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Variation of sesamin, sesamolin and tocopherols in sesame (*Sesamum indicum* L.) seeds and oil products in Thailand

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ABSTRACT

Sesame (*Sesamum indicum* L.) seed and oil contain abundant lignans, including sesamin, sesamolin and lignan glycosides. The aim of the present study was to determine sesamin, sesamolin and tocopherol contents in sesame seed and oil available in Thailand. The results showed that there was a large variation of sesamin and sesamolin contents in products. The distribution plot of sesamin and sesamolin contents in seeds showed that the mean values of sesamin and sesamolin were 1.55 mg/g (SD = 1.63; range n.d.–7.23 mg/g) and 0.62 mg/g (SD = 0.48; range n.d.–2.25 mg/g), respectively. The range of total tocopherols of these sesame lines was 50.9–211 μg/g seed. In commercial sesame oils, the ranges of sesamin and sesamolin were 0.93–2.89 mg/g oil and 0.30–0.74 mg/g oil, respectively, and tocopherol contents were 304–647 μg/g oil. The study reveals the extensive variability in sesamin, sesamolin and tocopherol contents among sesame products.

1. Introduction

Sesame (*Sesamum indicum* L.) has long been used, extensively, as a traditional food in eastern countries. Sesame seed and oil are widely used in cooking and as ingredients of sweet and confectionery foods. Sesame oil was the preferred cooking oil in India until the introduction of peanut oil (Moazzami, 2006). Sesame flavour is also very popular in Korean cuisine. The leaves of the sesame plant are also used in Korean food as a type of wrap, eaten with meat and other vegetables.

Sesame seeds contain a group of compounds, called lignans, which play an important role in health-promoting effects. Sesamin and sesamolin (Fig. 1) have been reported to have many pharmacological properties, e.g. antioxidant activity (Suja, Jayalekshmy, & Arumughan, 2004), antiproliferative activity (Yokota et al., 2007), enhancing antioxidant activity of vitamin E in lipid peroxidation systems (Ghafoorunissa, Hemalatha, & Rao, 2004), lowering cholesterol levels (Visavadiya & Narasimhacharya, 2008), increasing hepatic fatty acid oxidation enzymes (Ashakumary et al., 1999), and showing antihypertensive effects (Lee et al., 2004; Nakano et al., 2008) and neuroprotective effects against hypoxia or brain damage (Cheng, Jinn, Hou, & Tzen, 2006). Sesame ingestion (50 g sesame seed powder daily for 5 weeks) also positively affected sex hormones, antioxidant status, and blood lipids in postmenopausal woman (Wu, Kang, Wang, Jou, & Wang, 2006). Apart from sesame lignans, sesame seed and oil also contain other important biologically active compounds, such as vitamin E (tocopherol homologues), especially γ-tocopherol (Fig. 1; Williamson, Morris, Pye, Kamat, & Hensley, 2008; Hemalatha & Ghafoorunissa, 2004). Vitamin E occurs naturally as eight structurally related forms that include four tocopherols (α-, γ-, δ-, β-tocopherols) and four tocotrienols (α-, γ-, δ-, β-tocotrienols) (Dietrich et al., 2006). α-Tocopherol is the only form of vitamin E in vitamin supplements whereas γ-tocopherol is the predominant form of vitamin E in the US diet (Jiang, Christen, Shigenaga, & Ames, 2001). γ-Tocopherol has many beneficial properties, such as antiproliferative effects in human cancer cells, e.g. prostate cancer and breast cancer (Guthrie, Gapor, Chambers, & Carroll, 1997; Gysin, Azzi, & Visarius, 2002), anti-inflammatory activity (Jiang & Ames, 2003) and partial prevention of age-associated transcriptional changes in heart and brain of mice (Park et al., 2008). In addition, Cooney, Custer, Okinaka, and Franke (2001) found that the consumption of moderate amounts

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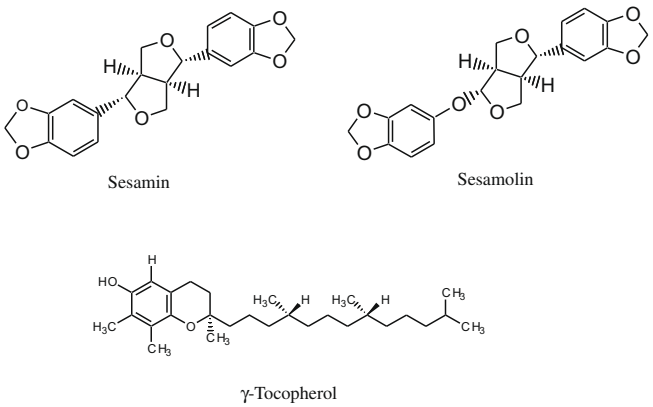

Fig. 1. The structures of sesamin, sesamolin, and γ-tocopherol.

of sesame seeds appeared to significantly increase plasma γ-tocopherol and alter plasma tocopherol ratios in humans.

Since sesamin, sesamolin and tocopherol are responsible for several pharmacological activities ascribed to sesame products, it is important to determine the variability in the contents of these antioxidant compounds among sesame lines and products. There are some reports of sesamin and sesamolin analysis in sesame seeds and oils (Wu, 2007; Kim, Lee, & Lee, 2006; Shirato-Yasumoto, Komeichi, Okuyama, & Horigane, 2003). In Thailand, sesame seed and oil are widely used for cooking and in cosmetic preparations (e.g. shampoo, lotion, cream and soap). Generally, breeding programmes for sesame have focused mostly on the crop's productive capacity and high oil contents (Arslan, Uzun, Ülger, & Çağirgan, 2007; Baydar, Marquard, & Turgut, 1999). Recently, the functional activities of lignans and tocopherols presented in sesame have become of major interest. The determination of lignans and tocopherols in a large number of samples could be beneficial for the selection of sesame lines for breeding programmes to develop the best sesame line with the highest contents of different major health-promoting compounds in the future. Therefore, this study aimed to determine the contents of lignans (sesamin and sesamolin) and tocopherols in sesame seeds (both landrace and developed lines) and commercial oil products available in Thailand.

