

# Influence of some additives on dough and bread properties of a wheat-lupin flour blend

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# RESEARCH ARTICLE

# Abstract

Effects of some enzymes (transglutaminase; TG and glucosoxidase; GO), emulsifiers (sodium stearoyl-2-lactylate; SSL, diacetyl tartaric acid esters of mono and diglycerides; DATEM, and SSL+DATEM combination) and oxidant (ascorbic acid; AA) on the properties of dough and bread made of a wheat flour (WF) and lupin flour (LF) blend were determined. LF replaced with WF at a 15% level in all dough formulation. All additives except TG increased development time and stability of dough containing 15% LF. While the highest dough energy obtained with AA addition, maximum resistance value increased from 217 BU up to 434 BU with TG usage in dough formulation. SSL+DATEM combination gave highest bread volume and lowest crumb hardness at 24 and 72 h. Generally, SSL+DATEM and AA had a more positive effect on symmetry, pore structure and crumb texture of the bread than the other additives. For improvement of the properties bread prepared with WF-LF blend, SSL+DATEM combination as emulsifier and AA as oxidant can be recommended. Effect of TG and GO on bread properties were found limited compared to AA.

Keywords: additive, bread, emulsifier, enzyme, lupin flour, oxidant

#### 1. Introduction

Lupin is an ancient leguminous plant Lupin seeds are a rich source of non-starch polysaccharides, oil, mineral and protein (Faluyi et al., 2000; Huyghe, 1997; Patel et al., 2012; Petterson and Mackintosh, 1994). Protein content of the lupin seeds (30-40%) are at the same levels or sometimes above that of soybean, according to genotype and location (Erbaş et al., 2005). Lupin proteins contain high amount of lysine and low amount of sulphur containing amino acids (Dervas et al., 1999). The usage of lupin and cereals together in food products had complementary effect in terms of essential amino acid. Lupin flour (LF) can be used as egg/ butter substitute or high protein ingredient in fermented foods, pasta, spaghetti, noodle, crisps, bread, biscuits, cookie, cakes, pancakes, gluten free and functional foods (Dervas et al., 1999; Erbaş et al., 2005; Jayasena and Nasar-Abbas, 2011; Kohajdová et al., 2011; Lampart-Szczapa et al., 1997; Papavergou et al., 1999; Pollard et al., 2002; Rayas-Duarte et al., 1996; Sironi et al., 2005). Mubarek (2001) reported that sweet LF could be used up to 6% level in bread

formulation without any observed detrimental effect on sensory properties of bread. Addition of LF also increased protein and total essential amino acids, especially lysine. According to Campos *et al.* (1978) 5% LF substituted bread had similar quality to control prepared with wheat flour (WF), but usage of 10% LF resulted in a slight reduction in volume and quality of breads. In another study, substitution of WF by increasing amount of sweet LF (5-15%) leads to a reduced breadmaking potential (Dervas *et al.*, 1999).

LF is an excellent ingredient with high protein, dietary fibre, mineral and functional component content for nutritional enrichment of bread. However, there are some limitations for usage of LF in bread formulation. Bitter lupin seeds contain some antinutrients, allergens and alkaloids which decreased the nutritional and sensory quality of LF. Alkoloids can be removed long debittering processes which contain heat treatment and soaking (Yorgancılar *et al.*, 2009). Sweet lupin seeds contain low level of alkaloids and can be used directly without debittering process. The other limitation for usage of LF in breadmaking is the absence of

gluten in LF. When LF was used in bread formulation over 5% level, dough and bread properties was deteriorated due to diluted gluten content and degraded gluten network in bread dough (Dervas *et al.*, 1999; Kohajdová *et al.*, 2011). Some additives as enzymes, emulsifiers and oxidants can be used to overcome these quality losses of LF enriched bread.

