ANTIOXIDANT ACTIVITY OF MILLING FRACTIONS OF SELECTED CEREALS

EVA IVANIŠOVÁ[1], MIROSLAV ONDREJOVIČ[2], STANISLAV ŠILHÁR[2]

[1]*Department of Storing and Processing Plant Products, Slovak University of Agriculture, Tr. A. Hlinku 2, Nitra, SK-949 76, Slovak Republic (eva.ivanisova@post.sk)*
[2]*Department of Biotechnology, University of SS. Cyril and Methodius, J. Herdu 2, Trnava, SK-917 01, Slovak Republic (miroslav.ondrejovic@ucm.sk)*

Abstract: The aim of this study was to evaluate antioxidant potential of four milling fractions of selected cereals grew in the year 2009 and 2010. Free radical scavenging activity of samples was measured using DPPH assay and reducing power was determined using FRAP assay. Secondary, total phenolic and flavonoid content of cereal extracts was evaluated. We found that flour fractions (break flour and reduction flour) showed the lower proportion of the total antioxidant potential than bran fractions (fine bran and coarse bran), which was balanced in observed years. Extract from barley had the highest values of antioxidant activity and polyphenol content.

Keywords: cereals, milling fractions, polyphenols, antioxidant activity

1. Introduction

In many countries, cereals are main ingredient for human foods or animal feeds (CASTRO-RUBIO *et al.*, 2006). Epidemiological studies indicate that the consumption of whole-grain and whole-grain products is related to reduction in total mortality, coronary heart disease mortality, diabetes and cancer incidence (SERPEN *et al.,* 2008). These beneficial effects are attributed to the bioactive factors in cereal grain such as non digestible carbohydrates and phytochemicals. The important part of phytochemicals with low molecular weight present in cereal grain is group of antioxidants such as tocopherols, lignans, flavonoids and phenolic acids. Antioxidants are defined as molecules that, at low concentration and specific assay conditions, can delay or prevent oxidation of an oxidizable substrate (VAHER *et al.,* 2010). Higher concentrations of these compounds are found in the outer layers of the kernel which constitute the bran (KIM *et al*., 2006).

Cereal grains are rich in phenolic acids and saponins, while phytoestrogens and flavonoid are presented in small quantities (DORDEVIĆ *et al.,* 2010). Studies have shown that dietary phenolics have high antioxidant activity, which may contribute to their health benefits. In cereals, the predominant phenolic acid is ferulic acid, representing up to 90 % of total polyphenols. Other phenolic acids including vanilic, syringic, chlorogenic, *p*-coumaric, *m*-coumaric and *OH*-cinnamic acid have also been reported in cereals (HOSSEINIAN and MAZZA, 2009). Total amount of polyphenols in cereals is highly variable both in whole grain and in bran and also depends on the cereal variety and milling procedure (ADOM *et al*., 2005).

DOI 10.2478/v10296-012-0005-0

The main objective of the present work was to evaluate antioxidant potential of selected cereals by Free Radical Scavenging Activity (DPPH) and The Ferric Ion Reducing Antioxidant Power (FRAP), and its distribution into the dry milling fractions. In addition, the content of flavonoids and phenolics was also determined, in order to refer unutilized potential of naturally occurring antioxidants in cereals, which leaving in the form of bran during the production of flour.

2. Materials and methods

2.1 Plant material

Cereals such as wheat (Torysa, PPRI), oat (Cacko, DEPOF), spelt wheat (Roquir, PPRI), triticale (Kandar, PPRI), rye (Dankovské nové, PPRI), barley (Ezer, DEPOF) were grown in the years 2009 and 2010 on a field nursery at Department of Environmental Protection and Organic Farming (DEPOF) Spišská Belá (SK) and on a field at Plant Production Research Institute (PPRI) Piešťany (SK).

2.2 Sample preparation

The samples were milled by laboratory mill (Brabender Quadrumat Senior) gaining four milling products: break flour (MF I.), reduction flour (MF II.), fine bran (MF III.), and coarse bran (MF IV.). The milling fractions were extracted with methanol in ratio 1:80 (w/v) for 20 hours at laboratory temperature. After centrifugation at 3000 g (Himac CT 6E, Hitachi Ltd., Japan,) for 20 min, the supernatant was evaporated at 40 °C. Residue was solubilised in methanol to a final volume 2 mL.

2.3 Free Radical Scavenging Activity

Free radical scavenging activity of samples was measured using the 2,2-difenyl-1-picrylhydrazyl (DPPH) according to the procedures described by YEN and CHEN (1995). The extracts (25 µL) were mixed with 100 µL of DPPH solution (0.012 g DPPH in 100 mL methanol) and incubated for 10 min at laboratory temperature. After them, reaction mixture absorbance was determined at 550 nm using BioTek Microplate Reader (ELx800). Free radical scavenging activity of the samples was expressed as mg of Trolox equivalent antioxidant capacity per g of dry matter (mg TEAC/g DM).

