International Journal of Agricultural Research

ISSN 1816-4897

Academic Journals Inc.

www.academicjournals.com

International Journal of Agricultural Research 9 (5): 219-230, 2014
ISSN 1816-4897 / DOI: 10.3923/ijar.2014.219.230

Comparative Screening of Phytochemicals in Egyptian and Hungarian Wheat Varieties

[1]Safaa S. Abozed, [1]Zahra S. Ahmed, [2]Mariann Rakszegi and [2]Làszló Làng
[1]Food Science and Nutrition Division, National Research Centre, Egypt
[2]Centre for Agricultural Research, Agricultural Institute, Hungarian Academy of Sciences, Hungary

Corresponding Author: Zahra S. Ahmed, Food Science and Nutrition Division, National Research Centre, Egypt

ABSTRACT

Four bread wheats grown in Egypt and Hungary were evaluated for their chemical composition, physical properties, phytochemical content and antioxidant activity. The thousand kernel weight, the wet gluten content, the Zeleny sedimentation as well as the titratable acidity were significantly ($p<0.05$) affected by the varieties and/or the growing sites. Chemical composition varied significantly among tested samples from which moisture and fat contents of Egyptian samples were significantly lower compared to the Hungarian varieties. The evaluation of the antioxidant activity was performed *in vitro* by the measurement of the DPPH radical scavenging activity, while quantitative analysis were also carried out on the total tocol, total phenolic and total flavonoid content of whole wheat. Results showed that all wheat samples exhibited significant antioxidant activities and contained significant levels of phenolic compounds. Among the tested samples, DPPH-RSA ranged from 19.94±0.9% (Mv-Karizma) to 29.9±0.8% (Ben-Su1) dw. Total phenolic content ranged from 1.65 (Mv-Karizma) to 2.21 (Ben-Su3) expressed as mg Gallic Acid Equivalent (GAE) per gram dry sample, while the flavonoid content ranged from 2.18 (Mv-Karizma) to 8.42 (Ben-Su1) μg rutin equivalent/g dry sample. Lycopene content was also significantly different ($p<0.05$) among cultivars. Mainly α and β-tocopherols and α and β-tocotrienols were found in all varieties of whole wheat, though γ-tocopherol was detected in traces. β-tocotrienol was the predominant form of vitamin E found in all varieties and ranged from 23.13 (Ben-Su1) to 36.77 (Ben-Su3) μg g^{-1} dw. According to the total tocol content, tested varieties were ranked ascending 37.65 (Ben-Su1) <41.77 (Mv-Karizma) <41.95 (Mv-Suba) <53.66 (Ben-Su3) μg g^{-1} dw. This wide range in the phytochemical content of the screened wheat cultivars refers to the presence of the genetic potential for the development of specific wheat genotypes with health promoting properties.

Key words: Wheat, phenolic, flavoniods, tocopherol, antioxidants, DPPH, HPLC

INTRODUCTION

Wheat is a staple food worldwide contributing to the basic nutrition with nutrients such as carbohydrates, proteins, dietary fibers, vitamins and minerals as well as phytochemicals. The well-known phytochemicals in whole grains of wheat are: Dietary fibres, vitamins, minerals, lignans, phytoestrogens, phenolic compounds and phytic acids, such as benzoic and cinnamic acids, anthocyanidins, quinines, flavonols, chalcones, flavones, flavonones and phenolic compounds, flavonoids, coumarin derivatives, polyphenols, phytosterines, saponins, catechins, tocotrienols and tocopherols, tannin, cartenoids, ferulic acid and diferulates (Adom and Liu, 2002; Zielinski and

Kozlowska, 2000; Slavin, 2003; Adom *et al*., 2003; Jones *et al*., 2004). Antioxidants are one group of wheat phytochemicals including carotenoids, tocopherols, tocotrienols, phenolic acids, phytic acid, phytosterols and flavonoids (Halliwell and Gutteridge, 1990).

Wheat phytochemical compounds attracted substantial attention as protective functional food components in the last decade. Such compounds exhibit antioxidant activity i.e., the ability to scavenge free radicals that may oxidize biologically relevant molecules (Liu *et al*., 2008; Nurmi *et al*., 2008).

These antioxidative components may prevent life important molecules such as DNA and enzymes from oxidative damages through different mechanisms. For instance, wheat antioxidants may directly react with the Reactive Oxygen Species (ROS), such as hydroxyl radicals or singlet oxygen molecules, to terminate the attack of the latter on biological molecules. Increased consumption of whole wheat has been correlated with a reduced risk of cardiovascular diseases and certain cancers (Liu, 2007; Lampi *et al*., 2008).

The phytochemical content of whole grains needs closer examination due to their potential health benefit in the prevention of chronic diseases. A more complete analysis of the phytochemical content and antioxidant activity of a range of diverse whole wheat samples have already been analyzed in the Healthgrain EU FP6 project (Ward *et al*., 2008). In the frame of this project a GxE study was also carried out and those components were identified which could possibly be good candidates for breeding purposes (Shewry *et al*., 2010). These properties are mostly genetically determined and less affected by the environment. Recently, ten different Egyptian bread wheat cultivars were screened for its content of antioxidant capacities within the joint Egyptian-American research project. Results showed significant differences in the antioxidant activities of the screened cultivars (Ahmed *et al*., 2013).

