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Variability in Coconut (*Cocos nucifera* L.) Germplasm and Hybrids for Fatty Acid Profile of Oil

S. Naresh Kumar*

Plant Physiology, Biochemistry and Post Harvest Technology, Central Plantation Crops Research Institute, Kasaragod 671 124, Kerala, India

ABSTRACT: Coconut oil, the main product of coconut fruit, is the richest source of glycerol and lauric acid and hence is called lauric oil. This paper reports the fatty acid profile of oil from 60 Talls, 14 Dwarfs, and 34 hybrids. These include collections from 13 countries covering a large coconut-growing area of the world, apart from the indigenous ones. Capillary gas chromatography analysis of oil indicated a wider variation for the fatty acid profile than earlier reported. Apart from this, for the first time other fatty acids such as behenic and lignoceric acids were detected. Oil from cultivars and hybrids of coconut has significantly differed, particularly for commercially important fatty acids such as lauric acid and unsaturated fatty acids. However, coconut oil seems to have a conserved fatty acid profile, mainly because of low unsaturated fatty acids, indicating the possibility of grouping cultivars on the basis of their fatty acid profiles. The cluster analysis based on fatty acid profile indicated grouping together of geographically and typically closely related cultivars. Cultivars with high concentrations of specific fatty acids can be of potential use for industrial exploitation, whereas those with high concentrations of short- and medium-chain fatty acids and unsaturated fatty acids are more suitable for human consumption. Cultivars and hybrids with high and low values for each of the fatty acids are also identified.

KEYWORDS: *coconut, copra, fatty acids, oil, oleochemicals, breeding*

■ INTRODUCTION

Coconut (*Cocos nucifera* L.), mainly a plantation crop of the humid tropics, is grown by ~11 million farmers, mostly resource poor, with major production being contributed by countries such as The Philippines, Indonesia, India, Brazil, and Sri Lanka. Globally it is grown on ~12.9 million hectares in over 90 countries, with annual production of ~61.2 billion nuts. India is one of the leading producers, with ~15.73 billion nuts from 1.894 million hectares at 8300 nuts ha^{-1} during 2009. Even though almost all parts of the coconut palm are economically important, with an annual global market of U.S. $~1.2 billion, oil is the most sought after high-value product with an annual global consumption of ~3.5 million tonnes. The European Union countries and the United States account for about 51 and 33%, respectively, of global imports.

Generally, coconut palm starts flowering about five years after field planting and flowers at almost monthly intervals. After fertilization, the fruit (nut) matures by about 12 months and has a solid endosperm. Dried mature endosperm, called copra, consists of about 64−70% of oil, yielding ~1.7−2.96 tonnes oil ha^{-1}; hybrids yield more oil.[1] In a developing coconut, fat synthesis starts 5 months after fertilization, and then solid endosperm begins to develop up to the 12th month. Coconut oil mainly contains saturated fatty acids such as caproic (C6), caprylic (C8), capric (C10), lauric (C12), myristic (C14), palmitic (C16), stearic (C18), and arachidic (C20). It also contains unsaturated fatty acids such as oleic (18C:1), palmitoleic (16C:1), and linolenic (18C:3) in lesser quantities.[1−6] Just-forming endosperm in tender nuts (5−6 months after fertilization) has more unsaturated fatty acids than mature nuts, and varietal variations also exist.[2] Lauric acid content among nine hybrids varied from 47.3 to 50.5%,[8] whereas it ranged from 44 to 49% in another set of 18 cultivars,[1] indicating the existence of variability for fatty acid composition.

Coconut oil and its derivatives are widely used in various industries. Fatty acids are important for industrial uses[3,4] and for human nutrition.[5,6] Because coconut oil is rich in lauric acid (~45%), industrially an important fatty acid, it is called lauric oil. Coconut oleochemicals are mainly used in soap and nonsoap detergent industries, for pretreatment of fibers in the textile industry, in painting and varnish manufacturing industries, and as surfactants in the petroleum industry.[7] Myristic acid, the second major fatty acid (~20%) in coconut oil, is widely used in the cosmetic industry. Methyl esters of coconut oil are used as biofuel as well. Furthermore, coconut oil is a base substance for many pharmaceutical products such as ointments and monolaurin. Oleochemicals of coconut oil are used in the food industry.[7]

On the basis of morphology, coconut cultivars are broadly classified into two groups, Tall and Dwarf. There have been several hybrids derived from crosses between Dwarfs and Talls. In general, Talls are high yielding and grown in large areas. For instance, in India, West Coast Tall, East Coast Tall, and Tiptur Tall are predominantly grown, and apart from these several Dwarf varieties, mainly for tender nut purpose, and hybrids are grown in relatively less area. Copra of different cultivars and thus oil gets mixed at various stages of processing. Earlier studies conducted on a few cultivars indicated the existence of

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significant variability in fatty acid profiles, providing large commercial scope for fetching premium prices for oils with desirable fatty acid profiles.[1,8,9] More awareness about health care led to consumer preference for oils with desirable fatty acid profiles, apart from industrial exploitation of individual fatty acids. However, no comprehensive information or database on fatty acid profiles of all cultivars and hybrids is available for optimal commercial and industrial exploitation. Thus, a study was conducted to analyze the fatty acid profile of 108 available and yielding cultivars and hybrids in India. Because variations in fatty acid profiles in oil crops arise due to genotypic[10−12] and weather factors,[2,13] sampling was done from the same season to delineate genotypic variability. This paper reports analysis of the variability in fatty acid profile of oil from 60 Talls, 14 Dwarfs, and 34 hybrids and identifies cultivars with extreme concentrations of each of the fatty acids. This information is anticipated to be useful to breeders in crop quality improvement programs, to the industry in assessing nutritional value from a human health point of view, and for specific commercial exploitation.