Enzymes and emulsifiers can be added to bread formulation to improve dough and bread properties. Emulsifiers influence rheological properties of bread by interacting with the proteins and carbohydrates resulting in a better quality of dough. Sodium stearoyl-2-lactylate (SSL) is an anionic oil-in-water emulsifier and is commonly used in bakery industry. SSL is also called dough strengthener and dough improver (Ribotta et al., 2010). Diacetyl tartaric acid esters of mono and diglycerides (DATEM) constitute an emulsifier that has been widely used as a bread improver. It is effective on bread volume and texture and also dough stability (Mettler and Seibel, 1993). Transglutaminase (TG) enzyme is known to catalyse the transfer reaction between an amide group in a protein-bound glutamine and ε-amino group in a protein-bound lysine side chain, resulting in cross-link between the protein molecules (Babiker, 2000; Nonaka et al., 1989). Beneficial effects of TG in bread making process have been reported by Gerrard et al. (2000). Başman et al. (2003) found that TG, even at very low levels, could be successfully incorporated in dough formulation to improve breadmaking quality. Glucosoxidase (GO) is an enzyme with oxidizing effect due to the hydrogen peroxide released from its catalytic reaction (Bonet et al., 2006). GO and ascorbic acid (AA) are widely used additives in bakery industry for improving dough and bread properties (Elgün and Ertugay, 1995).

The purpose of this research was to improve dough and bread properties of WF-LF blend with emulsifiers, enzymes and oxidant.

# 2. Materials and methods

### **Materials**

Commercial white wheat flour with 0.51% ash and compressed baker's yeast were purchased from local marked in Konya, Turkey. Additives, SSL (Palsgaard A/S, Juelsminde, Denmark), DATEM (Norbaran, Konya, Turkey), TG (AB Enzymes GMBH, Darmstadt, Germany), AA (HBI, Shangai, China) and GO (VatanGıda, Istanbul, Turkey) were used in breadmaking.

# Preparation of flour blends

LF replaced with WF at 15% level in all flour blends. Each emulsifiers, enzymes and oxidant were used separately in flour blends except DATEM and SSL which were used as individually and in combination. Emulsifiers were used as

0.5% level according to flour blend basis. In SSL+DATEM combination, each emulsifier was used as 0.25% level. TG, AA and GO were added to flour blend as 0.7%, 0.002% and 75 mg/kg levels, respectively. Control sample blend was prepared with 15% LF replacement by WF without additives (emulsifiers, enzymes and oxidant).

# Rheological properties

In the measurements of the rheological properties of flour blends, farinograph (AACC 54-21) and extensograph (AACC 54-10) analyses were performed according to AACC (1990). Water absorption, development time, stability and softening degree in farinogram, and energy, resistance to extension, extensibility and maximum resistance in extensogram (at 135 min) were determined.

#### Colour measurement

Colour values (L\*, a\* and b\*) of bread crust and crumb were measured by Minolta chroma meter CR-400 (Konica Minolta Sensing, Inc., Osaka, Japan) at five different points and the means were used. Hue angle (if a\*>0 and b\*>0, hue angle = arctan [b\*/a\*]; if a\*<0 and b\*>0, hue angle = arctan [b\*/a\*] + 180°) and saturation index ([a\*2 + b\*2] $^{1/2}$ )) were calculated using a\* and b\* colour parameters (Francis, 1998).

#### **Breadmaking**

Breads were obtained by the modification of AACC method 10-10B (AACC, 1990). Fermentation times were used as bulk fermentation for 30+30 min and proofing for 60 min. Loaf weight and volume of the bread samples were determined according to AACC (1990). Specific volume values were estimated by calculating loaf volume/loaf weight. At the 24<sup>th</sup> and 72<sup>nd</sup> h of storage at room conditions, the crumb hardness of bread samples was measured as Newtons per square centimetre on texture analyser using the procedure of Aydın and Öğüt (1991).

#### Sensory analysis

Sensory evaluation of bread in terms of symmetry, pore structure and texture was carried out after 24 h by twelve panellists (30-53 aged) from the Food Engineering Department at Selçuk University. Bread characteristics were rated on a 1-10 scale, 10 being the most desirable.

#### Statistical analysis

Results were expressed as the mean  $\pm$  standard deviation. TARIST (version 4.0; Ege University, İzmir, Turkey) software was used to perform the statistical analyses. Means that were statistically different from each other were compared by using Duncan's multiple comparison tests significance at P=0.05 level.

