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Effects of Lipase, Phospholipase and DATEM on some Quality Characteristics of Bugget

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ABSTRACT

The effect of lipase, phospholipase and, diacetyl tartaric esters of mono-glycerides (DATEM) on the bugget made from white wheat flour was studied. The aim of this study was to investigate the effect of microbial lipase and phospholipase enzymes in different concentration (10,30 and 50 ppm) on the quality of bugget and compare their effect with DATEM (0.25,0.5 and 0.75%) on bugget`s physicochemical characteristics such as volume, crust colour, crumb texture and colour. Results showed that Addition of 0.75% DATEM and all concentration of lipase significantly (P< 0.05) increased the bread volume. Addition of lipase and phospholipase enzymes in all tested concentrations resulted in significant difference of the crust color compared to the control (P<0.05), also Addition of all improvers in different concentrations resulted in significant difference of the crumb color. Regarding to staling the bread, 0.5% and 0.75% DATEM as well as 10 ppm lipase, significantly could retard stailing phenomena in bread. Lipase might be introduced as a natural alternative for replacing syntetic DATEM in future.

Keywords: DATEM, Lipase, Phospholipase, Emulsifier, Bugget

INTRODUCTION

Bread is a basic dietary item dating back to the Neolithic era, which is prepared by baking, which is carried out in oven. The first bread was made around 10,000 years BC or over 12,000 years in the past, which may have been developed by deliberate experimentation with water and grain flour [13].

In the past few decades, demands for longer shelf-life and consistent quality in baked goods lead to application of a wide range of additives in the baking industry. These additives, including emulsifiers, enzymes, oxidants and reductants, are essential for improving dough machinability, reducing resting time, and improving baked goods' shelf-life [12]. Diacetyl tartaric esters of mono-glycerides (DATEM) constitute an emulsifier that has been widely used as a bread improver. DATEM improves bread volume, texture and also dough stability [6]. Compared to emulsifiers and chemical agents which are detectable in the final loaf, enzymes become denatured during baking and become undetectable in the final products. This allows enzymes to be clean label improvers; they provide bread

improving functions without appearing on the label. Thus, partial or complete replacement of traditionally used volume-improving agents, such as emulsifiers with enzyme-based volume improvers, is desirable [12]. Lipases have been used over the past two decades, along with other enzymes and emulsifiers, to improve some characteristics of baked goods. They hydrolyze triglyceride esters and produce mono- or di-glycerides, glycerol and free fatty acids. Most lipases act only on specific positions in triglycerides and have fungal or bacterial sources. Lipases strengthen dough stability and increase bread volume, texture and shelf-life [9].

In the 1990s, the first generation of lipases was introduced to the market. These lipases hydrolyze the ester bond between glycerides and fatty acids in positions 1 and 3 of triglycerides, producing free fatty acids and mono-glycerides, and producing more polar lipids in the dough [15]. Lipase strengthened the gluten network resulting an increase in loaf volume. Flour lipids, though representing around 2% of flour mass, play an important technological role because they interact with proteins and starch in dough, influencing the rheological properties of dough, bread quality and its freshness. The properties of lipids and particularly of phospholipids are given by the structural and functional particularities of molecules. It is considered that phospholipids are amphipathic substances because there is no polarity in the core of molecule, and because the extremities have opposite poles. A phosholipase is an enzyme that converts phospholipids into fatty acids and other lipophilic substances. It is known that phospholipids contain in their molecules glycerol, fatty acids, phosphoric acid and preferential nitrogenous bases, reason for which they are amphophilic properties [9]. Depending on the type of polyalcohol in their structure - glycerol, inositol or sphingosine, phospholipids have been classified as, glycerophospholipids, inosiphospholipids and sphingphospholipids Phospholipids also contain in their molecules both saturated and unsaturated fatty acids. Usually, in the phospholipid molecule we can only find a single rest of phosphoric acid, scarcely two. Colin, ethanolamine and serine are predominant as nitrogenous bases in phospholipids. The extremity that contains the rest of phosphoric acid and the nitrogenous base forms the hydrophilic part of the molecule due to the existent polar groups, whereas the acyl groups resulted from the fatty acids form the hydrophobic part. Depending on the phospholipases' action on phospholipids (fig 1), these enzymes are divided into four groups as phospholipases A1 (PLA1), phospholipases A2 (PLA2), phospholipases B (PLB), phospholipases C (PLC) also named lecithinases C or glycerophosphatase and phospholipases D (PLD) [9].

