

Variations in fatty acid composition, glucosinolate profile and some phytochemical contents in selected oil seed rape (*Brassica napus* L.) cultivars

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RESUMEN

Variación de la composición de ácidos grasos, perfil de glucosinolatos y contenido de algunos fitoquímicos en cultivos seleccionados de semillas oleaginosas de colza (*Brassica napus* L.)

La colza (*Brassica napus* L.) es hoy en día el tercer cultivo más importante de aceites comestibles en el mundo tras el aceite de soja y de palma. En este estudio semillas de cinco cultivos diferentes de colza, denominadas pactol, silvo, topas, serw 4 y serw 6, fueron evaluadas mediante su composición en ácidos grasos, perfil de glucosinolatos, aminoácidos, tocoferoles totales y contenido fenólico. En todos los cultivos, una significativa variabilidad en los ácidos grasos fue observada. El ácido oleico (C18:1) varió desde un 56.31% a un 58.67%, ácido linoleico (C18:2) desde un 10.52% a un 13.74%, ácido α -linolenico (C18:3) desde 8.83% a un 10.32% y ácido erúrico (22:1) desde un 0.15% a un 0.91%. El perfil de glucosinolatos de colza fue también separado e identificado usando cromatografía líquida de alta eficacia. Una baja variación en el perfil de glucosinolatos fue observado en todos los cultivos ensayados; sin embargo, progoitrina y gluconapina fueron los principales glucosinolatos encontrados en todos los cultivos. Además, el cultivo silvo mostró el contenido de glucosinolatos totales más alto (5.97 $\mu\text{mol/g dw}$). Generalmente, el contenido de aspártico, glutámico, arginina y leucina fue alto, mientras que el contenido de tirosina y isoleucina fue bajo en todos los cultivos. En los tocoferoles totales, los resultados indican que tanto los cultivos serw 6 y pactol tuvieron el contenido de tocoferoles más alto (138.3 and 102.8 mg/100 g de aceite, respectivamente). El contenido total de fenoles varió entre 28.0 a 35.4 mg/g peso seco. El contenido total de fenoles más alto fue encontrado en topas, mientras que el valor más bajo fue detectado en serw 6. Estos parámetros, contenido de ácidos grasos, perfil de glucosinolatos y aminoácidos junto al contenido total de tocoferoles y fenoles, podrían ser tenidos en consideración por los mejoradores de semillas de colza como criterio de selección para desarrollar genotipos que modifiquen los rasgos de calidad de la *Brassica napus* L.

PALABRAS CLAVE: Colza – Composición de ácidos grasos – Fenoles totales – Glucosinolatos – Tocopheroles.

SUMMARY

Variations in fatty acid composition, glucosinolate profile and some phytochemical contents in selected oil seed rape (*Brassica napus* L.) cultivars.

Rapeseed (*Brassica napus* L.) is now the third most important source of edible oil in the world after soybean and palm oil. In this study seeds of five different rapeseed

cultivars namely; pactol, silvo, topas, serw 4 and serw 6 were evaluated for their fatty acid composition, glucosinolate profile, amino acids, total tocopherols and phenolic content. Among all cultivars significant variability in fatty acids were observed. The oleic acid (C18:1) ranged from 56.31% to 58.67%, linoleic acid (C18:2) from 10.52% to 13.74%, α -linolenic acid (C18:3) from 8.83% to 10.32% and erucic acid (22:1) from 0.15% to 0.91%. The glucosinolate profile of rapeseed was also separated and identified using high-performance liquid chromatography. Small variations in the glucosinolate profile were observed among all tested cultivars; however, progoitrin and gluconapin were the major glucosinolate found. Additionally, silvo cultivar showed the highest total glucosinolate contents (5.97 $\mu\text{mol/g dw}$). Generally, the contents of aspartic, glutamic, arginine and leucine were high, while the contents of tyrosine and isoleucine were low among all cultivars. For total tocopherols, the results indicated that both serw 6 and pactol cultivars had the highest total tocopherol contents (138.3 and 102.8 mg/100 g oil, respectively). Total phenolic contents varied from 28.0 to 35.4 mg/g dw. The highest total phenolic content was found in topas while the lowest value was detected in serw 6. These parameters; fatty acid contents, glucosinolate profile and amino acids together with total tocopherols and phenolic contents, could be taken into consideration by oilseed rape breeders as selection criteria for developing genotypes with modified seed quality traits in *Brassica napus* L.

KEY-WORDS: Fatty acid composition – glucosinolate-rapeseed – total phenolic – tocopherols.

