

# SO<sub>2</sub> Management in Winemaking

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# Roles of SO<sub>2</sub> in Wine

- Antimicrobial
  - knock down yeast (esp. wild) and bacteria
  - Wild yeast and bacteria potential source of off flavors
- Anti-oxidant
  - Oxidation of wine leads to brown color, decrease in varietal aromas, development of nutty, sherry aromas
  - In juice, SO<sub>2</sub> inhibits enzymatic oxidation (PPO)
  - In wine, SO<sub>2</sub> reacts with chemical oxidation intermediates & oxidized compounds to reduce apparent effect
- The chemistry behind how SO<sub>2</sub> performs these activities fairly complicated

# Three Areas to Cover Today

- Basic Chemistry of SO<sub>2</sub>
- Managing SO<sub>2</sub> during Winemaking
- The A/O Method for Testing Free SO<sub>2</sub>

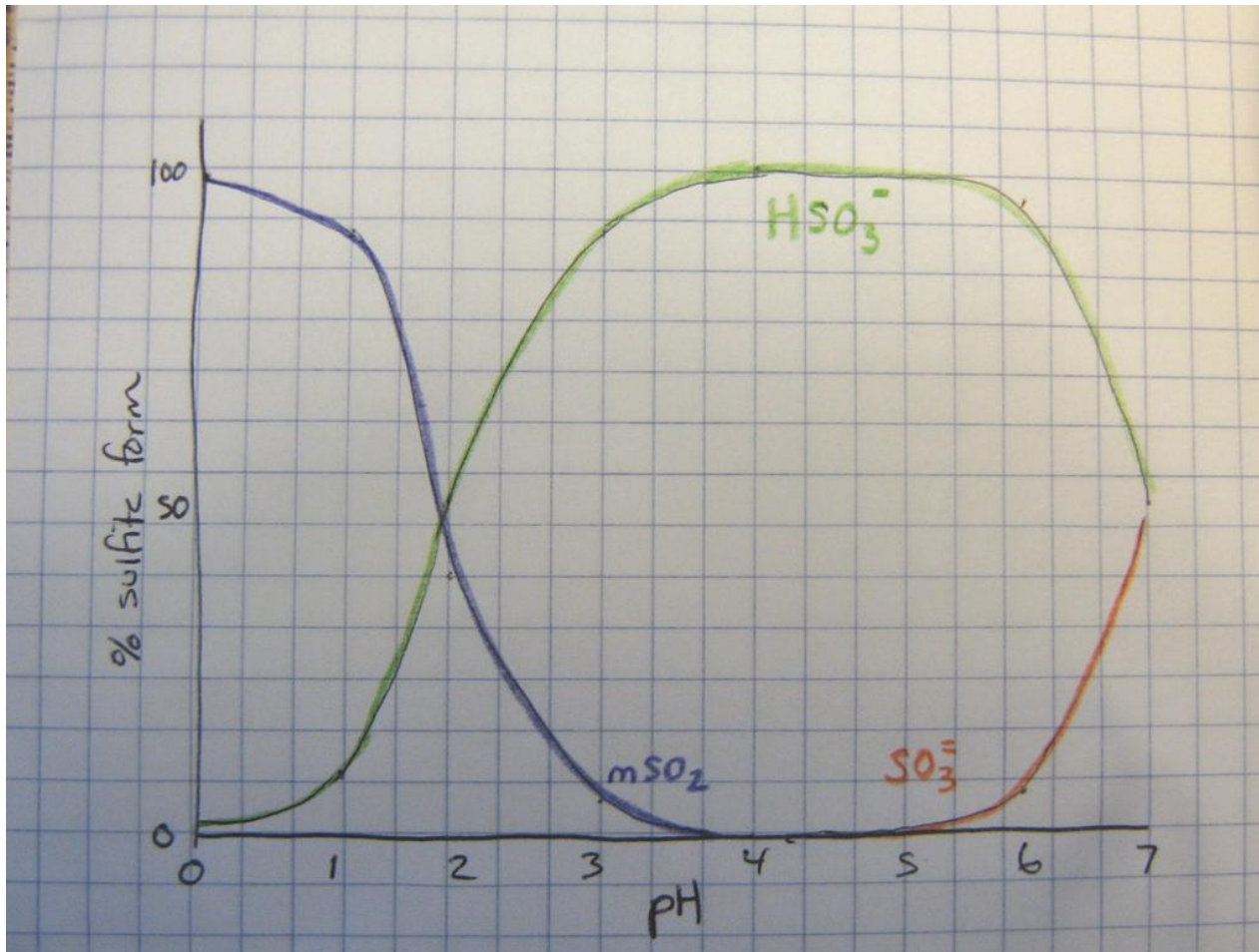
# Basic Chemistry of SO<sub>2</sub>

- SO<sub>2</sub> in wine exists as free SO<sub>2</sub> equilibrium, some free SO<sub>2</sub> can bind certain wine compounds to become bound SO<sub>2</sub>
- All the SO<sub>2</sub> in the wine is referred to as the total SO<sub>2</sub>
- Total SO<sub>2</sub> = free SO<sub>2</sub> + bound SO<sub>2</sub>