2.4 Reducing power

Reducing power of samples was determined according to the procedure by OYANIZU (1986). The mixture of cereal extract (20 µL), phosphate buffered saline (50 µL, pH 6.6) and 1 % potassium ferricyanide (50 µL) was incubated at 50 °C for 20 min, then rapidly cooled, mixed with 50 µL of 10 % trichloacetic acid, and centrifugated at 11 000 g (Eppendorf MiniSpin) for 10 min. 50 µL of the supernatant was mixed with 50 µl of distilled water and 10 µL of 0.1 % ferric chloride. The absorbance at 700 nm using BioTek Microplate Reader (ELx800) was detected.

Reducing power was expressed as mg of Trolox equivalent antioxidant capacity per g of dry matter (mg TEAC/ g DM).

2.5 Total phenolic content

Total phenolic content of cereal extracts was measured photometrically, using the modified Folin-Ciocalteu method as described by SINGLETON *et al.,* (1965). Each cereal extract (0.1 mL) was mixed with 0.1 mL of the Folin-Ciocalteau reagent and 1 mL of 20 % sodium carbonate, and centrifuged at 11 000 g (Eppendorf MiniSpin) for 10 min. Supernatant (240 μL) was used for measuring the absorbance at 700 nm using BioTek Microplate Reader (ELx800). The total phenolics content was expressed as mg of gallic acid equivalent (GAE) per g dry matter (DM).

2.6 Total flavonoid content

Total flavonoid was determined using the modified method by QUETTIER-DELEU *et al.*, (2000). Cereal extract (0.1 mL) was mixed with 20 μL of 5 % methanolic solution of aluminum chloride and centrifuged at 11 000 g (Eppendorf MiniSpin) for 10 min. Supernatant (120 μL) was used for measured the absorbance at 405 nm on a BioTek Microplate Reader (ELx800). The total flavonoid content was expressed as mg of quercetin equivalent (QE) per g dry matter (DM).

3. Results and discussion

3.1 Yield of Milling Fraction

The wheat and rye are main commodities of milling industry for production various type of flours. The oat, barley, triticale and spelt are used mainly as additives and for production of flour (MUCHOVÁ *et al.,* 2011). The main component of cereal grain is endosperm, which constitutes about 83 % of grain, and is source of white flour during the milling. Bran represents 14.5 % of grain, and they are part of whole meal flour during the milling, but nowadays bran are often removed from flour, and used for animal feed. Germ constitutes about 2.5 % of grain and from flour is often removed (MUCHOVÁ *et al.,* 2011). The purpose of milling is to separate bran and germ from endosperm so as to extract as much flour as possible, at minimum operating cost, while maintaining high and consistent flour quality (CAMPBELL *et al.,* 2007).

The moisture of selected cereals was in range 12.29 – 13.42 % and these values corresponding with limit values for cereals (max. 14 %). The samples of wheat, spelt, rye and triticale showed higher yields of flour fractions than bran fractions, and sample of oat and barley showed higher yield of bran fractions, what is caused mainly by different structure of grains. A comparison of results from different studies can be difficult due to variability in milling conditions.

3.2 Free Radical Scavenging Activity

DPPH$^{\bullet}$ is a stable free radical and accepts an electron or hydrogen radical to become a stable diamagnetic molecule. The reductions capability of DPPH$^{\bullet}$ is

determined by the decrease in its absorbance induced by antioxidant (LIU & YAO, 2007). The scavenging effect of cereal extracts in flour milling fractions on DPPH radical decreased in the year 2009 in the order: barley (26%) > spelt (19%) > oat (16%) > triticale (14%) > rye (13%) > wheat (12%), and in the year 2010 in the order: barley (29%) > spelt (22%) > oat (15%) > rye (13%) > triticale (12%) > wheat (9%). The scavenging effect of cereal extracts in bran milling fractions on DPPH radical decreased in the year 2009 in the order: barley (22%) > triticale (17%) > rye (17%) > wheat (16%) > oat (14%) > spelt (14%), and in the year 2010 in the order: barley (20%) > triticale (18%) > wheat (17%) > oat (15%) > rye (15%) > spelt (14%) (Fig. 1 and 2). These results indicated that all extracts had a noticeable effect on scavenging of free radicals. The higher activities of all extracts were measured in bran (MF III. and MF IV.). Bran is a composite material made of several layers, such as pericarp, testa and aleurone (HEMERY *et al.,* 2011). The main components of the fine bran (MF III.) are aleurone layer and the germ. Pericarp is dominant in coarse bran (MF IV.) (SCHNÜRER, 1991). It is known that cereals bran are a rich source of fatty acids and several substances, such as tocopherol, vitamins, and phenolic compounds, possessing antioxidant properties (PRISENŽŇÁKOVÁ *et al.,* 2010).

