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INFLUENCE OF HYDROTHERMAL TREATMENT ON DIETARY FIBER AND PHENOLIC COMPOUNDS CONTENT AS WELL AS ANTIOXIDATIVE ACTIVITY OF LEGUMES SEEDS

Agnieszka Filipiak-Florkiewicz[1]✉, Adam Florkiewicz[1], Ewa Cieślik[1], Maria Walczycka[2], Joanna Kapusta-Duch[3], Teresa Leszczyńska[3]

[1]Department of Catering Technology and Consumption, University of Agriculture in Krakow
Balicka 122, 30-149 Cracow, **Poland**
[2]Department of Animal Product Technology, University of Agriculture in Krakow
Balicka 122, 30-149 Cracow, **Poland**
[3]Department of Human Nutrition, University of Agriculture in Krakow
Balicka 122, 30-149 Cracow, **Poland**

ABSTRACT

Background. Dry legumes seeds are food of high nutrient density. Besides the biologically precious protein they contain essential unsaturated fatty acids, vitamins of group B, dietary fiber and antioxidants. The aim of the research was to assess the influence of different soaking and cooking methods on dietary fiber and phenolic compounds content, as well as antioxidative activity of selected legumes' seeds.

Material and methods. The experimental material was dry kidney bean originating from collection of Krakow's Plants and Seeds Horticulture POLAN LTD, and soya bean seeds bought at the market store. The technological treatment of seeds was performed in two stages: 1) soaking in cold and hot water, 2) cooking with three different kinds of equipment – electrical stove, induction stove and microwave oven. In the above described ways prepared material was examined for the dietary fiber and phenolic compounds content, as well as antioxidative activity.

Results. The highest amount of dietary fiber was found in soya bean seeds cooked on an induction stove (27.4 $g \cdot 100\ g^{-1}$ d.m.) and electric stove (27.3 $g \cdot 100\ g^{-1}$ d.m.) after previous soaking in hot water. The dry soya bean and kidney bean seeds were characterised by a high content of phenolic compounds 163.2 mg $GAE \cdot 100\ g^{-1}$ d.m. and 173.1 mg $GAE \cdot 100\ g^{-1}$ d.m. respectively. The dry matter of cooked (on induction and electric stove) kidney bean and soya seeds was characterised by a higher phenolic compounds level (232.7--311.6 mg $GAE \cdot 100\ g^{-1}$ and 224.4-315.6 respectively) than raw material. Antioxidative activity of untreated seeds of kidney bean and soya bean was 4.5 and 4.1 µmol $Tx \cdot g^{-1}$ d.m. respectively. The high ability of free radicals scavenging was observed in kidney bean i.e. 8.7 µmol $Tx \cdot g^{-1}$ d.m. after traditional soaking and 9.5 µmol $Tx \cdot g^{-1}$ d.m. after "hot" soaking.

Conclusions. Soaking and cooking of seeds (despite the species) influenced growth of phenolic compounds content in dry matter. More advantageous way of soaking of kidney bean seeds was "hot" soaking. That way of treatment caused, first of all, shortening of the whole heat treatment process. Cooking of soya and kidney bean seeds with electric and induction stoves favoured keeping of dietary fiber, phenolic compounds and antioxidative activity of seeds in comparison to thermal treatment in combined microwave oven.

Key words: soya bean, kidney bean, soaking, cooking, induction stove, dietary fiber, phenolic compounds, antioxidative activity

✉afilipiak-florkiewicz@ar.krakow.pl

INTRODUCTION

Still underestimated in Poland, but very valuable and rich in pro health components, are dry legumes seeds. They are a food of high nutrient density. Besides the biologically precious protein they contain essential unsaturated fatty acids, vitamins of group B, dietary fiber and antioxidants. The important characteristic of dry legumes seeds is a fact that their mineral composition could influence on body's acid-base homeostasis. They are rich in calcium, phosphorus and magnesium [Korus 2002]. Opposite to other vegetables, legumes dry seeds are not consumed in a dry state. To make them edible it is necessary to conduct a few hydrothermal processes such as soaking, cooking or steaming. These processes give the consumption softness and also can cause lowering of anti-nutritive factors (e.g. trypsin inhibitors, tannins, phytic acid, flatulence-causing oligosaccharides) restricting high nutritive value. The hydrothermal treatment can diminish also levels of nutrient substances and non-nutritive compounds with pro health features [Filipiak-Florkiewicz et al. 2011].

