WINERY SANITATION

<u>Learning Outcomes:</u> This review covers practical issues regarding winery sanitation. This is an area of great importance, but is often over-looked. Sanitation, or lack of optimum sanitation, can have a significant impact on wine quality. This examination outlines the differences between cleaning and sanitation, and provides a list of advantages and disadvantages of various agents used in the wine industry. It also outlines some of the newer tools available to aid in sanitation, and stresses the importance of sanitation monitoring.

Chapter Outline

Introduction

Green Chemicals and Sustainability

Biofilms

Water Quality

Cleaning

Detergents

Sanitizers

Ultraviolet light (UVL) and Photon sterilization technology (PST)

Dry Ice Blasting and Ultrasound/HPU

Sanitation Monitoring

Practical Summary of Winemaking Issues

Section 1.

Introduction

Uncontrolled proliferation of microorganisms can lead to wine deterioration and spoilage. Winery Sanitation consists of two, and potentially, three levels of effort:

- Surface Cleaning: The cleaning of the surface to be sanitized and/or sterilized as warranted. Cleaning refers to the removal of mineral and organic material or debris from surfaces.
- Surface Sanitation: Sanitizing is the reduction or elimination of the viable cell populations to acceptably lower numbers which may, depending upon agents used and conditions of application, be zero.
- Sterilization is 100% kill (or removal) of viable cells and associated spores. This level of treatment is usually restricted to bottling lines.

Combined, cleaning and sanitation not only eliminates or reduces viable cell populations but hospitable environments (grape "bloom," tartrates, biofilms) for growth. Depending upon the stage in processing, the winemaker may want to sterilize the equipment.

Green Chemicals and Sustainability

There is a justifiable concern regarding both energy and water utilization in the winery. As outlined in the module titled Sustainable Winery Design, winery energy and water use can be substantial. In a study done on the West Coast, winery water use was as follows:

- wine push water 34%
- sanitation 25%
- refrigeration 11%
- landscaping 30%

With regard to winery sanitation, several practices are being implemented to help lower winery water use and biological oxygen demand (BOS) of wastewater, including the following:

- CIP (clean in place), pigging systems
- capture solutions, storage tanks and solution recovery
- nanofiltration
- green chemistry solutions
- rainwater harvesting

Each winemaker should be asked the question, “How many times do you use your water?” Even in regions like Virginia, where water is not a limiting factor, water should be recycled when possible.

CIP can be automated and allows for a significant reduction in solution volume. This, coupled with nanofiltration, allows for the recycling of solutions, significantly lowering winery wastewater volumes. Recycled water can be monitored a number of ways, including by measuring the NTUs. Water recycling aids in reducing the winery wastewater BOD (see Sustainable Winery Design module).

In many wineries, there is a move to eliminate not just chlorine due to possible environmental taints (see module on Environmental Taints), but also many salts, including sodium. Such reductions significantly lower wastewater BOD. Chemicals, such as hydrogen peroxide products, are gaining favor because they break down completely without negatively impacting the BOD.

Chemical additives for cleaning and sanitization are a primary source of “problem” ions such as sodium in winery wastewater, apart from product (wine) losses. The concentration of these ions has potential implications for wastewater re-use, particularly regarding the sustainability of irrigation practices using wastewater with elevated salinity concentrations. Alternatives to caustics (sodium hydroxide) have been introduced to improve the quality of wastewater. These include the use of hydrogen peroxide, rather than ozone or chlorine dioxide. The use of potassium hydroxide (pH 11.5) and potassium bisulfate (pH 2.5) with hydrogen peroxide is now a common replacement for sodium salts.

Biofilms

Biofilms are well-known to form when microbial populations in liquids/slurries adhere/attach to solid surfaces. Resulting from microbially-produced extracellular polysaccharides, biofilm formation confers increased colony resistance to detergents. This resistance can be 1,000 times greater than the resistant to chemical cleaners and sanitizers (Kumar and Anand, 1998). The physical properties of biofilms make them difficult to remove using to routine sanitation efforts.

Biofilm-forming species include yeast (*Brettanomyces bruxellensis)* and bacteria (*Oenococcus oeni)* found in wine (Nel et al., 2002). About 50% of species studied were found to form biofims (Joseph and Bisson 2004).

- The effectiveness of a sanitizing agent is dependant, to a large degree, on how well it breaks down biofilms.
- Sanitation cycles using caustics (75°C/167°F for 30 minutes), followed by citric acid rinse have proven successful in removing biofilms (Parkar et al., 2004).

Water Quality

Water is important to most detergent/sanitizer formulations and, thus, consideration should be given to its chemical as well as sensory properties. Water contains varying amounts of calcium, magnesium and other alkali metals, that, collectively, contribute to “hardness.” The United States Geological Survey definition for hardness levels is shown in Table 1.

