Master Brewers Association of the Americas

Dedicated to the technology of brewing.

Barrel Dwellers: Microbiology of Barrel Aged Beers

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"The only way to sanitize a wooden barrel is with gasoline and a match"

Dr. MJ Lewis, 1982 UC Davis brewing lecture

Charring vs Toasting

Whiskey barrels

Wine barrels

Why Barrels Harbor Microbes

- Rough surfaces easy to attach to and are protective
- Porous nature gives additional shelter and nutrient source (wood sugars & product)
- Very difficult to clean, sanitize and inspect
- How many of you have taken a barrel apart?

We cannot control barrel microbiology but we can manage it to try to control its growth

Key Microbes in Barrels

- Bacteria
  - *Acetobacter, aerobe*
  - *Pediococcus, aerotolerant anaerobe*
  - *Lactobacillus, aerotolerant anaerobe*
- Yeast
  - *Brettanomyces/Dekkera*

Oxygen and Microbes

- *Aerobes* can only grow in presence of oxygen, will not grow anaerobically
- *Aerotolerant* will not grow in presence of oxygen but can tolerate oxygen
- *Obligate anaerobes* cannot grow with oxygen, it is toxic to them
- *Facultative* can grow aerobically or anaerobically

Catalase Test for Aerobes

$$2\ H_2O_2 \xrightarrow[\text{Catalase}]{} 2\ H_2O + O_2\ (\text{fizz})$$

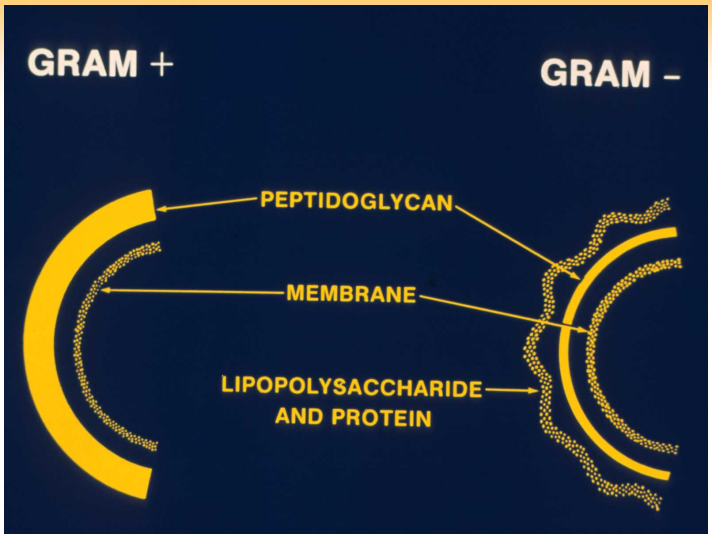
- Add one drop of Hydrogen Peroxide (H_2O_2) to colony
- Catalase produced by aerobes will fizz
- Anaerobes do not produce Catalase, no fizz

Gram Stain Reaction

Bacteria can be sub-classified by reaction to the Gram Stain:

Gram Positive = Violet/Purple Colored

Gram Negative = Red/Pink Colored



The Lipopolysaccharide/protein membrane that resists the stain also makes Gram Neg bacteria resistant to hop alpha acids

Quick Easy Test

- Slurry the colony onto a slide and add a drop of Potassium Hydroxide (KOH)
- Gram Negative bacteria will form a gooey mass
- Gram Positive will not be gooey