2. Materials and methods

2.1. Sesame seed and oil products

Air-dried *S. indicum* L. seeds (% moisture = 5.4 ± 0.8%), both landrace and developed lines (black and white sesame seeds) in Thailand and other countries, were gifted from Prof. Wasana Wongyai, Kasetsart University, Bangkok, Thailand (58 lines). Sesame oils were purchased from local markets in Thailand. The names of sesame lines and characteristics of each line are shown in Table 1.

2.2. Chemicals

Sesamin and sesamolin, used as reference standards in this study, were purified and identified using TLC, UV spectrum, IR and NMR by the Laboratories of Pharmacology and Natural Products, Chulabhorn Research Institute. The extraction and purification of these two compounds were modified from the previous published methods of Shimizu et al. (1991) and Marchand, Kato, and Lewis (1997). Gallic acid, γ-, β-, and α-tocopherols, sodium carbonate and Folin–Ciocalteu's phenol reagent were purchased from Sigma Chemical Company (St. Louis, MO, USA). HPLC grade methanol, acetonitrile and nitric acid were obtained from Merck (Darmstadt, FR, Germany). Milli-Q water (Millipore, Bedford, MA, USA) was used throughout this experiment.

2.3. Sample preparation and analysis

2.3.1. Sample extraction for sesamin and sesamolin determination

Air-dried sesame seeds were ground into powder and weighed (0.4–0.5 g) into 15 ml plastic tubes (two replicates per sample). Of 80% methanol (5.0 ml) was added and the whole extracted for 30 min. The samples were then centrifuged at 2000*g*, for 3 min at 25 °C. The supernatant was transferred into a 10 ml volumetric flask. The residues were then re-extracted with 4.0 ml of 80% methanol. All extracts were combined, volume adjusted with 80% methanol, and filtered through a 0.45 μm PVDF membrane (Chrom Tech, Apple Valley, MN) prior to HPLC analysis. For extraction of sesame oil, the oil sample (~4.0 ml) was weighed (3.5–4.5 g) into 15 ml plastic tubes (two replicates per sample), and the whole extracted with 80% methanol by the same process as in the previous seed extraction.

2.3.2. Sample extraction for tocopherol determination

Samples of sesame oils were accurately weighed (100 mg) and dissolved in 5.0 ml of ethanol. Vortex-mixed samples were filtered with a 0.45 μm Nylon membrane (Chrom Tech, Apple Valley, MN) and injected into an HPLC column. Sesame seeds were ground into powder and accurately weighed (300 mg) and 5.0 ml of ethanol was added to each sample. Samples were agitated for 30 min and then centrifuged at 2000*g* for 5 min. The supernatant was transferred into a 10 ml volumetric flask and the residue was re-extracted with 5.0 ml of ethanol. All extracted solutions were combined and filtered through a 0.45 μm Nylon membrane prior to HPLC analysis. Care was taken to exclude air and light exposure of the sample and standard solutions throughout the analytical procedure.

2.3.3. HPLC analysis for sesamin and sesamolin

The HPLC analysis of sesamin and sesamolin was performed using the external standard method by Agilent 1100 high-performance liquid chromatography (Agilent Technologies, Baudrats, Germany) with a thermostatically controlled column oven, a binary pump, and a diode-array detector. A reversed-phase column, Hypersil BDS C_{18} 5 μm, 150 × 4 mm i.d. (Thermo Electron Co., Southend-on-Sea, UK), was used in this study. The mobile phase consisted of water (solvent A) and methanol (Merck, Darmstadt, Germany) (solvent B) with a gradient system: 0 min, 5%B; 0–5 min, 5–18%B; 5–10 min, 18–35%B; 10–15 min, 35–62%B; 15–18 min, 62–80%B, 18–22 min, 80%B; 22–23 min, 80–5%B, and equilibrated at this condition (5%B) for 3 min at 25 °C. The flow rate was 1.0 ml/min (injection volume 20 μl) with detection at 280 nm. Total run time was 26 min. Lignan glucosides were identified by collecting the peaks from HPLC and analysis using a mass spectrometer (MicroTOF, Bruker Daltonics, Bremen, Germany).

2.3.4. HPLC analysis for tocopherols

The simple and rapid reversed-phase liquid chromatography method for determination of tocopherols in sesame seed and oil was modified from Gliszczyńska-Świgło and Sikorska (2004) who determined tocopherols in edible plant oils (corn, peanut, grapeseed, rapeseed, sunflower, soybean and olive oil). HPLC analysis of tocopherols was performed using the external standard method by Agilent 1100 high-performance liquid chromatography (Agilent Technologies, Baudrats, Germany) equipped with Zorbax Eclipse XDB-C18 (4.6 × 150 mm, 5 μm) fitted with an Eclipse XDB-C18 (4.6 × 12.5 mm) guard column (Agilent Technologies, Santa Clara, CA). For determination of tocopherols in seeds and oils, a mobile phase consisting of 50% acetonitrile (A) and 50% methanol (B) was used with a flow rate of 1.0 ml/min. The column compartment

Table 1
The contents of tocopherols in sesame seed lines.