Heating of plant products, in water, causes someway fast heat conduction into tissues, what is the cause of longer exposition on heat of the whole processed product and big losses in antioxidative substances [Gumul et al. 2005]. It is known, that cooking induces significant changes in chemical composition, affecting the bioavailability and content of chemopreventive compounds in vegetables. Cooking methods were shown to affect the contents of nutrient and health-promoting compounds such as vitamin C, carotenoids, polyphenols, and glucosinolates [Yuan et al. 2009]. For thermal cooking/processing of food different methods can be used. Microwave ovens are widely used in food-service establishments. The microwave oven has high thermal efficiency in comparison with conventional gas and electric ovens. Approximately, 75% less energy is required for microwave cooking or heating as compared to conventional methods [Nikmaram et al. 2011]. Induction cookers (also referred to as induction hobs) are electrical cooking appliances that use intermediate-frequency magnetic fields to heat the cooking vessel directly without heating the contact surface of the appliance itself. Their main advantages are rapid cooking times and higher energy efficiency compared to conventional (resistive) and glass-ceramic (infrared) cookers. With decreasing prices, induction cookers are gaining in popularity and up to 300 thousand units yearly are predicted for sale in Europe alone [Kos et al. 2011].

The aim of this research was to estimate the influence of different soaking and cooking methods on changes in contents of dietary fiber and phenolic compounds, as well as, antioxidative activity of selected legumes dry seeds.

MATERIAL AND METHODS

The experimental materials were dry legumes seeds i.e. kidney beans (vs. Piękny Jaś) originating from collection (2010 y.) of Krakow's Plants and Seeds Horticulture POLAN LTD and soya (the commercial mixture without declaration of eventual GMO origin) bought at market store. Initial average humidity of seeds of kidney and soya beans was 18.1 and 9.5% respectively. The dimensions of kidney beans: average width 13.5 ±0.7 mm, length 22.0 ±1.5 mm and soya beans: average Ø 5.78 ±0.38 mm.

The technological treatment of seeds was performed in two stages:

1. Soaking
   - "cold" – traditional soaking of seeds in cold-room temperature water (ratio 1:4); left for 12 hours
   - "hot" – soaking of seeds in hot water – temperature about 95°C (ratio 1:4); left for 2 hours.
2. Cooking (without sodium chloride addition) to consumption softness (the level of softness was checked in 10 min intervals and accepted by sensory panellists) with usage of:
   - electric stove with cast iron hotplate (Mastercook 5E1/0/4A type)
   - combined microwave oven (Panasonic NN--C703/C753 type)
   - induction stove with glass-ceramic hotplate (Stalglast, BT-180K type).

The ratio of water to seeds in preparation process was 4:1. The hydrothermal processes were conducted in duplicate. Information concerning timing of technological processes (soaking and cooking) and process efficiency was presented in previous paper [Filipiak--Florkiewicz et al. 2011]. The content of dietary fiber according to AOAC 985.29 [2003] (with Fibertec by Foss), and phenolic compounds with Folin-Ciocialteu reagent by Singleton et al. [1999], were assessed. Two-stage, methanol-acetone, extraction was performed. First, 1 g of the powdered sample was extracted with 40 cm^3 of HCl (0.16 $mol \cdot dm^{-3}$) in 80% methanol for 2 h, gently stirring the sample in a water bath with a shaking device at a temperature of 20°C ±2°C. Then the samples were centrifuged (4000 g) and the supernatant was collected. The residue was re-extracted with 40 cm^3 of 70% acetone in the same conditions, and centrifuged as above. Both extracts were combined and stored at temperature –20°C [Korus et al. 2007]. The prepared extracts were used for phenolic content, as well as antioxidant activity determination. The results of phenolic content were expressed as gallic acid equivalents (GAE), in milligrams of gallic acid per 100 gram of the dry matter of sample, on the basis of the calibration curve ($r^2 = 0.9939$).

