

## Industrial Rapeseed and Sunflower Meal as Source of Antioxidants

František Kreps, Lenka Vrbíková, Štefan Schmidt

Faculty of Chemical and Food Technology, Slovak University of Technology, Slovak Republic

Correspondence author: Štefan Schmidt, Department of Food Science and Technology, Faculty of Chemical and Food Technology, Slovak University of Technology in Bratislava, Radlinského 9, 812 37 Bratislava, Slovak Republic

### Abstract

In the processing of oilseeds for purposes of vegetable oil's production solid residues such as flakes, cakes and meal are formed. Those by-products are mainly used to produce feed for animals. But they may also enrich human food with proteins, minerals, valuable vitamins A, E, phenolic acids, polyphenols, flavonoids and condensed tannins. The expeller cakes, extracted meal or extracts obtained by extraction with organic solvents, are best applied by in bakery products, spiced meat products potato-based products. Fats are mainly stabilized by synthetic antioxidants BHT or BHA. In this work we proved that synthetic antioxidant BHT was not the best stabilizer of the stored sunflower oil. Typical by-products of fat industry were studied as a potential source of cheap natural antioxidants. The whole sunflower or rapeseed meal added into stored sunflower oils in the amount of 20 g kg<sup>-1</sup> increased oil's stability more than 0.1 g kg<sup>-1</sup> addition of BHT. In order to increase antioxidant activity of whole meal we carried out its fractionation to obtain five fractions with different particles size. Out of these fractions, fraction 4 with particles size of 0.05 – 0.15 10<sup>-3</sup> m was very effective, second only to whole meal, especially to whole sunflower meal. They increased the stability of sunflower oil 1.4 times during storage. By-products such as sunflower or rapeseed meal added directly into sunflower oil can extend its shelf life.

**Keywords:** by-products, expelled cake, extracted meal, BHT, fractions

### I. INTRODUCTION

The utilization of natural antioxidants of plant seems promising in view of the economic accessibility of by-products formed in the production of vegetable oils. The average annual (1996/2000) production of the 17th most produced oils and fats in the world was 105.06 10<sup>6</sup> t. The highest proportion represent soybeans oils (23.14 10<sup>6</sup> t). The production of palm oil was the second largest (18.72 10<sup>6</sup> t) followed by rapeseed oil (12.64 10<sup>6</sup> t) and sunflower oil (9.11 10<sup>6</sup> t).<sup>1</sup> The by-products obtained in vegetable oils production are mainly expelled cakes or extracted meal. The cakes are formed by heating-crushing of seeds and subsequent pressing. The meal represents by-products after extraction of lipids from crushed or flaked cakes.<sup>2</sup> Processing of the 10th most cultivated oilseeds in the world the average annual (1996/2000) production produced 242.71 10<sup>6</sup> t of crushed seeds and 164.65 10<sup>6</sup> t of meal.<sup>1</sup> Those by-products remaining after partially de-fating of oilseeds are generally rich in protein and find a ready market in production of animal feed. Some by-products (solids), especially from soybean, are added to human food as flours, concentrates, textured particles or protein isolates. But some oilseed solids contain toxins or allergens that make them unfit for

animal feeds, for example solids from processing of tung nut and castor bean. Some processes use a chemical additive and extrusion to detoxify and deallergenate castor meal and thus make it suitable for animal feed.<sup>2</sup> Addition of natural antioxidant should also satisfy the following requirements. Such antioxidants are supposed to originate from an abundant and inexpensive vegetable matrix, produced by a suitable and economical technology. Finally natural antioxidants should be properly effective.

The liposoluble antioxidants are extracted into crude oil during oilseed processing and are partially recovered from a deodorization condensate. More polar antioxidants remain in expelled cakes or extracted meal which may be also used as food additives to increase the oxidative stability of food. Oilseed meal industrially extracted with hydrocarbons may subsequently be extracted by more polar organic solvents to obtain concentrates of phenolic substances.<sup>3</sup>

By-products of oil processing contain phenolic compounds of various chemical structures such as tocopherols, carotenoids, flavonoids, lignans, lignins, phenolic acids and tannins. These sources of cheap natural antioxidants could substitute synthetic additives and also play an important role in

preventing of many diseases.<sup>4</sup> Natural derived phenolic extracts would be used as antioxidants in lipid and lipid-containing systems, e.g. meat products with higher fat content.<sup>5</sup> Their activities depend on the systems they're stabilized, as hydrophilic antioxidants are more effective in bulk oil, whereas hydrophobic antioxidants are more effective in oil-in-water emulsion or in liposomes.<sup>6,7</sup>

## II. A formation of by-products in processing of oilseeds in the fat industry

In the production of edible oils the seeds are crushed and heated to inactivate enzymes and to destroy the bonds between lipid-protein moieties in membranes and lipoproteins. Sunflower seeds can be dehulled before crushing and heating. The hulls remain as a by-product. Crushing and heating are omitted in the production of virgin oils.<sup>3</sup> Then oil is subsequently partially removed from the crushed and heated material by expeller pressing under high pressure and at temperatures up to 100 °C. After this process the cake (solid residue) contains some residual fat. Oilseeds which contain a lot of fat and are low in solids produce solid residues containing only a small residue of the original fat, approximately 2.5 – 5 %. However, oilseeds low in fat and high in solid residues still contain 15 – 20 % of the original fat.<sup>2</sup> In oilseeds with low oil content the pressing may be omitted. Further processing can produce more fat from cake, e.g. by extraction, most often performed by hexane or mixtures of hydrocarbons. Miscella containing oil dissolved in the solvent is then removed and the solvent is distilled off. Crude oil obtained in this way is usually combined with crude oil obtained by expeller pressing. Solvent-extracted meal typically has less than 1 % of residual fat. The residual solvents are removed from the meal. In the production of virgin (cold-pressed) oils, the second stage of solvent extraction is omitted.<sup>2,3</sup> During the processing, natural antioxidants present in original oilseeds are fractionated into the liposoluble and hydrophilic fractions. Mostly lipophilic antioxidants are extracted into crude oil during the expeller pressing and solvent extraction. Some antioxidants of medium polarity are also partially extracted. More polar natural antioxidants in crude oils are partially removed during the oil refining. Antioxidants remaining in the extracted meal are not significantly damaged during the oilseed processing.<sup>3</sup>

