THE ROLE AND THE INFLUENCE OF ENZYMES IN THE OPTIMIZATION OF WORT PRODUCTION FOR BEER

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ABSTRACT

Biochemical changes during the entire brewing process mostly involve the action of different enzymes which are essential in catalyzing these changes. Enzymes are present in mature barley (amylase and carboxypetidases), finished malt (α-amylase, limit dexrinase, proteases, glucanases, pentosanases) and yeast. They have optimal activity under specific conditions of pH and temperature and if these conditions are not optimal, enzymes do not perform their action. For beer production the most important spectrum of enzymes including amylase, protease and β-glucans. Using these enzymes in the boiling process, we complete the enzyme activity of malt supplement, or replace them when the boiling process does not allow the effectively action of natural enzymes of malt. The purpose of boiling process is to reduce the viscosity of wort, as well as reduce the resistance of the mass filter in order to significantly meliorate the circulation time in boiling process. The most important enzymes responsible for filtering process is β-glucanase. B-glucanase acting on gum substances of malt to improve the reduction of viscosity (liquefaction of wort) and clarity of beer. In this paper are presented the results of measurements performed for characteristic like viscosity, turbidity and filtration time. In the first test measurements were performed after the addition of β-glucanase enzyme. In the second test measurements were performed after the addition of breakbright enzyme. These results are compare with the values obtained when these enzymes are not added.

Keywords: Activity, β-glucanase, boiling process, enzyme, filtration time, viscosity, wort.

INTRODUCTION

Enzymes are catalysts, which speed up chemical reactions without change their nature or structure. Some of them degraded the starch, other degraded various components into smaller molecules and other new compounds synthesized from different precursors. The traditional source of enzymes used for the conversion of cereals into beer is barley malt. If the conditions of use or storage of enzymes (pH and temperature) are extreme, the enzyme can irreversibly lose its catalytic activity. In this case the enzyme is denatured. If wort is submitted to high temperatures, most enzymes will be denatured. Complex proteins of barley embryo contain several enzymes, however, most of them are formed during malting process. For example, α-amylase, does not exist in barley however it exists in malt. Each enzyme catalyzes only a specific chemical reaction. They have optimal activity under specific conditions of pH and temperature. The enzymes will denature in conditions not optimal, if they are extreme and in soft conditions they can retrieve again their catalytic activity. In caramel malts which are submissive to high temperatures, most of the enzymes are denatured. European malts for Pilsener beers are generally malts with good enzymatic content. The most important enzymes in brewing are amylase, protease and β-glucanase.[2]

The main representatives of amylases are α- amylase and β-amylase. During the wort production they work at the same time to make the breakdown of starch into simple sugars. The optimal activity of amylases extends in the interval 60-70ºC.

Proteases decompose the proteins by breaking the links between aminoacids. This is the mechanism by which are released the protein reserves present in barley. Aminoacids as peptides, play an important role in the chemistry of beer, above all the metabolism of yeast and in foam head. Proteases include over 40 enzymes that act in different ways on proteins.

β-glucanase operates on gum materials of malt to improve the reduction of viscosity (liquefaction of wort) and clarity of beer. The β-glucan carbohydrate is composed mainly from simple molecules of sugars as well as starch. Gum character of β-glucan increases the viscosity of the wort and brings poor filtering and bad clarity of wort. Best operating temperature of β-glucanase is 45ºC.

If too little enzyme activity is present in the mash, there will be several undesirable consequences: the extract yield will be too low; wort separation will take too long; the fermentation process will be too slow; too little alcohol will be produced; the beer filtration rate will be reduced; the flavour and the stability of beer will be inferior [7]. Using these enzymes in boiling process, we complete enzyme activity of malt supplement, or replace them when the process of boiling prevents the natural enzymes of malt act effectively.

