PRO Tech Notes For Craft Brewers
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Enzymes in Brewing Series
Part 2- Mashing

Preface

In the last edition (Issue 4, Volume 2) we discussed the basics of creating the enzymes in malt for the brewer. As we progress into this discussion we should begin by looking at our overall objectives in mashing and discuss the properties of each. This will help set the groundwork for our understanding as we close in on the control of the enzymes. In this material we will use the mashing profile of a single malt mash. Obviously there are many different methods to mash and additional materials that can be added. Each in itself can add complexity to our understanding. However, if you understand the basics, much of this is transferrable to other methods and materials as long as you understand the specific impacts of each variation. For instance, decoction brewing is a wonderful process to build flavor. But one needs to note that each pump-over and boil results in a loss of enzymatic power. Additionally, during pump-back, if you do not use extreme care you also will lose enzymatic power due to thermal degradation within the proximity of the hot liquid until blended. It's not that these processes cannot be used; one just needs to understand their constraints.

Introduction

To begin, only 15-25% of barley malt is soluble in water. Mashing is the process during which a much greater fraction of the malt is made soluble. During this process a predetermined (based upon the product definition by the brewer) fraction, of the solubilized compounds, is converted from starch to yeast fermentable sugars. The end product of mashing is wort. Wort contains fermentable and non-fermentable carbohydrates, proteins, amino acids, and other organic and inorganic compounds that are extracted from the malt. The primary goal of mashing is to produce as much high quality extract as possible from the grain, with some desired fraction of this extract being converted to fermentable sugars.

Amylose and Amylopectin

Also in the last edition we briefly discussed starches as a mix of Amylose and Amylopectin. Barley malt contains about 60% starch. Approximately 75-80% of this starch is Amylopectin; the complex, highly branched (1-6 glucosidic bonds) polysaccharide that can contain over 100,000 glucose monomers, with 20-25 glucose unit strands (1-4 glucosidic bonds) between the branch points. The remaining 20-25% of the malt starch is Amylose; linear strands (1-4 glucosidic bonds) of a 1000 or more glucose monomers in sequence.

Fermentable Sugars of Starch Digestion

When we discuss the digestion of these starches we break the resulting molecules into two fractions; those that are fermentable by yeast and those that are considered non-fermentable. The fermentable sugars created from barley malt via enzyme digestion are: glucose (single monomer), maltose (dimer of glucose joined by 1-4 glucosidic bonds), and maltotriose (trisaccharide of glucose that is joined by 1-4 glucosidic bonds), and that is it! One should note that not all yeast ferment maltotriose, or at least, not all of it. So when determining your fermentability from a sugar profile one needs to know what your yeast will do with the molecule. A good basis to start from is to consider it not more than 50% fermentable. We should also note that there are other sugars that yeast will ferment, namely: fructose and sucrose. These sugars do not originate in malt; they may enter your process through the addition of other fermentable materials like honey, fruits and table sugar.

Free Amino Nitrogen (FAN)

Also in the previous edition we introduced a discussion on digested protein and its importance to yeast growth. In barley the protein content can vary from 7 to about 15% by weight. About 30% of this will pass through into the finished beer, primarily as breakdown products of the initial complex protein structures inside the barley kernel. Protein breakdown products are; amino acids (single monomers of these nitrogenous compounds) and peptides (short chains of amino acids). As previously noted, proteins and high molecular weight peptides are important to beer foam and body, but they can also result in haze. Smaller peptides are basically neutral with no discernible effect. However, amino acids are vital yeast nutrients often referred to as amino nitrogen or FAN (Free Amino Nitrogen). The amount needed is a direct reflection upon the amount of yeast growth that occurs in your process. This can be a subject all to itself.

But briefly, the 25 different amino acids can be broken into 4 categories. The first category is absolutely essential for yeast growth, the second is important, the third is less important and used only if needed, and the fourth is a funny group. The yeast will typically leave these alone and only use them for construction materials if it is in a starving for FAN condition. Therefore, in rough terms, whatever amount you start with for FAN, you need to assure that you will not use it all and expect at the very least about 25-30% and preferably closer to 50% to pass through as unused and unwanted FAN. A good starting point for use and numbers would be 1 ppm FAN needed for every 1 million yeast cells you grow. Therefore, a safe place to be would be a starting FAN of about 200+ ppm if you were going to grow 100 million cells, this leaving you with around 100 ppm. As your yeast progresses through the third and into the fourth categories of amino acids you will begin to see other issues develop in your flavor profiles, this too is another opportunity for discussion.