As a part of our ongoing investigations on natural antioxidants of cereals, this work aims to study the phytochemical profiles and antioxidant activity of four varieties originating from two different geographic regions namely Egypt and Hungary. The health related benefits of two Egyptian varieties were investigated and analyzed in relation to two Hungarian varieties.

This study will finally contribute to the identification of the nutritional benefits of wheat related to phytochemicals and gives the possibility to support plant breeders and food processing industries with this additional information.

MATERIALS AND METHODS

Experimental materials: The 100 g grain samples of four bread wheat cultivars were used as experimental materials for this study, from which two originated from Egypt: Beni-Sueif-1 (Ben-Su1), Beni-Sueif-3 (Ben-Su3) and two from Hungary: Mv-Suba and Mv-Karizma.

Growing conditions: Mv-Suba and Mv-Karizma samples were harvested in the field at the AI CAR HAS (2011, Martonvàsàr, Hungary). The plots were 2 m long, with six rows spaced at a distance of 20 cm. Samples received complex chemical fertilizer during autumn (80 kg ha^{-1} 34% N, P, K) and they received ammonium nitrate (60 kg ha^{-1} 34% N) during spring.

After the extremely wet 2010 (>1,000 mm rain), 2011 proved to be the driest year in the last several decades (305 mm) compared to the average of 550 mm (Table 1). Due to the dry conditions, no serious disease epidemics occurred. The national wheat average reached only 4.04 t ha^{-1} which is equal to the average of the last 20 years. The quality of harvested wheat was good with relatively low protein content.

Table 1: Climatic conditions for the two growing sites during October to 30 July

Parameters	2010/2011 Egypt	2010/2011 Hungary
Precipitation (average) (mm)	26.0	305.0
Mean temperature (mm)	22.0	8.6
No. of days with T min≤0 (°C)	0.0	107.0
No. of days with T min≤-10 (°C)	0.0	14.0
No. of days with T max≥25 (°C)	90.0	57.0
No. of days with T max≥30 (°C)	30.0	13.0
No. of days with T max≥35 (°C)	30.0	3.0
Absolute min temperature (°C)	9.0	-23.7
Absolute max temperature (°C)	42.7	36.3

Ben-Su1 and Ben-Su3 Egyptian wheat cultivars were grown at the experimental station belongs to Agriculture Research Centre, Giza-Egypt and was harvested in the season 2011. Wheat seeds were planted in rows spaced 12-15 cm between adjacent plants in the row and 3-5 cm depth. Side dressing was applied with 85 to 115 kg of ammonium nitrate per hectare when plants reach 35 to 65 cm in height.

Sample preparation: Wheat samples were harvested at maturity and manually cleaned to remove all non-grain debris present following harvest. All cultivars were tempered to 14% moisture and milled using a Brabender Quadrumat Junior (Brabender GmbH and Co. KG, Duisburg, Germany) according to AACC (2000) Method 26-50. All milled samples were stored in plastic containers at -20°C until analysis.

Analytical methods: Experimental samples were compared using a range of widely used standard methods and instruments: Thousand kernel weights was calculated as grams per thousand kernels according to Hungarian standard (MSZ6367/4-86), moisture content was measured by ICC 109/1 while ash content was analyzed by ICC 104/1 method. Crude protein content was determined by Kjeltec 1035 Analyzer (ICC105/2), crude fat content was measured according to ICC 136 method. Gluten content was measured by Glutomatic 2200 (ICC 137/1, ICC 155) while bread volume was estimated by Zeleny sedimentation test (ICC 116/1). Acidity of wheat samples were measured by standard No. ICC 145. Starch content was estimated by near infrared spectroscopy method ICC 202.

Phytochemical composition

Total phenolic content (TPC): Total phenolic contents of flour extracts were determined using Folin-Ciocalteu method (Yu *et al*., 2002). The reaction mixture contained 100 μL of flour extracts and 500 μL of the Folin-Ciocalteu reagent and 1.5 mL of 20% sodium carbonate. The final volume was made up to 10 mL with water. After 2 h of reaction, the absorbance at 765 nm was determined and used to estimate the phenolic contents using a standard curve prepared using Gallic acid.

Total falvonoid content (TFC): Flavonoid contents of wheat samples were determined using the aluminum chloride colorimetric method (Chang *et al*., 2002) based on the method of Woisky and Salatino (1998). The appropriate dilution of extracts (0.5 mL) were mixed with 1.5 mL of 95%

ethanol, followed by 0.1 mL of 10% aluminum chloride, 0.1 mL of 1 M potassium acetate and 2.8 mL of distilled water. After incubation at room temperature for 30 min, the absorbance of the reaction mixture was measured at 415 nm with Shimadzu UV-160A spectrophotometer. The flavonoid content was calculated using a standard calibration of rutin solution and expressed as micrograms of Rutin Equivalent (RE) per gram of sample.