Enzymes derived from sources other than malt may be used at various stages during brewing, provided that this is allowed by local regulations [2]. The preparations available have a wide range of characteristics. Different suppliers describe their preparations in different ways so that it is difficult to make comparisons between them. The temperature and pH optimal of enzymes are so influenced by incubation conditions, and the conditions used in different breweries and at different stages of brewing are so varied, that it is not possible to give useful values. Consequently, the effectiveness of the addition of an enzyme preparation must be determined by brewers under their particular processing conditions. Protein coagulants are typically added to remove free-floating proteins from the brew. Protein coagulants bind free-floating proteins that may exist in the aqueous brew into protein-coagulant masses. When the density of the protein-coagulant masses becomes greater than that of water, the protein-coagulant masses sink to the bottom of the aqueous brew, facilitating their removal [1]. Examples of commercially available protein coagulants include breakbright enzyme.

To study the role of enzymes in the production of wort, are performed some tests adding β-glucanase and breakbright enzyme. Then was measured the viscosity, the time of filtration and the turbidity of wort. The same measurements performed in cases when are added enzyme and when the wort contains only natural enzymes malt. Therefore, the objective of this study is to investigate the influence of enzymes in the quality of wort and the proper quantity that should added to take a wort with much better characteristics.

EXPERIMENTAL

The data on which this paper is performed are obtained from the analyses that are made for different samples of wort. The period analyzed in this study was 2014-2015.

Test with breakbright enzyme

In a 500 ml balloon prepare a breakbright solution by mixing one gram breakbright in 400 ml of demineralized water. We keep the temperature constant in 70-80ºC and realize complete digestion of breakbrightit by mixing continuously the solutionn. After this procedure, we bring the balloon to tare mark. We take a sample from boiling wort before adding breakbrightit enzyme. The sample boils for 10 minutes. In 9 test tubes add about 100 ml of boiling wort and add the breakbright solution as follows:

Table 1. The sample prepared for breakbright test

Nr. of sample	1	2	3	4	5	6	7	8	9	10
ml sample	100	100	100	100	100	100	100	100	100	100
ml solucion BB	0	1	1.5	2	2.5	3	3.5	4	4.5	5

After we prepared the samples according to table, for each of them measure the turbidity. The turbidimeter is an instrumentation used for determining the turbidity. 800 and 800-P model performs the specifications of EPA for measurement of turbidity in drinking water. Turbidimeter is pre-calibrated and requires only do zero system before the test. Notice that the unit of turbidity is EBC.

Test with β-glucanase enzyme

β-glucanase is used to solve specific problems during the boiling process that stop a good filtration. This enzyme has effect on cellulose, hemicelluloses and β-glucan. Fungal β-glucanase preparations (e.g. from *Aspergillus* spp.) have varied properties, but usually have inconveniently low temperature optima (45-60ºC) for mashing but have convenient pH optima in the range 3.5-6.0 [4].

One of the critical physical properties is viscosity. Concerning breweries and processing laboratories, viscosity is monitored in several different stages of beer production (supplied malt quality tracing, malt and wort quality determination, filtration monitoring, and final product evaluation). Viscosity also plays an important role in theory of filtration. Viscosity is taken into account when designing the filters and setting the working pressures. A high viscosity makes beer filtration more difficult and may lead to starch hazes in the final beer [6]. The viscometer is an instrumentation used for determining the viscosity. Viscosity was determined for wort produced with different methods. The results are taken from wort prepared in 65ºC, in 45ºC after added the β-glucanase during the mashing process, and from wort prepared with the same methods but in this case are not added the β-glucanase.

Mashing is the process of extracting the goods by mixing them with water at suitable temperatures and in proper relative quantities, preparatory to boiling in the kettle. The mash should be so conducted as to secure the desired composition from the goods employed. Different methods of applying temperatures to a mash supply the following systems:

1. Infusion method, the mash is bought to its final temperature by the admixture of water of suitably high temperature.
2. Decoction method, part of the mash itself is raised to a boil and then returned to the mash-tun.
3. Mixed method, a combination of decoction and infusion methods. [5]

The filtration time is determined for samples of worts prepared through three methods. In the first case we add β-glucanase before the mashing process, and in the second case measure the filtration time for sample of worts without adding β-glucanase.

