

## EFFECT OF ROSELLE SEEDS FLOUR ADDITION ON THE QUALITY CHARACTERISTICS OF PAN BREAD

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### ABSTRACT

The effect of defatted Roselle seeds flour (DRSF) addition at different levels (5, 10 and 15%) to wheat flour on the flour characteristics, dough rheological properties, bread chemical, physical and sensory characteristics were investigated. Mixolab analysis showed that the addition of DRSF led to a reduction in the flour water absorption and an increase in the dough development time and consequently decreased the dough stability compared to the control. Also, the addition of DRSF decreased the bread loaf volume and increased its protein and fibers contents and the antioxidant activity compared to the control. Sensory evaluation showed that all fortified bread samples were acceptable up to 10% DRSF.

**Keywords:** Bread fortification, Roselle seed flour, Rheological properties, Mixolab.

### INTRODUCTION

Bread is considered the most famous backed products in the world, its quality is affected by several factors including flour type, and additives (Dall'Asta *et al.*, 2013). Wheat flour is rich source of energy, dietary fiber, minerals, vitamins and many other bioactive compounds, and no other cereal flour could achieve its baking properties. Wheat flour protein is deficient in some essential amino acids such as lysine and threonine. Fortification of wheat flour with protein-rich materials including cereal and non-cereal sources had been applied by many researchers to improve its protein content and quality and to add more bioactive compounds to the resultant baked products, especially bread, and to prevent worldwide protein-energy malnutrition. Consumer acceptability of new developed products is a key point of its success. (Amir *et al.*, 2013 and Ogur, 2014).

Roselle (*Hibiscus sabdariffa* L.), regionally known as karkade, is an important annual plant belongs to Family *Malvaceae* (Yagoub *et al.*, 2008). Roselle grows successfully in tropical and sub-tropicals. Roselle seeds contained 30.6-35.4% protein, 22.1-29.6% lipids, 26-33% carbohydrates and 18.3-25.5% total dietary fiber and high minerals contents including potassium, magnesium and calcium. Roselle seed protein is rich in lysine, arginine, leucine, phenylalanine and glutamic acid. Roselle seeds were indicated to be a potential source of functional ingredients (Hainida *et al.*, 2008b). Amino acids pattern of Roselle seeds is higher than FAO/WHO (1974) requirement, seeds protein fractions and isolates (with its low molecular weight poly peptide) are considered as functional ingredients with high nutritional properties (Toukara *et al.*, 2013). Defatted dried roselle seeds lowered

plasma cholesterol and low density lipoprotein cholesterol levels in rats with induced hypercholesterolemia (Hainida *et al.*, 2008a). Roselle seeds are a good source of lipid-soluble antioxidants, particularly  $\gamma$ -tocopherol and Vitamin E. The seeds contained total tocopherols of 2000 mg/kg, including  $\alpha$ -tocopherol (25%),  $\gamma$ -tocopherol (74.5%), and  $\delta$ -tocopherol (0.5%) (Mohamed *et al.*, 2007). These antioxidants reduced lipid oxidation in food and were better when compared to Butylated hydroxytoluene (BHT) (Mohd-Esa *et al.*, 2010).

This study aimed to investigate the effect of defatted Roselle seeds flour (DRSF) on the quality properties of pan bread

## **MATERIALS AND METHODS**

Commercial wheat flour (72% extract), and baking ingredients were purchased from the local markets of Zagaig city, El-Sharquia governorate, Egypt. Roselle (*Hibiscus sabdariffa* L.) seeds were obtained from the Agricultural Research Center, Giza, Egypt.

### **Preparation of Roselle seeds Flour**

Roselle seeds were cleaned, dried in shade and ground into powder using a grinder (Moulinex, France). Roselle seeds flour was defatted by soxhlet extraction using n-hexane as a solvent. The solvent was removed by hot air drying at (45°C  $\pm$ 1) for 12 hours. The Defatted Roselle seeds flour (DRSF) was sieved through a 60 mesh screen until fine powder was obtained and kept frozen at -18 °C in polyethylene pages until used.