The extract from barley had the strongest scavenging activity in flour milling fractions. From the literature it is known, that the barley is an excellent source of natural antioxidant either for food preservation (to inhibit lipid oxidation), or for disease prevention (FARDET *et al.,* 2008). LIU and YAO (2007) determined scavenging activity of barley seed extracts and found strong activity, which was dominant in 70 % acetone extracts and shown similar activity to BHT at the amount of 200 µg. In bran fractions, extracts from rye and wheat showed the strongest activity. High activity was also determined in flour extracts of spelt and in bran extract of triticale. In a recent study, HOSSEINIAN and MAZZA (2009) described triticale bran as a potential new source of antioxidant compounds.

Data variation in the scavenging activity of cereals in the year 2009 and 2010 is to be expected, as many factors such as genetics, agrotechnical processes and environmental conditions can influence the presence of antioxidant compounds. In addition, a comparison of results from different studies can be difficult due to variability in the experimental conditions amongst the method used (ALVAREZ-JUBETE *et al.,* 2010).

3.3 Reducing Power

For measurement of the reductive ability, the Fe^{3+} - Fe^{2+} transformation in the presence of cereal extracts (year 2009 and 2010) was investigated. Reductive capabilities of cereal extracts are shown Fig. 3 and 4. Increase in absorbance of the reaction mixture indicated the reducing power of the samples. Reducing power of flour milling fractions of cereal extracts in the year 2009 exhibited the following order: barley (33%) > spelt (21%) > oat (15%) > wheat (11%) > rye (10%) > triticale (9%), and in the year 2010 exhibited the following order: barley (50%) > spelt (16%) > oat (13%) > rye (10%) > wheat (7%) > triticale (3%). Reducing power of bran milling fractions of cereal extracts in the year 2009 exhibited the following order: barley

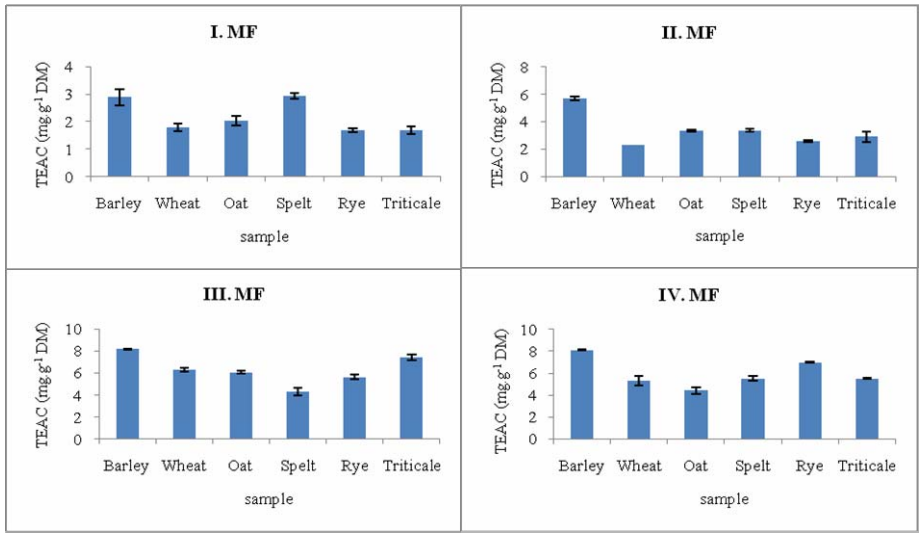

Fig. 1. Free radical scavenging activity of cereal extracts (in the year 2009) expressed as mg of Trolox equivalent antioxidant capacity per g of dry matter (mg TEAC/ g DM), (I. MF – break flour, II. MF – reduction flour, III. MF – fine bran, IV. MF – coarse bran).

