

Effects of protein variation on starch crystallinity and bread staling

M. Salehifar¹

ABSTRACT. Three flours with proteins of 9.4 , 11.6 and 13.5% were prepared and Flat breads were baked according to formulation. Wheat starch crystallinity was studied using differential scanning calorimetry (DSC). The crystallization enthalpy, peak temperature (T_p) and onset point temperatures (T_o) of the DSC thermograms were analyzed during storage in breads. Bread strips were submitted to Kramer shear test, and firming of breads were determined during storage. Results indicated that high protein breads showed lower extent of retrogradation. Study showed that low protein flours staled faster due to high concentration of starch and low water absorption.

Keywords: Flat bread, protein, starch crystallinity, staling

1. Introduction

Starch is a key factor in the staling phenomenon. Native starch is present as semi crystalline granules (Faergestad ,2000). During the starch gelatinization, the granules swell and lose their molecular order. (Carcea, 1996). During the baking, the starch is gelatinized, and the semi crystalline structure changed to amorphous structure. During cooling and storage of the bread, the starch crystalline structure is recovered limitedly which caused significant changes in the mechanical properties of the bread. (Primo martin, 2007). Generally the consumer acceptability of bread depends in part on its mechanical properties such as firming and staling. It was suggested that starch retrogradation, is also implicated in staling (Martin 1991). Gluten and its interactions with starch may have a role in the staling process (Armero 1998 and Roa 1992). Studies showed that the mechanism of staling, deals with the interactions between starch granules and proteins and it was indicated that staling rate can be delayed by increasing the amount of

DSC(Differential Scanning Calorimetry) was recently used to investigate the a crystallization and retrogradation of starch (Abd karim,2000).

In this study the crystallinity of starch and the staling, in breads which were produced by three different flours varied in protein contents, was studied.

2. Material and Methods

2.1. Chemical Analysis of flours

Three flours with proteins of 9.4. (t_1), 11.6 (t_2) and 13.5% (t_3) were prepared and the protein, ash and moisture content and farinograph test were determined according to AACC procedures 46-12,08-01, 16-44 A, and 54-21 respectively (AACC, 2000).

2.2. Bread baking

Flat breads were prepared at the baking lab using: wheat flour, dry yeast (0.4%), salt (1.5%) (Primo martin, 2007). Ingredients were blended for 1.5 min at low speed (150 rpm). Water was added and mixed at medium speed (200-220 rpm) for 6 min until final dough prepared. The dough was allowed to rest for 45 min

¹ Academic member of Islamic Azad University , Food Science and Technology department, shahriyar- shahre ghods branch.
Tel(00982122428135), Fax(00982122428134)
Email address: M_salehi1978@yahoo.com

and was then divided and rounded and rest again for 12 min. Proofing was performed at 30-32 C and 80% RH until a fixed volume of gas was produced. Breads were baked at 330 C for 75 sec , allowed to cool at ambient temperature (25) C and packed in a sealed polyethylene films and stored in 25 C.

2.3. Differential Scanning Calorimetry (DSC)

30±5 mg of samples were weighted in a hermetically sealed aluminum pan and analyzed. Samples were heated at 25-200 C and the rate of 5 C/min. The onset (T_o) and peak (T_p) temperatures and the transition enthalpy (J/g) of recrystallization were calculated (AACC 2000).

2.4. Bread staling

Bread strips (6 × 7.5 cm²) were submitted to Kramer shear test on an Instron universal Testing machine according to AACC 74-09, load cell: 500 N, extension range: 25 mm, test end point: 12 mm, test speed: 120 mm/minute and maximum point in curves was read and firming of breads were determined in 1,3,5,7 days of storage. (AACC., 2000).

2.5. Statistical analysis

The significance of each treatments in samples was analyzed using analysis of variance (ANOVA), and if ANOVA was significant, Tukey test were used. SPSS V.13 performed at 95% confidence level.