Lycopene: Lycopene was determined according to the method of Nagata and Yamashita (1992). A fine dried powder (150 mg) was vigorously shaken with 10 mL of acetone-hexane mixture (4:6) for 1 min and filtered through Whatman No. 4 filter paper. The absorbance of the filtrate was measured at 453, 505, 645 and 663 nm. Contents of lycopene were calculated according to the following equation:

$$\text{Lycopene (mg/100 mL)} = 0.0458\times A663\pm0.204\times A645\pm0.372\times A505\pm0.0806\times A453$$

Assay of DPPH radical scavenging activity: DPPH radical scavenging capacities of wheat extracts were determined by the reduction of the reaction color between DPPH solution and sample extracts as previously described by Huang *et al.* (2005). A final concentration of DPPH solution used was 0.15 mM DPPH solution (3.9 mL) was mixed with sample solution (0.1 mL). The mixture was kept in the dark at ambient temperature. The absorbance of the mixtures was recorded at 515 nm for exactly 30 min. Blank was made from 3.9 mL of DPPH and 0.1 mL methanol and measured absorbance at t = 0. The scavenging of DPPH was calculated according to the following equation (Liyana-Pathirana and Shahidi, 2007):

$$\text{DPPH scavenging (\%)} = \frac{\text{Abs}_{(t=0)} - \text{Abs}_{(t=30)}}{\text{Abs}_{(t=0)}}\times 100$$

where, $\text{Abs}_{(t=0)}$ is absorbance of DPPH radical + methanol at t = 0 min, $\text{Abs}_{(t=30)}$ is absorbance of DPPH radical + phenolic extracts at t = 30 min.

Tocols: Total tocol content and its components, the α-, β-tocopherols and α-, β-tocotrienols were quantitatively determined by the NP-HPLC method of Lampi *et al.* (2008). After the saponification and extractions of the samples, HPLC separation was carried out. Separation was performed using an Inertsil silica column (5 μm, 250 mm×4.6 mm; Varian Chromapack, Middelburg, The Netherlands) with a silica guard column (Guard-Pak Silica, Waters, Milford, MA) and a mobile phase containing 3% of 1, 4-dioxane in heptane at a flow rate of 2 mL min^{-1} at 30°C. FLD was set at λex = 292 nm and λem = 325 nm. Each whole meal sample was worked up in duplicate and each tocol standard solution was analyzed twice by HPLC.

Statistical analysis: All tests were conducted in triplicate. Data were reported as Mean±SD. One way analyses of variance Duncan test were conducted to identify differences among means. Statistical significance was declared at $p<0.05$.

RESULTS AND DISCUSSION

Proximate composition: Basic chemical compositions, such as moisture, ash, protein, crude fat and starch contents (%) of the tested whole wheat flours were measured (Table 2).

Table 2: Chemical composition of Egyptian and Hungarian wheat samples (g/100 g dw)

Wheat samples	Moisture	Ash	Fat	Protein*	Starch**
Ben-Su1	10.00±0.11^{b}	1.59±0.11^{b}	1.69±0.51^{b}	13.21±0.00^{a}	73.7±0.6
Ben-Su3	9.36±0.01^{c}	2.06±0.09^{a}	1.47±0.61^{b}	11.42 ±0.01^{c}	75.6±0.8
Mv-Suba	11.29±0.02^{a}	1.52±0.10^{b}	3.01±0.22^{a}	13.26±0.11^{a}	56.1±1.1
Mv-Karizma	11.36±0.07^{a}	1.32±0.02^{c}	3.67±0.11^{a}	12.05±0.17^{b}	58.5±1.3

*N ×5.7, **Measured by FOSS Tecator 1241, values are averages of three repetitions, values within columns followed by different upper case letters are significantly different ($p<0.05$)

Table 3: Agronomic and physical properties of Egyptian and Hungarian wheat samples

Wheat samples	*TKW (g)	Extraction yield (%)	Zeleny (mL)	Wet gluten content (%)	pH	Titratable acidity (%)
Ben-Su1	56.5±0.9^{a}	31.7	16.6±0.7^{d}	23.6±0.7^{d}	6.57±0.00^{a}	0.01±0.0^{c}
Ben-Su3	39.0±0.3^{c}	23.9	21.7±0.1^{c}	33.0±1.0^{c}	6.41±0.00^{a}	0.01±0.0^{c}
Mv-Suba	46.0±1.0^{b}	49.3	61.0±0.9^{a}	36.4±0.9^{b}	6.59±0.00^{a}	0.02±0.0^{b}
Mv-Karizma	42.0±0.7^{b}	57.1	57.0±1.1^{b}	38.2±0.9^{a}	6.54±0.10^{a}	0.06±0.0^{a}

*TKW: Thousand kernel weight, values are averages of three repetitions, values within columns followed by different upper case letters are significantly different ($p<0.05$)

Wheat grain grown in Hungary had significantly higher moisture content than those grown in Egypt, which should be resulted by the different climatic conditions of the two countries.

