



Full Length Article

Improving Bread Quality by Carboxymethyl Cellulase Application

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ABSTRACT

The concept of organic foods is getting popularity these days. Chemical additives have shown ill effects on human health. Food processing employing biochemicals can compensate the problem as these are easily denatured by heat treatment therefore, render the foods chemical free and safer. Locally produced carboxymethyl cellulase was used in bread making process that showed significant effects on farinographic and mixographic parameters. The enzyme addition during bread making caused an increase in the bread volume up to 250 IU/100 g flour (T₂) and the maximum volume observed at this level was $635 \pm 25.30 \text{ cm}^3$ as compared to the control ($550 \pm 15.50 \text{ cm}^3$). Likewise, the highest specific volume ($3.99 \pm 0.14 \text{ cm}^3/\text{g}$) was observed in 250 IU/100 g flour with $0.265 \pm 0.010 \text{ g/cm}^3$ density. The use of CMCase treated flour resulted in improved internal and external characteristics of bread loaves. In most of the cases, 250 IU/100 g flour showed better performance. The results showed that CMCase can effectively be used in bread making for improvement in quality.

Key Words: CMCase; Bread; Volume; Enzyme; Sensory evaluation

INTRODUCTION

Enzymes applications have grown to be a common practice in the baking industry with advantage of being considered as natural additives (Penstone, 1996). The hydrolytic enzymes are being used in the baking industry to improve dough-handling properties; xylanases and cellulases (β -glucanases) have gained popularity during the last few years (Harada *et al.*, 2005).

CMCase has pre-dominant role in the bread industry as their imperative role in pentosans hydrolysis. No doubt, pentosans are minor component of wheat flour, but play an important role in dough rheology and bread quality due to their high water binding capacity. Soluble pentosans are implicated in the dough elasticity and the hydrolysis of the insoluble pentosans promotes changes on dough rheology due to the release of water, which can be used to form the gluten network. It has been observed that enzymatic hydrolysis of non-starch polysaccharides like pentosans leads to an improvement of the rheological properties of dough, bread loaf volume and crumb firmness (Martinez-Anaya & Jimenez, 1997). Although some researchers used CMCase in bread production, these studies employed commercial enzymes. Moreover, these studies do not provide information regarding the optimum level of enzyme application rather these comprised the comparative studies of cellulase at single level with other hydrolases that may be insufficient to evaluate the enzyme efficiency. This study was therefore conducted to attain best levels of locally

produced enzyme for addition in the bread making process to increase volume and improve quality.

MATERIALS AND METHODS

The raw materials including wheat flour, sugar, shortening, yeast etc. were purchased from the local market. CMCase produced in the Microbiology and Biotechnology Laboratory of the National Institute of Food Science and Technology was used in the bread making process.

Proximate analysis. For proximate analysis of flour samples, the respective methods of AACC (2000) were followed to determine various parameters including moisture (Method No. 44-15 A), ash (Method No. 08-01), crude fat (Method No. 30-10), crude protein (Method No. 46-10), crude fiber (Method No. 32-10) and NFE.

Dough rheological studies. The rheological behavior of supplemented flours was evaluated by farinographic and mixographic studies following the AACC Method 54-21 and 54-40, respectively (AACC 2000).

Bread production. The breads were prepared in the Bakery Section of the National Institute of Food Science and Technology, University of Agriculture, Faisalabad, using straight-dough method (AACC, 2000). Carboxymethyl cellulase (CMCase) was used in the recipe in different concentrations as mentioned below in Table I.

Sensory evaluation. The sensory evaluation of the enzyme treated bread was conducted by a trained taste panel of 10 judges to find out the effect of the enzyme addition on

different external as well as internal characteristics of bread following the procedure of Matz (1972).

RESULTS

Proximate analysis of flour. The Table II shows the proximate composition of flour used for the bread making.

Farinographic studies. The farinographic parameters like water absorption (WA), arrival time (AT), dough development time (DDT), dough stability time (DST) and mixing tolerance index were studied under the influence of different doses of CMCase. The mean squares Table III explicated that the enzymatic treatment showed significant effect on all of the farinographic parameters except water absorption that showed non-significant variations among the treatments (Table IV).