The antioxidative activity measured as ability of scavenging of ABTS free radical was performed by the method of Re et al. [1999]. An aqueous solution of ABTS (2,2'-azino-bis(3-ethylbenzotialozline-6-sulfonic acid), Sigma) of 7 $mmol \cdot dm^{-3}$ was mixed with aqueous solution of potassium persulfate (2.45 $mmol \cdot dm^{-3}$) (both heated to 30°C ±0.5°C) and left for the night to produce the $ABTS^{\bullet+}$ cation-radical. The $ABTS^{\bullet+}$ solution was diluted with the methanol to obtain a working solution of 0.74-0.75 absorbance. Then, 2 cm^3 of the working solution of $ABTS^{\bullet+}$ was mixed either with 1 cm^3 of methanol (blank) or with the extract appropriately diluted with methanol (so that the radical-scavenging degree would not exceed 60%), and after 15 min, absorbance at 734 nm was measured. Based on the calibration curve ($r^2 = 0.9989$), the results of antioxidative activity were expressed as TEAC in micromoles of Trolox per gram of the dry matter of sample.

All the above mentioned analyses were performed after each stage of hydrothermal treatment (soaking and cooking – in triplicate).

Obtained results were analysed with Statistica v. 7.0. The significance of differences was estimated with post hoc Duncan test with $p < 0.05$.

RESULTS AND DISCUSSION

The highest amount of dietary fiber was found in soya bean seeds cooked with induction stove (27.4 $g \cdot 100 g^{-1}$ d.m.) and electric stove (27.3 $g \cdot 100 g^{-1}$ d.m.) after previous soaking in hot water (Table 1). In other treatments no significant changes of that component were observed in comparison to raw seeds. Interesting results concerning content of dietary fiber were noticed for kidney bean seeds. It was shown that dry seeds, soaked and then cooked (despite the technique) had comparable amounts of that component, whereas application of traditional soaking caused significant (almost doubled) lowering of dietary fiber level. It could be caused by changes of starch (presence of resistant starch – RS) caused by high temperature activity during "hot" soaking what influenced growth of dietary fiber level. RS has been categorized as RS_1, RS_2, RS_3 and RS_4. The processes used in our investigations caused, most probably, the origin of RS_3. This kind of starch – retrograded or recrystallized- is found in most of the heat processed and cooled foods [Yadav et al. 2010].

The dry soya bean and kidney bean were characterised by high content of phenolic compounds 163.2 mg $GAE \cdot 100 g^{-1}$ d.m. and 173.1 mg $GAE \cdot 100 g^{-1}$ d.m. respectively. Borowski et al. [2008] showed the content of phenolic compounds in raw kidney beans equalled 50.52 mg of galic acid (GAE) per 100 of product. According to the authors the lower content of phenolic compounds was found also in bean seeds – 34.08 mg $GAE \cdot 100 g^{-1}$ of seeds. The lower amount of assessed compounds (36.84 mg of catechin per 100 g) was found also in green bean examined by Melo et al. [2006]. Remiszewski et al. [2006] examined the seeds of John Tall kidney and John Dwarf kidney and found that content of phenolic compounds was 107 mg and 108 mg of $GEA \cdot 100 g^{-1}$ d.m., respectively. A bit lower level of those compounds equalled 86 mg $GEA \cdot 100 g^{-1}$ d.m. the authors noticed for multiflower John Dwarf kidney [Remiszewski et al. 2007]. According to research of Bieżanowska-Kopeć and Pisulewski [2006] the average content of phenolic compounds in ordinary bean with white colour of seed integument was 165 mg of $catechin \cdot 100 g^{-1}$ d.m.

