Antioxidant Activity of Rye Caryopses and Embryos Extracts

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Abstract

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Phenolic compounds were extracted with 80% methanol from caryopses and embryos of rye (cv. Dańkowskie Złote and Amilo). In all extracts, reducing power, scavenging effect on DPPH radical, and antioxidant activity in a β-carotene-linoleate model system were examined. The highest content of total phenolic compounds was noted in the extract from caryopses of Amilo (7.93 mg/g of extract). UV spectra of all extracts were characterised by maxima originated from phenolic acids (320, 326 and 328 nm), and by maxima at shorter wavelengths (272 and 274 nm) attributed to other phenolic compounds. All extracts showed a good antioxidant activity in a β-carotene-linoleate model system. This activity was similar to that reported before in leguminous seeds extracts. The antioxidant activities of the extracts from the caryopses of Dańkowskie Złote and the embryos of both cultivars were very similar, especially during the second part of the incubation period. The extract of Amilo embryos showed a slightly weaker antioxidative effect. The weak antiradical effects in the experiments with DPPH radical and a weak reducing power were characteristic for all the extracts investigated.

Keywords: antioxidant activity; rye caryopses; rye embryos; free radical scavengers

Phenolic compounds are listed as a major natural antioxidants present in cereals grains (DANIELS & MARTIN 1967; TIAN &WHITE 1994; ANDLAUER & FURST 1998; YANG *et al*. 2001). In this group, phenolic acids are the best recognised components. They are present in grains as free, esterified, and glucosided compounds (WEIDNER *et al*. 2000, 2001; ZIELIŃSKI *et al*. 2001). Phenolic acids are known as active antioxidants (GRAF 1992; SHAHIDI & WANASUNDARA 1992). The antioxidant activity of grain extracts was confirmed in several studies (WHITE & XING 1995). ONYENEHO and HETTIARACHCHY (1992) observed the antioxidant activity in ethanol extracts of wheat bran in bulk oils system. Total antioxidant activity of water and methanol soluble extracts from cereal grains was determined by ZIELIŃSKI *et al*. (2000) and ZIELIŃSKI and KOZŁOWSKA (2000). A strong antioxidant effect of phenolic compounds in the extracts from triticale and wheat was observed in a β-carotene-linoleate model system (AMAROWICZ *et al*. 2002b,c). Water extracts obtained from wheat products inhibited a phosphatidylcholine liposome oxidation induced by iron/ascorbic acid (BAUBLIS *et al*. 2000). The extracts of triticale were capable of scavenging DPPH free radical (AMAROWICZ *et al*. 2002d).

The present study was undertaken to determine the antioxidant activity of rye caryopses and embryos extracts.

MATERIAL AND METHODS

Materials. Materials investigated were caryopses and embryos of rye (cv. Dañkowskie Z³ote and Amilo) which were collected from an experimental field of the University of Warmia and Mazuria in Olsztyn (Poland).

Extraction. Phenolic compounds were extracted from ground caryopses and embryos twice with 80% (v/v) methanol for 15 min at 80°C (AMAROWICZ *et al*. 1995). After evaporating the organic solvent in a rotary evaporator at 45°C, the remaining aqueous solution was lyophilised.

Total phenolics compounds and UV spectra. The contents of phenolic compounds in extracts were determined using the Folin-Ciocalteau reagent (NACZK & SHAHIDI 1989) while (+)-catechin was used as a standard. UV spectra of the extracts were recorded with a Beckman DU 7500 diode array spectrophotometer. The extracts were dissolved in methanol at a concentration giving absorbance less than 1.0.

Antioxidant activity. The antioxidant activity of the extracts was evaluated using a β-carotene-linoleate model system (MILLER 1971). Methanolic solutions (0.2 ml) containing 4 mg of extracts or 0.3 mg of butylated hydroxyanisole (BHA) were added to a series of tubes containing 5 ml of previous prepared emulsion of linoleate and β-carotene stabilised with Tween 40. Immediately after the addition of the emulsion to the tubes, the zero-time absorbance at 470 nm was recorded. Samples were kept in a water bath at 50°C and their absorbance values were recorded over a 120 min period at 15 min intervals.