The means indicated that the water absorption (WA) increased non-significantly with increase in enzyme concentration and maximum water absorption was $59.6 \pm 0.11\%$ in T_3 (500 IU/100 g flour) followed by $59.1 \pm 0.12\%$ in T_2 (250 IU/100 g flour), while minimum value for the parameter was calculated in case of control i.e., $57.9 \pm 0.8\%$. Arrival time (AT) of all the treatments decreased with increase in enzyme concentration. The control flour showed the maximum arrival time 1.9 ± 0.08 min, while minimum arrival time 1.3 ± 0.07 min was computed in T_3 (500 IU/100 g flour). As concerned with dough development time, it decreased gradually with increase in CMCase units. The flour without enzyme addition exhibited maximum dough development time (4.7 ± 0.02 min), whereas T_3 showed minimum DDT (3.9 ± 0.08 min). Likewise, when the enzyme units were increased gradually in the flour, the dough stability decreased accordingly. The control showed maximum dough stability (10 ± 0.13 min), while T_3 exhibited minimum (8.1 ± 0.08 min) stability time.

Mixographic studies. Table V depicts the mean squares of the mixographic studies. It can be seen that the treatments exerted significant effect on the mixing time and peak height. The mean values in Table VI elaborated the effect of different doses of enzyme (CMCase) on mixographic parameters. It is obvious that addition of the enzyme decreased the mixing time of the dough during experiment. The flour sample without enzyme exhibited the maximum mixing time as 5.50 ± 0.09 min followed by T_1 (5.1 ± 0.11 min), while T_3 (500 IU/100 g flour) exhibited minimum mixing time (4.25 ± 0.09 min). There were non-significant variations between T_1 (5.1 ± 0.11 min) and T_2 (5.00 ± 0.1 min), whereas T_0 and T_3 showed highly significant differences. In contrast to mixing time, a gradual increase in peak height was detected with the increase in level of enzyme; T_3 (500 IU/100 g flour) exhibited maximum peak height ($60.2 \pm 4.20\%$) followed by T_2 (60.10 ± 3.80) however, minimum value for peak height was calculated in T_0 i.e., $57.50 \pm 2.80\%$. It is obvious from the data that T_1 (60.00 ± 2.50) and T_2 (60.10 ± 3.80) showed non-significant

Table I. Different doses of CMCase used in bread production

| Treatment | CMCase (IU/100 g flour) |
|--------------------|-------------------------|
| Control | - |
| 100 IU/100 g flour | 100 |
| 250 IU/100 g flour | 250 |
| 500 IU/100 g flour | 500 |

Table II. Proximate composition of wheat flour

| Parameter | Value (%) |
|---------------|------------------|
| Moisture | 10.5 ± 0.02 |
| Total ash | 0.51 ± 0.01 |
| Crude fat | 0.98 ± 0.02 |
| Crude protein | 10.52 ± 0.03 |
| Crude fiber | 0.38 ± 0.01 |
| NFE | 87.61 ± 2.34 |

Table III. Mean squares for farinographic characteristics

| SOV | df | WA | AT | DDT | DST | MTI |
|------------|----|--------------------|--------|--------|--------|----------|
| Treatments | 3 | 1.95 ^{ns} | 0.21** | 0.368* | 2.008* | 386.75** |
| Error | 8 | 20.334 | 0.018 | 0.118 | 0.524 | 27.4259 |
| Total | 11 | | | | | |

*Significant; **Highly significant; ^{ns}Non-significant

Table IV. Means for farinographic characteristics

| Treatments | WA (%) | AT (min) | DDT (min) | DST (min) | MTI (BU) |
|--------------------|-----------------|------------------|------------------|------------------|---------------|
| Control | 57.9 ± 0.8 | $1.9 \pm 0.08a$ | $4.7 \pm 0.02a$ | $10 \pm 0.13a$ | $60 \pm 2.8a$ |
| 100 IU/100 g flour | 58.3 ± 0.11 | $1.8 \pm 0.04ab$ | $4.5 \pm 0.06ab$ | $9.5 \pm 0.11ab$ | $65 \pm 2.2a$ |
| 250 IU/100 g flour | 59.1 ± 0.12 | $1.6 \pm 0.04b$ | $4.2 \pm 0.07ab$ | $8.9 \pm 0.12ab$ | $77 \pm 2.8b$ |
| 500 IU/100 g flour | 59.6 ± 0.11 | $1.3 \pm 0.07c$ | $3.9 \pm 0.08b$ | $8.1 \pm 0.08b$ | $85 \pm 3.2b$ |

