academicJournals

Vol. 16(23), pp. 1308-1313, 7 June, 2017
DOI: 10.5897/AJB2016.15688
Article Number: 5629FC164640
ISSN 1684-5315

http://www.academicjournals.org/AJB

African Journal of Biotechnology

Full Length Research Paper

Evaluation of sesame (*Sesamum indicum* L.) germplasm collection of Tamil Nadu for α-linolenic acid, sesamin and sesamol content

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Received 21 September, 2016; Accepted 11 November, 2016

A germplasm collection of Tamil Nadu comprising of nine varieties and six landraces were analyzed for their ω3 fatty acid content, lignans such as sesamin and sesamol. The percentage of ALA ranges from 0.1 to 0.55%. A wide variation was observed in sesamol content. The highest sesamol content of 0.547 mg/g was found in a variety SVPR1. The sesamin content ranged between 2.03 and 6.45 mg/g. A negative relationship was observed between total oil content and ALA. White sesame seeds had higher sesamol content whereas black varieties revealed higher sesamin content.

Key words: ω3 fatty acid, sesame, sesamin, sesamol, Tamil Nadu germplasm.

INTRODUCTION

Sesame (*Sesamum indicum* L.) is an ancient oil yielding crop known as “Queen of Oilseeds” (Nayar, 1984). In the world production, Myanmar ranks first with 6, 20,000 metric tonnes (mt) of sesame while India ranks second with 6, 10,000 mt. In area under production, India ranks first with about 18, 20,000 ha whereas Myanmar stands second with 15, 70,000 ha (FAOSTAT, 2011). In India, Uttar Pradesh, Madhya Pradesh, Rajasthan, Orissa, Gujarat, Andhra Pradesh, Tamil Nadu and Maharashtra are the states which are suitable for sesame cultivation. Chemical composition of sesame seed shows 58% oil, 25% protein, 13.5% carbohydrate and 5% ash. Sesame oil has high percentage of desirable mono 18:1 and poly unsaturated fatty acids 18:2. The fatty acids present in sesame oil are oleic acid (43%), linoleic acid (35 %), palmitic acid (11%) stearic acid (7%) and trace amounts of α-linolenic acid which together comprises of about 96% of total fatty acids (Elleuch et al., 2007). Sesame is also used as nutraceutical and phytoceuticals as it has significant effect on preventing several diseases such as cancer, cardiovascular disease, atherosclerosis and the process of ageing in human being (Suja et al., 2004; Pathak et al., 2014a). Sesame seeds are described as the “seeds of immortality” for its resistance to oxidation and rancidity when stored at ambient temperature (Bedigian and Harlan, 1986; Pathak et al., 2014b).

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Table 1. Details of sesame varieties and landraces used in this study.

S/N	Crop/variety	Parentage	Place of collection	Colour of seed
1	TMV3	South Arcot variety x Malabar variety	Tindivanam	Dark brown
2	TMV4	Pureline selection sattur (local)	Tindivanam	Brown
3	TMV5	Purline selection from Srivaikuntam(local)	Tindivanam	Brown
4	TMV6	Selection from Andhra local	Tindivanam	Brown
5	TMV 7	Si 250 x ES 22	Tindivanam	Brown
6	CO 1	(TMV 3 x Si 1878) x Si 1878	Coimbatore	Dark brow/black
7	SVPR 1	Selection from “Western Ghats White”	Srivilliputhur	White
8	VRI 1	Pureline selection from Tirukattupali local	Vridhachalam	Brown
9	VRI 2	VS9003 X TMV6	Vridhachalam	Reddish brown
10	VS07023	Landraces	Vridhachalam	Brown
11	MD1	Landraces	Madurai	White
12	MD2	Landraces	Madurai	White
13	MD3	Landraces	Madurai	White
14	MD4	Landraces	Madurai	White
15	MD5	Landraces	Madurai	White

Sesamin is a major lignan compound present in sesame seeds. If the intake of sesamin is around 100 to 150 mg, it can preserve vitamin E in the human body. It also acts as a fatty acid metabolism modifier (Wang et al., 2012). Sesamolin is another major lignan present in the sesame seeds which is thermally unstable. During sesame oil processing, sesamolin is converted into sesamol and some other products by heat. Sesamol has very strong antioxidant activity. The sesamol content is very high in roasted sesame seed oil than unroasted sesame seed oil. It acts as a scavenger of free radicals (Singh et al., 2015). The present study was carried out to evaluate the α-linolenic acid (omega 3), sesamin and sesamol content in *Sesamum indicum* L. of Tamil Nadu germplasm for enhancing oil quality and anti-oxidant properties.

