# NUTRIENTS FOR ALCOHOLIC FERMENTATION

## What, when, and how – Guidelines for North America

This document was updated in the spring of 2002. The field of alcoholic fermentation nutrition is dynamic. Please check www.lallemandwine.com for the latest information.

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## Recommended Addition Rates and Feeding Schedule

### (For 22° brix must)

| Initial Must YAN (Yeast Assimiliable Nitrogen) | | At Active Dried Yeast Rehydration | At End of Lag Phase (around 6-12 hours after inoculation) | At 1/3 Sugar Depletion |
|---|---|---|---|---|
| High | >225 mgN/L | GO-FERM 30g/hL (2.5 lb/1,000 gal) | | |
| Medium | 125–225 mgN/L | GO-FERM 30g/hL (2.5 lb/1,000 gal) | | Fermaid K 25g/hL (2 lb/1,000 gal) |
| Low | <125 mgN/L | GO-FERM 30g/hL (2.5 lb/1,000 gal) | Add DAP to bring total YANC to approx. 150 mgN/L | Fermaid K 25g/hL (2 lb/1,000 gal) |

*Note: The above chart has been developed on the basis of treating 22°Brix must and should be adjusted to reflect the actual Brix.*

*In addition to Brix, there are other factors that affect the yeast strain's ability to uptake nitrogen including pH, temperature and osmotolerance.*

Adjustments for high Brix musts (above 25°Brix) can be successfully accomplished in three ways.

1. Lallemand recommends inoculating with 25 g/hL (2 lb/1,000 gal) of properly rehydrated and handled yeast in low to medium sugar must concentrations. When sugars are above 25°Brix, we recommend increasing the inoculation rate to 35 g/hL (2.8 lb/1,000 gal). Starting at a higher inoculation level will yield higher initial viable cell counts having a direct effect on the maximum cell density achieved at the end of the exponential growth phase. This higher cell density is needed in order to successfully convert all sugar to alcohol during the course of the fermentation.

2. Select a yeast with lower relative nitrogen demands as well as higher alcohol tolerance (i.e. DV10, QA23, or T73). For more information, please refer to the Yeast Quick Reference Chart and Yeast Strain Descriptions on www.lallemandwine.com .

3. In high sugar must (above 25°Brix), an alternative to increasing the inoculation rate or selecting another yeast is to add additional DAP at 1/3 sugar depletion. In most musts, low or high sugar, a total of 150 mgN/L is a sufficient amount of nitrogen to take the yeast through the exponential growth phase. It is during the stationary phase when the yeast in high sugar conditions benefit from an additional boost of nitrogen to avoid sluggish fermentations. For this reason, in high sugar must (25°Brix), follow the same recommendations outlined in the above chart with two modifications. Add an additional 25g/hL (2lb/1,000 gal) of DAP at 1/3 sugar depletion when YAN is below 225 mgN/L, and add Fermaid K at 1/3 sugar depletion to any must under 300 mgN/L YANC.

# The Most Important Wine Yeast Nutrients – What they do and when to add them

## GO-FERM

Nitrogen deficiency in must is one of the most understood and easily corrected causes of sluggish and stuck fermentations. While researchers have determined that there are several key micronutrients for healthy yeast growth and survival, unfortunately many winemakers only address the importance of nitrogen. Lallemand's recent collaboration with the INRA in Montpellier confirms the critical role of yeast micronutrients but more importantly, it identifies the most effective way to ensure that these micronutrients benefit the selected yeast.

As a result of this recent research, Lallemand developed GO-FERM (patent-pending), a natural yeast nutrient to avoid sluggish and stuck fermentations that contains a balance of micronutrients formulated. GO-FERM is particularly effective for helping the yeast survive fermentation of high sugar musts.

GO-FERM is added to the rehydration water before adding the selected dried yeast. The yeast then acts like a "sponge" to soak up the bio-available micronutrients in GO-FERM critical for increased viability and strong fermentation finish. If these micronutrients were added directly to the must, competitive microorganisms would use a significant number of them and others would be chelated by polyphenols or inactivated by $SO_2$.

**NEVER** use nutrients containing ammonia salts such as DAP during yeast rehydration – they are not as effective and they are **toxic** to the yeast at high levels!

• Use 30g/hL (2.5lb/1,000 gal) GO-FERM when rehydrating yeast to supply critical micronutrients. *Note: This recommendation is based on a yeast inoculum of 25 g/hL (2 lb/1,000 gal). If using more or less yeast, respect a ratio of 1 part yeast : 1.25 parts GO-FERM.*

## Diammonium Phosphate (DAP):

DAP is the least expensive source of inorganic nitrogen allowed in wine ferments and an important contributor of Yeast Assimilable Nitrogen (YAN).

Nitrogen is an essential element needed to avoid sluggish and stuck fermentations. A nitrogen deficiency will lead to problems in four fundamental ways. The first three are related to each other as follows: (1) protein synthesis is limited; (2) then because the proteins are the bricks used to build new cells, cell numbers are limited; and (3) the fewer number of cells negatively affect the fermentation kinetics. The fourth way that nitrogen deficiencies can cause sluggish fermentations is through a decrease in the efficiency of the sugar transporters in the membrane causing a significant decrease in fermentation kinetics (Basturia and Lagunas, 1986).

Typically grape musts contain somewhere between 80 and 400 mg/L of yeast assimilable nitrogen. High sugar must (above 25° Brix) are considered deficient if they have less than 225 mgN/L (Ortiz-Julien and Sablayrolles, 2000). In general, DAP additions will reduce the development of sulphur compounds, however, excessive DAP additions can actually lead to high levels of sulphur taint, Watson, 2000); harsh characters in wine (Delteil, 1997) and can even burn the fermentation out. These problems are especially evident when DAP additions are not supplemented with complex nutrient formulations such as Fermaid K and GO-FERM.