Name	Sources	Seed colour	Tocopherol contents (μg/g seed)			
			δ-Tocopherol	γ+β-Tocopherol	α-Tocopherol	Total tocopherol
China[a]	People's Republic of China	Black	0.72 ± 0.14	115 ± 10.04	ND	108 ± 4.44
China[a] (WH)	People's Republic of China	White	2.99 ± 0.01	191 ± 1.65	ND	194 ± 1.66
High Lignan[a] (WH)	People's Republic of China	White	ND	133 ± 0.05	ND	133 ± 0.05
Lao[a]	Lao People's Democratic Republic	Black	1.59 ± 0.20	105 ± 7.32	ND	107 ± 7.53
Lao[a]	Lao People's Democratic Republic	White	0.95 ± 0.03	84.6 ± 1.61	ND	85.6 ± 1.64
Guatemala[a]	Guatemala	White	1.30 ± 0.09	168 ± 3.26	ND	169 ± 3.34
Chachursoa[b]	Thailand	Black	ND	50.9 ± 1.00	ND	50.9 ± 1.00
Nakhonsawan[c]	Thailand	Black	0.55 ± 0.04	123 ± 1.72	ND	123 ± 1.76
Maehongson[c]	Thailand	Black	0.84 ± 0.02	86.7 ± 3.64	ND	87.5 ± 3.66
Maehongson[c]	Thailand	White	ND	99.8 ± 3.36	ND	99.8 ± 3.36
Saraburi[c]	Thailand	White	ND	69.7 ± 6.42	ND	69.7 ± 6.42
Nan[c]	Thailand	White	0.93 ± 0.10	114 ± 4.31	ND	115 ± 4.40
Loei[c]	Thailand	White	1.87 ± 0.00	121 ± 1.51	ND	122 ± 1.51
Chaiyaphum[c]	Thailand	White	ND	71.4 ± 1.48	ND	71.4 ± 1.48
Khoksamrong[c]	Thailand	White	0.63 ± 0.03	93.0 ± 1.97	ND	93.7 ± 1.99
Ubon Ratchathani-1[d]	Thailand	Red	ND	55.2 ± 2.58	ND	55.2 ± 2.58
Bantai, Black[c]	Thailand	Black	1.00 ± 0.14	105 ± 4.05	ND	106 ± 4.19
Bantai, Yellow[c]	Thailand	Yellow	6.65 ± 0.02	195 ± 0.60	9.22 ± 0.23	211 ± 0.86
MR-13[d]	Thailand	Brown	ND	85.3 ± 2.45	ND	85.3 ± 2.45
DamdangKaset[c]	Thailand	Black-red	0.62 ± 0.04	111 ± 1.95	ND	112 ± 2.00
Tongkam KUNS-7003[e]	Thailand	White	ND	91.6 ± 2.53	ND	91.6 ± 2.53
KU-18[e]	Thailand	Black	0.54 ± 0.04	108 ± 4.40	ND	108 ± 4.44
KU-19[e]	Thailand	White	1.28 ± 0.03	114 ± 2.35	ND	115 ± 2.38
KU-20[e]	Thailand	White	ND	60.3 ± 5.45	ND	60.3 ± 5.45
C-Plus-1[e]	Thailand	White	0.80 ± 0.00	98.3 ± 0.45	ND	99.1 ± 0.45
C-Plus-2[e]	Thailand	White	ND	82.0 ± 0.05	ND	82.0 ± 0.05
KUns-7020[e]	Thailand	White	ND	95.6 ± 2.85	ND	95.6 ± 2.85
KU-Aox-1[e]	Thailand	White	ND	72.7 ± 2.03	ND	72.7 ± 2.03
KU-Aox-2[e]	Thailand	White	ND	63.6 ± 3.77	ND	63.6 ± 3.77
Aox9001-2[e]	Thailand	Black	ND	69.7 ± 5.65	ND	69.7 ± 5.65
A7240-4[e]	Thailand	White	ND	89.9 ± 2.14	ND	89.9 ± 2.14
A7242-1[e]	Thailand	White	2.55 ± 0.04	193 ± 1.65	ND	195 ± 1.69
A7242-2[e]	Thailand	White	0.79 ± 0.03	146 ± 0.97	ND	146 ± 1.00
A7242-3[e]	Thailand	White	0.88 ± 0.01	145 ± 0.04	ND	146 ± 0.03
A7242-6[e]	Thailand	White	0.67 ± 0.04	132 ± 4.11	ND	133 ± 4.15
A7242-7[e]	Thailand	White	ND	154 ± 0.69	ND	154 ± 0.69
A7247-3[e]	Thailand	White	1.35 ± 0.04	173 ± 0.20	ND	174 ± 0.24
A7248-8[e]	Thailand	White	0.58 ± 0.06	130 ± 3.36	ND	131 ± 3.42
A7249-1[e]	Thailand	White	–	–	–	–
A7249-2[e]	Thailand	White	–	–	–	–
A7249-3[e]	Thailand	White	0.95 ± 0.04	135 ± 2.33	ND	136 ± 2.37
A7249-4[e]	Thailand	White	ND	84.8 ± 2.30	ND	84.8 ± 2.30
A7249-5[e]	Thailand	White	ND	104 ± 7.99	ND	104 ± 7.99
A7250-3[e] (BR)	Thailand	Brown	–	–	–	–
A7250-8[e]	Thailand	White	0.61 ± 0.02	94.0 ± 0.06	ND	94.6 ± 0.08
A7251-7[e] (BR)	Thailand	Brown	2.73 ± 0.04	190 ± 0.85	ND	192 ± 0.89
A7251-9[e]	Thailand	White	1.73 ± 0.15	119 ± 7.67	ND	120 ± 7.81
A7252-4[e]	Thailand	White	0.95 ± 0.03	84.6 ± 1.61	ND	82.0 ± 0.05
A7252-6[e]	Thailand	White	0.89 ± 0.11	111 ± 7.91	ND	112 ± 8.01
TQ7001-3[e]	Thailand	Black	ND	71.6 ± 8.91	ND	71.6 ± 8.91
TQ7001-5[e]	Thailand	Black	ND	98.4 ± 4.75	ND	98.4 ± 4.75
TQ7002-5-1[e]	Thailand	Black	ND	76.5 ± 1.91	ND	76.5 ± 1.91
TQ7003-5-2[e]	Thailand	White	ND	78.4 ± 0.59	ND	78.4 ± 0.59
TQ7003-2[e]	Thailand	White	ND	71.3 ± 3.45	ND	71.3 ± 3.45
TQ7003-3[e]	Thailand	Black	ND	88.6 ± 2.77	ND	88.6 ± 2.77
TQ9005[e]	Thailand	Black	ND	72.6 ± 0.73	ND	72.6 ± 0.73
TQ9009-1[e]	Thailand	Black	ND	85.6 ± 0.76	ND	85.6 ± 0.76
TQ9009-2[e]	Thailand	Black	ND	71.6 ± 2.10	ND	71.6 ± 2.10