1. INTRODUCTION

Rapeseed (*Brassica napus* L.) is an important source of edible oil in many countries. In Egypt, it was introduced as an oil crop which may reduce the gap between local production and the consumption of edible oil. It might also provide a low-cost renewable resource of high value-added compounds such as tocopherol and phytosterols (Moyad, 2005). Moreover Rapeseed (*Brassica napus*), also known as rape, oilseed rape, rapa, rapeseed and (in the case of one particular group of cultivars) canola, is a bright yellow flowering member of the family Brassicaceae (mustard or cabbage family). The oil content of the seed varies from 30-45% depending on the species, the variety and climatic conditions under which it is grown. The

B. napus has been known as a rich source of oil with a low content of saturated fatty acids (5-7%) and a high content of polyunsaturated fatty acids with about 7-10% α -linolenic and 17-21% linoleic acids. It is therefore considered as very healthy edible oil (Baux *et al.*, 2008). The improvement of seed quality is one of the most important objectives in Brassica breeding for satisfying future edible oil requirements (Shengwu *et al.*, 2003). The functional and nutritional values of different vegetable oils are dependent on the nature of the different fatty acids, which are incorporated into the oil (triacylglycerols). On the other hand, erucic acid in the oil is one of the important characteristic components associated with *Brassica spp.* High erucic acid oilseed rape cultivars may increase health risks (Khan *et al.*, 1985). After oil extraction, the remaining meal contains different nutritional and anti-nutritional compounds. Among these, glucosinolates are the most important anti-nutritional compounds. The glucosinolates are nitrogen and sulphur containing natural plant products that have become increasingly important as flavor precursors, cancer prevention agents and crop protectants (Graser *et al.*, 2000). Variation in the amount and pattern of glucosinolates in Brassica plants has been attributed to genetic and environmental factors, including plant age, temperature, water stress, and soil type (Rosa, 1997). They are found in all plant parts, but their quantities may vary considerably among organs (Kjaer 1976, Font *et al.*, 2005). The content of seed glucosinolate is controlled by multiple genes and is complexly regulated in the cell (Uzunova *et al.*, 1995). In oilseed rape, however, the level of genetic diversity in adapted winter oilseed rape breeding material with double low seed quality (zero erucic, low glucosinolate content) is relatively low due to a strong selection for these vital nutritional traits for the seed oil and meal, respectively. Oilseed rape (*Brassica napus L.*) is a major oil crop that also supplies proteins for the feed. Proteins represented the major class of storage compounds and their average amino acid composition was found to be very close to that of rapeseed in various environmental conditions. Indeed, oil-free rapeseed meal contains 38-40% of crude proteins that display a well-balanced amino acid composition with high levels of essential sulfur containing amino acids (Nesi *et al.*, 2008). Oil seed crops, particularly those containing high percentages of polyunsaturated fatty acids (PUFA) and vitamin E (tocopherols) are thought to show high antioxidant capacity. The consumption of foodstuffs rich in antioxidants provides protection against cancer and several diseases. Other classes of natural antioxidants found in all vegetable oils are tocopherols. High levels of α -tocopherol were found in soybean, corn, sesame and also rapeseed oils (Koski *et al.*, 2002). Moreover, both α - and γ -tocopherol with their important antioxidant properties in oilseed rape play an important role in the development of oilseed rape seeds (Volker *et al.*, 2004). Phenolic compounds exist widely in

plants. They are plant secondary metabolites, and they play an important role as defense compounds. Rapeseed contains more phenolic compounds than any other oilseed plant (Nowak *et al.*, 1992). The most significant of these are sinapic acid and its derivatives, most notably sinapine (Kozłowska *et al.*, 1990). Rapeseed phenolics include esterified phenolic acids, free phenolic acids, and insoluble-bound phenolic acids (Krygier *et al.*, 1982). The total contents of phenolic acids vary between 6400 and 18400 $\mu\text{g/g}$ depending on the variety of the plant and oil processing method. However, the content of phenolics in rapeseed flour is nearly 30 times higher than that of soybean. Currently, research is oriented towards an improvement in the yield quality of rapeseed oil. Hence, to find new source of edible oil with a desirable quality and possibly new material for breeding purposes, the present study was undertaken to evaluate the nutritional value of some selected oilseed rape (*Brassica napus L.*) cultivars.

2. MATERIALS AND METHODS

2.1. Chemicals

Folin-Ciocalteu's phenol reagent and gallic acid were obtained from Sigma Chemical Co., Ltd (St. Louis, MO, USA) and sodium carbonate from Riedel-de Haën. Acetonitrile, methanol and acetic acid were HPLC grade from Merck. Ferrous chloride and α -tocopherol were obtained from Sigma Chemical Co. (St Louis, USA). All other chemicals and reagents were of analytical grade.

