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*original scientific paper*

# Effect of Extrusion on the Phenolic Composition and Antioxidant Activity of Dry Beans of *Phaseolus vulgaris* L.

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**Summary**

The phenolic composition of dry beans and extrudates was investigated to evaluate the effect of extrusion process on their antioxidant activity. Myricetin, quercetin, kaempferol, cyanidin, chlorogenic acid, caffeic acid, ferulic acid and *p*-coumaric acid were identified in raw and processed bean seeds. The effect of extrusion on the total phenolic content of beans depended on the cultivar – one variety showed a 14 % increase in the amount of phenolics in extrudates compared to raw beans, while the other two exhibited a decrease by 19 and 21 %. Extracts from bean extrudates showed a faster initial free radical scavenging activity than the extracts from raw beans; however, the final values were similar. The least active extrudates were obtained by using the initial moisture of 20 % and the temperature of 180 °C. Extrusion also decreased the antioxidant activity, compared to the raw material.

*Key words*: bean, *Phaseolus vulgaris*, extrusion, antioxidant activity, scavenging activity

## Introduction

Natural antioxidants occurring in food products play a double role: first, they preserve the foods, and second, they affect human health through scavenging free radicals, chelating the metal ions that catalyse oxidation reactions, inhibiting the activity of oxidant enzymes, and activating antioxidant enzymes (*1–5*). Those who consume foods rich in antioxidants are less prone to cardiovascular diseases and various forms of cancer, and the rate of degenerative changes in their organisms due to ageing is slower (*3,6*). Among natural antioxidants, phenolics form the largest group. Such compounds are secondary metabolites synthesised solely in plants. Phenolics comprise several groups of compounds differing in structure: phenolic acids (benzoic acids and hydroxycinnamic acids), flavonoids (flavonols, flavones, flavanols, isoflavones), stilbenes and tannins (*3,7–9*).

The raw materials that are rich in antioxidants include dry beans of *Phaseolus vulgaris* L. which, among other components, contain flavonoids and phenolic acids (*10–14*). A high consumption of beans is believed to reduce the incidence of cardiac diseases, diabetes and colon cancer, to mention a few (*13,15*). In the course of food processing, however, phenolics may undergo various changes, thus altering the antioxidant activity of the products (*3,16*).

The present study attempts to describe the phenolic composition of *Phaseolus vulgaris* L. dry beans and extrudates (which may be eaten as snacks) in qualitative and quantitative terms, and to establish the effect of the extrusion process on their antioxidant activity.

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## Material and Methods

Dry beans of three Polish cultivars of *Phaseolus vulgaris* L., differing in the seed coat colour: Rawela (dark--red), Tip-Top (black-brown) and Toffi (cream; ZHiNO PlantiCo Szymanów, Poland) were grown in year 2004 in the central part of the country. The beans were pulverised in a laboratory mill Pulverisette 14 (Fritsch, Idar--Oberstein, Germany), sieved through a 0.5-mm sieve and moistened to the 14 or 20 % moisture content. Extrusion was performed using a 20DN single-screw extruder (Brabender, Duisburg, Germany). The extrusion conditions were: the screw speed was constant at 90 rpm, screw compression 3:1, round die hole 3 mm, feeding screw speed 70 rpm. Two temperature profiles, 80/100/ 120 °C and 120/160/180 °C, were employed. The extrudates were cooled at room temperature, pulverised and sieved as described above.

### *Preparation of extracts*

Two-stage methanol-acetone extraction was conducted. One gram of the sample was extracted for 2 h using 40 mL of 0.16 M HCl in 80 % methanol (by volume) with gentle stirring in a shaker equipped with a water bath of (20±2) °C. Then the samples were centrifuged (4000 × *g*), the supernatant was collected, and the residue was reextracted under the same conditions, using 40 mL of 70 % acetone (by volume). After centrifuging (as above), both extracts were combined and stored at a temperature of –20 °C until they were used.

### *HPLC analysis of phenolic compounds after enzymatic hydrolysis*

#### Preparation of samples

A mixture of enzymes (5 mg of each, Drum pectinase 263 from Seclin, France; β-glucosidase, hesperidinase and sulphatase type H-2 from Sigma) dissolved in 5 mL of citrate buffer with pH=5.5 was added to 0.5 g of the sample. The whole was incubated for 1 h in a water bath at the temperature of 40 °C and then left for 20 h in the dark chamber for enzymatic hydrolysis. After that, 5 mL of pure methanol were added to the samples which were then placed in an ultrasonic bath for 10 min. After centrifuging in a laboratory centrifuge (10 000 × *g*), the samples were subjected to HPLC analysis.