Performed soaking processes influenced the growth of phenolic compounds in dry mass of soya bean and kidney bean (177.7-313.3 mg $GAE \cdot 100 g^{-1}$ d.m.) in comparison to dry seeds (respectively 163.2 and 173.1 mg $GAE \cdot 100 g^{-1}$ d.m.; Table 1, 2). Many literature data show that technological processes influence

Table 1. Influence of hydrothermal process on dietary fiber and phenolic compounds content, as well as antioxidant activity of soya bean

Kind of proccesing	Content in 100 g of dry matter		Antioxidant activity (TEAC)
	fiber	total phenolic compound*	
	g	mg GEA	µmol Trolox·g^{-1} d.m.
Raw material (dry seeds)	21.7^{a} ±0.74	163.2^{a} ±4.74	4.1^{a} ±0.10
"Hot" soaked	25.0^{ab} ±0.11	313.3^{e} ±4.71	5.9^{b} ±0.14
"Cold" soaked	21.0^{a} ±0.68	223.0^{c} ±4.38	6.0^{bc} ±0.09
"Hot" soaked			
Traditional cooking	27.3^{b} ±0.29	249.3^{d} ±9.48	5.7^{b} ±0.06
Microwave oven cooking	21.1^{a} ±0.03	315.6^{e} ±8.67	6.2^{c} ±0.01
Induction stove cooking	27.4^{b} ±0.53	224.4^{c} ±9.27	6.5^{d} ±0.03
"Cold" soaked			
Traditional cooking	22.5^{a} ±0.28	251.1^{d} ±8.26	5.7^{b} ±0.02
Microwave oven cooking	23.1^{a} ±0.91	198.3^{b} ±4.59	6.2^{c} ±0.11
Induction stove cooking	22.6^{a} ±0.15	229.1^{cd} ±3.25	6.2^{c} ±0.06

*Total phenolic compounds value is expressed as gallic acid (GAE).
The values in the same columns denoted with different letters: a, b, c, d, e differ statistically significantly at $p \leq 0.05$.

the contents of biologically active compounds such as phenolic compounds and on antioxidative characteristics of vegetables tissues [Anton et al. 2008, Korus et al. 2007, Remiszewski et al. 2007, Siddhuraju and Becker 2007, Troszyńska et al. 2002, Xu and Chang 2008]. The dry matter of cooked kidney bean seeds was characterized by higher phenolic compounds level (232.7-311.6 mg GAE·100 g^{-1}) in comparison to dry and soaked seeds. The exception were seeds cooked in microwave oven after former "cold" soaking (Table 2).

Similar tendencies were observed for antioxidative activity. Above conclusions can be explained by fact that during different ways of treatment the proportions between individual compounds of dry matter were changing, because i.e. of migration of some easy solved substances to water [Winiarska-Mieczan and Koczmara 2006]. It could also arise because of changes in products structure which probably allowed for release of combined polyphenols from glycosidic into aglicons compounds. It caused obtaining of higher amounts of phenolic compounds or higher antioxidative activity in seeds after treatment in comparison to model material [Przeciwutleniacze... 2007]. The different tendency was observed by Boateng et al. [2008] when examining ordinary bean with mottle integument. They noticed no significant lowering of phenolic compounds content in dry matter of seeds in comparison to material not submitted to culinary treatment. Also Bieżanowska-Kopeć and Pisulewski [2006] when examining the ordinary bean with white integument observed average lowering of 23% of phenolic compounds sum in comparison to raw seeds. A similar tendency was noticed by Remiszewski et al. [2008] when estimated the influence of soaking on content of polyphenols in red ordinary bean. According Stasiak and Ulanowska [2008] traditional soaking and cooking of kidney bean seeds led to lowering of phenolic compounds, at about 4%, in comparison to raw seeds. Xu and Chang [2008] proved that steam cooking of green peas caused growth of polyphenols

Table 2. Influence of hydrothermal process on dietary fiber and phenolic compounds content, as well as antioxidant activity of kidney been seeds

Kind of proccesing	Content in 100 g of dry matter		Antioxidant activity (TEAC)
	fiber	total phenolic compound*	
	g	mg GEA	mmol Trolox·g^{-1} d.m.
Raw material (dry seeds)	23.0^b ±0.12	173.1^a ±6.93	4.5^b ±0.02
"Hot" soaked	22.02^b ±0.31	177.7^a ±5.68	9.5^h ±0.13
"Cold" soaked			
"Hot" soaked	14.10^a ±0.68	178.3^a ±3.42	8.7^g ±0.09
Traditional cooking	22.0^b ±0.23	274.7^c ±1.29	4.8^c ±0.05
Microwave oven cooking	23.8^b ±1.06	232.7^b ±3.51	3.2^a ±0.06
Induction stove cooking	22.7^b ±0.96	311.6^d ±2.59	6.3^e ±0.02
"Cold" soaked			
Traditional cooking	15.7^a ±0.56	248.8^b ±7.01	6.8^f ±0.10
Microwave oven cooking	12.6^a ±0.29	171.4^a ±2.37	3.4^a ±0.02
Induction stove cooking	12.9^a ±0.36	268.4^c ±5.21	5.6^d ±0.06