Table V. Mean squares for effect of treatments on mixographic characteristics

| SOV | df | Mixing time | Peak height |
|------------|----|-------------|-------------|
| Treatments | 3 | 1.8475** | 138.00* |
| Error | 8 | 0.0725 | 21.966 |
| Total | 11 | | |

*Significant; **Highly significant

Table VI. Means for mixographic characteristics

| Treatments | Mixing time (min) | Peak height (%) |
|--------------------|-------------------|--------------------|
| Control | $5.50 \pm 0.09a$ | $57.50 \pm 2.80a$ |
| 100 IU/100 g flour | $5.10 \pm 0.11b$ | $60.00 \pm 2.50ab$ |
| 250 IU/100 g flour | $5.00 \pm 0.10b$ | $60.10 \pm 3.80bc$ |
| 500 IU/100 g flour | $4.25 \pm 0.09c$ | $60.20 \pm 4.20c$ |

differences with each other significant variations were observed between T_0 ($57.50 \pm 2.80\%$) and T_3 ($60.20 \pm 4.20\%$) for peak height.

Effect of CMCase on bread quality. Bread loaves were prepared from respective flour samples containing different concentration of enzyme and evaluated for various quality attributes.

Volume and density of bread. After the bread making process, the volume of the loaves was measured by rapeseed displacement method. Maximum volume was observed in

Fig. 1. Volume of breads treated with different concentrations of CMCase (the results are average of triplicate samples \pm SD)

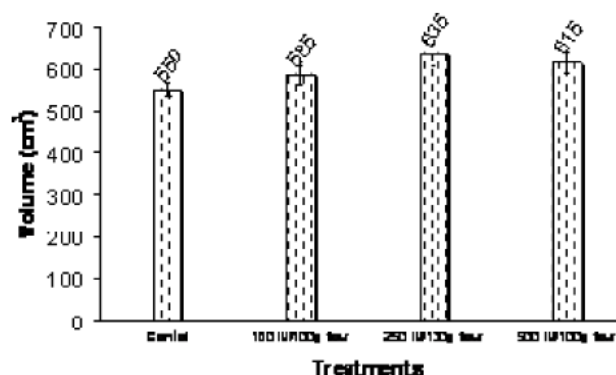


Fig. 2. Percent increase in volume of breads treated with different concentrations of CMCase (the results are average of triplicate samples \pm SD)

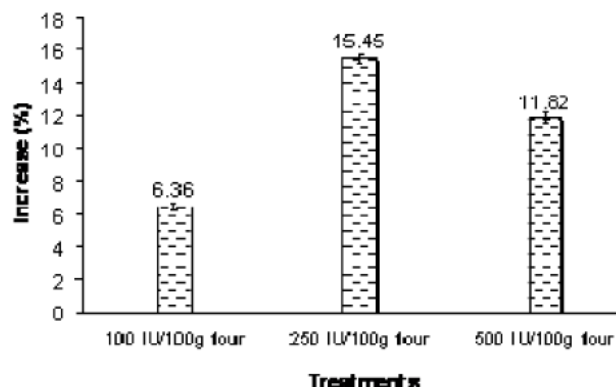
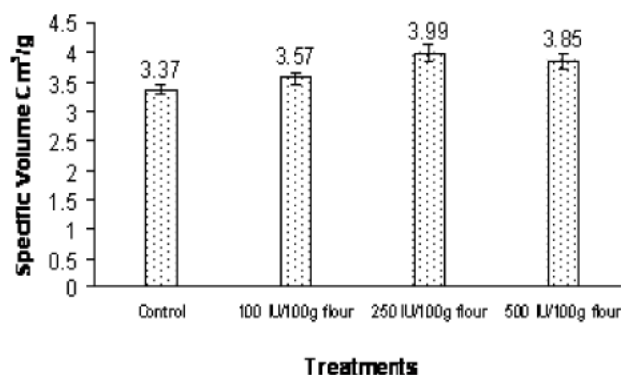
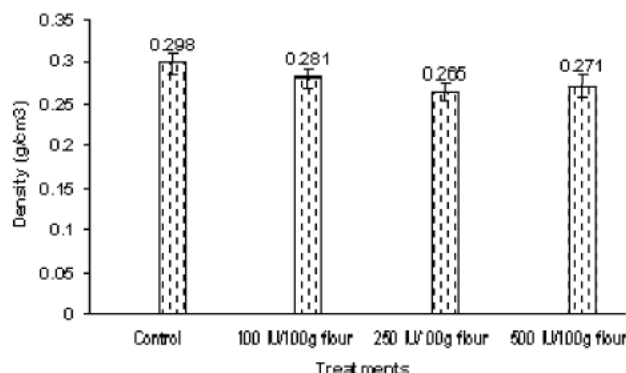