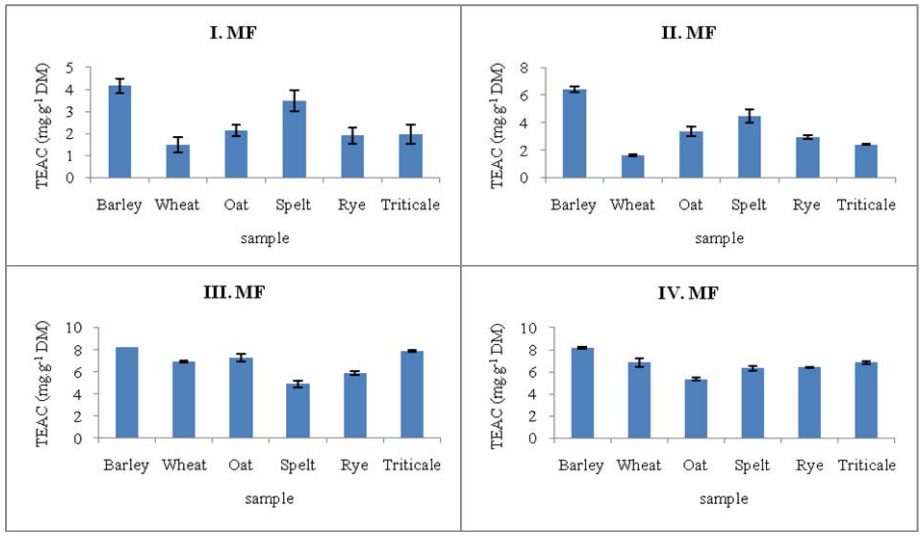

Fig. 2. Free radical scavenging activity of cereal extracts (in the year 2010) expressed as mg of Trolox equivalent antioxidant capacity per g of dry matter (mg TEAC/ g DM), (I. MF – break flour, II. MF – reduction flour, III. MF – fine bran, IV. MF – coarse bran).

(38%) > rye (15%) > triticale (14%) > oat (14%) > wheat (13%) > spelt (6%), and in the year 2010 exhibited the following order: barley (32%) > triticale (16%) > wheat (16%) > oat (14%) > rye (13%) > spelt (9%). The higher activities of all extracts were measured in bran (MF III. and MF IV.).

The reducing capacity of a compound may serve as significant indicator of its potential antioxidant activity (LIU and YAO, 2007). The reducing properties are generally associated with the presence of reductones (PIN-DER, 1998). It is reported that the antioxidant action of reductones is based on the breaking of the free radical

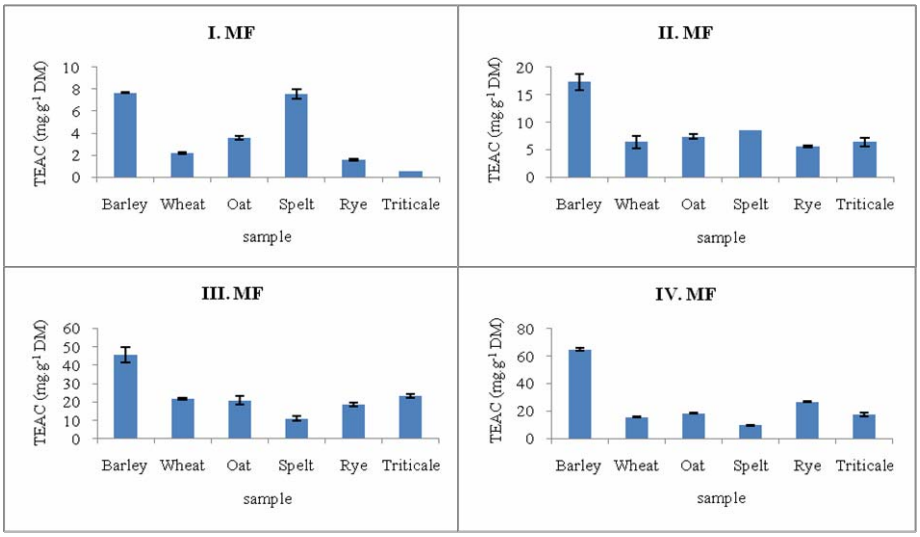

Fig. 3. Reducing power of cereal extracts (in the year 2009) expressed mg of Trolox equivalent antioxidant capacity per g of dry matter (mg TEAC/ g DM), (I. MF – break flour, II. MF – reduction flour, III. MF – fine bran, IV. MF – coarse bran).