2.2. Plant materials

Five cultivars of rapeseed (*B. napus*) were used in this study namely, pactol, silvo, topas (French origin cultivars), serw 4 and serw 6 (Egyptian origin cultivars). Seeds were obtained from Egyptian Ministry of Agriculture, and cultivated under Egyptian environmental conditions (Mekki, 2003), by Prof. Bahaa Mekki at the experimental farm of National Research Center, Egypt along a couple of successive winter seasons (2004-2005 and 2005-2006). When seeds matured, the plants were harvested and then seeds of uniform size were collected and kept in a cool, dry place until used for analysis as follows:

2.3. Total lipid extraction

Total lipid extraction was performed according to AOAC (2000); briefly, seeds were dried overnight at 50°C and ground to fine powder using a grinder prior to oil extraction. 10 g dry samples were extracted continuously in the Soxhlet apparatus using petroleum ether as the extraction solvent. The volume of solvent used was enough to soak the seeds completely. The number of reflux cycles was 50 and the complete extraction procedure lasted from 8 to 10 h. In turn, samples were extracted in

triplicate and then filtered through whattman filter paper No 1 in order to remove any solid particles. All collected filtrates were combined together and the solvent was evaporated under reduced pressure using a rotavapor model R-114 (Büchi, Flawil, Switzerland), with a water bath model B-480 (Büchi, Flawil, Switzerland) then the lipid fraction residues were weighed and dried for 1 h at 105°C. The residue was converted to its fatty acid methyl esters as described by Bhardwaj and Hamama (2000) with slight modification. 250-500 mg of lipids were mixed with 0.5 M sodium hydroxide in methyl alcohol, saponified by refluxing with heating, and then transesterified with boron trifluoride in methyl alcohol. After extraction and dehydration, the fatty acid methyl esters were analyzed by gas chromatography with a mass spectrometer (GC-MS).

2.4. Determination of fatty acid composition by (GC-MS)

A Trace GC Model 2000 gas chromatograph with a mass spectrometer (GC-MS), detector model (electron impact, 70eV) and a DB-5 (30 m by 0.25 µm by 0.25 mm) capillary column was used. Injection was carried out at 250°C. Helium was the carrier gas. The oven temperature was programmed from 180°C for 3 min, and then from 180 to 210°C at 25°C min⁻¹, from 210 to 280°C at 25°C min⁻¹. The final temperature was maintained for 3 min. The target fatty acids were identified by mass spectrometric in both SCAN (total number of ions) and selected ion monitoring (SIM) mode. Spectra of the fatty acids were obtained and compared with those in the US. National Institute of Standards and Technology (NIST) library according to Jiang *et al.* (2006).

2.5. Determination of glucosinolate profile

Glucosinolate were analyzed by HPLC at the Federal Agricultural Research Center (FAL), Institute of Plant Nutrition and Soil Science (FAL, Germany) according to Wathelet *et al.* (1991). About 100 mg dry sample of seeds was ground in a mixer (PT3000 Polytron-kinematica) for about 20 sec. The ground samples were extracted in boiling methanol (70%) in a water bath at 70°C for 20 min. Subsequently, the extract was centrifuged (1000 × g, 10 min) and the supernatant were collected. The pellet was re-extracted three times following the same procedure. An aliquot of the supernatant was loaded onto ion-exchange mini-columns (DEAE Sephadex A-25) and the glucosinolates were desulphated on-column without disturbing the resin surface and allowed to drain. Desulphation was carried out by the addition of 75 µl of purified sulphatase (E.C. 3.1.6.1, Sigma) solution. The column was capped for 12 h. The desulphoglucosinolates were eluted with water and separated by gradient system high performance liquid chromatography (Thermo Separation Products) using a Nova Pak C18 (5 mm) reverse phase

column. The filtrate was filtered and analyzed using a liquid chromatograph LaChrom (Merck Hitachi) coupled with a variable wavelength UV detector LaChrom L 7400. The desulphoglucosinolates were monitored by UV-absorption at 229 nm and quantified against the internal standard (sinigrin-Sigma). Identification of individual glucosinolates was done by comparing retention times with pure internal standards and expressed as µmol /g dw. The total glucosinolate content was computed as the sum of all the individual glucosinolate present in the sample.

