

THE SCAVENGING EFFECT AND FLAVONOID GLYCOSIDES CONTENT IN FRACTIONS FROM FRUITS OF HOT PEPPER *CAPSICUM ANNUUM* L.

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ABSTRACT

Background. The aim of this study was to isolate phenolic compounds fraction (PCF) from hot pepper and to characterise their composition and antiradical activity.

Material and methods. PCF was isolated from hot pepper fruits by solid phase extraction technique and separated by medium pressure liquid chromatography on Büchi system. HPLC quantitative analysis of sub-fractions constituents and of standards was made on Empower-Pro chromatograph (Waters) with DAD detection. Total phenolics content was assessed by Folin-Ciocalteu method. Antioxidant activity was measured according to superoxide radical, generated in NADH/PMS model and DPPH radical.

Results. The separation yielded three subfractions with different composition. In fraction 1, the predominant component was quercetin 3-*O*-rhamnoside-7-*O*-glucoside. The presence of luteolin 7-*O*-apiosyl-glucoside was observed in fraction 2, and the key component of fraction 3 was quercetin 3-*O*-rhamnoside. The highest antioxidant activity was noted for fraction 2, both in reducing superoxide radicals ($IC_{50} = 97 \mu\text{g}\cdot\text{cm}^{-3}$) and DPPH $^{\cdot}$ ($IC_{50} = 43 \mu\text{g}\cdot\text{cm}^{-3}$).

Conclusion. Pepper fruits are a rich source of phenolic compounds with a considerable lipophilicity and different chemical activity. They can be used as supplements to enhance food products.

Key words: antioxidant activity, *Capsicum annuum*, phenolic compounds, preparative chromatography

INTRODUCTION

The main purpose of food is to satiate hunger, supply energy and essential nutrients for growth and health, and to provide a satisfactory sensory experience. Contemporary food science searches for foods that not only supply vital nutrients, but also deliver health benefits. Intensive research efforts undertaken in the past decade in combination with analyses of correlations between diet and human health indicate that in addition to essential nutrients, food products also contain various non-nutritive substances that may contribute to the prevention and, in some cases, the

treatment of various diseases, including atherosclerosis and cancer [Halliwell 1996, Chen et al. 2008].

The group of plant constituents that activate the body's natural defense mechanisms against free radicals is inclusive of phenolic compounds, fat-soluble vitamins A and E and water-soluble vitamin C [Kaur and Kapoor 2001, Prior 2003]. Those compounds participate in various metabolic processes to boost immunity, and when incorporated into the diet on a regular basis, they can directly or indirectly influence homeostatic regulation [Halliwell 1996, Prior 2003].

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In vegetables, the highest content of vitamin C is noted in peppers [Marin et al. 2004]. *Capsicum* fruits are also a rich source of vitamin E, a first-line antioxidant, provitamin A (β -carotene and β -cryptoxanthin), phenolic compounds, mainly flavonoids and phenolic acids, as well as alkaloids – capsaicinoids which are responsible for the hot taste of chili peppers [Marin et al. 2004]. Owing to their high bioactive compounds content, the consumption of peppers may prevent various diseases caused by reactive oxygen species or some chemical carcinogens and mutagens [Howard et al. 2000]. Peppers are consumed raw, as well as in powdered form as spice.

Spices modify the sensory attributes of food, and they have been long used as natural food preservatives [Hinneburg et al. 2006, Suhaj 2006]. In contemporary market practice, synthetic food preservatives are often replaced with natural substances. Spices can be used as natural food preservatives to a limited extent due to their effect on sensory properties of food products. Phenolic compounds isolated from edible plants in the form of complex fractions or in pure form seem to offer an attractive alternative to synthetic substances. Acting as antioxidants they can limit free radical reactions in food. As an additional advantage, they have a less profound effect on the taste of food products than spices.

In the experiment discussed, phenolic compounds were extracted from the fruits of hot pepper *Capsicum annuum* L. The objective of the study was separation of fractions containing phenolic compounds and description of their composition based on reference substances as well as on their antiradical activity.

MATERIAL AND METHODS

The experimental material comprised the pericarps of hot pepper *Capsicum annuum* L., cv. Capel Hot, supplied by the Seminis Seed Farm near Lublin in Poland.