Scavenging of DPPH radical. Scavenging effect of phenolic compounds present in the extracts on DPPH radical was monitored according to the method described by YEN and CHEN (1995). A 0.1 ml methanolic solution containing from 0.4 to 2.0 mg extracts was mixed with 2 ml of methanol and a methanolic solution of α,α-diphenyl-β-picrylhydrazyl (DPPH) (1mM, 0.250 ml) was then added. The mixture was vortexed for 15 s, then left to stand at room temperature for 30 min; the absorbance of this solution was then read at 517 nm.

Reducing power. The reducing power of phenolic compounds in the extracts was determined as described by OYAIZU (1986). The suspension of extracts (0.4–2.0 mg) in 1 ml of distilled water was mixed with 2.5 ml of 0.2M phosphate buffer (pH 6.6) and 2.5 ml of 1% potassium ferricyanide. The mixture was incubated at 50°C for 20 min. Subsequently, 2.5 ml of trichloroacetic acid was added and the mixture was then centrifuged at 1750 g for 10 min. A 2.5 ml aliquot of the upper layer was mixed with 2.5 ml of distilled water and 0.5 ml of 0.1% $FeCl_{3,}$ and the absorbance of the mixture was read at 700 nm.

RESULTS AND DISCUSSION

In Fig. 1 the content is shown of total phenolic compounds in the materials investigated. The extracts of Amilo were characterised by a higher content of total phenolic compounds (caryopses 7.93 mg/g of extract; embryos 7.60 mg/g of extract) than the extracts of Dańkowskie Złote (caryopses 6.09 mg/g of extract; embryos 4.42 mg/g of extract). In our previous investigations, the contents of total phenolic compounds in caryopses of two cultivars of wheat were 4.62 and 4.42 mg/g of extract. In the extracts of wheat embryos, the contents of total phenolic compounds were higher than in caryopses (6.05 and 5.25 mg/g of extract) (AMAROWICZ *et al*. 2002c). In the extracts of two cultivares of triticale, we noted 5.02 and 4.96 mg of total phenolic compounds per g of extract (caryopses) and 5.25 and 6.05 mg of total phenolic compounds per g of extract (embryos) (AMAROWICZ *et al.* 2002b). The content of total phenolic compounds in leguminous seeds extracts ranged from 0.97 to 8.09 mg/g of extract (AMAROWICZ & RAAB 1997). A higher content of phenolic compounds was observed in extracts of rapeseed (AMAROWICZ *et al*. 2001).

Fig. 2 depicts UV spectra of phenolic compounds extracted from rye caryopses and embryos. These spectra are characterised by maxima originating from phenolic acids at 320 nm (caryopses and embryos of both cultivars), and by maxima at shorter wavelengths: 282 nm (caryopses of both cultivars), 272 nm (embryos of Amilo), and 374 nm (embryos of Dańkowskie Złote) attributed to other phenolic compounds. Slight differences in the UV spectra of extracts at longer wavelengths were due to different contents of individual phenolic acids as reported previously (WEIDNER *et al*. 2000; AMAROWICZ & WEIDNER 2001). UV spectra reported are similar to those recorded for phenolic compounds in extracts from triticale and wheat caryopses and embryos (AMAROWICZ *et al*. 2002b,c).