■ MATERIALS AND METHODS

Coconut cultivars (Tall and Dwarf types) and hybrids used for the analysis of fatty acid composition of oil include collections from Camaroon (CMR), Fiji Islands (FJI), Indonesia (IDN), India (IND), Guam (GUM), Jamaica (JAM), Kenya (KEN), Sri Lanka (SL), Malysia (MYS), Nigeria (NGA), Philippines (PHL), Solomon Islands (SLB), Seychelles (SYC), and Thailand (THA). The Talls (60) include (1) Andaman Giant, (2) Andaman Ordinary Tall, (3) Aliyarnagar Tall, (4) Arsampatti Tall, (5) Arsikeri Tall, (6) Assam Giant, (7) Assam Yellow, (8) Ayiramkachi, (9) Benaulim Tall, (10) Bengal Hazari (HAZT01; IND), (11) Bengal Selected (WBT 01; IND), (12) Benaulim Green Long (BGL; IND), (13) Borneo (BONT; Borneo), (14) Benaulim Red Round (BRR; IND), (15) British Solomon Islands Tall (BSIT; SLB), (16) Benaulim Yellow Long (BYL; IND), (17) Car Nicobar (CART; IND), (18) Cochin China (CCNT, VNM), (19) Ceylon Tall (SLT; SL), (20) East Coast Tall (ECT; IND), (21) Fijian Tall (FJT; FJI), (22) Federated Malayan States Tall (FMST; MLY), (23) Gangapani, (24) Gon Thembili Tall (GTBT; SL), (25) Guam Tall (GUAT; GUM), (26) Hazari Tall (HAZT; IND), (27) Jamica Tall (JAMT; JAM), (28) Java Tall (JVT; IDN), (29) Java Gaint (JGT; IDN), (30) Kamrupa (Assam Tall-AST; IND), (31) Kappadam Tall (KPDT; IND), (32) Karkar Tall (KKT; PNG), (33) Kenya Tall (KENT; KEN), (34) Klapawangi (KWGT; MYS), (35) Laccadive Ordinary (LCT; IND), (36) Lifou Tall (LFT; NCL), (37) Laccadive Micro Tall (LMT; IND), (38) Laccadive Small, (39) Mysore Tall, (40) Nufella (NUFT; NC), (41) Newguinea (NGAT; PNG), (42) Newe Huang (NWHT; NC), (43) Nizerian Tall, (44) Nugili Tall, (45) Philippines Lono Tall, (46) Philippines Ordinary Tall, (47) Rajapalayam (RJPT; IND), (48) Sakhigopal Tall (SKT; IND), (49) San Ramon Tall (SNRT; PHL), (50) Seychelles Tall, (51) Siam, (52) Spicata Tall (SPIT; WCT01; PHL), (53) Strait Settlement Apricot (SSAT; MYS), (54) Strait Settlement Green Tall (SSGT; MYS), (55) St. Vincent Tall (STVT; TTO), (56) Thailand Tall (THT; THA), (57) Tiptur Tall (TPT, IND), (58) Verrikobbari (VKBT; IND), (59) West Coast Tall (WCT; IND), and (60) Zanzibar Tall (ZAT; TZA). The Dwarfs (14) include (61) Chowghat Green Dwarf (CGD; IND), (62) Chowghat Orange Dwarf (COD; IND), (63) Cameroon Red Dwarf (CRD; CMR), (64) Ceylon Yellow Dwarf (CYD; SL), (65) Kenthali (KTOD; IND), (66) Gangabondam (GBGD; IND), (67) Green Dwarf, (68) Gudanjali, (69) Kulashekaram Green Dwarf (KGD; IND), (70) Laccadive Dwarf (LCD; IND), (71) Malayan Green Dwarf (MGD; MYS), (72) Malayan Orange Dwarf (MOD; MYS), (73) Malayan Yellow Dwarf (MYD; MYS), and (74) Niu leka (NLAD; FJI). The hybrids (34) include (75) ADOT × GBGD, (76) ADOT × PHOT, (77) CCNT × GBGD, (78) CCNT × LCT, (79) COD × WCT, (80) COD × LCT, (81) COD × SKT, (82) ECT × MOD (VHC 3), (83) ECT × COD, (84) ECT × GBGD, (85) FJT × GBGD, (86) GBGD × ECT, (87) GBGD × FJT, (88) GBGD × LCT, (89) GBGD × PHOT, (90) JVT × GBGD, (91) LCT × COD, (92) LCT × CCNT, (93) LCT × GBGD, (94) LCT × PHOT, (95) LMT × GBGD, (96) MOD × WCT, (97) MYD × ECT, (98) MYD × TPT, (99) MYD × WCT, (100) PHOT × GBGD, (101) SKT × COD, (102) VHC-1 (ECT × MYD), (103) VHC-2 (ECT × MGD), (104) WCT × CGD, (105) WCT × COD, (106) WCT × GBGD, (107) WCT × MYD, and (108) WCT × WCT.

At least 6−10 palms from each collection were selected, and freshly opened inflorescences were tagged during April. Twelve months after fertilization, mature nut samples (4 nuts/palm; ∼40 nuts/cultivar or hybrid) from selected palms were harvested from pretagged bunches. Sampling was done during April for three years. Therefore, for overall analysis, more than 10000 nuts were sampled from about 1000 palms. Harvested nuts were processed for obtaining solid endosperm, which was dried to 6% uniform moisture in aseptic condition to make copra. For extracting oil, chopped copra samples (300 mg) were ground finely along with an equal quantity of anhydrous sodium sulfate for absorbing moisture in the copra. Oil extraction was made in a Soxhlet apparatus using petroleum ether (60−80 °C) as the extraction solvent for 3 h. Petroleum ether was evaporated at room temperature, and percentage of oil was estimated gravimetrically. Samples from each palm were pooled in each cultivar for fatty acid analysis. Therefore, about 4000 samples were analyzed for fatty acid profile. For this, oil samples were methyl esterified, and 1 μL of methyl-esterified sample was injected into a GC for fatty acid analysis under conditions as described earlier.[14]

Preparation of Fatty Acid Methyl Esters (FAMEs). Methyl esters of fatty acids were prepared following the method described earlier.[14,15] The HCl reagent (5%) was prepared by adding 8.3 mL of acetyl chloride dropwise to 100 mL of absolute methanol. An ice jacket was used to prevent bumping due to exothermic reaction. This reagent (2 mL) was added to 0.2 g of coconut oil in a 15 mL screw-capped glass vial, vortexed well, and incubated at 70 °C in a hot air oven for 10 h and then cooled to room temperature. To this incubated mixture were added 5 mL of distilled water and 1 mL of hexane, and the mixture was vortexed thoroughly. When two layers were separated, the top layer of hexane was aspirated into microtubes and stored for GC analysis. Furthermore, precaution was taken to avoid evaporative losses of hexane to maintain concentrations of FAMEs.