# 3. Results and discussion

## Rheological properties of bread dough

Farinogram properties of bread dough containing 15% LF and different additives are given in Table 1. Water absorption, development time, stability and softening degree of WF which was used in WF-LF blends were found as 62.3%, 9.5 min, 15 min and 10 BU, respectively (data not shown). Same parameters of flour blends containing 15% LF (control) were found as 74.3%, 6.2 min, 12.0 min and 18 BU, respectively. LF usage in flour blend increased water absorption and softening degree of dough but decreased development time and stability compared to WF. Similar effect of LF with different forms (full fat, concentrated or defatted concentrated) on water absorption of flour blend have been previously reported by Dervas et al. (1999). Campos and El-Dash (1978) found an increase in water absorption and reduce in dough stability with increased full fat LF incorporation (0-15%) into bread dough.

SSL and SSL+DATEM combination decreased the water absorption of the dough compared to control (Table 1). While all of the additives except TG increased development time, the highest development time (9.3 min) was obtained with GO addition. Emulsifiers GO and AA improved the dough stability, only TG decreased the dough stability from 12.0 min to 9.3 min. Indrani *et al.* (2003) found that GO increased the dough stability and dough development time compared to control without enzyme.

Extensogram properties of bread dough containing 15% LF and different additives are presented in Table 2. Energy, resistance to extension, extensibility and maximum resistance of WF were found as 63 cm<sup>2</sup>, 370 BU, 85 mm and 380 BU, respectively (data not shown). 15% LF addition decreased energy, resistance to extension and maximum resistance value of dough due to diluting gluten content in dough formulation with LF/WF replacement. The adverse effect of increasing LF substitution levels into WF on extensogram parameter have been previously reported in literature (Campos and El-Dash, 1978; Dervas *et al.*, 1999).

Table 1. Effect of some additives on faringgram properties of bread dough containing lupin flour (mean values ± standard deviation)<sup>1</sup>.

Additives <sup>2</sup>	Water absorption (%)	Development time (min)	Stability (min)	Softening degree (BU)
Control	74.3±0.42 <sup>a</sup>	6.2±0.14 <sup>d</sup>	12.0±0.28 <sup>c</sup>	18±0.28 <sup>f</sup>
SSL	72.3±0.42 <sup>c</sup>	8.0±0.28 <sup>b</sup>	13.3±0.14 <sup>b</sup>	30±0.28 <sup>b</sup>
DATEM	73.5±0.28 <sup>ab</sup>	7.3±0.14 <sup>c</sup>	14.8±0.28 <sup>a</sup>	16±0.28 <sup>9</sup>
SSL+DATEM	72.9±0.42bc	8.0±0.14 <sup>b</sup>	13.4±0.14 <sup>b</sup>	25±0.14 <sup>c</sup>
TG	73.9±0.57 <sup>ab</sup>	6.5±0.28 <sup>d</sup>	9.3±0.28 <sup>d</sup>	37±0.14 <sup>a</sup>
GO	74.3±0.42 <sup>a</sup>	9.3±0.14 <sup>a</sup>	13.2±0.14 <sup>b</sup>	20±0.28e
AA	73.9±0.42 <sup>ab</sup>	7.5±0.00bc	13.2±0.14 <sup>b</sup>	21±0.28 <sup>d</sup>

<sup>&</sup>lt;sup>1</sup> Values followed by the same letter within a column are not significantly different (P>0.05).

Table 2. Effect of some additives on extensogram properties of bread dough containing lupin flour (mean values ± standard deviation)<sup>1</sup>.

