that spoils the nose of some wines does not come directly from S02, but is made by yeasts under stress.

*MEASURING S02*

The iodine (Ripper) procedure described here uses iodine to combine with the S02 in the sample. A starch solution indicates the endpoint by turning purple when a slight excess of iodine is present. To test free S02, the wine is acidified with sulfuric acid; for total S02, the sample is first treated with strong sodium hydroxide to release bound S02, then acidified with sulfuric and titrated as for free S02.

Though used in many wineries, the iodine method has sources of error. The starch color change is not sharp and fades rapidly, making the endpoint hard to determine. A more serious problem is that other components in red wine besides S02 react with iodine to give a falsely high result (5-20 ppm too high, which may be more than the free S02).

Our basic recommendation is to ADD THE SMALLEST AMOUNT OF SULFITE NECESSARY TO PREVENT DETERIORATION OF WINE QUALITY.

General suggestions:

*At harvest: none (No-S02 method) or 40-60 ppl71 total* S02

Most of the amount added at harvest will be lost during fermentation, but what does remain will be present as bound S02. Adding more than 50 ppm at harvest may result in lower quality wine because high S02 in juice encourages production of acetaldehyde, and can delay growth of ML bacteria in wines inoculated for malolactic fermentation.

A number of premium wine producers are adding no S02 at all before fermentation on reds or whites. This method must be done properly to be successful, but it can result in wine that is fruitier, is less likely to brown, and has less total S02 than wine fermented with S02.

*At first racking or after Ml.fertmentation:* 50 *ppm total* S02

If MLF is to be encouraged, adding S02 after fermentation before MLF is completed can delay or prevent proper growth of the bacteria.

*During cellaring: under pH* 3.4: *keep at 0.5-0.8 ppm molecular* S02 *over pH* 3.4: *adjust according to spoilage tendencies*

In general, delicate wines are kept at higher molecular levels, heavy reds at lower levels. In higher-pH wines, some S02 protection is sacrificed to avoid the chemical taste of too much bound S02. Test free and total S02 (and pH) several times a year; adjust to desired molecular S02 level. The free S02 binds slowly as the wine ages, but total S02 falls little if at all except when lees are still dropping.

*At bottling: under pH* 3.4: *bring to 0.5-0.8 ppm molecular; over pH* 3.4: *adjust according to spoilage tendencies*

Again, whites usually have higher levels than reds. If at all possible, total S02 at bottling is kept under 100 ppm.

*When consumed: 0.4-0.6 ppm, NEVER more than 0.8 ppm molecular*

The normal (non-asthmatic) consumer's reaction to too much S02 is to dislike the wine, though unaware of the reason. At high levels of molecular S02, the taster may cough or feel an irritation in nose and throat. Some people do not find 0.8 ppm molecular S02 or higher to be excessive, but many others do.

Asthmatic winemakers may experiment with their own sensitivities to find the molecular S02 level they can safely tolerate.

Wine aroma and flavor is best when there is enough S02 to bind up the acetaldehyde (so it does not smell cooked, stale, or like sherry) but not so much as to cover the fruitiness or offend the palate.

Standardization procedure:

Use 10 ml of sodium thiosulfate 0.025 N (0.' 5 ml 0.1 N) instead of the wine sample, and proceed as for free S02. Titrate as soon as the sulfuric acid is added, or the solution will turn milky white during the titration (if this happens, throw it out and try again). The purple endpoint color will not fade as it will for a wine sample, so it is OK to go drop by drop near the end. We suggest running the standardization twice if possible.

If the iodine has not changed strength, the amounts required for standardization are:

| Iodine | Na2S203 0.025 N | (10 ml) | (5ml) | Na2S203 0.100 N (5 m)) |
|---|---|---|---|---|
| 0.0156N | ml iodine: | 16.0 ml | 8.0 ml | 32.0 ml |
| 0.0200N | ml iodine: | 12.5 ml | 6.25 ml | 25.0 ml |

If you use less than the above amounts, do not change your calculation factor; the cause is usually a variation in pipeting technique, since the iodine cannot increase in strength. If you use more iodine than listed above, calculate the new normality of iodine:

$$\text{Normality of iodine} = \frac{\text{normality of Na2S203 x volume of Na2S203 (ml)}}{\text{volume of iodine used (ml)}}$$

Use the new normality in your calculations. To figure the new calculation factor:

New Factor = old factor (10 or 12.8) x new normality + old normality

(for example, if you use 10 ml of 0.025 N Na2S203 and the titration took 16.4 ml of iodine, the new normality of the iodine is 0.0152. The new calculation factor would be 10 x 0.0152 + 0.0156 = 9.7)

*INTERPRETATION OF RESULTS*

Calculating an appropriate amount of S02 to add is very important, especially with increased consumer awareness of sulfites. The various forms of S02 in wine are closely related to pH.