### **Flour quality**

Mixolab (Chopin, Tripette and Renaud, Villeneuve-la-Garenne, France) was used to analyze flour quality as described by Jia *et al.* (2011).

### **Bread preparation**

Pan bread preparation was based on the official method 10-09 (AACC, 2002) with some modifications. The formula of control pan bread is reported in Table (1). The addition levels of DRSF were 5%, 10% and 15% based on wheat flour. Water, salt, sugar, oil, fermented starter and dry yeast were added and mixed in Kenwood Major mixer (Kenwood, Hampshire, UK) at medium speed for 2 min. Flour was added and mixed at high speed for 6 min. After the mixing time, the dough was divided into 750 g pieces and put into 30 x 10 x 10 cm baking pans. Then, the pans were rested in trays at room temperature for 10 min, it was covered by a plastic film to prevent dehydration during proofing. The pans were placed in a thermostatically controlled proofing oven at 35°C and 95% relative humidity for 45 minutes for final proofing. Baking was performed in an electrical oven at 200°C for 30 minutes. Bread loaves obtained were cooled to 25°C for 3 hours before use in further evaluation.

**Table 1. Control dough of pan bread formula**

<b>Ingredient</b>	<b>Weight (g)</b>
Flour	1000
Sugar	50
Oil	40
Fermented starter	150
Dry yeast	30
Salt	5
Water	600
Total	1875

**Sensory evaluation:**

Bread samples with or without DRSF were organoleptically evaluated for aroma, crust color, crumb color, texture, taste and overall acceptability according to Tuorila (2015). Bread samples were introduced in random order to 35 untrained panelists (staff of Food Science Department, Faculty of Agricultural, Zagazig University, Egypt) within 24 hours of bread preparation. The panelists were chosen randomly and the samples were subjected to sensory evaluation using a 9-point hedonic scale, where (1=dislike extremely, 2=dislike very much, 3=dislike moderately, 4=dislike slightly, 5=neither like nor dislike, 6=like slightly, 7=like moderately, 8=like very much, and 9= like extremely, and the mean of 35 values was taken.

**Physical properties of bread:**

**Specific Volume:**

Volume of bread loaves was measured using Tex-vol instrument BVM-L370. Specific volume of bread was calculated from the ratio between volume and weight of loaves (Sciarini *et al.*, 2012).

**Texture analysis:**

Fresh bread samples supplemented with different levels of DRSF (5, 10 and 15%) were analyzed for their texture profile (hardness, stickiness, adhesiveness, chewiness, cohesiveness, gumminess, springiness and stringiness) according to the methods described by O'Brien *et al.*, (2000) with minor modifications. Texture analyzer (TVT-300XP) was used on two bread slices of 12 mm height (24 mm total), two compression cycles with 5 seconds relaxation; the probe was a 35 mm cylindrical coded (P-Cy35S), compression cell 5 kg, pretest speed 5 mm/min, compression rate 40% of original height, force in grams required to accomplish compression was recorded as hardness.

**Proximate chemical composition:**

Proximate chemical composition of crude DRSF as well as prepared pan bread was determined. Moisture, ash contents were determined according to the approved method of AACC (2002). Crude protein and fiber contents were determined according to AOAC (2005).

**Antioxidant activity:**

Antioxidant properties including extraction of flour and DPPH scavenging activity percentage of flour samples, defatted Roselle seeds flour and all treated bread samples were determined according to Lilei *et al.* (2013).

**Statistical analysis:**

Analysis of variance (ANOVA) was used by SAS software version 9.2 (SAS Institute, 1990). Differences between means were determined by the least significant difference test, and significance was defined at  $P < 0.05$ . All measurements were carried out in triplicates.