RESULTS AND DISCUSSIONS

Test results by adding breakbright enzyme

Samples are prepared as given in the Table 1, were left in repose for two hours. The turbidity was measured for all samples after 30 minutes, after one hour and after two hours. The results obtained are as follows:

Table 2. Measurements taken after 30 minutes

Sample	1	2	3	4	5	6	7	8	9	10
Turbidity (EBC)	77,1	88,5	87,3	88,3	84,8	82,0	81,6	83,8	81,9	86,1

Table 3. Measurements taken after 1 hour

Sample	1	2	3	4	5	6	7	8	9	10
Turbidity (EBC)	76,9	13,6	7,2	67,9	69,9	101,2	112,4	98,9	94,4	95,3

Table 4. Measurements taken after 2 hour

Sample	1	2	3	4	5	6	7	8	9	10
Turbidity (EBC)	77,0	5,44	5,23	56,6	62,3	74,7	65,9	62,8	62,4	64,2

Samples 2 and 3 respectively with 1 and 1.5 ml breakbright give very good results and the difference from the other samples is very visible. Samples 4-10 start to clarified after more than two hours. Sediment taken from samples 2 and 3 is relatively compact. The best result gives simple 3. Hair of protein coagulation for samples 2 and 3 are great. Sedimentation time for samples 2 and 3 is relatively short. Turbulence is good especially for sample 3.

In Figure 1 it is clear that with increasing time of sedimentation is reduced the turbidity of the wort. The most convincing samples is number 3, in which was added 1.5 ml breakbright. With this amount of enzyme not only achieve optimal value of clarity (minimum value of turbidity), but achieved even the best time of sedimentation. Performance of protein coagulation within the first 30 minutes is almost linear. After 1 hour and 2 hours in turbidity performance of wort is given by a fourth order polynomial. Certainly after 2 hours are obteined lowest value of turbidity for all samples compared with that after 1 hour. This experiment clearly shows that the enzymes should be used in completely defined amount. Inappropriate use of their not only brings high costs in the process, but is ineffectual.

