

## Wheat-Legumes Composite Flours. 2. Nutritional Value of Bread

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### Abstract

This study demonstrated significant effects of legume flours addition to wheat flour on gross chemical composition, phenolic compounds, phytic acid, antioxidant activity and essential amino acids of pan bread baked from wheat-legume composite flours. The results showed that the substituting of wheat flour with different ratio (10, 20 and 30%) of each defatted soybean, sweet lupine, and (5, 10, and 15%) fenugreek flours, separately, increased the protein, ash, crude fiber and fat contents of wheat-legume pan bread with increment the addition of legume flour while the available carbohydrate contents decreased. The bread baked from wheat-legume composite flours exhibited higher phenolics content and antioxidant activity than that determined for wheat bread as control. The 70%wheat-30% sweet lupine pan bread showed higher antioxidant activity than other treatments. Slight increases were observed in phytic acid content in bread made from wheat-soybean and wheat-fenugreek composite flours while it decreased in bread made from wheat-sweet lupine flour blends when compared with wheat bread as control. The substitution of wheat flour with legumes flours increased the essential amino acids contents in baked pan bread except the methionine. However, methionine and lysine values were lower than the FAO/WHO (2007) recommended values.

**Keywords:** *Wheat, Legume, Bread, Phytic, Phenolics, Antioxidant, Amino Acids.*

### 1. Introduction

Bread consumption has increased continuously in many developing countries due to changing eating habits, a steadily growing population (Seibel, 2011). Bread plays an important role in the human diet and relatively large amounts are consumed worldwide (Henchion *et al.*, 2017). A major nutritional limitation of the wheat bread, an inexpensive staple food, is its low content of essential amino acids, particularly lysine. Keeping aside the animal protein, with protein contents double than that of other cereal crops, legumes have been emerged as an economical and environmentally sustain-

able protein source having potential to improve the nutritional value of breads (Xiao *et al.*, 2015). Composite flour is considered advantageous in developing countries as it reduces the importation of wheat flour and encourages the use of locally grown crops as flour (Hasmadi *et al.*, 2014). Local raw materials substitution for wheat flour is increasing due to the growing market for confectioneries (Noor Aziah and Komathi, 2009). Thus, several developing countries have encouraged the initiation of programmes to evaluate the feasibility of alternative locally available flours as a substitute for wheat flour (Abdelghafor *et al.*, 2011). The use of a cereal/leguminous blend may be

real/leguminous blend may be nutritionally convenient in bread and bakery products manufacturing. The legume flour addition to the wheat flour involves the incorporation of a higher protein content but affects the functional and viscoelastic properties of wheat flour dough (Angioloni and Collar 2012). Addition of legume to cereal based products could be a good option for increasing the intake of legumes. In addition, legume proteins are rich in lysine and deficient in sulphur containing amino acids, whereas cereal proteins are deficient in lysine, but have adequate amounts of sulphur amino acids (Eggum and Beame 1983).

The combination of grain with legume proteins would provide better overall essential amino acid balance, helping to overcome the world protein calorie malnutrition problem (Livingstone *et al.*, 1993). Among the legumes tested flour are, defatted soybean (Riaz 1999), Lupine (Aniess *et al.*, 2015) and fenugreek (Afzal *et al.*, 2016). Such legumes are good source of protein, dietary fiber, some vitamins (thiamine, niacin) minerals (Ca, Fe, K, Mg and P), unsaturated fatty acids (linoleic, oleic) and the essential amino acids which are deficient in wheat flour (Zafra *et al.*, 2015). The use of soy flour to improve the nutritional value of wheat bread has been well recognized. Soy enhances the protein quality of wheat bread because of its lysine content which is deficient in wheat. (Mafirimbo *et al.*, 2006). Lupine flour can be incorporated into wheat flour to improve the nutritional value of the final products without detrimental effects on the quality (Pollard *et al.*,