Hard water interferes with the effectiveness of detergents, particularly bicarbonates, and contributes to precipitate or "scale" formation on equipment. Such precipitates serve as sites for accumulation of organic debris and microorganisms and thus makes sanitation more difficult. With time, these may also damage stainless steel surfaces.

Table 1. USGS Definitions for Water Hardness

Hardness Designation	**Parts per Million (ppm)**	**Grains per Gallon (gpg)[a]**
Very Hard	> 180	> 10.5
Hard	120 - 180	7.0 - 10.5
Moderately Hard	60 - 120	3.5 - 7.0
Soft	0 - 60	0 - 3.5

[a]gpg x 17.1 = mg/L (ppm)

Hard water is best addressed by installation of a water softener. Although detergent formulations may include chelating agents that help mitigate the problem, there extensive use is often more expensive than initially treating the water. Water temperature is important with respect to scale formation. Maximum scale formation occurs at 82°C (180°F). With soft water, this is not an issue.

It is recommended that winery water be analyzed regularly depending on the source (well or city). Water testing should include pH, alkalinity, calcium, hardness, iron, silica, total dissolved solids, and a standard plate count for microorganisms. It is not uncommon for winery to water to have a high calcium content that can cause the formation of insoluble calcium tartrate crystal formation post-bottling. Additionally, winery well water may contain enologically-viable microorganisms, including yeasts.

Cleaning

It is necessary to remove as much of the visible debris as possible before using detergents. This can be accomplished either manually or by automated cleaning systems that apply low volumes of warm (38°C/100°F to 43°C/109°F) water at high pressure (600 to 1,200 lb/in^2). Some considerations regarding cleaning include the following:

- Application of hot water to remove organic materials may "cook on" the debris, thereby requiring greater effort and costs to effect removal.
- Proper clean increases the effectiveness of sanitizing agents and lowers the amount needed.
- Avoid scratching stainless steel surfaces. Because of the protective oxide coating on stainless steel, only soft–bristle brushes should be used in cases where scrubbing is required

Detergents

Once visible debris and film has been removed, detergents are used to solubilize any remaining deposits. Each detergent has unique properties of action, as well as formulation, that may make it more appropriate for one application versus another. Generally, increasing the concentration beyond recommended levels provides little additional benefit and is not cost effective.

Warm (38-43°C, 100-110°F) water delivered at high pressure improves cleaning operations while decreasing the time required. It is best to direct the spray at an angle to the surface being cleaned. To avoid violent reaction during formulation, dry chemicals should always be added to cold water rather than to hot. When it is necessary to apply cleaning agents hot, it is recommended that the mixture be heated after formulation.

Alkalies

Strong alkalies, including NaOH (caustic soda or lye) or KOH (caustic potash) are the most commonly used detergents. Other alkali compounds also find application in the winery. These include sodium *ortho*– and *meta*–silicates (Na_2SiO_3). Although less caustic than NaOH, the silicates possess better detergent properties, and are less corrosive towards equipment.

- Alkalies have excellent detergent properties and are strongly antimicrobial.
- Alkalies may be corrosive, even to stainless steel, if recommended application levels are exceeded.
- Handling strong alkalies requires use of personal protective equipment.

Where the relative amount of organic material is not heavy, mild alkalies such as sodium carbonate (soda ash) or trisodium phosphate (TSP) find application. Although relatively inexpensive, frequent use of sodium carbonate (Na_2CO_3) in hard water may contribute to precipitate formation and build-up.

Sequestering Agents

Many detergent formulations include chelating agents that “soften” water by binding with calcium and magnesium. Polyphosphates are widely used and include sodium hexametaphosphate (e.g., Calgon®) and sodium tetraphosphate (Quadrofos). Another chelating compound used is ethylenediamine tetraacetic acid (EDTA). While more expensive than polyphosphates, EDTA has the advantage of being relatively heat stable.

Surfactants

Detergent formulations often contain surfactants which act at the boundary between aqueous and organic phases, facilitating contact between the detergent and the surface being cleaned. One end of the surfactant molecule is compatible with water (hydrophilic), the other end with oil (hydrophobic, sometimes also called lipophillic). Physically, surfactants remove organics by surrounding the molecule, thereby lifting debris and microorganisms from the surface (Wirtanen and Salo, 2003).

Various anionic, nonionic and cationic surfactants are available. Nonionic surfactants have the broadest range of properties since these can be either wetting or emulsifying agents depending on their chemical structure. Nonionic surfactants also vary in their ability to foam, a characteristic that can be important for cleaning some pieces of equipment.

Acids

Acids are used in specialized detergent formulations (at approximately 0.5%) to reduce mineral deposits and soften water. Maximum effectiveness occurs at pH 2.5. At low pH, acid solutions are very corrosive toward stainless steel (and other metals). Among these, phosphoric acid is preferred because of its relatively low corrosiveness and compatibility with nonionic wetting agents also present in the formulation. Other mineral acids (i.e., nitric) are also used but these tend to be more corrosive to metal equipment.