Extract preparation. Chopped pericarps (1100 g) were homogenized in 80% aqueous ethanol solution ($3 \times 1 \text{ dm}^3$) in the Diax 900 homogenizer. The ethanolic extract (EE) was filtered, and the filtrate was condensed in a rotary evaporator at 40°C to remove solvents. The condensed EE was diluted with redistilled water to a known volume (50 cm^3) and one milliliter of EE was taken. It was dried, weighed and frozen before later analyses. The remaining EE was put into

a sintered funnel filled with conditioned modified silica gel C18 (LichroPrep, Merck, $40\text{-}63 \mu\text{m}$). Hydrophilic substances were eluted with redistilled water and next, the phenolic compound fraction (PCF) was eluted with 40% aqueous methanol solution in a quantity required to produce a colorless filtrate. Nitrogen was passed through the filtrate to remove air. The PCF was diluted in redistilled water to reduce methanol concentration and enrichment on C18 gel. PCF was eluted from gel matrix with a small volume of 80% methanol-water solution and the eluate was evaporated in a rotary evaporator at 40°C to dryness. Dry residue was weighed, dissolved in water (50 cm^3) and one milliliter of PCF was taken for the subsequent analyses. PCF in water was subjected to preparative separation.

Preparative chromatography. The PCF was separated by medium pressure liquid chromatography (MPLC) using the Büchi system which comprised medium pressure pump (C-601), a manual injector unit, preparative column, UV detector (C-635) and fraction collector (C-660). A preparative column with 4.5 cm diameter and 49 cm of length, filled with modified silica gel C18 (LichroPrep, Merck $40\text{-}63 \mu\text{m}$) was used. Fractions eluted from the column with 20%, 40% and 100% methanol-water solution were collected into 15 cm^3 test tubes. A scheme of the experimental procedure is shown in Figure 1. The absorbance

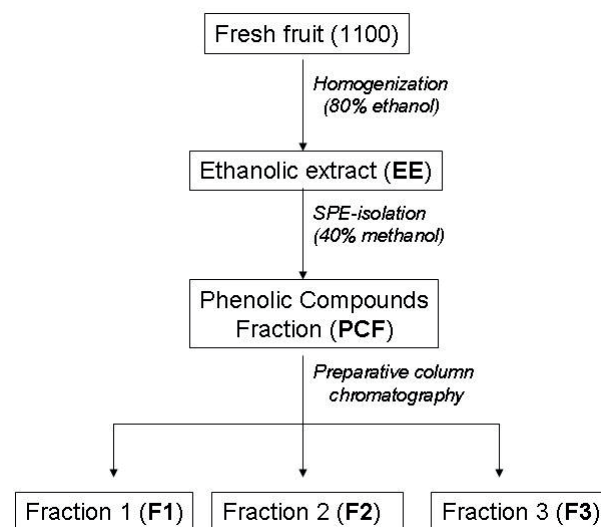


Fig. 1. Diagram of experimental procedure

at $\lambda = 330$ nm of each collected tube was recorded and the phenolics profile in tubes with the maximum and half of maximum absorbance was checked by HPLC method. Samples with similar phenolics profile were combined and put again to HPLC analysis.

HPLC analysis. A quantitative analysis of standard compounds and qualitative and quantitative analyses of fraction constituents were performed by HPLC on the Empower-Pro chromatograph (Waters) consisting a quaternary pump (M2998 Waters) with degasser and UV-Vis diode array detection (DAD) system. Separation was done on a column filled with a modified silica gel RP-18 (Atlantis T3 – Waters, 3 μm , 4.6 mm \times 150 mm), gradient elution consisting of mobile phase A (1% H_3PO_4 in water) and B (40% acetonitrile in solvent A) in such a proportion that the concentration of solvent B was: till the 10th min – 8%, in the 40th min – 20% and in the 55th min – 40%; the flow rate was 1 cm^3/min , and the detection was at 330 nm and 280 nm.

For a qualitative evaluation, flavonoid derivatives were compared with the chromatograms of analytical standards performed in identical conditions. Reference compounds were isolated from pepper fruits and identified by MS and NMR techniques in earlier analyses [Materska et al. 2003 a, b]. Quantitative analyses were based on standard calibration curves, and the content of main fraction components was determined on the basis of surface area of chromatogram peaks.