ND = not detectable (detection limit = 0.01 μg/ml for δ-tocopherol and 0.05 μg/ml for α-tocopherol); for A7249-1, A7249-2, and A7250-3, there were not enough samples for analysis.

[a] Introduction.
[b] Wild species.
[c] Thailand landraces.
[d] Field Crop Research Institute (Ubon Ratchathani).
[e] Kasetsart University Sesame Breeding Program.

was controlled at 25 °C and the injection volume was 20 μl. The eluate was detected using a fluorescence detector set at an emission wavelength of 325 nm with an excitation wavelength at 295 nm. This RP-HPLC-FLD method can be used for the routine analysis of tocopherol homologues in sesame and other edible plant oils.

2.3.5. Preparation of sesame seed for trace element determination

Sesame seed powders were accurately weighed (0.25 g) into PTFE vessels. Two millilitres of H_2O_2 and 6.0 ml of concentrated HNO_3 were added to the samples. The vessels were closed and placed on the rotating turntable of a microwave oven, and then the digestion process was started. The digestion was done at

11.72 bar and 190 °C over 30 min and then maintained at 190 °C for 40 min. After microwave digestion, the digested solutions were filtered through filter paper (Whatman No. 42) and diluted to 50 ml with de-ionised water. All samples were then analysed for Ca, Mg, Se, Mn, Pb, Cd and As by ICP-MS (Agilent Technologies 7500c, Palo Alto, CA), following the previous method reported by Nookabkaew, Rangkadilok, and Satayavivad (2006).

2.3.6. Determination of total phenolic content in sesame seed extract

One hundred grammes of sesame seed powders were extracted with 500 ml of 80% methanol for 30 min (extraction ratio 1:5). The extract was then filtered and collected. The residue was re-extracted twice with 500 ml of 80% methanol, each time. The extracts were combined, and then concentrated using a rotary evaporator (temperature < 40 °C). The percentage yield of extract was 4.7–6.2%, depending on sesame lines. Total phenolic content was determined using Folin–Ciocalteu reagent, following the method of Singleton, Orthofer, and Lamuela-Raventos (1999) and SOP for total phenolic content, from the Botanical Research Center, Purdue University and University of Alabama-Birmingham (SOP No. CB0101, 2001), with some modifications. Aliquots of 50 μl of sesame seed extract (40 mg/ml in 80% methanol) were placed in screw-cap test tubes containing 3.95 ml of distilled water. Folin–Ciocalteu reagent (250 μl) was then added and, after 1 min, 750 μl of sodium carbonate solution (20%) were added. The tube contents were mixed thoroughly by inverting several times and then allowed to stand at room temperature for 2 h. The absorption was measured at 760 nm. The same solution, without the plant extract solution, was used as a blank solution. Total phenolic content was expressed as gallic acid equivalents in milligrams per gramme of dry material.

3. Results and discussion

3.1. Determination of sesamin and sesamolin in sesame seeds and oils

The HPLC analysis indicated that the main sesame lignans in seeds and oils were sesamin and sesamolin (Fig. 2). In addition, sesame samples also contained some lignan glucosides, e.g. sesaminol diglucoside and triglucoside. Sesamin and sesamolin are partially insoluble in water. Therefore, 80% methanol was used to extract these compounds from sesame seeds and oils. The retention time of sesamin was 20.1 min, sesamolin 20.5 min, and sesaminol glucosides 16.1–17.1 min. However, there were only two reference sesame lignan standards available in the market, sesamin and sesamol. Therefore, we extracted and purified sesamolin in our laboratory to use as a reference standard for the determination of sesamolin content in sesame samples. The percentage recoveries of sesamin and sesamolin, at the added concentrations of 5 and 10 mg, were 97–98% and 98–101%, respectively. Calibration