Moisture has significant effect on wheat quality during storage. The flour produced from dry and sound grains can be kept for longer periods if properly stored but the one produced from wet grains deteriorates dramatically within a few days (Pomeranz, 1988). The variation in the moisture content of different wheats might be attributed to climatic factors experienced during growth period, harvest time and storage (Slaughter *et al*., 1992; Mahmood, 2004).

The ash content is one of the quality indicators of flour yield; hence the wheat with lower content of ash may have more endosperm and ultimately good yield after flour extraction (Williams *et al*., 1986). The ash contents for wheat grains showed significant difference between wheat samples where Ben-Su3 exhibited the highest ash content (2.06%) and Mv-Karizma gave the lowest values (1.32%).

The main quality determinant components of wheat are the storage proteins and their composition. In our study the crude protein content of the tested varieties varied significantly and ranged from 11.42 to 13.26% for Ben-Su3 and Mv-Suba, respectively.

Starch is a major component of wheat flour, accounting for 65-70% of the dry weight of the wheat grain. The average starch content of wheat samples grown in Egypt (74.65% dw) was significantly higher than those grown in Hungary (57.30% dw).

Wide range of crude fat content were also found in the studied lines (1.47-3.67%) where Mv-Suba and Mv-Karizma exhibited the highest fat contents with 3.01 and 3.67% values, respectively.

Processing properties: Properties related to the processing quality of wheat, such as the thousand kernel weight TKW, flour yield, gluten content, Zeleny sedimentation and titratable acidity were measured in the four studied lines (Table 3).

Table 4: Total Phenolic Content (TPC), Total Flavonoid Content (TFC) and lycopene content of Egyptian and Hungarian wheat varieties (mg/100 g)

Wheat samples	TPC gallic acid equation (mg g^{-1} dw*)	TFC rutin equation (µg g^{-1} dw)	Lycopene (µg/100 g dw)
Ben-Su1	1.72±0.00^{b}	8.42±0.50^{a}	0.13±0.01^{a}
Ben-Su3	2.12±0.10^{a}	3.52±0.50^{b}	0.12±0.00^{ab}
Mv-Suba	1.69±0.00^{b}	2.25±0.10^{b}	0.11±0.01^{bc}
Mv-Karizma	1.65±0.00^{b}	2.18±0.10^{b}	0.10±0.07^{c}

*dw: Dry weight, values are averages of three repetitions, values within columns followed by different upper case letters are significantly different ($p<0.05$)

Based on the results of the analyses of variances, the mean TKW value of Ben-Su1 (56 g) was significantly higher than that of the Hungarian varieties while Ben-Su3 (39 g) had significantly lower TKW value compared to Mv-Suba (46 g) and Mv-Karizma (42 g).

Flour yield was around 50% in the Hungarian wheat varieties (49.3 and 57.1% in Mv-Suba and Mv-Karizma, respectively) but the yield was half of this from the Egyptian varieties (31.7 in Ben-Su1 and 23.9 in Ben-Su3).

The wet gluten content of the Hungarian wheat varieties were rather similar although evaluated as significantly different values ($p<0.05$) (Mv-Karizma (38.2%), Mv-Suba (36.4)). At the same time Ben-Su3 have 33.0% gluten content while the lowest value was found in Ben-Su1 (23.6%).

Zeleny sedimentation of the tested samples varied significantly ($p<0.05$), where the highest values were observed for varieties Mv-Suba and Mv-Karizma (61 and 57 mL), while the lowest Zeleny value was reported for variety Ben-Su1 (16.6 mL) in relation to its low wet gluten content. Although Ben-Su3 has high wet gluten content but its bread making quality should be low as the Zeleny sedimentation was found to be also low (21.7%).

No significant difference in pH values while the titrable acidity values ranged from 0.01 to 0.06%. These results are in accordance with several studies where the pH and titrable acidity showed similar values (Finney *et al*., 1987; Mahmood, 2004).

These results show that Mv-Suba and Mv-Karizma belongs to the same quality group from processing point of view. They have high protein and gluten content and Zeleny sedimentation values, which means that their seed is excellent for bread making purposes. Ben-Su1 and Ben-Su3 represents different quality groups. They differ not only from the Hungarian varieties but also differ from each other. Ben-Su1 have very low gluten content and Zeleny sedimentation but have very big kernels, so from processing point of view it is similar to the Hungarian biscuit making quality category. Ben-Su3, have similar Zeleny sedimentation but it has small kernel and relatively higher gluten content at the same time. This variety is appropriate for typical flat balady bread production in Egypt.

Phytochemical composition: Total phenol content, total flavonoid content, total tocol content and lycopenes are among the antioxidant compounds that were evaluated, while DPPH radical scavenging activity measurement were used for *in-vitro* assessment of the total antioxidant activity.