Antioxidant activity and total phenolic content. To determine the antioxidant activity and the total phenolic content of fractions isolated in a preparative column, each fraction was dissolved to produce a solution with a known concentration ($\text{g}\cdot\text{dm}^{-3}$). For this purpose, 5 to 10 cm^3 of each fraction was condensed in a vacuum evaporator under reduced pressure at 40°C, and next was dried in a nitrogen stream. Dry samples were weighed and used to prepare stock solutions which were then diluted to produce fraction concentrations ranging from 0.1 to 1.5 $\text{mg}\cdot\text{cm}^{-3}$. Stock solutions of initial EE and PCF were prepared simultaneously in an analogical way from 1 cm^3 of collected fraction.

Determination of total phenolic content. The total content of phenolic compounds in fractions from preparative column as well EE and PCF was determined by the Folin-Ciocalteu method [Zheng and

Wang 2001]. The total phenolic content was calculated on the basis of a reference curve, and the results were expressed as milligrams of chlorogenic acid equivalent per gram of analysed fraction [Conforti et al. 2007].

Antioxidant activity. The antioxidant activity of diluted fractions was determined as ability to scavenge superoxide radicals and DPPH radicals. The assays were performed in triplicate for each dilution. Simultaneously with prepared fractions, antiradical activity of quercetin, luteolin and vitamin C standards was evaluated.

Superoxide radicals were generated in the NADH/PMS system [Valentao et al. 2001]. The reaction mixture comprised a reduced form of adenine dinucleotide (NADH; 10.64 mM), nitro blue tetrazolium (NBT; 0.24 mM), phenazine methyl sulfate (PMS; 5.39 μM) and the tested sample (0.1 cm^3). The components were dissolved in 19 mM phosphate buffer with a pH of 7.4. The reaction was initiated through the addition of PMS. Reaction cuvettes were kept at 37°C for 30 minutes. The absorbance was measured at 560 nm wavelength using Cary 50 (Varian) spectrophotometer. Antioxidant activity was calculated based on formula (1). IC_{50} values, i.e. the concentrations at which fractions demonstrated 50% activity, were determined based on the correlations between antioxidant activity and fraction concentrations.

$$\%AA = 1 - \frac{\text{sample absorbance}}{\text{control absorbance}} \cdot 100\% \quad (1)$$

Evaluation of scavenging effect on DPPH radicals is described in detail in Materska and Perucka [2005]. A methanol stock solution of DPPH at a concentration of 0.1 $\text{mol}\cdot\text{dm}^{-3}$ was prepared. The absorbance of the mixture of analysed samples and DPPH working solution was measured at 517 nm wavelength. The reference was a test tube containing methanol instead of the sample. Antioxidant activity was determined as percentage discoloration of the DPPH solution once the plateau was reached in all samples (30 min) in comparison with the reference. The calculations were performed using formula (1). Next, IC_{50} values were evaluated on the basis of dependence between antiradical activity and sample concentration $f(c) = \%AA$.

Statistical analysis. Data were expressed as a means \pm SD. Statistical analysis was performed by using

Tukey multiple range test, with 5% error probability. The dose-response curve was obtained by plotting the percentage of inhibition versus concentration. The inhibitory concentration of 50% (IC_{50}) was calculated by linear or nonlinear regression analysis. Statistical comparisons were performed using Statgraphic Centurion software, version VXI.

RESULTS AND DISCUSSION

Preparative separation of PCF produced five fractions. HPLC analysis of obtained fractions showed that two were marked by an absence of significant peaks, and they were excluded from further analyses. The composition of the remaining three fractions named F1, F2 and F3 varied significantly (Fig. 2 B, C, D).