MATERIALS AND METHODS

Sesame seeds

Sesame seeds cultivated in Tamil Nadu were collected from Tamil Nadu Agricultural University (TNAU) and oil seed Research stations of TNAU located in Madurai, Coimbatore, Vridhachalam, Srivilliputhur and Tindivanam. Totally 15 accessions were collected. Out of them, nine were varieties and six of them were landraces. The parentage, seed colour and other information regarding the germplasm were collected from TNAU Agritech portal and presented in Table 1.

Lipid extraction

One gram of each seed variety was crushed in a mortar and pestle using liquid nitrogen to make a fine powder. Lipids were extracted from each seed variety with chloroform: methanol (2:1) as described by Bligh and Dyer (1959).

Percentage of oil in ground sample = [Weight of oil (g) / Weight of samples (g)] × 100

Fatty acid methyl esterification

Oil sample of each accession was converted into fatty acid methyl esters (FAME). Lipids were trans esterified with 6% potassium hydroxide in methanol. Lipids were incubated in water bath at 60°C for 3 h. The FAME were dried under nitrogen gas and dissolved with dichloromethane.

Gas chromatography analysis

FAME were analysed on Systronics gas chromatography instrument equipped with capillary column of 30 m × 0.3 mm internal diameter and Flame ionization detector. The oven was programmed to hold the temperature at 100°C and increased up to 200°C/5 min. Nitrogen was used as a carrier gas. The Flame ionization detector and injector temperature was maintained at 260°C. The peak was identified by comparison with the retention time of a commercial standard mixture of FAME (Supelco 37-component FAME mix Sigma, USA). Lipid analysis was performed independently for three times. Values were reported as average and standard deviation (n=3).

Sesamin and sesamol extraction

Sesame seeds were roasted and ground into powder using liquid nitrogen. Methanol was added and centrifuged at 4000 rpm for 5 min by a modified protocol (Rangkadilok et al., 2010). The supernatant was filtered through 0.22 µm nylon membrane filter (Pall Corporation).

HPLC analysis

High performance liquid chromatography was performed to analyze the lignans such as sesamin and sesamol. Sesamol and sesamin content were determined by direct injections using C18 reverse phase column equipped with UV detector at 290 nm. The mobile

Table 2. Total oil content and Alpha linolenic acid content of sesame germplasm in Tamil Nadu.

Sesame varieties	Total oil content (%)	Alpha linolenic acid content (%)
TMV3	51.09	0.17
TMV4	51.55	0.23
TMV5	51.91	0.24
TMV6	55.2	0.18
TMV7	50.15	0.55
CO1	51.56	0.19
SVPR1	51.83	0.13
VRI 1	51.19	0.18
VRI 2	51.75	0.13
VS07023	51.44	0.1
MD1	51.43	0.22
MD2	53.8	0.12
MD3	50.25	0.17
MD4	52.87	0.21
MD5	53.07	0.19

phase for both lignin was methanol: water (80:20) at the flow rate of 0.8 ml/min. Peaks were identified and quantified by comparison with commercial standard Sesamol (Sigma chemical,USA) and Sesamin (Cayman chemical, USA):

$$\text{Sample Concentration (mg/g of seeds)} = \frac{\text{Sample peak area} \times \text{standard concentration}}{\text{Standard peak area}}$$