The Folin-Ciocalteu (FC) method was used to determine Total Phenolic Contents (TPC) of the tested wheat samples (Table 4). Total phenolic content expressed as mg GAE/g sample dw varied significantly ($p<0.05$) and ranged from 1.65 (Mv-Karizma) to 2.21 (Ben-Su3). Egyptian variety,

Table 5: Tocol content (μg g^{-1} dw*) from wheat of different wheat varieties

Samples	α-tocopherol AT	α-tocotrienol ATT	β-tocopherol BT	β-tocotrienol BTT	Total tocol content
Ben-Su1	6.61±0.41^{b}	4.00±0.10^{b}	3.86±0.10^{b}	23.13±0.31^{b}	37.65
Ben-Su3	6.68±0.30^{b}	6.32±0.31^{a}	3.99±0.21^{b}	36.77±1.70^{a}	53.66
Mv-Suba	9.42±0.12^{a}	3.26±0.17^{c}	5.38±0.10^{a}	23.89±0.51^{b}	41.95
Mv-Karizma	9.93±0.30^{a}	3.16±0.10^{c}	5.54±0.11^{a}	23.14±0.21^{b}	41.77

*dw: Dry weight, values are averages of three repetitions, values within columns followed by different upper case letters are significantly different ($p<0.05$)

Ben-Su3 has significantly higher phenolic content than the other three varieties. Results are in accordance with several studies where, TPC values for wheat grains have ranged from 0.23 to 9.28 mg Gallic Acid Equivalents (GAE) per gram (Zhou *et al*., 2004; Moore *et al*., 2005; Yu *et al*., 2002). In addition, it was stated previously that phenolic compounds have significant antioxidant activity which was verified by the DPPH scavenging capacity values (Adom *et al*., 2003).

Total flavonoid content, expressed as micrograms of Rutin Equivalent (RE) per gram of sample, were determined for the tested samples (Table 4). The flavonoid content ranged from 2.18 μg rutin equivalent/g sample (Mv-Karizma) to 8.42 μg rutin equivalent/g (Ben-Su1). Ben-Su1 had significantly higher flavonoid content than all the other three varieties, although Ben-Su3 also had a bit higher flavonoid content than the Hungarian varieties. We need to note here however, that the quantity of flavonoids and the antioxidant activity varies much depending on the cultivar and the location effects (Oomah and Mazza, 1996). Lycopene contents were significantly different ($p>0.05$) among tested cultivars and ranged from 0.10 (Mv-Karizma) to 0.13 (Ben-Su1) μg/100 g dw (Table 3). Lycopene content reported in this study is in accordance with previously reported values (Mashaba and Barros, 2011).

Total tocols content ranged from 37.65 to 53.66 μg g^{-1} dw. In all samples, β-tocotrienol was the predominant form of vitamin E accounting for roughly half of the total tocol content and ranged from 23.13 to 36.77 μg g^{-1} dw (Table 5). The content of α-tocopherol, α-tocotrienols and β-tocopherol ranged from 6.61 to 9.42; from 3.16 to 6.32; from 3.86 to 5.54, respectively. Figure 1 shows the typical chromatogram of the contents of individual tocols in the four tested wheat samples. Several reports suggested that the vitamin E activity depends on its chemical structure and physiological factors. Isomers of tocols exhibit vitamin E activity as follows: α-TP>β-TP>α-TT>β-TT>δ-TP or no activity for γ-TT and δ-TT (Panfili *et al*., 2008; Sheppard *et al*., 1993). The α-TP has the greatest vitamin E activity but α-TT also possesses excellent antioxidant activity (Cahoon *et al*., 2003) and contributes to the nutritive value of cereal grains in the human diet. The vitamin E contents reported in the present study is consistent with previously reported values (Hidalgo *et al*., 2006; Lampi *et al*., 2008).

Total DPPH radical scavenging activity DPPH-RSA was used to evaluate the antioxidant activity of the four different wheat varieties (Fig. 2). DPPH-RSA was calculated as percentage discoloration, with higher percentages indicating higher antioxidant activity. Based on the results Ben-Su1 has outstanding, significantly higher antioxidant activity level, than the other varieties (29.9±0.8%) but even Ben-Su3 has significantly higher activity than the Hungarian varieties (19.94±0.9%).