# Free SO<sub>2</sub> in Wine

- Exists in three forms: molecular SO<sub>2</sub>, bisulfite ion (HSO<sub>3</sub><sup>-</sup>), sulfite ion (SO<sub>3</sub><sup>=</sup>)
- The forms are in equilibrium, amounts of each dependent on pH
- At wine pHs, mostly in bisulfite form
- Eyes on the Prize: molecular SO<sub>2</sub> is the form that is antimicrobial

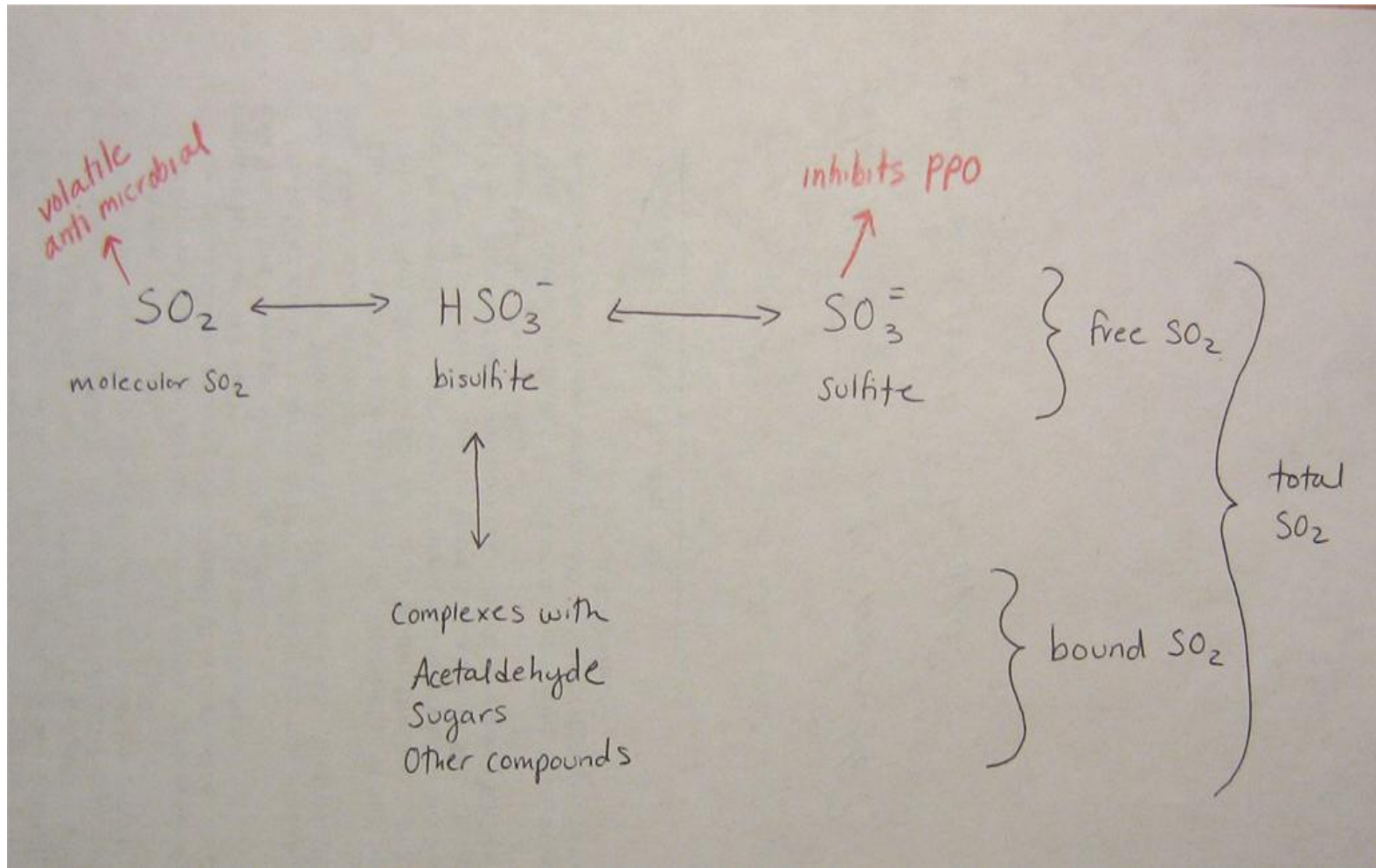
# The Ever Present Free SO<sub>2</sub> Graph