### By-products from processing of rapeseed oil

In the production of rapeseed oil, the most active antioxidants from seeds remain in the by-products which have the form of flakes, expelled cakes or extracted meal.<sup>8</sup> Rapeseeds usually contain more active phenolic antioxidants compounds than

other oilseeds.<sup>9</sup> Rapeseed contains approximately 17.5 g kg<sup>-1</sup> of total phenolic compounds. The typical by-products of rapeseed oil processing are expelled cake and extracted meal containing approximately 18.5 and 15.8 g kg<sup>-1</sup> of total phenolic compounds, respectively.<sup>10</sup> Rapeseed meal commonly used as animal feed contains 40 % protein and its amino acid composition has a high nutritive value. The level of glucosinolates in rapeseed is less than 30 10<sup>-6</sup> mol g<sup>-1</sup>, resulting in good quality meal. It has high fiber content and is rich in minerals such as calcium, magnesium, zinc and copper. Extracted rapeseed meal contains only negligible residues of liposoluble antioxidants (vitamin E).<sup>3</sup> More significant is the amount of polar active phenolic antioxidants, vitamin A and several B vitamins which make the meal nutritionally very valuable.<sup>11,12</sup> The rapeseed meal contains phenolic compounds in the range 1–2 %, which is 5 times higher content compared to soybean meal.<sup>13</sup> The content of phenolic acids in dehulled and defatted rapeseed meal is 10 - 30 times higher than in meals obtained from other oilseeds.<sup>11</sup> Phenolic compounds in the extracts of rapeseed meal mostly contain derivatives of phenolic acids, flavonoids and condensed tannins.<sup>13,14</sup> These phenolic acids, especially the derivatives of benzoic acid and cinnamic acid are present both as free, insoluble, bound form and as soluble esters (glycosides).<sup>5,9,15</sup> In rapeseeds the most significant phenolic compound is sinapine, the choline ester of sinapic acid (about 80 % of the total phenolic compounds). Typically, the amount of sinapic acid derivatives in rapeseed meal varies from 6.39 to 18.37 g kg<sup>-1</sup> depending on the variety of oilseed plant and the oil processing method.<sup>16</sup> Sinapine Fig. 1. is mainly concentrated in kernels, their content in hulls is lower. The content of phenolic compounds increases during ripening of rapeseed (*Brassica napus*). The amount of water soluble phenols, including sinapine increases to maximum value in the last phase of green seeds ripening and at the beginning of their browning.<sup>11</sup> Sinapic acid in rapeseeds also exists as the glucopyranosyl sinapate (Fig. 1.).<sup>17</sup> This sinapic acid derivate was identified as the most antioxidative active component of rapeseed meal.<sup>18</sup> Free phenolic acids represent 6.5 - 9.0 % of total phenols in rapeseed flour and more than 15 % in rapeseed meal. However the sinapic acid is the main free phenolic acid (70 – 80 %).<sup>16</sup> Investigations on the free-radical-scavenging activity of sinapic acid and sinapine indicated that sinapine had a significant but lower activity as sinapic acid. Sinapine was not able to inhibit the formation of hydroperoxides, compared with sinapic acid. This indicated that sinapic acid-rich extracts could better inhibit the lipid oxidation in bulk lipid systems compared with sinapine-rich fractions.<sup>19,20</sup> Other minor phenolic compounds in the

rapeseed may include *p*-hydroxybenzoic, vanillic, gentisic, protocatechuic, syringic, *p*-coumaric,

ferulic, caffeic and chlorogenic acids<sup>10</sup> and tannins which are mainly concentrated in rapeseed hulls.<sup>5</sup>