Fig. 3. Specific volume of the breads treated with different concentrations of CMCase (the results are average of triplicate samples \pm SD)



case of T_2 (635 ± 25.30 cm³) followed by T_3 (615 ± 25.60 cm³), whereas minimum value for this parameter was recorded in control (550 ± 15.50 cm³) as obvious from Fig. 1. At higher level, CMCase resulted in a decreased volume because of poor mixing and sheeting properties and dough became stickier. The dough becomes weaker and unable to

Fig. 4. Density of the breads treated with different concentrations of CMCase (the results are average of triplicate samples \pm SD)



retain the gas evolved during the fermentation, proofing and subsequently during baking process. Such dough may collapse during baking and results in decreased volume. Fig. 2 depicted the percent increase in the bread volume as compared to control; the maximum increase was calculated in T_2 ($15.45 \pm 0.35\%$) followed by T_3 ($11.82 \pm 0.30\%$), while minimum increase was recorded in T_1 ($6.36 \pm 0.25\%$).

Similarly, the specific volume of the bread samples was calculated and the best treatment in this regard was found to be T_2 (3.99 ± 0.14 cm³/g) followed by T_3 (3.85 ± 0.12 cm³/g), whereas T_0 was at the bottom with specific volume 3.37 ± 0.10 cm³/g (Fig. 3). The results pertaining to the specific volume of bread indicated that CMCase treatment resulted in breads with less density and high volume and hence, preferred by the consumers.

DISCUSSION

Hydrolytic enzymes are being used in the baking industry to improve dough-handling properties and enhance bread quality. The improvement at optimum levels of these enzymes has been attributed to both non-specific and specific effects. The former effect seems to be associated with improved (softer) dough-handling characteristics, improved oven spring and softer crumb due to the hydrolytic release of water from the respective polymeric substrates (Kulp, 1993; Rouau *et al.*, 1994; Martínez-Anaya & Jiménez, 1997; Harada *et al.*, 2000). The latter effect has been attributed to interactions between the enzyme-specific hydrolysis reaction products and other dough or bread components, resulting in better processing and bread characteristics (Martin & Hoseney, 1991; Biliaderis *et al.*, 1995; Bombara *et al.*, 1997). The relative importance of these effects is currently not well understood owing to a lack of studies comparing the performance of different types and sources of hydrolytic enzymes (Ranum & DeStefanis, 1990; Rouau *et al.*, 1994; Martínez-Anaya & Jiménez, 1997) and the confounding effects of other factors such as baking process and flour characteristics that strongly influence

response (Cauvain & Chamberlain, 1988; Rouau *et al.*, 1994).

For farinographic parameters, similar trend was observed by Haros *et al.* (2002), whereas Al-Suaidey *et al.* (1973) did not find any significant modifications in the wheat flour treated with CMCase. For mixing tolerance index (MTI), little variations among the treatments were observed, T_0 showed the minimum MTI as 60 ± 2.8 BU, whereas T_2 (250 IU 100 g⁻¹ flour) exhibited a value of 65 ± 2.2 BU however, maximum value for the mixing tolerance index (85 ± 3.2 BU) was calculated in T_3 (500 IU/100 g flour).

Regarding the volume and density, the results are supported by Gebhardt *et al.* (1982) and Diaconescu (2006) who reported increased bread volume after enzymatic treatment of the flour. The results are also in harmony to the findings of Harada *et al.* (2005), who prepared breads with volume up to 1050 cm³ by CMCase treatment of flour, whereas Laurikainen *et al.* (1998) recorded an increase of 18-19% in the bread specific volume by treating the flour with CMCase and hemicellulase. Correa *et al.* (1997) also found pronounced effect of the CMCase on the volume of bread made from flour with 10% wheat bran addition. Haros *et al.* (2002) applied CMCase at the time of tempering of wheat. When the flour obtained from this wheat was used in bread production, significant increase in the volume was obtained. The enzyme treatment of flour resulted in lighter breads whereas; the control had higher density. Minimum value for the density was observed in T_2 (0.265 ± 0.010 g/cm³), whereas maximum was calculated in the control with 0.298 ± 0.013 g/cm³ density (Fig. 4). The lesser density of bread reflects the good texture of the loaves with better grain formation and also optimum baking of the product.

