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The monitoring of enzyme activity of glucoamylase and endoxylanase on the bread dough with consitographic method

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Abstract

This study presents the action of two enzyme preparation: based on glucoamylase and endoxylanase, in the bread dough. The determination of the rheological characteristics of the dough is obtained by consistographic method and baking test. The addition of glucoamylase and endoxylanase enzymes allows adjustment of the rheological characteristics of dough according to the needs of the technological process. The specific effect of glucoamylase and endoxylanase on the physical properties of dough show a positive influence on the volume and porosity of the bread, also on reducing the kneading process of the dough and the energy consumption for the technological process.

Keywords: bread, glucoamylase, endoxylanase, consitographic method, baking test.

1. Introduction

Enzymes applications have grown to be a common practice in the baking industry with advantage of being considered as natural additives. The enzymes preparation are being used in the baking industry to improve dough-handling properties. The synthetically additives can be replaced with natural additives, as enzymes.

Bread is the most comun traditional food product in the entire world. It has a high nutritive value due to the content of easily retainable sugars, lipids and proteins.

γ-amylase (glucoamylase or α-1 ,4-D-glucan-glucohidrolaza) is an exoamylase used in food industry, that catalyzes the hydrolysis of α-1 ,4-linkages of glycosidic starch / glycogen (from the end of the unreduced polisugar) to form glucose. Compared with other amylases (α or β) γ-amylase is active at acidic pH (pH optimum is 3). Most glucoamylases can split and bond α-1 ,6-glycosidic, but at a rate lower than the connection α-1, 4. A single species of fungs, *Aspergillus oryzae* is used on an industrial scale preparation of α-amylase and glucoamylase. Glucoamilase from *A. Niger* contains 3 regions: (i) the region containing the catalytic (amino acids 1-470), which is the portion glycosylated region 441-470, (ii) a region binder (amino acids 471-508) in which serine or threonine residues are O-manosylate and region (iii) attaching insoluble starch chain (amino acids 509-616).

Glucoamylase acts against aging bread, but also helps prevent steakiness of core. Also works during Maillard reactions between low molecular weight dextrins and protein materials, and can lead to a darker bread crust.

Endoxilanaza is most effective hemicellulases in the manufacture of bread. It hydrolyses araboxylans both water-soluble and water-insoluble, in relatively large oligomers. Degradation into smaller fragments occurs strongly with these types of enzymes.

Adding endoxylanase of *Aspergillus niger* for bread leads to increased disolving araboxylans up to 10 to 30%. Without addition of enzyme, 15 to 20% of non-extractable pentosans are solubilized by endogenous

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xylanase present in wheat flour. Adding endoxylanase double this amount, leading to an increase of 40-65% for viscosity of dough.

The breaking of pentosans chains and their disolving helps to improve dough tolerance and a higher volume of the bread.

Endoxylanase contribute to greater stability through effects against aging, increased warranty period, improved freshness and improve the texture of bread with a softer core, fine and uniform pore structure, stability of frozen dough, increased crispiness and decreased wettability. Are also used to increase the volume of bread products with high content of fiber, to improve the quality of bread flour. The color and flavor are also improved due to the browning effect of the crust and whitening effect of the core.

2. Materials and Methods

2.1. Samples preparation

In order to determine the consistogrames following tests were performed: the constant hydration test (CH) and adapted hydration test (AH).

Enzyme preparations used are:

- ALPHAMALT GA 5071 - based on glucoamylase enzyme preparation; Features: improves browning features, increase the power of slow fermentation; Dosage: 1-10g/100kg flour

- ALPHAMALT HCJ – based on endoxylanase enzyme preparation; Features: improved extensibility characteristics of dough, increase volume; Dosage: 5-15g/100kg flour

Constant hydration is essential in order to make adapted hydration.

In order to prepare the divice for the tests it is needed to include the double-mixing auger into the mixing space.

Sample preparation is done by weighing 250 g flour with a precision of 0.5 g sample. Then it is determined the moisture with the analytical balance. After this it is prepared a 2.5% NaCl solution which is placed in the burette of the consistograph. The temperature of the solution is between 18 to 22 ° C. The volume of this solution is equal to the initial specified moisture. Then the mixing phase is started and after 1 min the divice is stopped and cleaned on the side walls of the kneading space, taking care not to touch the sensor and not longer than 2 minutes for cleaning. Then it starts again and after 250 seconds of mixing the consistograph and indicates the quantity of sample that will take part in the next test and the salt solution required to determine the adapted hydration.

The beginning of the adapted hydration test is conducted by weighing the suitable quantities of flour and the required amount of water indicated, then the operation is started again by doing the mixing according to the first test. After 480 seconds the consistograph stops and the consistographic results for the sample is recorded by Alvoelink.