Gas Chromatography of FAMEs. Methyl-esterified samples were diluted (40 μL of FAME sample + 960 μL of *n*-hexane, HPLC quality) in a sample vial, and 1 μL was injected into a gas chromatograph (GC-2010, Shimadzu, Japan), fitted with an autoinjector (AOC-20i) and capillary column (BPX 70). Elutants were detected on a flame ionization detector (FID). Conditions set for analysis include split mode of injection (split ratio −50). High-grade nitrogen was used as carrier gas at a pressure of 114.9 kPa with a column flow rate of 1.29 mL min^{-1}. The initial temperature of the column was set to 100 °C and then was increased at a rate of 5 °C min^{-1} to a terminal temperature of 225 °C. Amplified signals were transferred and recorded in a computer with GC-Solutions software (Shimadzu, Japan). A quantitative method was followed using an external standard of a mixture of fatty acids (C6−C24). For this, a cocktail of FAME standards (C:6−C24; Sigma, USA) was loaded into the column under conditions similar to those used for sample analysis. The concentrations and peak areas were computed by using different concentrations of standard FAMEs and employing a data analysis method. Data thus acquired were analyzed using GC-Postrun analysis software (Shimadzu, Japan). By using this software, the type of fatty acid and the concentration of each fatty acid in unknown samples were determined precisely.

Data Processing. Data acquired by the GC-Solution Real Time Analysis software were analyzed using GC-Solution Postrun Analysis software to get concentrations and relative percentages of each fatty acid in oil. Processed data were transferred to a MicroSoft Excel spreadsheet for further analysis. SPSS v10 (IBM-SPSS, USA) and Axum software (Mathsoft, U.K.) were used for data exploration, cluster analysis, box-plot analysis, and correlation analysis and for ANOVA in CRD for comparing means using CD at $P = 0.05$.

■ RESULTS AND DISCUSSION

The analysis of oil from 60 Talls, 14 Dwarfs, and 34 hybrids indicated wide variation for the fatty acid profile of oil. In general, coconut oil contains saturated fatty acids such as caproic (C6), caprylic (C8), capric (C10), lauric (C12), myristic (C14), palmitic (C16), stearic (C18), and arachidic (C20). It also contains unsaturated fatty acids such as palmitoleic (C16:1), oleic (C18:1), linoleic (C18:2), and linolenic (C18:3) acids (Figure 1). Apart from these, behenic (C22), lignoceric acid (C24), and some other long-chain fatty acids with retention times (RT) of 26.773 min (unidentified fatty acid, UIFA1), 27.524 min (UIFA2), and 29.487 min (UIFA3) are present in traceable amounts. The *F* test of significance indicates that variation in fatty acid profile is mainly due to genotypic factor ($n = 108$) rather than year factor ($n = 2$) in this experiment due to sampling from corresponding periods across years. However, some trace fatty acids such as C20, C22, and UIFA3 did show significant variations due to year rather than genotypic factor. This study reports not only the extension of a range of concentrations of fatty acids but also new fatty acids (Table 1). The lower range of concentrations of short- and medium-chain fatty acids is extended compared to that reported in the literature, whereas concentrations of myristic acid, palmitic acid, and stearic acid are greater than reported. The presence of behenic, lignoceric, and other long-chain fatty acids with RT 26.773, 27.524, and 29.487 min are newly reported. Palmitoleic acid, even though present in appreciable levels, is not present in many cultivars and hybrids. Similarly, behenic acid is present in a few Talls and hybrids, but it is totally absent in Dwarfs. Earlier methods mostly used packed columns, but the use of capillary columns and more sensitive GC detectors made it possible to detect traces of new fatty acids.

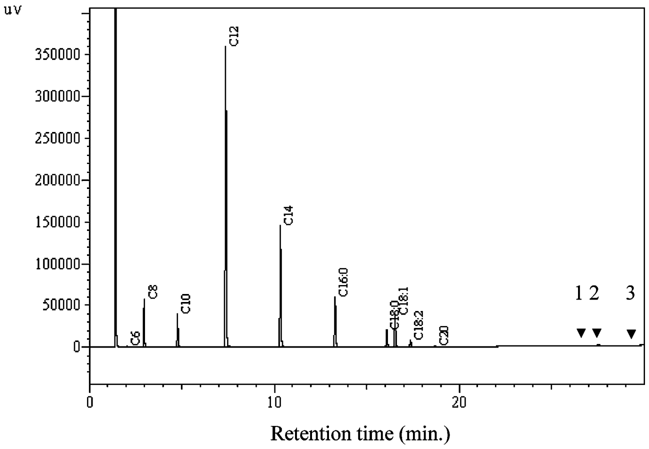

Figure 1. Typical chromatogram of fatty acids in coconut oil. Unidentified fatty acids: (1) UIFA1, fatty acid with a retention time (RT) of 26.773 min; (2) UIFA2, RT 27.524 min; (3) UIFA3, RT 29.487 min.

Relative concentrations of each fatty acid varied significantly among cultivars and hybrids (Table 2). Among the Talls, cultivars NWHT and ASYT had minimum percentages of caproic and caprylic fatty acids, and as a consequence these cultivars also had the lowest percentage of short0 and medium-chain fatty acids (SMFAs). They also had the least concentrations of saturated fatty acids (SFAs) and more long-chain unsaturated fatty acids (LUSFAs) and thus had the least saturated to unsaturated fatty acid ratio. On the other hand, cultivar Hazari had highest percentages of caproic, caprylic, and capric acids and thus the highest concentration of SMFAs. In cultivar Klapawangi, the saturated to unsaturated fatty acid ratio was highest. The concentration of lauric acid was highest in Klapawangi and lowest in VKBT, whereas the concentration of myristic acid was highest in BYL and lowest in Hazari. The concentration of long-chain saturated fatty acids was lowest in Hazari and highest in SS Apricot.

Table 1. Maximum and Minimum Values for the Concentrations of Fatty Acids in Coconut Oil As Reported and Comparison with Values from the Current Experiment

	concentration (%) of fatty acids		
	reported in the literature		this experiment
fatty acid	minimum	maximum	range
C6	0.20	0.80	0.08−0.49
C8	4.70	9.50	2.77−7.21
C10	4.50	9.70	3.46−5.94
C12	45.00	55.06	42.42−52.52
C14	16.89	21.60	18.12−23.05
C16	7.36	11.4	7.59−12.99
C18	1.00	3.70	2.45−4.07
C20	ND[a]	trace	0.02−0.31
C22	ND	ND	0.02−0.19
C24	ND	ND	0.02−1.36
UIFA1[b]	ND	ND	0.06−0.22
UIFA2[c]	ND	ND	0.05−6.01
UIFA3[d]	ND	ND	0.07−1.51
C16:1	ND	ND	0.05−3.90
C18:1	4.01	7.60	4.92−10.86
C18:2	trace	2.20	0.14−2.80
C18:3	ND	ND	0.05−0.14

[a] ND, not detected. [b] UIFA1, fatty acid with RT 26.773 min. [c] UIFA2, RT 27.524 min.. [d] UIFA3, RT 29.487 min.