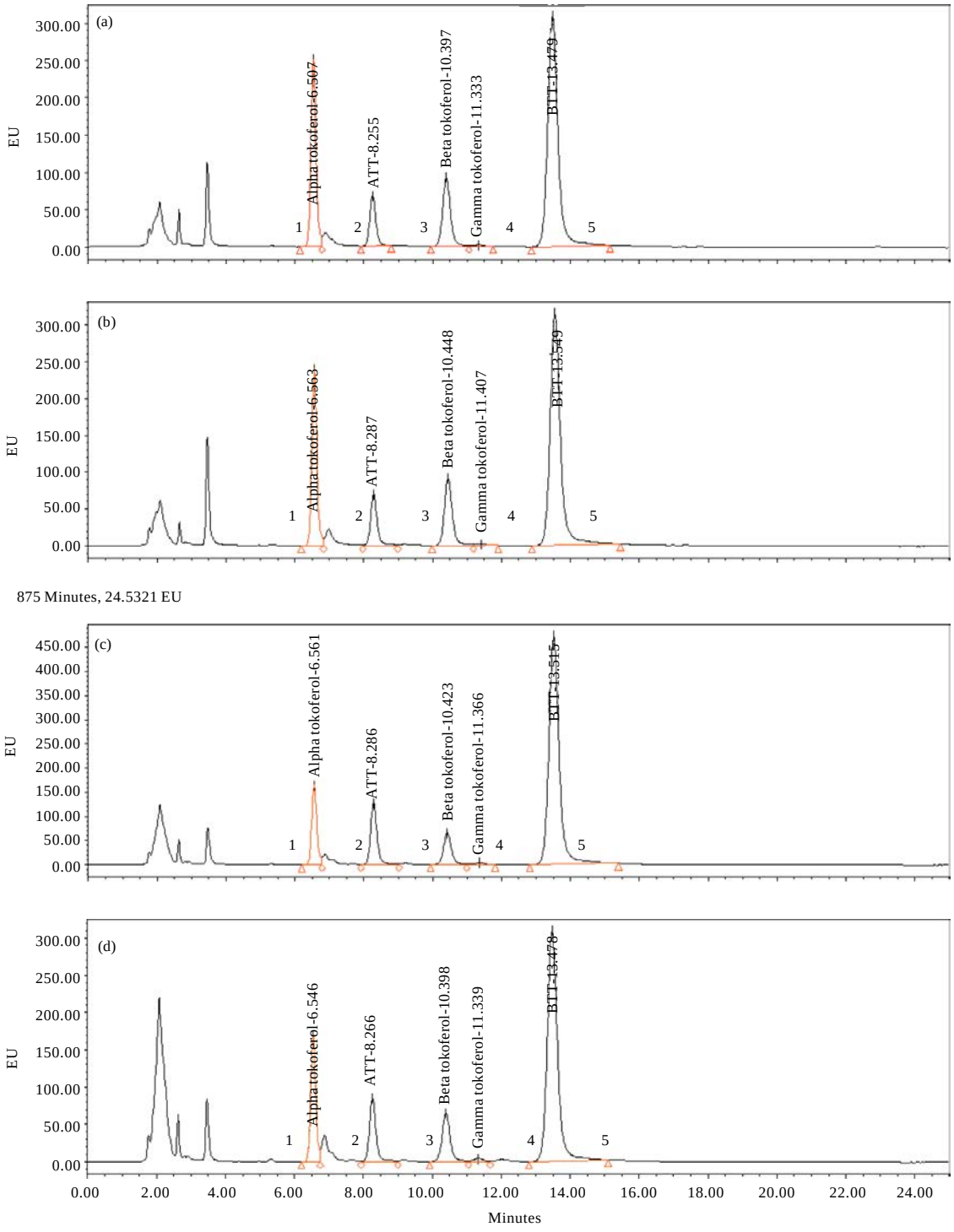

Fig. 1(a-d): Typical chromatogram of tocochromanols in the four wheat samples, Peaks: 1.α-tocopherol, 2.α-tocotrienol, 3.β-tocopherol, 4.γ-tocopherol and 5.β-tocotrienol, (a) Mv-Karizma, (b) Mv-Suba, (c) Ben-Su1 and (d) Ben-Su3

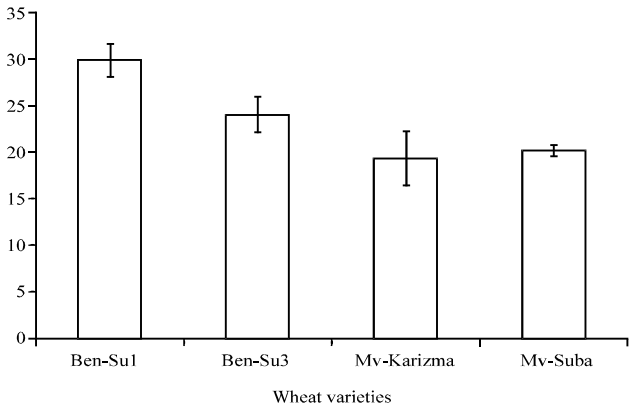

Fig. 2: DPPH radical scavenging activity of the tested wheat samples

CONCLUSION

Four wheat varieties, including Ben-Su1, Ben-Su3, Mv-Suba and Mv-Karizma, were studied for compositional properties, processing quality and phytochemical profile. The Hungarian varieties had excellent breadmaking qualities while the Egyptian cultivars had much lower protein and gluten content and Zeleny sedimentation. At the same time the health related benefits of the Egyptian varieties was reported and high phytochemicals as well as antioxidants were found in these lines.

The wheat cultivars examined showed significant differences in their contents of TPC, TFC, tocols, as well as in their free radical scavenging capacities. Ben-Su1 had significantly higher total flavonoid content and its DPPH radical scavenging activity was also significantly higher compared to the other remaining three varieties. Ben-Su3 was also outstanding with respect to its antioxidant composition. In this variety the total phenolic content, total tocols and the ratio of the α-tocotienol and β-tocotrienol were significantly high. Ben-Su3 has also higher total flavonoid content and antioxidant activity compared with the Hungarian varieties.