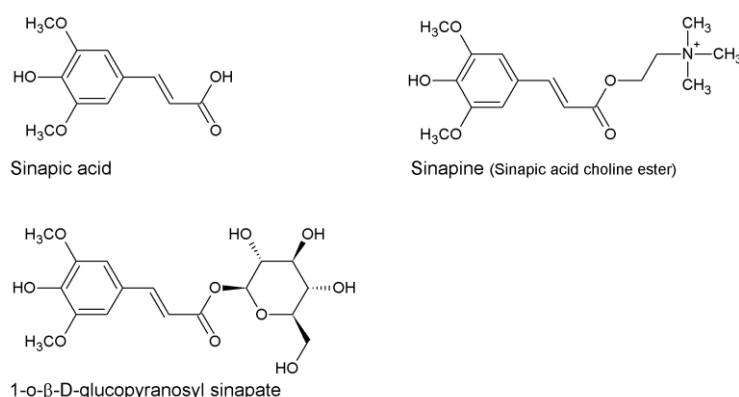


Fig. 1. Main sinapic acid derivatives of rapeseeds

### III. Antioxidant activity of rapeseed cake and meal

The ground oilseeds can without difficulty be used in bakery or meat products, where the presence of spices would mask their eventual off-flavours.<sup>3</sup> The antioxidant activity of rapeseed by-products can be measured in the lard (contains any antioxidants) or in the vegetable oils where there is synergism of phenolic compounds with other compounds. Antioxidative effect of flakes, meal and their fractions in the lard has been demonstrated. The best antioxidative was observed in the fraction of rapeseed flakes (size <math>0.15 \cdot 10^{-3}</math> m), 1.2 times higher than that of 0.1 g kg<sup>-1</sup> BHT addition. Also lard with direct addition of fraction rapeseed flakes (size <math>0.15 \cdot 10^{-3}</math> m) had 3 times higher stability than addition of the same size fraction of rapeseed meal. Moreover, direct addition of flakes or meal was more efficient than its lard macerate.<sup>5,21</sup> Other authors observed that addition of 10, 20 or 40 g kg<sup>-1</sup> the non-sieved rapeseed meal improved the stability of lard 1.2 – 2 times. Adding 80 – 150 g kg<sup>-1</sup> of meal into the lard has increased its stability 3 – 8 times. Rapeseed meal was fractionated in order to find a fraction of meal that would stabilize the lard best. The fraction with size 0.15 – 0,315  $\cdot 10^{-3}$  m has stabilized lard better than other fractions of sieved meal. It has been discovered under microscope that this fraction with size 0.150 – 0,315  $\cdot 10^{-3}$  m contained a mixture of hulls and rapeseed kernels. Non-sieved rapeseed meal has had similar but lower stabilization effect on the lard owing to the presence of dust particles.<sup>22</sup> It was found that extracts from rapeseed meal had higher antioxidant activity than non-extracted by-products obtained from oilseeds in the fat industry.<sup>23</sup> A very promising antioxidant appears to be ethanol extract of evening primrose meal, possessing antioxidant activity comparable to ethanolic extract of rapeseed meal.<sup>24</sup> Further extracts obtained from rapeseed by-

products showed better antioxidant activity as direct addition of by-products in the lard.<sup>5,22</sup> The antioxidant activity of those extracts depends on the cultivar of oilseeds and the solvent used for extraction.<sup>8</sup> Solvents of medium polarity are more useful for the extraction of antioxidants but they are relatively expensive.<sup>5</sup> Solvents such as ethanol or methanol (80 - 90 % v/v) are suitable for the extraction of polar antioxidants, e.g. flavonoids or phenolic acids from rapeseed meal or cake. Antioxidants obtained by these solvents would be acceptable for fat dispersions, such as in meat product.<sup>3</sup> Ethanol extract of rapeseed meal has been added to rapeseed oil in the concentration of 0.5 – 1 g kg<sup>-1</sup> and has had higher effect than 0.2 g kg<sup>-1</sup> addition of synthetic antioxidants BHT, BHA and mixed antioxidant BHA / BHT / monoacylglycerol citrate.<sup>13</sup> On the other hand it has been demonstrated that fraction of rapeseed meal low in phenolic compounds has had better antioxidant activity than rapeseed meal with high phenolic content. That synergism of phenolic compounds with one or more other components present in each fraction may contribute to the high antioxidant activity of rapeseed fractions containing low phenolic compounds.<sup>25</sup>

### IV. By-products from processing of sunflower oil and their antioxidant activity

The production of sunflower oil generates by-products such as hulls, flakes, expelled cakes or extracted meal. These by-products are a good source of non-lipophilic antioxidants. The fat industry produces three types of sunflower meal: dehulled meal (28 % protein and 25 – 28 % fiber), partially dehulled meal (35 – 37 % protein and 18 % fiber) and double-dehulled sunflower meal (40 – 42 % protein and 12 – 14 % fiber). Thus, the composition of sunflower meal depends on the efficiency of the

dehulling process. Sunflower meal is, unlike other oilseed meals, a valuable source of calcium, phosphorus as well as an excellent source of water-soluble B-complex vitamins, namely nicotinic acid, thiamine, pantothenic acid, riboflavin and biotin.<sup>26</sup> It is known that sunflower seeds are rich in phenolic compounds and the total phenols content in sunflower seeds is in the range 10 - 42 g kg<sup>-1</sup>.<sup>27</sup> The sunflower meal formed after extraction of lipids from cakes had similar content of phenolic antioxidants as sunflower seeds. The content of phenolic compounds in meal may vary depending on the content of hulls in meal and variety (regions) cultivate of sunflower.<sup>28</sup> Chlorogenic and caffeic acids (Fig. 2.) compose 70 % of phenolic compounds in sunflower

flour.<sup>29</sup> Chlorogenic acid is described as a major phenolic compounds in sunflower seeds while caffeic acid is present in lower concentration.<sup>30,31</sup> Nevertheless it was evaluated that the caffeic and protocatechuic acids (3,4-Dihydroxyphenolic acids) were more active antioxidants than monohydroxyphenolic acid (p-hydroxybenzoic acid), 2,5-dihydroxyphenolic acid (gentisic acid) and 3-methoxy-4-hydroxyphenolic acids (vanillic and ferulic acids) and their corresponding alkyl esters.<sup>32</sup> In extracts of sunflower seeds several minor phenol acids: *p*-hydroxybenzoic acid, *m*-hydroxybenzoic, *p*-coumaric, cinnamic, vanillic and syringic have been identified.<sup>33</sup>

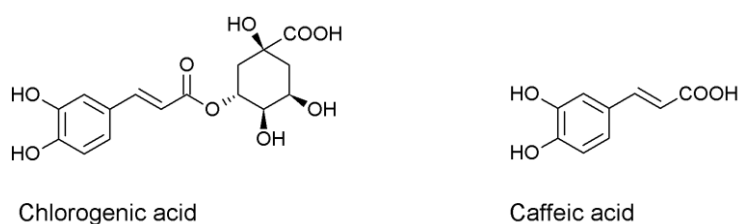


Fig. 2. Main phenolic acids of sunflower seeds

Chlorogenic acid can be bonded to polypeptides and oligonucleotides with low molecular weight.<sup>34</sup> In the production of sunflower oil, mainly in alkaline conditions, chlorogenic acid reacts with certain protein fraction, giving the meal of dark green color.<sup>35</sup> The undesirable browning of cakes or meal can be negatively affected by the oxidation of chlorogenic acid, manifested by undesirable browning of cakes and meal depends on the amount of polyphenol oxidase present in the sunflower seeds.<sup>36</sup> Several methods have been proposed for the production of protein concentrates obtained from sunflower seeds, cakes or meal through extraction or inactivation of chlorogenic acid. Sunflower meal (blended with wheat flour) can be used for human nutrition. Despite their dark color, sunflower protein (71 %) concentrates have excellent digestibility.<sup>35</sup>