# Bound SO<sub>2</sub> in Wine

- The bisulfite form binds to wine components such as sugar, acetaldehyde, and phenolic compounds.
- Amount SO<sub>2</sub> bound varies (25 to 60%)
- When bisulfite ion reacts with these compounds, no longer takes part in free SO<sub>2</sub> equilibrium

# Forms of SO<sub>2</sub> in Wine



# Molecular SO<sub>2</sub> is Antimicrobial

- A level of 0.8 ppm molecular SO<sub>2</sub> generally viewed as sufficient for microbial stability
  - Amount of SO<sub>2</sub> added that is in free form is dependent on how much is bound
  - Amount of free SO<sub>2</sub> that is in molecular SO<sub>2</sub> form is dependent on pH
    - At pH 3.0, molecular SO<sub>2</sub> is 6% of free SO<sub>2</sub> and therefore need 13 ppm free SO<sub>2</sub> to have 0.8 ppm molecular SO<sub>2</sub>
    - At pH 3.8, molecular SO<sub>2</sub> is 1% of free SO<sub>2</sub> and therefore need need 79 ppm free SO<sub>2</sub> to have 0.8 ppm molecular SO<sub>2</sub>
- If goal is to ensure microbial stability, need to know pH & amount of free SO<sub>2</sub>

# Vinquiry – Free SO<sub>2</sub> Chart

## DISTRIBUTION OF FREE SO<sub>2</sub> From pH 3.0 – 4.0 (pKa = 1.81)

pH	% Molecular SO <sub>2</sub> (m)	% Bisulfite (HSO <sub>3</sub> <sup>-</sup> )	% Sulfite (SO <sub>3</sub> <sup>=</sup> )	Minimum ppm of Free SO <sub>2</sub>	
				0.8 molecular	0.5 molecular
3.00	6.1	93.9	0.012	13	8
3.05	5.3			15	9
3.10	4.9	95.1	0.015	16	10
3.15	4.3			19	12
3.20	3.9	96.1	0.019	21	13
3.25	3.4			23	15
3.30	3.1	96.8	0.024	26	16
3.35	2.7			29	18
3.40	2.5	97.5	0.030	32	20
3.45	2.2			37	23
3.50	2.0	98.0	0.038	40	25
3.55	1.8			46	29
3.60	1.6	98.4	0.048	50	31
3.65	1.4			57	36
3.70	1.3	98.7	0.061	63	39
3.75	1.1			72	45
3.80	1.0	98.9	0.077	79	49
3.85	0.9			91	57
3.90	0.8	99.1	0.097	99	62
3.95	0.7			114	71
4.00	0.7	99.2	0.122	125	78

# SO<sub>2</sub> as Antioxidant

- In juice, SO<sub>2</sub> reduces enzymatic oxidation by inhibiting the activity of polyphenol oxidase (PPO)
- In wine, chemical oxidation of compounds
- In wine, sulfite ion is the antioxidant form but at wine pH ineffective as antioxidant

# Oxidation Reactions in Wine

- Phenolic compounds -> Oxidized compounds
  - $\text{H}_2\text{O}_2$  is an oxidation intermediate, enables oxidation of ethanol to acetaldehyde
  - $\text{SO}_2$  scavenges  $\text{H}_2\text{O}_2$ , preventing further oxidation of phenolics and ethanol
  - $\text{SO}_2$  can recycle quinone oxidation intermediate back to phenolic compound
  - $\text{SO}_2$  can bleach brown compounds
- Ethanol -> Acetaldehyde
  - $\text{SO}_2$  binds strongly to acetaldehyde, removes nutty, sherry like aroma

# SO<sub>2</sub> as Anti-Oxidation Agent in Wine

- SO<sub>2</sub> masks effect of oxidation by binding acetaldehyde and bleaching brown pigments
- SO<sub>2</sub> curtails oxidation reactions by scavenging oxidation intermediates
- SO<sub>2</sub> recycles oxidation intermediates

# Amount of $\text{SO}_2$ is not Stable

- Will lose free  $\text{SO}_2$ :
  - in headspace (molecular  $\text{SO}_2$  is volatile)
  - as binds oxidation reactants/products
  - as binds solids in wine
- Test free  $\text{SO}_2$  to ensure protection

