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Chemical Characterization of Canola and Sunflower Oil Deodorizer Distillates

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In the present work deodorizer distillates of canola and sunflower oil were studied for their chemical characterization. Tocopherols, sterols, hydrocarbons and fatty acid composition were determined by Gas chromatography Mass spectrometry (GC–MS). Results indicated that deodorizer distillates of canola oil (DDCLO) studied had higher saponification value in the range of 164.2–175.8 mg/g and all deodorizer distillates of sunflower oil (DDSFO) had high iodine value of 126.2–127.1 g/100 g. Peroxide value and free fatty acids were present in the range between 7.0–8.9 mEq/Kg and 35.7–54.4 g/100 g in DDSFO and DDCLO samples, respectively. Contents of oleic acid (49.3–51.1 g/100 g) and linolenic acid (6.2–7.5 g/100 g) were significantly greater in DDCLO samples. Conversely, DDSFO contained concentrated amount of linoleic acid (52.8–53.3 g/100 g). Tocopherols and hydrocarbons were significantly higher in all DDSFO samples while, sterols were dominant in DDCLO samples. GC–MS provided excellent results for simultaneous determination of tocopherols, sterols, hydrocarbons and fatty acid composition of deodorizer distillates of canola and sunflower oil.

INTRODUCTION

The most important by-product of edible oil refining is the deodorizer distillate (DD) obtained in the deodorization stage [Ruiz-Méndez & Dobarganes, 2007]. Basically deodorization is the final key step of the refining process accountable for removing targeted volatile compounds which are liable for producing unacceptable odor, color, taste and flavor in the oil. The unwanted volatile compounds removed during the deodorization are FFAs, oxidation products of fat and oil namely aldehydes, ketones, alcohols, peroxides and carbohydrates. These volatile odoriferous compounds are stripped off by direct injection of heated steam at a temperature of 180–270°C. The fraction of the volatile species is then collected in a condensed form of DD. Owing to the vigorous conditions of deodorization process unfortunately some fractions of valuable minor components are also distilled off and become the part of DD. It is considered to be most concentrated source of the various valuable tocopherols, tocotrienols, phytosterols (free and esterified), hydrocarbons, squalene, mono and di-glycerides, neutral oil and free fatty acids [Dumont & Narine, 2007]. DD could be used as a cheap source for the retrieval of natural tocopherols and tocotrienols, while 0.3–0.5% of the deodorizer feedstock ends up as the DD [Chu *et al*., 2002].

Tocopherols and tocotrienols are value-added natural antioxidants collectively known as vitamin E. These components are important because they impede the oxidation reaction and prevent material from oxidative damage. These are broadly applicable in cosmetics and pharmaceutical preparations. Phytosterols are used as an essential precursor for the preparation of pharmaceutical medicines and steroids. Phytosterols have the ability to inhibit cholesterol absorption by the small intestine. Furthermore, phytosterols possess anti-inflammatory, anti-atherogenic, anti-cancer and anti-oxidative activities [Yang *et al*., 2010].

Another important bioactive component is squalene (2,6,10,15,19,23-hexamethyl-2,6,10,14,18,22-tetracosahexaene, ($C_{30}H_{50}$), *all-trans*) a terpenoid hydrocarbon used in cosmetic formulations and for ensuring stability of vegetable oil; it additionally accounts for the biosynthesis of cholesterol [Mendes *et al*., 2005]. Depending on the quality of crude vegetable oil and physical/chemical refining processes, fatty acids constitute 25–75% of the DD. These may be used as preliminary material for the synthesis of phytosteryl esters showing the hypocholesterolemic effect added to functional foods [Durant *et al*., 2006]. Simple esters of fatty acids with alcohol are used as an alternative source for fuel production. Recently more interest has been focused on the production of biodiesel from various vegetable oil deodorizer distillates (DDs), which is environmental friendly [Liu & Wang, 2009]. The importance and use of DD have been changing in many years, even periodically it was used only for its phytochemicals

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content, mainly sterols. Sterols belong to the class of steroids having biological activity and used as precursors for the preparation of pharmaceuticals. Currently, an industrialist retailed the DD by its tocopherols and phytosterols amount [Verleyen *et al*., 2001].

DD is not only considered as a waste material because it is a rich source of naturally occurring bioactive and nutritive compounds that make it economically valued. DD obtained from chemically refined oil is generally rich in tocopherols in a broad proportion (9.2–15%) and phytosterols (9.0–17.6%) and has the lowest quantity of FFAs (10–24.5%). However, the distillates resulting from physically refined vegetable oils mainly comprised of a large percentage of FFAs (76.2–83.6%), therefore the minimum percentage of tocopherols (1.4–4.3%) and phytosterols (1.8–6.9%) were attained [Verhé *et al*., 2006]. It has been observed that variation in the composition of DD is basically dependent on the nature of the oil, type of refinement process, design of deodorizer column, deodorization time, the operating parameters of deodorizer include volume of stripping steam, the ratio of reduced pressure and temperature [De Greyt & Kellens, 1996; Kellens & De Greyt, 2000].