*Total phenolic compounds value is expressed as gallic acid (GAE).
The values in the same columns denoted with different letters: a, b, c, d, e, f, g, h differ statistically significantly at $p \leq 0.05$.

in dry matter equalled 48%. Similar conclusions obtained Remiszewski et al. [2007] submitting the Dwarf John beans to extrusion. The same tendency was observed by Turkmen et al. [2005] examining green (not matured) bean. They noticed 14% growth of share of phenolic compounds in dry matter of examined material after its cooking when comparing to raw seeds. Boateng et al. [2008] proved that toasting of ordinary bean seeds caused 18% growth of share of the examined compounds.

Antioxidative activity of untreated seeds of kidney bean and soya bean was respectively 4.5 and 4.1 µmol Tx·g^{-1} d.m. The lowest antioxidative activity, equalled 2.38 µmol Tx·g^{-1} d.m. was found in bean seeds examined by Hunter and Fletcher [2002]. The higher ability of free radicals scavenging in multiflower Dwarf John bean was measured by Remiszewski et al. [2006, 2007] amounting respectively 8.19 and 11.48 µmol Tx·g^{-1} d.m. Also much higher results were obtained by Gumienna et al. [2007] in red bean seeds. The antioxidative activity of the examined vegetable was almost 20 µmol Tx·g^{-1} d.m. Such a significant difference in ability of scavenging of free radicals by seeds was caused by the seeds colours, since as had been proved in many experiments [Boateng et al. 2008, Remiszewski et al. 2007, Stasiak and Ulanowska 2008], the kinds with lighter seed's integument were characterised by lower antioxidative activity. Soaking of seeds (both "cold" and "hot" method) caused the growth of antioxidative activity of dry matter in comparison to raw material, whereas after their cooking the activity decreased. High ability of free radicals scavenging was observed in kidney bean seeds – 8.7 µmol Tx·g^{-1} d.m. after traditional soaking and 9.5 µmol Tx·g^{-1} d.m. after shortened soaking and they had grown significantly of 93.3% and of 111.1% respectively. Statistical analysis showed that differences in antioxidative activity of soya bean seeds subjected to different methods of soaking were also significant. Stasiak and Ulanowska [2008] obtained different results. The ability of free radicals scavenging with the examined white ordinary bean seeds was lowered of 37%. In examination of Remiszewski et al. [2008] 35% lowering of that parameter was shown. The authors examined red ordinary bean seeds. Other conclusions were presented by Bieżanowska-Kopeć and Pisulewski [2006]. Those researchers obtained 6% loss of antioxidative activity of extracts gathered from white bean Longina, after their soaking with shortened "hot" method, in comparison to raw seeds. Whereas, after cooking the growth of 30% of antioxidative activity was observed [Bieżanowska-Kopeć and Pisulewski 2006].

Cooking of kidney bean seeds with induction and electrical stoves caused lowering of antioxidative activity losses (independently the soaking method) in comparison to microwaves cooking where the bases of comparison were the soaked seeds. In Boateng et al. [2008] report the traditional soaking also favoured the growth of that parameter of 11% whereas the subsequent toasting of seeds caused 13% loss of antioxidative activity in comparison to raw material.

CONCLUSIONS

1. Dry matter of soya bean and kidney bean were characterised by high levels of dietary fiber, phenolic compounds and antioxidative activity.
2. Soaking and cooking of seeds (independently the species) influenced on growth of phenolic compounds.
3. Soaking of soya bean and kidney bean seeds with "hot" method was characterized by lowest losses of non-nutritive compounds and of antioxidative activity.
4. Cooking of soya bean and kidney bean with electrical or induction stove favoured preservation of dietary fiber, phenolic compounds and antioxidative activity of seeds in comparison to thermal treatment conducted with combined microwave oven.