The effect of the rye extracts investigated on the coupled oxidation of linoleic acid and β-carotene is presented in Fig. 3. The antioxidant activities of the extracts from the caryopses of Dańkowskie Złote and the embryos of both cultivars were very similar, especially during the second part of the incubation period. The extract of Amilo embryos showed a slightly weaker antioxidative effect. A similar

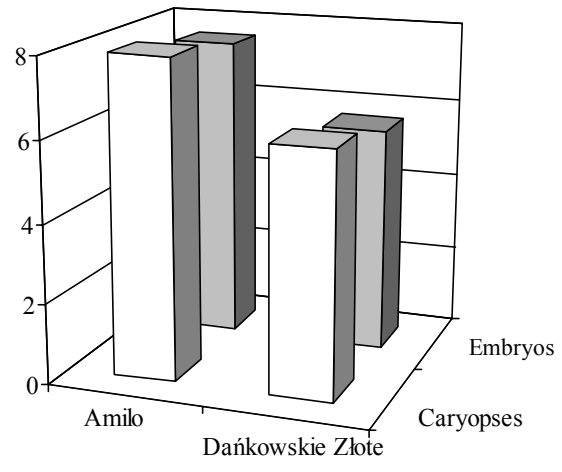

Fig. 1. Content of total phenolics in rye caryopses and embryos

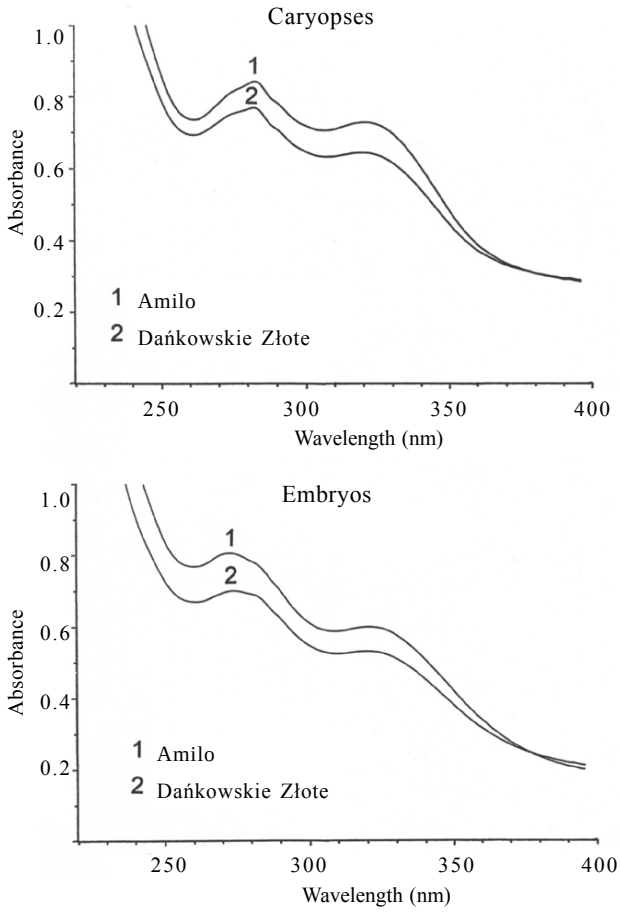

Fig. 2. UV spectra of extracts from rye caryopses and embryos

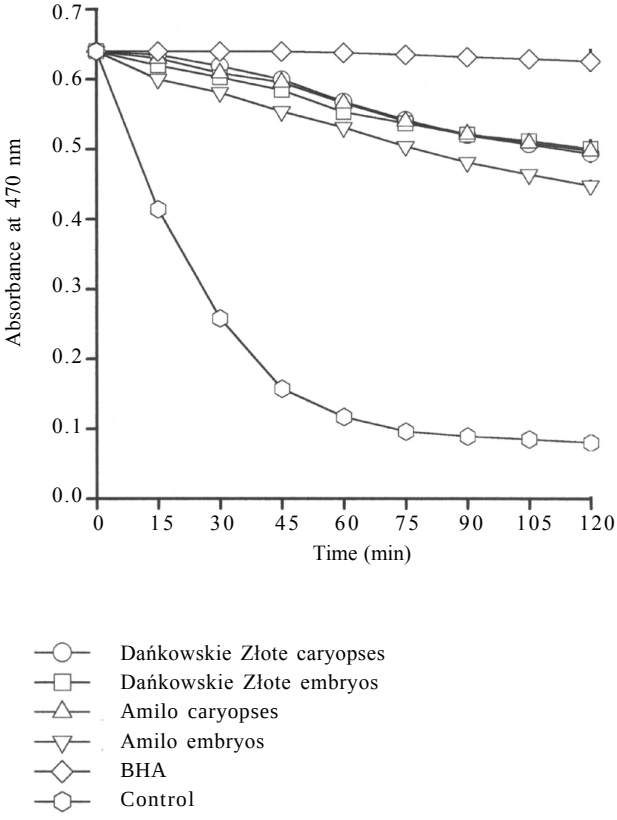

Fig. 3. Antioxidative activity of extract from rye caryopses and embryos in β-carotene-linoleate model system

antioxidant effect in a β-carotene-linoleate model system was observed in the extracts of wheat and triticale but the extracts obtained from the embryos of these cereals were more active than those from caryopses (AMAROWICZ *et al*. 2002b,c). Extracts of lentil, faba bean, and broad bean (AMAROWICZ *et al*. 1996) and extracts of rapeseed and rapeseed oil cake (AMAROWICZ & FORNAL 1995; AMAROWICZ *et al.* 2001) also exhibited an activity similar to that presented in this study. A much stronger antioxidant activity was determined in the extracts of phenolic compounds from canola hulls (AMAROWICZ *et al*. 2000a,b), condensed tannins of beach pea, canola hulls, and faba beans (AMAROWICZ *et al*. 2000c) but the contents of phenolic compounds in the studies cited were several times higher than those in the extracts evaluated in the present work.