#### Methods

Phenolic compounds were determined by a high-performance liquid chromatography (HPLC) using a liquid chromatograph equipped with a Merck-Hitachi L-7455 diode array detector (DAD) working in tandem with an L-7100 pump and a D-7000 HSM Multisolvent Delivery System (Merck-Hitachi, Tokyo, Japan) to mix the reagents. The separation was performed on a LiChroCART® 125-3 Purospher® RP-18 (5 μm) column (Merck) thermostated at a temperature of 30 °C. An 80 % solution of acetonitrile in 4.5 % formic acid (reagent A), and 2.5 % acetic acid (reagent B) were used as eluents. The flow rate was set at 1 mL/min, and the solvent gradient was used as follows: the proportion of reagent A was increased from 0 to 15 % in 7 min, then to 20 % in further 8 min. A column clean-up stage was used by passing reagent A only for a further 1 min, followed by final re-equilibration for 10 min only with reagent B. During the analysis, the solutions were degassed in an ultrasonic bath (Merck).

The recording was conducted at the following wavelengths ($\lambda$): phenolic acids 320 nm, flavones 340 nm, flavonols 360 nm, and anthocyanins 520 nm. The compounds were identified on the basis of the spectra in the range from 200 to 600 nm and the retention times were compared to standards. Recoveries from samples were measured by spiking prior to analysis the bean samples in extraction solution with pure *p*-coumaric acid at the level of 50–100 % of the measured content. Recoveries were determined in triplicate. Recoveries of the *p*-coumaric acid standard from bean samples were 85–90 %. The results were expressed in mg per 100 g of sample on a dry matter basis.

### *Electron Paramagnetic Resonance (EPR) measurement of antioxidant activity*

Measurements were made on a home-made EPR L-band (1.2 GHz) spectrometer constructed in cooperation with Dartmouth Medical School, Hanover, New Hampshire, USA, equipped with a surface-coil resonator tunable in the range of modulation frequency ±12 MHz by means of a varactor diode in tandem with an automatic frequency control (AFC). The conditions of spectrometry were as follows: maximum power of microwaves 16 mW, modulation amplitude 2.2 G, time constant of the phase discriminator 20 ms, field modulation frequency 33 kHz. The magnetic field scan was adjusted to 100 G. Antioxidant activity was determined using DPPH (2,2-diphenyl-1-picrylhydrazyl Sigma), which is a stable free radical. The volume of 0.250 mL of the extract was added to 0.750 mL of 33 mM DPPH methanol solution. The time of scan was set at 20 s, and each spectrum was the average of four individual scans. Measurements were performed in duplicates at room temperature. All spectra were gathered, stored and manipulated using a specially developed PC software that was designed at the EPR Center of Dartmouth Medical School, Hanover, New Hampshire, USA. The computer program for data acquisition was written in the *C* programming language. It collects and stores up to 32 arrays of 1024–4096 data points, which contain the observed spectra. Further details are given by Walczak *et al.* (*17*).

### *Determination of antioxidant activity in the β-carotene/linoleic acid system*

Antioxidant activity by oxidation of the β-carotene/linoleic acid system was determined using the method of Al-Saikhan *et al.* (*18*). In brief, 4 mg of β-carotene (Sigma) were dissolved in 40 mg of chloroform, then 3 mL of linoleic acid (Fluka) and 400 mg of Tween 40 (Sigma) were added. The chloroform was evaporated under reduced pressure (vacuum pump PL2, AGA Labor, Poland) at a temperature of (45±2) °C while heating the sample in a water bath. After that, 100 mL of 10 % $H_2O_2$ (by volume) were added to the obtained emulsion, 3 mL of it were taken, and 0.12 mL of the extract were added. The samples were placed in a water bath with a temperature of (50±2) °C, and absorbance was measured

at a wavelength of 470 nm (Helios γ spectrophotometer, Thermo Electron Corp., UK) at 10-minute intervals. The blank contained 0.12 mL of distilled water instead of the extract. The degradation rate of β-carotene and the antioxidant activity (AA) were calculated according to Al-Saikhan *et al.* (*18*), and the oxidation rate ratio (ORR) and the antioxidant activity coefficient (AAC) were computed following Oomah *et al.* (*13*).