Although, significant differences were found in the bioactive component levels of the Hungarian and Egyptian varieties, the environmental conditions need to be addressed. Several authors have concluded that both the genotype and the environment in which wheat is grown, manifest significant differences in the total phenolic content, antioxidant activity and tocols composition of wheat. Yu *et al.* (2004) reported significant effects of growing conditions, including the number of hours exceeding 32°C, on the antioxidant properties of hard red winter wheat variety. Moreover Beta *et al.* (2005) stated that growing location had a strong influence on the antioxidant activity of pearled wheat fractions of a leading Canadian bread wheat cultivar. The potential effects of environmental factors on the chelating activities of wheat flour were also reported by Yu *et al.* (2003, 2004). They proved the correlations between the chelating activity and the total solar radiation, the daily average solar radiation, or the total hours exceeding 32°C. Wang and Zheng (2001) also suggested that the irrigation may alter the chelating property of white hard winter wheat. Egypt generally has much warmer weather conditions, with much higher solar radiation and less precipitation than Hungary. These conditions significantly influence the size and compositions of the kernel, so the quantity of the bioactive components of wheat varied accordingly.

In the Healthgrain EU program, high genotypic variance values were shown for total tocols and total sterols. Thus, the ratio of genotypic variance to total variance was high (0.77 and 0.57 for

tocols and sterols, respectively). The high heritabilities of the contents of dietary fiber components, tocols and sterols indicate that they are potentail targets for selection in plant breeding. In contrast, the lower ratios of genetic to total variance for folates (0.24) and total phenolic acids (0.05) indicate that stable increases in content will not readily be achieved by breeding (Shewry *et al*., 2010).

Taking into consideration these previous results, it could be concluded that the outstanding flavonoid content and DPPH activity of Ben-Su1 and the outstanding total tocol content of Ben-Su3 should be genetically determined and that was not the environment which imparted such variation in wheat kernel composition. These results highlight the challenges facing breeders to select appropriate wheat varieties with optimizing agronomic quality as well as nutrition and health benefit. The Egyptian Ben-Su1 could be a good genetic resource to breed for high flavonoid content, although its agronomic quality needs to be elucidated. Moreover, Ben-Su3 might be a good genetic resource to increase the tocol content in wheat breeding programs.

ACKNOWLEDGEMENTS

We gratefully acknowledge research grant No. 58-3148-7-023 from the U.S.-Egypt Science and Technology joint fund. The authors would also like to say thank to the Agricultural Research Centre for providing grain samples. CAR-HAS also acknowledged the support of the National Research Funds HT cereal (TECH-08-A3/2-2008-0425) and OTKA 80292 CK.

REFERENCES

AACC, 2000. Approved Methods of the American Association of Cereal Chemists. 10th Edn., American Association of Cereal Chemists Press, St. Paul, MN., USA.

Adom, K.K. and R.H. Liu, 2002. Antioxidant activity of grains. J. Agric. Food Chem., 50: 6182-6187.

Adom, K.K., M.E. Sorrells and R.H. Liu, 2003. Phytochemical profiles and antioxidant activity of wheat varieties. J. Agric. Food Chem., 51: 7825-7834.

Ahmed, Z.S., S.F. Hamed and S.S. Abozed, 2013. Phytochemical content and antioxidant activity of different milling extractions of diverse varieties of Egyptian wheat. Int. J. Agric. Food Sci. Technol., 4: 11-23.

Beta, T., S. Nam, J.E. Dexter and H.D. Sapirsteini, 2005. Phenolic content and antioxidant activity of pearled wheat and roller-milled fractions. Cereal Chem., 82: 390-393.

Cahoon, E.B., S.E. Hall, K.G. Ripp, T.S. Ganzke, W.D. Hitz and S.J. Coughlan, 2003. Metabolic redesign of vitamin E biosynthesis in plants for tocotrienol production and increased antioxidant content. Nat. Biotechnol., 21: 1082-1087.

Chang, C.C., M.H. Yang, H.M. Wen and J.C. Chern, 2002. Estimation of total flavonoid content in propolis by two complementary colorimetric methods. J. Food Drug Anal., 10: 178-182.

Finney, K.F., W.T. Yamazaki, V.L. Youngs and G.L. Rubenthaler, 1987. Quality of Hard, Soft and Durum Wheats. In: Wheat and Wheat Improvement, Heyne, E.G. (Ed.). ASA, CSSA, SSSA Publishers, Madison, WI., USA., pp: 677-748.

Halliwell, B. and J.M. Gutteridge, 1990. The antioxidants of human extracellular fluids. Arch. Biochem. Biophys., 280: 1-8.

Hidalgo, A., A. Brandolini, C. Pompei and R. Piscozzi, 2006. Carotenoids and tocols of einkorn wheat (*Triticum monococcum* ssp. *monococcum* L.). J. Cereal Sci., 44: 182-193.

Huang, D., B. Ou and R.L. Prior, 2005. The chemistry behind antioxidant capacity assays. J. Agric. Food Chem., 53: 1841-1856.

Jones, J.M., M. Reicks, J. Adams, G. Fulcher and L. Marquart, 2004. Becoming proactive with the whole-grains message. Nutr. Today, 39: 10-17.