The intricate nature of DDs makes it very difficult for their full analysis [Verleyen *et al*., 2001]. Conversely few analytical methods have been reported which provide the comprehensive analysis of DD. Durant *et al*. [2006] worked on the separation of multicomponents present in the canola DDs *via in situ* silylation by gas chromatography (GC) with mass spectrometry (MS) for their final detection. The compounds separated included tocopherols, sterols, fatty acids and glycerides. Dumont & Narine [2007] reported the characterization of soybean oil deodorizer distillate through GC with flame ionization detector (FID) after the preparation of silyl derivatives. However, only rare data about the physico-chemical analysis of DDs are available. [Khatoon *et al*., 2010]. To the best of our knowledge no one reported the physicochemical parameters and fatty acid profile of sunflower and canola distillate.

Increased use of industrial waste and byproducts fits the requirement of industry to fulfill with environmental rules. The replacement of natural products for synthetic materials has gained worldwide consideration in the food, pharmaceutical and other industries. Therefore, DD has been utilized as a natural source of tocopherols, sterols, squalene as well as fatty acids in many fields. To find the potential use of DD is very essential to characterize its composition. Therefore this study was aimed at characterizing the sunflower and canola oil DD for their tocopherols, phytosterols, hydrocarbons, fatty acids content and other parameters like peroxide value (PV), iodine value (IV), moisture content, saponification value (SV) and acid value (AV).

MATERIALS AND METHODS

Samples and reagents

Two DD samples of sunflower and three DD samples of canola oil were obtained from vegetable oil refining industries located in Karachi, Pakistan. Samples were collected from three different industries. The initial material consisted of one DDCLO and one DDSFO of two industries. From the third industry only one DDCLO sample was collected as it was not processed for sunflower oil. Each individual sample of DD was collected in triplicate and therefore analyzed in triplicate then reported as mean with the standard deviation (SD). All the samples were kept in the refrigerator at 4°C until analyzed. All chemicals, reagents and solvents used were of analytical grade.

Physico-chemical characterization

Moisture content

The moisture content in DD was determined by the Association of the Official Analytical Chemists [AOAC, 1990] method *via* sample heating in an oven at 105ºC till constant weight was obtained.

Free fatty acid value

Free fatty acids (FFAs) amount in the DDs was determined by standard titration method Ca 5a-40 [AOCS, 1998]. DD sample was titrated against the standardized aqueous solution of sodium hydroxide (KOH), about 2.35±0.001 g of DD was placed in a 250-mL conical flask which contained 10 mL of neutral ethanol. The mixture was warmed on a heating plate, shaken well and titrated against 1 N NaOH after addition of a phenolphthalein indicator.

Iodine value (IV)

The IV was determined by Standard method of oil and fats 993.20 [AOAC, 1997]. Iodine numbers determination is an indication of unsaturation of oil samples and it is expressed as the amount in grams of iodine absorbed by 100 gram of oil. DD sample was dissolved in carbon tetrachloride CCl_4 (15 mL), Wijs' reagent (25 mL) and fresh solution of potassium iodide 10 mL of 5% KI. The solution was mixed well and kept in the dark for 30 min to complete the process. Iodine liberated from sample mixture was then titrated by adding starch as an indicator with 0.1 N standard sodium thiosulfate solutions.

Saponification value (SV)

The SV was determined by AOCS standard method Cd 3–25. SV is the amount of potassium hydroxide in mg necessary to saponify one gram of fat and oil. The SV depends on the type of fatty acid contained in the fat and oil. About 2 g of the DD samples was refluxed with 25 mL of 95% ethanolic potassium hydroxide for at least 60 min. After refluxing, the sample was titrated against a standardized solution of 0.5 N HCl in addition to a phenolphthalein indictor.

Peroxide value (PV)

PV was determined by method Cd 8b-90 [AOCS, 1998]. The DD sample 2.25±0.005 g was dissolved in 15 mL of a glacial acetic acid: chloroform mixture (3:2 v/v %) and saturated potassium iodide solution (0.25 mL). Titration of the resulting solution was performed against standardized solution of sodium thiosulphate (0.1N) in the presence of 1% starch indicator solution.

TABLE 1. Chemical analysis results of canola and sunflower deodorizer distillates.