The scavenging effect of wheat extract on DPPH radical is shown in Fig. 4. The weakest effect was noted in the extract of Amilo caryopses. The remaining extracts were more active, however, their effect on DPPH radical was also weak. In other our investigations (AMAROWICZ *et al*. 2002b,c) the phenolic compounds extracted from wheat and triticale caryopses and embryos also possessed a weak activity to scavenge DPPH radical. Much stronger scavenging effects on DPPH radical were found for green and black tea (YEN & CHEN 1995), extracts of legume seeds (AMAROWICZ *et al.* 2002a), canola hulls extract (AMAROWICZ *et al*. 2000a,b), condensed tannins of beach pea, canola hulls, and faba beans (AMAROWICZ *et al*. 2000c). The DPPH radical scavenging effect observed in this work is in agreement with literature data. According to BRAND-WILLIAMS *et al*. (1995) and SÁNCHEZ-MORENO *et al*. (1998), ferulic acid, the main phenolic acids in cereal grains, showed a weak antiradical effect in experiments with DPPH radical. In our previous study (WEIDNER *et al*. 1999), caffeic, *p*-coumaric, ferulic, and sinapic acids were the dominant phenolic acids detected in rye caryopses. The majority of phenolic acids were found in the form of soluble esters.

Fig. 5 shows the reducing powers of phenolic compounds extracted from rye caryopses and embryos. The extract of embryos of Amilo possessed the strongest reducing power. The extract of caryopses of Amilo was more active than both extracts of Dańkowskie Złote. The lowest reducing power was observed in the extract of cary-

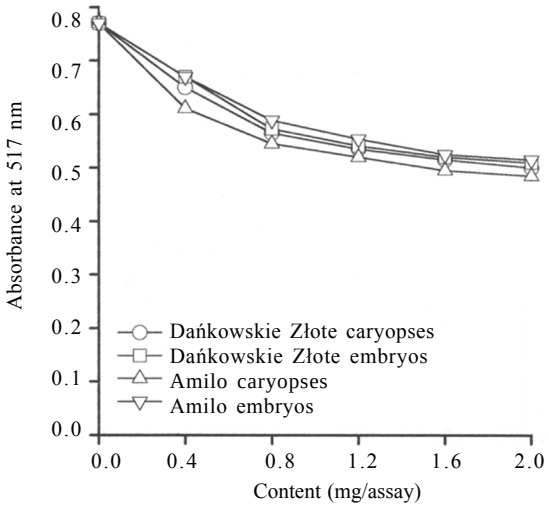

Fig. 4. Scavenging effect of extract from rye caryopses and embryos on DPPH radical