Enzyme Overview

Moving on to enzymes, we also previously noted that they are high molecular weight complex proteins that act as catalysts for organic reactions. Typically a catalyst enables a reaction to occur, or it considerably accelerates the rate at which it occurs. Enzymes are extremely specific, catalyzing only one chemical reaction. There is a whole science around how enzymes work, beginning with a naming convention. In simple terms, the substrate affected by the enzyme becomes the basis of the enzyme's name, with an "ase" attached to the end. Thus proteases affect protein; beta glucanases affect beta glucans, etc. The science of enzymes theorizes a lock and key concept of attachment and reaction, feedback and regulation, environmental effects, etc. For our discussion we are really only interested in the effects of time, temperature and pH.

pH

Let's begin with the easy one of these variables, pH. As complex protein structures, enzymes possess very defined polarized (charged) areas. This polarization determines much of their shape and can be a major part of how they function. The ion characteristics of the solution they reside in will have a major effect upon their polarization, thereby affecting their ability to hold shape and function. In basic terms, the pH of a solution expresses the balance of negative and positive ions expressed as a concentration. In looking at mashing, we normally look at a pH range of roughly 5.0-5.7. It's nice for us that all of the natural enzymes we are concerned about in mashing function well within this range. So for the sake of this discussion, we no longer need to

be concerned. Notable, there are some specialty mashing processes that take us outside this normal pH range, resulting in opportunities for greater discussion.

Time and Temperature

We now need to look at the effects of time and temperature on enzyme function. We begin by looking in greater detail at what we are trying to achieve in the mashing process. Our objectives, simply stated, are to influence the enzymes created in malting to digest the barley malt extract in such a manner as to make the wort for the beer type we are trying to create. To accomplish this we use the variables of temperature and time dependence to establish that influence, as we require, for our process. Again, in simple terms, all enzymes have a temperature of greatest activity. Unfortunately, this high rate of activity normally occurs very close to the temperature at which the enzyme becomes deactivated. Therefore, time also plays a role in our discussion. The closer we get to the optimum activity temperature for the enzyme, the shorter the life span for the enzyme. Thus, balance is needed, as well as an understanding of which enzymes are critical, at each stage in mashing.

Seven Critical Enzymes

With this background we can now move into the mashing process. As we have stated, this process is a sequence of time and temperatures designed to influence the enzymes we need for the beer we are making. There are many enzymes that are active within a mashing process. Many of these enzymes act on substrates that are either in such a large quantity that their very level defines a process, or the substrates are so small they can be considered *de minimis*. For the sake of this discussion we will consider only the seven enzymes that are really important to control for our processes.

The First Three Enzymes

Normally, mashing begins at a temperature designed to either fit our equipment design or because we believe it has some benefit. In any event, it is important to fully wet the grains and begin the process of freeing up the enzymes into solution. To begin, there are three of the seven enzymes that concern us at mash in. These three enzymes have optimal activity temperatures around where most brewers begin the mashing process, 100-120 dF. These enzymes are: Beta Glucanase (optimal 113-122 dF, deactivated 140 dF), Ferulic Acid Esterase (optimal 100-113 dF, deactivated 149 dF), and Proteases (optimal 113-130 dF, deactivated 158-167 dF). As you can see some optimal activities

overlap in temperature so, often, multiple activities are occurring at the same time at specific temperatures.

Protein Rest Benefits

If the maltster has done their job correctly we have well modified malt and therefore little concern about Beta Glucan and FAN content. However, in the real world we know this is not always the case. It should be noted that after kilning much of the Beta Glucanase and Protease capabilities in the malt have been eliminated. Therefore in attempting to further reduce Beta Glucans or increase FAN by using extended protein rest holds around 120 dF, we will have only a small effect (perhaps about a 10% shift). What is critical to note is the Ferulic Acid Esterase activity. Extended holds can result in an increase of upwards of 100% in Ferulic Acid.

Ferulic Acid

Ferulic Acid, by itself is not of great concern. **However, if your wort possesses high levels of ferulic acid, and your process allows for long hot temperature holds post kettle boil, you can experience a flavor impact in your beers.** As ferulic acid sits at high temperature, it will naturally convert to 4 vinyl guaiacol (4VG). 4VG is highly volatile and possesses a strong clove aroma and flavor. For wheat beers this flavor is considered normal, but for other beers this is considered a defect. Therefore if you are short on FAN or long on Beta Glucans, there are commercially available solutions to address these issues. We will discuss this in our next edition.