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WPŁYW OBRÓBKI HYDROTERMICZNEJ NA ZAWARTOŚĆ BŁONNIKA POKARMOWEGO I ZWIĄZKÓW FENOLOWYCH ORAZ AKTYWNOŚĆ ANTYOKSYDACYJNĄ NASION ROŚLIN STRĄCZKOWYCH

STRESZCZENIE

Wprowadzenie. Suche nasiona roślin strączkowych należą do produktów o dużej wartości żywieniowej. Oprócz białka o wysokiej wartości biologicznej, zawierają niezbędne nienasycone kwasy tłuszczowe, witaminy z grupy B, błonnik pokarmowy i przeciwutleniacze. Celem badań było określenie wpływu różnych metod moczenia i gotowania na zawartość błonnika pokarmowego i związków fenolowych oraz aktywność antyoksydacyjną nasion roślin strączkowych.
Materiał i metodyka. Materiałem badawczym były suche nasiona fasoli (odmiana 'Piękny Jaś') pochodzące z kolekcji Krakowskiej Hodowli i Nasiennictwa Ogrodniczego POLAN S.A. oraz nasiona soi zakupione w handlu detalicznym. Obróbka technologiczna nasion przebiegała w dwóch etapach: 1) moczenie na zimno i na gorąco, 2) gotowanie z wykorzystaniem trzech różnych urządzeń: trzonu elektrycznego, indukcyjnego i aparatu mikrofalowego. W materiale badawczym oznaczono zawartość błonnika pokarmowego, związków fenolowych oraz aktywność antyoksydacyjną.
Wyniki. Największą zawartość błonnika pokarmowego stwierdzono w nasionach soi oraz fasoli gotowanych na trzonie indukcyjnym (27,4 $g \cdot 100\ g^{-1}$ s.m.) i elektrycznym (27,3 $g \cdot 100\ g^{-1}$ s.m.) po uprzednim namoczeniu ich w wodzie gorącej. Suche nasiona soi i fasoli charakteryzowały się dużą zawartością związków fenolowych, odpowiednio 163,2 mg $GAE \cdot 100\ g^{-1}$ s.m. i 173,1 mg $GAE \cdot 100\ g^{-1}$ s.m. Sucha masa gotowanych (trzon indukcyjny i elektryczny) nasion fasoli i soi odznaczała się wyższym poziomem związków fenolowych (232,7-311,6 mg $GAE \cdot 100\ g^{-1}$ i 224,4-315,6 mg $GAE \cdot 100\ g^{-1}$) w porównaniu z nasionami suchymi. Aktywność antyoksydacyjna suchych nasion fasoli i soi wynosiła odpowiednio 4,5 i 4,1 $\mu mol\ Tx \cdot g^{-1}$ s.m. Największą zdolnością wygaszania wolnych rodników odznaczały się nasiona fasoli, np. 8,7 $\mu mol\ Tx \cdot g^{-1}$ s.m. (moczone metodą tradycyjną) i 9,5 $\mu mol\ Tx \cdot g^{-1}$ s.m. (moczone w wodzie gorącej).
Wnioski. Pod wpływem moczenia oraz gotowania nasion (niezależnie od gatunku) obserwowano wzrost udziału związków fenolowych w suchej masie. Korzystniejszym sposobem okazało się moczenie nasion fasoli metodą na gorąco. Sposób ten przede wszystkim wpływał na znaczne skrócenie całego procesu. Gotowanie nasion soi oraz fasoli na trzonie elektrycznym i indukcyjnym sprzyjało lepszemu zachowaniu włókna pokarmowego, związków fenolowych oraz aktywności antyoksydacyjnej nasion w porównaniu z obróbką termiczną prowadzoną w kombinowanym urządzeniu mikrofalowym.

Słowa kluczowe: soja, fasola, moczenie, gotowanie, kuchnia indukcyjna, błonnik pokarmowy, związki fenolowe, aktywność antyoksydacyjna

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