Rinses

Once the cleaning cycle using detergents is completed, equipment surfaces should be thoroughly rinsed to remove residual chemicals. A mild acid (e.g., citric) is often incorporated into the rinse to neutralize alkaline detergent residues.

A sensory evaluation of the rinse water helps assure the winemaker that there are no latent aroma/flavors that could be incorporated into the wine.

Sanitizers

Once surfaces are visibly clean, tanks and equipment can be sanitized. Sanitizing agents include the following:

- halogens (chlorine, iodine and bromine-containing agents)

- quaternary ammonium compounds (QUATS)
- acidulated sulfur dioxide
- hot water
- ozone
- peroxides

Table 2 compares commonly-used chemical sanitizers with respect to their relative advantages and disadvantages. Like caustic detergents, sanitizing chemicals represent health and safety concerns and, thus, winery staff should be thoroughly trained in their use as well as the importance of wearing personal protective equipment. Close attention should be paid to manufacture's and supplier's recommendations regarding preparation and application. Deviation from prescribed and approved formulation is not only a safety concern, but may be a regulatory violation.

- Once the sanitation process has been completed, surfaces are rinsed to remove sanitizer, drained and tested (chemically, and/or sensorially) for residual sanitizer.

Chlorine, Iodine, and Bromine-Containing Agents

Historically, the two most important halogens formulated for use in sanitation were chlorine, and chlorine-based agents, as well as Iodine (iodophors). More recently, bromine-containing agents are being utilized for sterilization.

Chlorine. Because of chlorine's role as a precursor in microbial formation of chloroanisoles, its use is not recommended (see module on Environmental Taints).

One exception that continues to find interest and application is the gas chlorine dioxide (ClO_2). Chlorine dioxide has a long history as a biocide and sterilizing agent. Chlorine dioxide has the following properties:

- Like ozone, chlorine dioxide is an oxidant. Because ClO_2 cannot be compressed and stored as a gas, it is can be generated, as need, on-site by reaction of sodium hypochlorite with an acid. Small wineries using chlorine dioxide usually use commercially available tablets.
- Chlorine dioxide is approximately 10 times more effective than chlorine as a sanitizer.
- Unlike other forms of chlorine, ClO_2 does not hydrolyze in aqueous solution; it remains as a highly reactive free radical.
- Compared to chlorine where the "active" forms are influenced by pH, the bactericidal impact of ClO_2 is unaffected at pH levels between 4 and 10.
- The contact time required for ClO_2 is less than for chlorine.
- Chlorine dioxide actively degrades the polysaccharide matrix of biofilms.
- At concentrations required for disinfection, chlorine dioxide is not corrosive.
- Given its volatility, the use of ClO_2 in confined spaces or poorly-ventilated areas raises significant health and safety concerns. Like chlorine, inhalation of chlorine dioxide may cause nose, throat, and lung irritation. Chronic exposure to ClO_2 may cause bronchitis. The exposure limit for chlorine dioxide is 0.1 ppm (ATSDR, 2004).
- When chlorine dioxide concentrations reach 10% or more in air, the gas becomes explosive.

Iodine. Formulations including iodine, acid (commonly phosphoric acid) and nonionic wetting agents are called iodophors. Iodophors are most effective at <pH 4, where the concentration of I_2 is maximum. Iodophor product labels normally have a "titratable iodine" statement, which represents the minimum amount of iodine available in the product.

- Iodophors are broad spectrum, with demonstrated effectiveness against a variety of bacteria, yeasts, and molds.
- Iodophors are effective at relatively low concentrations.

- I_2 volatilizes at >49°C/120°F, so formulations are not compatible with hot water applications.
- Iodophores are frequently used for bottling line sanitation, followed by a cold-water rinse.
- Formulations containing iodophores may foam excessively and may stain polyvinylchloride and other surfaces.
- Residual iodophore can be detected using inexpensive test strips.
- To date, there are no data indicating the involvement of iodine-containing compounds in formation of wine aroma/flavor taint.

Quaternary Ammonium Compounds

Quaternary ammonium compounds (QUATS) have the basic structure of a nitrogen covalently bound to four alkyl or aromatic groups.

- QUATS are surface-active agents that function by disrupting microbial cell membranes.
- The group exhibits differential activity toward microbes, with Gram-positive bacteria such as (LAB) most affected, whereas Gram-negatives (*Acetobacter*) are less impacted. The agents have no activity against bacteriophage (viruses that attack bacteria).
- QUATS have extended activity over a broad pH range.
- They have additional advantages of being heat stable and noncorrosive and, if not rinsed away, have residual activity.
- In addition, activity is not compromised by hard water or poorly-prepared surfaces.
- At typical application levels of 200 to 400 mg/L, a thorough post-application rinsing is required.
- QUATS find application in controlling mold growth on walls and tanks. The formulation is sprayed on the surface and left without rinsing. Depending on environmental conditions and extent of mold growth, a single application may last for several weeks.