Conversion Temperatures

Moving further along in the mashing process we normally see use of a one or two temperature holds to influence the optimal activity of the starch converting enzymes, typically known as the Conversion Temp. Remember, our objective is to create a blend of fermentable and non-fermentable sugars for the beer type we are making. The next four of the seven enzyme groups now become of interest. We can break these down into two basic categories. The first three are starch converting enzymes: Limit Dextrinase (optimal 131-140 dF, deactivated 145 dF), Beta Amylase (optimal 140-149 dF, deactivated 158 dF), and Alpha Amylase (optimal 162-167 dF, deactivated 176 dF). The last enzyme of interest at these temperatures is Beta Glucan Solubilase (optimal 140-158 dF, deactivated 163 dF).

Beta Amylase
Let's begin with the starch digesting enzymes. Typically the ratio of fermentable to non-fermentable sugars in a mash is set by the conversion temperature. This temperature should be chosen to express the blend of the two principle starch enzyme activities; Beta Amylase and Alpha Amylase. The Beta Amylase digests strands of starch and dextrins, breaking them down to maltose molecules and undigestible fragments. As it works, it can only break linear 1-4 glucosidic bonds and therefore cannot digest Amylopectin due to the branch points. Secondly it can only address the starch chain from one end, the non-reducing end. Therefore, its activity is one molecule at a time, working along the chain until it reaches a branch point where it must stop.

Alpha Amylase
For Beta Amylase to be effective, more non-reducing ends must be made available for its attack. This is where Alpha Amylase comes into play. The Alpha Amylase also only break 1-4 glucosidic bonds, but it can attack the starch molecule at any point. This random attack results in a large array of smaller dextrins and a small amount of fermentable sugars. However, the starch molecules have been broken down for effective digestion to fermentable sugars, primarily maltose, by the Beta Amylase.

Sense of Balance
This would be great if it were that easy. But if you note, the optimal activity temperature for the Beta Amylase is much lower than the Alpha Amylase. It is therefore possible to deactivate the fermentable sugar-making enzyme before the Alpha Amylase can break down the starch for its attack. Thus we need some balance. Keep in mind most enzymes are active at some level up to their deactivation temperature. They are just not as active as they are at their optimal temperature. So at the optimal temperature for Beta Amylase, 145 dF, Alpha Amylase is digesting starch, just not at the same high rate it would be if the temperature was 165 dF, roughly seven degrees higher than the deactivation temperature of Beta Amylase.

Time
Given this concept, we must now introduce the variable of time into the discussion. The Beta Amylase works by chewing up a linear strand of starch/dextrin, one maltose molecule at a time. On a relative basis, it is a fairly slow process. The Alpha Amylase cares little about where it attacks the starch, and therefore, it is considered relatively

fast. If a brewer is looking for more fermentability (**higher alcohol, thinner and faster beer)**, they will need to place the conversion temperature closer to the optimal activity of the Beta Amylase. Additionally, they will need to consider the relative rate of reaction and the hold time for the conversion will need to be longer, again on a relative basis. Conversely, if one is looking for less fermentability (**lower alcohol, greater body and sweetness**), you would choose a higher temperature to favor the Alpha Amylase activity, while carefully considering the deactivation temperature of the Beta Amylase and noting the reaction will come quickly.

Words of Caution

As you look at the opportunity before you I offer a couple words of caution and advice. When dealing with mash temperature close to or below 147 dF one needs to consider the micro characteristics of your process. As your drop near to 145 dF you are now allowing for the incubation of a large amount of naturally occurring micro-flora that comes with the grain. This is nothing to worry about, unless you hold for extended periods of time, as with long holds to create high RDF beers. Holds that exceed 3-4 hours can be problematic due to the micro-flora contributing veggie, especially radish type flavor notes to your beers, be cautious. Secondly, the longer you hold the more damage your filter bed is seeing. Please consider coarser grinds and slow agitation speeds.

Our Last Two Critical Enzymes

To finish our discussion we need to look at the last two of the seven enzymes we discussed; Limit Dextrinase and Beta Glucan Solubilase. Keeping with the starch discussion we will look at the Limit Dextrinase first. This enzyme possesses the ability to break the 1-6 glucosidic bonds in the branch points of Amylopectin. As with most enzymes of its type it is extremely temperature sensitive and is deactivated rapidly at or above its deactivation temperature of 145 dF. Additionally, there is very little to begin with in barley and much less, after kilning, in malt. However, it does exist and does have an impact, albeit insignificant as it relates to the Amylase activity. I mention it here to help stimulate thoughts of “what if” as we look toward the next edition where we take a look at commercial enzymes. During that discussion we will refer to Limit Dextrinase as Pullulanase as they are typically known within this industry. So as you ponder scenarios, consider that there are other tools available to help you achieve what you are trying to accomplish.

