# Vutrition i

ne of the most overlooked aspects of creating a sound, rapid and complete fermentation is the nutritional package needed to maintain healthy and viable yeast.

Developing a proper nutritional strategy is difficult and many questions result. What should I add? Do I really need it? What type of nutrition is best? What commercial nutrient packages are available? These are all valid questions.

Over the past few years of extensive growth in the ethanol industry, low input costs and particularly high revenue from ethanol and co-products were the norm. It was fairly easy to overlook overall plant efficiency or yield and focus on making the maximum number of gallons over a given time. This is generally regarded as throughput.

During this period of a throughput mindset, many plants operated well beyond design capacity with some wasted substrate but were still profitable. Now that economic times have changed, and the growth of the ethanol industry has slowed, the focus of plant operations have shifted to increased yield and efficiency.

With a yield-focused mentality, plants maximize the quantity of alcohol with minimal inputs. Plants are utilizing less corn (maize) and still producing large quantities of ethanol. They are investigating ways to improve efficiency and maximize total profitability. This lowering of inputs (primarily substrates and energy) with maintenance of good yields of final product has a huge impact on plant profitability in the economic hard times of today and the economic good times of yesterday.

# IMPORTANCE OF NUTRITION

The basic fermenter ingredients are grains, sugars or starches (which are, if necessary, broken down by enzymes into dextrins and subsequently into glucose), a diluent for proper dry solids requirements (combinations of thin stillage, process condensate and other recycled waters) and yeast. Other requirements include usable nitrogen, trace oxygen or essential fatty acids and sterol, vitamins (growth factors), and inorganic ions. During the fermentation process, yeast will then grow in this nearly anaerobic environment and usually multiply twenty-fold to approximately 2 x 10<sup>8</sup> cells for the most efficient fermentations.

Many of the grains used in ethanol fermentation contain nutrients needed for successful fermentation, but not all of the nutrients in the grain come in usable formats. Also, not all substrates are the same.

Healthy yeast can increase fermentation, yields and profitability by Cody Snyder and Mike Ingledew

There are many sources of grain — especially fractionated grain components — that have differing quantities of nutrients so the make up for optimal fermentation response will have to be determined. Incoming grain has a multitude of vitamins and minerals as well as a fair amount of useable nitrogen components. Many of the fresh water sources within the industry also contain minerals. However, all of these inputs do not provide consistent quantity, quality or availability of the specific nutrients needed for optimal fermentation performance. Yeast "foods" are therefore provided to assist the cellular reactions of energy production, cell growth, ethanol production, CO, production and glycerol production (oxidation/reduction balance) that occur in concert in the cell.

# **YEAST GROWTH**

Additional nutrients are important for optimal yeast growth. As stated above, yeast must multiply many times over to reach critical mass. Typical yeast growth has three phases. Growth starts in the lag phase as the yeasts acclimate to environment, while maximum growth occurs in the exponential phase. The growing yeasts produce ethanol 33 times faster than stationary phase yeasts. Nutrition is important during growth for maintaining a good start during the log phase and later, in the final stages

Usable nitrogen levels in mg/L mash from various grains normalized to 22% w/v dissolved solids

Mash	FAN Total	Usable FAN
Wheat	82	64
Barley	84	62
Hulless Barley	124	100
Oats	193	159
Hulless Oats	184	130
Rye	103	83
Molasses	267	141
Corn	70	58
Starch Slurry	~0	~0

Source: Ingledew, 2005

# Metallic ions

Ingredient	Importance	Optimal concentrations
Magnesium	Insulates the cell against stress factors of temperature, alcohol, and osmotic pressure	2-4 ppb
Zinc	Helps the cell produce fermentation enzymes. Lack can cause slow/sluggish fermentation and poor yield	4-8 ppb
Calcium	Helps stimulate cell growth and cell wall permeability	4-8 ppb
Copper	Assists with cells internal enzyme production	1-2 ppb
Potassium	Assists with storage of ATP inside the cell	2-4 ppm