# Review Basic Chemistry of SO<sub>2</sub>

- SO<sub>2</sub> exists as free and bound forms in wine
- Free SO<sub>2</sub> consists of molecular SO<sub>2</sub>, bisulfite, and sulfite ion; amounts of each dependent on pH of wine
- The molecular SO<sub>2</sub> form inhibits yeast and bacteria, 0.8 ppm molecular SO<sub>2</sub> good for stability
- To ensure microbial stability, need to know wine pH and how much SO<sub>2</sub> is present as free SO<sub>2</sub>
- SO<sub>2</sub> can inhibit enzymatic oxidation in juice
- SO<sub>2</sub> not a true anti-oxidant in wine, but does help to curtail oxidation reactions and covers up acetaldehyde smell quite nicely

# Three Areas to Cover Today

- Basic Chemistry of  $\text{SO}_2$
- Managing  $\text{SO}_2$  during Winemaking
- The A/O Method for Testing Free  $\text{SO}_2$

# Using SO<sub>2</sub>

- SO<sub>2</sub> is typically added to wine in the form of Potassium Metabisulfite.
- By weight, only 57% of Potassium Metabisulfite is released as SO<sub>2</sub>
  - 1 gram of PMBS contains 0.57 grams SO<sub>2</sub>
- SO<sub>2</sub> concentration measured in parts per million (ppm) which is equivalent to milligrams per liter (mg/L)
  - 1 gram of PMBS in 5 gallons gives 30 ppm SO<sub>2</sub>

# Adding SO<sub>2</sub> to Juice/Must

- SO<sub>2</sub> gives a little jump-start to wine yeast (around 30 ppm SO<sub>2</sub>)
- SO<sub>2</sub> lowers activity of PPO in juice and must
  - Add SO<sub>2</sub> to prevent browning, oxidation of fruity & floral aroms (oh, let's see, 30 ppm SO<sub>2</sub> for good inhibition)
  - Be stingy with the SO<sub>2</sub> to promote browning, oxidation (0 ppm SO<sub>2</sub>)
    - Brown pigments will be removed by lees
    - Resulting wine more color stable, resistant to oxidation
- Add more SO<sub>2</sub> to moldy must/juice (>50 ppm SO<sub>2</sub>)

# SO<sub>2</sub> During Fermentation

- Yeast produce a small amount SO<sub>2</sub>, 10 ppm, during fermentation
- Wine yeast neutralize SO<sub>2</sub> during fermentation with acetaldehyde
- SO<sub>2</sub> not bound to acetaldehyde is bound to solids
- At the end of alcoholic fermentation there is essentially no free SO<sub>2</sub>

# Adding SO<sub>2</sub> After Alcoholic Fermentation

- Add 0.8 ppm molecular SO<sub>2</sub> to prevent yeast and malolactic bacteria growth (also sufficient for anti-oxidation purposes).
  - 1 gram of PMBS dissolved in 5 gallons of wine would give 30 ppm SO<sub>2</sub>
  - At a wine pH of 3.35, 30 ppm free SO<sub>2</sub> would provide 0.8 ppm molecular SO<sub>2</sub>
  - But that assumes no SO<sub>2</sub> in bound form, 25% to 60% is typically bound
  - Need to determine amount of free SO<sub>2</sub> to ensure protection

# SO<sub>2</sub> and Malolactic Fermentation

- Malolactic bacteria highly sensitive to free SO<sub>2</sub>
- Delay additions of SO<sub>2</sub> until after MLF complete
- ML Bacteria also sensitive to bound SO<sub>2</sub>, use a light hand when adding SO<sub>2</sub> to juice/must (>25 ppm total SO<sub>2</sub> may inhibit)
- Keep ML bacteria happy (temp, pH, alcohol) to ensure timely completion while wine not protected by SO<sub>2</sub>

# Adding SO<sub>2</sub> before Bottling

- Ensure have enough free SO<sub>2</sub> for 0.8 ppm molecular SO<sub>2</sub>
- Add a little bit extra - to account for free SO<sub>2</sub> loss during bottling
  - I typically target a free SO<sub>2</sub> that is 10 to 15 ppm higher than the level of free SO<sub>2</sub> needed for 0.8 ppm molecular SO<sub>2</sub>
  - Target more or less depending on trauma of bottling method (O<sub>2</sub> pick-up)

# Let's give SO<sub>2</sub> a Helping Hand

- The combination of limiting O<sub>2</sub> exposure, using clean equipment, and maintaining free SO<sub>2</sub> leads to healthy wines
  - O<sub>2</sub> needed in oxidation of wine components
  - SO<sub>2</sub> doesn't prevent this oxidation, helps cover and curtail
  - Clean equipment and full containers helps limit yeast and bacteria contamination/growth