### USE DAP in 22°Brix must when FAN levels are below 125 mgN/L

• Add 30 g/hL (2.5lb/1,000 gal) GO-FERM during rehydration. *Note: This recommendation is based on a yeast inoculum of 25 g/hL (2 lb/1,000 gal). If using more or less yeast, respect a ratio of 1 part yeast : 1.25 parts GO-FERM.*

• Add enough DAP at the end of lag phase (beginning of alcoholic fermentation) to bring the FAN to approximately 150 mgN/L.

• Add 25 g/hL (2lb/1,000 gal) Fermaid K at 1/3 sugar depletion.

• When YAN levels are below 225 mgN/L and Brix levels are high (25° Brix), add an additional 25 g/hL (2 lb/1,000 gal) of DAP at 1/3 sugar depletion.

## FERMAID K

FERMAID K is a blended complex yeast nutrient developed by Clayton Cone and Cliff Caron of Lallemand. Fermaid K supplies inorganic nitrogen (DAP), organic nitrogen (alpha amino nitrogen derived from yeast extract), key nutrients (magnesium sulfate, thiamine, folic acid, niacin, biotin and calcium pantothenate) and inactive yeast.

The best time to add Fermaid K is at 1/3 sugar depletion. This timing corresponds to the end of exponential growth and the beginning of the stationary phase. With proper rehydration and handling, the yeast cell population should be at least 4-million cells/mL at inoculation. At the time we recommend Fermaid K addition, this population will have hopefully grown to about 100-million cells/mL. It is critical to remember that yeast are still reproducing during the stationary phase

and to do this effectively, especially in high sugar musts, they need nitrogen, oxygen, sterols, fatty acids and micronutrients.

At 1/3 sugar depletion:

• Nitrogen is needed for protein synthesis and to maintain cellular growth. Nitrogen from alpha amino acids coming from Fermaid K is utilized significantly more efficiently than inorganic nitrogen (DAP).

• Fermaid K also provides unsaturated fatty acids and sterols important to maintain alcohol resistance and permease activity.

• Micronutrients in Fermaid K help the yeast maintain their cellular integrity throughout the remainder of the fermentation.

• The cell wall fractions in Fermaid K absorb medium chain fatty acids that are toxic to yeast as well as provide nucleation sites to help keep the yeast in suspension.

### Use Fermaid K in 22°Brix must when YAN levels are below 225 mgN/L or in 25°Brix must when YAN levels are below 300 mgN/L

• Add 30 g/hL (2.5lb/1,000) GO-FERM during rehydration. *Note: This recommendation is based on a yeast inoculum of 25 g/hL (2 lb/1,000 gal). If using more or less yeast, respect a ratio of 1 part yeast : 1.25 parts GO-FERM.*

• If needed, supplement with DAP at the end of lag phase (beginning of alcoholic fermentation) to bring YAN to 150 mgN/L.

• Add 25 g/hL (2 lb/1,000 gal) Fermaid K at 1/3 sugar depletion.

# Determining the Nitrogen Content of Must

Yeast Available Nitrogen (YAN) is composed of two portions: (1) organic or assimilable amino nitrogen and (2) inorganic nitrogen (ammonia). When determining the nitrogen concentration in must it is critical to take both of these portions into account. The best fermentations contain a balance of yeast available nitrogen from both assimilable amino nitrogen and inorganic nitrogen (ammonia).

### Approximate YAN Contribution for the important yeast nutrients

| Nutrient | 12.5 g/hL (1 lb/1,000 gal) | 25 g/hL (2 lb/1,000 gal) | 30 g/hL (2.5 lb/1,000 gal) | YAN Source |
|---|---|---|---|---|
| DAP | 25 mgN/L | 50 mgN/L | 63 mgN/L | Inorganic nitrogen |
| Fermaid K | 12.5 mgN/L | 25 mgN/L | Exceeds ATF limits for thiamine addition | Inorganic nitrogen (from DAP) and organic nitrogen (alpha amino nitrogen) resulting from autolyzed yeast |
| Go-Ferm | | | 10 mgN/L | Only organic nitrogen resulting from autolyzed yeast (micronutrient carrier) |

### Determining Assimilable Amino Nitrogen (Organic Nitrogen)

There are two popular methods for determining organic nitrogen, formol titration and the o-phthaldialdehyde/N-acetyl-L-cysteine spectrophotometric assay (NOPA) method (Dukes and Butzke,1998). The two methods of analysis yield similar results (Shively and Henick-Kling,2001).

• Formol Titration:

This method only requires a pH meter, reagents and titration capabilities. The drawbacks are that it is time consuming and uses formaldehyde. Formaldehyde is difficult to purchase in small quantities, requires proper handling and hazardous disposal.

• NOPA:

This procedure uses an UV spectrophometer. The sample is run against a standard curve based on known reagents. The reagents have a short shelf -life and the standard curve needs to be established every time a group of samples are analyzed. Once the procedure is set-up, however, many samples can be run quickly.

## Determining Inorganic Nitrogen (Ammonia):

• Ammonia Probe:

The ammonia probe is attached to a special pH meter with ion specific capabilities. After standardization, the procedure is similar to reading the pH.

• Enzymatic Analysis:

There is an enzymatic reagent kit for use with a UV spectrophometer. If a winery has already made the investment for doing NOPA analysis then this may be the best test to use.

## Determining Yeast Assimilable Nitrogen (YAN)

Add the result obtained from the organic nitrogen analysis to results obtained from the inorganic nitrogen (ammonia) analysis and the result is the YAN.