2002). Sweet lupine is a legume high protein and dietary fiber but lower in energy than refined wheat flour (Hall and Johnson, 2004). Sweet lupine seeds also contain carotenoids (Wang *et al.*, 2008) and phenolics and have good antioxidant capacity (Siger *et al.*, 2012). Sweet lupine flour addition to refined wheat flour in bread making reduced the bread glycaemic index (Hall *et al.*, 2005). The intake of fenugreek products may be beneficial for patients who suffer from iron deficiency anemia owing to their nutritive and restorative values (Mahmoud *et al.*, 2012). Fenugreek flour can be also used as supplement to enhance the low nitrogen content of traditional products of cereals and tubers (El Nasri and El Tinay 2007). In the same way 10% germinated fenugreek seed flour has been incorporated into the wheat based biscuits formula resulted in improving their chemical and nutritional quality and additionally also complimented the deficiency of lysine, isoleucine, leucine, threonine and valine, hence neutralizes the amino acid imbalance (Ibrahim and Hegazy 2009).

Bread is the backbone of the Egyptian diet. Cheap, filling and ingrained into Egypt's culture, it's also the only food most poor Egyptian can afford. The most bread makes from wheat flour of various extraction rates (82% for Baladi bread and 72% for Fino and pan bread). Bread baked from wheat flour is a good source of calories and other nutrients but its protein is of low in some indispensable amino acids as lysine and threonine when compared to milk, soybean and lupine proteins. Therefore, the aim of this investigation was

carried out to use the legumes (soybean, sweet lupine and fenugreek) in form of flour to mix them with wheat flour for bread making in order to enhance the protein content and improve the balance of essential amino acids of bread. We have shown in a previous study reported about chemical composition, functional properties and anti-oxidant activity of wheat-legume composite flours (Abdel-Gawad *et al.*, 2016a). In the present study we reported about chemical composition, phenolic compounds, phytic acid, antioxidant activity, and essential amino acids of wheat-legume composite bread.

## 2. Materials and Methods

**2.1 Materials:** Wheat flour extraction rate 72% (cv. *Misr 1*), flours of defatted soybean (*Glycine max*, cv. *Giza 111*), sweet lupine (*Lupinus albus L.*, cv. *Giza*) and fenugreek (*Trigonella foenum-graecum*, cv. *Giza 30*) were used in this study. The legume flours were prepared as described by Abdel-Gawad *et al.*, (2016a). Composite flours were prepared by substituting wheat flour with various portions of each legume flour as shown in Table 1. All prepared samples were used for pan bread making.

**Table 1: Composite flour formulation**

Abbreviation of sample	Blends composition
WF	100% Wheat flour
WF-SF10	90% Wheat flour + 10% soy flour
WF-SF20	80% Wheat flour + 20% soy flour
WF-SF30	70% Wheat flour + 30% soy flour
WF-LF10	90% Wheat flour + 10 lupine flour
WF-LF20	80% Wheat flour + 20% lupine flour
WF-LF30	70% Wheat flour + 30% lupine flour
WF-FF5	95% Wheat flour + 5% fenugreek flour
WF-FF10	90% Wheat flour + 10% fenugreek flour
WF-FF15	85% Wheat flour + 15% fenugreek flour

### 2.2 Preparation of pan bread:

Pan bread was prepared by straight dough method as described by Mostafa and Othman (1986).

**2.3 Analytical Methods:** Protein, ash, crude fiber and fat contents of bread were determined according to official methods as described in A.O.A.C. (2000) and the carbohydrate content was calculated by differences. Total, free and bound phenolic compounds were estimated by the methods described by Abdel-Gawad (1982) by Folin-Ciocalteu method spectrophotometrically (Singleton and Rossi 1965) and the results were

expressed as milligrams of gallic acid equivalents (GAE) per 100 gram of sample on dry weight basis. The phytic acid (IP6) was determined in terms of its phosphorous content, using the method described by Kent-Jones and Amos (1957). The phytic acid was calculated from phytate phosphorus from the weight ratio of phosphorus atoms per molecule of IP6 (1:3.52) according to Abdel-Gawad (2016b). Amino acid analysis were performed on a high performance amino acid analyzer Biochrom 20 (Auto sample version) Pharmacia Biotch constructed at National Center for

Radiation Research and Technology (NCRRT), Cairo, Egypt. The data of each chromatogram was analyzed by EZ Chrom™ chromatography data system tutorial and user's Guide- Version 6.7. Tryptophan amino acid was determined using spectrophotometer method as described by Sastry and Tummuru (1985). The chemical score (CS) was defined and calculated by the equation:

$$CS = \frac{\text{mg of essential amino acid in 1 g of test protein}}{\text{mg of essential amino acid in 1 g of reference protein}} \times 100$$

according to Bhanu *et al.*, (1991).