### *Statistical analysis*

The results were analysed using Snedecor's F-test and Student's *t*-test. The least significant difference (LSD) was computed at p=0.01.

## Results and Discussion

Among the three cultivars of *Phaseolus vulgaris* L., Rawela contained the highest amount of identified phenolics, 90.28 mg per 100 g of dry mass of sample (some of the peaks were not classified). The values for the other cultivars, 35.61 mg/100 g for Tip-Top and 30.48 mg/100 g for Toffi, were significantly lower (Table 1). The bean cultivars differed significantly in the kaempferol content, which was 50.20 mg in Rawela, 3.24 mg in Tip-Top and 0.91 mg in Toffi. This last cultivar also had the smallest quercetin and caffeic acid contents and the highest ferulic acid content. The Tip-Top cultivar was the richest in chlorogenic, caffeic and *p*-coumaric acids, while Rawela had the lowest levels of chlorogenic and *p*-coumaric acids and the highest level of quercetin and kaempferol. The bean cultivars contained from 1.75 to 67.18 mg of flavonoids (identified by HPLC), and from 23.10 to 29.46 mg of phenolic acids (Table 1). For comparison, the amounts reported by Heimler *et al.* (*1*) were 3–127 mg of flavonoids and 2–9 mg of phenolic acids per 100 g of beans in the common bean cultivars they studied. The levels of flavonoids established by Oomah *et al.* (*13*) in various bean cultivars were 41–102 mg/100 g, and those found by Romani *et al.* (*14*) in four cultivars were 30–71 mg/100 g. Luthria and Pastor-Corrales (*19*) noted 19.1 to 48.3 mg of phenolic acids in fifteen bean cultivars commonly consumed in the United States.

Processing is usually thought to cause substantial losses of antioxidants in food products. Some authors, however, have observed its positive influence on antioxidant levels (*3,16*). In the present study, the effect of extrusion on the total phenolic content of beans depended on the cultivar. Rawela showed a 14 % increase in the amount of phenolics in extrudates compared to raw beans, while Tip-Top and Toffi exhibited a decrease by 19 and 21 %, respectively. Cultivar also affected significantly the degree of retention of the studied phenolic compounds. In Rawela, only the levels of chlorogenic and caffeic acids fell on average by 33 and 9 %, respectively, whereas those of the other compounds increased due to extrusion, with the rise being the greatest for quercetin (by 84 %) and ferulic acid (by 40 %). The Tip-Top cultivar showed an increase solely in ferulic acid, by 10 % on average, and the Toffi cultivar, in caffeic acid, by 17 %. The levels of the other phenolics decreased in both cultivars. The losses were the greatest (on average) for myricetin, by 80 % in Tip-Top, quercetin, by 50 % in Tip-Top and by 49 % in Toffi, chlorogenic acid, by 33 % in Rawela and by 31 % in Toffi, and kaempferol, by 32 % in Tip-Top.

The conditions of extrusion had an unclear effect on the levels of antioxidants. Considering the total amount of the identified phenolic compounds, it can be seen, however, that the beans of all three cultivars extruded at a lower temperature, *i.e.* 120 °C, retained a higher amount of phenolics in total than those extruded at 180 °C. At both extrusion temperatures, smaller losses were noted in samples of higher initial moisture (except for Tip-Top beans extruded at 120 °C), although the effect produced by this parameter on individual phenolics varied greatly, suggesting that water probably protects phenolic compounds during extrusion. A beneficial effect of a higher moisture content of the raw material on the degree of retention of chemical compounds was observed by Ismail and Zahran (*20*) and Korus *et al.* (*21*).

The initial rate of the DPPH radical scavenging was the highest for Tip-Top (reduction by 33 % within 5 min and by 52 % within 10 min); the values for Toffi and Rawela were lower (respectively, 26 and 11 % within 5 min, 49 and 38 % within 10 min, Fig. 1). After 40 min, the kinetics of antiradical activity was similar in all cultivars: the free radical inhibition by the extracts was 92 % for Rawela, 96 % for Tip-Top and 90 % for Toffi. After 60 min, the respective values increased to 97, 99 and 94 %.