CONCLUSION

Application of CMCase in the bread production can result improvement in the white pan bread as found in this case. In case of pan bread loaf volume is of utmost importance as it determines the acceptability of the end product. The enzyme caused increase in volume of the bread loaves that advocates its suitability to be used in bread production to replace chemical bread conditioners.

REFERENCES

- AACC, 2000. *Approved Methods of American Association of Cereal Chemists*. AACC, Inc., St. Paul, Minnesota
- Al-Suaidey, M.A., J.A. Johnson and A.B. Ward, 1973. Effects of certain biochemical treatments on milling and baking properties of hard red winter wheat. *Cereal Sci. Today*, 18: 174-179
- Biliaderis, C.G., M.S. Izydorczyk and O. Rattan, 1995. Effect of arabinoxylans on bread-making quality of wheat flours. *Food Chem.*, 53: 165-71
- Bombara, N., M.C. Añón and A.M.R. Pilosof, 1997. Functional properties of protease modified wheat flours. *Lebensm. Wiss. Technol.*, 30: 441-447
- Cauvain, S.P. and N. Chamberlain, 1988. The bread improving effect of fungal α -amylase. *J. Cereal Sci.*, 8: 239-248
- Correa, G. and R.P. Tengerdy, 1997. Production of cellulase on sugar cane bagasse by fungal mixed culture solid substrate fermentation. *Biotechnol. Lett.*, 19: 665-667
- Diaconescu, D., 2006. *The Effect of Glucose Oxidase, Hemicellulases, Xylanases, Cellulases, Amyloglucosidase and α -Amylase Upon Bread Quality*. COST Action 928 P9 WG 1-3 meeting, Iceland
- Gebhardt, E., C. Neumann and I. Eckert, 1982. Modification and derivatization of gluten and influence on the manufacture of baked goods. *Baecker Konditor*, 30: 37-39
- Harada, O., E.D. Lysenko and K.R. Preston, 2000. Effects of commercial hydrolytic enzyme additives on Canadian short process bread properties and processing characteristics. *Cereal Chem.*, 77: 70-76
- Harada, O., E.D. Lysenko, N.M. Edwards and K.R. Preston, 2005. Effects of commercial hydrolytic enzyme additives on japanese-style sponge and dough bread properties and processing characteristics. *Cereal Chem.*, 82: 314-320
- Haros, M., M.R. Cristina and B. Carmen, 2002. Effect of different carbohydrases on fresh bread texture and bread staling. *European Food Res. Technol.*, 215: 425-430
- Kulp, K., 1993. Enzymes as dough improvers. In: Kamel, B.S. and C.E. Stauffer (eds.), *Advances in Baking Technology*, pp: 152-178. Blackie Academic and Professional, London
- Laurikainen, T., H. Harkonen, K. Autio and K. Poutanen, 1998. Effects of enzymes in fibre-enriched baking. *J. Sci. Food Agric.*, 76: 239-249
- Martin, M.L. and R.C. Hoseney, 1991. A mechanism of bread firming II. Role of starch hydrolyzing enzymes. *Cereal Chem.*, 68: 503-507
- Martinez-Anaya, M.A. and T. Jimenez, 1997. Functionality of enzymes that hydrolyse starch and non-starch polysaccharide in breadmaking. *European Food Res. Technol.*, 205: 209-214
- Matz, S.A., 1972. *Bakery Technology and Engineering*. The AVI Publishing Company Westport, CT
- Penstone, K., 1996. Zooming in on enzymes. *Food Rev.*, 23: 36-41
- Ranum, P. and V.A. DeStefanis, 1990. Use of fungal α -amylase in milling and baking. *Cereal Foods World*, 35: 931-933
- Rouau, X., M.L. El-Hayek and D. Moreau, 1994. Effect of an enzyme preparation containing pentosanases on the bread-making quality of flours in relation to changes in pentosan properties. *J. Cereal Sci.*, 19: 259-272

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