The sunflower seeds may be dehulled after clearing and climatizing of seeds. The hulls comprise 22 – 28 % of the total weight of sunflower seeds and may be removed before or immediately following oil extraction or may remain in the meal. Dehulling is omitted when the fat industry has not this technology and the raw oil is dewaxed. Sunflower hulls contain 60 – 65 % fiber, around 4 % crude protein, 5 % lipidic matter (including waxes, hydrocarbons, fatty acids, sterols and triterpenic acid), 50 % carbohydrates (mainly cellulose and lignin), 26 % reducing sugars (mainly xylose) and 2 % ash.<sup>26,35</sup> It is reported that sunflower hulls contain approximately 32 g kg<sup>-1</sup> of total phenols. Chlorogenic acid was the

most abundant phenol (79.4 %) in sunflower hulls. Other remarkable phenols are the *o*-cinnamic (5.8 %), protocatechuic (5.2 %), caffeic acid (4.1 %) and under 2 % is the content of ferulic and syringic acid.<sup>37</sup> Sunflower hulls contain a large amount of raw fiber of practically no nutritional value, so that they are almost exclusively used as ruminant feed. They can be utilized as a source of fuel. The heat value of hulls is 19.2 10<sup>6</sup> J kg<sup>-1</sup>, whereas the heat value of hulls and meal together is 23.6 10<sup>6</sup> J kg<sup>-1</sup>.<sup>26</sup>

In many studies it has been demonstrated that sunflower meal has high antioxidant potential, which could be beneficial for further technological utilization.<sup>28</sup> Numerous sunflower polyphenols such as caffeic, chlorogenic and ferulic acids have been shown in many studies to exert a high antioxidative potential.<sup>28,32</sup> They can be used as effective antioxidants for stabilization of sunflower oil.<sup>27</sup> By-products of sunflower oil's production, meal and hulls are valuable sources of phenolic compounds that might be recovered and used as natural antioxidants.<sup>28</sup> Mixtures of solvents, e.g. acetone/water 60:40 (v/v) or ethanol/water 60:40 (v/v) proved to be suitable for extraction of phenolic compounds from sunflower seed hulls. From 25 g of partially defatted sunflower hulls, around 90 mg of powdery antioxidant product, consisting of 58 % caffeic acid, was obtained. From 1 kg of sunflower hulls around 14 g of chlorogenic acid can be recovered and after its alkaline hydrolysis 7 g of caffeic acid was recovered. The accelerated oxidation test (Rancimat 130 °C) was performed by using

powdery sunflower antioxidant's product, caffeic acid standard and propyl gallate. It was determined that the propyl gallate was 15 – 20 % more effectiveness than powdery sunflower antioxidant's product which was similar effective than the caffeic acid standard.<sup>37</sup>

## V. EXPERIMENTAL

### Conditions and procedures

We investigated the antioxidative potential of rapeseed and sunflower meal and their fractions in sunflower oil.

Sunflower, rapeseed meal and sunflower oil were obtained from Slovak fat industry (Palma Group Inc. Bratislava, Slovakia). The meal was solid residues after extraction of oils (with hexane) from flaked sunflower and rapeseed cake. Sunflower and rapeseed meal was fractionated on sieves to obtain five fractions with different particle sizes. The rapeseed

(Fig. 3 A) and sunflower (Fig. 3 B) meal fractions 1

consisted mainly of hull conglomerates and some seed kernels with size longer than  $0.75 \cdot 10^{-3}$  m (max  $1 \cdot 10^{-3}$  m). Fractions 2 contained more kernels than fraction 1, but still dominated hulls. These rapeseed (Fig. 4 A) and sunflower (Fig. 4 B) meal fractions contained particles with size  $0.63 - 0.75 \cdot 10^{-3}$  m. Fractions 3 of rapeseed (Fig. 5 A) and sunflower (Fig. 5 B) meal with size  $0.32 - 0.63 \cdot 10^{-3}$  m were a mixture of hulls and kernels. The rapeseed (Fig. 6 A) and sunflower (Fig. 6 B) meal fractions 4 contained less hulls than fraction 3 and more seed kernels with size  $0.15 - 0.32 \cdot 10^{-3}$  m. Fractions 5 of rapeseed (Fig. 7 A) and sunflower (Fig. 7 B) meal with size  $0.05 - 0.15 \cdot 10^{-3}$  m contained mainly fragments of kernels, some hulls and dust particles.

The best method to determine the antioxidant activity of natural compounds is monitoring the changes of oils and fats during storage. This method is expensive and time-consuming, that is a reason why the accelerated method of fat oxidation is usually used.<sup>5</sup>

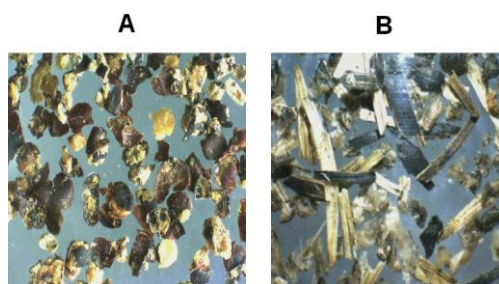


Fig. 3. Rapeseed (A) and sunflower (B) meal fractions 1 with particles longer than  $0.75 \cdot 10^{-3}$  m. Magnification 10x

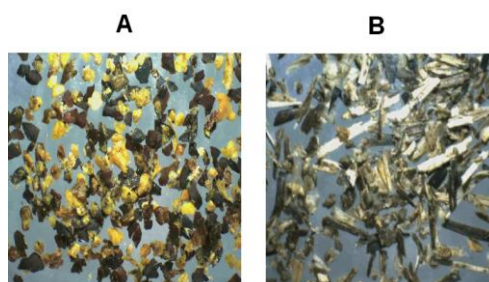


Fig. 4. Rapeseed (A) and sunflower (B) meal fractions 2 with particles size  $0.63-0.75 \cdot 10^{-3}$  m. Magnification 10x