Antiradical activity depends not only on the amount, but also on the kind of free radical scavengers present in the material. Such a dependence was evidenced by Oomah *et al.* (*13*): in their research, the dark-red bean cultivar ROG 802, containing the highest amount of phenolics among the cultivars they examined (16.6 *vs.* 3.3–8.5 mg/g), showed the lowest DPPH radical scavenging degree. Karamać *et al.* (*22*) established the DPPH radical scavenging power of individual phenolic acids. Taking the power of gallic acid for 100 %, they obtained figures of 49.58 % for caffeic acid, 25.57 % for ferulic acid and as little as 0.04 % for *p*-coumaric acid. Similar proportions between the activities of the three latter acids had earlier been noted by Gadow *et al.* (*23*). In the varieties Tip-Top and Toffi the dominating poliphenols were phenolic acids, while in Rawela they were flavonoids (Table 1). The most active cultivar, Tip-Top, contained a considerably greater amount of caffeic acid, a strong scavenger, than Rawela and Toffi (by 49 and 55 %, respectively). Toffi, in turn, had the highest ferulic acid content (46 and 17 % higher than Rawela and Tip-Top, respectively). On the other hand, Rawela contained approx. 24 times more kaempferol than the other two cultivars and approx. 14 times more quercetin, which compares to caffeic acid in the DPPH radical scavenging power (*23*). A similar lack of correlation between the DPPH radical scavenging power, and the flavonoid and phenolic acid contents was observed by Heimler *et al.* (*1*). As was mentioned above, some of the peaks were not identified, thus they were not calculated into total phenolics. Additionaly, the contribution to the antiradical and antioxidant capacities is also made by other components contained in the beans, which is not covered in the present study, such as by procyanidins, which show antiradical activity, and, to a smaller extent, tocopherol, proteins and aroma compounds (*13,24–27*).

Table 1. Contents of identified phenolic compounds in raw beans and extrudates of three *Phaseolus vulgaris* L. cultivars

| Conditions of extrusion* | *w*(myricetin) mg/100 g | | *w*(quercetin) mg/100 g | | *w*(kaempferol) mg/100 g | | *w*(chlorogenic acid) mg/100 g | | *w*(caffeic acid) mg/100 g | | *w*(ferulic acid) mg/100 g | | *w*(*p*-coumaric acid) mg/100 g | | *w*(total) mg/100 g | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | A** | | A** | | A** | | A** | | A** | | A** | | A** | | A** |
