UC DAVIS VITICULTURE AND ENOLOGY




Best fermentation management practices

WF101: CURRENT ISSUES IN FERMENTATION MANAGEMENT

JULY 27TH, 2018

ANITA OBERHOLSTER

Know your grapes

- Brix – potential EtOH production
- YAN – nutritional needs
- TA – acid balance
- pH – antimicrobial control, taste
- Organic acid composition
  - For ex. malic acid contribution to predict pH change in the case of red wine production

Know your grapes

- Condition of the grapes
  - Raisons – Brix measurement will be inaccurate
  - Mold or rot – what is the risk?
  - Uneven ripeness
  - Unripe/over-ripe

Brix

- Why measure?
  - Indication of fruit ripeness
  - Potential ethanol production in wine
    - Yeast vary in their efficiency to convert sugar to alcohol – could differ up to 0.8 % v/v
    - Conversion factor of Brix to EtOH% 0.55-0.64
  - Determine optimal yeast to use for fermentation
  - Follow progress of fermentation

Brix adjustments

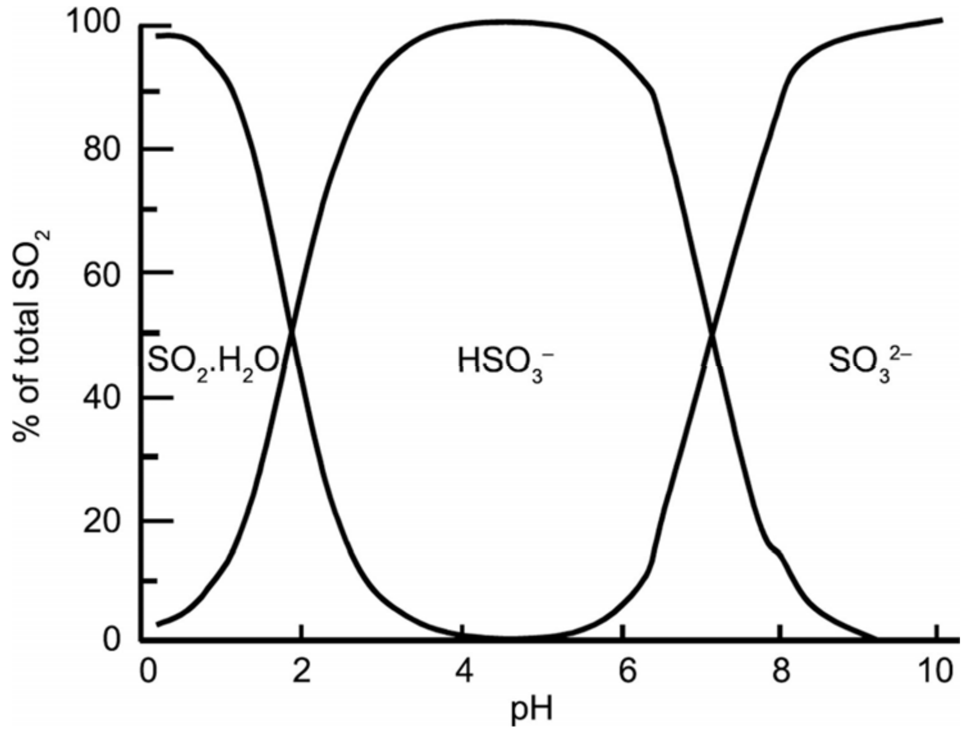
- Water back
- Grape concentrate
  - Use Pearson's Square
    - Vol of water/concentrate = V(D-A)/(C-D)
      - V = vol juice
      - D = desired Brix
      - A = initial Brix
      - C = Brix of water/concentrate
- Saignee
- Remember changing in some cases more than Brix - TA, pH, YAN
  - Retest after any adjustments

Brix adjustments

- Error in calculation
  - Accuracy of Brix determination depends on homogeneity of grapes
    - Influenced by grape ripeness
    - Dehydrated and raisoned berries

pH

- **Why is knowing the pH important?**
  - **Microbial stability (pH < 3.7)**
  - **Effectiveness of SO_2**
    - **Molecular SO_2 is the form effective against microorganisms**



pH

- Juice pH levels range from 2.8 to 4+
  - *Saccharomyces* optimal pH of 6, but grows well below and above this value
  - pH > 3.5 favors acetic acid bacteria and *Oenococcus* (LAB)
  - pH > 3.6 enables growth of larger diversity of LAB

pH

- Mouth-feel ("flabby") – wine style
- Color of red wine
- High pH ↑oxidation risk