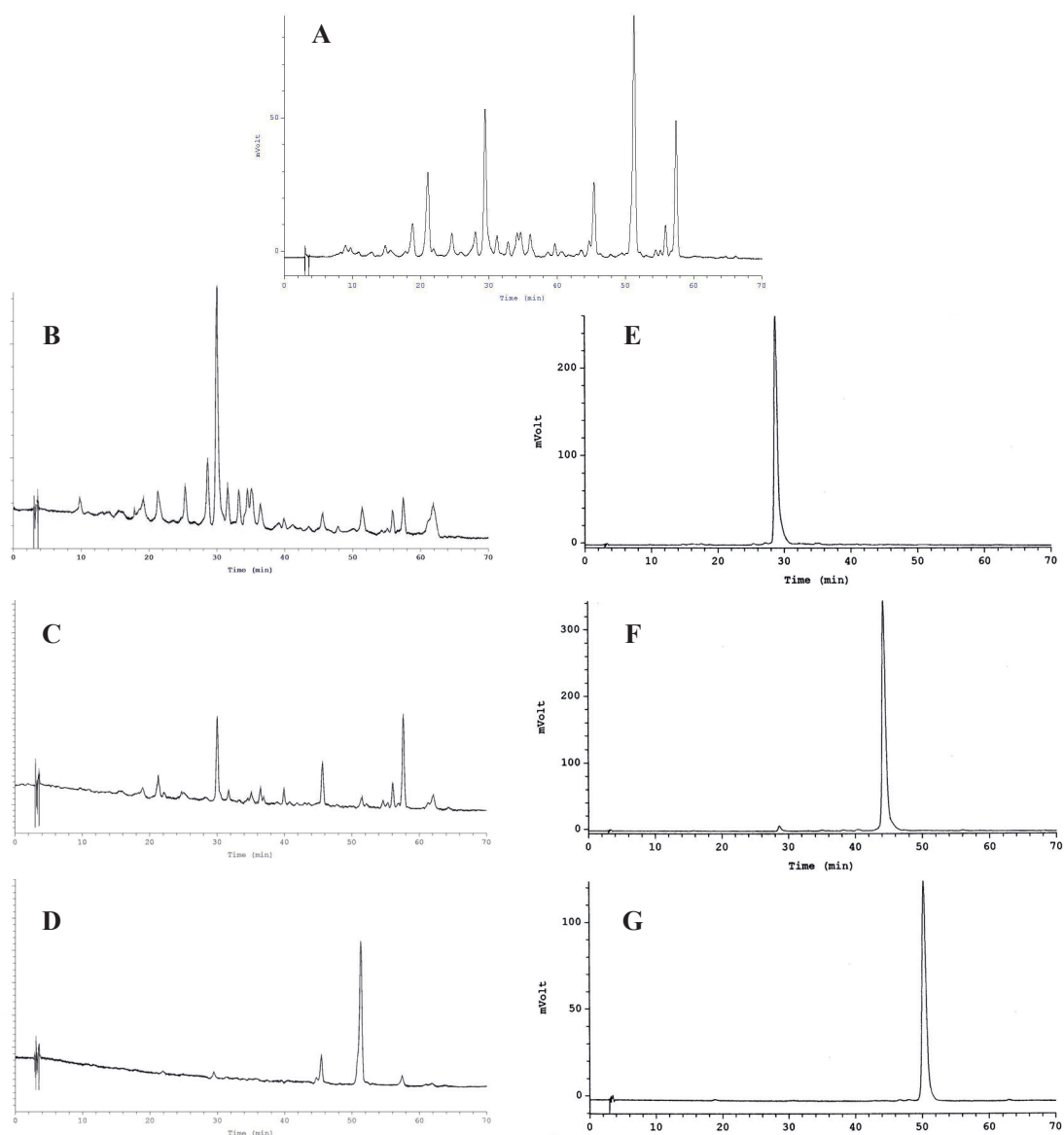


Fig. 2. Chromatograms of PCF (A) and fractions isolated by preparative separation. B – F1, C – F2, D – F3 and standard solutions of flavonoids derivatives, E – quercetin 3-*O*-rhamnoside-7-*O*-glucoside, F – luteolin 7-*O*-apiosylglucoside, G – quercetin 3-*O*-rhamnoside

Calculated extraction yields on each step of the presented procedure (Table 1) can give an overall view on the mass of individual fractions. Because the separation was done in a preparative scale, the obtained results may be considered only as an approximation, although Conforti et al. [2007] obtained similar extraction yield in the first step of extraction.

Table 1. Yield of extraction steps and total phenolics (TP) content

Fraction	Mass g	Yield %	TP*
EE	92	8.4	2 986
PCf	1.3	0.12	274
F1	0.0702	0.006	11
F2	0.0072	0.00065	4.5
F3	0.119	0.01	77

*Values expressed as mg of chlorogenic acid in 1000 g of fresh fruit.

