

Wine Yeast*

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It is often said that a "wine is made in the vineyard" or "you can't make a silk purse out of a sow's ear." These comments emphasize the point that good quality fruit is needed for good quality wine. Although the importance of quality grapes in making superior wine cannot be denied, it is equally important to realize that it is one of the critical components but not the only component necessary to make good wine. There are many other factors that are vital to wine quality, otherwise, how can one explain poor quality wine from high quality grapes?

Fermentation is the key process that transforms grapes into wine. In addition to "ideal fruit composition," a successful fermentation depends on (1) yeast, and (2) vinification conditions. A sound understanding of these factors and their skillful manipulation is essential if one is to achieve a successful fermentation and consequently, a better wine.

Ecology and fermentation

Wine yeasts are a group of fungi that are predominantly uni-cellular (one cell) organisms which are widely distributed in nature. They are fermentative organisms that can metabolize a variety of sugars. Many types of yeasts naturally occur on the surface of grapes. Some of the commonly found yeasts on grapes and in wines belong to the following genera: *Saccharomyces*, *Kloeckera*, *Hanseniaspora*, *Candida*, *Hansenula*, *Pichia*, and *Brettanomyces*.

It is commonly believed that in a natural or spontaneous fermentation non-*Saccharomyces* yeasts such as *Kloeckera*, *Hansenula*, *Candida* and many others grow and participate in earlier stages of fermentation. As the fermentation proceeds the ethanol concentration increases. This (high ethanol) limits the growth and activity of native non-*Saccharomyces* yeasts thus creating a condition favorable to the growth and domination of native *Saccharomyces* yeast, which then conducts the fermentation. Advocates of natural fermentation believe that a mixed culture fermentation produces more complex flavors in wine. Other winemakers find natural fermentations unpredictable and prone to the development of off odors. For these and other reasons mentioned later, they prefer to use pure yeast strains in the form of a "starter culture" or as an active dry wine yeast in order to conduct the alcoholic fermentation.

Inoculation of a must by pure yeast strains assumes that the fermentation will be carried out by the inoculated yeast. However, recent research indicates that in many cases, non-*Saccharomyces* and native *Saccharomyces* strains also contribute (in various degrees) to the fermentation, even though a pure strain is added to the must. The degree to which native yeasts contribute to the fermentation varies greatly. Certain vinification variables such as the amount of inoculum, the fermentation temperature, and must clarification can be manipulated to diminish the role of native yeast in fermentation. However, to what extent these factors are effective in reducing the contribution by indigenous yeasts in the fermentation is not clear.

It is reasonable to assume that in many cases native yeast strains do participate and contribute to the alcoholic fermentation, even if the must has been inoculated with a pure starter culture.

It appears that with the present state of our knowledge one can either choose a natural fermentation (mixed culture) or a predominantly (but not absolutely) pure culture fermentation. It should be noted that spontaneous natural fermentations have largely been replaced by pure culture fermentations in many wine regions of the world.

Using pure yeast culture

The benefits of using a pure yeast culture to conduct an alcoholic fermentation are widely recognized. The pure yeast is used as a liquid yeast starter culture or as an active dry wine yeast. The preparation of a liquid yeast starter culture requires an investment in special equipment, is labor intensive, and may not be convenient for a small scale winery operation. For these reasons the use of pure yeast in the active dry form has become very common in the wine industry.

There are many advantages to using active dry wine yeast:

- (1) rapid onset of fermentation,
- (2) better fermentation control, thus permitting more efficient use of fermentors,
- (3) consistent and reproducible wine quality,
- (4) reduced possibility of off flavor formation and stuck fermentation, and
- (5) taking advantage of certain yeast characteristics to obtain specific results (for example, de-acidification using 71B yeast).

Due to the advancement in the technology of yeast production, many strains with different desirable properties are commercially available. A list of some of the yeast suppliers is given below:

1. Cellulo Co., 2949 E. Townsend Ave., Fresno, CA 93721. (209)485-2692.
2. Scott Laboratories, 2220 Pineview Way, PO Box 750249, Petaluma, CA 94975. (800)821-7254.
3. Universal Foods Corp., 433 East Michigan St., Milwaukee, WI 53201. (414)347-3886.
4. Vinqury, 16003 Healdsburg Ave., PO Box 695, Healdsburg, CA 95448. (707)433-8869.
5. The Wine Lab, 477 Walnut St., Napa, CA 94559. (707)224-7903.
6. Presque Isle Wine Cellar, 9440 Buffalo Rd., North East, PA 16242. (800)488-7492.

NOTE: Providing the names of the suppliers listed above does not imply their endorsement.

To obtain the best results from the use of active dry wine yeast, a good understanding of the rehydration procedure (technique) and inoculation method is essential.

The active dry wine yeast is prepared from a pure culture strain having a proven track record of producing quality wine. Before drying, the desired yeast is first propagated under special conditions. During this phase yeast is supplied with adequate nutrients and oxygen. The aeration (oxygen exposure) is important since it promotes sterol synthesis. Sterols such as ergosterol play a vital role in semi-permeable membrane function. At the end of the growth stage the healthy yeast cells contain adequate amounts of protein, ergosterol and unsaturated fatty acids in the cell membrane. They also contain glycogen and trehalose which serve as a source of energy for growth and survival. During the drying process the moisture content of a yeast cell is reduced from about 70% to about 8 percent. The dry yeast is then tested for its quality. The quality assurance tests include checking for the following items:

- (1) percent solids, proteins and phosphorus
- (2) fermentation activity
- (3) stability
- (4) total viable population
- (5) total bacteria
- (6) wild yeast count
- (7) presence of a killer factor

After the quality of the dry yeast is assured, it is packaged under vacuum or nitrogen. Since the microbiological quality of the dry yeast can vary depending on the source, it is important that only the highest quality product be used in conducting a fermentation.