Lampi, A.M., T. Nurmi, V. Ollilainen and V. Piironen, 2008. Tocopherols and tocotrienols in wheat genotypes in the HEALTHGRAIN diversity screen. J. Agric. Food Chem., 56: 9716-9721.

Liu, R.H., 2007. Whole grain phytochemicals and health. J. Cereal Sci., 46: 207-219.

Liu, H., N. Qiu, H. Ding and R. Yao, 2008. Polyphenols contents and antioxidant capacity of 68 Chinese herbals suitable for medical or food uses. Food. Res. Int., 41: 363-370.

Liyana-Pathirana, C.M. and F. Shahidi, 2007. Antioxidant and free radical scavenging activities of whole wheat and milling fractions. Food Chem., 101: 1151-1157.

Mahmood, A., 2004. Acid-PAGE gliadin composition and cluster analysis for quality traits for different wheat varieties. Ph.D. Thesis, University of Agriculture, Faisalabad, Pakistan.

Mashaba, C.S. and E. Barros, 2011. Screening South African potato, tomato and wheat cultivars for five carotenoids. South Afr. J. Sci., 107: 34-39.

Moore, J., Z. Hao, K. Zhou, M. Luther, J. Costa and L. Yu, 2005. Carotenoid, tocopherol, phenolic acid and antioxidant properties of Maryland-grown soft wheat. J. Agric. Food Chem., 53: 6649-6657.

Nagata, M. and I. Yamashita, 1992. Simple method for simultaneous determination of chlorophyll and carotenoids in tomato fruit. J. Jap. Soc. Food Sci. Technol., 39: 925-928.

Nurmi, T., L. Nystrom, M. Edelmann, A.M. Lampi and V. Piironen, 2008. Phytosterols in wheat genotypes in the HEALTHGRAIN diversity screen. J. Agric. Food Chem., 56: 9710-9715.

Oomah, B.D. and G. Mazza, 1996. Flavonoids and antioxidative activities in buckwheat. J. Agric. Food Chem., 44: 1746-1750.

Panfili, G., A. Fratianni, T. Di Criscio and E. Marconi, 2008. Tocol and β-glucan levels in barley varieties and in pearling by-products. Food Chem., 107: 84-91.

Pomeranz, Y., 1988. Wheat Chemistry and Technology. Vol. 2, American Association of Cereal Chemists Inc., St. Paul, MN., USA.

Sheppard, A.J., J.A.T. Pennington and J.L. Weihrauch, 1993. Analysis and Distribution of Vitamin E in Vegetable Oils and Foods. In: Vitamin E in Health and Disease, Packer, L. and J. Fuchs (Eds.). Marcel-Dekker Inc., New York, USA., pp: 9-31.

Shewry, P.R., V. Piironen, A.M. Lampi, M. Edelmann and S. Kariluoto *et al.*, 2010. The HEALTHGRAIN wheat diversity screen: Effects of genotype and environment on phytochemicals and dietary fiber components. J. Agric. Food Chem., 58: 9291-9298.

Slaughter, D.C., K.H. Norris and W.R. Hruschka, 1992. Quality and classification of hard red wheat. Cereal Chem., 69: 428-432.

Slavin, J., 2003. Why whole grains are protective: Biological mechanisms. Proc. Nutr. Soc., 62: 129-134.

Wang, S.Y. and W. Zheng, 2001. Effect of plant growth temperature on antioxidant capacity in strawberry. J. Agric. Food Chem., 49: 4977-4982.

Ward, J.L., K. Poutanen, K. Gebruers, V. Piironen and A.M. Lampi *et al.*, 2008. The HEALTHGRAIN cereal diversity screen: Concept, results and prospects. J. Agric. Food Chem., 56: 9699-9709.

Williams, P.C., F.J. El-Haramein, H. Nakkoul and S. Rihawi, 1986. Crop quality evaluation methods and guidelines. Technical Manual No. 14, International Center for Agriculture Research in the Dry Areas, Alspoo, Syria.

Woisky, R.G. and A. Salatino, 1998. Analysis of propolis: Some parameters and procedures for chemical quality control. J. Apicult. Res., 37: 99-105.

Yu, L., S. Haley, J. Perret, M. Harris, J. Wilson and M. Qian, 2002. Free radical scavenging properties of wheat extracts. J. Agric. Food Chem., 50: 1619-1624.

Yu, L., J. Perret, M. Harris, J. Wilson and S. Haley, 2003. Antioxidant properties of bran extracts from Akron wheat grown at different locations. J. Agric. Food Chem., 51: 1566-1570.

Yu, L., S. Haley, J. Perret and M. Harris, 2004. Comparison of wheat flours grown at different locations for their antioxidant properties. Food Chem., 86: 11-16.

Zhou, K., J.J. Laux and L. Yu, 2004. Comparison of swiss red wheat grain and fractions for their antioxidant properties. J. Agric. Food Chem., 52: 1118-1123.

Zielinski, H. and H. Kozlowska, 2000. Antioxidant activity and total phenolics in selected cereal grains and their different morphological fractions. J. Agric. Food Chem., 48: 2008-2016.