RESULTS AND DISCUSSION

Total oil content

The total oil content and α-linolenicacid of sesame seeds from Tamil Nadu germplasm were analyzed and given in Table 2. The highest oil content of 55.2% was found in the sesame variety TMV6 whereas TMV7 recorded the lowest of 50.15%. The oil content of TMV7 was low when compared to other varieties of Tamil Nadu whereas TMV6 has recorded with high oil content and low yield of 611 kg/ha compared toTMV7 variety with an average yield of 802 kg/ha. TMV7 is a recently released variety which is also tolerant to root rot disease (Unal and Yalcin, 2008). Similarly, the oil content of four varieties of sesame collected from Turkey showed an average of 54.26% (Alege and Musapha, 2013). There was no significant difference in the oil content of Tamil Nadu sesame varieties in comparison to Turkish varieties. In another study, the oil content of 23 sesame samples from Nigeria were analyzed from 18 traditional and five developed accessions collected from 10 states in Nigeria. Among them, IBA II sesame variety from South West had high oil content of 58.85%. Nigerian varieties showed higher oil content than the Tamil Nadu varieties (Azeez and Morakinyo, 2014). The Nigerian sesame varieties were crossed for obtaining good oil, yield and fatty acid content, the highest oil content observed in S530 x PACH was only 57.58% (Mondal and Bhat, 2010). Genetic diversity based studies on Indian germplasm were used to estimate genetic differences among populations for seed yield and other characters (Bisht et al., 1998). However, there are no reports on oil content and quality parameters with genetic diversity on Indian germplasm. Genetic variation in Indian germplasm belonging to Rajasthan and North-eastern parts of India was observed through RAPD analysis to evaluate the genetic potential of Indian sesame varieties (Bhat et al., 1999). A RAPD study showed genetic distance coefficient of 0.35 among 58 accessions of sesame in Indian subcontinent and other countries (Bhat et al., 1999). The findings of this study would enable a breeder to link the genotype and oil parameters to the genetic cluster. Besides genetic factors, environmental factors and cultivation conditions may influence physiological and biochemical parameters (Bhat et al., 1999). Turkish sesame varieties showed variation coefficient ranging from 0.14 to 0.40 on RAPD analysis (Ercan et al., 2004). An ISSR based study on sesame germplasm from Korea and other countries showed minimum variation on polymorphism and distance coefficient of about 0.25 (Pham et al., 2011). Similarly, amplified fragment length polymorphism study also showed low genetic diversity of 0.14 to 0.21 among 32 sesame germplasm (Laurentin and Karlovsky, 2006). These suggest that the genetic diversity observed in several different germplasm collection are low hence other factors could play a role in oil content and yield.

Alpha linolenic acid content

Quantification of Alpha linolenic acid was performed in

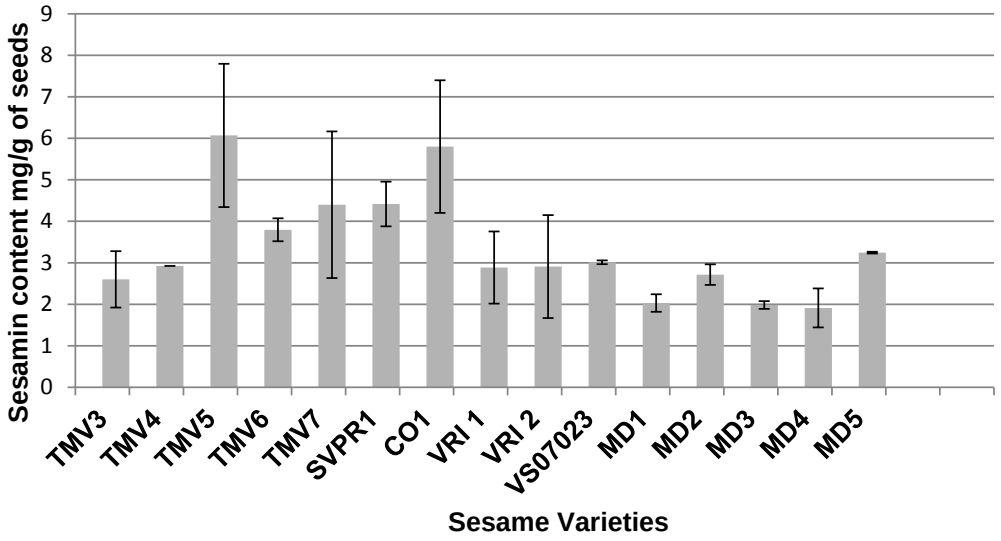

Figure 1. Sesamin content of *Sesamum indicum* L. germplasm collection of Tamil Nadu. Bars represent the amount of sesamin present in mg/g of seed. Error bars indicates mean ± SD, n=3 experiments.