Rehydration of active wine dry yeast

In order to resume the fermentative activity, the moisture content of a dried yeast cell must be restored. This is best achieved by following a proper rehydration procedure. When a yeast cell is rehydrated, it rapidly (within seconds) takes up water. At the time of rehydration the cell membrane is permeable and improper rehydration can cause leaching of valuable cell constituents. This in turn results in the loss of cell viability and leads to poor fermentation activity. Rehydration also affects the physical dispersibility of the yeast. When poorly dispersed, clumping of the yeast cells occur and the cell viability is reduced. For the reasons given above it should be obvious that proper rehydration of the dry yeast is vital to ensure a sound fermentation. Monk (1986) has suggested the following procedure to properly rehydrate the must:

1. Use water in the amount of 5 to 10 times the weight of the yeast. For example, for 500 grams of dry yeast use 3 to 5 liters of water for rehydration.
2. Rehydrate in warm water at 40-45°C (104-115°F).
3. Slowly add yeast to water to obtain even rehydration. Do not add water to yeast, this will cause clumping.
4. Allow yeast to remain in warm water for 5 to 10 minutes before stirring.
5. Do not leave yeast in water for more than 30 minutes. Longer duration will reduce activity.
6. Do not add rehydrated yeast to cold must. The temperature difference between the yeast starter and the

must should not be more than 10°C.

7. To reduce the possibility of cold shock, gradually cool the starter and then add it to the must.

8. Preferably, use warm water rather than must to rehydrate the yeast.

Although the above rehydration procedure will give successful results, it is important to check the manufacturer's directions on the rehydration procedure.

Inoculating the must

In white wine production the must is chilled and clarified to reduce the suspended solids content. It is then held at a lower temperature until inoculation. Some winemakers may prefer to ferment their must below 15°C (59°F), but in such a case the inoculation should not be done below 15°C (59°F). It is desirable to add a yeast starter at a higher temperature (above 15°C), then slowly lower the must temperature to the desired level to conduct a controlled low temperature fermentation. To avoid cold shock to the yeast and obtain a sound fermentation, Monk (1986) suggested slowly cooling the starter to 20°C (68°F), then adding it to the white must at a temperature no lower than 15°C (59°F). In a laboratory trial we were able to lower the temperature of a starter culture to 24°C (75.2°F) in 30 minutes by using the following procedure:

- a. The must was rehydrated at 40°C (104°F) and held at room temperature (75°F) for 15 minutes.
- b. The starter culture was then placed in the refrigerator (about 40°F) for another 15 minutes.
- c. At the end of 30 minutes, the temperature of the starter culture was lowered to 24°C (75.2°F).

It would be possible to further lower the temperature of the starter culture to a suggested level of 20°C (68°F) by extending storage time in the refrigerator. This would obviously bring the starter culture temperature close to the must temperature (59°F) and minimize the possibility of cold shock.

At this point, the effect of cooling the starter culture on cell viability and the ensuing fermentation is not clearly known. However, considering the importance of avoiding cold shock, such a procedure would be acceptable.

In red wine production the must is usually fermented at a higher temperature (25-30°C or 77-86°F). Therefore, inoculating with yeast at the proper must temperature can be easily managed.

To ensure the rapid onset of fermentation, the yeast population in the must at inoculation should be 5 million cells/mL (5 x 10⁶ cells/mL). In a high sugar or difficult-to-ferment must, inoculating the must with even higher populations should be considered. To achieve a cell density of 5x10⁶ cells/mL, the addition of about 25g of reconstituted yeast per 100 liters is suggested. During the fermentation, the yeast grows under anaerobic conditions and with every generation the yeast population doubles. This multiplication of yeast usually occurs for four to five generations. Inoculation at 5 million cells/mL will grow to 160 million cells/mL in five generations. This assumes that ideal conditions for yeast growth exist. However, in reality, the conditions for yeast growth are less than ideal. High sugar, low pH, ethanol, SO₂ and low temperature, all limit yeast growth; consequently, the cell population is usually below the 160 million cells/mL level.

Some winemakers use fermenting must to inoculate other musts. This practice (pitching) is not recommended and should not be followed. When using fermenting must, one should realize that the yeast cells have gone through several generations (cell divisions or budding). With each generation, the survival factors (sterols and fatty acids) are reduced. The yeast cells with lower amounts of survival factors may cease to grow before all the sugar is used. This will result in an incomplete or a stuck fermentation.

Also, if the fermenting must is contaminated, then the contamination will spread to other inoculated must. Considering the cost of other ingredients, yeast is relatively inexpensive and only high quality yeast in the amount and manner recommended should be used. In one case (not in Missouri), a winemaker inoculated by simply sprinkling the dry yeast on the surface of the must. As one may expect, this approach would lead to poor rehydration and dispersion. Both of these points are critical when using dry yeast. Needless to say, this approach is not recommended.

Yeast storage

The yeast loses its activity during storage. For this reason only enough yeast to last through the season should be purchased. Surplus or leftover yeast should be stored dry at 4°C (39.2°F), preferably under vacuum or anaerobic conditions. At 4°C the yeast loses activity at a rate of about 5% per year. At higher temperatures, e.g., 20°C (68°F), yeast activity is reduced by 20% in one year.

Reference:

1. Monk, P.R. 1986. Rehydration and propagation of active dry wine yeast. Australian Wine Industry J. 1:3-5.

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