**2.4 Statistical Analysis:** Data were statistically analyzed by analysis of variance analysis (ANOVA) using a completely randomized factorial design. Basic statistics and ANOVA were performed to test the significance within replications and between treatments (MSTAT-C 1989). LSD tests were used to determine the differences among means at the level of 0.05%

### 3. Results and Discussions

**3.1 Chemical composition of pan bread baked from wheat-legume composite flours:** The chemical composition of pan bread baked from wheat flour substituted with different levels of soybean, lupine and fenugreek flours, individually, are presented in Table 2. The control of wheat bread contained 33.61% moisture, 12.16% Protein, 1.94% ash, 0.85% crude fiber, 4.60% fat, and 80.45% carbohydrate. The chemical composition of wheat bread as control reported in this work are agreement with the results reported by Ahmed (2013) and Afzal *et al.*, (2016). Moisture content of pan bread substituted with legume flours was ranged from 34.16 to 36.68%. It can be observed that pan bread baked from wheat-legume flours had high

moisture content compared to wheat bread as control. Likewise, Ismail (2007) found that present of legumes flour increased the water required for the optimum bread making absorption. Similar observation have been previously also reported by Kasaye and Jha (2015) and Afzal *et al.*, (2016). It can be seen also from Table 2 that protein content of the pan bread increased as proportion of legume flours raised in flour blends.

The pan bread baked from 70% wheat and 30% defatted soybean flour (WF-SE30) had the highest protein content, followed by WF-LF30, WF-SF20 and WF-LF20 bread samples. The lowest protein content for bread made from wheat-legume composite flour was recorded for WF-FF5 bread sample. This could obviously be due to the significant quantity of protein in legume flours (Kasaye and Jha 2015 and Afzal *et al.*, 2016). The high protein content in soy supplemented breads would be of nutritional importance in most developing countries, such as Egypt, because a large sector of low income peoples cannot afford the high price of animal protein sources.

Ash and crude fiber contents of all pan bread made from composite flours were higher than that of wheat bread and increased significantly ( $P < 0.05$ ) with increment the substituting of wheat flour with legume flours (Table 2). Among the bread made from composite flours, WF-SF30 pan bread showed high ash content (3.87%) followed by WF-SF20 bread sample (3.16%). The highest crude fiber content (2.15%) was recorded for WF-LF30, while the lowest content (1.20%) was found in WF-SF10 bread. The increase in ash and crude fiber contents of composite flours may be due to the higher ash and fiber contents of legumes flours than in wheat flour. Similar findings were reported previously by Ahmed

(2014) and Kasaye and Jha (2015). Significant differences in fat content of pan bread made from wheat-legumes flours were also observed, the mean values showed high fat content for WF-LF30 (6.96%), followed by WF-LF20 (6.25%), WF-FF15 (5.57%) and WF-LF10 (5.53%) bread samples. The lowest of fat content (4.51%) was recorded for

WF-SF 10 bread. On the other hand, the available carbohydrate content of pan bread decrease as proportion of legumes flour increased in the flour blends. This may be due to the higher carbohydrate content of wheat flour compared to legume flours. Such trend was supported previously by Ahmed (2014) and Kasaye and Jha (2015).

Table 2: Chemical composition of pan bread baked from wheat flour and wheat-legume composite flours