Tamil Nadu sesame germplasm collection to evaluate the existing genotypes for improvement. Its quantity was very low in sesame accessions and ranged from 0.1 to 0.55 % in Tamil Nadu sesame seeds. The Maximum ALA content was found in TMV7 (0.55%) and the minimum ALA content was observed in VS07023 (0.1%) (Table 2). The AKT 101 variety (N 62 × N12-19) from Maharashtra had high ALA content of 1.17% (Mondal and Bhat, 2010). Sesame wild species like *S. mulayanum* (CAZRI, ABB-99-33)*, S. malabaricum* (IC 253971) and *S. alatum* (IC 253950) had no alpha linolenic acid (Mondal and Bhat, 2010). Domestication and selection might have enhanced ALA to a certain extent. The ALA content of sesame seeds from different countries like China, Egypt, India, Japan, Mexico, Myanmar, Thailand and Burkina Faso varies from 0.1 to 0.2% (Crew et al., 2006). The ALA content of Turkish varieties ranges from 0.53 to 0.60% (Unal and Yalcin, 2008). The Cambidi variety from Turkish had high level of ALA (0.60%). Worldwide studies indicate a wide variation in ALA content. A Chinese study on seed coat colour and ALA content reported no correlation between ALA content and seed colour (Philip, 2011).

Determination of sesamin content in sesame seeds

Sesaminis present in high levels compared to other lignans in sesame. The sesamin content of Tamil Nadu sesame varieties ranges from 2.03 to 6.45 mg/g of sesame seeds shown in Figure 1. The highest content of sesamin was found in TMV5 variety (6.45 mg/g of seeds). Similarly quantification of sesamin content was done in sesame seeds collected from different parts of North India and in commercial sesame oils. Sesame seeds from Assam show high sesamin content of 18.6 g/kg of oil. Low levels of sesamin content were found in Gujarat variety 3.1 g/kg of oil. Among commercial oils, AS brand (Agmark labeled oil) produced by crushing hulled sesame seeds had high level of sesamin content (9.0 g/kg).Similarly wide variation of sesamin content was seen in commercial oils of Assam and Gujarat sesame seeds (Hemalatha and Ghafoorunissa, 2004). These two states come under different geographical region with different soil and agro climatic conditions. Hence environmental factors could have a role on the sesamin content. However, Tamil Nadu germplasm grown in same geographical zone indicate higher difference suggesting a possible genetic contribution. Sesamin content in different colored of sesame seeds obtained from National Bureau of Plant Genetic Resources, New Delhi, India were analyzed. A white seeds variety, phuletil had high sesamin content of 38.43 g/kg whereas praghti another white seed variety showed the minimum level of sesamin 15.43 g/kg (Dar et al., 2014). Broad variation of sesamin content from 2.49 to 18.01 mg/g was observed in 215 sesame lines from core collection of China. They observed higher sesamin content in white sesame seeds than that of black, yellow, brown colours of sesame seeds (Wang et al., 2012). Sesamin content of black and white sesame seeds of Karnataka showed only slight variation of 3.031 and 4.663 g/kg of seed. Similarly commercially available sesame oils of Karnataka also showed slight variation in sesamin content of about 6.724 and 6.759 g/kg of sesame seed oil (Bhatnagar et al., 2013). In contrast, Tamil Nadu germplasm showed higher sesamin content in brown seeds (Figure 1). TMV5 and CO1 of Tamil Nadu sesame seeds recorded 6.45 and 5.9 mg/g of sesamin, respectively.