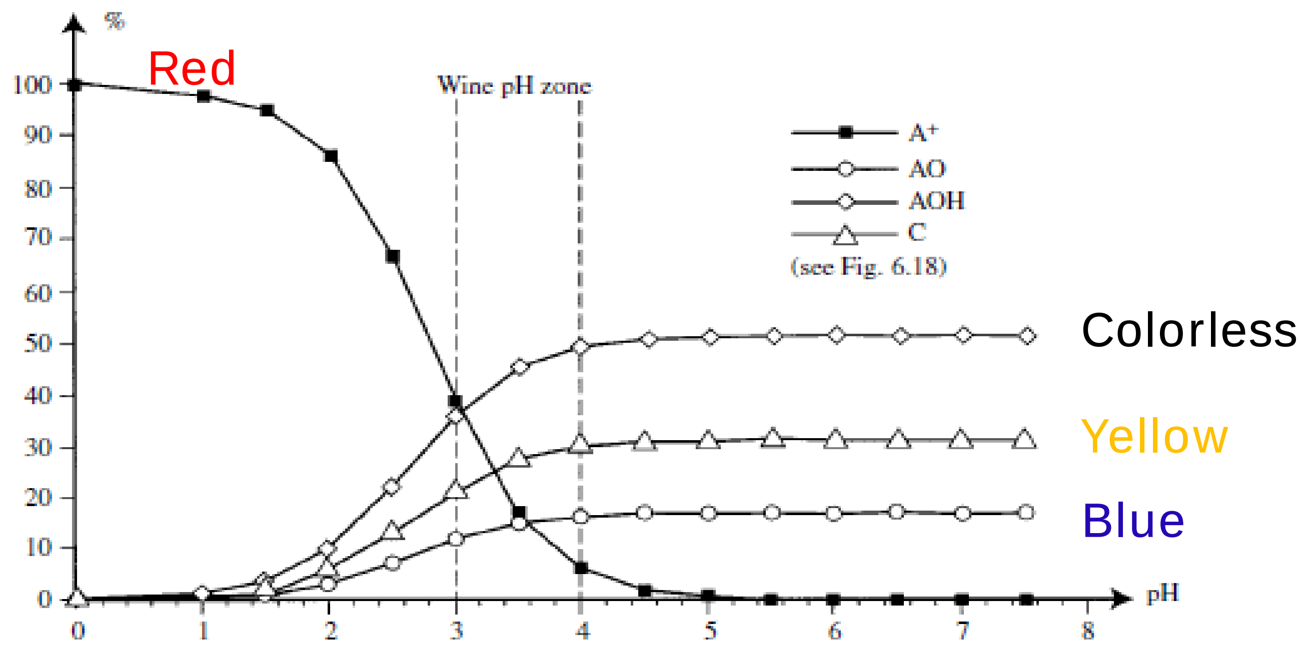


Fig. 6.19. Changes in the proportion of different forms of anthocyanins according to pH: $pK_a = 3.41$, $pK_h = 2.93$, $K_t = 0.61$ (Glories, 1984)

Titratable acidity (TA)

- Tartaric and malic acid are the main organic acids present in grapes
  - Citric acid is 3rd most prevalent
    - Rest formed during fermentation by yeast or bacteria
- Why is knowing the TA important?
  - Guide to acid taste of wine
    - Desired amounts depends on wine style
    - Wine balance
  - Adjust pH of juice/wine
  - How much to add to adjust pH?
    - Difficult to predict

Titratable acidity (TA)

- Adjust potential wine pH
  - Change in pH not directly related to acid addition
    - Depends on wine's buffer capacity
    - Rule of thumb: 1 g/L of tartaric acid, decrease pH by 0.1
    - Efficiency of acidification can improve with $CaSO_4$ addition
  - Add L(+)-tartaric acid
    - DL-tartaric acid addition increase calcium tartrate instability
  - De-acidify when pH <2.9 + TA > 10 g/L
    - Add 1.3 g/L of $KHCO_3$ – lower TA 1 g/L H_2T
  - Ion-exchange

Titratable acidity

- Measure malic acid (and lactic acid) conc
  - High malic conc will impact pH if planning on MLF
    - ↓ 1 g/L malic acid, ↓ TA 0.56 g/L and ↑ pH
  - Also if high conc of malic and very low pH
    - MLF at pH<3 difficult

Nitrogen (NH_3, NH_4^+ and amino acids)