3. Results and discussion

General composition analysis of wheat flours showed in Table 1. Ash content of flours had no significant differences and variations in moisture content was just for different environment humidity conditions and was not affect the flour specifications, but differences in protein content were detectable and affect flour characteristics.

Table 1. composition analysis of flours.

sample	Chemical Analysis		
	Moisture (%)	Ash (%)	Protein (%)
t1	11.3 ^a	1.3 ^a	9.5 ^a
t2	11.4 ^a	1.1 ^a	11.5 ^b
t3	11.9 ^b	1.1 ^a	13.5 ^c

* In each column averages with the same characters (a,b,..) have no significant differences at 5%level

The relative recrystallization of starch in breads was shown in Table 2. The DSC thermograms acquired in 1 day storage and 3 days storage showed that wheat starch recrystallization enthalpy increased during storage time which was due to the retrogradation of starch. Fig 1 Shows the effect of storage on retrogradation, indicated the increasing in retrogradation during bread staling (Ribotta, 2007).

Table2. Enthalpy variations during storage.

Sample	Storage time	Enthalphy (J/g)
Bread	1 day storage	318.35 ^a
	3 days storage	481.35 ^b

* In each column averages with the same characters (a,b,..) have no significant differences at 5%level

During storage, crystals have time to recovered and regain their crystallinity. So by increasing the storage time, there could be much more recovered crystals which need high energy for melting . (Jocobs., 1998).

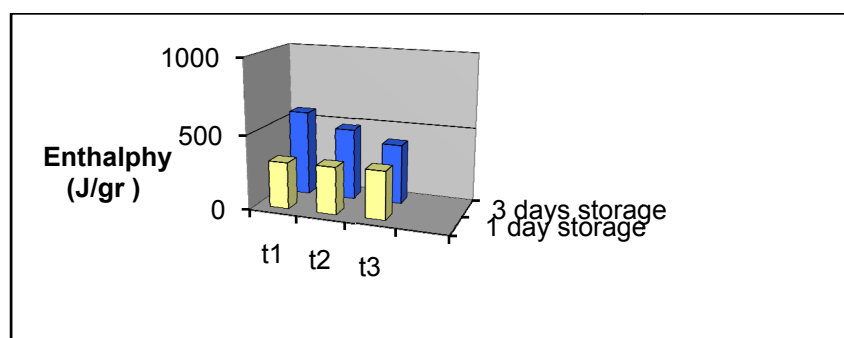


Fig 1. Effect of bread storage on retrogradation.

Table 3 showed peak temperature and onset point temperatures in B₂ and B₃ breads.

Table 3. Peak and onset point temperatures of breads during storage time.

Bread	1 day storage		3 days storage	
	T _p (c)	T _o (c)	T _p (c)	T _o (c)
	102.45 ^b	96.79 ^a	112.03 ^b	85.125 ^b

T_p: peak temperature, T_o: onset point Temperature All values were means of three replicates.

Typical DSC thermograms of starch-water mixture showed three endothermic peaks (Fukuoka., 2002). The first peak (55-60C) corresponds to gelatinization of starch crystallites. Second peak (about 65-75 C) is the melting of remaining crystallites and the third peak (about 100C) has been attributed to dissociation of amylase- lipid complex. Bread thermograms, showed a single peak at about 100C which was due to dissociation of recovered crystals (recrystals). The absence of first and second peak and presence of third peak at the temperatures about 100C indicated that starch had completely gelatinized during baking.

DSC thermograms of the flours showed gelatinization peak at about 65-70 C while DSC thermograms of bread showed the melting of remaining crystals (which have higher melting temperatures and could not be gelatinized during baking) or the melting of recrystals (that need the highest melting temperatures). Breads had high peak and onset point temperatures which showed that due to the longer time of baking, more starch granules gelatinized so during storage most crystals were recovered crystals which were stronger and had higher melting temperatures, reflecting in high peak and onset point temperatures. The effect of protein on the extent of retrogradation was studied by DSC. Table 4.