Standard solutions and the PCF were subjected to chromatographic analysis, and the resulting chromatogram is presented in Figure 2. The PCF was characterised by five main peaks, of which three coincided with the standards chromatograms (Fig. 2 E, F, G). The peak with a retention time of 29.4 corresponded to quercetin 3-*O*-rhamnoside-7-*O*-glucoside (Fig. 2 E), the peak

with a retention time of 45.4 coincided with the peak reported for luteolin 7-*O*-apiosylglucoside, (Fig. 2 F), whereas the peak with a retention time of 51.3 corresponded to quercetin 3-*O*-rhamnoside (Fig. 2 G). The presence of a large number of compounds whose peaks were characterised by a smaller surface area was determined in the analysed fraction in the range of 8 to 58 minutes. The chromatograms of fractions obtained during PCF separation in the preparative column are presented in Figure 2 B, C and D. Fraction F1 had a complex composition, and the predominant component was quercetin 3-*O*-rhamnoside-7-*O*-glucoside which had a 32% share of the peak area (Fig. 2 B). This fraction also contained small quantities of luteolin 7-*O*-apiosylglucoside and quercetin 3-*O*-rhamnoside which had a 2.8% and 3.2% share of the peak area, respectively. The main components of F2 were not identified, but the presence of luteolin 7-*O*-apiosylglucoside was found with a 16.1% share of the peak area (Fig. 2 C). The predominant compound in F3 was quercetin 3-*O*-rhamnoside whose content accounted for 83.8% of the peak area (Fig. 2 D). The reported results indicate that the PCF isolated from pepper pericarps is a mixture of substances of varying lipophilicity that can be separated on a preparative column. A higher resolution chromatographic system is needed to isolate pure substances. Based on the mass of the separated fractions and % peak area of predominant compounds, the content of those substances was determined. Quercetin 3-*O*-rhamnoside had the highest share of phenolic compound fractions (Table 2).

Table 2. Contents and composition of fractions from preparative chromatography

Fraction	Identified compounds	Retention time min	Compounds mass in fraction* mg
F1	quercetin 3- <i>O</i> -rhamnoside-7- <i>O</i> -glucoside	29.95	22.72
	luteolin 7- <i>O</i> -apiosylglucoside	45.59	1.96
	quercetin 3- <i>O</i> -rhamnoside	51.37	2.23
F2	luteolin 7- <i>O</i> -apiosylglucoside	45.65	1.16
F3	luteolin 7- <i>O</i> -apiosylglucoside	45.44	14.70
	quercetin 3- <i>O</i> -rhamnoside	51.28	100.27

*Calculated for all fractions.

The reported results are consistent with the phenolic compound profile of the PCF solution (Fig. 2 A). Previous research investigating the phenylpropanoid content of pepper pericarps validates the results noted in this study [Marin et al. 2004, Materska and Perucka 2005]. Quercetin 3-*O*-rhamnoside was the predominant flavonoid in the fruits of both sweet and hot peppers, while quercetin was the main aglycone of PCF.

The PCF consisted mostly of flavonoid derivatives bound to sugar molecules. The presence of sugar residues in a flavonoid molecule increases its hydrophilicity which significantly affects the transport of the compound inside plant cells [Rice-Evans et al. 1996]. Both the number and the location of glycosidic bonds alter the properties of free aglycones from lipophilic to hydrophilic. In general, the higher the number of glycosidic bonds in a flavonoid particle, the more water-soluble it becomes. The above is validated by the discussed experiment where the reference with the highest number of glycosidic bonds (quercetin 3-*O*-rhamnoside-7-*O*-glucoside) was characterised by the shortest retention time (Fig. 2 E, Table 2). Since the derivatives of phenolic compounds have varied affinity for water, the selected solvent had to guarantee the highest extraction efficiency and eliminate typically hydrophilic compounds, including vitamins and sugars, as well as typical lipophilic substances, such as carotenoids. Solvents are selected based on the popular aphorism of "like dissolves like". The majority of polar compounds (phenolic acids, water-soluble vitamins) are isolated with the use of water as the solvent. Our experience and other authors' findings suggest that polar compounds are most effectively separated in 80% aqueous ethanol solution [Howard et al. 2000, Marin et al. 2004]. The use of ethanol solution in preparative-scale extraction eliminates the threats related to the contact with the reagent, which is unavoidable at this stage of the experiment, and it supports the extraction of all hydrophilic compounds and substances with intermediate lipophilicity from the pericarp. The chromatogram of PCF resembles previous chromatogram profiles [Marin et al. 2004, Materska and Perucka 2005]. A chromatographic analysis of fractions yielded by the preparative column suggests relatively low resolution (Fig. 2 B, C, D). In this study, methanol concentrations were marked by excessive gradient. To increase column resolution, the gradient of

methanol concentrations should be decreased; alternatively, a linear gradient for 15-40% methanol concentrations should be applied. The above solutions could not have been deployed in the discussed experiment due to equipment limitations.