- N_2 deficiency (< 100 mg/L)
  - Stuck fermentation
  - Utilization of sulfur containing amino acids – formation of H_2S
- Too much N_2
  - Modify aroma character
    - ↑Fusel alcohols, esters
  - Formation of ethyl carbamate
- N_2 needed
  - Depended on yeast needs and starting Brix
  - 21-27° Brix, need 200-350 YAN (mg N/L)

Yeast Assimilable Nitrogen (YAN) levels in juice

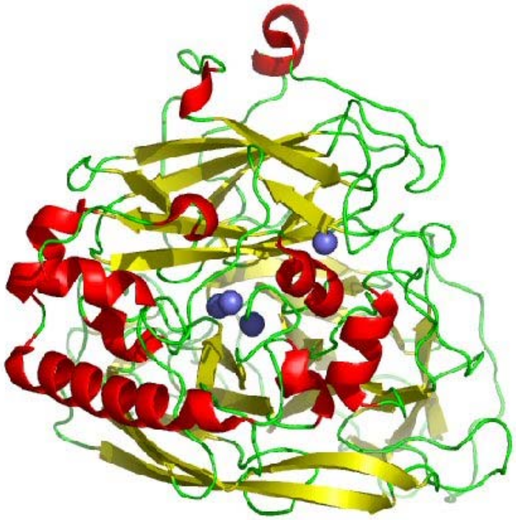
- Need to measure YAN of each fermentation
- Can vary greatly across fermentation lots even if from same vineyard
- YAN vary by season, varietal, rootstock and region

Adjusting N levels

- DAP (di-ammonium phosphate) most popular
  - Easy and cheap
  - Approximately 5 mg/L DAP for 1 N mg/L added
  - Adjustment needed before inoculation for cell growth
  - Any additional addition must be made before the half way point (< 8% alc)

Laccase activity

- **Why measure?**
  - **If Botrytis in the vineyard – knowing the number and potential risk**
  - **Better adaptation during the winemaking process**



So what is needed for a healthy fermentation?

- Yeast goal is survival
- Enable max cell growth
  - Meet nutritional needs
  - Minimize inhibitors
  - Minimize shocks like temp
- Sustain permissive conditions ~ minimize stress
  - Enhance ability to resist premature metabolic arrest

Yeast nutrition

- Growth is dependent upon available nutrients
- Insufficient nutrients causes:
  - Limit cell numbers (biomass)
  - Impact fermentation speed
  - Result in premature arrest of fermentation
  - Decrease ethanol tolerance due to increase in stress sensitivity
- Macronutrients are building blocks for new cell material
- Micronutrients catalyzes biochemical reactions