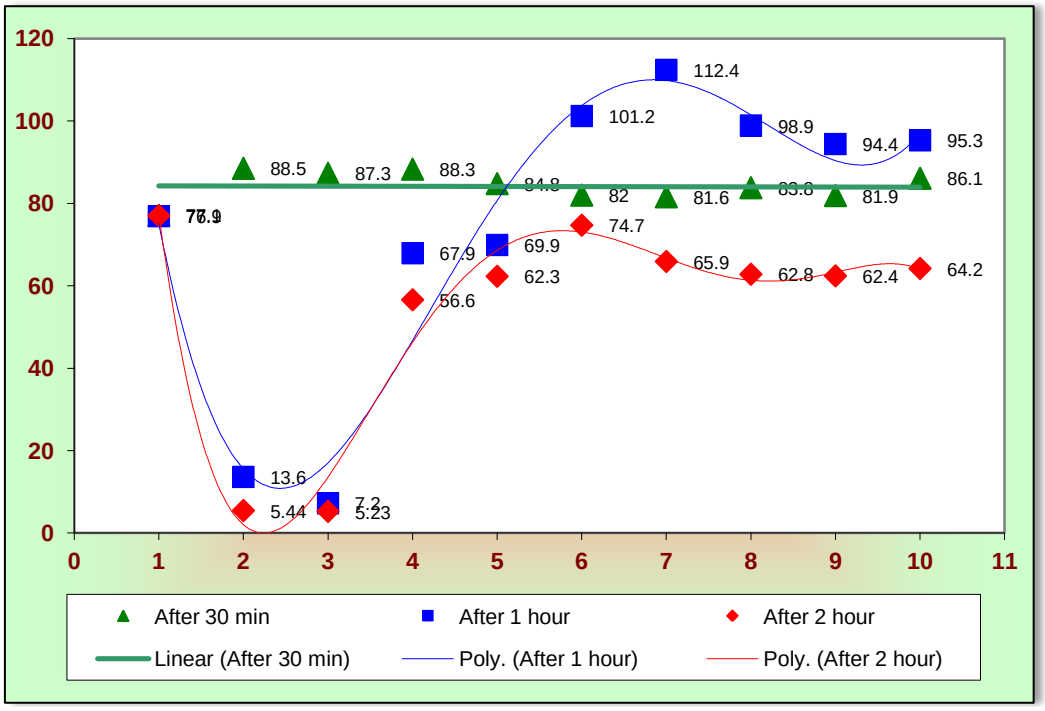

Figure1. Turbidity after 30 min, 1 and 2 hours of wort treated with breakbright.

Test results by adding β-glucanase enzyme

The first test done for studying the role of the β-glucanase in wort, is the measurement of viscosity of wort produced by different methods. For this is compared the viscosity of wort produced by the method in 45ºC and 65ºC without adding the β-glucanase and the viscosity of wort produced by the same method but in this case add β-glucanase in the mashing process. For each malt are taken 3 samples from different beer production. The results obtained are summarized in Figure 2. This figure shows that the worts in which β-glucanase was added, have lower value of viscosity. The lowest viscosity is taken in the worts produced with method in 45ºC adding β-glucanase, because the best operating temperature of β-glucanase is 45ºC. Worts that have viscosity higher than 1.75 cP, the filtration time is over one hour caused from a bed filtration. As we can see the wort produced by the method in 65ºC has viscosity higher than 1.75 cP in all three measurement and presents difficulties in filtering. If this method will be used for brewing is necessary the addition of β-glucanase in mashing process.

The second test done to determinate the influence of β-glucanase in wort is the measurement of filtration time of wort produced by tree different methods. In this case is compared the filtration time of wort produced by infusion method, decoction method and mix method without adding β-glucanase and the filtration time of wort produced by the same method by adding β-glucanase in the mashing process. The results of the measurements done are given in Figure 3. From the chart it is clear that the filtration time of the worts where is added β-glucanase compared with the worts without β-glucanase is almost half. The difference between these two times decreases significantly when enzyme activity of the malt is increased.