2.6. Determination of amino acid composition

Amino acids were extracted from the seeds using modified methods as described by Cohen *et al.* (1990). Dry and defatted samples containing 50 mg protein were weighed in the ampoules and 5 ml of 6 N HCl were added. Ampoules were sealed under vacuum and the contents were digested at 110°C for 24 h. The sealed tubes were then opened and samples were filtered and the residue was filtered. Five ml of the filtrate were evaporated under vacuum at room temperature. The residue was dissolved in 5 ml sodium citrate buffer (pH 2.2) and the mixture was centrifuged at 15,000 × g 15 min at 4°C (Hermle Z323 K, Germany), filtered and made up to 50 ml with deionized water. The amino acids were separated by reverse phase HPLC using the method of Cohen *et al.* (1990) and modified by Puziah *et al.* (1998). The amino acids were detected using a Waters 486 Tunable Absorbance Detector at 254 nm. The mobile phase A of the gradient elution consisted of 940 ml of sodium acetate buffer containing 0.05% triethylamine (TEA), pH 5.7 with 60 ml acetonitrile (HPLC grade) and mobile phase B consisted of acetonitrile: water (60:40 v/v). Waters Pico-Tag free amino acid column, 3.9 x 300 mm was used for the analysis. The calculations were carried out according to the external standard.

2.7. Determination of total tocopherols

The modified Emmerie-Engel procedure for total tocopherols was used according to Backer *et al.* (1980). Tocopherol were extracted from seed material with acetone: methanol (7:3, v/v) and were determined by spectrophotometer (Unicam UV-300A model) based on ferrous -dipyridyl color reagent. Briefly, aliquots of the extracted solution were mixed with 1 ml of 2, 2'-dipyridyl reagent (0.125 g in 25 ml of absolute EtOH) then, a 1-ml portion of ferric chloride reagent (0.2 g in 100 ml of absolute EtOH) was added and the mixture was shaken for 10 sec. The absorbance of the mixture was read at 522 nm in a 1-cm cell 50 sec after adding the ferric chloride. A blank was run, using CHCl₃, 2, 2'-dipyridyl reagent, and ferric chloride reagent. The absorbance of this solution was measured at 522 nm against a blank. Then,

the standard curve was drawn and α -tocopherol contents in the extracts were calculated from the regression equation of the standard curve.

2.8. Determination of total phenolic content

Phenolic compounds were determined based on a method described by Singleton *et al.* (1999). Briefly, 1 ml of methanolic extract was mixed with 1 ml of Folin Ciocalteu reagent. After 3 min, 1 ml of saturated sodium carbonate solution (20%) was added to the mixture and adjusted to 10 ml with distilled H₂O. The reaction mixture was kept in the dark for 1 h with intermittent shaking. The absorbance was measured at 725 nm using a spectrophotometer. Phenolic contents were calculated on the basis of the standard curve for gallic acid (GAL). The results were expressed as mg of gallic acid equivalent per g of dry extract.

2.9. Statistical analysis

All tests were conducted in triplicate. Data are reported as means \pm standard deviation (SD). Analysis of variance and significant differences among means were tested by one-way ANOVA using the COSTAT computer package (Cohort Software, CA, USA). The least significant difference (LSD) at P = 0.05 level was calculated.

3. RESULTS AND DISCUSSION

3.1. Fatty acids

The results of the fatty acid composition analysis of five rapeseed cultivars are presented in Table 1. Oleic acid (18:1) was determined to be the pre-dominant fatty acid among all cultivars. Its amounts ranged from 56.31% (in pactol cultivar) to 58.67% (in serw 6 cultivar) but these differences were not statistically significant. For α -linolenic acid (18:3) acid, its highest level has been found in serw 4 (10.32%) whereas, its lowest amount has been detected in topas (8.83%). Other cultivars