**RESULTS AND DISCUSSION**

**Flour characteristics and dough rheological properties:**

Rheological properties of composite flour characteristics are shown in Table 2. The moisture content of the flour ranged from 12.70% for 15% DRSF to 13.62% for control. Water absorption of flour, one of its fundamental quality parameters, can be defined as the amount of water needed to hydrate flour to produce dough with optimum consistency. Water absorption of wheat flour was influenced by the extraction rate, protein (gluten) content, starch properties (damaged and gelatinized starch granules) and flour particle size (Perten, 1990). Water absorption of control wheat flour was 56.17%. Proportional addition of DRSF to wheat flour decreased the water absorption values comparing to that of the control. Wheat flour with 15% DRSF had the lowest water absorption value (51.2%). The decrease in water absorption capacity of DRSF may be related to the denaturation of protein during the defatting or fat trimming process. Rakszegi *et al.* (2014) stated that water absorption of wheat flour was affected by soluble protein and damaged starch contents. Similar results were reported by Sibanda *et al.* (2015), since they found that a decrease in the water absorption ability of the flour from 57.73% for 100% wheat to 53.03% for the 30% sorghum flour.

Dough development time (DDT) (C1 time in Table 2) of control treatment was ideal as it was minimal at a time of 1.42 minutes while the addition of DRSF increased DDT to reach 4.11, 5.31 and 6.23 minutes in 5, 10 and 15% DRSF, respectively. This might be due to the dilution of wheat gluten by the addition of gluten free additive (DRSF) and also is due to the higher fiber contents in DRSF comparing to wheat flour. Those findings were similar to those found by Sedej *et al.* (2011), who reported an increased in DDT with higher fiber additives to bread flour.

**Table 2. Rheological properties of control and composite wheat flour measured using Mixolab**

Properties Treatment	Flour moisture (%)	Water absorption (%)	Dough development time (min.)	Dough Stability (min.)
Control	13.62 a	56.17a	1.42 d	8.95 a
R5	13.10 b	53.70b	4.11 c	8.22 b
R10	12.93c	51.82c	5.31 b	7.82 c
R15	12.70d	51.20d	6.23 a	7.3 d

R5: 5%DRSF, R10: 10% DRSF, R15: 15% DRSF

Dough stability represents the period of time in which the dough is able to stand with the applied deformation, subsequent decrease in torque value during further kneading and heating is measure of protein weakening (Rosell *et al.*, 2007). Application of DRSF to the flour caused a decrease in dough stability from 8.95 minutes in control flour to 8.22, 7.82 and 7.3 minutes in 5, 10 and 15% DRSF, respectively. This decrease in dough stability was also noted by Sibanda *et al.* (2015) who reported a decreased dough stability with the addition of sorghum to the dough.

### **Sensory evaluation**

Sensory evaluation is considered to be one of the limiting factors for consumer acceptability. Sensory evaluation of control and DRSF enriched breads (aroma, crust and crumb colors, texture, taste and overall acceptability) is shown in Table (3) and Figure (1).

Gradual increase in DRSF addition percentage to bread decreased aroma scores from 9.1 in control to 8.6, 8.2 and 8 in the 5, 10 and 15% added DRSF. Color of bread including crust and crumb color scores decreased with the proportional increase in DRSF. Texture of the DRSF added bread samples got lower scores (7.8, 6.2 and 5.6 for 5, 10 and 15% added DRSF respectively), which comes in agreement in texture profile results in Table 4. As the bitter taste of DFRS affected the taste of prepared bread, sensory scores of taste of bread decreased from 8.5 in control sample to 8.2, 7.0 and 5.8 in the 5, 10 and 15% added DRSF bread samples, respectively.

Pan bread containing 15% DRSF had the lowest overall acceptability score (5.8) compared to control (8.4) and was not favored by most of the panelists because of the bitter taste, while the 5 and 10% of added DRSF was preferred and accepted for panelist comparing to the 15% added DRSF bread with scores of 8.2 and 7.2, respectively. These results are in harmony with these found by Lee and Choi (2013).