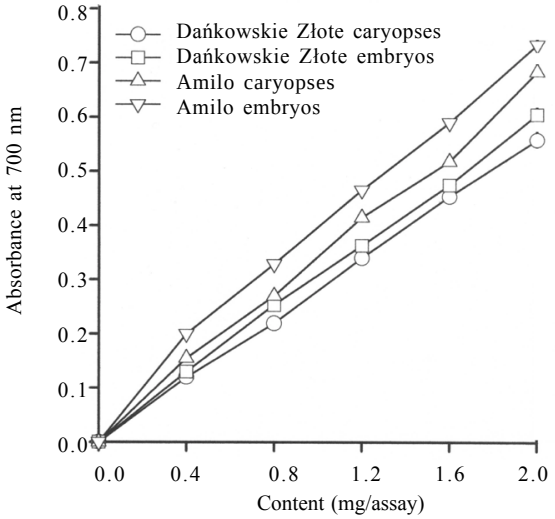

Fig. 5. Reducing power of extracts from rye caryopses and embryos

opses of Dańkowskie Złote. The lower reducing power of the extracts of Dańkowskie Złote can be caused by lower contents of phenolic acids than those present in the extracts of Amilo (WEIDNER *et al.* 1999). In our previous work (AMAROWICZ *et al.* 2002c), the reducing power of the extracts of wheat embryos proved to be much stronger than that of the extracts obtained from wheat caryopses. In the case of triticale, the extracts of embryos also exhibited a higher reducing power than the extracts of caryopses (AMAROWICZ *et al.* 2002b). The values obtained in the present work were much lower than those found for leguminous extracts (AMAROWICZ *et al.* 2002a), canola hulls extracts (AMAROWICZ *et al.* 2000a,b), and the extracts of condensed tannins (AMAROWICZ *et al.* 2000c).

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Souhrn

KARAMAĆ M., AMAROWICZ R., WEIDNER S., ABE S., SHAHIDI F. (2002): **Antioxidační aktivita extraktů žitných obilek a embryí.** Czech J. Food Sci., **20**: 209–214.

Ze žitných obilek a embryí (odrůdy Dańkowskie Złote a Amilo) byly extrahovány fenolické sloučeniny 80% metanolem. Ve všech extraktech byla sledována redukční schopnost, schopnost pohlcovat (scavenge) radikál DPPH a dále antioxidační aktivita v modelovém systému β-karoten-linoleát. Nejvyšší obsah fenolických sloučenin byl zjištěn v extraktu z obilek Amilo (7,93 mg/g extraktu). Pro UV spektra všech extraktů byla charakteristická maxima daná přítomností fenolických kyselin (320, 326 a 328 nm) a maxima v oblasti kratších vlnových délek (272 a 274 nm) přisouzená jiným fenolickým sloučeninám. Všechny extrakty prokázaly dobrou antioxidační aktivitu v modelovém systému β-karoten-linoleát. Tato aktivita byla obdobná jako aktivita extraktů luštěninových semen, o které bylo referováno již dříve. Antioxidační aktivity extraktů z obilek Dańkowskie Złote a z embryí obou odrůd byly

velmi podobné, zejména během druhé části inkubační doby. Antioxidační účinek extraktů z embryí Amilo byl poněkud nižší. U všech sledovaných extraktů byl charakteristicky slabý protiradikálový účinek při pokusech s radikálem DPPH i malá redukční schopnost.

Klíčová slova: antioxidační aktivita; žitné obilky; žitná embrya; pohlcovače (scavengers) volných radikálů

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