Similarly among Dwarfs, KGD had the least concentrations of SMFAs (C6−C12), C20, and SFAs, but had the highest concentrations of LSFAs and LUSFAs and, thus, the least saturated to unsaturated fatty acid ratio (Table 2). The concentration of lauric acid was highest in Camroon Red Dwarf and Gudanjali. The Cameroon Red Dwarf also has a higher percentage of SMFAs. The percentage of myristic acid was highest in Green Dwarf and lowest in Niulekha. The concentrations of oleic acid and other LCUFAs were greater in KGD and least in Niulekha. The concentration of SFAs and their ratio to unsaturated fatty acids were low in KGD and high in MGD.

Among hybrids, COD × LCT had the least concentration of caproic and lauric acids and SMFAs, while it had the highest percentages of myristic acid and LSFAs (Table 2). On the other hand, the concentrations of lauric acid and SMFAs were highest in ECT × COD, but it had the least concentration of LSFAs. The concentrations of oleic acid and LUSFAs were least in WCT × CGD, whereas the concentrations of palmitoleic acid and LUSFAs were significantly high in CCNT × GBGD. The concentration of oleic acid was highest in MYD × TPT. Hybrid CCNT × GBGD had the lowest percentage of SFAs and their ratio to unsaturated fatty acids, whereas the percentage of SFAs was highest in WCT × CGD.

Table 2. Range of Percentage of Fatty Acids and Their Groups in Coconut Tall and Dwarf Germplasm and Hybrids

fatty acid/ group	Talls		Dwarfs		hybrids	
	minimum	maximum	minimum	maximum	minimum	maximum
C6	0.08 (NWHT; HAZT01; WDT01)	0.49 (HAZT)	0.09 (KGD)	0.34 (MGD)	0.13 (COD × LCT)	0.40 (COD × SKT)
C8	3.73 (ASYT)	7.21 (HAZT)	2.77 (KGD)	5.39 (NLAD)	3.97 (SKT × COD)	5.88 (COD × SKT)
C10	4.07 (VKBT)	5.94 (HAZT)	3.46 (KGD)	5.22 (NLAD)	3.81 (SKT × COD)	4.82 (CCNT × LCT; ECT × GBGD)
C12	46.26 (VKBT)	52.52 (KWGT)	42.42 (KGD)	51.10 (CRD, GGD)	45.60 (COD × LCT)	50.06 (ECT × COD)
SMFAs[a]	54.49 (ASYT)	64.34 (HAZT)	48.74 (KGD)	61.22 (CRD)	54.22 (COD × LCT)	59.85 (ECT × COD)
C14	18.12 (HAZT)	22.69 (BYL)	19.18 (NLAD)	22.12 (Green Dwarf)	19.77 LCT × CCNT)	23.05 (COD × LCT)
C16	7.59 (Siam; HAZT)	10.79 (ASYT)	8.18 (CRD)	12.99 (KGD)	8.33 (ECT × COD)	10.17 (WCT × WCT; COD × LCT)
C18	2.55 (AROPT)	3.58 (ASYT)	2.88 (KTOD)	4.07 (KGD)	2.77 (MOD × WCT)	3.66 (LM × GBGD)
C20	0.02 (HAZT)	0.09 (NWHT; BGL; BYL; NIT)	0.02 (KGD)	0.31 (NLAD)	0.03 (COD × LCT; PHOT × GBGD)	0.09 (ADOT × GBGD, WCT × CGD)
C22	0.02 (ADOT; AGT; Siam NIT; ASYT)	0.05 (CCNT)	ND[i]	ND	0.02 (COD × WCT, GBGD × PHOT)	0.19 (LCT × GBGD, LCT × PHOT)
C24	0.02 (GTBT)	1.36 (Siam)	0.04 (BYD)	0.41 (KGD)	0.03 (ECT × GBGD, VHC 1)	0.32 (MYD × TPT)
UIFA1[b]	0.06 (BSIT)	0.12 (CCNT, JAMT)		0.11 (MGD)	0.06 (LCT × GBGD, LCT × PHOT)	0.22 (WCT × COD)
UIFA2[c]	0.04 (AROPT)	6.01 (SSAT)	0.32 (GGD)	5.32 (NLAD)	0.05 (ECT × COD)	1.95 (MYD × TPT)
UIFA3[d]	0.07 (ECT)	1.51 (BENT)		0.10 (MGD)	0.08 (VHC)	0.15 (WCT × GBGD)
LSFAs[e]	29.05 (HAZT)	37.70 (SSAT)	31.00 (CRD)	40.13 (KGD)	31.95 (ECT × COD)	36.62 (COD × LCT)
C16:1	0.05 (GUAT; JVT; KENT)	1.24 (KPDT)	0.05 (MOD)	2.43 (GBGD)	0.05 (LCT × CCNT)	3.90 (CCNT × GBGD)
C18:1	4.92 (KWGT)	9.58 (NWHT)	5.78 (NLAD)	9.36 (KGD)	5.43 (WCT × CGD)	8.04 (MYD × TT)
C18:2	0.58 (BYL)	2.28 (NWHT)	0.66 (MGD)	2.17 (Green Dwarf)	1.00 (VHC-1)	1.88 (Cochin China × LCT)
C18:3	0.05 (Siam, KWGT, THT, LFT)	0.11 AROPT, ASYT)	0.06 (NLAD, Green Dwarf)	0.14 (KGD)	0.06 CCNT × LCT)	0.11 (WCT × WCT, VHC, WCT × CGD)
LUSFAs[f]	5.68 (KWGT)	11.88 (NWHT)	6.24 (MGD)	11.14 (KGD)	6.32 (WCT × CGD)	9.95 (CCNT × GBGD)
SFAs[g]	88.12 (NWHT)	94.32 (KWGT)	88.87 (KGD)	93.76 (MGD)	90.05 (CCNT × GBGD)	93.68 (WCT × CGD)
sat/unsat FA ratio[h]	7.75 (NWHT)	17.60 (KWGT)	8.05 (KGD)	14.05 (MGD)	9.17 (CCNT × GBGD)	16.45 (WCT × CGD)

[a]SMFAs, short- and medium-chain fatty acids. [b]UIFA1, fatty acid with RT 26.773 min.. [c]UIFA2, RT 27.524 min.. [d]UIFA3, RT29.487 min.. [e]LSFAs, long-chain saturated fatty acids. [f]LUSFAs, long-chain unsaturated fatty acids. [g]SFAs, saturated fatty acids. [h]Sat/unsat FA ratio, saturated to unsaturated fatty acid ratio. [i]ND, not detected.