Additives <sup>2</sup>	Energy (cm <sup>2</sup> )	Resistance to extension (BU)	Extensibility (mm)	Maximum resistance (BU)
Control	32±0.71 <sup>d</sup>	216±1.41 <sup>9</sup>	101±0.42 <sup>b</sup>	217±1.41 <sup>9</sup>
SSL	35±0.85c	224±1.41 <sup>f</sup>	105±0.57 <sup>a</sup>	225±1.41 <sup>f</sup>
DATEM	35±0.71c	244±0.71 <sup>d</sup>	98±0.42 <sup>c</sup>	245±1.13 <sup>d</sup>
SSL+DATEM	35±0.85c	229±1.13 <sup>e</sup>	103±0.42a	230±1.13 <sup>e</sup>
TG	38±0.71 <sup>b</sup>	392±0.99 <sup>a</sup>	66±0.71 <sup>e</sup>	434±1.27 <sup>a</sup>
GO	38±0.57 <sup>b</sup>	255±1.27°	100±0.57 <sup>b</sup>	257±0.71 <sup>c</sup>
AA	48±0.42 <sup>a</sup>	382±1.13 <sup>b</sup>	91±0.99 <sup>d</sup>	385±1.27 <sup>b</sup>

<sup>&</sup>lt;sup>1</sup> Values followed by the same letter within a column are not significantly different (P>0.05).

<sup>&</sup>lt;sup>2</sup> AA = ascorbic acid; DATEM = diacetyl tartaric acid esters of mono and diglycerides; GO = glucosoxidase; SSL = sodium stearoyl-2-lactylate; TG = transglutaminase.

<sup>&</sup>lt;sup>2</sup> AA = ascorbic acid; DATEM = diacetyl tartaric acid esters of mono and diglycerides; GO = glucosoxidase; SSL = sodium stearoyl-2-lactylate; TG = transglutaminase.

While all additives increased dough energy compared to control, the highest increment was obtained with AA which hinders the disulfide-to-sulfide interchange caused by glutathione and thereby strengthens the dough after its resting time (Aamodt *et al.*, 2003). Oxidative enzymes and oxidants have a strong impact on the dough thiol-disulphide system and hence, on the properties of the dough (Goesaert *et al.*, 2005).

All of the additives increased resistance to extension values of dough. Minimum increment ratio (3.7%) was obtained with SSL, the maximum increment (81.5%) was found with TG addition. As expected GO and AA also increased the resistance to extension of dough. SSL and SSL+DATEM combination increased the dough extensibility compared to control. Maximum resistance values showed similar trend to resistance to extension values and the highest value obtained with TG addition. Regarding the rheological properties of dough, TG showed the most remarkable effect on maximum resistance value. Increment in maximum resistance might be a result of transformation of weak gluten into strong one due to the TG catalysed cross-linking reactions between wheat and LF proteins or within each one (Başman et al., 2003). Bauer et al. (2003) reported that increasing concentrations of TG caused a drastic decrease on extensibility and an increase on the resistance to extension of dough.

# **Bread properties**

Effect of some additives on bread properties of WF-LF blend are given in Table 3. Bread prepared with WF had higher volume and specific volume compared to control bread containing 15% LF without additives (data not shown). Similar to dough rheological properties, LF addition adversely affected bread properties with its diluting effect on gluten amount in flour blend (Doxastakis *et al.*, 2002). Ribotta *et al.* (2010) reported the negative effect of soy products on gluten network in soy-wheat bread dough.

In present study, some additives were used to overcome these negative effects, and all additives showed positive effect on volume of bread compared to control samples containing 15% LF.

DATEM with and without SSL gave superior volume and specific volume values. There are numerous studies in the literature for improving effect of SSL and DATEM on volume of bread containing different cereal, pseudocereal and legume flours (Atalay et al., 2013; Elgün and Ertugay, 1995; Gül, 2007; Koca and Anıl, 1999). Campos and El-Dash (1978) found that calcium stearyl-2-lactylate improved volume and other characteristics of bread containing full fat sweet LF. AA as an oxidant also improved the volume of the bread and increased up to 430 ml from 370 ml. Dağdelen and Göçmen (2007) reported that specific loaf volume of bread showed a significant enhancement when GO and AA combinations were used. SSL+DATEM combination and AA showed more softening effect on bread crust with lower hardness value at the end of 24 and 72 h measurement. Emulsifiers are widely used in bakeries as dough strengtheners and crumb softeners (Gómez et al., 2004). TG usage did not show marked improvement on bread properties. Crust and crumb colour values of breads are summarised in Table 4. While TG gave lighter and yellowish bread crust compared to other additives and control, SSL+DATEM caused more increment on crust redness. Başman et al. (2003) reported that lighter crust colour of wheat breads supplemented with TG might be the result of limited amount of Maillard reaction due to the decreasing amount of available lysine after TG enzymatic reaction. GO increased crumb lightness, yellowness and saturation index values compared to control. In contrast to crust colour, TG decreased the lightness of the bread crumb. Indrani et al. (2003) found that an improvement in crust and crumb colour scores of bread dough with GO usage compared to control samples.

