

“Film Organisms”

(Acetic Acid Bacteria and Yeast)
Or, Do I really have to keep all of my
storage containers completely full?
Michael Ramsey, Teaching Lab Manager
UC Davis

This is a brief re-cap, in my words and interpretation, of his “Wine Flavor 101” presentation in the new **Jess Jackson, LEED Platinum, “Sustainable Building”**, next to the new student winery.



Employing thermal mass and a high performance cladding system, the building does not have a traditional heating and air conditioning system and is completely heated and cooled by passive strategies including night time ventilation and an underground thermal rock bed. Lighting and plug load requirements are met by photovoltaic panels on the roof and the building is pursuing net-zero energy certification from the Living Building Challenge. The building also captures and stores rain water from the roof. The Jess S. Jackson Sustainable Winery building will serve as a test bed facility for faculty currently collaborating with industry partners to develop innovative new systems to demonstrate net-zero water and net-zero energy in the commercial production of wine.

You can [read more project details in the building brochure \(PDF file\)](#).

Some Background: (http://en.wikipedia.org/wiki/Ethyl_acetate) Ethyl acetate (smells like nail polish) is the most common **ester in wine**, being the product of the most common volatile **organic acid** — **acetic acid**, and the ethyl alcohol generated during the **fermentation**. The aroma of ethyl acetate is most vivid in **younger wines and contributes towards the general perception of "fruitiness"** in the wine. Sensitivity varies, with most people having a perception threshold around 120 mg/L. **Excessive amounts of ethyl acetate are considered a wine fault** . Exposure to **oxygen** can exacerbate the fault due to the **oxidation** of ethanol to **acetaldehyde**, which becomes acetic acid.

Volatile Acidity (VA) measurement using a “Cash Still”, measures more than just the acetic acid (vinegar smell) content in the wine. A cash still is not really a still. It is a steam stripping device that also measures trapped CO₂ (as carbonic acid); SO₂ (as sulfurous acid); sorbate, lactic, formic, butyric, and propionic acids. **It does NOT measure the Ethyl Acetate (nail polish smell) nor the aldehyde content**. Legal limits for VA, via this method, are **1.4 g/L (.014 g/100ml) (14% by volume) in red wines** and **1.2 g/L (.012 g/100ml) (12% by volume) in white wines**. (<http://www.ttb.gov/ssd/pdf/tm503.pdf>).

Acetic Acid (Aroma threshold of acetic acid at around 1 g/L) can be formed by:

- Lactic Acid Bacteria (*anaerobic* ferment of glucose and producing lactic acid, CO₂).
- Oxidation of ethanol to acetaldehyde, becoming acetic acid.
- From *Saccharomyces* wine yeast and is very strain dependent. (*Saccharomyces* also produces Ethyl Acetate (glue-like smell), but most Ethyl Acetate come from “wild”, non-*saccharomyces* yeast)
- Film Yeast.

Film Yeast:

- **Require O₂ (aerobic) plus a growth media** (ethanol, organic acids (especially Malic Acid)).
- Can synthesize negative aroma compounds as aldehydes (stale smell, becoming Acetic Acid); Ethyl Acetate (glue-like); Acetoin (buttery cheese).
- Types of film yeast: *Candida vini* (formerly *Candida mycoderma* and often identified incorrectly by Kombucha makers as *Saccharomyces mycoderma*); *Pichia kluyveri*; *Candida parapsolosis*; *Acetobacter aceti*; *Acetobacter pasteurianus*.
- Can initially appear as **“Flowers of Wine”**/dusty and may rapidly become pellicular (skin like, thickening with time and O₂ exposure).

Acetic Acid Bacteria:

- **Present on the grapes and in the winery environment.**
- Several genera and many species. Most decline as ethanol is produced.
- **However, acetic acid is inhibitory to *Saccharomyces*, leading to stuck fermentations.**
- **These *acetobacter* species survive through to aging, storage, and bottling.**
 - ✓ Can survive periods of anerobic (without oxygen) conditions.
 - ✓ **Will begin growing with O₂ pickup** during fining, racking, stirring, filtering.
 - ✓ *A. pasteurianus* requires less O₂ than *A. aceti*.
 - ✓ **Can survive in the wine bottle and regrow in days in an opened bottle.**

A Fall Quarter Experiment

- Carboys filled 9/10 full and *Acetobacter pasteurianus* added to all.
- Nutrients and other film yeasts added.
- **Visible surface film yeast began to form on all carboys, within one week** in control and “plus nutrients”.
- Added organisms were **quickly overwhelmed by the indigenous *Acetobacter pasteurianus*** and by week five, all surfaces were completely covered.
- No cells of any of the other added film yeast could be seen under the microscope.

Could this spoilage have been prevented strictly through sanitation?

- **No!**
- You can not hope to kill 100% of all organisms through normal sanitation

Could this spoilage have been prevented by gassing the surface?

- **No!**
- **Neither Argon, nor Nitrogen, nor CO₂ is a substitute for a completely full container**, with the least (or none) surface area as possible.
- Perhaps replenishing the gas, often, might buy you some time. The larger the headspace, the less gassing works.
- **There is no way to completely replace the O₂ in a container by using another gas.**
- Check out *“Henry’s Law”*, *“Law of Partial Pressures”*, and the *“Ideal Gas Law”*.
- Add to this, leaking connections and fittings.
- Especially vulnerable are Variable Capacity Tanks (potentially large surface area and leaking gas fittings)!
- If in a **constant storage temperature**, fill to ¼” of the stopper/high point. **Headspace is only for heat expansion requirements.**

What about SO₂?

- **Largely ineffective if there is any headspace**, and it doesn’t protect the surface.
- Generally ineffective once the film is formed.
- Organisms create aldehyde, which binds SO₂.

Conclusions:

- **Avoid moldy and damaged grapes.**
- **Maintain proper SO₂ from Crush forward.**
- **Good sanitation, at all times, helps.**
- **Use of low cellar temperatures (less than 60F) can slow growth.**
- **pH of below 3.6 favors non-spoilage.**

The number one component of the creation of acetic acid was oxygen/headspace.