Bread Samples	Protein* (%)	Ash* (%)	Crude fiber* (%)	Fat* (%)	Carbohydrate** (%)
WF	12.16 ± 1.10 <sup>f</sup>	1.94 ± 0.05 <sup>g</sup>	0.85 ± 0.10 <sup>f</sup>	4.60 ± 0.18 <sup>g</sup>	80.45 ± 1.86 <sup>a</sup>
WF-SF10	15.82 ± 1.06 <sup>cd</sup>	2.71 ± 0.014 <sup>cd</sup>	1.20 ± 0.071 <sup>e</sup>	4.51 ± 0.15 <sup>h</sup>	75.76 ± 2.16 <sup>c</sup>
WF-SF20	17.48 ± 1.16 <sup>b</sup>	3.16 ± 0.039 <sup>b</sup>	1.71 ± 0.065 <sup>cd</sup>	4.76 ± 0.24 <sup>f</sup>	72.89 ± 2.64 <sup>d</sup>
WF-SF30	19.56 ± 1.36 <sup>a</sup>	3.87 ± 0.031 <sup>a</sup>	2.05 ± 0.013 <sup>ab</sup>	5.07 ± 0.57 <sup>e</sup>	69.45 ± 2.57 <sup>e</sup>
WF-LF10	15.16 ± 1.13 <sup>cd</sup>	2.44 ± 0.28 <sup>e</sup>	1.26 ± 0.067 <sup>e</sup>	5.53 ± 0.14 <sup>c</sup>	75.61 ± 1.90 <sup>c</sup>
WF-LF20	16.19 ± 1.23 <sup>c</sup>	2.86 ± 0.081 <sup>c</sup>	1.82 ± 0.078 <sup>c</sup>	6.25 ± 0.28 <sup>b</sup>	72.88 ± 2.36 <sup>d</sup>
WF-LF30	19.38 ± 1.55 <sup>a</sup>	2.91 ± 0.016 <sup>c</sup>	2.15 ± 0.13 <sup>a</sup>	6.96 ± 0.12 <sup>a</sup>	68.60 ± 3.11 <sup>e</sup>
WF-FF5	13.39 ± 0.99 <sup>e</sup>	2.17 ± 0.087 <sup>f</sup>	1.24 ± 0.09 <sup>e</sup>	5.04 ± 0.22 <sup>e</sup>	78.16 ± 2.22 <sup>b</sup>
WF-FF10	14.82 ± 1.14 <sup>cd</sup>	2.41 ± 0.02 <sup>e</sup>	1.61 ± 0.027 <sup>d</sup>	5.35 ± 0.37 <sup>d</sup>	75.81 ± 2.44 <sup>c</sup>
WF-FF15	15.83 ± 1.18 <sup>cd</sup>	2.57 ± 0.059 <sup>de</sup>	2.01 ± 0.16 <sup>b</sup>	5.57 ± 0.71 <sup>c</sup>	74.02 ± 3.00 <sup>d</sup>

Values are the mean of triplicate determinations with standard division.

The different letters at the column means significant differences at ( $p \leq 0.05$ ) and the same letters means No significant differences.

\* Determined on dry weight basis.

\*\* Carbohydrates were calculated by difference.

### 3.2 Phenolic compounds, phytic acid and antioxidant activity:

The phenolic compounds, phytic acid and antioxidant activity of pan bread baked from wheat flour and wheat-legume composite flours are shown in Table 3. The free, bound and total phenolics contents of bread made from composite flours showed increasing with increment the proportion of legume flours. This indicated the high phenolics contents in legume flours than in wheat flour. The previous investigations of Dhingra and Jood (2001) on defatted soybean, Sosulski and Dabrowski (1984) on lupine flour and Afzal *et al.*, (2016) on

fenugreek flour indicated higher content of phenolic compounds in these legume flours than that of wheat flour. The bread baked from WF-SF30 flour showed highest free phenolics, while the bread made from WF-LF10 flour exhibited the lowest value comparing to breads made from other wheat-legume composite flours. Moreover, the bound phenolics showed highest value in WF-SF30 bread and lowest in WF-FF5 bread as compared with breads of other wheat-legume composite flours (Table 3). Among the bread from wheat-legume composite flours, the total phenolics were the highest in WF-SF30 bread



(255.97 mg/100g) and the lowest in WF-FF5 bread (227.96 mg/100g).