The cluster analysis grouped cultivars and hybrids into four major clusters. In the first cluster, three hybrids, namely, FJT × GBGD, PHOT × GBGD, and WCT × CGD, and one cultivar (HAZT) were grouped. In the second cluster 23 Talls (AGT, AROPT, ASYT, WB 01, BONT, BSIT, CCNT, FMST, GUAT, JVT, KAMRUPA, KPDT, KKT, KWGT, LFT, NUFT, NGAT, NWHT, NUGT, PLNT, PHOT, SCT, and SSAT), 11 Dwarfs (CGD, COD, CRD, CYD, KTOD, GBGD, GGD, KGD, MGD, MOD, and MYD), and 11 hybrids (COD × WCT, GBGD × ECT, GBGD × FJT, GBGD × LCT, GBGD × PHOT, JVT × GBGD, LCT × CCNT, LMT × GBGD, MOD × WCT, MYD × ECT, and MYD × TPT) were grouped together. In the third group, 36 Talls, 2 Dwarfs (Green Dwarf and LCD), and 19 hybrids were grouped. In cluster four, NLAD was grouped distinctly, indicating uniqueness of the fatty acid profile of oil from this cultivar. Analysis indicated that cultivars related geographically or typically were grouped closely. For example, Benaulim cultivars from Maharastra were grouped closely. Similarly, cultivars such as Aliyarnagar Tall and Rajapalem Tall arising from Tamil Nadu also grouped closely. Dwarfs such as COD, MYD, MOD, GGD, and GBGD also were grouped closely, whereas the majority of the Talls were closely related. Differences in lauric acid concentration among cultivars from different regions, namely, Philippines, Papua New Guinea, Vanuatu, North Sulawesi, and Sri Lanka, are reported to range from 45 to 52.6%.[9,16]

The frequency distribution analysis indicates that the majority of cultivars fall in a narrow range of each fatty acid concentration. For instance, more than 90 cultivars have a concentration of caproic acid between 0.1 and 0.3%, caprylic acid between 4 and 6%, capric acid in a range of 4−5%, lauric acid between 46 and 50%, myristic acid in a range of 20−22%, palmitic acid in a range of 8−10%, and stearic acid in a range of 2.5−3.5%. Similarly, more than 90 cultivars have unsaturated fatty acids, namely, oleic, linoleic, and linoleneic acids, in ranges of 6−8, 1−2, and 0.05−0.1%, respectively. Therefore, mean percent values for the fatty acid profile of coconut oil are as follows: C6, 0.22 ± 0.01; C8, 5.01 ± 0.06; C10, 4.69 ± 0.03; C12, 48.54 ± 0.12; C14, 20.65 ± 0.08; C16, 9.31 ± 0.06; C16:1, 0.36 ± 0.06; C18, 3.07 ± 0.02; C18:1, 6.71 ± 0.08; C18:2, 1.44 ± 0.03; C18:3, 0.08 ± 0.001; C20, 0.06 ± 0.001; C22, 0.04 ± 0.001; C24, 0.09 ± 0.01; UIFA1 (RT 26.7 min), 0.10 ± 0.001; UIFA2 (RT 27.5 min), 0.60 ± 0.08; and UIFA3 (RT 29.4 min), 0.23 ± 0.04. Only a few cultivars have very low or very high concentrations of fatty acids. For example, the concentration of lauric acid was <46% in 2 cultivars and >50%

in 10 cultivars. The concentration of myristic acid was <20% in 20 cultivars and >22% in 6 cultivars. Similarly, the concentration of oleic acid was >8% in 6 cultivars, linoleic acid was >2% in 4 cultivars, and linolenic acid was >0.1% in 15 cultivars. The analysis indicated that about 85 of 108 cultivars and hybrids have oil containing SMFAs in a range of 56−60% (Figure 2); 17 of them have >60% ,whereas the remaining have <56%.

The box-plot analysis indicates the relative variability among cultivars and hybrids for each of the fatty acids varied (Figure 3). Analysis showed (1) the concentration of each fatty acid that is present in the majority of cultivars relative to the respective maximum value for each fatty acid and (2) cultivars having extremely low or high values for each of fatty acids. Coconut cultivars and hybrids varied maximally for concentrations of C6 and C18:2 fatty acids followed by C18:3, C18:1, C8, and C20 fatty acids. Relative variation for concentrations of C10, C12, C14, C16, C22, C24, and other unidentified fatty acids is less. The majority of cultivars had just about 50% of maximum concentration of C6 fatty acid; 70% of maximum recorded concentration for C8, C16, C18, C18:1; C18:2; 80% of concentration for C10; 90% of concentration for C12 and C14; 50−60% of concentration for C18:1; and 15% of concentration for C20. Analysis also indicated that the majority of cultivars had <10% of respective maximum values recorded for concentrations of C16:1, C22, C24, and UIFA2 fatty acids.

Analysis revealed cultivars with high and low concentrations for each fatty acids beyond normal variability. The list of cultivars having the highest value for each fatty acid is C6, HAZT, JAMT, Nizerian Tall, SSGT, VKBT, and COD × SKT; C8, Aliyarnagar Tall, HAZT, Nizerian Tall, RJPT, and Siam; C10, Andaman Giant, Arsampatti Tall, HAZT, LFT, Nizerian Tall, and Nugili Tall; C12, Arsampatti Tall, HAZT, KWGT, Lifou Tall, and CRD; C14, BYL, VKBT, KGD, COD × LCT, and SKT × COD; C16, Assam Yellow, COD, CYD, KGD, and MYD; and C18, Assam Yellow, Laccadive Small, GBGD, KGD, GBGD × ECT, LMT × GBGD, and MYD × TPT. Among unsaturated fatty acids, the concentration of C18:1 is higher in Mysore Tall, NWHT, CGD, Green Dwarf, and KGD; C18:2 is higher in Gangapani, Mysore Tall, NWHT, KTOD, and Green Dwarf; and C18:3 is higher in Arsampatti Tall, Assam Yellow, KGD, and WCT (selfed).