pH and Microbes

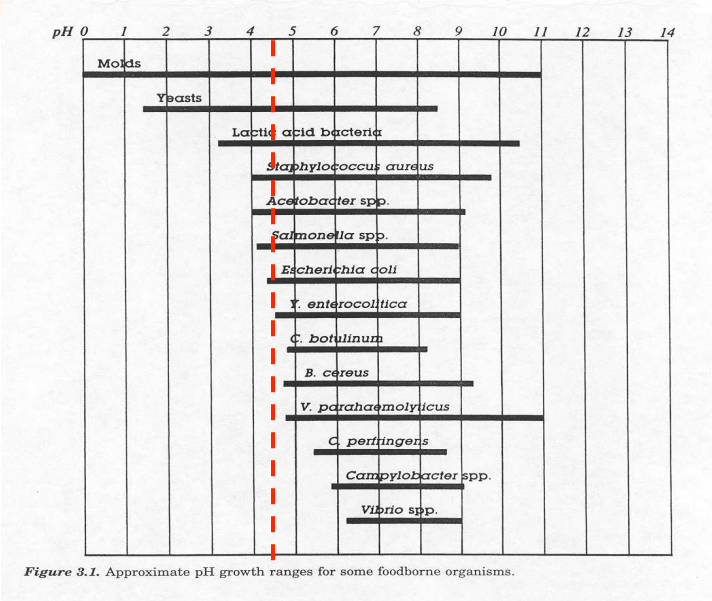


Figure 3.1. Approximate pH growth ranges for some foodborne organisms.

Factors Affecting Pathogens in Beer

- Oxygen (beer is anaerobic)
- pH (<4.6 in beer)
- Temperature (low temperature)
- Nutrients (carbohydrate/nitrogen depleted)
- Selectors:
  - CO_2
  - Ethanol (>2.5% ABV)
  - Hop Acids (>20 BU's)

Spoilage or Innovation?

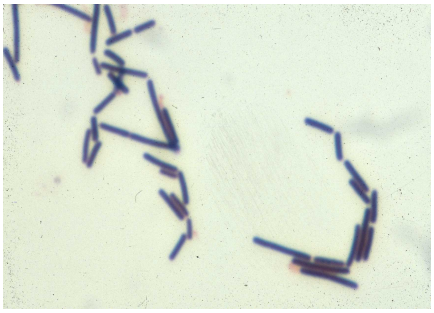
Quality = Flavors and Characters present in the beer that the brewer intended to be there

- Acidity, Diacteyl, Phenloic, Fusel Alcohols, Esters, etc
- Considered spoilage in some styles and brands but desirable in others
- New flavor boundaries are being explored

Acetobacter

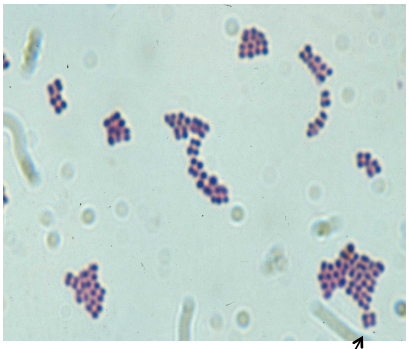
- Gram Neg, Short rods, Aerobe
- Oxidizes ethanol to acetic acid (vinegar)
- Aerobic growth seen as a surface film or around bung area
- Will produce acetaldehyde in low oxygen atmosphere

Lactobacillus



- Gram Pos. Rods in pairs or chains, aerotolerant anaerobe
- Produces lactic & acetic acids from sugars
- Some produce diacetyl and phenols
- Prefers pH >3.5

Pediococcus

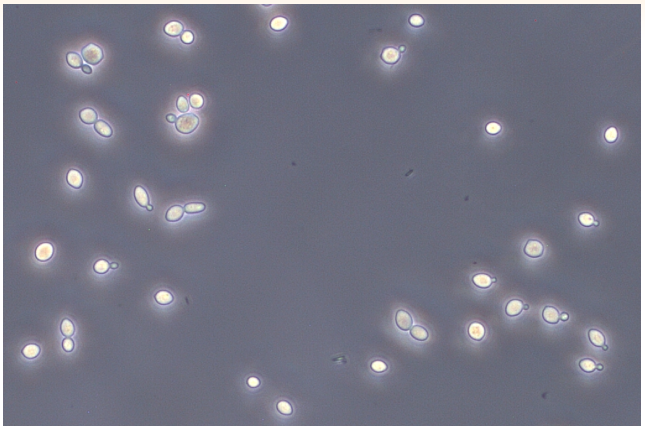


- Gram Pos. Round cocci in tetrads or clumps of cells, aerotolerant anaerobe
- Prefers pH >3.5
- Produces lactic acid from sugars and also diacetyl
- May also produce "ropiness", and slimy growth