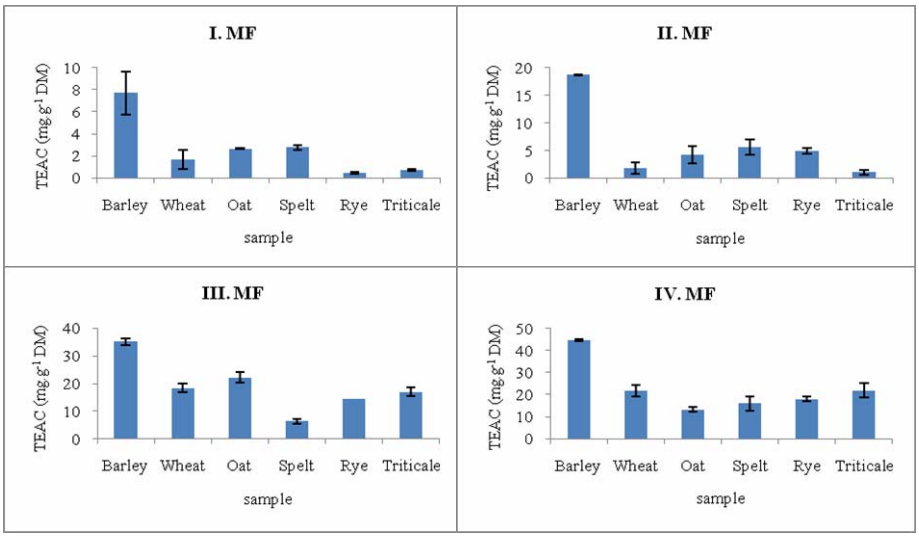

Fig. 4. Reducing power of cereal extracts (in the year 2010) expressed as mg of Trolox equivalent antioxidant capacity per g of dry matter (mg TEAC/ g DM), (I. MF – break flour, II. MF – reduction flour, III. MF – fine bran, IV. MF – coarse bran).

chain by donating a hydrogen atom, or reacting with certain precursors of peroxide to prevent peroxide formation. It is presented that the phenolic compounds in cereals may act in a similar fashion as reductones by donating electrons and reacting with free radicals to convert them to more stable products and terminating the free radical chain reaction (LIU and YAO, 2007). The data presented here indicates that the marked reducing power of cereal extracts seems to be the result of their antioxidant activity.

The extract from barley had the strongest reducing power in both years in all fractions. LIU and YAO (2007) determined reducing power of different solvent extracts of barley and found that 70 % methanol extract exhibit the highest activity. ZHAO *et al.* (2008) measured reducing power of malting barley extracts. They confirmed strong antioxidant activity of barley. The high activities were also determined in flour fractions of spelt, and in bran fractions of triticale and rye. ZIELIŃSKI *et al,* (2007) reported that rye is an excellent raw material for healthy and tasty foods. The ray grain contains a large variety of substances, especially those that are biologically active and demonstrate antioxidant properties, which include free radical-scavengers, reducing agents, and potential complexes of prooxidant metals and quenchers of the formation of singlet oxygen.

3.4 Cereal polyphenols

Phenolics are compounds with one or more aromatic ring and one or more hydroxyl groups. Structurally, phenolics in cereals can be subdivided into acids derived from either benzoic acid or cinnamic acid. Vanillic and salicylic acids are derivates of benzoic acid while ferulic acid, the dominant phenolic acid in cereals, and caffeic acid are derivates of cinnamic acid (ABDEL-AAL *et al.,* 2001). Phenolic acids are predominantly found in the outer bran layer.

Flavonoids are one group of phenolics, which consists of two aromatic rings linked by 3 carbons that usually in an oxygenated heterocycle ring. In cereals flavonoids are located mainly in the pericarp (DYKES and ROONEY, 2007) but their content is very low (PETERSON, 2001). KIM *et al*. (2006) reported that wheat is a good source of phenolic acid and flavonoids. KING (1962) isolated two related flavone glycosides from wheat germ. Three major flavones, apigenin, luteolin and tricin were identified in oat flour (PETERSON, 2001). In barley grains are dominant catechin and epicatechin (SHAHIDI and NACZK, 2004).

The majority of phenolics in cereals are insoluble and bound by ester and ether linkages with polysaccharides, such as arabinoxylan and lignin, in the cell wall (LIYANA-PATHIRANA and SHAHIDI, 2006), while a smaller portion is soluble (STALIKAS, 2007). The bran layer is highly stratified not only in phenolic composition, but also in the degree of ester and ether bonds and the compounds to which the phenolics are cross-linked (VERMA *et al.,* 2009). Several studies have shown that methanol is an effective solvent in extracting phenolics and other polar substances from cereals (RAGAEE *et al.,* 2006). In this study, methanol extracts from cereals were used for the determination of phenolic (mg GAE/g) and flavonoid content (mg QE/g DM).

3.4.1 Total Phenolic Content

The total phenolic content was determined by the Folin-Ciocalteau assay. The results (year 2009 and 2010) are presented in Tab. 1 and 2. It is evident that bran has higher content of total phenolic than flour. This is not surprising because it is well known that phenolic compounds are concentrated in the bran fractions of cereals that are removed during the milling of cereals into white flour (KIM *et al.,* 2006; VAHER *et al.,* 2010). ABDEL-AAL *et al.,* (2001) investigated the distribution of phenolic acids in wheat milling fractions. About 73 % of grain phenolic acids were found in the bran, but only 5 % in first and second milling fraction.

Table 1. The phenolic contents (mg GAE/g DM) of milling fractions from cereals grew in the year 2009.