have close levels of α -linolenic acid (pactol: 9.37%, silvo: 9.31%). Palmitic, stearic and gadoleic acids were determined in all cultivars. Their amounts ranged from 2.18-7.91, 11.09-14.93, and 0.93-1.69%, respectively. Similarly, the cultivars with the highest level of erucic acid were topas (0.91%) and silvo (0.69%); while those with the lowest level were serw 6 (0.15%) and pactol (0.17%). The total amount of unsaturated and saturated fatty acids and the ratio of saturated to unsaturated are given in Table (2). The highest percent of total saturated fatty acids (TS) was observed in the topas cultivar, (the value was 20.5%). The proportion of total unsaturated fatty acids (TUS) was higher in serw 6 and serw 4 (83.73% and 84.33% respectively) compared to topas (78.99%), while total saturated fatty acids in the same cultivars were lower (16.42% and 15.85%) compared to topas (20.5%). With regards to saturated fatty acids, other researchers have reported differences among cultivars. Auld *et al.* (1992) stated that a reduced level of polyunsaturated FA (especially linolenic acid, 18:3) and an increased content of monounsaturated FA (oleic acid, 18:1) provided higher oil stability and the resulting product can be used for food applications requiring high cooking and frying temperatures. Therefore, breeding rapeseed with high 18:1 and low 18:3 contents is a major goal. Additionally, the nutritional quality of the rapeseed oil can still be improved by increasing the dietary essential linoleic acid (C18:2) contents and decreasing the α -linolenic acid (C18:3) contents (Beare-Roger, 1988). The great variability in seed oil contents in Brassica cultivars showed their potential for use in future breeding programs and supported the findings of Getinet *et al.* (1997), Rabiee *et al.* (2004) and Ashraf *et al.* (1999) who also recorded great variations in seed oil contents among different Brassica species. An inverse relationship between monounsaturated (oleic acid) and polyunsaturated fatty acids (linoleic acid) in sunflower has been reported by Flagella *et al.* (2002), which were mainly attributed to the maturing of crops at low temperatures. Therefore, the findings of the present study are consistent with those of Flagella *et al.*

Table 1
Fatty acid contents (%) of different rapeseed (*Brassica napus L.*) cultivars

Cultivars	Relative % of fatty acid							
	Palmitic acid (16:0)	Stearic acid (18:0)	Oleic acid (18:1)	Linoleic acid (18:2)	α -Linolenic acid (18:3)	Gadoleic acid (20:1)	Erucic acid (22:1)	Nervonic acid (24:1)
Pactol	7.91 ^a \pm 0.19	11.09 ^c \pm 0.15	56.31 ^a \pm 2.03	13.41 ^a \pm 0.16	9.37 ^b \pm 0.10	1.40 ^b \pm 0.07	0.17 ^c \pm 0.01	0.34 ^a \pm 0.03
Silvo	6.44 ^b \pm 0.12	11.54 ^c \pm 0.19	57.00 ^a \pm 2.95	13.33 ^a \pm 0.33	9.31 ^b \pm 0.17	1.69 ^a \pm 0.04	0.69 ^b \pm 0.03	0.12 ^c \pm 0.03
Topas	5.57 ^c \pm 0.27	14.93 ^a \pm 0.52	57.49 ^a \pm 2.51	10.52 ^b \pm 0.26	8.83 ^b \pm 0.42	0.93 ^d \pm 0.05	0.91 ^a \pm 0.04	0.31 ^a \pm 0.02
Serw 4	3.59 ^d \pm 0.24	12.26 ^b \pm 0.48	58.48 ^a \pm 1.95	13.74 ^a \pm 0.39	10.32 ^a \pm 0.30	1.40 ^b \pm 0.13	0.21 ^c \pm 0.03	0.18 ^b \pm 0.02
Serw 6	2.18 ^e \pm 0.16	14.24 ^a \pm 0.42	58.67 ^a \pm 2.31	13.53 ^a \pm 0.65	10.03 ^a \pm 0.21	1.20 ^c \pm 0.12	0.15 ^c \pm 0.04	0.15 ^b \pm 0.02
LSD	0.36	0.72	4.32	0.71	0.48	0.16	0.05	0.112

Values are expressed as the means \pm SD of three independent assays. Values with different letters in the same column were significantly different (P \leq 0.05).

Table 2

Total saturated and unsaturated fatty acid contents of different rapeseed (*Brassica napus L.*) cultivars.

Cultivars	TS ¹	TMUFA ²	PUFA ³	TUS ⁴	US/S ⁵
Pactol	19.00 ^b ± 0.34	58.22 ^a ± 2.13	22.78 ^a ± 0.26	81.00 ^a ± 2.39	4.26 ^d ± 0.05
Silvo	17.98 ^b ± 0.07	59.53 ^a ± 3.03	22.64 ^a ± 0.50	82.17 ^a ± 3.52	4.57 ^c ± 0.21
Topas	20.50 ^a ± 0.79	59.64 ^a ± 2.62	19.35 ^b ± 0.68	78.99 ^a ± 3.30	3.85 ^e ± 0.01
Serw 4	15.85 ^c ± 0.72	60.27 ^a ± 2.11	24.06 ^a ± 0.69	84.33 ^a ± 2.80	5.32 ^a ± 0.06
Serw 6	16.42 ^c ± 0.58	60.17 ^a ± 2.47	23.56 ^a ± 0.86	83.73 ^a ± 3.33	5.10 ^b ± 0.03
LSD	1.02	4.53	1.14	5.63	0.18

¹Total saturated fatty acids, ²Total mono-unsaturated fatty acids, ³Polyunsaturated fatty acids, ⁴Total unsaturated fatty acids, ⁵Ratio of unsaturated fatty acids to saturated fatty acids. Values are expressed as the means ± SD. Values with different letters in the same column were significantly different (P ≤ 0.05).