The total phenolic compounds content expressed as chlorogenic acid equivalent in 1 g of examined fraction (Fig. 3) clearly show changes in their composition. EE as a product of first step of extraction contain different compounds from pepper fruit and only few react with the Folin-Ciocalteu reagent. In turn, the fractions obtained after PCF separation, there are consist of compounds which react with Folin-Ciocalteu reagent. The highest content of phenolic compounds was found in F2 and F3 (Fig. 3).

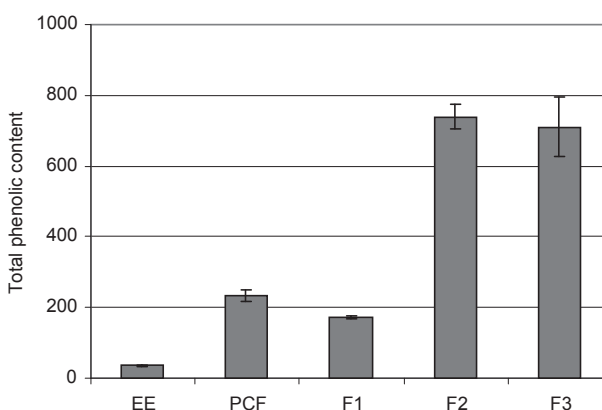


Fig. 3. Total phenolic content expressed as mg of chlorogenic acid·g⁻¹ of analysed fraction

The antioxidant properties of fractions from preparative column isolation were analysed by two methods. In the first, absorbance changes induced by the reaction between the superoxide radical (generated in the NADH/PMS) system and NBT was measured, where the presence of the analysed sample limited free radical synthesis, thus lowering NBT reduction and absorbance. In the second method, the drop in absorbance values during the reduction of a 2,2-diphenyl-1-picrylhydrazyl (DPPH) stable radical in the studied phenolic compound solutions was recorded. To compare the antiradical power of isolated fractions, analysis of quercetin and luteolin standards, as well

as vitamin C and PCF was performed simultaneously. The scavenging effects of fractions from preparative column on superoxide radicals and DPPH[•], examined at different sample concentrations showed a dose-response relationship (Fig. 4 and 5). The highest activity of F2 both in NADH/PMS and DPPH[•] model systems was noted (Table 3). In NADH/PMS system the antioxidant activity of F2 was comparable to the activity of PCF and of luteolin, while according to DPPH radical, activity of F2 was lower than that of standard compounds, but the highest among analysed subfractions (Table 3).

Antioxidant activity assessment methods used in presented work may be classified to single electron transfer (SET) [Conforti et al. 2007]. The difference between them lies in the nature of radicals. Superoxide radicals are hydrophilic, because they are generated in buffered environment. DPPH radicals are insoluble in water and its solutions are prepared in methanol. Because of similar mechanism of antioxidants action, differences in activity between analysed samples come rather from hydro-liphophilic character, than from their electron donating ability. In NADH/PMS method the highest activity of hydrophilic vitamin C

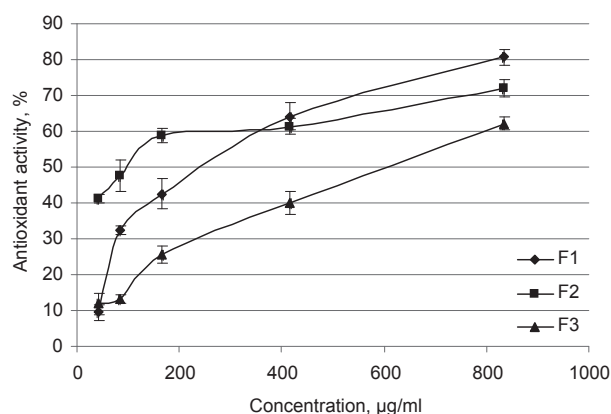


Fig. 4. Antioxidant activity of phenolic compound fractions from pepper fruit in NADH/PMS model system n = 3, ±SD

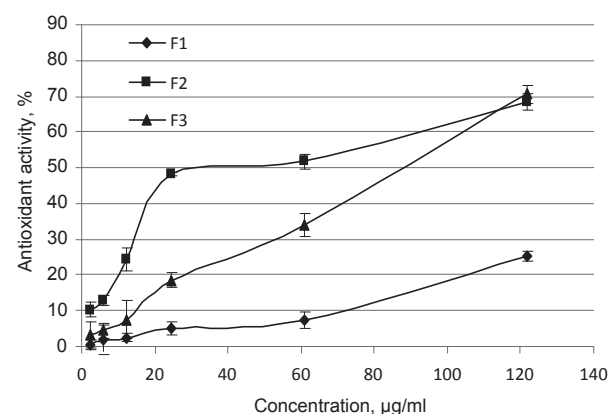


Fig. 5. Antioxidant activity of phenolic compound fractions from pepper fruit in DPPH model system n = 3, ±SD