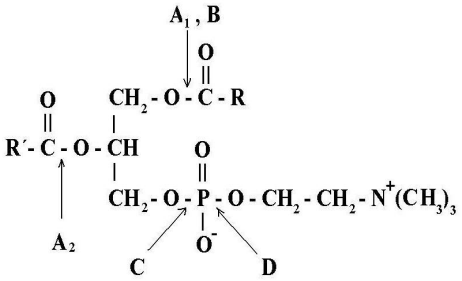

Fig 1. Phospholipases n phospholipids[9]

Because the application of lipase and phospholipase enzymes, as a bread improver, is relatively new and there have been only a few investigations of this category of baking enzymes, in this study we compare different concentration of lipase, phospholipase and DATEM to each other in order to study their effects on the baking main characteristics of final bread [12].

MATERIALS AND METHODS

2-1. Materials

2-1-1. Flour

Wheat flour was supplied by khoy avren Milling. The chemical composition of flour is shown in Table1.

Table1: Chemical composition of wheat flour

Chemical composition	%
Fat	1.15
Protein	11.09
Wet Gluten	24.60
Ash	0.56
Moisture	14.30

2-1-2. Improvers

The lipase enzyme was supplied by Sigma-Aldrich company (Germany) and phospholipase enzyme was supplied by DSM company (Netherlands). DATEM emulsifier, was supplied by Valencia company (Netherlands). Baking recipes with addition of DATEM (0.25 ,0.5and 0.75%), lipase and phospholipase (10, 30and 50ppm for each one) were test-baked to study their effects on the baking characteristics (Table 2).

Table2: Different treatments for bread formulation

Trade name	Unit	Unit	Unit	unit	%	ppm	Ppm
	Flour	Salt	Active dried yeast	Water	DATEM	Lipase	Phospholipase
R_1	**100**	**1**	**2**	**50**	-	-	-
R_2	**100**	**1**	**2**	**50**	**0.25**	-	-
R_3	**100**	**1**	**2**	**50**	**0.5**	-	-
R_4	**100**	**1**	**2**	**50**	**0.75**	-	-
R_5	**100**	**1**	**2**	**50**	-	**10**	-
R_6	**100**	**1**	**2**	**50**	-	**30**	-
R_7	**100**	**1**	**2**	**50**	-	**50**	-
R_8	**100**	**1**	**2**	**50**	-	-	**10**
R_9	**100**	**1**	**2**	**50**	-	-	**30**
R_{10}	**100**	**1**	**2**	**50**	-	-	**50**

2-2. Methods

2-2-1. Baking Test

The flour and salt as dry ingredients well blended in a spiral mixer for 20 s. Water was added, followed by 30 s of mixing at low speed and then yeast and improvers were added. After a mixing time of 12.5 minutes the dough were cut into 250 g pop loaves. The pop loaves then were molded and rested in a cabinet at room temperature for 20 min. fo the final proofing the trays were placed into a proofer by controlled condition of 35-38 ^{o}C and 70-75% relative humidity for 45 minutes. Breads were baked in an electric oven at 180^{o}C for 15 min.

2-2-2. Bread analysis

Bread volume, crust color, crumb texture and color of the baked bread as the main factors were considered.

After cooling the breads, volume of loaves were measured using the seed replacement method [2]. The color of bugget was measured using Hunter Lab (colorflex4.510usa), and using the L*a*b* system. color was determined by value of $\Delta E = \{(L_{Standard} - L_{Sample})^2 + (a_{standard} - a_{sample})^2 + (b_{standard} - b_{sample})^2\}^{/5}$[4].

Crumb firmness was determined on the same loaves according to 74-09 AACC method, using a texture analyser, model H5KS (Hounsfeild , England). The crumb firmness test was performed on the one ,three and five days after baking. The maximum force was 500 (N), 100 mm/s speed, end point 4(mm) and Extension range 6, and finally compression of one bread slices (2×2 cm thickness) was defined as firmness.