Table 4. Enthalpy variations in breads produced by flours of different protein contents during storage.

Flour	Enthalpy (J/g)	
	1 day storage	3 days storage
t ₁	312.69 ^a	571.42 ^c
t ₂	317.68 ^a	478.29 ^b
t ₃	323.62 ^a	400.24 ^a

** In each column averages with the same characters (a,b,..) have no significant differences at 5%level*

Table 4 Shows the effect of bread staling on enthalpy or retrogradation, indicated the increasing in enthalpy during bread staling (Ribotta, 2007). During storage, crystals have time to recovered and regain their crystallinity. So by increasing the storage time, there could be much more recovered crystals which need high energy for melting (Jacobs., 1998). It was found that the recrystallization enthalpy of starch as measured by DSC, decreased with increasing levels of protein (Eliasson., 1995). This was explained by the distribution of water within the mixed system(see table 5).

Table 5. Farinograph results.

Flour	Farinograph parameters			
	Water absorption (%)	Dough development time (min)	Dough resistancy (min)	Dough softening (20 min)
t1	64.05 ^a	1.875 ^a	2.25 ^a	117 ^{ab}
t2	66.05 ^b	3.625 ^b	3.62 ^b	117 ^a
t3	74.50 ^c	3.625 ^b	4.50 ^c	110 ^a

* In each column averages with the same characters (a,b,..) have no significant differences at 5%level

As can be seen, increasing protein content of flours caused increase the water absorption of flour. Protein could alter the amount of water available to the starch. (Eliasson.,1997, Osella., 2007). It can be suggested that this was due to the dilution of starch by increasing protein, and so the retrogradation decreased (Ottenhof., 2004). Water absorption in t1 flour with protein content of 9.5% was 64.05% compared with flour t3 with 13.5%, protein was 74.5% which showed significant difference. p2 and p3 with the same protein quality, showed different water absorption, around 66.05 and 74.5% respectively.

Differences in the water absorptions of t2 and t3 were due to the different protein contents. Studies showed that higher protein quantity, affect dough rheological parameters. Dough development time and softening in t2 and t3 flours had no significant difference but dough resistance showed detectable difference, which was 3.62 and 4.5 respectively. Table 6 showed that the minimum and maximum Enthalpy (retrogradation) was observed in breads produced by t3 and t1 flours respectively.

table 6. Total Enthalpy variation of breads produced by flours of different proteins

Flour	Total Enthalpy (J/g)
	Bread
t ₁	442.05 ^c
t ₂	397.99 ^b
t ₃	361.93 ^a

* In each column averages with the same characters (a,b,..) have no significant differences at 5%level

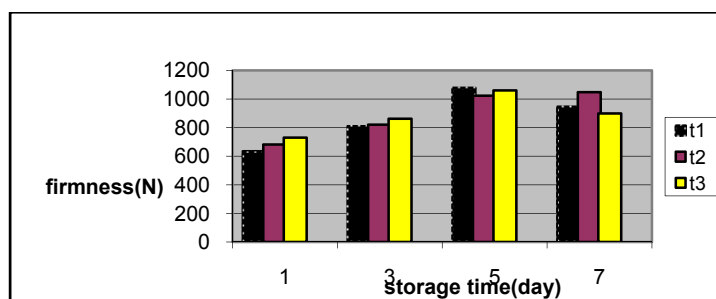


Fig 2. The rate of staling in breads with different proteins

Fig 2 showed the lower rate of staling and firming in the breads with higher protein. Fessas showed that one of the reasons for decreasing in the perceived firmness of breads with higher protein during storage must be due to the dilution of starch that delayed retrogradation, but there is a positive relationship between protein content and water absorption (Fessas, 1998). Flours with higher protein content have more water absorption.