2.2. Methods of analysis

1.The Consistographic method is used to determine the consistency of dough and water absorption capacity of flour and observing changes in the process of kneading dough.

The equipment used to determine consistogrames is Alveo-Consistograph NG.

From the consistograme the following indicators were obtained:

• flour moisture (H_2O)

• maximum pressure (Pr MAX)

• hydration potential of the flour (HYDRA)

• time to reach the maximum pressure (Pr T MAX)

• tolerance or dough stability (TOL)

• pressure drop compared to PrMax after 250 seconds or the degree of softening of the dough after 250 seconds (D 250)

• pressure drop compared to PrMax after 450 seconds or the degree of softening of the dough after 450 seconds (D 450)

• water absorption capacity WAC.

2. To determine the characteristics of flours and baking influence of various additives on the quality of their bread, the most reliable method is the baking test.

Flour quality assessment and its behavior in the presence of additives is based on the quality of bread produced. The baking test allows tracking the rheological properties of dough during the technological process.

Baking tests were performed using both equipment and laboratory facilities, specific, and industrial equipment.

Baking process requires a baking oven and a fermenting room. Samples after kneading are put in fermenting room for 45-50 min at a temperature of 33-35 ° C. After that samples are placed in the baking oven at a temperature of 245 ° C for 50min. After baking, the breads are left to cool for 20 min. After cooling, the breads are cut in the middle and measure with a ruler and determined the section height.

3. Results and Discussion

a) The dough samples consistograms are represented in Fig. 1, Fig. 2, Fig. 3. Each dough sample consitograme show the parameters registered at the testing moment.

The following classification of flour based on the time to reach maximum pressure (Pr T MAX) and dough tolerance (TOL)

- Weak flour: T Pr MAX between 1-3 minutes TOL between 1 - 4 minutes

- Medium flour: T Pr MAX between 3-8 minutes TOL between 4 - 5 minutes

- Strong flour: T Pr MAX between 8-15 minutes and 10-15 minutes TOL

In Fig. 1 the consistograme of the standard Sample 0 - no added enzyme preparation are shown the dough's rheological characteristics of a sample that contains no enzyme.

- Humidity is 14.30%;

- The time at which the maximum pressure is reached 135s;

- Tolerance or stability of the dough is 198s;

- pressure drop compared to PrMax after 250s and after 450s, or the degree of softening at 250 and 450 seconds is high (349 or 596 mb) and water absorption capacity is 53.6% base 15% H_2O.

- When the maximum pressure is reached ($TPRMAX_0$) and dough tolerance (TOL_0) is lower compared to the optimal conditions, indicates that the Sample 0 has weaker features due to lack of enzymatic activity.

The dough obtained from this type of flour shows a low extensibility and low tensile strength, suggesting that it is not resistant and bread obtained is undeveloped, flat shape with cracks in the upper crust.

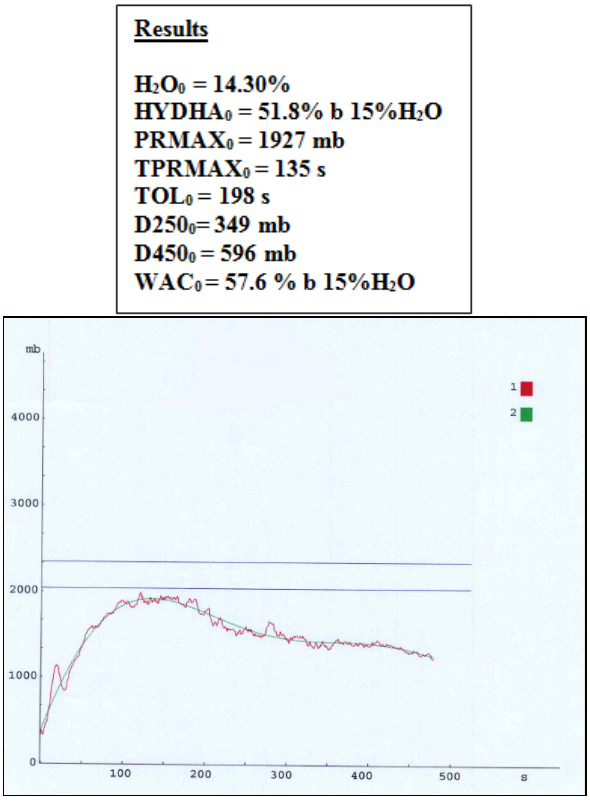

Figure 1. Consistograme for Sample 0 - no added enzyme preparation

In the consitograme of Sample 1 - with the addition of glucoamylase we can notice an improvement of rheological properties of dough due to the addition of this enzyme compared to the standard Sample 0. It can be seen that:

- The humidity remains constant (14.30%),

- Maximum pressure ($PRMAX_1$) shows an increase 164mb;

- Time for complete hydration ($TPRMAX_1$) is 214S.