**Table 3. Sensory evaluation of pan bread treated with DRSF**

Properties Treatments	Aroma* (9)	Crust color (9)	Crumb color (9)	Texture (9)	Taste (9)	Overall Acceptability (9)
Control	9.1 a	8.9 a	8.7 a	8.7 a	8.5 a	8.4 a
R5	8.6 a	8.0ab	8.4a	7.8 a	8.2 a	8.2ab
R10	8.2 ab	7.5 b	6.6b	6.2b	7.0 b	7.2 b
R15	8.0 b	5.8c	6.0b	5.6b	5.8c	5.8c

R5: 5%DRSF, R10: 10% DRSF, R15: 15% DRSF

- all parameters were given a score on 9 points headonic scale

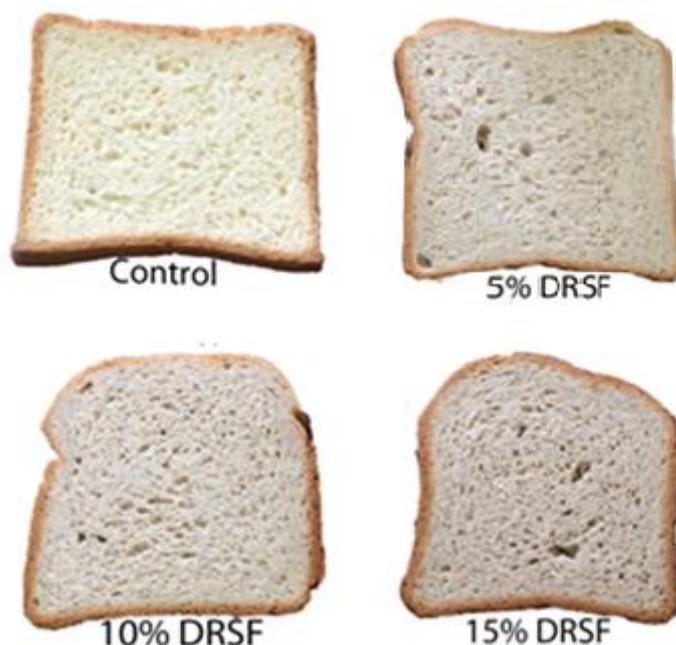


Figure 1. Images of control and enhanced pan bread with the DRSF

**Physical properties of bread:**

Physical properties of control and DRSF enhanced bread including bread texture, loaves specific volumes, bread hardness, stickiness, adhesiveness, chewiness, gumminess, springiness, and stringiness as well as bread color are presented in Table (4).

As for specific volumes of control and DRSF treatments, control bread had the highest specific volume (2.83 g/cm<sup>3</sup>) with a perfect texture and best gluten network comparing to other samples, while 15% DRSF had the lowest specific volume (2.47 g/cm<sup>3</sup>). A significant reduction in loaf volume was observed as the levels of DRSF increased. Lower specific volume values of bread with the addition of DRSF may be due to the lower gluten content in the additives (DRSF) which gives less active gluten network and gas trapping (Doxastakis *et al.*, 2002).

Texture is a manifestation of the rheological properties of a food product, texture then affects processing, handling, shelf stability and consumer acceptance of food products (Agyare *et al.*, 2005). Texture of bread slices was analyzed and all texture parameters merged from the texture analyzer curves are reported in Table (4).

Hardness is the peak force of the first compression of the product or force required to compress a food between the molars, defined as force necessary to attain a given deformation (40%). Those definitions describe the

first bite feeling in mouth. From the data it can be noticed that the addition of different levels of DRSF gradually increased hardness values. Control treatment showed the lowest hardness (6108 g), while the addition of 5, 10 and 15% of DRSF gradually increased bread hardness values to 6814, 7834 and 8532 g, respectively. These results may be attributed to the lower loaf volume of DRSF enhanced bread compared to the control which made the bread components more condensed therefore it gave harder texture. It was found that the smaller loaf volume has a negative effect on its quality attributes, such as crumb grain and tenderness (Doxastakis *et al.* 2002).