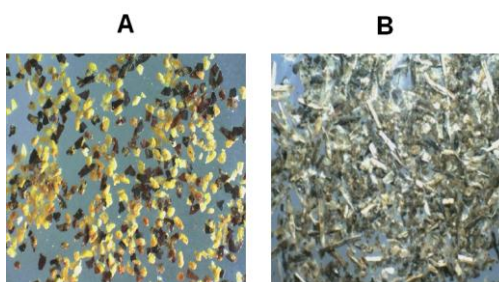


Fig. 5. Rapeseed (A) and sunflower (B) meal fractions 3 with particles size  $0.315-0.63 \cdot 10^{-3}$  m. Magnification 10x

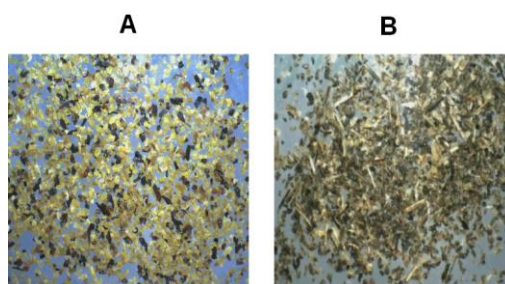


Fig. 6. Rapeseed (A) and sunflower (B) meal fractions 4 with particles size  $0.15\text{--}0.315 \cdot 10^{-3}$  m. Magnification 10x

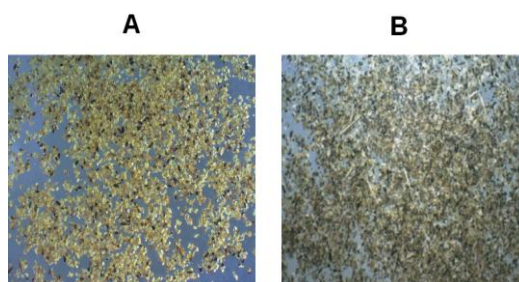


Fig. 7. Rapeseed (A) and sunflower (B) meal fractions 5 with particles size  $0.05\text{--}0.15 \cdot 10^{-3}$  m. Magnification 10x

We combined these two methods for the purpose of examination of antioxidant activity of rapeseed and sunflower meal. Rancimat test as accelerated method of sunflower oil oxidation with added  $20 \text{ g kg}^{-1}$  of sunflower and rapeseed meal fractions was investigated. The fraction with highest antioxidative activity was compared with whole meal ( $20 \text{ g kg}^{-1}$ ) and synthetic antioxidant BHT ( $0.1 \text{ g kg}^{-1}$ ) added into sunflower oil. Then we carried out a 56 days storage of sunflower oils with addition of  $20 \text{ g kg}^{-1}$  whole meal, fraction 4 which was the best antioxidant of all fractions and  $0.1 \text{ g kg}^{-1}$  addition of synthetic antioxidant BHT. The sunflower oil was stored near the window (160 cd) at ambient temperature ( $26 \text{ }^\circ\text{C}$ ) in glass jars with  $500 \text{ dm}^3$  volume, which were covered by filter paper. For reasons of mixture homogeneity, these oils were stirred by sticks once a day. At the beginning of experiment, the sunflower oil without added stabilizers had the peroxide value  $0.43 (10^{-3} \text{ mol } 0.5 \text{ O}_2 \text{ kg}^{-1} \text{ of fat})$ , iodine value was  $128.5 \text{ g I}_2 100 \text{ g}^{-1}$  of fat and the acid value was  $0.21 \text{ g KOH kg}^{-1}$  of fat.

#### VI. Analytical methods

The peroxide value was determined iodometrically and expressed in  $10^{-3} \text{ mol } 0.5 \text{ O}_2 \text{ kg}^{-1}$  of fat.<sup>38</sup> The acid value was analyzed by alkalimetric titration and expressed in  $\text{g KOH kg}^{-1}$  of fat.<sup>39</sup> The analysis of iodine value was carried out iodometrically according to Hanuš and expressed in  $\text{g}$

$\text{I}_2 100 \text{ g}^{-1}$  of fat.<sup>40</sup> The stability of fats was measured by the method of accelerated oxidation with the apparatus Rancimat 743 at a constant temperature of fat ( $110 \text{ }^\circ\text{C}$ ) and air flow ( $20 \text{ dm}^3 \text{ h}^{-1}$ ) which bubbled through the sample. The stability of lipids is expressed by induction period in hours as resistance of lipids to oxidation.<sup>41</sup> The antioxidant activity of the sunflower oil enriched with meal or synthetic antioxidant BHT is expressed as a protective factor (PF) calculated as a ratio of the induction period of sunflower oil with the addition of meal or BHT ( $\text{IP}_A$ ) and without the addition of meal or BHT ( $\text{IP}_0$ ):  $\text{PF} = \text{IP}_A / \text{IP}_0$

#### VII. Results and discussion

In the first experiment the protective effect of whole rapeseed, sunflower meal and their five fractions before oxidation of sunflower oil was investigated. For comparison of their relative protective effects, one of the most used synthetic antioxidant BHT was applied. Sunflower oil was enriched with  $20 \text{ g kg}^{-1}$  of meal or  $0.1 \text{ g kg}^{-1}$  of BHT addition and compared with pure sunflower oil. Figure 8 shows that the most effective stabilizer before oxidation of sunflower oil was the synthetic antioxidant. The whole rapeseed and sunflower meal has 1.13 and 1.04 times lower protective factor than BHT, respectively. Sunflower meal was more effective than rapeseed meal before oxidation of sunflower oil.