The phytic acid as an anti-nutrient interferes with the availability of some important minerals e.g. calcium, iron and zinc has assumed greater significance from nutrition point of many years ago. Phytate phosphorus and phytic acid contents increased significantly in breads with increasing the substitution of wheat flour with soybean or fenugreek flours (Table 3). Dhingra and Jood (2001) reported increase in phytic acid content (252.2 mg/100g) of pan bread baked from 90% wheat-10% defatted soybean composite flour when compared with 225.6 mg/100g in wheat bread. In contrast, the all bread made from wheat-lupine composite flours showed lower phytic acid content comparing to bread made from wheat flour as control or bread baked from wheat-soybean or wheat-fenugreek composite flours (Table 3); which indicating to low phytate in prepared lupine flour because the soaking of lupine seeds dur-

ing the preparation of lupine flour. Abdel-Gawad (2016b) determined the different forms of inositol phosphates in some soaked legumes and described the degradation of inositol hexaphosphate (phytic acid) during soaking was due to the activation of the endogenous enzyme phytases and leakage to water of soaking.

The scavenging activity of methanolic extracts against DPPH in pan bread were increased significantly ( $p < 0.05$ ) with increment the proportion of legumes flour in wheat-legume composite flours (Table 3). The pan bread baked from WF-SF30 and WF-LF30 samples had high antioxidant activity 6.73 and 6.67%; respectively; while wheat pan bread as control had the lowest value of antioxidant activity (2.97%). These results indicated that, generally, the legumes had higher scavenging activity than wheat flour. Similar observations were reported by Zhang *et al.*, (2011) for soy bean, Siger *et al.*, (2012) for lupine and Afzal *et al.*, (2016) for fenugreek.

**Table 3: Phenolic compound, phytic acid and antioxidant activity of pan bread baked from wheat-legume composite flours.**

Bread samples	Phenolic compound mg/100g			Phytate phosphorus mg/100g	phytic acid mg/100g	Antioxidant activity%
	Free	Bound	Total			
WF	2.94 ± 0.26 <sup>f</sup>	221.21 ± 0.85 <sup>h</sup>	224.15 ± 1.20 <sup>e</sup>	66.09 ± 0.78 <sup>e</sup>	234.49 ± 1.56 <sup>e</sup>	2.97 ± 0.15 <sup>h</sup>
WF-SF10	3.45 ± 0.20 <sup>de</sup>	230.43 ± 0.86 <sup>e</sup>	233.88 ± 1.36 <sup>e</sup>	69.35 ± 0.48 <sup>c</sup>	246.07 ± 0.89 <sup>c</sup>	4.15 ± 0.22 <sup>e</sup>
WF-SF20	4.01 ± 0.33 <sup>c</sup>	238.20 ± 0.75 <sup>b</sup>	242.21 ± 1.19 <sup>b</sup>	73.43 ± 0.43 <sup>b</sup>	260.54 ± 0.67 <sup>b</sup>	4.81 ± 0.16 <sup>c</sup>
WF-SF30	5.66 ± 0.18 <sup>a</sup>	250.31 ± 0.56 <sup>a</sup>	255.97 ± 0.95 <sup>a</sup>	76.70 ± 0.57 <sup>a</sup>	272.12 ± 1.13 <sup>a</sup>	6.73 ± 0.45 <sup>a</sup>
WF-LF10	3.15 ± 0.46 <sup>ef</sup>	225.47 ± 0.62 <sup>e</sup>	228.62 ± 1.15 <sup>f</sup>	65.26 ± 0.33 <sup>f</sup>	231.55 ± 0.83 <sup>e</sup>	3.75 ± 0.26 <sup>ef</sup>
WF-LF20	3.57 ± 0.37 <sup>d</sup>	232.18 ± 0.51 <sup>d</sup>	235.75 ± 1.53 <sup>d</sup>	63.55 ± 0.28 <sup>e</sup>	225.47 ± 0.76 <sup>f</sup>	4.46 ± 0.35 <sup>d</sup>
WF-LF30	4.36 ± 0.52 <sup>c</sup>	238.52 ± 0.57 <sup>b</sup>	242.88 ± 1.62 <sup>b</sup>	62.01 ± 0.38 <sup>h</sup>	220.01 ± 0.95 <sup>e</sup>	6.67 ± 0.53 <sup>a</sup>
WF-FF 5	3.38 ± 0.27 <sup>de</sup>	224.58 ± 0.39 <sup>e</sup>	227.96 ± 1.10 <sup>f</sup>	68.13 ± 0.64 <sup>d</sup>	241.72 ± 1.23 <sup>d</sup>	3.50 ± 0.61 <sup>e</sup>
WF-FF10	4.31 ± 0.45 <sup>c</sup>	228.12 ± 0.48 <sup>f</sup>	232.43 ± 0.87 <sup>e</sup>	72.58 ± 0.51 <sup>b</sup>	257.65 ± 1.17 <sup>b</sup>	3.93 ± 0.25 <sup>ef</sup>
WF-FF15	4.83 ± 0.19 <sup>b</sup>	235.73 ± 0.58 <sup>c</sup>	240.56 ± 1.43 <sup>c</sup>	76.26 ± 0.46 <sup>a</sup>	270.57 ± 0.97 <sup>a</sup>	5.57 ± 0.49 <sup>b</sup>