Sample	Milling Fraction			
	I.	II.	III.	IV.
Barley	31.4 ± 1.2	62.1 ± 0.7	161.0 ± 4.1	291.7 ± 3.5
Wheat	17.6 ± 0.1	19.7 ± 0.5	159.1 ± 2.6	128.2 ± 0.3
Oat	31.3 ± 0.2	39.1 ± 0.7	99.2 ± 2.2	61.3 ± 2.7
Spelt	40.4 ± 0.7	36.4 ± 0.3	66.4 ± 1.0	151.0 ± 3.7
Rye	17.6 ± 0.3	30.3 ± 0.6	95.9 ± 1.6	159.5 ± 12.6
Triticale	11.8 ± 0.5	16.3 ± 1.0	178.0 ± 1.7	133.7 ± 3.4

I. – bread flour, II. – reduction flour, III. – fine bran, IV. – coarse bran

Table 2. The phenolic contents (mg GAE/g DM) of milling fractions from cereals grew in the year 2010.

Sample	Milling Fraction			
	I.	II.	III.	IV.
Barley	32.4 ± 1.1	53.7 ± 0.1	111.2 ± 1.4	232.9 ± 3.6
Wheat	9.2 ± 0.6	12.3 ± 0	130.7 ± 3.2	190.3 ± 3.4
Oat	29.3 ± 0.6	35.1 ± 2.3	121.8 ± 3.6	53.6 ± 1.7
Spelt	38.7 ± 1.3	47.6 ± 3.4	71.1 ± 0.7	168.7 ± 3.3
Rye	23.6 ± 0.4	30.9 ± 0.1	110.1 ± 3.6	153.9 ± 4.1
Triticale	10.7 ± 1.0	9.2 ± 0.5	122.5 ± 1.6	155.9 ± 1.6

I. – bread flour, II. – reduction flour, III. – fine bran, IV. – coarse bran

Total phenolic content of cereal extracts in flour milling fraction in the year 2009 decreased in the order: barley > (26 %) > spelt (22 %) > oat (20 %) > rye (14 %) > wheat (11 %) > triticale (7 %), and in the year 2010 in the order: barley > (26 %) > spelt (26 %) > oat (19 %) > rye (15 %) > wheat (8 %) > triticale (6 %). Total phenolic content of cereal extracts in bran milling fraction in the year 2009 decreased in this order in the order: barley (27 %) > triticale (18 %) > wheat (17 %) > rye (15 %) > spelt (13 %) > oat (10 %), and in the year 2010 in the order: barley > (21 %) > wheat (20 %)

> triticale (17 %) > rye (16 %) > spelt (15 %) > oat (11 %). The results showed that phenolics were found in all cereal extracts, but bran fractions content higher values of phenolics. The sample of barley and spelt in flour fractions showed higher amounts of phenolics than extract from oat and wheat. In bran fractions were the highest amounts of phenolics determined in sample of barley, wheat and triticale. A comparison of results from different studies can be difficult, because in our study cereals were milling into four milling fraction, while in other works, sample of cereals were milling into flour and bran.

3.4.2 Total Flavonoid Content

The total flavonoid content of cereal extracts is shown in Tab. 3 and 4. Higher concentration was found in bran (MF III. and MF IV.), but in smaller amounts than the total phenolic content. Bran flavonoids may be important for the miller because bran is introduced into flour during the milling process. Increasing amounts of bran will decrease the grade of the flour (FENG *et al.,* 1988).

In flour milling fractions of cereal extracts content of flavonoid in the year 2009 decreased in the order: barley (33%) > oat (28%) > spelt (14%) > rye (14%) > triticale (5%) > wheat (5%), and in the year 2010 in the order: oat (31%) > barley (25%) > spelt (16%) > rye (15%) > triticale (6%) > wheat (6%). In bran milling fractions of cereal extracts content of flavonoid in the year 2009 decreased in the order: rye (22%) > wheat (20%) > oat (19%) > barley (16%) > spelt (12%) > triticale (11%), and in the year 2010 in the order: wheat (20%) > rye (20%) > oat (19%) > barley (17%) > spelt (12%) > triticale (12%).

Table 3. The flavonoid contents (mg QE/ g DM) of milling fractions from cereals grew in the year 2009.

Sample	Milling Fraction			
	I.	II.	III.	IV.
Barley	0.75 ± 0.04	1.02 ± 0.03	1.08 ± 0.07	2.14 ± 0.07
Wheat	0.12 ± 0.01	0.16 ± 0.01	0.93 ± 0.15	3.07 ± 0.06
Oat	0.64 ± 0.04	0.84 ± 0.05	2.60 ± 0.02	1.23 ± 0.05
Spelt	0.33 ± 0.03	0.42 ± 0.04	0.81 ± 0.02	1.55 ± 0.04
Rye	0.24 ± 0.04	0.51 ± 0.02	2.39 ± 0.02	1.91 ± 0.03
Triticale	0.11 ± 0.02	0.18 ± 0.02	1.01 ± 0.02	1.24 ± 0.04

I. – bread flour, II. – reduction flour, III. – fine bran, IV. – coarse bran

From literature is known that flavonoids are concentrated mainly in pericarp. Our results shown that flavonoids are presented also in flour fractions; this is very important information, because products from endosperm are basic in human nutrition. In flour milling fractions, high amount of total flavonoid showed extract of barley and oat. Oat is a source of many compounds that exhibit antioxidant activity, but is consumed in considerably lower quantities worldwide than wheat (PETERSON, 2001). In bran milling fractions high amount of total flavonoid was determined in

extract of rye and wheat. These results indicate that the flavonoids of cereals were mostly concentrated in the outer layer of grains. ADOM and LIU (2002) found similar results for cereal grains including wheat and oat.