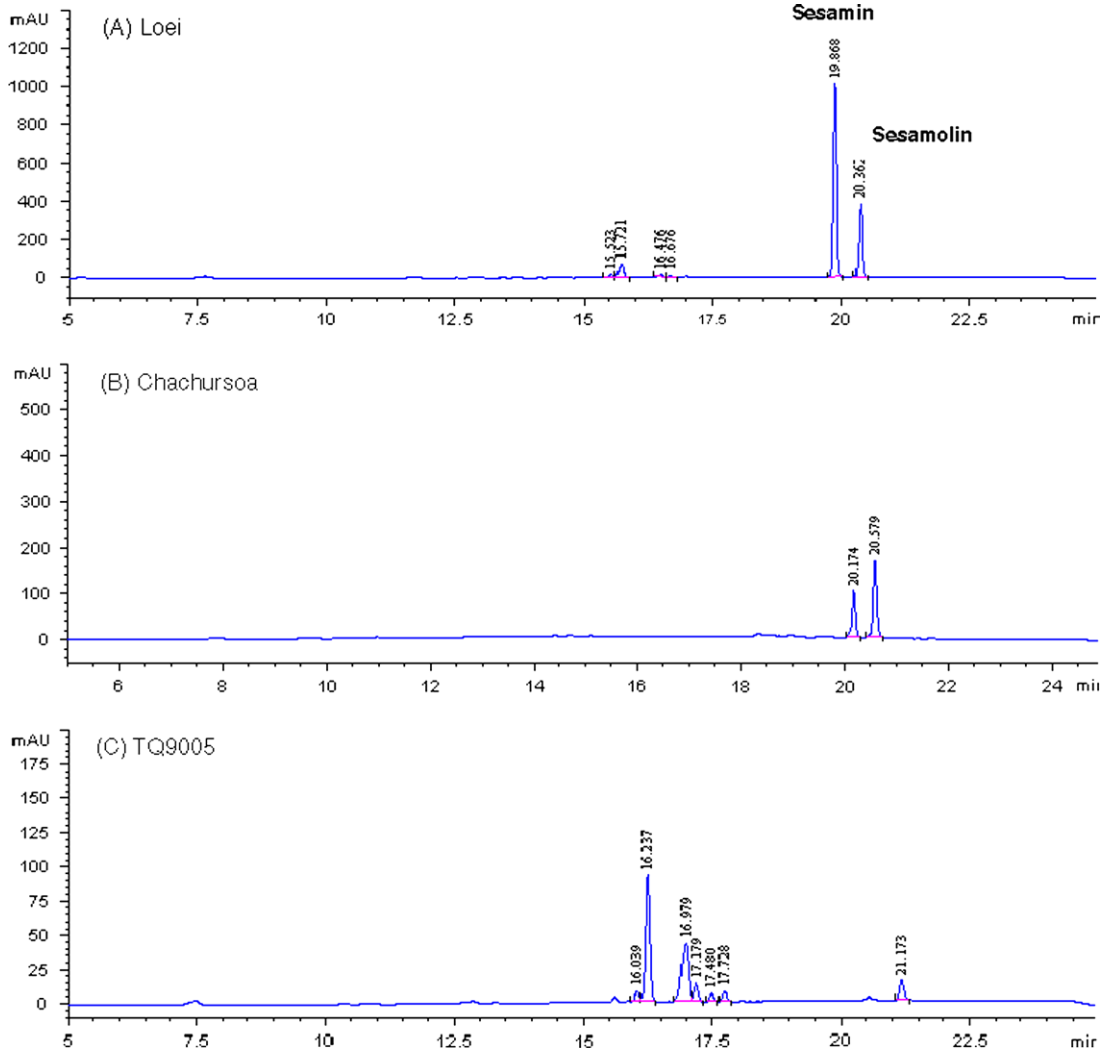

Fig. 2. HPLC fingerprints of sesame seed line (A) Loei, (B) Chachursoa, and (C) TQ9005.

curves were linear with R^2 = 0.9991 (sesamin; concentration range 0.05–0.8 mg/ml) and 0.9993 (sesamolin; concentration range 0.07–0.7 mg/ml). The precision of the method was determined for inter-day assay (3 days). The coefficients of variation (%CV) for sesamin and sesamolin were 1.5% and 1.9%, respectively. The detection limits of the method were 0.04 μg/ml for sesamin and 0.1 μg/ml for sesamolin. There was a large variation of sesamin and sesamolin contents in seeds. Generally, sesame seeds (both white and black seeds) contained sesamin as the highest, followed by sesamolin, and other glucosides (Fig. 2A). However, some sesame lines, such as Chachursoa (wild species), had a higher level of sesamolin (Fig. 2B) and TQ9005 had a high level of lignan glucosides with less sesamin and sesamolin (Fig. 2C). The differences in genetic, geographical and growing conditions, such as soil type, irrigation, fertiliser, and weather, may contribute to variable amounts of sesamin and sesamolin in seeds. In addition, seed size and position of capsules, including harvesting time, also affect the contents of these compounds in seeds. Sesame oil, also had high levels of both sesamin and sesamolin with less lignan glucosides or none.

For the landrace black seed lines, sesame seeds from Lao had the highest contents of both sesamin and sesamolin, followed by Nakornsawan and Bantai (black) (Thailand landrace) (sesamin range of 0.05–4.71 mg/g seed and sesamolin range of n.d.–1.28 mg/g seed) (Fig. 3A). For white sesame seeds, the high lignan line from China had the highest contents of sesamin and sesamolin, followed by Nan and Loei (Thailand landrace) (sesamin range of 0.74–7.23 mg/g seed and sesamolin range of 0.32–2.25 mg/g seed) (Fig. 3B). However, for black sesame seeds, Maehongsong and Chachursoa had a higher level of sesamolin than of sesamin (Fig. 3A). For the breeding lines 'A72–'; A7242-3, A7247-3 and A7242-7 had the highest contents of sesamin (range 0.05–5.85 mg/g seed) while A7249-2 and A7249-1 had the highest content of sesamolin