RESULTS AND DISCUSSION

3-1. Volume

Addition of 0.75% DATEM and all concentration of lipase significantly ($P< 0.05$) increased the bread volume compared with the control but addition of phospholipase in higher concentration (30 and 50 ppm) decreased the bread volume significantly (fig 2).The maximum volume was observed for 0.75% DATEM and 30 ppm lipase. DATEM and lipase enzyme increases the expansion of the gluten network, increases the wall thickness and decreases cell density and improving the gas retention in bread [6, 15]. Application high concentration of lipase

causes decreasing the volume, which could be explained by longer action of this strong lipase on the dough, producing stiffer dough, which leads to decrease in bread volume [12].

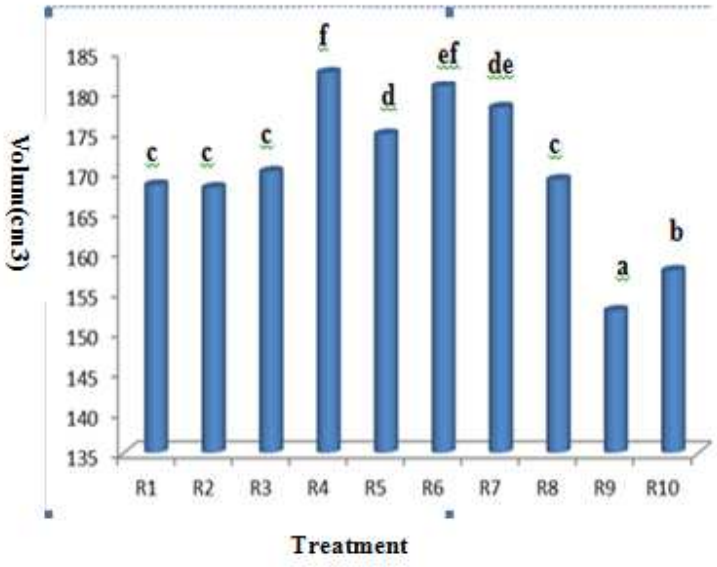

Fig 2. Effect of bread improvers on bread volume

3-2.Crust Color

Addition of lipase and phospholipase enzymes in all tested concentrations resulted in significant difference of the crust color compared to the control ($P<0.05$). Brown crust in bread is as a result of non-enzymatic browning reaction (Maillard type) between amino acids and reducing sugars [12,5]. Unlike lipase and phospholipase, which produces more lipids in the dough, resulting in crust darkness, DATEM doesn't change the carbohydrate profile of the dough (fig 3).

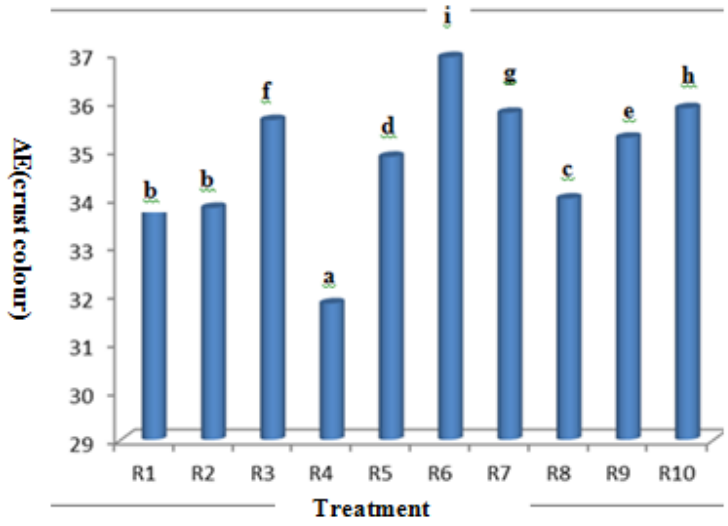

Fig 3. Effect of bread improvers on bread crust color

3-3. Crumb color

Addition of all improvers in different concentrations resulted in significant difference of the crumb color compared to the control (fig4). There were significant differences among all the samples' crumb color ($P<0.05$). Crumb color is related to flour type and to crumb air cell structure which affect the way that light reflects from a piece of bread.

DATEM, lipases and phospholipase change the structure of the bread crumb and make the air cells smaller and more evenly distributed [16].