Table 3. Antioxidant activity of the PCF and fractions isolated by preparative column chromatography, expressed in terms of IC₅₀ values, µg·cm⁻³

	Superoxide radical			DPPH [•]		
	regression line	R ²	IC ₅₀	regression line	R ²	IC ₅₀
PCF	y = 0.353 x + 22.332	0.997	78	y = 2.612 x - 15.153	0.995	25
F1	y = 22.883 ln(x) - 73.235	0.991	218	y = 0.197 x - 0.525	0.940	256
F2	y = 9.81 ln(x) + 5.123	0.961	97	y = 15.555 ln(x) - 8.33	0.937	43
F3	y = 0.063 x + 11.007	0.979	615	y = 0.561 x + 1.68	0.995	86
Quercetin	y = 14.405 ln(x) + 15.642	0.930	10.9	y = 13.545 x + 18.866	0.971	2.3
Luteolin	y = 15.603 ln(x) - 22.051	0.896	101.3	y = 7.749 x + 1.324	0.995	6.3
Vitamin C	y = 32.922 ln(x) + 13.93	0.962	3	y = 10.399 x + 0.967	0.9997	4.7

was confirmed, while in DPPH the highest activity of quercetin was noted (Table 3). Both PCF and fractions from preparative chromatography separation contained compounds with medium lipophilicity. They are still complex fractions and activity of their may be explained in a number of ways. F2, which was the most complex and consisted of compounds of medium lipophilicity, showed the highest activity in two model systems. On the contrary, F3 was the purest fraction and it showed higher activity in lipophilic medium, while it was inactive in relation to superoxide radical (Table 3). F1 showed low activity in both analysed model systems. Probably in this fraction the compounds presented on chromatogram were not active as antioxidants. Additionally, in DPPH model system antioxidant activity of prepared subfractions correlated well with total phenolic compounds content ($r = 0.934$; data not shown). A number of authors confirmed high correlation between total phenols content and antioxidant activity in this model system [Conforti et al. 2007, Alvarez-Parilla et al. 2011].

CONCLUSION

Pepper fruits are a rich source of phenolic compounds with intermediate lipophilicity. Those compounds are difficult to separate due to their physical similarities. The choice of appropriate conditions for chromatographic separation, i.e. column size, sorbent type, eluent concentration and flow rate, supports the successful isolation of pure substances.

The chemical activity of phenolic compounds varies subject to the applied oxidation factor. The results of this study indicate that pepper pericarp is a rich source of antioxidants which can be used as supplements to enhance food products, delivering significant benefits when consumed in unprocessed form.

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REFERENCES

- Alvarez-Parrilla E., De La Rosa L., Amarowicz R., Shahidi F., 2011. Antioxidant activity of fresh and processed Jalapeno and Serrano peppers. J. Agric. Food Chem. 59, 163-173.
- Chen Z.Y., Jiao R., Ma K.Y., 2008. Cholesterol-lowering nutraceuticals and functional foods. J. Agric. Food Chem. 56, 8761-8773.
- Conforti F., Statti G.A., Menichini F., 2007. Chemical and biological variability of hot pepper fruits (*Capsicum annuum* var. *acuminatum* L.) in relation to maturity stage. Food Chem. 102, 1096-1104.
- Halliwell B., 1996. Antioxidants in human health and disease. Ann. Rev. Nutr. 16, 39-50.
- Hinneburg I., Dorman D.H.J., Hiltunen R., 2006. Antioxidant activities of extracts from selected culinary herbs and spices. Food Chem. 97, 122-129.
- Howard L.R., Talcott S.T., Brenes C.H., Villalon B., 2000. Changes in phytochemical and antioxidant activity of selected pepper cultivars (*Capsicum* species) as influenced by maturity. J. Agric. Food Chem. 48, 1713-1720.
- Kaur Ch., Kapoor H.C., 2001. Antioxidants in fruits and vegetables – the millennium's health. Int. J. Food Sci. Tech. 36, 703-725.
- Marin A., Ferreres F., Tomas-Barberan F.A., Gil M.I., 2004. Characterization and quantitation of antioxidant constituents of sweet pepper (*Capsicum annuum* L.). J. Agric. Food Chem. 52, 3861-3869.
- Materska M., Perucka I., 2005. Antioxidant activity of the main phenolic compounds isolated from hot pepper fruit (*Capsicum annuum* L.). J. Agric. Food Chem. 53, 1750-1756.
- Materska M., Perucka I., Stochmal A., Piacente S., Oleszek W., 2003 a. Quantitative and qualitative determination of flavonoids and phenolic acid derivatives from pericarp of hot pepper fruit cv. Bronowicka Ostra. Pol. J. Food Nutr. Sci. 12/53, SI 2, 72-76.
- Materska M., Piacente S., Stochmal A., Pizza C., Oleszek W., Perucka I., 2003 b. Isolation and structure elucidation of flavonoid and phenolic acid glycosides from pericarp of hot pepper fruit *Capsicum annuum* L. Phytochemistry 63, 893-898.
- Prior R.L., 2003. Fruits and vegetables in the prevention of cellular oxidative damage. Am. J. Clin. Nutr. 78, 570-578.
- Rice-Evans C., Miller N.J., Paganga G., 1996. Structure-antioxidant activity relationships of flavonoids and phenolic acids. Free Rad. Biol. Med. 20, 933-956.