The chart on the next page shows that the percentage of free S02 existing as molecular S02 goes down as the pH goes up (see 2nd column), showing *that for the same amount of free S02, wines with higher pH have less protection from molecular S02.* The last two columns show the free S02 levels needed at different pH's to achieve two levels of molecular S02, 0.5 ppm and 0.8 ppm (normal ranges for S02 in many bottled wines).

To calculate the amount to add: *It is impossible to add free S02 or molecular S02 per se.* You can add a sulfiting agent to raise the total S02; some of the amount added remains as free (and a tiny bit is molecular) while some becomes bound, often about half and half, but the exact amount must be tested after a few days.

*When adding S02, even in liquid form it tends not to mix but to stay where it is added in juice, or to layer near the bottom in wine. Always stir well, then again the next day. Test S02 in* 2-3 *days.*

## R E L A T ION S HIP 0 F **pH** AND S 0 2

FREE S02 NEEDED TO ACHIEVE MOLECULAR S02 OF:

| | molecular S02 (% of free S02) | 0.8 PPM | 0.5 PPM |
|---|---|---|---|
| 2.90 | 7.5% | 11 ppm free S02 | 7 ppm free S02 |
| 2.95 | 6.6 | 12 | 7 |
| 3.00 | 6.1 | 13 | 8 |
| 3.05 | 5.3 | 15 | 9 |
| 3.10 | 4.9 | 16 | 10 |
| 3.15 | 4.3 | 19 | 12 |
| 3.20 | 3.9 | 21 | 13 |
| 3.25 | 3.4 | 23 | 15 |
| 3.30 | 3.1 | 26 | 16 |
| 3.35 | 2.7 | 29 | 18 |
| 3.40 | 2.5 | 32 | 20 |
| 3.45 | 2.2 | 37 | 23 |
| 3.50 | 2.0 | 40 | 25 |
| 3.55 | 1.8 | 46 | 29 |
| 3.60 | 1.6 | 50 | 31 |
| 3.65 | 1.4 | 57 | 36 |
| 3.70 | 1.3 | 63 | 39 |
| 3.75 | 1.1 | 72 | 45 |
| 3.80 | 1.0 | 79 | 49 |
| 3.85 | 0.9 | 91 | 57 |
| 3.90 | 0.8 | 99 | 62 |
| 3.95 | 0.7 | 114 | 71 |
| 4.00 | 0.7 | 125 | 78 |

Adapted From: *Enology Briefs 1(#* 1), F eblMar 1982. University of California Cooperative Extension.

*RECOMMENDED S02 LEVELS*

To make specific recommendations for S02 additions or even appropriate levels to maintain, it helps to know about the particular wine and winemaking conditions. Since molecular S02 is the active portion, *we express recommendations as molecular S02, not free or total S02.*

A molecular S02 0[0.8 ppm is an acceptable maximum for some white wines (below pH 3.4); in general, we suggest lower molecular S02 for reds than whites, perhaps around 0.5 ppm at bottling. Wineries that adjust to a standard free S02 level instead of monitoring molecular S02 will have varying S02 impact on their wines, with some wines having too little and others too much. Adjusting to a desired level of molecular S02 provides consistency in S02 management.

Wines with relatively high pH, red (over 3.5) or white (over 3.4), may require too high a level of total S02 to achieve appropriate molecular S02 levels. *Whenever possible, keep total S02 under 100 ppm.* Rather than have excessive bound S02, we rely on a combination of factors, including likelihood of spoilage, to determine an appropriate S02 level. Some pH problems can be relieved by adjusting the pH downward with tartaric acid, if the balance of the wine will permit. *High acid/high pH wines are best adjusted in the vineyard by modifying vine structure, rather than depending on S02 to prevent problems.*

*NOTES:*

- Once you have added the sulfuric acid, the titration must be performed as quickly as possible, or S02 will be lost. Overzealous swirling or shaking of the sample also loses S02; if a sloshing noise is heard, it is too vigorous.