| **Rawela** | | | | | | | | | | | | | | | | |
| Raw material | 0 | – | 16.98 | – | 50.20 | – | 17.31 | – | 0.49 | – | 4.41 | – | 0.89 | – | 90.28 | – |
| 14/120 | 0 | – | 30.00 | 77 | 51.36 | 2 | 11.72 | –32 | 0.53 | 9 | 6.57 | 49 | 1.40 | 57 | 101.59 | 13 |
| 20/120 | 0 | – | 36.35 | 114 | 58.91 | 17 | 12.69 | –27 | 0.49 | 0 | 5.79 | 31 | 1.00 | 12 | 115.23 | 28 |
| 14/180 | 0 | – | 27.76 | 63 | 47.08 | –6 | 10.40 | –40 | 0.45 | –9 | 6.22 | 41 | 0.93 | 4 | 92.83 | 3 |
| 20/180 | 0 | – | 30.75 | 81 | 51.20 | 2 | 11.76 | –32 | 0.32 | –35 | 6.16 | 40 | 0.90 | 1 | 101.09 | 12 |
| LSD (p=0.01) | – | – | 1.311 | – | 1.210 | – | 1.115 | – | 0.111 | – | 0.862 | – | 0.137 | – | 4.185 | – |
| **Tip–Top** | | | | | | | | | | | | | | | | |
| Raw material | 1.37 | – | 1.53 | – | 3.24 | – | 19.20 | – | 0.73 | – | 5.51 | – | 4.02 | – | 35.61 | – |
| 14/120 | 0.33 | –76 | 1.25 | –18 | 2.76 | –15 | 16.33 | –15 | 0.71 | –3 | 6.73 | 22 | 3.61 | –10 | 31.72 | –11 |
| 20/120 | 0.23 | –83 | 0.50 | –67 | 2.20 | –32 | 16.70 | –13 | 0.75 | 3 | 6.00 | 9 | 3.69 | –8 | 30.08 | –16 |
| 14/180 | 0.31 | –77 | 0.83 | –46 | 1.82 | –44 | 12.94 | –33 | 0.62 | –15 | 5.28 | –4 | 3.28 | –18 | 25.08 | –30 |
| 20/180 | 0.23 | –83 | 0.47 | –69 | 2.08 | –36 | 15.15 | –21 | 0.69 | –6 | 6.27 | 14 | 3.59 | –11 | 28.48 | –20 |
| LSD (p=0.01) | 0.448 | – | 0.266 | – | 0.653 | – | 1.311 | – | n.s. | – | n.s. | – | 0.120 | – | 3.619 | – |
| **Toffi** | | | | | | | | | | | | | | | | |
| Raw material | 0 | – | 0.84 | – | 0.91 | – | 17.87 | – | 0.47 | – | 6.44 | – | 3.95 | – | 30.48 | – |
| 14/120 | 0 | – | 0.19 | –77 | 0.74 | –19 | 12.41 | –31 | 0.52 | 10 | 6.44 | 0 | 3.46 | –12 | 23.76 | –22 |
| 20/120 | 0 | – | 0.49 | –42 | 0.59 | –35 | 15.58 | –13 | 0.65 | 39 | 6.67 | 3 | 3.92 | –1 | 27.89 | –8 |
| 14/180 | 0 | – | 0.59 | –29 | 1.00 | 11 | 7.83 | –56 | 0.54 | 15 | 5.96 | –8 | 2.87 | –27 | 18.80 | –38 |
| 20/180 | 0 | – | 0.45 | –47 | 0.64 | –29 | 14.03 | –21 | 0.49 | 4 | 6.59 | 2 | 3.84 | –3 | 26.04 | –15 |
| LSD (p=0.01) | – | – | 0.125 | – | 0.236 | – | 1.215 | – | n.s. | – | n.s. | – | 0.132 | – | 2.298 | – |
| Total LSD (p=0.01) | – | – | 2.372 | – | 3.160 | – | 2.665 | – | 0.231 | – | 0.425 | – | 0.383 | – | 6.789 | – |