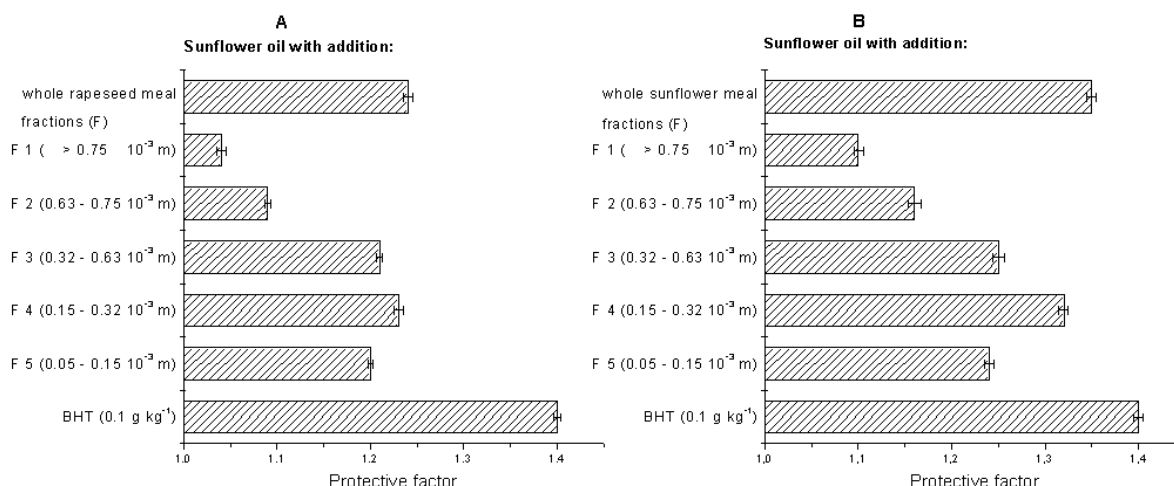


Fig. 8. Protective factor of 20 g kg<sup>-1</sup> whole rapeseed meal (A), sunflower meal (B) and their fractions into sunflower oil

According to some authors, the reason may be higher content of total phenols (10 – 42 g kg<sup>-1</sup>) in sunflower meal<sup>27</sup> and lower concentration of total phenols (16 g kg<sup>-1</sup>) in rapeseed meal.<sup>10</sup> The whole rapeseed and sunflower meal had better protective factor than fractions 4 (size 0.05 – 0.15 10<sup>-3</sup> m) which were more effective before oxidation than other fractions (Fig. 8). Whole rapeseed and sunflower meal contain dust particles, nevertheless they may contain more phenol compounds than fractions 4 with lower hulls content compared with whole meal. Defatted rapeseed meal contains 1.8 % total phenols but dehulled rapeseed flours contain 1.1 – 1.3 % total phenols.<sup>10</sup>

In the second part of experiment we have made the storage test of sunflower oil with addition of 20 g kg<sup>-1</sup> whole rapeseed, sunflower meal and fractions 4 (Fig. 9). During the storage we measured

the peroxide value which expressed the content of primary oxidation products, namely lipoperoxides in the sunflower oil. The peroxide value also predicts the oxidation stability of this oil.<sup>5</sup> Stability of sunflower oil with addition of meal was compared with oil without stabilizers and with addition of 0.1 g kg<sup>-1</sup> BHT. From Fig. 9. resulted that the synthetic antioxidant was not the best stabilizer during storage of sunflower oils. Sunflower oils with 20 g kg<sup>-1</sup> whole sunflower or rapeseed meal had lower peroxide value even at 30th day of storage. That indicated higher stability than oil with an addition of BHT. The presence of natural antioxidants may be the reason of their better regeneration. Activities of polar-phenolic antioxidants depend on the systems to be stabilized and they are more effective in bulk oil.<sup>6,7</sup> Sunflower oils stored with fractions 4 were not so stable as sunflower oils with whole meal addition.

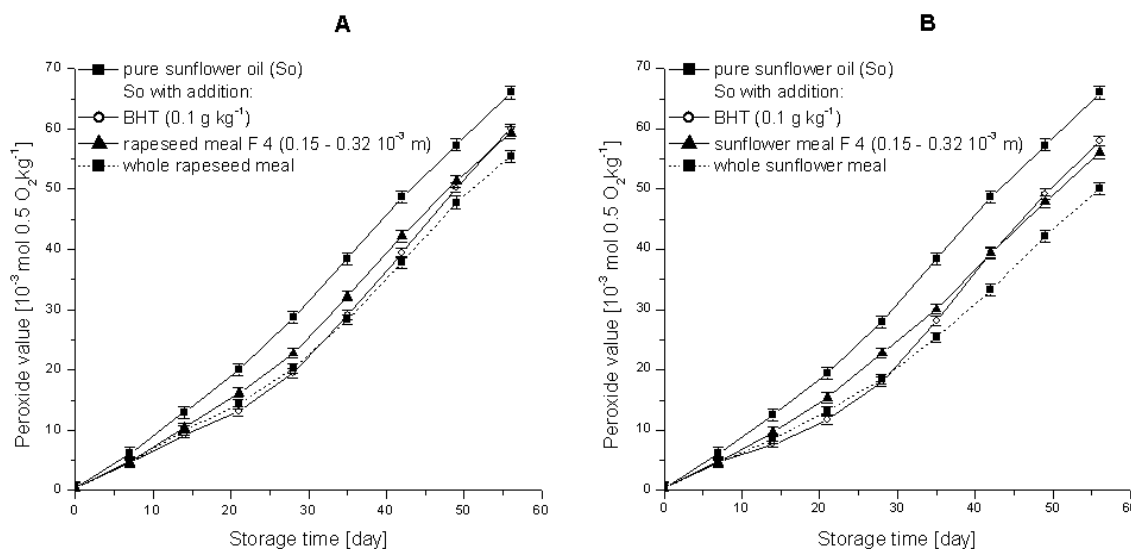


Fig. 9. Storage of sunflower oil with added 20 g kg<sup>-1</sup> of whole rapeseed meal (A), sunflower meal (B) and their fractions

The sunflower oil with rapeseed meal fractions 4 had lower peroxide value than sunflower oil with BHT addition just at the end of the experiment (56th day). But sunflower meal fraction 4 added into the sunflower oil decreased the peroxide value after 42th day on lower value than sunflower oil with content of BHT. Sunflower oil with whole sunflower meal had in average 1.4 times lower peroxide value than sunflower oil without whole sunflower meal. The Fig. 9. describes that the stability of sunflower oil was the best with an addition of whole meal and especially with whole sunflower meal.