3.2. Glucosinolate profile

Variations in the glucosinolate profile of different rapeseeds were observed as shown in (Table 3). The major glucosinolate found in the rapeseed cultivars were progoitrin (with the mean value 2.83 ± 0.041 µmol/g dw), gluconapin (with the mean value 1.58 ± 0.19 µmol/g dw) and glucobrassicinapin (with the mean value 0.44 ± 0.05 µmol/g dw). While other minor glucosinolates were gluconasturtiin (mean being 0.082 ± 0.048 µmol/g dw), 4-hydroxyglucobrassicin (mean being 0.038 ± 0.14 µmol/g dw) and epi-progoitrin (mean being 0.036 ± 0.005 µmol/g dw). However, low variations were found for total glucosinolate contents. The total glucosinolate contents ranged from 4.4 to 5.97 µmol /g dw in serw 6 and silvo respectively. Generally, the lowest total glucosinolate content was found in serw 6. Also, the same cultivar showed low progoitrin contents (aliphatic glucosinolate) and 4-hydroxyglucobrassicin (Indolyl glucosinolate).

This cultivar might be considered a useful rapeseed to include in the diet as it contains low specific glucosinolate. These contents are lower than those found in other *B. rapa* varieties (Kim *et al.*, 2003). The seed glucosinolate content has been drastically reduced to contents of 10 µmol g⁻¹ seed and lower (Raney *et al.*, 1999) by conventional plant breeding, taking advantage of spontaneously arisen mutants. Considerable effort has gone into breeding out glucosinolates from the seeds of commercial varieties of canola (*Brassica campestris*) and oilseed rape (*Brassica napus*) because these compounds reduce the nutritional value of rapeseed meal. However, this is detrimental because low glucosinolate contents of the cotyledons of emerging seedlings encourages feeding by slugs and other non-specialist herbivores

3.3. Amino acids content

The amino acids compositions (mg/100g N) in different rapeseed cultivars are illustrated in

Table 3

Glucosinolate (GLs) profiles of different rapeseed (*Brassica napus L.*) cultivars.

Glucosinolate (Abbreviation)	Trivial name	Glucosinolate (GLs) content (µmol/g dw)					
		Pactol	Silvo	Topas	Serw 4	Serw 6	Means
Aliphatic							
3-Methylsulfinylpropyl (3MSOP)	Glucobrassicin	0.01	0.00	0.00	0.00	0.00	0.002 ± 0.004
4-Pentenyl	Glucobrassicinapin	0.46	0.46	0.45	0.47	0.34	0.44 ± 0.054
2-(R)-2-Hydroxy-3-butenyl	Progoitrin	2.80	3.42	2.77	2.88	2.28	2.83 ± 0.41
2-(S)-2-Hydroxy-3-butenyl	epi-Progoitrin	0.04	0.04	0.03	0.04	0.03	0.036 ± 0.005
2-Propenyl	Sinigrin	0.00	0.02	0.00	0.00	0.00	0.004 ± 0.009
5-Methylsulfinylpentyl (5MSOP)	Glucoalyssin	0.00	0.00	0.01	0.00	0.00	0.002 ± 0.004
2-Hydroxy-4-pentenyl	Gluconapoleiferin	0.00	0.00	0.00	0.01	0.00	0.002 ± 0.004
4-Methylthiobutyl (4MTB)	Glucorucin	0.00	0.00	0.00	0.03	0.00	0.004 ± 0.009
3-Butenyl	Gluconapin	1.37	1.88	1.62	1.48	1.56	1.58 ± 0.19
Indolyl							
3-Indolylmethyl (I3M)	Glucobrassicin	0.02	0.01	0.00	0.00	0.00	0.006 ± 0.009
1-Meoxo-3-indolylmethyl (1MOI3M)	Neoglucobrassicin	0.00	0.00	0.00	0.00	0.01	0.002 ± 0.004
4-Hydroxy-3-indolylmethyl (4OHI3M)	4-Hydroxyglucobrassicin	0.06	0.03	0.04	0.04	0.02	0.038 ± 0.014
Aromatic							
2-Phenylethyl (2PE)	Gluconasturtiin	0.00	0.1	0.09	0.09	0.13	0.082 ± 0.048
4-Meoxo-2-Phenylethyl (4MO2PE)	4-Methoxygluconasturtiin	0.00	0.01	0.00	0.00	0.00	0.002 ± 0.004
Total µmol / g dw		4.75	5.97	5.02	5.01	4.40	5.03 ± 0.58

Mean ± SD of rapeseed cultivars.