The concentration of short- and medium-chain fatty acids is about 90% of maximum value in the majority of cultivars (Figure 4). The concentration of LSFAs is about 85% of maximum, wheres the concentration of LUSFAs is about 70% of maximum values in the bulk of the cultivars. The variability of coconut germplasm for concentration of LUSFAs is high followed by that for LSFAs and SMFAs. Consequently, about 65% of maximum values for the SFA/USFA ratio is present in most cultivars. The list of top-ranking cultivars and hybrids for highest concentration of all these parameters is as follows: SMFAs, Arsampatti Tall, GUAT, HAZT, KWGT, and LFT; LSFAs, Assam Yellow, SSAT, KGD, COD × LCT, and SKT × COD; LUSFAs, Mysore Tall, NWHT, CGD, Green Dwarf, and KGD; SFAs, BRR, BYL, KWGT, and WCT × CGD. Cultivars having the least concentration of SFAs are Mysore Tall, NWHT, CGD, Green Dwarf, and KGD, and cultivars having the least concentration of LUSFAs are BGL, BRR, BYL, KWGT, and WCT × CGD. Cultivars with the highest ratio of saturated to unsaturated fatty acids are BGL, BRR, BYL, KWGT, and WCT × CGD, whereas those with the least value for ratio are Mysore Tall, NWHT, Green Dwarf, KGD, and CCNT × GBGD.

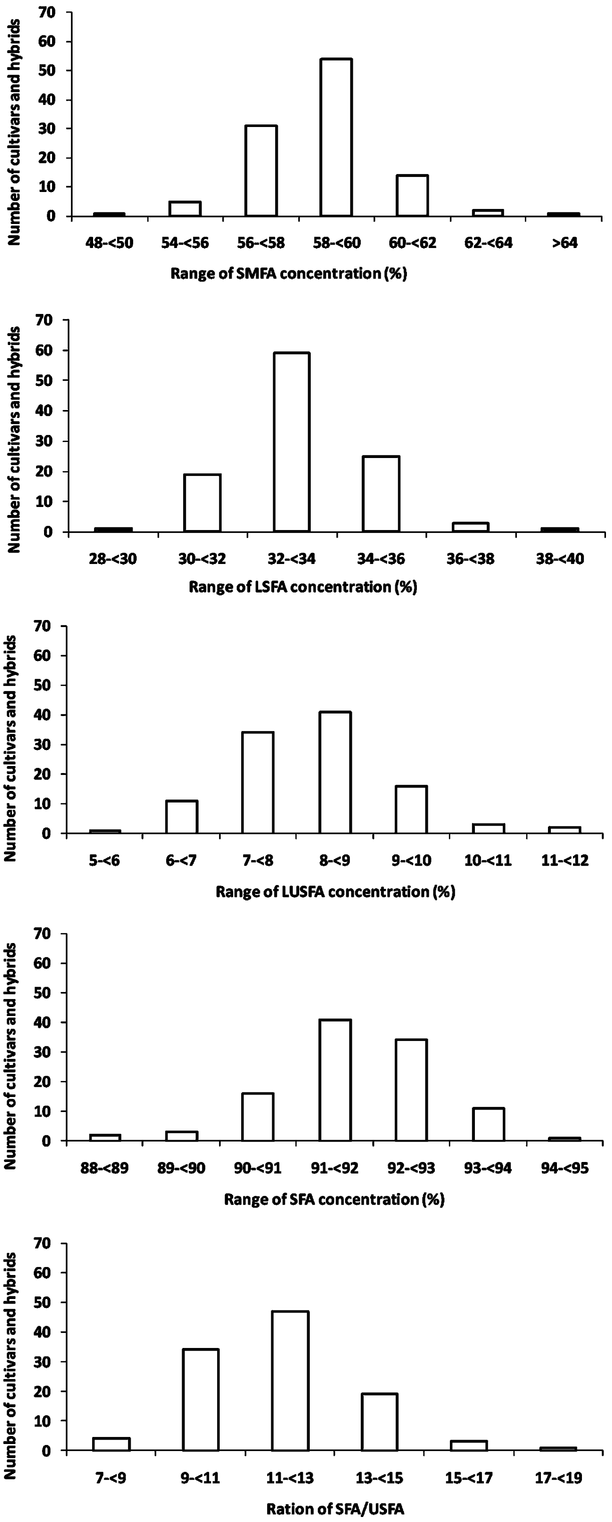

Figure 2. Frequency distribution of cultivars and hybrids in different concentrations of various groups of fatty acids. *n* = 108. SMFAs, short- and medium-chain fatty acids; LSFA, long-chain saturated fatty acids; LUSFAs, long-chain unsaturated fatty acids; SFAs, saturated fatty acids; SFA/USFA, saturated to unsaturated fatty acid ratio.

The correlation matrix indicates that concentrations of C6, C8, and C10 are positively correlated (Figure 5). The concentration of C10 is positively correlated with that of