Table 4. The flavonoid contents (mg QE/ g DM) of milling fractions from cereals grew in the year 2010.

Sample	Milling Fraction			
	I.	II.	III.	IV.
Barley	0.62 ± 0.04	0.71 ± 0.02	1.24 ± 0.04	2.35 ± 0.05
Wheat	0.12 ± 0.02	0.17 ± 0.02	1.11 ± 0.02	3.20 ± 0.02
Oat	0.73 ± 0.04	0.89 ± 0.04	2.65 ± 0.05	1.30 ± 0.02
Spelt	0.34 ± 0.04	0.49 ± 0.01	0.92 ± 0.05	1.68 ± 0.02
Rye	0.29 ± 0.02	0.55 ± 0.04	2.32 ± 0.04	2.00 ± 0.01
Triticale	0.16 ± 0.04	0.24 ± 0.04	1.02 ± 0.06	1.56 ± 0.05

I. – bread flour, II. – reduction flour, III. – fine bran, IV. – coarse bran

4. Conclusion

In this article, we prepared and evaluated milling fractions from selected cereals. Antioxidant activities were determined by DPPH and FRAP assay and total phenolic and flavonoid content was also determined. We found that flour fractions (break flour and reduction flour) showed the lower proportion of the total antioxidant potential, which was balanced in observed years. Bran fractions (fine bran and coarse bran) showed higher antioxidant activity, but 30 – 80 % of these fractions are unused in food industry, they are used mainly as animal feed. Extract from barley showed the highest values in all methods in observed years. It is evident, that bran fractions can be evaluated in the future and used for fortification of flours.

References

ABDEL-AAL, E.S.M., HUCL, P., SOSULSKI, F. W., GRAF, R., GILLOTT, R., PIETRZAK, L.: Screening spring wheat for midge resistance in relation to ferulic acid content. J. Agric. Food Chem., 49, 2001, 3559-3566.

ADOM, K. K., LIU, R. H.: Antioxidant activity of grains. J. Agric. Food Chem., 50, 2002, 6182-6187.

ADOM, K.K., SORRELLS, M.E., RUI, H.L.: Phytochemicals and antioxidant activity of milled fractions different wheat varieties. J. Agric. Food Chem., 53, 2005, 2297-2306.

ALVARE-JUBETE, L., WIJNGAARD, H., ARENDT, E.K., GALLAGHER, E.: Polyphenol composition and *in vitro* anioxidant activity of amaranth, quinoa, buckwheat and wheat as affected by sprouting and baking. Food Chem., 119, 2010, 770-778.

CAMPBELL, G.M., FANGI, C., MUHAMAD, I.I.: On predicting roller milling performance VI: Effect of kernel hardness and shape on the particle size

distribution from first break milling of wheat. Food Bioprod. Process., 85, 2007, 7-23.

CASTRO-RUBIO, A., GARCIA, M.C., MARINA, M.L.: Rapid separation of soybean and cereal (wheat, corn, and rice) proteins in complex mixtures: Application to the selective determination of the soybean protein content in commercial cereal-based products. Anal. Chim. Acta, 558, 2006, 28-34.

DORDEVIĆ, T.M., ŠILER-MARINKOVIĆ, S.S., DIMITRIJEVIĆ-BRANKOVIĆ, S.I.: Effect of fermentation on antioxidant properties of some cereals and pseudo cereals. Food Chem., 119, 2010, 957-963.

DYKES, L., ROONEY, L.W.: Phenolic compounds in cereal grains and their health benefits. Cereal Food World, 52, 2007, 105-111.

FARDET, A., ROCK, E., RÉMÉSY, CH.: Is the *in vitro* antioxidant potential of whole-grain cereals and cereal products well reflected *in vivo*? J. Cereal Sci., 48, 2008, 258-276.

FENG, Y., MCDONALD, C.E., VICK, B.A.: *C*-Glycosylflavones from hard red spring wheat bran. Cereal Chem., 65, 1988, 452–456.

HEMERY, Y., HOLOPAINEN, U., LAMPI, A.M., LEHTINEN, P., NURMI, T., PIIRONEN, V., EDELMANN, M., ROVAN, X.: Potential of dry fractionation of wheat bran for the development of food ingredients part II: Electrostatic separation of particles. J. Cereal Sci., 53, 2011, 9-18.