Values are the mean of triplicate determinations with standard division.

The different letters at the column means significant differences at ( $p \leq 0.05$ ) and the same letters means No significant differences.

### 3.3 Essential amino acid, chemical score and limiting amino acids:

The value of food quality is judged by its protein content, the digestibility of protein, number and amounts of essential amino acids. The essential amino acids content of pan bread baked from wheat and wheat-legume composite flours are presented in Tables 4, 5 and 6. The content of amino acids lysine and threonine of all pan bread baked from wheat-legume composite flours were increased as legume flours increment in the formulation of bread. The amino acid lysine in bread of wheat flour was 1.92 g/100g protein, and increased by 16.70, 20.31 and 25.52% in bread made from wheat-soybean composite flours at levels of 10, 20 and 30% defatted soy flour; respectively (Table 4). However, the lysine content in bread made from wheat-soybean composite flour was lower than the value (6.9g/100g protein) recommended by FAO/WHO (2007). The threonine content of wheat bread was 2.91g/100g protein, and increased by 5.50, 7.56 and 10.31% in bread made from wheat-defatted soybean composite flours at levels of 10, 20 and 30% defatted soybean flour; respectively. The content of threonine amino acid in bread of wheat and wheat-defatted soybean composite flours was lower than the

value (4.4g/100g protein) recommended by FAO/WHO (2007). Valine, isoleucine, leucine, phenylalanine and tyrosine contents showed increasing in bread made from wheat-soybean composite flours with increment the substitution of wheat flour with defatted soybean flour than that determined in wheat bread as control. As expected, the content of sulfur-containing amino acids (methionine and cystine) in bread was decreased as the ratio of defatted soybean in composite flours increased (Table 4). Generally, it's known that legumes are poor in the sulfur-containing amino acids methionine and cystine (Khan *et al.*, 2009). The amino acid with lowest value is the first limiting amino acid and this value also is the amino acid score for the test protein; and the amino acid scoring pattern methods are based on assumption that there is a direct relationship between the concentration of a limiting amino acid and utilization of the limiting amino acid in a protein Young and Pellett (1991). The data in Table 4 showed that the first limiting amino acid in wheat bread as control and bread from wheat-defatted soybean composite flours was the lysine and the second limiting amino acid was tryptophan.

Table 4: Essential amino acids and chemical score of pan bread baked from wheat flour and wheat-soybean composite flours.

Essential amino acids (EAA)	Bread samples								Average EAA content of Human Milk*** (FAO/WHO, 2007)
	WF		WF-SF10		WF-SF20		WF-SF30		
	EAA*	CS**	EAA*	CS**	EAA*	CS**	EAA*	CS**	
Valine	4.76	87	4.79	87	4.81	88	4.87	89	55
Methionine +Cystine	2.90	88	2.81	85	2.74	83	2.66	81	33
Isoleucine	3.85	70	4.04	73	4.09	74	4.14	75	55
Leucine	6.53	68	6.57	68	6.65	69	6.69	70	96
Phenylalanine +tyrosine	8.60	102	8.67	92	8.82	94	9.14	97	94
Lysine	1.92	<u>28</u>	2.24	<u>32</u>	2.31	<u>33</u>	2.41	<u>35</u>	69
Threonine	2.91	66	3.07	70	3.13	71	3.21	73	44
Tryptophan	0.81	<u>48</u>	0.81	<u>48</u>	0.79	<u>46</u>	0.78	<u>46</u>	17
Total EAA	32.28		33.00		33.34		33.90		

\*EAA: Essential amino acid (g EAA/ 100g protein). \*\*CS: Chemical score (mg EAA/g protein). \*\*\* Human milk: mg EAA/g protein (FAO/WHO, 2007).