- Suhaj M., 2006. Spice antioxidants isolation and their antiradical activity: a review. J. Food Comp. Anal. 19, 531-537.
- Valentao P., Fernandez E., Carvalho F., Andrade P.B., Seabra R.M., Bastos M.L., 2001. Antioxidant activity of *Centaureum erythraea* infusion evidenced by its superoxide radical scavenging and xanthine oxidase inhibitory activity. J. Agric. Food Chem. 49, 3476-3479.
- Zheng W., Wang S., 2001. Antioxidant activity and phenolic compounds in selected herbs. J. Agric. Food Chem. 49, 5165-5170.

AKTYWNOŚĆ ANTYOKSYDACYJNA ORAZ ZAWARTOŚĆ GLIKOZYDÓW FLAWONOIDOWYCH WE FRAKCJACH Z OWOCÓW PAPRYKI OSTREJ *CAPSICUM ANNUUM* L.

STRESZCZENIE

Cel. Badania miały na celu rozdzielanie frakcji związków fenolowych na mniej złożone podfrakcje i ich charakterystykę pod względem jakościowym i ilościowym oraz aktywności antyoksydacyjnej.

Materiał i metody. Frakcję związków fenolowych izolowano z owocni papryki metodą ekstrakcji do fazy stałej. Następnie rozdzielano ją na podfrakcje metodą cieczowej chromatografii średniociśnieniowej na zestawie firmy Büchi. Analizę ilościową składników podfrakcji oraz substancji wzorcowych wykonano metodą HPLC na chromatografie Empower-Pro firmy Waters z detektorem DAD. Sumę związków fenolowych w uzyskanych frakcjach oznaczono metodą Folina-Ciocalteu'a, a aktywność antyoksydacyjną oznaczono w odniesieniu do rodnika ponadtlenkowego generowanego w układzie NADH/PMS oraz do rodnika DPPH.

Wyniki. W wyniku rozdziału frakcji związków fenolowych otrzymano trzy podfrakcje o różnym składzie. We frakcji 1 dominującym związkiem był 3-*O*-ramnozyd-7-*O*-glukozyd kwercetyny. We frakcji 2 stwierdzono obecność 7-*O*-apoizydo-glukozydu luteoliny, natomiast związkiem dominującym we frakcji 3 był 3-*O*-ramnozyd kwercetyny. Największą aktywność antyoksydacyjną zanotowano dla frakcji 2, zarówno w odniesieniu do rodnika ponadtlenkowego ($IC_{50} = 97 \mu\text{g} \cdot \text{cm}^{-3}$), jak i rodnika DPPH ($IC_{50} = 43 \mu\text{g} \cdot \text{cm}^{-3}$).

Podsumowanie. Owoce papryki są bogatym źródłem związków fenolowych o pośredniej lipofilności i zróżnicowanej aktywności chemicznej. Mogą być one stosowane jako korzystne dla zdrowia dodatki do żywności.

Słowa kluczowe: aktywność antyoksydacyjna, *Capsicum annuum*, związki fenolowe, chromatografia preparatywna

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