Staling and firming of breads is supposed to depend on the moisture content, and in breads richer in water, staling was delayed (Scalon,2000).

4. Conclusion

DSC results indicated lower recrystallization enthalpy (retrogradation) in breads with higher protein contents. Results showed that proteins affect dough farinograph parameters, bread firmness and staling. Higher protein content caused flours to have more water absorption. Studies showed that more protein reduce firming and staling of bread during storage. Breads with higher protein content staled later than lower ones due to higher water absorption and starch dilution.

5. Acknowledgements

We are grateful to Cereal Research Center , Tehran Iran, for their laboratory support.

6. REFERENCES

- [1] E.M. Faergestad , and E.M, Magnus. Interrelationship of protein composition, protein level, baking process and characteristics of Hearth and Pan bread. *J. Cereal. Sci.* 2000, 31: 309-320.
- [2] M.,Carcea, and D.Schofield . Protein- lipid interactions in wheat gluten: Reassessment of the occurrence of lipid-mediated aggregation of protein in gliadin fraction. *J. Cereal. Sci.* 1996, 24: 101- 113.
- [3] C.Primo-Martin, N.H .Van Nicuwenhuijzen, R.J.Hamer, and T.Van Vliet, Crystallinity changes in wheat starch during the bread making process: starch crystallinity in the bread crust. *J. Cereal. Sci.* 2007, 45: 219-226.
- [4] M.L. Martin , K.J. Zeleznak, , and R.C. Hosney. A mechanism of bread firming. I. Role of starch swelling. *Cereal Chem.* . 1991, 68: 198-503.
- [5] E. Armero, and C. Collar. Crumb firming kinetic of wheat breads with anti-staling additives. *J. Cereal. Sci.* 1998, 28: 174- 195.
- [6] P.A. Roa. A.Nosiniwitch, and P.Chinachotti, Effect of selected surfactants on amylopectin recrystallization and on recoverability of bread crumb during storage. *Cereal Chem.* 1992, 69:613-619.
- [7] A.Abd Karim, M.H.Norzoah, and C. Seow. Methods for study of starch retrogradation. *Food Chem.* 2000, 71: 9-36.
- [8] AACC. Approved methods of Analysis of the American Association of Cereal Chemists. 10th ed. St. the American Association of Cereal Chemists. St: Paul. MN, 2000
- [9] P.D.Ribotta. Thermo-physical assessment of bread during staling. *LWT.* 2007, 40: 879-884.
- [10]H. Jacobs , J.A. Delcour. Hydrothermal modifications of granular starch, with retention of granular starch: A review. *J. Agric . Food Chem.* 1998, 46: 2895-2905.
- [11]M. Fukouoka, K.I.Ohta. H.Watanbe. Determination of the thermal extent of starch gelatinization in a limited water system by DSC. *J. Food. Engineering.* . 2002, 53:30-42.
- [12]A.C.Eliasson, M.Gudmundsson, G.Svensson,. Thermal behaviour of wheat starch in flour relation to flour quality. *Food Sci & Tech.* 1995, 28(2): 227-235.
- [13]C.A. Osella , H.D.Sanchez, , C.R.Carrara, M.A.De la Torre, M.P. Buera,. Water redistribution and structural changes of starch during storage of gluten free bread. *Starch.* 2005, 57:208-216.
- [14]M.A.Ottenhof, and I.A.Farhat, The effect of gluten on the retrogradation of wheat starch. *J. Cereal Sci.* 2004, 40: 269-274.
- [15]D.Fessas , A. Schiraldi,. Texture and staling of wheat bread crumb: Effect of water extractable proteins and pentosans. *Thermochemica Acta.* 1998, 323:17-26.
- [16]M.G. Scalon, H.D. Sapirstein, and D.Fahloul. Mechanical properties of bread crumb prepared from flours of different dough strength. *J. Cereal. Sci.* 2000, 32:235-243.