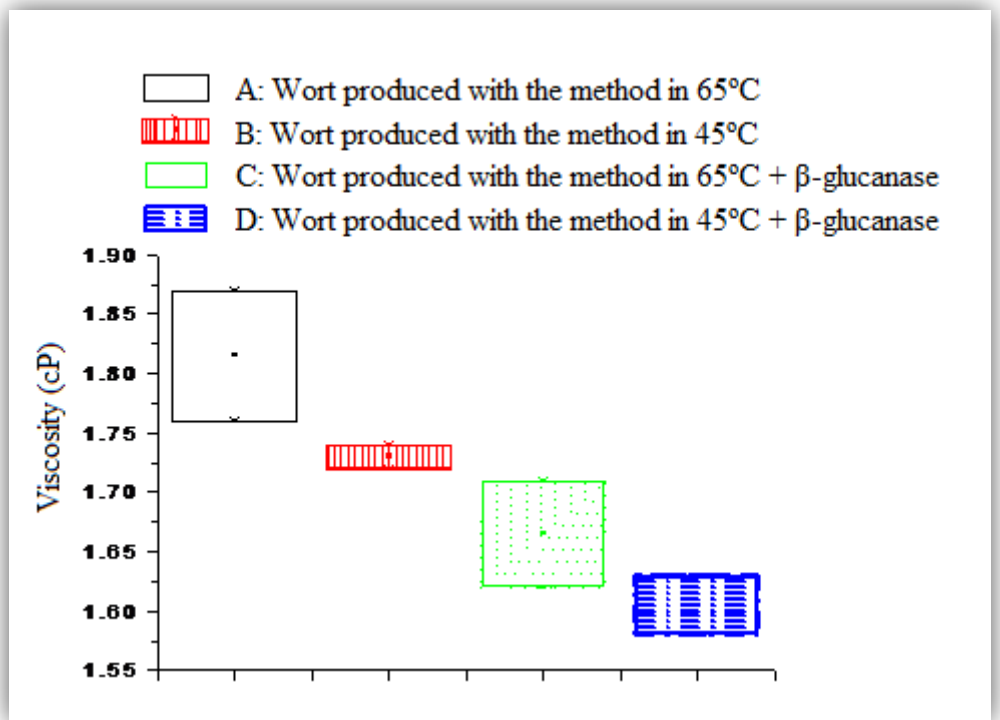

Figure 2. Viscosity of worts produced with different methods.

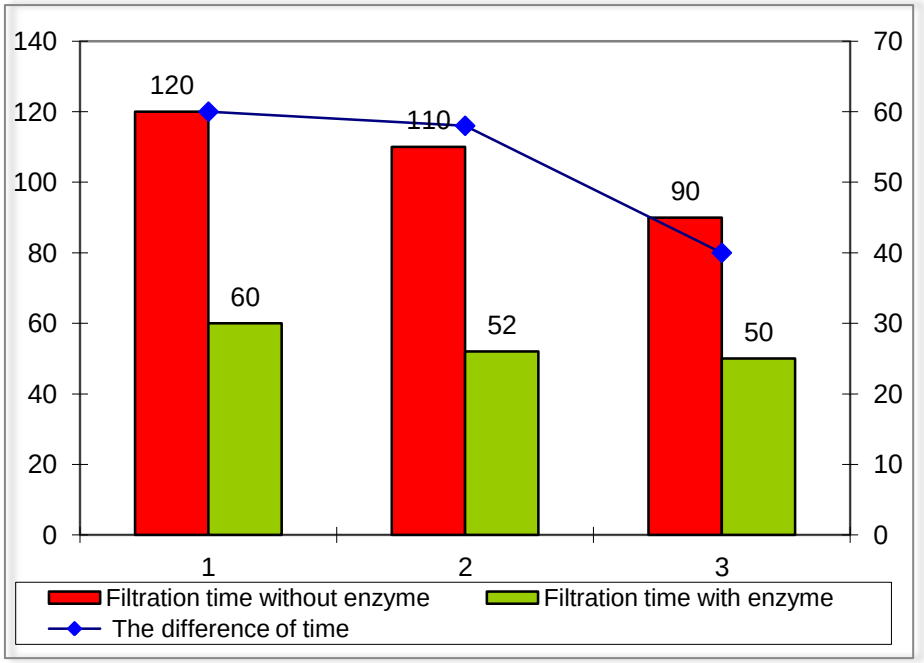

Figure 3. The diference of filtration time

CONCLUSIONS

By tests that are made we have noted that enzymes have a great impact on the wort production. Enzymes affect the turbidity, viscosity and filtration time of wort. If we want to produce a wort with turbulence enough order not to prevent the filtration process, is required the addition of enzymes in the mashing process. For a good filtration is recommended viscosity of wort less than 1.75 cP. The wort produced by the method in 65º have a viscosity higher than 1.75. In this is necessary the addition of β-glucanase in mashing process that reduces the viscosity of the wort. In conclusion we can say that, if the malt in the wort production not degrade at the appropriate level as a result of the poor quality or as a result of a method not accurate (this means incomplete activation of enzymes in the proper time and temperature), the only way to get a clear wort, filterable and with low viscosity is the addition of this enzyme.

LITERATURE

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