Table (4). Basically there was no change in the total essential amino and non-essential acid contents, whereas a distinct difference in the amounts and types of amino acids among cultivars occurred. In serw 6, it is important to note that aspartic (17.91 mg/100g N), glutamic (8.94 mg/100g N), and arginine (7.90 mg/100g N) acids were detected in high concentrations compared to other amino acids. Aspartic and glutamic acids were the prevailing amino acids in all cultivars while tyrosine and isoleucine were in very small amounts. Quality breeding of oilseed rape has been largely orientated by nutritional concerns driven by consumer and food industry demands. Several promising attempts have been made in rapeseed to increase cysteine, methionine and lysine contents, with these two latter amino acids being essential (Altenbach *et al.*, 1992). Current goals for improving seed quality in rapeseed deal with the development of yellow-seeded cultivars that are high yielding and display higher levels of oil and proteins.

3.4. Total tocopherols

Total tocopherol content of rapeseed was assayed and data presented in Table (5). Wide variations in tocopherol content were observed among tested cultivars. Tocopherol contents ranged from 73.02 to 138.3 g/100 g oil. The maximum content was recorded in serw 6 (138.3 mg/100

g oil), followed by pactol (102.8 mg/100 g oil), while the minimum content was recorded in topas (73.02 mg/100 g oil). Significant differences in the content of tocopherol in rapeseed cultivars have been detected. Rapeseed oils are a good source of tocopherol which has varying antioxidative abilities. Besides this well characterized function as lipid-soluble antioxidants that are able to scavenge oxygen radicals and to quench singlet oxygen (Kamal-Eldin and Appelqvist, 1996), presumably tocopherols preserve the integrity of membranes by forming complexes with products of membrane lipid hydrolysis and have a role in the regulation of transcription and post-translational processes (Quinn, 2004). The content of tocopherols among different cultivars of seed oils is known to be representative, and related to the similar habitats where these species grow. Moreover, the qualitative and quantitative evaluations of tocopherols along with the prediction of other relevant seed quality traits like oil and glucosinolate content and fatty acid composition in Brassicaceae, could be important among the plants belonging to this family.

3.5. Total phenolic contents

The concentration of total phenolic compounds in rapeseed cultivars are shown in Table (5). This phytochemical content ranged from 28.0 to 35.4 mg/g dw. The significant variation in phenolic

Table 4
Amino acid composition of different rapeseed (*Brassica napus L.*) cultivars

Amino acids mg/ 100g N	Rapeseed cultivars				
	Pactol	Silvo	Topas	Serw 4	Serw 6
Isoleucine (Ile)	3.60 ± 0.21	3.63 ± 0.26	3.44 ± 0.26	3.29 ± 0.33	3.02 ± 0.23
Leucine (Leu)	7.29 ± 0.39	7.40 ± 0.40	7.47 ± 0.37	7.55 ± 0.35	7.62 ± 0.41
Lysine (Lys)	6.71 ± 0.47	6.41 ± 0.40	6.54 ± 0.33	6.82 ± 0.33	6.33 ± 0.41
Cysteine (Cys)	4.39 ± 0.12	4.50 ± 0.30	4.21 ± 0.19	4.49 ± 0.26	4.42 ± 0.29
Methionine (Met)	4.15 ± 0.18	4.93 ± 0.26	3.81 ± 0.17	3.91 ± 0.20	3.93 ± 0.28
Total sulphur amino acids	8.54	9.43	8.02	8.40	8.35
Tyrosine (Tyr)	2.71 ± 0.25	3.15 ± 0.25	3.30 ± 0.29	3.25 ± 0.28	3.27 ± 0.29
Phenylalanine (Phe)	4.10 ± 0.19	3.85 ± 0.27	3.94 ± 0.34	3.79 ± 0.33	3.66 ± 0.32
Total aromatic amino acids	6.80	7.00	7.24	7.04	6.93
Threonine (Thr)	4.24 ± 0.34	4.45 ± 0.30	4.97 ± 0.39	4.26 ± 0.37	4.68 ± 0.47
Valine (Val)	5.48 ± 0.29	5.36 ± 0.32	5.87 ± 0.38	5.75 ± 0.35	5.70 ± 0.32
Histidine (His)	4.50 ± 0.22	4.66 ± 0.32	4.60 ± 0.33	4.69 ± 0.31	4.75 ± 0.35
Total essential amino acids	47.16	48.34	48.15	47.80	47.38
Arginine (Arg)	7.75 ± 0.38	7.56 ± 0.45	7.59 ± 0.39	7.77 ± 0.38	7.90 ± 0.47
Aspartic (Asp)	17.61 ± 0.52	17.78 ± 0.62	16.83 ± 0.50	17.30 ± 0.69	17.91 ± 0.72
Glutamic (Glu)	8.61 ± 0.29	8.47 ± 0.42	8.76 ± 0.45	8.77 ± 0.48	8.94 ± 0.48
Serine (Ser)	5.86 ± 0.39	5.48 ± 0.42	5.53 ± 0.37	5.32 ± 0.32	5.09 ± 0.41
Proline (Pro)	5.04 ± 0.27	4.93 ± 0.37	4.52 ± 0.34	5.03 ± 0.38	4.85 ± 0.43
Glycine (Gly)	6.52 ± 0.28	6.89 ± 0.38	6.79 ± 0.39	6.08 ± 0.43	6.96 ± 0.36
Alanine (Ala)	5.47 ± 0.33	5.56 ± 0.38	5.82 ± 0.35	5.73 ± 0.38	5.47 ± 0.42
Total non-essential amino acids	56.86	56.67	55.84	56	57.12
Total amino acids	104.02	105.01	103.99	103.80	104.50