*14, 20 – *w*(initial humidity of extruded materials)/%; 120, 180 – final temperature of extrusion/°C
**A – change relative to raw material/%
n.s. – not significant

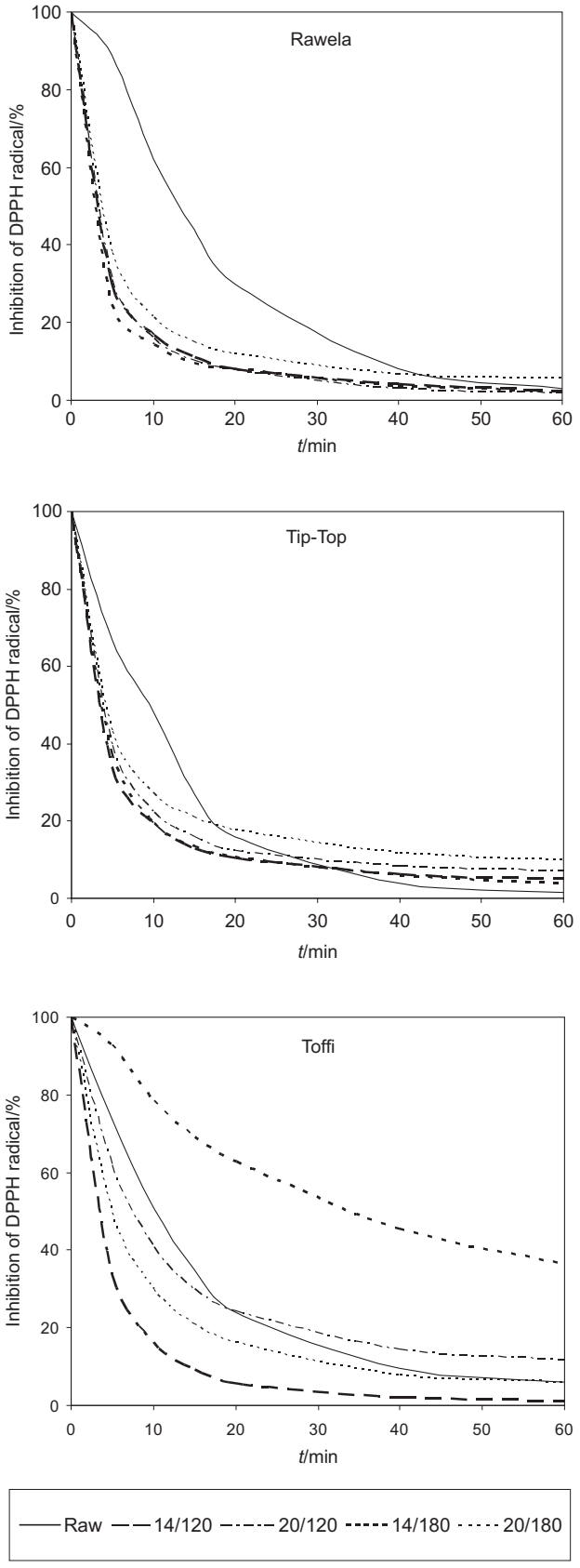


**Fig. 1.** Dynamics of DPPH radical scavenging by extracts from raw beans and extrudates of three *Phaseolus vulgaris* L. cultivars (14, 20 – initial humidity of extruded materials/%; 120, 180 – final temperature of extrusion/°C)

The picture of the DPPH radical scavenging dynamics indicates that Rawela contained the least active scavengers of free radicals (slow initial kinetics) among the bean cultivars studied. However, owing to the high total content of such compounds, it was possible to monitor antiradical activity in a longer time and observe that the kinetics levelled out after approx. 40 min, and the percent inhibition of the free radicals after approx. 60 min was higher for Rawela than for Toffi. On the other hand, Oomah *et al.* (*13*) suggest that the differences in the antiradical activity of various bean cultivars may arise from the amount and composition of procyanidins, which exhibit a great free radical scavenging power.

Extracts from bean extrudates showed a faster initial free radical scavenging activity than the extracts from raw beans (Fig. 1). This probably resulted from the destruction of cell walls in the hydrothermal process and the release of compounds that have antiradical potential. There were no significant differences in the effects of extrusion parameters on the antiradical activity of extrudates in the Rawela cultivar, either in terms of the DPPH scavenging rate or in the final degree of free radicals inhibition after 60 min, except for the extrudate obtained at 20 % initial moisture and 180 °C temperature, which displayed a lower degree of final inhibition (Fig. 1, Table 2). The latter observation was also true for the Tip-Top cultivar (Fig. 1, Table 2). In this cultivar, the DPPH inhibition was the highest for the extract from raw beans, suggesting that the free radical scavengers in extrudates were depleted faster. The Toffi cultivar showed a wider scatter of results (Fig. 1, Table 2). The dynamics of the DPPH radical scavenging reaction considerably varied among extrudates from the very beginning; after 5 min, the degree of inhibition ranged from 7.2 % (14 %/180 °C extrudate) to 66.1 % (14 %/120 °C extrudate). The extrudate produced at 14 % moisture and 180 °C temperature deserves closer attention as one exhibiting the lowest antiradical activity both in terms of scavenging dynamics and of the final degree of inhibition. This may result from the smallest total phenolic content, 18 mg/100 g (Table 1). The results indicate that the least active extrudates were obtained by using the initial moisture of 20 % and the temperature of 180 °C.

The results of the antioxidant activity of bean samples largely confirm the patterns revealed in the studies of antiradical activity (Fig. 2, Table 2). Antioxidant activity (AA) was the lowest in the Rawela cultivar, while the other two cultivars showed a similar activity both in terms of dynamics and of final value. This could be a result of poliphenol composition, as mentioned above. In Rawela there was prevalence of flavonoids, and in the other varieties of phenolic acids (Table 1). In contrast, Oomah *et al.* (*13*) found that the ROG 802 cultivar, with the highest total phenolic and flavonol content, exhibited the strongest antioxidant activity. The results of our other studies (unpublished), however, confirmed the poorest antioxidant activity of Rawela, as determined by the FRAP method.