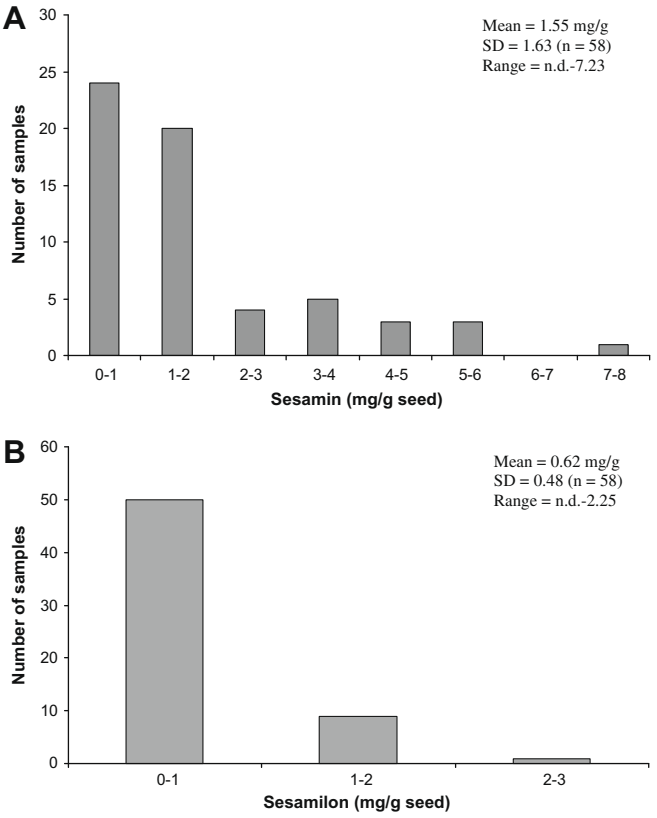

Fig. 4. The distribution plot of sesamin (A) and sesamolin (B) contents of total sesame seeds of 58 lines.

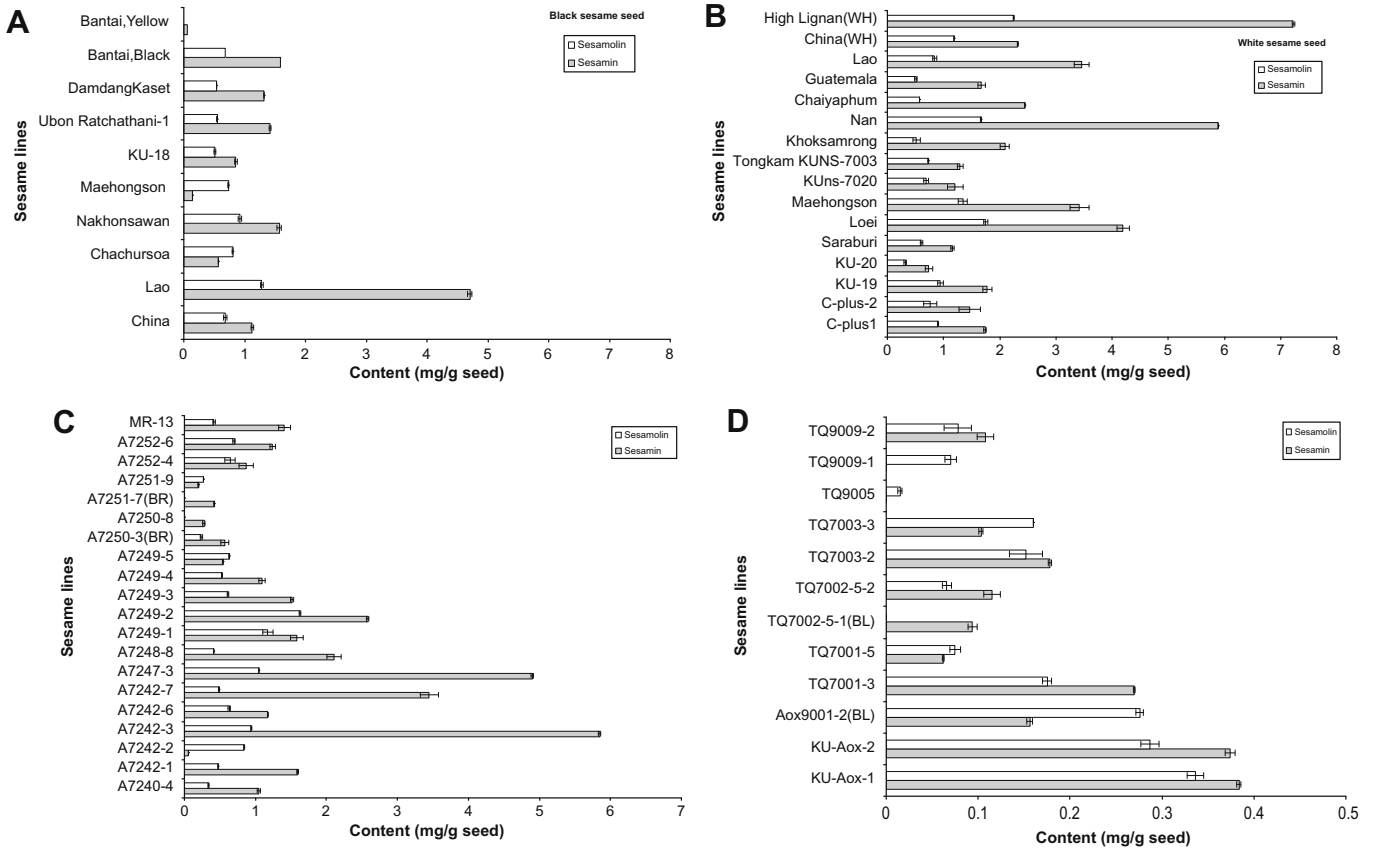

Fig. 3. Sesamin and sesamolin contents in (A) black sesame seed lines, (B) white sesame seed line, (C) line 'A72', and (D) lines 'TQ' and 'KU-Aox'.