**Table 4. Physical properties of pan bread treated with DRSF**

Treatment	Control	R5	R10	R15
<b>Properties</b>				
Volume (cm <sup>3</sup> )	1892 a	1788 b	1699c	1638 d
Weight (g)	667.77 b	673.87a	668.02b	663.19 b
Specific volume (g/cm <sup>3</sup> )	2.83 a	2.65 b	2.54 c	2.47 d
Hardness (g)	6108 d	6814 c	7834 b	8532 a
Stickiness (g)	1511d	1671 c	2345 b	2570 a
Adhesiveness	3502 d	3719 c	3917 b	4377 a
Chewiness (g)	1227d	1818 c	2264b	2617a
Cohesiveness	0.44 c	0.51b	0.52ab	0.53a
Gumminess (g)	2634 d	3441c	4020 b	4475a
Springiness	0.48 d	0.54 c	0.57 b	0.59 a

R5: 5%DRSF, R10: 10% DRSF, R15: 15% DRSF

As it mentioned above, hardness is the first positive peak force as bread resist compression by the probe, exactly the same when it resist compression under teeth, while the negative peak simulate the stickiness or adhesiveness of bread samples to teeth. Adhesiveness is defined as the work necessary to overcome the attractive forces between the surface of the food and the surface of other materials with which the food comes into contact (e.g. tongue, teeth, palate), or the work required to pull food away from a surface. It was noticed that stickiness values had the same trends of hardness values. That might be because of the higher contents of protein in roselle seeds flour, comparing to wheat flour. Increased stickiness with increased protein contents were also reported by Zhu *et al.*, (2001) who found that, stickiness of steamed bread and dough were higher with the gradual increase of protein contents.

Gluten dilution by the addition of DRSF caused less active gluten network and a less gas trapping within dough caused decreased gas to whole volume ratio which led to an increase in hardness of bread samples. Increase in hardness through the compression and decreased gas trapping and increased stickiness makes bread samples more chewable as could also be easily merged from data in Table (4).

Chewiness is the chewing ability of bread slices samples; it can be calculated from the texture analyzer (TVT-300XP) software from multiple

analyses of gumminess and springiness. Following the increased of both parameters gumminess and springiness, chewiness of bread enhanced with DRSF were higher than control samples. Control bread sample recorded the lowest chewiness value (1227 g), while 15% DRSF gave the highest being 2617g. The addition of different levels of DRSF gradually increased chewiness, Table (4).

Cohesiveness is how well the product withstands a second deformation relative to how it behaved under the first deformation. It is measured as the area of work during the second compression divided by the area of work during the first compression. The same trend of all texture parameters was also reported in cohesiveness which ranged between 0.44 to 0.53 for control and 15% DRSF enhanced bread, respectively.

Springiness measure the recovery of food structure after it has been deformed during the first compression (Guine and Barroca, 2012). Data showed that the springiness gradually increased with the addition of DRSF from 0.48 in control bread to 0.54, 0.57 and 0.59 in the 5, 10 and 15% DRSF enhanced bread, respectively. Also, an increase in cohesiveness and springiness as well were clearly observed when DRSF was applied to bread formula. These results agree with those reported by Onyango *et al.* (2015).

**Proximate chemical composition:**

Crude DRSF contains 9.21% moisture, 33.45% protein, 1.04% fat, 5.17% ash, 4.42% crude fiber and 46.72% total carbohydrate

Proximate chemical composition of control and DRSF enhanced bread are shown in Table (5). The results indicated that the moisture contents of all bread samples gradually decreased compared to control (41.3%). The addition of 5, 10 and 15% of DRSF decreased the bread moisture content to 37.66, 36.21 and 35.6%, respectively. The high protein content of crude DRSF used in the fortification of the wheat flour was reflected in the high content of protein in the produced bread. Protein content ranged from 12.11% in control to 15.36% in 15% added DRSF. The addition of DRSF to wheat flour was expected to increase protein content of produced bread, science they have high content of protein (33.45%). Similar findings were observed by Anton *et al.* (2008) who stated that the addition of bean flour to wheat flour was expected to increase the protein content of tortillas, since legumes generally contain more proteins than cereals.