As with antiradical activity, extrusion parameters did not have an unequivocal effect on the antioxidant activity of beans (Table 2). However, it should be stressed that, in fact, extrusion decreased antioxidant activity, compared to the raw material. This suggests that the

Table 2. Comparison of antiradical activity (expressed as EPR and TEAC) and antioxidant activity (AA, AAC, ORR) of raw beans and extrudates of three *Phaseolus vulgaris* L. cultivars

| Conditions of extrusion[1] | Rawela | | | | | Tip-Top | | | | | Toffi | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | EPR[2] | TEAC[3] | AA[4] | AAC[5] | ORR[6] | EPR[2] | TEAC[3] | AA[4] | AAC[5] | ORR[6] | EPR[2] | TEAC[3] | AA[4] | AAC[5] | ORR[6] |
| Raw material | 97.0b | 77.94b | 42.7d | 335d | 0.573a | 98.6e | 79.22e | 59.4b | 416d | 0.406a | 94.0c | 75.53c | 60.4d | 435a | 0.396a |
| 14/120 | 97.6bc | 78.42bc | 34.4b | 177a | 0.656bc | 95.0c | 76.33c | 53.1a | 329b | 0.469b | 98.9d | 79.46d | 43.8a | 325a | 0.563c |
| 20/120 | 98.3c | 78.98c | 38.5c | 293c | 0.615ab | 92.8b | 74.56b | 53.4a | 282a | 0.466b | 88.4b | 71.03b | 57.3c | 410a | 0.427b |
| 14/180 | 97.6bc | 78.42bc | 37.5bc | 224b | 0.625ab | 96.2d | 77.30d | 53.4a | 338b | 0.466b | 63.6a | 51.10a | 51.5b | 341a | 0.485b |
| 20/180 | 94.2a | 75.69a | 28.1a | 181a | 0.719c | 90.1a | 72.39a | 51.9a | 350c | 0.481b | 93.9c | 75.45c | 54.2b | 336a | 0.458b |

[1]14, 20 – *w*(initial humidity of extruded materials)/%; 120, 180 – final temperature of extrusion/°C
[2]EPR – inhibition of DPPH free radical/% after *t*/min=60
[3]TEAC – Trolox Equivalent Antioxidant Capacity, μM Trolox/g sample after *t*/min=60
[4]AA - antioxidant activity after *t*/min=40
[5]AAC - antioxidant activity coefficient after *t*/min=40
[6]ORR - oxidation rate ratio after *t*/min=40
Means in each column with different letter differ significantly at p=0.01