Table 3. Effect of different additives on some bread properties (mean values ± standard deviation)1.

Additives <sup>2</sup>	Weight (g)	Volume (ml)	Specific volume (ml/g)	24h hardness (N/cm²)	72h hardness (N/cm²)
Control	157.5±0.42 <sup>bc</sup>	370±1.41 <sup>f</sup>	2.35±0.01 <sup>f</sup>	0.348±0.01a	0.472±0.01 <sup>a</sup>
SSL	156.6±0.57 <sup>c</sup>	395±1.41 <sup>d</sup>	2.52±0.00e	0.292±0.01c	0.391±0.01 <sup>b</sup>
DATEM	158.1±0.28 <sup>b</sup>	430±0.71 <sup>b</sup>	2.72±0.01 <sup>b</sup>	0.228±0.00 <sup>d</sup>	0.344±0.00 <sup>c</sup>
SSL+DATEM	157.6±0.57bc	440±0.71 <sup>a</sup>	2.79±0.01 <sup>a</sup>	0.185±0.01e	0.264±0.01 <sup>d</sup>
TG	159.8±0.28 <sup>a</sup>	375±1.41e	2.35±0.01 <sup>f</sup>	0.321±0.01 <sup>ab</sup>	0.479±0.01 <sup>a</sup>
GO	158.5±0.42ab	415±1.41 <sup>c</sup>	2.62±0.01 <sup>d</sup>	0.295±0.00bc	0.380±0.01 <sup>b</sup>
AA	157.8±0.42 <sup>b</sup>	430±0.71 <sup>b</sup>	2.72±0.01 <sup>c</sup>	0.249±0.01 <sup>d</sup>	0.270±0.01 <sup>d</sup>

<sup>&</sup>lt;sup>1</sup> Values followed by the same letter within a column are not significantly different (P>0.05).

<sup>&</sup>lt;sup>2</sup> AA = ascorbic acid; DATEM = diacetyl tartaric acid esters of mono and diglycerides; GO = glucosoxidase; SSL = sodium stearoyl-2-lactylate; TG = transglutaminase.

Table 4. Effect of different additives on crust and crumb colour of bread (mean values ± standard deviation)1.