The contents of essential amino acids lysine and threonine (g/100g protein) in bread of wheat-sweet lupine composite flours were increased as the substitution of wheat flour with lupine flour raised (Table 5), but the content of lysine amino acid was lower than recommended by FAO/WHO (2007). Leucine content was increased in bread of wheat-sweet lupine composite flour by 2.6%, 4.4% and 9.2% with raising the proportion of sweet lupine flour at level of 10, 20 and 30%; respectively, comparing to wheat flour bread as control, but the obtained data were lower than the value (9.6g/100 g protein) recommended in FAO/WHO (2007). The essential amino acids valine, methionine+cystine, isoleucine, and phenylalanine+tyrosine were decreased in bread of wheat-sweet lupine composite flour as proportion of sweet lupine flour increased in flour blends (Table 5). However, valine, methionine+cystine and isoleucin values were higher than recommended by FAO/WHO (2007). The chemical score and limiting amino acids for bread baked from wheat-sweet lupine composite flours

showed another pattern depending on the ratio of sweet lupine flour in flour blend (Table 5). In the bread baked from 90% wheat-10% lupine flour blend, the first limiting amino acid was lysine and the second was tryptophan, whereas in bread baked from 80% wheat-20% lupine or 70% wheat-30% lupine flour blends, the first limiting amino acid was tryptophan and the second was lysine.

The essential amino acid composition of bread made from wheat and wheat-fenugreek composite flours are shown in Table 6. The essential amino acids isoleucine, leucine, phenylalanine+tyrosine, threonine and tryptophan contents were increased as the proportion of fenugreek flour increased in flour blends, and the determined values of these acids were higher for isoleucine, threonine and equal for phenylalanine+tyrosine, while lower for tryptophan than recommended by FAO/WHO (2007). Lysine was also increased in bread of wheat-fenugreek composite flours as the fenugreek flour increment in flour blends.



Table 5: Essential amino acids and chemical score of pan bread baked from wheat flour and wheat-sweet lupine composite flours.

Essential amino acids (EAA)	Bread samples								Average EAA content of Human Milk*** (FAO/WHO, 2007)
	WF		WF-LF10		WF-LF20		WF-LF30		
	EAA*	CS**	EAA*	CS**	EAA*	CS**	EAA*	CS**	
Valine	4.76	87	4.32	79	4.10	75	3.91	71	55
Methionine +Cystine	2.90	88	2.76	84	2.72	82	2.68	81	33
Isoleucine	3.85	70	3.76	68	3.69	67	3.64	66	55
Leucine	6.53	68	6.70	70	6.82	71	7.13	74	96
Phenylalanine +tyrosine	8.60	102	8.16	87	7.64	81	7.33	78	94
Lysine	1.92	<u>28</u>	2.81	<u>41</u>	3.53	<u>51</u>	3.80	<u>55</u>	69
Threonine	2.91	66	3.15	72	3.36	76	3.53	80	44
Tryptophan	0.81	<u>48</u>	0.81	<u>48</u>	0.79	<u>46</u>	0.78	<u>46</u>	17
Total EAA	32.28		32.47		32.65		32.80		

\*EAA: Essential amino acid (g EAA/100g protein). \*\*CS: Chemical score (mg EAA/g protein). \*\*\* Human milk: mg EAA/g protein (FAO/WHO, 2007).

However, the determined values of lysine in all bread studied were lower than that recommended by FAO/WHO. Valine and methionine+cystine contents showed decreasing in bread with the substitution of wheat flour with high proportions of fenugreek flour but the content of valine still higher than recommended for FAO/WHO. The limiting amino

acids for the bread baked from wheat-fenugreek composite flours (Table 6) were similar to that observed for bread from wheat-soybean composite flours, where the lysine was the first and tryptophan the second limiting amino acid. These findings are good agreement with that found by Mubarak (2001) and Mahmoud *et al.*, (2012).

Table 6: Essential amino acids and chemical score of pan bread baked from wheat flour and wheat-fenugreek composite flours.