(range n.d.–1.63 mg/g seed) (Fig. 3C). Lines A7242-2, A7249-5, and A7251-9 had sesamolin contents higher than sesamin but A7250-8 and A7251-7 (BR) did not contain any sesamolin. For breeding lines 'TQ-' and 'KU-Aox', both KU-Aox-1 and KU-Aox-2 showed the highest contents of sesamin and sesamolin (range n.d.–0.38 mg/g seed and n.d.–0.34 mg/g seed, respectively) (Fig. 3D). Line TQ7002-5-1 did not contain sesamolin while TQ9005 did not contain sesamin. It should be noted that the contents of sesamin and sesamolin in all of the 'TQ-' and 'KU-Aox' lines were lower than those of other sesame seed lines (<0.5 mg/g). Fig. 4A and B shows the distribution plot of sesamin and sesamolin contents in total sesame seeds of 58 lines. Mean values of sesamin and sesamolin were 1.55 mg/g (SD = 1.63) and 0.62 mg/g (SD = 0.48), respectively. The mean contents of both sesame lignans were classified as low content sesame samples (<10 mg/g) by Hemalatha and Ghafoorunissa (2004) from India. These sesamin and sesamolin values are in close agreement with the data reported in 65 sesame seed samples harvested in TX, USA, by Moazzami (2006) (1.63 ± 1.41 mg/g seed for sesamin and 1.01 ± 0.58 mg/g seed for sesamolin). In addition, the sesamin content in our study (range n.d.–7.23 mg/g seed) is also in the same range as the recent study by Williamson et al. (2008) who reported sesamin contents for 11 genotypes conserved in the United States Department of Agriculture (USDA), Agricultural Research Service (ARS) and Plant Genetic Resources Conservation Unit (PGRCU) in Griffin, GA, USA (0.67–6.35 mg/g). These data included one landrace from Thailand (PI 490026; sesamin = 3.17 mg/g). However, sesamin and sesamolin contents of 403 sesame landraces of Korea were 2.23 ± 0.92 mg/g and 1.74 ± 0.53 mg/g, respectively (Kim et al., 2006).

There was also a variation of sesamin and sesamolin in sesame oils. Oils 1, 2 and 5 are dark-brown in colour (roasted sesame seed) while other oils are light yellow in colour (cold-pressed oil). The range of sesamin content in oil was 0.93–2.89 mg/g of oil (average 1.67 ± 0.04 mg/g) and sesamolin was 0.30–0.74 mg/g of oil (average 0.58 ± 0.01 mg/g). The highest content of sesamin was found in oil 2 (2.89 ± 0.12 mg/g of oil) and the lowest in oil 1 (0.93 ± 0.01 mg/g oil) (Fig. 5A). The contents of both lignans in sesame oils in the present study were lower than the sesamin and sesamolin contents in sesame oil from India determined by using HPTLC (7.20 and 4.00 mg/g of oil, respectively) (Sukumar, Arimboor, & Arumughan, 2008). Our mean sesamin and sesamolin contents were also lower than those of Taiwanese sesame oil (9.47 ± 2.28 mg/g of oil and 1.74 ± 0.76 mg/g of oil, respectively). These differences may be due to the uses of different sesame lines and oil processing methods, including the techniques for phytochemical analysis in these studies.

Our HPLC method is a simple and practical technique for the quantification of sesamin and sesamolin contents in sesame seeds and oils. In addition, it can also be applied as a sesame fingerprinting for the quality assurance of other herbal products that contain sesame oil.

3.2. Determination of tocopherols in sesame seeds and oils

We have developed the RP-HPLC-FLD method which can determine tocopherol (*T*) in three different forms, α-T, γ+β-T, and δ-T in one sample injection. The percentage recoveries for δ-T and γ+β-T were 91.7% and 97.9%, respectively. Calibration curves were linear with $R^2 = 0.9999$ (δ-T; concentration range 0.1–25 μg/ml) and 0.9999 (γ+β-T; concentration range 0.1–25 μg/ml). The coefficients of variation (% CV) were 0.7% for δ-T and 0.8% for γ+β-T. The detection limit of the method was 0.01 μg/ml for both δ-T and γ+β-T. All sesame seeds had high levels of γ+β-T with little δ-T (Table 1) except Bantai (yellow) which had all forms of tocopherols. For Thailand landrace lines, Bantai (yellow) had the highest content of γ+β-T, 195.30 ± 0.60 μg/g of seed (total T = 211 ± 0.86 μg/g of seed), followed by Nakhonsawan and Loei (total T; 123 ± 1.76 and 122 ± 1.51 μg/g of seed, respectively). The range of total T in 58 sesame lines was 50.9–211 μg/g of seed. The results also indicated that, although the high lignan line from China (white seed) had the highest contents of sesamin and sesamolin, it had a lower level of tocopherol (133 ± 0.05 μg/g of seed) than had the other white seed line from China (194 ± 1.66 μg/g of seed). Chachursoa and Ubon Ratchathani-1 had the lowest total T levels (50.9 ± 1.00 and 55.2 ± 2.58 μg/g of seed, respectively). For breeding lines, A7242-1 and A7251-7 had the highest contents of total tocopherols (195 ± 1.69 and 192 ± 0.89 μg/g of seed, respectively). It should be noted that all A7242 lines contained tocopherols > 130 μg/g of seed while all TQ lines had total tocopherols < 100 μg/g of seed. Williamson et al. (2008) reported ranges of α-T, δ-T, and γ-T in sesame seeds of 11 genotypes, 0.034–0.175 μg/g, 0.44–3.05 μg/g, and 56.9–99.3 μg/g, respectively.