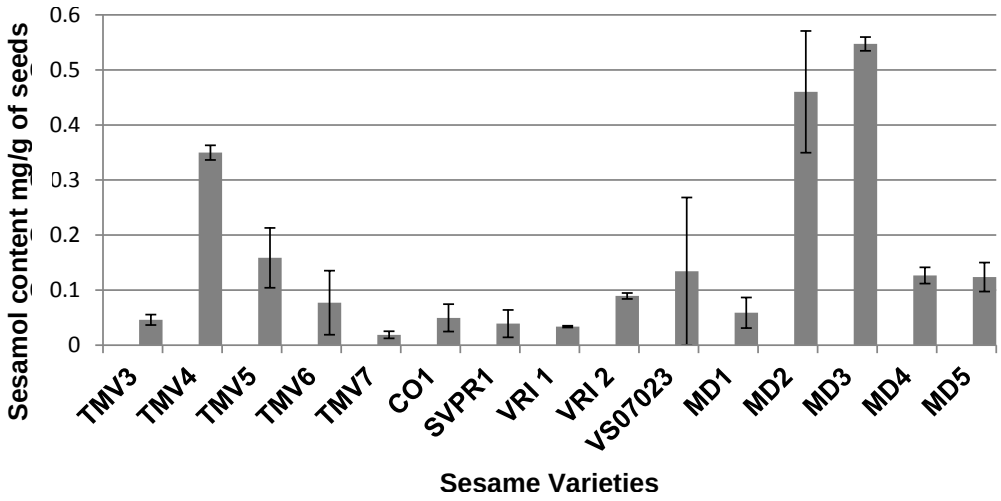

Figure 2. Sesamol content of *Sesamum indicum* L. germplasm collection of Tamil Nadu. Bars represent the amount of sesamol present in mg/g of seed .Error bars indicates mean ± SD, n=3 experiments.

Determination of sesamol content in sesame seeds

Sesamolin is another lignan present in sesame seeds. Sesamolin is hydrolysed during heating leading to the production of sesamol. Sesamol is the main reason for oxidative stability of sesame seed oil (Wan et al., 2014). Sesamol quantity in sesame varies between 0.03 and 0.55 mg/g of seeds in Tamil Nadu varieties and landraces (Figure 2). The highest sesamol content of 0.547 mg/g of seed was found in landrace MD3. The sesamol content of Burma black sesame seeds was less than 0.1 mg/g of seeds (Shyu and Hwang, 2001). The sesamol content of black sesame variety was 730 mg/kg and white sesame variety was found to be 68 mg/kg. The commercial expeller pressed sesame oils (CSO-A, CSO-B) showed variation of sesamol content ranging between 274 and 618 mg/kg of sesame oil (Bhatnagar et al., 2013). In Tamil Nadu accessions the sesamol content was high in white sesame seeds when compared to brown sesame seeds. Sesame oil produced by roasted sesame seeds at high temperatures before mechanical pressing can promote the flavor and colour of sesame oil (Singh et al., 2015). The roasted sesame seed oil has a higher sesamol content of 36 mg/kg of sesame seed when compared to unroasted which has only 7 mg/kg of seed (Wan et al., 2014). The sesamol content of unroasted sesame seeds was not detectable while those roasted at 180°C showed 235 mg/kg of sesame oil which further increased 10 fold when roasted at 250°C (Yoshida, 1994).

The total oil content of sesame germplasm of Tamil Nadu ranged between 50 and 56%. The alpha linolenic acid content of these sesame varieties was found to be 0.1 to 0.55% which is lesser than the Maharastra AKT101 which had 1.17% of ALA. Sesamin and Sesamol content were comparable to other germplasms from Turkey, China and North India. High sesamin content in Tamil Nadu varieties were discussed previously (6.45 mg/g) as they were close to *S. malabarium*. Quantification of these lignans would enable us to find the local varieties for further exploitation.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

ACKNOWLEDGEMENTS

The authors thank the Department of Biotechnology, PSG College of Technology and Department of Science and Technology, Government of India for providing the funding and infrastructure. They also thank Tamil Nadu Agricultural University and Research stations of TNAU for their generous contribution of sesame seeds.

Abbreviations

ALA, Alpha linolenic acid; **mt,** metric tonnes; **ha,** hectare.

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