Values are expressed as the means ± SD of three independent assays.

Table 5
The content of total tocopherols
and total phenolic compounds in different
rapeseed (*Brassica napus L.*) cultivars.

Rapeseed cultivars	*Total tocopherols (mg/100g oil)	**Total phenolic (mg/g dw)
Pactol	102.8 ^b ± 0.75	34.5 ^a ± 0.59
Silvo	90.0 ^d ± 0.7	28.4 ^c ± 0.43
Topas	73.02 ^e ± 0.45	35.4 ^c ± 0.45
Serw 4	97.22 ^c ± 0.51	33.4 ^b ± 0.23
Serw 6	138.3 ^a ± 0.57	28.0 ^a ± 0.20
LSD (0.05)	1.03	0.84

Values are expressed as the means ± SD of three independent assays. Values with different letters in the same column were significantly different ($P \leq 0.05$).

content was observed in different cultivars of rapeseeds ($p \leq 0.05$). The highest phenolic content (35.4 mg/g dw) was found in 'topas' whereas the lowest value (28.0 mg/g dw) was detected in 'serw 6'. Rapeseed meal has a high content of phenolic acid esters, mainly sinapate esters, which have been shown to cause a dark color and a bitter taste in rapeseed meal and derived protein products. The most active rapeseed meal phenolic fraction contained several classes of phenolic compounds including phenolic acids, flavones and flavonols (Koski *et al.*, 2003). Potential genotypes with desired phenolic levels may play an important role in the future for the development of new and improved cultivars delivering potential health benefits. In this concern El-Beltagi *et al.* (2007) tested five flax cultivars (Sakha 1, Sakha 2, Giza 8, Lithuania and Aryana) to assess their nutritional value using different parameters such as protein profile, fatty acid content, unsaponifiable matter, total tocopherols, total phenolics and total flavonoids. The authors found that Sakha 1, Sakha 2 and Aryana had lower total phenolics (162, 205 and 185 mg/100 g) than Giza 8 and Lithuania (362 and 352 mg/100 g).

4. CONCLUSION

The results of this study showed that rapeseeds cultivars differed in their fatty acids, glucosinolate and amino acid contents. The proportion of total unsaturated fatty acids (TUS) was higher in serw 6 and serw 4 compared to topas, while total saturated fatty acids in the same cultivars were lower compared to topas. The lowest total glucosinolate content, progoitrin and 4-hydroxyglucobrassicin were found in serw 6. In serw 6, it is important to note that aspartic, glutamic, and arginine acids were detected in high concentrations compared to other amino acids. Also, significant differences were observed between cultivars concerning total tocopherols and total phenolics with serw 6 showing the highest level of total tocopherols, and the lowest total phenolic content among all cultivars. Thus, our

results could be used for developing canola cultivars with increased health promoting compounds. Also, all the European genotypes are Canola types (low in erucic acid and low in glucosinolate) and since Canadian breeders were the first to develop Canola types, most likely, European genotypes have canola genes incorporated through breeding. The magnitude, genotype x environment interaction comes into play since the material is introduced into a new environment. So, to use these introductions in the Egyptian breeding program, accessions need to be subjected to molecular marker analysis to discriminate among them.

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