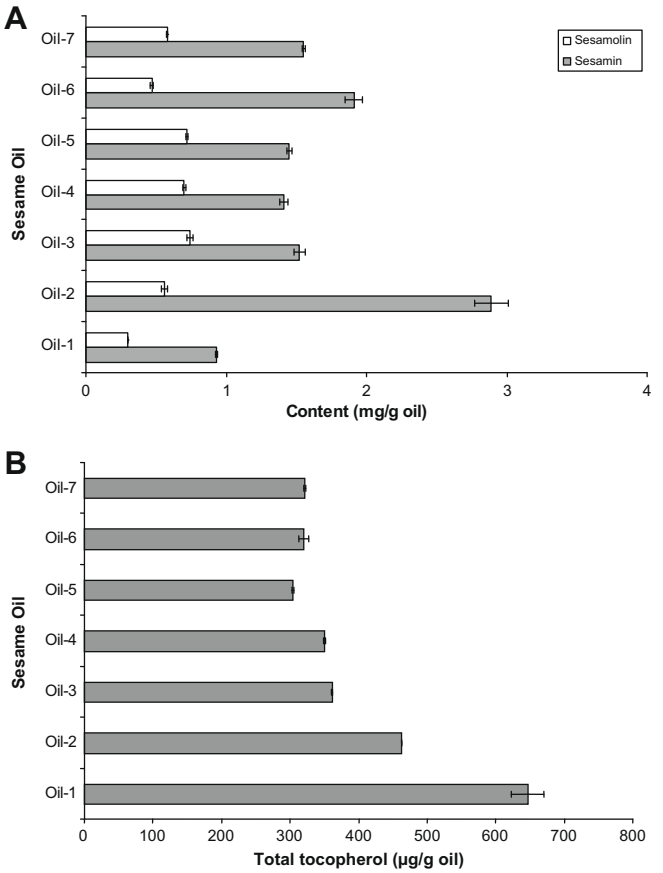

Fig. 5. Sesamin and sesamolin contents (A) and total tocopherol (B) in sesame oil products available in Thailand.

For sesame oils produced in Thailand, only oils 1, 2, and 5 contained all γ+β-T, δ-T, and α-T while others had only γ+β-T and δ-T. The total tocopherols of sesame oils were in the range of 304–647 μg/g of oil (average 395 ± 5.05 μg/g of oil) (Fig. 5B). As these three oils (1, 2, and 5) had dark-brown colours, the processing in oil production including the use of roasted sesame seed may affect the levels of tocopherol in oils or different lines (with different tocopherol levels) of sesame seeds. Sesame oils from cold-pressed processing (3, 4, 6, and 7) contained total tocopherol almost at the same level (334 ± 1.27 μg/g of oil). From our data, sesame oil 2 (from roasted seed) may be the best oil product as it had high levels of all antioxidant compounds, e.g. sesamin, sesamolin, and tocopherols.

The data from the present study indicated that sesame lines containing high levels of sesamin, sesamolin, and total tocopherols, such as Nan, Loei, A7242-3, A7242-7, and A7247-3, will be selected as superior lines for the breeding programme. Currently, we are undertaking field experiments for the development of new sesame lines with increases of sesamin, sesamolin and tocopherols in seeds.

3.3. Trace element analysis of sesame seeds

The results showed that white sesame seed, Loei (landrace line), contained more Ca (14,325 ± 337 mg/kg), Fe (1169 ± 149 mg/kg), and Se (0.63 ± 0.03 mg/kg) than those did black sesame seed, KU-18 (breeding line) (12,160 ± 306, 253 ± 21, and 0.16 ± 0.02 mg/kg, respectively). In addition, Loei contained some toxic metals, and especially more Cd (0.170 ± 0.00 mg/kg) than did KU-18 (about 7.7 times, 0.022 ± 0.00 mg/kg). Lead content was also higher in Loei (0.54 ± 0.08 mg/kg) than in KU-18 (0.12 ± 0.01 mg/kg). High Cd and Pb contents in sesame seed may result from the environment, e.g. soil, fertiliser and water where the plants were grown. Loei is the sesame landrace line usually grown in north-eastern Thailand while KU-18 is the sesame breeding line developed by Kasetsart University, Bangkok, Thailand. However, a larger number of sesame lines should be analysed for some toxic metals, especially Cd and Pb.

3.4. Determination of total phenolic content of sesame seed extracts

The results showed that the extract of white sesame seed (Loei) had the highest contents of both lignans (sesamin 30.6 ± 2.10 mg/g; sesamolin 20.8 ± 2.46 mg/g) and also total phenolics (13.2 ± 2.94 mg/g gallic acid equivalent). Black sesame seed extracts, KU-18 and TQ9005, had total phenolic contents of 7.20 ± 0.44 and 6.42 ± 0.62 mg/g gallic acid equivalent, respectively. Although the KU-18 extract (high sesamin) had higher levels of sesamin (23.1 ± 0.77 mg/g) and sesamolin (13.2 ± 0.61 mg/g), it had a total phenolic content at the same level as the TQ9005 extract (high lignan glucosides, low sesamin and sesamolin; 0.11 ± 0.04 and 0.45 ± 0.05 mg/g, respectively).

In conclusion, the evaluation of the antioxidant compounds, sesamin, sesamolin and tocopherols, present in 58 sesame seed lines in the present study, may be beneficial for the selection of the best sesame line in the breeding programme to develop a new sesame line with high levels of both lignans and tocopherols. The variations of lignans and tocopherols in sesame commercial oils should be also considered for possible health benefits and disease prevention when these products are used as functional foods.

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