- Tolerance of the dough (TOL_1) shows an increase of 154s.

- $D250_1$ has decreased to 25mb and $D450_1$ also decreased to 442mb compared to the Sample 0

Thereby demonstrating that the enzyme preparation based on glucoalylase improved dough quality compared with standard sample. When dough elasticity and extensibility is high enough, it results

in loose bread, with developed volume and a core containing pores with thin walls. This Sample 1contains the optimal conditions used in bread dough.

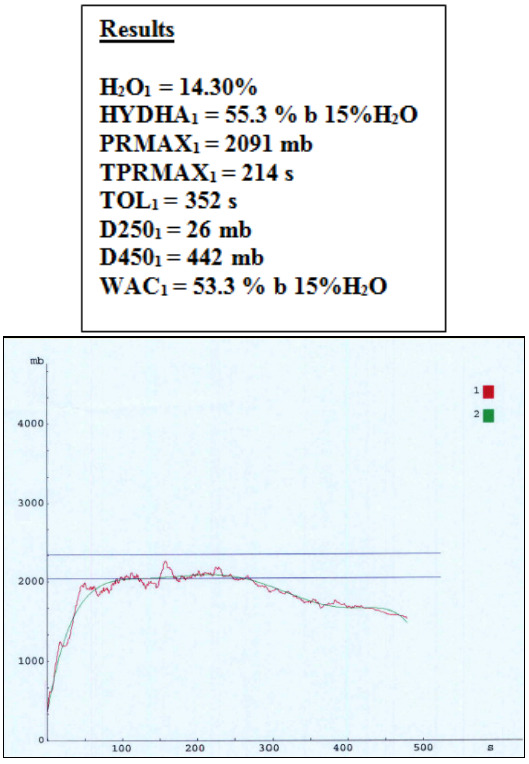

Figure 2. Consistograme for Sample 1 – with addition of enzyme preparation based on glucoamylase

In Fig. 3 the consistograme of Sample 2 - with added endoxylanase shows a small improvement in all indicators compared to Sample 0, but not achieving the parameters of Sample 1 or the normal conditions for bread dough. The results show:

- The same moisture content as the other two samples;

- The time at which it reaches the maximum pressure is 155s,

- Tolerance or stability of the dough is 246s

- The degree of softening at 250 and 450 seconds is 189, or 639 mb;

- Water absorption capacity is 54.7% base 15% H_2O.

Compared with standard sample parameters were improved, but compared to Sample 1 or optimal values for bread dough, these parameters do not qualify for bread dough. Bread made from this dough has a small volume, pale, and with a dense core.

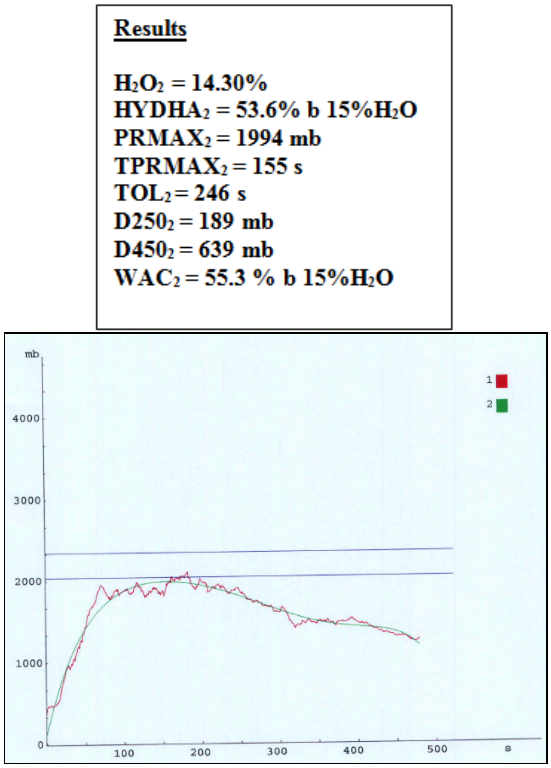

Figure 3. Consistograme for Sample 2 – with addition of enzyme preparation based on endoxylanase

Table 1. Consistograph results of the dough samples: Sample 0 - no added enzyme preparation; Sample 1 – with addition of enzyme preparation based on glucoamylase; Sample 2 – with addition of enzyme preparation based on endoxylanase

Sample	Sample 0 - no added enzyme preparation	Sample 1 – with addition of enzyme preparation based on glucoamylase	Sample 2 – with addition of enzyme preparation based on endoxylanase
H_2O(%)	14.30	14.30	14.30
HYDHA(%b	51.8% b	55.3 % b 15%	53.6% b 15%
15% H_2O)	15%		
PRMAX(mb)	1927	2091	1994
TPRMAX(s)	135	214	155
TOL(s)	198	352	246
D250(mb)	349	26	189
D450(mb)	596	442	639
WAC(%)	57.6 % b 15	53.3 % b 15%	55.3 % b 15%

In Tabel 1. there are presented the characteristics of dough samples obtained by consistographic method.