Additives <sup>2</sup>	L*	a*	b*	SI	hue
Crust					
Control	64.98±0.18 <sup>f</sup>	0.56±0.06 <sup>cd</sup>	25.41±0.11 <sup>f</sup>	25.42±0.08 <sup>f</sup>	88.74±0.10 <sup>bc</sup>
SSL	68.33±0.16 <sup>d</sup>	0.20±0.07 <sup>e</sup>	27.26±0.06d	27.26±0.06 <sup>d</sup>	89.58±0.10 <sup>a</sup>
DATEM	70.46±0.18 <sup>c</sup>	0.68±0.08bc	28.19±0.10 <sup>c</sup>	28.20±0.08c	88.62±0.16 <sup>c</sup>
SSL+DATEM	72.04±0.14 <sup>b</sup>	1.61±0.06a	28.45±0.06 <sup>b</sup>	28.50±0.08b	86.76±0.11 <sup>d</sup>
TG	72.88±0.11 <sup>a</sup>	0.42±0.04d	29.32±0.08a	29.32±0.07a	89.18±0.14 <sup>ab</sup>
GO	67.05±0.14 <sup>e</sup>	0.75±0.04 <sup>b</sup>	26.52±0.08e	26.53±0.07e	88.38±0.14 <sup>c</sup>
AA	70.45±0.16 <sup>c</sup>	0.66±0.04bc	28.32±0.11bc	28.33±0.13bc	88.66±0.14 <sup>c</sup>
Crumb					
Control	69.66±0.11 <sup>b</sup>	-1.43±0.03d	22.81±0.10e	22.85±0.11e	93.59±0.11b
SSL	67.69±0.13 <sup>e</sup>	-1.24±0.01b	22.85±0.10 <sup>de</sup>	22.88±0.11 <sup>de</sup>	93.11±0.08 <sup>c</sup>
DATEM	68.09±0.14 <sup>d</sup>	-1.57±0.03 <sup>e</sup>	23.77±0.07b	23.82±0.08b	93.78±0.13 <sup>ab</sup>
SSL+DATEM	69.03±0.14 <sup>c</sup>	-1.33±0.03c	23.76±0.07b	23.80±0.07b	93.20±0.11c
TG	68.77±0.10 <sup>c</sup>	-1.54±0.01e	23.41±0.08 <sup>c</sup>	23.46±0.08 <sup>c</sup>	93.76±0.10 <sup>ab</sup>
GO	71.08±0.14 <sup>a</sup>	-0.94±0.01a	25.39±0.06a	25.41±0.08 <sup>a</sup>	92.12±0.08d
AA	69.83±0.13 <sup>b</sup>	-1.64±0.01 <sup>f</sup>	23.05±0.06 <sup>d</sup>	23.11±0.08 <sup>d</sup>	94.07±0.13 <sup>a</sup>

<sup>&</sup>lt;sup>1</sup> Values followed by the same letter within a column are not significantly different (*P*>0.05).

Sensory properties of breads are presented in Figure 1. GO and AA gave the more symmetric breads than other additives and control. Pore structure improved with additives except TG compared to control. Most dominant additives

on pore structure of bread containing LF were found as emulsifiers. Breads containing SSL+DATEM combination or AA were rated highest texture scores, and these additives were generally gave superior sensory properties.

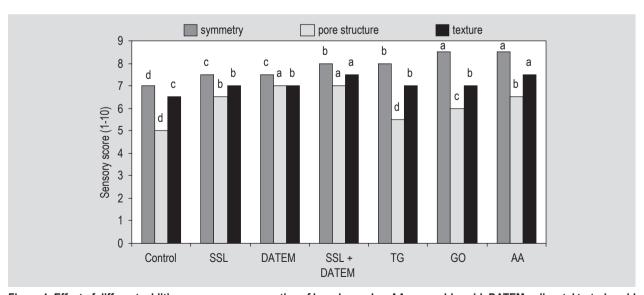


Figure 1. Effect of different additives on sensory properties of bread samples. AA = ascorbic acid; DATEM = diacetyl tartaric acid esters of mono and diglycerides; GO = glucosoxidase; SSL = sodium stearoyl-2-lactylate; TG = transglutaminase. Means with same letter are not significantly different (*P*>0.05; Duncan's multiple range test).

<sup>&</sup>lt;sup>2</sup> AA = ascorbic acid; DATEM = diacetyl tartaric acid esters of mono and diglycerides; GO = glucosoxidase; SSL = sodium stearoyl-2-lactylate; TG = transglutaminase; L\* = light/dark; a\* = green (negative values)/red (positive values); b\* = blue (negative values)/yellow (positive values); hue = hue angle; SI = saturation index.

# 4. Conclusions

LF is valuable ingredient with high protein, fat, mineral, dietary fibre and functional component content for nutritional enrichment of bakery products. Because of the some limitations, LF adversely affects dough and bread properties, and cannot be used in bread formulation over 5-10% level. In the present study, some additives (emulsifier, enzymes and oxidant) were used to overcome this problem. All additives increased the energy, resistance to extension and maximum resistance of the dough compared to control. SSL+DATEM combination and AA were found as the most effective additives on bread properties especially on volume and softness. Among the additives, TG had more dominant effect on increment of crust lightness and vellowness. For improvement of LF enriched bread properties, SSL+DATEM combination as emulsifier and AA as oxidant presented better properties than other additives.

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