Yeast Nutrition

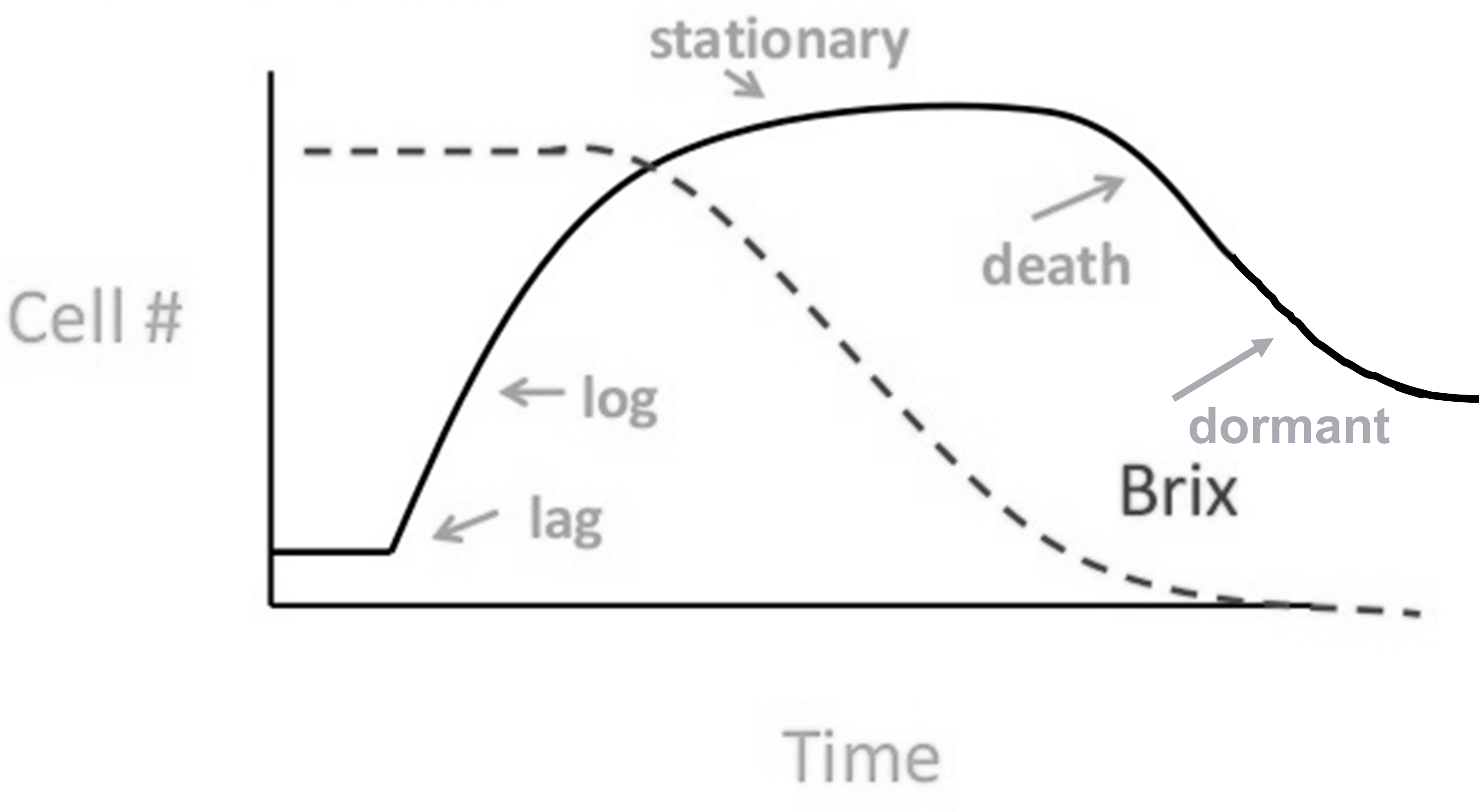
- Macronutrients are building blocks for new cell material
  - Carbon/energy sources - glucose, fructose, sucrose (never limiting)
  - Nitrogen Sources - amino acids, ammonia, nucleotide bases, peptides (often limiting)
  - Phosphate Sources - inorganic phosphate, organic phosphate compounds (occasionally limiting)
  - Sulfur Sources - inorganic sulfate, organic sulfur compounds (rarely limiting)

Yeast nutrition

- Micronutrients catalyzes biochemical reactions
  - Minerals and trace elements - Mg, Ca, Mn, K, Zn, Fe, Cu
  - Vitamins - biotin is the only required vitamin, but others are stimulatory
  - Sufficiency is site and varietal/rootstock dependent

UCDAVIS
UNIVERSITY OF CALIFORNIA

Yeast nutritional phases



Yeast Nitrogen Requirements

- Strain used
- Level of starting sugar/final ethanol
  - At ≤ 21Brix need starting YAN ~ 200
  - At > 25 Brix need starting YAN ~ 300
- Accompanying deficiencies
  - Presence of other microorganisms
- N needs increase with ↑alcohol
  - Decreased amino acid transport
  - Decreased ammonia uptake
- Stress can increase amino acid demand
-

Inoculation practices

- Active Dry Yeast
  - Rehydration is important
    - Can lose cytoplasmic components during rehydration
    - Will decrease viability if held too long prior to use
    - Water versus juice depends upon preparation: follow manufacturer instructions
  - Temperature equilibration is critical
  - Osmotic equilibration is critical for high Brix juices
  - Success of implantation depends upon conditions

Inoculation practices

- Starter culture – fermenting juice
  - May exacerbate nutritional deficiencies if juice is deficient
  - If too far along (too high in ethanol) return to high osmotic concentration poses a biological shock
  - Need to assess viability (stressors present)
  - Need to assess nature of organisms growing
- Starter success
  - Length of lag time before biomass production and fermentation initiation (↓2 Brix)

Use of sulfur dioxide

- It is an antioxidant
- Protects musts and wines from browning
- Binds oxygen and acetaldehyde
- Antiseptic activity
- Prevent microbiological spoilage in wines
  - Acetic acid and lactic acid bacteria, molds, wild yeast

Use of sulfur dioxide

- When to add SO_2
  - Crushing – amount depends on condition of grapes, temp and pH (50-80 mg/L)
  - Immediately after alcoholic fermentation
    - Amount based on wine style and variety
    - Usually aim for 30 mg/L free
- Other antimicrobials
  - Lysozyme
  - Heat treatments