Brettanomyces

- "British brewing fungus"
- *Dekkera* is the sporulating form
- Common species are:
  - *B. anomalous*
  - *B. bruxellensis*
  - *B. lambicus*
  - *B. claussenii*

Brettanomyces

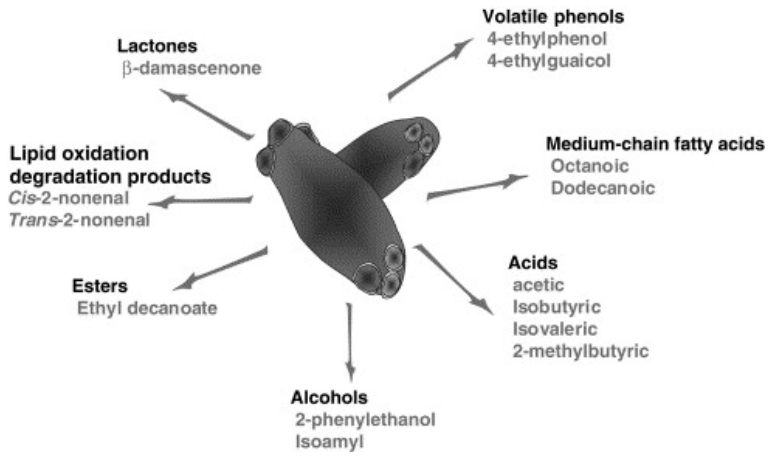


Can appear ovoid similar to S. cerevisae or elongated, depends on growth media and conditions.

Brettanomyces

- Can grow fast (weeks) in favorable conditions or slowly (years) in less favorable.
- Out-competed by *Saccharomyces* in beer fermentations because of low growth rate so usually seen post-primary fermentation, “waits”
- Very little oxygen required to stimulate growth
- Can form a surface film or “pellicle”
- Uses a range of carbon sources: sugars or ethanol, can survive in barrels utilizing wood sugars – cellobiose
- Can also super attenuate by utilizing dextrins via [illegible]-Glucosidase activity

Flavor compounds by Brett



Adapted from Suarez et al. 2007

Brett Flavors

'Positive' Brett aromas:

– Rancid: fatty acids (isovaleric acid)

– Clove, smoked, medicinal: volatile phenols

– Sweat, wet leather, goat-like, wet dog: caprylic, caproic and capric acid

'Negative' Brett aromas:

– Mousy (mouse or rabbit urine): 2-acetyl-1-pyrroline, ethyltetrahydropyridine, produced in early fermentation

Brettanomyces

- Most commonly found in used barrels
  - Found up to 8 mm deep in staves, can survive on the cellobiose, "wood sugar"
  - Almost impossible to remove
- Brett not usually found in brand new barrels
  - Toasting/charring processes an effective sterilizer but barrels can quickly become infected with Brett once filled
  - Newer barrels have higher cellobiose, make a good habitat for Brett to hide in

Barrel Cleaning

- Manage populations and control growth
  - Rinse with warm water after using
  - Clean with weak alkaline agents like ProxyClean
  - Use of Steam or hot water (180F for 20 minutes) are effective to knock down populations and have good penetration
  - Ozone is very effective, no residuals but poor penetration. Barrels must be very clean to work.

Barrel Maintenance

- Good practices:
  - Use continuously, keeping the barrels full
  - Avoid letting them dry out completely for a long period!
  - Top off or use of inert gas to avoid air space
  - Keep bung area clean
  - Empty barrel storage – wet (citric acid/SO_2 solution) or dry (gaseous SO_2 – sulfur wicks)
  - Track barrel uses, cycle number, issues with each barrel
  - Isolate and label infected barrels

Avoid Cross Contamination

- Isolate barrels, hoses, fittings and clamp gaskets from non-barrel production
- Run barrel beer operations, i.e. transfers and bottling, *after* non-barrel operations.
- Always clean and sanitize hoses, pumps, lines, etc. after processing barrel beers.
- Consider using a flash pasteurizer prior to entering the bright beer tank and the filler

Thank you!

Thanks to:

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MBAA – ASBC Brewing Summit

- June 5-7th 2014
- Chicago at the Palmer House Hilton
- Papers, Workshops and Exhibits
- www.mbaa.com