Beta Glucan Opportunities

Lastly, we look at Beta Glucan Solubilase, a very problematic enzyme when considering malt that may be borderline in modification (slightly high Beta Glucans). Additionally longer holds for conversion or delays in your brewhouse process, holding up your lautering can also play into this enzyme. Earlier we mentioned the optimal activity temperature of this enzyme at 140-158 dF, centered on typical conversions. Most brewers will mash off to lauter/strain at temperatures at or greater than 163, this enzyme's deactivation temperature. But often if one is stretching the limits of the Beta Amylase activity, some attempt to mash off in the 150's leading to extended time for the Beta Glucan Solubilase to work. Taking from its name, this enzyme solubilizes beta glucan, freeing it from the husk material, not only allowing more into solution but further degrading the integrity of you filter bed. It is a quandary on how to deal with this enzyme, except to offer two solutions if you venture down the path of long holds. The first deals with extremely well modified malt. Keep in mind this is not standard material available to the general public, one must request and probably pay for it. To do this, maltsters must play with the maximum ranges of their processes and flirt with PYF (premature yeast flocculation) potentials in the malt. Please refer to the last edition (Issue 4 Vol. 2). So if you take this path understand that there are no solutions to PYF yet, other than to avoid it. The second solution is commercial enzymes, small additions of thermostable Beta Glucanases. Often these come as combinations of Beta Glucanases and Xylanases not only offering you a solution to Beta Glucan Solubilase and undermodified malt, but as a means to increase your yields and stabilize the performance in your brewhouse. But that is enough said on the next issue's topic.

PRO Engineering Services

As we walk away from this discussion one should be able to see the critical nature of brewhouse control. This is not only control of the obvious, temperature, but time as well. Additionally agitation, insulation, pumping capacity, heating and cooling capacity all play roles in controlling what you are trying to do when you challenge yourself with something different. I'm not the engineer and cannot offer to you these services. However, your host for these articles can. PRO Engineering and Manufacturing Inc. can offer you a wide range of fully scalable and integrated solutions to your "opportunities". Please consider them to partner with you and help you grow with new possibilities.

Next Issue of PRO Tech Notes

In our next issue we will begin the discussion of "what if". As you can see from above, there are many challenges before you, many I am sure you all face daily. I know what comes to you in the form of grain; I was responsible for grain quality for several years in a past life dealing with the "opportunities" of nearly a dozen malt houses. Good quality malt is not only at the hands of the maltster; mother nature plays a massive role. She challenges us all, year to year, with all kinds of new events, etc. This creates variation in the grain and thus your malt. I believe in consistency because I believe in flavor control, matching the flavors that I want, not what a variable process will give me. Please take the time to read the next issue of PRO Tech Notes and at least consider the possibilities of the doors that can be opened, the issues resolved and the consistencies created. As always, I am open to your thoughts and questions, please feel free to share.

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The author, Mark Sammartino has over 35 years'experience in the brewing industry. Some of his experience includes:

- Brewmaster at Anheuser Busch
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Additionally, Mr. Sammartino is a consultant with Brewing Consulting Services, LLC The company provides a wide range of practical operational advice and solutions to clients in the Craft Brewing industry. The group also includes Founder, David Kapral and Associate, Pat Frost. Collectively this group has 100 years of experience in the industry.

Contact Mark Sammartino if you would like to discuss the issues raised in the article or if you want to explore further assistance from the firm of which he is a member:

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Ed Michalski (left) with brother Dave,checking specs for a customer

If you need brewing equipment repaired, or re-engineered to work better, fasteror more cost-effectively, contact Ed Michalski, CEO, at PRO Engineering and Manufacturing, Inc.

PRO Engineering and Manufacturing, Inc has a commitment to serving the Craft Brewing industry through tunnel pasteurizers and batch pasteurizers specifically tailored to craft brewers.

Edward A. Michalski Bio

Ed Michalski started his career in the beverage industry by designing stainless steel, higherflow, spray headers for Pabst Brewing. Along with the header design he also developed a process to produce the new headers.

Ed, along with his brother David, formed PRO Engineering/Manufacturing, Inc. Based on what they learned by re-designing and refurbishing other manufacturers' pasteurizers, Ed and PRO started to offer the pasteurizer marketplace superior newpasteurizers.

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