# Review Managing SO<sub>2</sub> Winemaking

- Add SO<sub>2</sub> to must/juice to help wine yeast establish & prevent enzymatic oxidation
- After alcoholic fermentation, maintain 0.8 ppm molecular SO<sub>2</sub> to for microbial stability (also sufficient for anti-oxidation purposes)
- Delay SO<sub>2</sub> additions to wines undergoing MLF
- Ensure > 0.8 ppm molecular SO<sub>2</sub> at bottling for microbial stability

# Three Areas to Cover Today

- Basic Chemistry of SO<sub>2</sub>
- Managing SO<sub>2</sub> during Winemaking
- The A/O Method for Testing free SO<sub>2</sub>

# Why We Test SO<sub>2</sub>

- Survey says: audience home winemakers either don't test or use Titrets to test for free SO<sub>2</sub>.
- Testing free SO<sub>2</sub> enables winemaker to know if wine is protected with 0.8 ppm molecular SO<sub>2</sub>.
- Testing for SO<sub>2</sub> enables the winemaker to troubleshoot wines that are not behaving properly.
- Testing enables winemaker to figure out how SO<sub>2</sub> is being consumed in *their* wines.

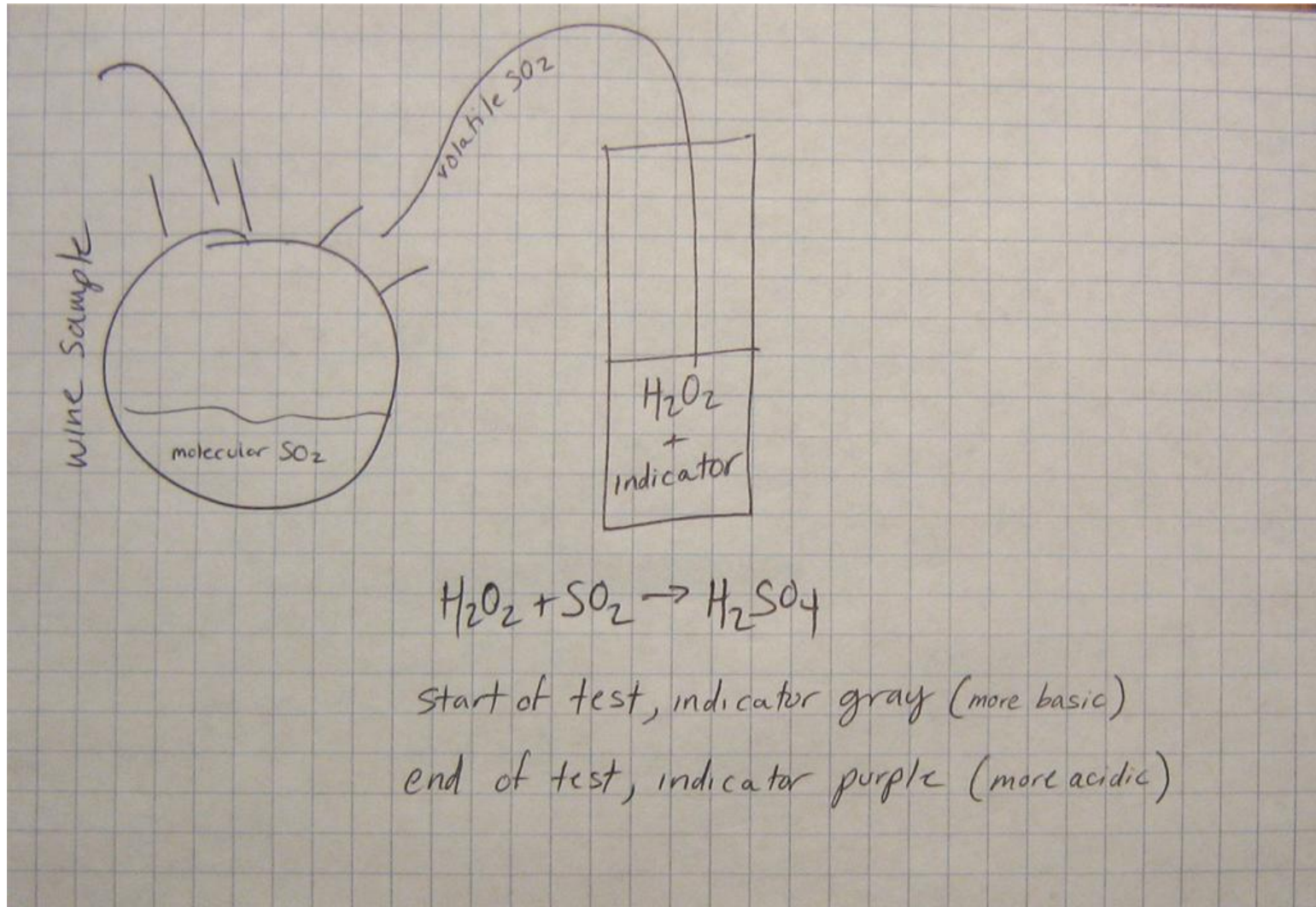
# A/O method is A-OK

- Titrations based on Ripper method
  - Easy to use and quick (just over \$1 per test)
  - Can only be used with whites
- Aeration/Oxidation method
  - Most accurate method
  - Can be used on reds
  - \$\$\$ investment in equipment
  - Time investment to set-up & learn method

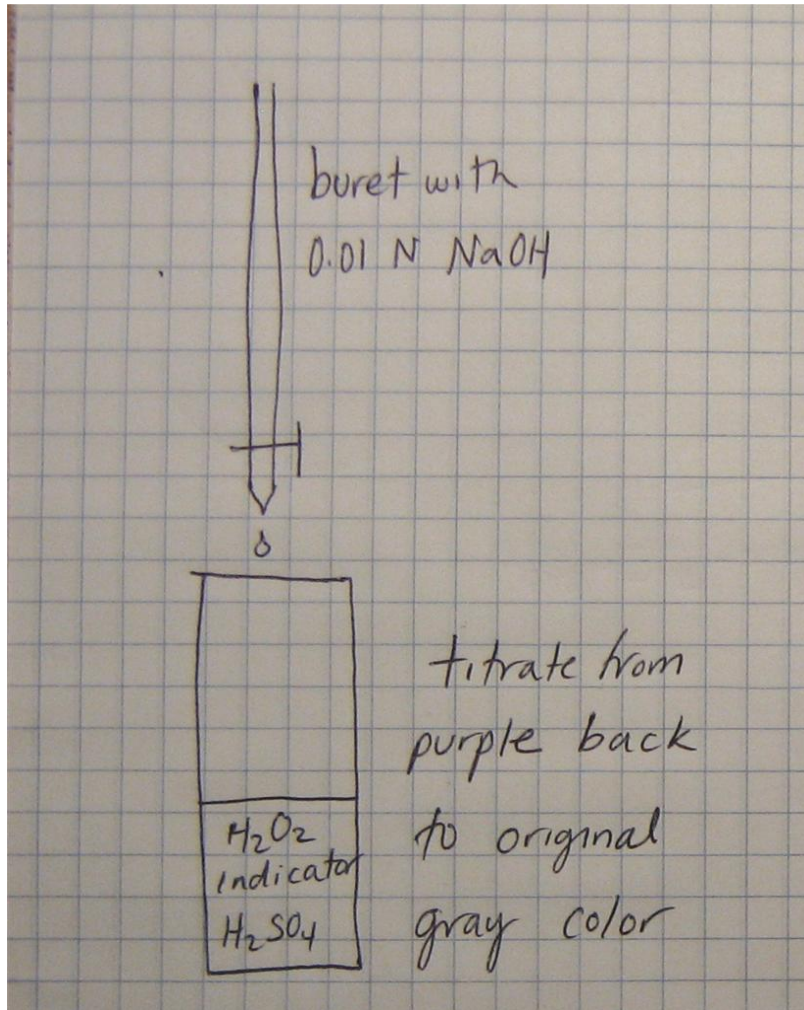
# A/O Method Theory

- Acidify wine sample to drive all free  $\text{SO}_2$  into the volatile molecular  $\text{SO}_2$  form.
- Allow the molecular  $\text{SO}_2$  to react with hydrogen peroxide ( $\text{H}_2\text{O}_2$ ) to form sulfuric acid ( $\text{H}_2\text{SO}_4$ )
- Titrate sulfuric acid ( $\text{H}_2\text{SO}_4$ ) with sodium hydroxide base ( $\text{NaOH}$ ) to calculate starting amount free  $\text{SO}_2$ .