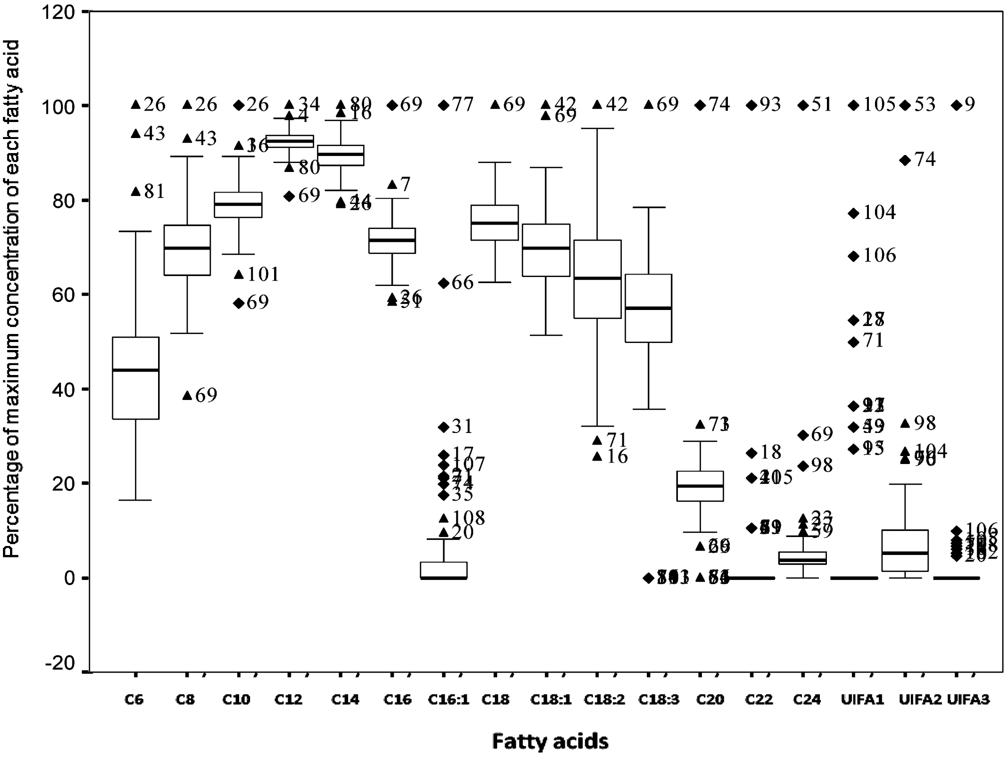

Figure 3. Box-plot analysis of fatty acid profile of coconut oil from different cultivars and hybrids. The *Y*-axis indicates the concentration of fatty acid as a percentage of maximum observed concentration for the respective fatty acid. UIFA1, fatty acid with RT 26.773 min; UIFA2, RT 27.524 min; UIFA3, RT 29.487 min. Data legends indicate cultivars/hybrids with concentration of respective fatty acid significantly different. For cultivar name, please refer to Materials and Methods.

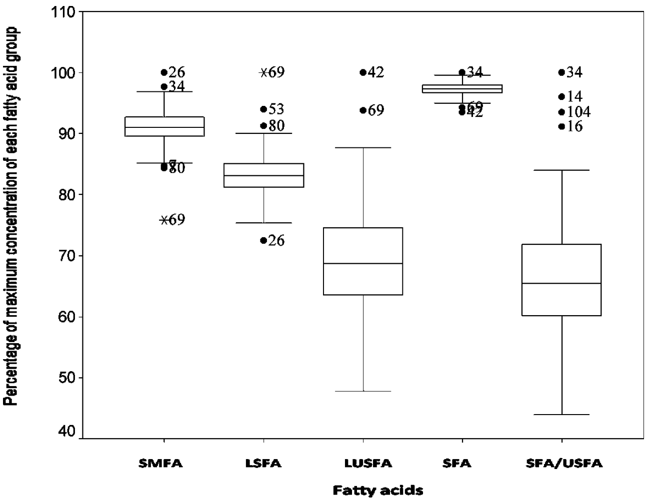

Figure 4. Box-plot analysis of fatty acid groups in coconut oil from different cultivars and hybrids. The *Y*-axis indicates the concentration of groups of fatty acids as a percentage of maximum observed concentration for respective fatty acid group. SMFAs, short- and medium-chain fatty acids; LSFAs, long-chain saturated fatty acids; LUSFAs, long-chain unsaturated fatty acids; SFAs, saturated fatty acids; SFA/USFA, saturated to unsaturated fatty acid ratio. Data legends indicate cultivars/hybrids with concentration of respective fatty acid significantly different. For cultivar name, please refer to Materials and Methods.

C12. On the other hand, concentrations of these two fatty acids are negatively correlated with long-chain saturated fatty acids such as C14 and C16. Concentrations of long-chain fatty acids are not correlated strongly except between C18:1 and C18:2 and between C18 and C18:3. Furthermore, concentrations of C10 and C12 strongly influenced the concentration of SMFAs, whereas the concentrations of C14 and C16 influenced the concentration of LSFAs. Results also indicate that the concentration of LSFAs is built up at the cost of SMFAs, particularly lauric acid. Even concentrations of USFAs are built up at the cost of SMFAs. The ratio of SFAs to USFAs is more influenced by the concentration of USFAs. Analysis also indicates that long-chain fatty acids have negative correlation with the concentration of fatty acid SMFAs, but correlations among concentrations of long-chain fatty acids are either positive or have no direction. Fatty acids having a chain length of ≥20 do not have any significant correlation with the concentration of other fatty acids.

The lack of significant correlation between concentrations of C16 and C18 and also between C18, C18:1, and C18:2 indicates independent or alternative metabolic pathways as suggested in the case of sunflower[17] and olive.[18] As observed in sunflower, in coconut also concentrations of oleic and linoleic acids are positively correlated. A negative correlation of long-chain fatty acids with SMFAs indicates a chain elongation process and therefore provides an opportunity to stop chain elongation to obtain a higher concentration of lauric acid. Fat deposits of a developing nut possess capacity to selectively synthesize chain length-regulating enzyme systems for any chain length at any time without reference to nature and proportion of other system present. Different chain length-elongating enzymes in fat deposits are not formed as a single composite unit but as a collection of different independent units. The correlation matrix also indicates a possibility of independent desaturation and elongation of SMFAs to form C18 mono- and polyunsaturates. Strong positive correlation

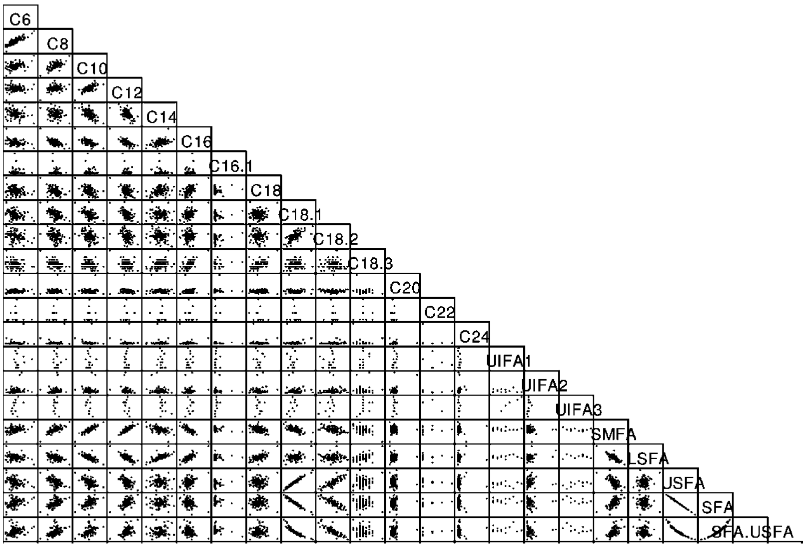

Figure 5. Matrix of correlation between fatty acids in coconut germplasm. UIFA1, fatty acid with RT 26.773 min; UIFA2, RT 27.524 min; UIFA3, RT 29.487 min; SMFAs, short- and medium-chain fatty acids; LSFA, long-chain saturated fatty acids; LUSFAs, long-chain unsaturated fatty acids; SFAs, saturated fatty acids; SFA/USFA, saturated to unsaturated fatty acid ratio.

between oleic and linoleic acid and also their negative correlation with SMFAs indicate the possibility of sequential unsaturation.