Essential amino acids (EAA)	Bread samples								Average EAA content of Human Milk*** (FAO/WHO, 2007)
	WF		WF-FF5		WF-FF10		WF-FF15		
	EAA*	CS**	EAA*	CS**	EAA*	CS**	EAA*	CS**	
Valine	4.76	87	4.05	74	3.97	72	3.89	71	55
Methionine +Cystine	2.90	88	2.78	84	2.96	82	2.64	80	33
Isoleucine	3.85	70	3.78	70	3.91	71	3.95	72	55
Leucine	6.53	68	6.68	70	6.90	72	7.08	74	96
Phenylalanine +tyrosine	8.60	102	8.80	94	8.88	94	8.91	95	94
Lysine	1.92	<u>28</u>	3.06	<u>44</u>	3.10	<u>45</u>	3.12	<u>45</u>	69
Threonine	2.91	66	2.99	68	3.13	71	3.24	74	44
Tryptophan	0.81	<u>48</u>	0.85	<u>50</u>	0.88	<u>52</u>	0.93	<u>55</u>	17
Total EAA	32.28		32.99		33.73		33.76		

\*EAA: Essential amino acid (g EAA/100g protein). \*\*CS: Chemical score (mg EAA/g protein). \*\*\* Human milk: mg EAA/g protein (FAO/WHO, 2007).

#### 4. Conclusion:

This study demonstrated that the nutritional and bioactive composition of refined wheat flour bread is sig-

nificantly improved with addition of legume flours, indicating that wheat-legume bread may have useful nutrition and health functionality. Signifi-

cant effects of legume type on the nutrition and bioactive composition of wheat-legume bread were also revealed. Our findings suggested that defatted soy bean, sweet lupine, and fenugreek flours may be added, separately, with different ratio to wheat bread to give improved protein, ash, dietary fiber contents, essential amino acid balance and bioactive component as phenolic compounds as well as antioxidant activity.

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## دقيق القمح والبقوليات المركب. ٢ . القيمة الغذائية للخبز

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## الملخص

أجريت هذه الدراسة بهدف معرفة تأثير إضافة دقيق البقوليات لدقيق القمح علي التركيب الكيميائي العام وحامض الفيتيك والمركبات الفينولية والنشاط المضاد للاكسده والأحماض الامينية الضرورية لخبز القوالب المصنع من الدقيق المركب للقمح والبقوليات. أظهرت النتائج إن أحلال دقيق القمح بنسب مختلفة (١٠ ، ٢٠ ، ٣٠%) من كل من دقيق فول الصويا منزوع الدهن ودقيق الترمس الحلو ودقيق الحلبة بنسب (٥ ، ١٠ ، ١٥%) كلا علي حده أدت إلي زيادة نسبة البروتين والرماد والألياف الخام والدهون ، بينما انخفضت نسبة الكربوهيدرات القابلة للهضم في خبز القوالب المصنع من دقيق القمح والبقوليات المركب بزيادة نسبة دقيق البقوليات المضافة. ولقد استبان من الدراسة أن خبز القوالب المصنع من الدقيق المركب للقمح والبقوليات احتوي علي نسبة مرتفعة من المركبات الفينولية والنشاط المضاد للاكسده مقارنة بخبز القوالب المصنع من دقيق القمح فقط كعينة كنترول. إضافة إلي ذلك فقد اظهر الخبز المصنع من ٧٠% دقيق قمح و ٣٠% دقيق ترمس حلو علي نشاط مرتفع لمضادات الاكسده مقارنة بالمعاملات الاخرى موضع الدراسة . كما اظهر الخبز المصنع من الدقيق المركب للقمح وفول الصويا او دقيق القمح والحلبة زيادة طفيفة في محتوى حامض الفيتيك بينما انخفض هذا المحتوى في الخبز المصنع من الدقيق المركب للقمح والترمس الحلو مقارنة بالخبز المصنع من القمح فقط. وادي إحلال دقيق القمح بدقيق البقوليات إلي ارتفاع نسبة الأحماض الامينية الضرورية في الخبز المصنع ماعدا الحامض الاميني الضروري الميثونين. وعموما فان نسبة الأحماض الامينية الضرورية الليسين والمثيونين في الخبز المصنع كانت اقل من النسب الموصي بها من قبل منظمة الفاو ومنظمة الصحة العالمية.