# A/O Method Free SO<sub>2</sub> – Step 1



# A/O Method Free SO<sub>2</sub> – Step 2



- Amount of 0.01 N NaOH used in titration directly proportional to amount of free SO<sub>2</sub> in original wine sample

# A/O Method in Practice

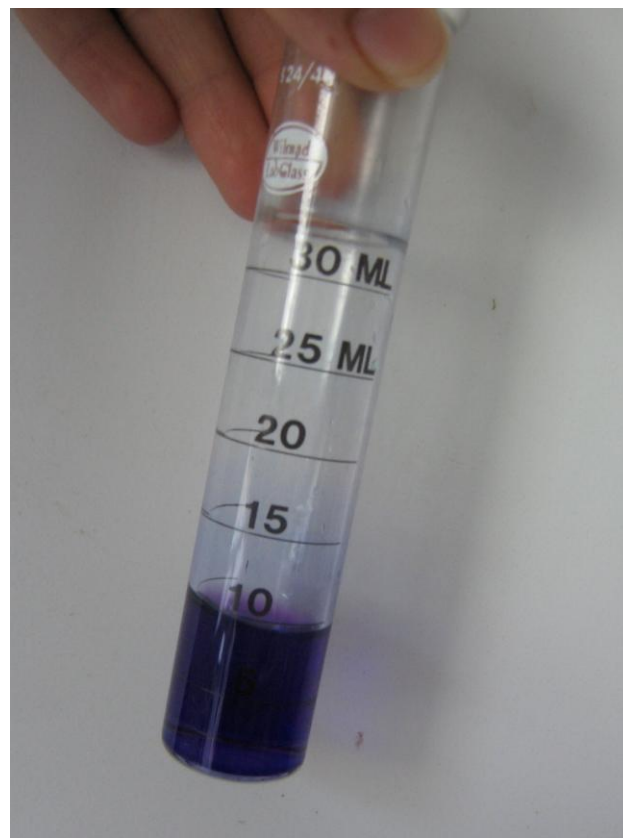
- 20 mL wine sample acidified with 10 mL 25% phosphoric acid to drive all the free  $\text{SO}_2$  to the molecular  $\text{SO}_2$  form (volatile form)
- Volatile  $\text{SO}_2$  leaves round bottom flask via vacuum suction (aspirator) or air pump into an impinger containing 3% hydrogen peroxide and a few drops of  $\text{SO}_2$  indicator
- Takes about 15 mins to complete,  $\text{SO}_2$  indicator is gray at the start (more basic) and turns purple as hydrogen peroxide reacts with  $\text{SO}_2$  to form sulfuric acid (more acidic)
- The sulfuric acid formed in the impinger is titrated with 0.01 N NaOH, as sulfuric acid is titrated to endpoint the indicator in the solution turns from purple back to initial gray color
- Amount of 0.01 N NaOH used in titration is proportional to free  $\text{SO}_2$  present in original sample

# Picture of A/O Method in Practice

$\text{H}_2\text{O}_2$  and  $\text{SO}_2$  indicator at start



After reaction with volatile  $\text{SO}_2$



# A/O Method – 0.01 N NaOH

- The 0.01 N NaOH prone to degrade overtime
- Using less than full strength 0.01 N NaOH affects accuracy of test
  - Use freshly opened 0.01 N NaOH
    - Expensive (16 oz is \$16 at [MoreWine.com](http://MoreWine.com))
  - Dilute 0.1 N NaOH to 0.01 N NaOH; check titre
    - Requires more equipment, time to test titre
    - Eventually inexpensive (16 oz 0.1 N NaOH is \$5 at [Presque Isle](http://PresqueIsle.com))

# Make & Check Titre of 0.01 N NaOH

- Determining titre strength of 0.01 N NaOH with potassium acid phthalate ensures accurate results
- Will need: buret, buret stand, flasks, pipets, 0.1 N NaOH, graduated cylinder,  $\text{dH}_2\text{O}$ , potassium acid phthalate, phenolphthalein indicator
- Use much of the same equipment set-up for titratable acidity

# A/O Method – Aspirator vs Pump

- Need to volatilize  $\text{SO}_2$  to enable reaction with  $\text{H}_2\text{O}_2$
- Aspirator requires a sink faucet with water flow to create vacuum
  - Can control flow rate easily by opening up the faucet
- Pump requires electricity
  - Can control flow rate via pump itself or inline valve
  - Nice not to waste water, worry about flooding

# A/O Method – Flowmeter vs Wing It

- The recommended 1 liter/min flowrate is to ensure one is not blasting the gaseous  $\text{SO}_2$  right through your hydrogen peroxide sample and out the other end
- Flowmeters are expensive (\$75 to \$120)
- Might just want to wing it and track bubbling rate by eye

# A/O Method – Bubble Rate



# Set up A/O at Home - EASY

- Morewine.Com has a starter kit ready from the box with user friendly instructions
  - Uses air pump so no water source needed
  - With flowmeter (\$490) and without (\$295)
  - Includes reagents for 2 to 3 tests
  - Uses a syringe to titrate w/0.01 N NaOH (requires fresh NaOH)
  - Upgrade to buret titration system to check titre of 0.01 N NaOH (enables one to make own 0.01 N NaOH solution)