### VIII. CONCLUSION

Rapeseed, sunflower meal as by-products from processing of vegetable oils can serve as a potential source of cheap natural antioxidants. We have confirmed that the addition of 20 g kg<sup>-1</sup> whole sunflower meal and rapeseed meal increased its protective effect by 1.35 and 1.24 times before sunflower oil oxidation, respectively. Based on the results, whole meal had higher protective factor than fractions obtained by sieving of meal. Only fractions 4 with particle size 0.15 – 0.32 10<sup>-3</sup> m had a similar effect as whole meal. During ageing of sunflower oils with whole meal, fractions 4 and with BHT, antioxidative activity of synthetic antioxidant did not surpass natural antioxidants. Thus whole rapeseed and sunflower meal added (20 g kg<sup>-1</sup>) into sunflower oil increased stability of sunflower oil better than a 0.1 g kg<sup>-1</sup> addition of BHT. Rapeseed meal which was worse stabilizer than sunflower meal added into sunflower oil increased after 30 days its stability 1.2 times compared with addition of BHT. Whole sunflower or rapeseed meal can be used either for recovery by extraction of antioxidants or directly added into vegetable oil. The use of these by-products in fat processing factories as direct addition into bulk vegetable oil can extend the shelf life of oils during their storage in batch operation.

### IX. Acknowledgement

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### Notation

BHA	butylated hydroxyanisole
BHT	butylated hydroxytoluene
cd	candela unit of luminous intensity
F	fractions of rapeseed or sunflower meal
IP <sub>0</sub>	induction period of oil without addition of stabilizers
IP <sub>A</sub>	induction period of oil with addition of stabilizers
PF	protective factor

### References

- [1] Gunstone FD, Production and trade of vegetable oils. *Vegetable Oils in Food Technology: Composition, Properties and Uses*, Blackwell Publishing Ltd., Osney Mead and CRC Press LLC., Boca Raton, pp. 1-17 (2002).
- [2] Shahidi F and Williams MA, Recovery of Oils and Fats from Oilseeds and Fatty Materials. *Bailey's industrial oil and fat products*, 6th ed.; John Wiley & Sons Inc., New Jersey, Vol. 5, pp. 99-189 (2005).
- [3] Schmidt Š and Pokorný J, Potential application of oilseeds as sources of antioxidants for food lipids – a review. *Czech J Food Sci* **23**:93-102 (2005).
- [4] Matthäus B, Antioxidant activity of extracts obtained from residues of different oilseeds. *J Agric Food Chem* **50**:3444-3452 (2002).
- [5] Schmidt, Š. *Antioxidants and oxidation changes in foods*, 1th ed; STU., Bratislava, p. 220 (2010).
- [6] Frankel EN, Huang S-W, Kanner J and German JB, Interfacial phenomena in the evaluation of antioxidants: bulk vs. emulsions. *J Agric Food Chem* **42**:1054-1059 (1994).
- [7] Schmidt Š, Niklová I, Pokorný J, Farkaš P and Sekretár S, Antioxidant activity of evening primrose phenolics in sunflower and rapeseed oils. *J Agric Food Chem* **105**:427–435 (2003).
- [8] Amarowicz R, Nacz M and Shahidi F, Antioxidant activity of various fractions of non-tannin phenolics of canola hulls. *J Agric Food Chem* **48**:2755-2759 (2000).
- [9] Nowak H, Kujava R, Zadernowski R, Rocznik B, and Kozłowska H, Antioxidative and bactericidal properties of phenolic compounds in rapeseeds. *Fat Sci Technol* **94**:149-152 (1992).
- [10] Khattab R, Eskin M, Aliani M and Thiyam U, Determination of sinapic acid derivatives in canola extracts using high-performance liquid chromatography. *J Am Oil Chem Soc* **87**:147-155 (2010).
- [11] Nacz M, Amarowicz R, Sullivan A, and Shahidi F, Current research developments on polyphenolics of rapeseed/canola: a review. *Food Chem* **62**:489-502 (1998).
- [12] Chen ZY, Chan PT, Ma HM, Fung KP and Wang J, Antioxidative effect of ethanol tea extracts on oxidation of canola oil. *J Amer Oil Chem Soc* **73**:375-380 (1996).
- [13] Wanasundara UN and Shahidi F, Canola Extract as an alternative natural antioxidant