**Table 5. Proximate composition of pan bread treated with DRSF**

Constituents Treatment	Moisture	Protein	Fiber	Ash
Control	41.30 a	12.11d	1.05d	0.70 d
R5	37.66b	13.65c	2.93c	0.84c
R10	36.21c	14.11b	3.60 b	0.90 b
R15	35.60d	15.36 a	4.25 a	0.98a

R5: 5%DRSF, R10: 10% DRSF, R15: 15% DRSF

Control bread sample had the lowest ash content (0.70%) comparing to those of other enhanced bread samples. As the levels of DRSF increased in

bread as its ash content increased to reach 0.98% in the 15% added DRSF. Roselle flour contained 4.42% fiber. Fiber contents of enhanced breads were higher than control bread samples which contained 1.05%. DRSF gradually increased fiber contents to be 2.93, 3.6 and 4.25% in 5, 10 and 15% added DRSF, respectively. These results are closed to that found by Seleem & Omran (2014) who stated that bread enhanced with beans had the highest values in protein, ash and fiber compared with wheat and sorghum bread.

**Antioxidant activity:**

Table. (6) shows the total phenolic compounds and antioxidant properties of bread samples as well as the raw materials used. Results of total phenolic compounds (TPC) in raw materials were found to be 5.25 mg gallic acid equivalent/100g (GAE/100g) in wheat flour, while TPC of DRSF was higher (10.41 mg Gallic acid equivalent/100g). DPPH scavenging activity of wheat flour and DRSF were 64.3 and 89.5%, respectively. Control bread contained the lowest TPC content (1.03 mg GAE/100g), while samples contained 15% DRSF had the highest TPC content of 1.41 GAE/100g bread.

DPPH scavenging activity percentages of bread samples are presented in Table (6). Bread samples with 15% of DRSF had the highest DPPH scavenging percentage (90.4%), while control bread had the lowest (82.1%). It was noticed that, the addition of DRSF increased the DPPH scavenging percentages. Similar results are found by Swieca *et al.* (2014) who reported that fortification of bread with coriander and quinoa leaves powder enhanced the antioxidant properties and phenolic content of bread.

**Table 6. Antioxidant activity of pan bread treated with DRSF**

Antioxidant Treatments	TPC mg Gallic acid equivalent/g	DPPH Scavenging Activity %
Wheat flour	5.25	64.3
DRSF	10.41	89.5
Control	1.03d	82.10 d
R5	1.14 c	87.37 b
R10	1.29 b	88.20 b
R15	1.41 a	90.40 a

**CONCLUSION**

Enhancement of wheat flour with DRSF decreased water absorption and weakened the rheological properties of the dough though the diluting and disrupting the gluten network. The loaf volume was significantly decreased, while hardness, cohesiveness and springiness increased as the levels of DRSF increased. The addition of DRSF led to slightly darker crumb (lower *L\** values) than control. Bread sample enhanced with DRSF was acceptable by panelists up to 10%. We recommend the application of DRSF in bread up to 10% to enrich its nutritional value and antioxidant activity without reducing consumers' acceptability.

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تأثير إضافة دقيق بذور الكركديه علي خواص الجودة لخبز القالب  
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تم دراسة تأثير إضافة مستويات مختلفة (5 و 10 و 15%) من دقيق بذور الكركديه منزوعة الدهن إلي دقيق القمح علي خواص الدقيق و الخواص الريولوجية للعجين و التركيب الكيماوي للخبز الناتج و كذلك خواصه الفيزيائية و الحسية. ولقد أو ضحت نتائج التحليل بجهاز المكسولاب أن إضافة دقيق بذور الكركديه منزوعة الدهن أدي إلي خفض أمتصاص الدقيق للماء و زيادة زمن العجن و بالتتابع أدي ذلك إلي خفض ثبات العجين مقارنة بالكنترول. ولقد أدي إضافة بذور الكركديه منزوعة الدهن أيضاً إلي خفض حجم رغيف الخبز الناتج و زيادة محتواه من البروتين و الألياف و زيادة نشاطه المضاد للأكسدة مقارنة بالكنترول. أوضحت نتائج التقييم الحسي أن جميع عينات الخبز المدعم المنتج كانت مقبولة حتي مستوي إضافة 10%.

**الكلمات المفتاحية:** تدعيم الخبز، دقيق بذور الكركديه، الخصائص الريولوجية، المكسولاب