# Set Up A/O at Home – Basic

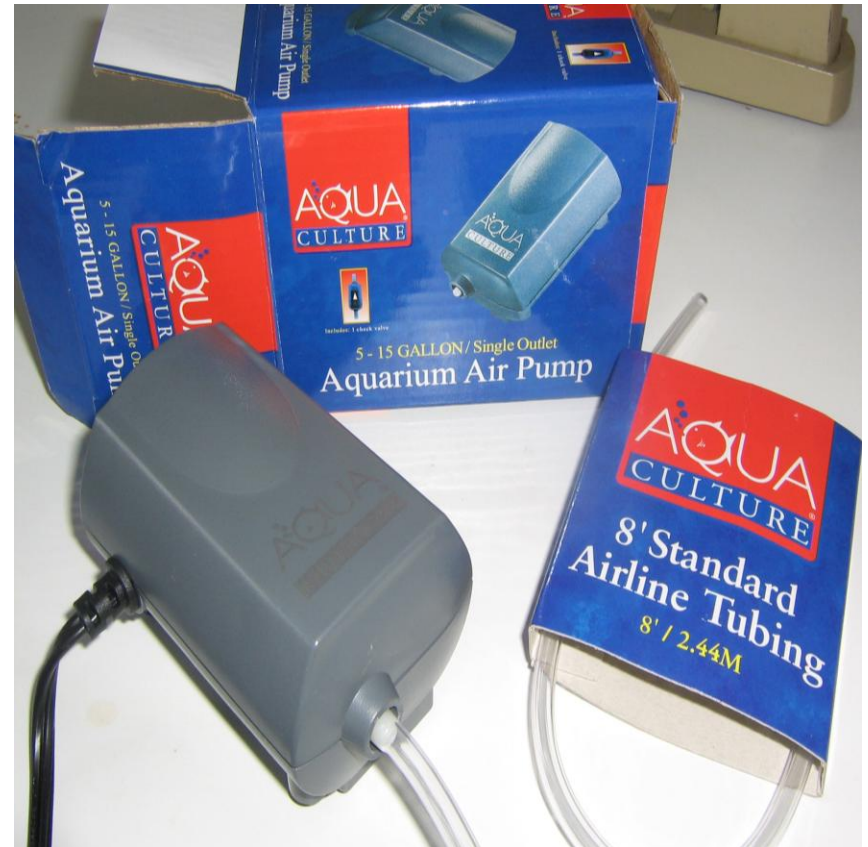
- Presque Isle has a starter kit with aspirator and flowmeter (\$326)
  - Low price for including a flowmeter
  - Does not include reagents
  - Does not include titration set-up, pipets

# Set Up A/O at Home – DIY

- Buy components separately to accommodate what is already owned
  - Round Bottom Flask (\$45 MoreWine)
  - Impinger (\$57 MoreWine)
  - Stand and Clamps (Cynmar)
  - Flowmeter (\$75 to \$120)
  - Aspirator or Pump (<\$20)
  - Buret, Buret Stand, Buret clamp, flasks, cylinders, pipets (\$60 Cynmar)
  - Reagents (varies)

# DIY Further \$\$ Savings

- Buy an air pump & tubing from Walmart (\$8)
- One for small fish tanks, 5 to 15 gallon, provides close to 1 L/min flowrate



# Further \$\$ Savings

- Purchase 3% Hydrogen Peroxide at drug store (\$1) instead of from wine suppliers (\$16).



# Further \$\$ Savings

- Cynmar is a great supplier for labware:
  - Support stands (<\$10)
  - Clamps (~\$5)
  - 25 mL Buret (\$17.50)
  - Buret stand with clamp (\$25)
  - 20 mL volumetric pipet (\$2.90)
  - 100 mL Graduated cylinder (\$9.80)

# Review Using A/O Method

- Most accurate method, it is the real deal
- Best for home winemakers wanting to ensure consistency & interested in wine chemistry
- Determine titre strength of 0.01 N NaOH

# Three Areas We Covered Today

- Basic Chemistry of  $\text{SO}_2$ 
  - Free, Bound, molecular  $\text{SO}_2$ , pH, oh my!
- Managing  $\text{SO}_2$  during Winemaking
  - After alcoholic fermentation maintain 0.8 ppm molecular  $\text{SO}_2$  for microbial stability
- The A/O Method for Testing free  $\text{SO}_2$ 
  - A/O and a pH meter is all one needs

# The End

- Come visit us at Keuka Lake Vineyards down in Hammondsport
- ?'s -> [stacinugent@klvineyards.com](mailto:stacinugent@klvineyards.com)