- for canola Oil. *J Amer Oil Chem Soc* **71**:817-822 (1994).
- [14] Zadernowski R, Nowak-Polakowska H and Konopka I, Effect of heating on antioxidative activity of rapeseed and evening primrose extracts. *Pol J Food Nutr Sci* **46**:13-20 (1996)
- [15] Krygier K, Sosulski F and Hogge L, Free, esterified, and insoluble-bound phenolic acids. 2. Composition of phenolic acids in rapeseed flour and hulls. *J Agric Food Chem* **30**:334-336 (1982)
- [16] Kozłowska H, Naczka M, Shahidi F, and Zadernowski R, Phenolic acids and tannins in rapeseed and canola. *Canola and Rapeseed: Production, chemistry, nutrition and processing technology*. Van Nostrand Reinhold, New York, Vol. 11, pp. 193-210 (1990).
- [17] Amarowicz R, and Shahidi F, Chromatographic separation of glucopyranosyl sinapate from canola meal. *J Am Oil Chem Soc* **71**:551-552 (1994).
- [18] Wanasundara U, Amarowicz R and Shahidi F, Isolation and identification of an antioxidative component in canola meal. *J Agric Food Chem* **42**:1285-1290 (1994).
- [19] Thiyam U, Stöckmann H, Felde T and Schwarz K, Antioxidative effect of the main sinapic acid derivatives from rapeseed and mustard oil by-products. *Eur J Lipid Sci Tech* **108**:239-248 (2006).
- [20] Vuorela S, Meyer AS, Heinonen M, Impact of the isolation method on the antioxidant activity of rapeseed meal phenolics. *J Agric Food Chem* **52**:8202-8207 (2004).
- [21] Schmidt Š, Sekretár S, Niklová I and Klvanová J, Defatted rapeseed flakes as a promising source of natural antioxidants. *Proceedings of the 27th International Conference of Slovak Society of Chemical Engineering*. Tatranské Matliare, Slovakia, 22. - 26. 5. 2000, ISBN 1 80-227-1350-3
- [22] Kreps F, Schmidt F, Vrbíková L, Tmáková L, Hlásniková J and Sekretár S, Influence of rapeseed meal on lard stability. *Acta Chimica Slovaca* **5**:131-138 (2012).
- [23] Kozłowska H, Zadernowski W and Sosulski FW, Phenolic acids in oilseeds flours. *Nahrung* **27**: 449-453 (1983).
- [24] Schmidt S, Niklova I, and Sekretár S, Antioxidant activity of extracts from rapeseed and evening primrose meal. *Bull Food Res* **37**:257-265 (1998).
- [25] Wanasundara U, Amarowicz R and Shahidi F, Partial characterization of natural antioxidants in canola meal. *Food Res Intern* **28**:525-530 (1995)
- [26] Shahidi F and Oickard MD, By-product utilization. *Bailey's industrial oil and fat products*, 6th ed.; John Wiley & Sons Inc., New Jersey, Vol. 4, pp. 391-416 (2005).
- [27] Leonardis AD, Macciola V and Rocco AD, Oxidative stabilization of cold-pressed sunflower oil using phenolic compounds of the same seeds. *J Sci Food Agric* **83**:523-528 (2003).
- [28] Weisz GM, Ka10-3 merer DR and Carle R, Identification and quantification of phenolic compounds from sunflower (*Helianthus annuus* L.) kernels and shells by HPLC-DAD/ESI-MS<sup>n</sup>. *Food Chem* **115**:758-765 (2009).
- [29] Sabir MA, Sosulski FW and Kernan JA, Phenolic constituents in sunflower flour. *J Agric Food Chem* **22**:572-574 (1974).
- [30] Sosulski F, Organoleptic and nutritional effects of phenolic compounds on oilseed protein products: A Review. *J Amer Oil Chem Soc* **56**:711-715 (1979).
- [31] Rhee KS, Ziprin YA and Rhee KS, Water-soluble antioxidant activity of oilseed protein derivatives in model lipid peroxidation systems of meat. *J Food Sci* **44**:1132-1135 (1979).
- [32] Hrádková I, Merkl R, Šmidrkal J, Kyselka J, and Filip V, Antioxidant Effect of Mono- and Dihydroxyphenols in Sunflower Oil with Different Level of Naturally Present Tocopherols. *Eur J Lipid Sci Tech*, in press, DOI: 10.1002/ejlt.201200293
- [33] Leung J, Fenton TW and Clandinin DR, Phenolic components of sunflower flour. *J Food Sci* **46**:1386-1388 (1981).
- [34] Sabir MA, Sosulski FW and Finlayson AJ, Chlorogenic acid-protein interaction in sunflower. *J Agric Food Chem* **22**:575-578 (1974)
- [35] Shahidi F and Grompone MA, Sunflower oil. *Bailey's industrial oil and fat products*, 6th ed.; John Wiley & Sons Inc., New Jersey, Vol. 2, pp. 655-730 (2005).
- [36] Singh N, Singh R, Kaur K and Singh H, Studies of the physico-chemical properties and polyphenoloxidase activity in seeds from hybrid sunflower (*Helianthus annuus*) varieties grown in India. *Food Chem* **66**:241-247 (1999)
- [37] Leonardis AD, Macciola V and Domenico ND, A first pilot study to produce a food antioxidant from sunflower seed shells (*Helianthus annuus*). *Eur J Lipid Sci Tech* **107**:220-227 (2005).

- [38] AOCS official Method Cd 8-53. *Peroxid Value Acetic Acid-Chloroform method*. 2 p. (1993)
- [39] AOCS official Method Cd 3d-63. *Acid Value*. 2 p. (1993)
- [40] ČSN 58 0101. *Methods of fats and oils analysis* 40 p. (1965)
- [41] AOCS official Method Cd 12b-92. *Oil Stability Index*. 5 p. (1993)

#### František Kreps



Dr. František Kreps working at the Slovak Technical University in Faculty of Chemical and Food Technology, Slovakia. He specializes in food and lipid technology, his recent research activities is focused on antioxidants, analysis of minor compounds in vegetable oils, by-products from processing of oil, lipids technology and refining of vegetable oils.

#### Lenka Vrbiková



Dr. Lenka Vrbiková working at the Slovak Technical University in Faculty of Chemical and Food Technology, Slovakia. She specializes in

lipid technology, especially in shelf life prediction of vegetable oils, analysis of bryndza (traditional aromatic soft sheep cheese) and honey compounds.

#### Štefan Schmidt



Professor Štefan Schmidt is subhead of the Institute of Biotechnology and Food Science, head of the Department of Food Science and Technology and guarantee of two study programs at the Slovak Technical University in Faculty of Chemical and Food Technology. He teaches courses of chemistry and technology of fats and food engineering where applies his experimental knowledge and experience from research projects, external internships and co-operation with praxis. In scientific field he is focused on the structures of fatty acids and triglycerides, but in present he orients on oxidative deterioration of fats and oils and research of natural antioxidants. He was responsible author of the national part of an international grant Copernicus Joint Research Project with the title Isolation and Investigation of Natural Antioxidants Effective in Preserving Food Quality. He graduated internships at various foreign universities focused on the study of modern methods for the analysis of lipids. He has published 27 scientific works registered in CC with significant international response (116 citations according to the Science Citation Index).