HOSSEINIAN, F.S., MAZZA, G.: Triticale bran and straw: Potential new sources of phenolic acids, proanthocyanidins, and lignans. J. Funct. Foods,1, 2009, 57-64.

KIM, K.H., TSAO, R., YANG, R., CUI, S.W.: Phenolic acid profiles and antioxidant activities of wheat bran extracts and the effect of hydrolysis conditions. Food Chem., 95, 2006, 466-473.

KING, H. G.C.: Phenolic compounds of commercial wheat germ. J. Food Sci., 27, 1962, 446-454.

LI, D., XIAO, G., DING, X.: Study on antioxidant effect of tartary buckwheat flavonoids. J. Wuxi Un. Light Ind., 20, 2001, 44-47.

LIU, Q., YAO, H.: Antioxidant activities of barley seeds extracts. Food Chem., 102, 2007, 732-737.

LIYANA-PATHIRANA, C.M., SHAHIDI, F.: Importance of insoluble-bound phenolics to antioxidant properties of wheat. J. Agric. Food Chem., 54, 2006, 1256-1264.

MUCHOVÁ, Z., FRANČÁKOVÁ, H., BOJŇANSKÁ, T., MAREČEK, J.: Cereals. The evaluation of raw materials and plant products, SAU, Nitra, SK, 2011, 50-84.

OYAIZU, M.: Studies on products of browning reaction prepared from glucoseamine. Jpn. J. Nutr., 44, 1986, 307-314.

PETERSON, D.M.: Oat antioxidants. J. Cereal Sci., 33, 2001, 115-129.

PIN-DER, D.: Antioxidant activity of Budrock (*Arctium lappa*, L.): its scavenging effect on free radical and active oxygen. J. Am. Oil. Chem., 75, 1998, 455-461.

PRISENŽŇÁKOVÁ, Ľ., NOSÁĽOVÁ, G., HROMÁDKOVÁ, Z., EBRINGEROVÁ, A.: The pharmacological activity of wheat bran polysaccharides. Fitoterapia, 81, 2010, 1037-1044.

QUETTIER-DELEU, CH., GRESSIER, B., VESSEUR, J., DINE, E., BRUNET, C., LUYCKX, M., CAZIN, M., CAZIN, J.C., BAILLEUL, F.: Phenolic compounds

and antioxidant activities of buckwheat (*Fagopyrum esculentum* Moench) hulls and flour. J. Ethnopharm., 1-2, 2000, 35-42.

RAGAEE, S., ABDEL-AAL, E.S. M., NOAMAN, M.: Antioxidant activity and nutrient composition of selected cereals for food use. Food Chem. 98, 2006, 32-38.

SCHNÜRER, J.: Distribution of fungal biomass among fine bran, coarse bran, and flour from wheat stored at four different moisture levels. Cereal Chem., 68, 1991, 434-437.

SERPEN, A., GOKMEN, V., PELLEGRINI, N., FOGLIANO, V.: Direct measurement of total antioxidant capacity of cereal products. J. Cereal Sci., 48, 2008, 816-820.

SHAHIDI, F., NACZK, M..: Phenolics in food and nutraceuticals. CRC Press, Washington, 2004, 566.

SINGLETON, V.L., ROSSI, J.A.: Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. Am. J. Enol. Agri., 6, 1965, 144 – 158.

STALIKAS, C. D.: Extraction, separation, and detection methods for phenolic acids and flavonoids. J. Sep. Sci. 30, 2007, 326-329.

VAHER, M., MATSO, K., LEVANDI, T., HELMJA, K., KALJURAND, M.: Phenolic compounds and the antioxidant activity of the bran, flour and whole grain of different wheat varieties. Procedia Chem., 2, 2010, 76-82.

VERMA, B., HUCL, P., CHIBBAR, R.N.: Phenolic acid composition and antioxidant capacity of acid and alkali hydrolysed. Food Chem., 116, 2009, 947-954.

YEN, G.C., CHEN, H. Y.: Antioxidant activity of various tea extracts in relation to their antimutagenicity. J. Agric. Food Chem., 43, 1995, 27-32.

ZHAO, H., FAN, W., DONG, J., LU, J., CHEN, J., SHAN, L., LIN, Y., KONG, W.: Evaluation of antioxidant activities and total phenolic contents of typical malting barley varieties. Food Chem., 107, 2008, 296-304.

ZIELIŃSKI, H., CEGLIŃSKA, A., MICHALSKA, A.: Antioxidant contents and properties as quality indices of rye cultivars. Food Chem., 104, 2008, 980-988.

Presented at the 3rd International Scientific Conference "Applied Natural Sciences - 2011", October 5–7, 2011, Častá Papiernička, Slovak Republic.