In coconut oil concentration of C18:1 and C18:2 increased mainly at the expense of SMFAs, unlike sunflower, where the increase in linolenic acid was at the expense of oleic acid.[19] Even though the literature indicates a significant influence of external factors such as weather[13,20,21] and water regimen[17] on the fatty acid profile, particularly on the oleic group of polyunsaturates,[22] no such significant influence is noted for the fatty acid profile of coconut oil as the concentration of unsaturated fatty acids is only ~10%. Seasonal variation in the fatty acid profile of developing coconut is proportional to the concentration of USFAs, which is greater in tender nuts.[2] However, by the time the nut matures, the fatty acid profile of coconut oil seems to attain a more conservative profile. Overall, genetic improvement for higher concentration of essential fatty acids such as C18:1, C18:2, and C18:3 has more scope due to the existence of higher genetic variability than scope for improving the concentration of C12. However, to improve the concentration of C12, it is necessary to arrest chain elongation of fatty acids. Reducing the omega 6:3 ratio in agricultural plants has been the focal issue for improving the nutritional value of oils and also to correct inadvertent shifts toward higher ratios of omega 6:3, which has taken place in the past 100 years.[10,23,24] Metabolic and protein engineering is being employed to design desirable fatty acids in plants.[25]

The saturated to unsaturated fatty acid ratio varied from 6.32 to 17.6, indicating a wider variation in the quality of coconut oil than earlier thought. This variation can be used for commercial exploitation in making value-added oleochemicals. Cultivars such as Klapawangi, Camaroon Red Dwarf, and Gudanjali and the hybrid ECT × COD are highest yielders of lauric acid, indicating that oil from these can be exploited in the pharmaceutical and lauric acid based industries. Cultivars and hybrids with high lauric acid (a conditionally essential fatty acid) concentration and low ratio of saturated to unsaturated fatty acids can be used for both edible and industrial purposes (the soap industry). In the medicinal and pharmaceutical industries for manufacturing trilaurin and ointment bases, oil with high concentrations of lauric acid and medium-chain fatty acids (MCFAs) is desirable. A high concentration of myristic acid in oil is useful for manufacturing binders and emollients used in cosmetics.[1,26] Among lauric oils and fats such as coconut oil, palm kernel oil, babassu, and tucum, the most commercially important oils are from coconut and palm kernel oil. Lauric oils have the advantage over other oils in the preparation of confectionary items due to low unsaturation. Furthermore, coconut oil is highly suitable as a biofuel because of its high ratio of saturated to unsaturated fatty acids. A high concentration of short-chain fatty acids and SFAs in coconut oil makes it more stable. It is softer than palm kernel oil with a lower slip melting point of 24−26 °C and a lower solid fat content at 20 °C apart from high stability against oxidative rancidity making it a better oil for use in confectionary items.

This study reports the presence of more fatty acids than reported by earlier studies.[1,8,9,15,27,28] Oleic acid and other unsaturated fatty acids and overall LUSFAs were present in higher quantities than reported, indicating that cultivars such as Nuhuang, KGD, and GBGD and hybrids MYD × TT, CCNT × GBGD, and CCNT × LCT are good for human consumption. Whereas vegetable oils with a high level of unsaturation are considered to be good for human health, a high level of saturated fatty acids is desirable in the food industry to avoid hydrogenation and transesterification processes in the production of margarines and shortenings. Moreover, industrial hydrogenation requires high energy and also results in the formation of trans fatty acids, a concern from the human health point of view. Apart from this, trans fatty acids are considered to be more atherogenic than saturated fatty acids.[29] Coconut oil with a high amount of saturated fatty acids is more suitable as a frying fat and is relatively stable against oxidative rancidity. Furthermore, high concentrations of SMFAs in coconut oil are easily digestible.[5] In fact, several efforts have focused on increasing the concentrations of saturated fatty acids in many oil crops.[30,31] Instead, blending of coconut oil with other oils[32] can be a suitable approach for human consumption.

Results also indicate that coconut germplasm varied for the content of fatty acids such as lauric acid and essential unsaturated fatty acids. These variations can be attributed more toward genotypic factors than climatic factors as samples are drawn from the same season for comparison. Climatic factors predominantly influence the degree of unsaturation, thereby influencing the concentration of unsaturated fatty acids as noted in sunflower[17,33] and olive oils.[34] Seasons and water regimens are also noted to influence concentrations of fatty acids in immature coconuts.[2,10] However, oil from mature nuts possesses the very least concentration of unsaturated fatty acids and, therefore, variation in fatty acid profile can be a useful criterion for grouping cultivars, if not for identification of a cultivar as suggested for olive cultivars by Perri et al.[35] As coconut flowers and yields nuts at almost monthly intervals throughout the year, grouping of cultivars based on fatty acid profile can be of potential use. However, samples need to be drawn from similar conditions of management and climate conditions.

It can be concluded from the present investigation that oils from cultivars and hybrids of coconut significantly differed, particularly for commercially important fatty acids such as lauric acid and unsaturated fatty acids. However, coconut oil seems to have a conserved fatty acid profile, mainly because of low unsaturated fatty acids, indicating the possibility of grouping cultivars on the basis of fatty acid profile. Fatty acid profile based cluster analysis grouped geographically and typically closely related cultivars together. There is a scope for genetic improvement for higher concentration of essential fatty acids and lauric acid through breeding, and metabolic engineering may be required to bring significant changes in the fatty acid profile of coconut oil. Cultivars with a high concentration of a specific fatty acid can be of potential use for industrial exploitation, whereas those with high concentrations of short- and medium-chain fatty acids and unsaturated fatty acids are more suitable for human consumption.

■ AUTHOR INFORMATION

Corresponding Author

*Present address: Division of Environmental Sciences, Indian Agricultural Research Institute, Pusa, New Delhi 110 012, India.
E-mail: nareshkumar.soora@gmail.com.

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