Inhibitory levels of sulfur dioxide

Microbe	Molecular SO_2
Saccharomyces	0.825
Acetobacter	0.05-0.6
Lactic Acid Bacteria	0.01-0.2
Brettanomyces	0.1-0.6
Non-Saccharomyces yeast	0.1-0.6

Temperature control

- Important to control fermentation rate
  - Viability of yeast
    - For red wine heat accumulates in the cap
      - Temp gradients of up to 12˚C have been observed
      - Yeast growth from 12 to 42 ˚C (53 to 107 ˚F)
      - Temp tolerance reduced at high EtOH
      - Yeast strains vary in their ability to adapt to temp shifts
- Non-Saccharomyces yeast and bacteria
  - LAB grow at temp 18 to 48 ˚C (64 to 118 ˚F) but varies by strain

Temperature control

- High temp > 30 ˚C can stress yeast resulting in more off-flavors such as H_2S
- Flavor stripping can occur at high temp
  - Cooked fruit character
- Temp control by jacketed tanks or submerged cooling/heating units

UCDAVIS
UNIVERSITY OF CALIFORNIA

Temperature control

- **Optimum temp for white and red wine fermentation is different**
  - **Whites ≤ 15 °C (59 °F)**
    - **Preserve aroma and flavor compounds**
  - **Reds 20-30°C (68-86°F)**
    - **For extraction of color, phenolics and tannins**
    - **In Cab we found opt temp to be 28-30 °C**
    - **At 35 °C or 95 °F signf decreases in color**

Lerno et al., 2015 AJEV 66:4

Cap management

- Mixing of the cap for red wine fermentations
  - Remove accumulated heat from cap
    - Prevent yeast inhibition in the cap
    - Remove extraction saturation close to the cap
  - Studies show the 1 vol twice a day are adequate to remove concentration and temp gradients
  - Complex pump over regimes with different frequencies and volumes have limited impact on extraction

Sacchi et al. 2005 AJEV 56:3; Lerno et al., 2018 AJEV 69:3; Lerno et al. 2017 AJEV 68:4

Oxygen management

- Amount of oxygen depends on fruit condition
- White vs red grape processing
- Crushing and pressing - two processes with potentially signf O_2 exposure
  - As soon as berries are damaged, numerous oxidase enzymes including polyphenoloxidase (PPO) activates starting oxidative chain reactions which form quinones that can lead to browning

Oxygen management

- **Protective inert gas blankets and SO_2 have shown to preserve thiols and glutathione (GSH), a natural grape antioxidant**
  - GSH 51% in juice and 40% in skins
- **Hyper-oxygenation – 50 mg/L just before cold settling**
  - Protects against future browning
  - Lack of scientific study
  - Sensory of Chardonnay indicated ↑banana and ↓herbaceous and floral
- O_2 exposure during red grape processing is less of concern for healthy fruit

Day et al., 2015 AJGWR 21:693

Oxygen management

- White or red wine fermentation
  - Oxygen good for yeast viability
    - Lipid production needed for cell growth
    - O_2 addition at end of exponential growth phase – most effective stimulation of fermentation
      - 5-10 mg/L O_2 is sufficient (macro-oxygenation)
        - When combined with DAP addition, faster fermentation completion to lower RS
    - In non-Saccharomyces– a range of aeration regimes, applied continuously for various durations starting at inoculation
    - Do not know how laboratory studies translate to large scale
    - Translation to native fermentations?

Oxygen management

- Both yeast and bacteria grow better under aerobic environment
- Anaerobic conditions best to control acetic acid bacteria

Oxygen management

- Excess O_2 removed by CO_2 during fermentation
- Too little O_2 can result in reductiveness
- Too much O_2 can result in oxidation
  - Loss of varietal character for aromatic whites

Oxygen management

- Optimal use of O_2 can impact wine style greatly
  - Enhance fruit characters, limit reductive characters
  - Develop mouthfeel profiles
  - O_2 during fermentation can have the same impact as MOX following fermentation to remove reductive aromas
  - The optimal amount of oxygen during red winemaking without leading to excessive addition and concomitant spoilage still has to be determined

Concluding remarks

- Successful fermentations
  - Selecting correct yeast strain for EtOH range
  - Meeting nutritional needs of yeast
  - Limit stress and follow fermentation progress
  - Intervene early if not 'normal'
- Most off-flavors can be minimized or prevented by
  - Using clean fruit
  - Sufficient nutrient, oxygen and temperature control during fermentation
  - Good winery sanitation and adequate SO_2 use

Thank you