The dough Sample 1- with addition of enzyme preparation based on glucoamylase show an improvement of the rheological characteristics compared to Sample 0, such as: the maximum pressure ($PRMAX_1$) which shows an increase of 164mb; the time for complete hydration ($TPRMAX_1$) which is 214s; the tolerance of the dough (TOL_1) that shows an increase of 154s, $D250_1$ has decreased to 26mb and $D450_1$ also decreased to 442mb.

Comparing Proba 1 - with the addition of glucoamylase with Sample 2 - with the addition of endoxylanase we observed that the addition of enzyme mixes based on glucoamylase had a positive effect on Sample 1 by increasing the maximum pressure ($PRMAX_1$) by 97 mb than $PRMAX_2$

The corresponding enzyme preparation enhances flour quality and provides an appropriate characteristics for bread dough by creating a dough which is more easly handled during the mechanical process, which becomes more dry, elastic, resulting in loose bread, developed volume and a core containing pores with thin walls.

b) regarding the baking test the results are as follows (Table 2):

The sample Proba 1 - with the addition of glucoamylase has the best volume and Proba 2 - with addition of endoxylanase is at 1.4 cm difference. Unlike Proba 0 – with no added enzyme preparation which has a very low volume and cracks in the crust, the other two samples show a significant increase in volume and pleasant look

Table 2. The baking test results of the dough samples: Sample 0 - no added enzyme preparation; Sample 1 – with addition of enzyme preparation based on glucoamylase; Sample 2 – with addition of enzyme preparation based on endoxylanase.

Sample / Parameter	Sample 0 - without enzyme	Sample 1 – with gluco-amylase	Sample 2 – with endo-xylanase
Volume (h – cm)	5.9	7.9	6.5

In the figures below we can notice the significant difference of the breads made from this samples:

Figure 4. Sample 0 - no added enzyme preparation

Figure 5. Sample 1 – with addition of glucoamylase

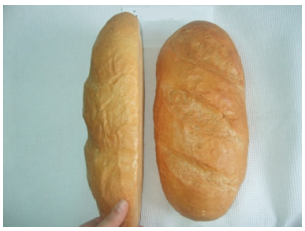

Figure 6. Sample 2 – with addition of endoxylanase

4. Conclusion

The additive actions of complex enzymes as ameliorator on flour have positive effects on the rheological characteristics of dough. The technological characteristics of the flour and the nutritive value of the bread are characterized by the following variables: initial volume, fermentation time, flexibility, the dough condition to fermentation, water retention, maximum resistance, extensibility, final rise to baking, final volume of the bread, nutritive value, and energy value. In order to improve these variables, different additives and substances are used in the bread manufacture, some of these being native components of the flour. The consistograph test provides results that are common specifications used by flour millers and processors to ensure a more consistent process and product. The consistograph is well suited for measuring the dough characteristics of weak gluten wheat.

The two enzyme preparations used are designed to improve the quality of dough used in the preparation of bakery products. They do not change the manufacturing process and do not have a high cost. Choosing suitable enzyme preparation is based on dough rheological characteristics and the correct amount to be added to the dough will be one that will bring maximum value for these features.

Glucoamylase acts against aging bread, but also helps prevent steakiness of core. Also works during Maillard reactions between low molecular weight dextrins and protein materials, and can lead to a darker bread crust.

Endoxylanase contribute to greater stability through effects against aging, increased warranty period, improved freshness and improve the texture of bread with a softer core, fine and uniform pore structure, stability of frozen dough, increased crispiness and decreased wettability. Are also used to increase the volume of bread products with high content of fiber, to improve the quality of bread flour. The color and flavor are also improved due to the browning effect of the crust and whitening effect of the core.

Enzyme preparations are used to obtain quality breads and more natural. These products are of most interest to all consumers.

Compliance with Ethics Requirements

Authors declare that they respect the journal's ethics requirements. Authors declare that they have no conflict of interest and all procedures involving human and/or animal subjects (if exists) respect the specific regulations and standards.

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