- Up to 50 ml of distilled water may be added to a red sample just before titration with iodine, if the color is very dark. A yellow light nearby can help, but do not shine the light directly through the sample. Another aid is to set up two identical beakers with red wine samples and other chemicals, but titrate only one. Compare the two beakers during titration to see when the titrated beaker darkens.

- The iodine method is not valid for free S02 on red wines, but an estimate can be made by running a "blank" titration. Run the test as described, then run once more, but before you add sulfuric acid, put in a few drops of hydrogen peroxide (3%, from a drugstore). The peroxide destroys the S02, so the result from the second run will be approximately equal to the error introduced by compounds other than S02 (pigment, tannin). Subtract the second result from the first result to arrive at a corrected value for free S02 *(+1-20%* or so).

*CALCULATIONS*

S02 is expressed in parts per million (ppm), also called milligrams per liter *(mg/L),* another way of expressing S02.

$$\text{PPM S02} = \frac{\text{NORMALITY OF IODINE x OF IODINE USED X 32,000}}{\text{VOLUME OF SAMPLE}}$$

*Ifsample volume is 50 ml (do not count water used as dilution):*

0.0156 N Iodine: ppm = ml of iodine used x 10
0.0200 N Iodine: ppm = ml of iodine used x 12.8

The calculation factors above would be valid until standardization indicates that iodine has lost strength.

*STANDARDIZATION OF IODINE*

Iodine solutions are very unstable since they are sensitive to light, especially sunlight. An unopened, full *glass* bottle (iodine escapes through plastic bottles) kept in the dark may not begin to decay for months, but as it is being used, it will gradually lose strength. When exposed to direct light, the decay is rapid. Iodine left in a buret loses much of its strength within hours, and in direct sunlight it will decay before even one titration is finished.

Iodine is standardized against sodium thiosulfate (Na2S203), a chemical that changes strength more slowly than iodine, whose strength must be checked ("standardized") with sodium thiosulfate at frequent intervals, usually once a week if used often. The 0.025 N sodium thiosulfate is good for around 4-6 months, depending on airspace in the bottle; 0.1 N lasts up to one year.

An iodine-based method such as is presented here gives a reasonable approximation for free and total S02 on white wines that have not been infected with Botrytis, undergone ML fermentation, or had ascorbic acid added. With practice, total S02 can be tested on red wine with this method but NOT free S02 on reds. The tremendous error for free S02 on red wine is avoided with vacuum aspiration (aeration/oxidation) apparatus, which we recommend for wineries.

*PROCEDURE*

*Note: This assembly uses a buret for the titration, which is not included in the Assembly components. See Titration Assembly, 5AOOO or 5AO J0 for buret, stand, clamps, and accessories.*

FREE S02 (non-ML white wines, rose, any light-colored juice or must)

1. Preparation: Clean off a well-lit work surface out of direct sunlight and spread paper to catch spills. If there is no sink, provide distilled water for rinsing. Have paper towels handy.

2. Fill a glass buret with iodine. Standardize it if more than a week has gone by since its last standardization. Do not let the buret stand in the buret for more than a few minutes.

3. Pipet 50 ml of sample into a 500 ml Erlenmeyer flask (a 250 ml flask is OK for free S02 test).

4. Add 5 ml of sulfuric acid (H2S04 1 + 3) and one squirt (2-5 ml) of starch from starch dispenser.

5. Titrate immediately with iodine until the sample turns a bluish-purple color that lasts at least 30 seconds but fades within two minutes. DO NOT swirl the sample more than necessary to mix in the iodine; sloshing loses S02.

6. Write down amount of iodine used and calculate ppm free S02.

TOTAL S02 (white, red, rose, and juice or must)

1. Fill buret as above.

2. Pipet 50 ml of sample into 500 ml Erlenmeyer flask (a 250 ml flask is OK for whites, not for reds).

3. Add 10 ml of sodium hydroxide 10% (or 25 ml of 1 N NaOH; 0.01 N NaOH will not work). Swirl once and let stand undisturbed for 10 minutes. White wines turn gold, reds green.

4. Add 10 ml sulfuric acid 1 + 3 (sample will return to close to its original color), and add one squirt of starch. If red wine is very dark, add up to 50 ml of water.

5. Titrate immediately with iodine until the sample turns a bluish-purple color that lasts at least 30 seconds but fades within two minutes. In red wines, the color change is seen as a darkening of the sample. Again, do not swirl too much or slosh.

6. Write down the amount of iodine used and calculate total S02.