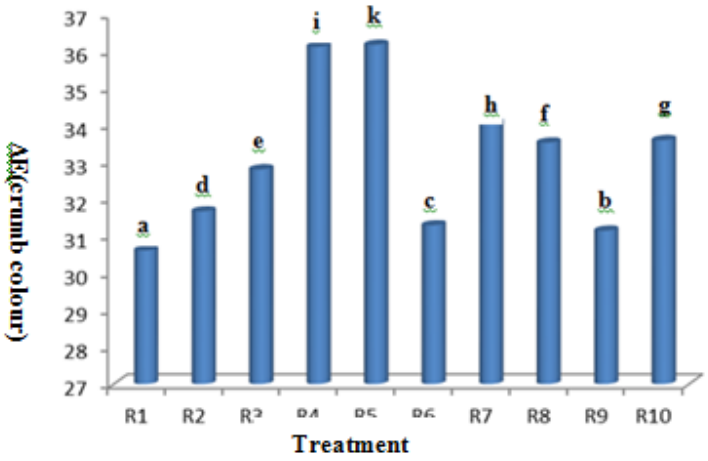

Fig 4. Effect of bread improvers on bread crumb color

3-4. Crumb texture

The results showed that 0.5% and 0.75% DATEM as well as 10 ppm lipase, significantly reduced the firmness ($P< 0.05$). Regarding the effect of DATEM and lipase on the staling rate of breads, it might be concluded that with the addition of these surfactants the staling rate of bread will be delayed compared to the control [2, 5, and 12]. Retardation of the staling rate of bread is in the highest level when 0.75% DATEM or 10ppm lipase is added into the dough (fig 5). Colakoglu 2012 reported that DATEM and lipase reduced the staling rate. But Phospholipase hydrolyze complex phospholipid - amylose in dough and expedite staling [14].

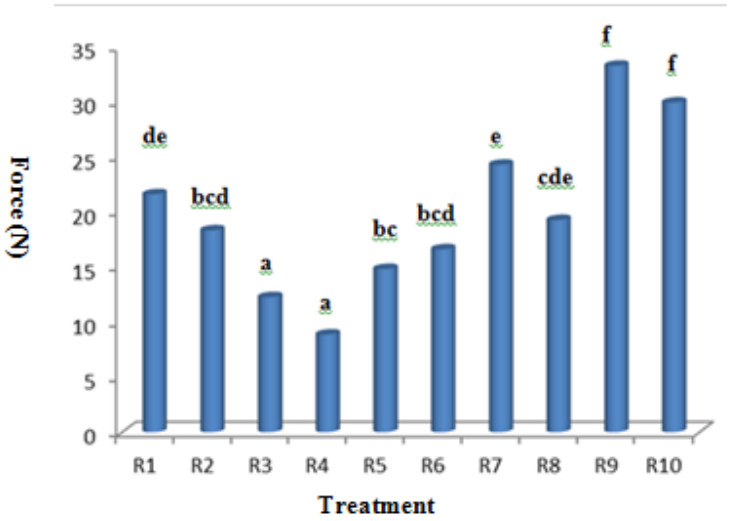

Fig 5. Effect of bread improvers on bread crumb texture

CONCLUSION

Nowadays chemical emulsifiers consumption, specially DATEM is increasing in order to improve the quality and extend the shelf life of bakery products. As these chemical components are pernicious for the consumer, using of bioemulsifiers to replace their advantages with chemical emulsifiers disadvantages might be efficient to have a

healthy product. Recently microbial Lipase and Phospholipase enzymes are being stuidied as a bioemulsifier in cereal industry. According to our results using 0.75 % DATEM has more positive effect on texture than other treatments and lipase in concentrations of 10 and 30ppm is better than using 0.25% DATEM, but by increasing keeping time, the effect of 10 ppm lipase on tough reduction is similar to 0.5 and 0.75% DATEM. Bread produced by 0.75% DATEM showed the maximum volume and the effect of 10 ppm lipase on volume was more than 0.25 and 0.5% DATEM. The color examination revealed that using 10 ppm lipase causes the best crumb color and application of 30 ppm lipase is the best crust color of the all treatments ($P<0.05$). Application of 10 ppm phospholipase, showed an amelioration in all examinations as compared with control experiment, but it was in a lower statistical group with regard to the two other ameliorators. Hence buggets might be produced by replacement of DATEM emulsifier with lipase enzyme by keeping it`s desired characteristics and more investigations on replacement of phospholipase enzyme is needed.

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