### **Vitamins**

Ingredient	Importance	Optimal concentrations
Biotin	Carboxylation and Decarboxylation reactions	0.56 ppm
Thiamin	Animo acid biosynthesis	60 ppm
Inositol	Structural Membranes	15-23 ppm
Niacin	Coenzymes in Redox reactions	1000-1200 ppm
Calcium Pantothenate	Coenzymes in Oxidation reactions. Fatty, amino acid and carbohydrate metabolism	45-60 ppm
Riboflavin	Coenzymes in Redox reactions	20-50 ppm

Source Alcohol Textbook, Fifth Edition

of fermentation, to sustain continued but lower rates of alcohol production.

Within the yeast itself, certain nutrients are needed. An example of this is usable nitrogen that the yeast cell uses to make structural proteins and enzymes. The cell has to maintain the integrity of its genetic make-up, produce structural proteins and produce functional enzymes to enable the cell to undergo metabolism. This nitrogen is sourced from the medium either from the grain or via external addition. There are other nutritional components that aid in cell membrane integrity or in the function of the cell. Below are the specific nutrients required and their effect on yeast during fermentation.

**Nitrogen.** The most common nutrient used in the ethanol industry is nitrogen. Yeast growth requires known amounts of usable or Free Amino Nitrogen (FAN). It is widely known that there is a deficiency in incoming grain. Usable nitrogen is needed to increase the rate of fermentation and to complete both "normal" and very high gravity fermentations. The chart on page 54 represents some of the typical FAN levels in substrates used.

When FAN and other usable nitrogen sources are deficient, more nitrogen needs to be added. Yeast typically can assimilate only low molecular weight nitrogenous

materials including ammonium ions, urea and amino acids. The most popular addition of nitrogen primarily comes in urea added directly to fermentors and ammonia added directly to the fermentor or to the slurry tank for pH adjustment.

Sterols and unsaturated fatty acids. These nutrients are important for the yeast membrane to maintain fluidity. During the aerobic process of yeast production, the yeast cell utilizes high levels of oxygen to produce cell mass. It also produces high levels of sterols and unsaturated fatty acids needed in the cell membrane. These two components help maintain yeast cell wall membrane integrity and help in the resistance of cells to stress. In anaerobic growth (ethanol production), without the presence of low levels of molecular oxygen, these components cannot be produced by the cell resulting in a lack of resistance to stress. Without oxygen, the mother cells donate these compounds to daughter cells, and eventually both are weakened and fermentation suffers.

Vitamins and minerals. The yeast cell needs vitamins in small quantities for enzyme structure and function, and minerals for enzyme stability and metabolism. Then, optimal fermentation performance can be achieved. The table on the left il-

lustrates the most important nutrients. It is also important to mention that phosphorous in the cell is present up to 2% and usable sulfate to about 0.8% w/w as incorporated into DNA, amino acids and other important molecules for metabolism.

Many of the micronutrients listed in the table can be added independently, but the minerals come in a salt format. meaning potentially adverse anions might be added to the mash, which ultimately negates any benefit. Also, creation of on-site nutrient packages is very labor intensive. On the other hand, there are many commercial nutrient packages (yeast foods) designed to assist in this deficiency. These nutrient packages are reliable and consistent so the plant gets the best fermentation performance. Consult with a fermentation ingredients expert to identify where potential weaknesses exist and to improve the fermentation consistency of the facility.

Ultimately, there are a variety of operational factors that impact ethanol yield and a plant's profitability. With this assumption and knowing that certain substrates within the industry (pure starch or sugar fermentations) contain few nutrients, a more comprehensive nutrient package is required. Implementing a proper nutrient package will drive increased consistency within the plant and aid in the functionality of the yeast.

The most important thing in fermentation is maintaining adequate yeast viability. Yeast nutrition is vital in ensuring that the yeasts are in their best state for alcohol production. The nutritional strategy employed by a plant does have a cost, but the performance benefits of increased nutrition and ultimately healthy yeast will lead to increased fermentation rate, higher yield, reproducibility and profitability.

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