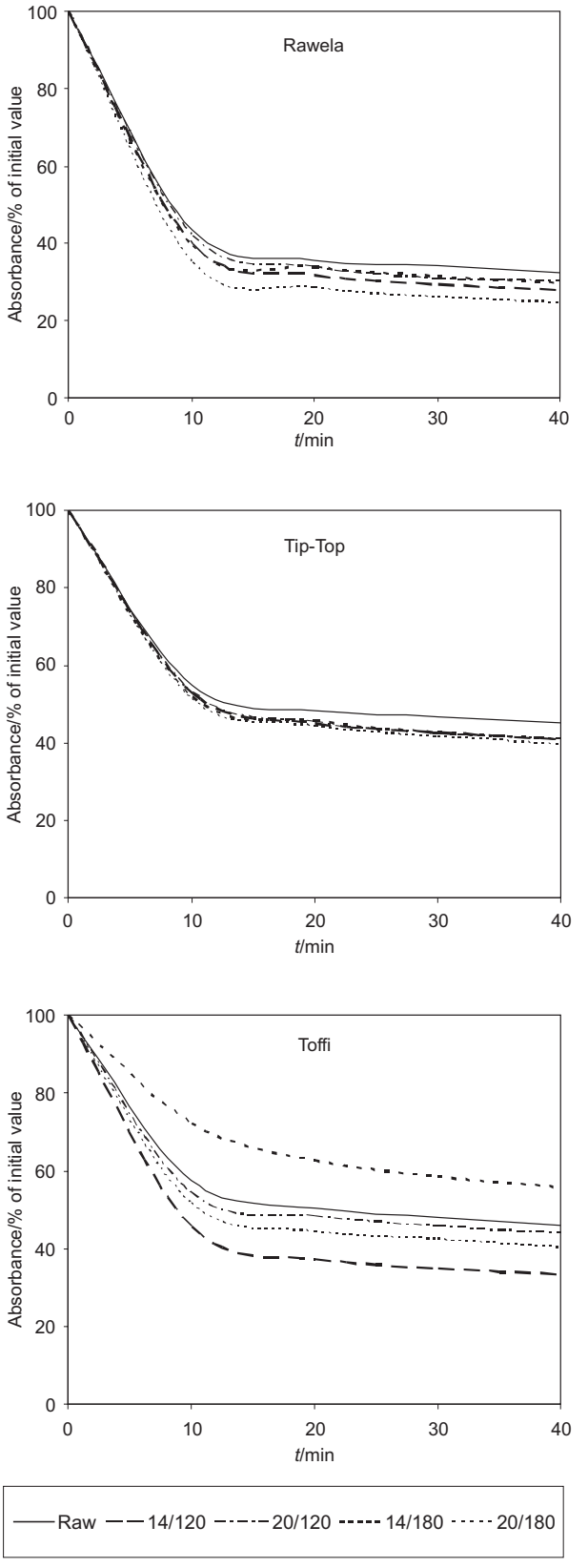


**Fig. 2.** Antioxidant activity of extracts from raw beans and extrudates of three *Phaseolus vulgaris* L. cultivars, as measured by dynamics of changes in β-carotene colour (14, 20 – initial humidity of extruded materials/%; 120, 180 – final temperature of extrusion/°C)

compounds responsible for this feature are more thermolabile than the components displaying antiradical activity. In the Tip-Top cultivar, the dynamics of β-carotene decomposition was similar for all extrudates, whereas in Toffi it varied most widely. Compared to antiradical activity, the antioxidant activity of extrudates showed a different pattern: the extrudate produced at 14 % moisture and 180 °C temperature was the most active (the lowest degree of β-carotene decomposition), while that obtained at 14 % moisture and 120 °C temperature exhibited the weakest activity.

The antiradical and antioxidant activities of beans depend on the amount and composition of the antioxidants they contain. The research conducted by Oomah *et al.* (*13*) with Canadian bean cultivars revealed differences between the cultivars in antioxidant and antiradical activities: some of them displayed high antioxidant activity but low antiradical activity, others were highly active in both respects, and one cultivar had a very low antioxidant activity and a very high antiradical activity. Similar observations were made by Cardador-Martínez *et al.* (*11*). The results of the present study also suggest that the antioxidant activity of extrudates was affected by cultivar rather than by extrusion parameters since within a cultivar the extrudates in most cases did not differ significantly in this respect.

When considering the role of antioxidants in foods and the influence of such compounds on the human organism, it seems that one should distinguish between antiradical and antioxidant activity because the same product may show a correlation between those two features ranging from strongly positive to strongly negative. Such a situation is connected with the specific phenolic composition of the products and the different antiradical and antioxidant activities of individual compounds, for example, caffeic acid makes a strong free radical scavenger but a weak antioxidant (*23*).

## Conclusions

Cultivar seems to produce a stronger effect on the antiradical/antioxidant activity of a product than the parameters of the hydrothermal process; the latter in most cases were insignificant. The change in the phenolic content of the material in the course of processing does not necessarily translate into a corresponding change in antiradical/antioxidant activity. In the Tip-Top and Toffi cultivars of *Phaseolus vulgaris* L., for example, the levels of the studied phenolics decreased by approx. 20 % due to extrusion, while the antioxidant activity decreased by 11 and 14 %, respectively, and the antiradical activity (percent inhibition of the free radical after 60 min) fell by 5 and 8 %, respectively. On the other hand, in the Rawela cultivar, although the phenolic content increased by 14 %, the antioxidant activity decreased by 19 % on average but the antiradical activity remained unchanged. As mentioned before, some other compounds not covered in the present work, *e.g.* condensed tannins, also contribute to both activities. In the investigations carried out by Fernandez-Orozco *et al.* (*25*), phenolic compounds contributed 92 % to the total antioxidant capacity (TEAC) of raw lentil seeds and 73 % to that of boiled seeds.

Noteworthy is a much higher initial dynamics of free radical scavenging by extracts from extrudates than those from raw beans. The percent inhibition of the DPPH radical after 10 min was in all extrudates greater than in raw beans: by 118 % on average for Rawela, by 50 % for Tip-Top, and by 20 % for Toffi. A faster depletion of the antiradical potential of extrudates (a high initial dynamics but a similar or lower percent inhibition after 60 min compared to raw beans) may indicate that antiradical activity is likely to quickly diminish in the final product when stored. Proving this hypothesis, however, would require further storage investigations.

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