Samples	DDCLO–1	DDCLO–2	DDCLO–3	DDSFO–1	DDSFO–2
Physical appearance	Dark brown semisolid	Dark brown semisolid	Dark brown semisolid	Dark brown semisolid	Dark brown semisolid
Moisture (%)	$0.08^{a}\pm0.01$	$0.15^{c}\pm0.01$	$0.12^{b}\pm0.01$	$0.16^{c}\pm0.01$	$0.11^{b}\pm0.01$
Saponification value (mg KOH/g sample)	$175.61^{d}\pm0.53$	$164.24^{c}\pm0.89$	$175.78^{d}\pm0.37$	$140.19^{a}\pm0.09$	$152.44^{b}\pm0.16$
Peroxide value ($mEqO_2$/Kg sample)	$6.97^{a}\pm0.11$	$8.94^{c}\pm0.02$	$6.95^{a}\pm0.23$	$7.08^{a}\pm0.06$	$8.00^{b}\pm0.11$
Iodine value (g I_2/ 100 g sample)	$107.55^{b}\pm0.14$	$99.83^{a}\pm0.09$	$107.38^{b}\pm0.42$	$127.12^{d}\pm0.04$	$126.19^{c}\pm0.12$
Free fatty acids (as oleic) (g/100 g sample)	$36.16^{a}\pm0.07$	$54.38^{d}\pm0.32$	$35.66^{a}\pm0.01$	$40.89^{c}\pm0.02$	$39.81^{b}\pm0.05$

Values are presented as mean ± SD of 3 measurements. Values within the same rows followed by the different letters are significantly different at 95% significance ($p<0.05$).

TABLE 2. Fatty acid composition of canola and sunflower deodorizer distillates by GC-MS.

Components (%)	DDCLO–1	DDCLO–2	DDCLO–3	DDSFO–1	DDSFO–2
Palmitic acid methyl ester (16:0)	6.87 ± 0.11^{a}	9.04 ± 0.23^{b}	7.31 ± 0.22^{a}	9.62 ± 0.13^{c}	$9.87\pm .28^{c}$
Palmitoleic acid methyl ester (16:1)	nd	nd	nd	0.23 ± 0.01^{a}	0.25 ± 0.01^{b}
Stearic acid methyl ester (18:0)	1.78 ± 0.02^{a}	3.37 ± 0.07^{c}	1.76 ± 0.04^{a}	2.87 ± 0.13^{b}	2.98 ± 0.04^{b}
Oleic acid methyl ester (18:1)	51.14 ± 0.4^{c}	49.29 ± 0.18^{c}	50.76 ± 0.36^{b}	30.12 ± 0.64^{a}	29.92 ± 0.13^{a}
Linoleic acid methyl ester (18:2)	26.92 ± 0.07^{b}	24.71 ± 0.13^{a}	26.70 ± 0.31^{b}	53.26 ± 0.42^{c}	52.78 ± 0.41^{c}
Linolenic acid methyl ester (18:3)	7.22 ± 0.32^{c}	6.18 ± 0.29^{b}	7.45 ± 0.04^{c}	0.61 ± 0.05^{a}	0.63 ± 0.03^{a}
Arachidic acid methyl ester (C20:0)	1.59 ± 0.02^{c}	3.47 ± 0.05^{d}	1.42 ± 0.05^{b}	0.53 ± 0.02^{a}	0.56 ± 0.04^{a}
Eicosenoic acid methyl ester (C20:1)	3.81 ± 0.11^{b}	3.14 ± 0.14^{a}	3.84 ± 0.18^{b}	nd	nd
Eicosadienoic acid methyl ester (C20:2)	nd	nd	nd	2.35 ± 0.24^{a}	2.62 ± 0.15^{a}
Docosanoic acid, methyl ester (C22:0)	0.66 ± 0.01^{b}	0.70 ± 0.05^{c}	0.74 ± 0.06^{bc}	0.40 ± 0.01^{a}	0.40 ± 0.02^{a}

nd: not detected, Values are presented as means ± SD of 3 measurements. Values within the same rows followed by the different superscript are significantly different at 95% significance ($p< 0.05$).

Chromatographic evaluation

Determination of fatty acid composition by GC-MS

Fatty acid methyl esters (FAMEs) for all the samples of DD were prepared and the fatty acid composition was determined by IUPAC standard method 2.301 [IUPAC, 1976]. Analysis of FAMEs of DD was carried out on the gas chromatography instrument coupled with mass selective detector (GC-MS) model 6890 N from Agilent Technology. The ChemStation 6890 Scale Mode software was installed for the chromatographic peak analysis. A capillary column RT-2560 Biscyanopropylsiloxane (100 m x 0.25 mm ID x 0.25 μm film thickness) was used for the separation of fatty acids. The initial oven temperature was 150°C; it was held for 2 min then raised to 245°C with ramp rate of 4°C/min. Helium was used as the carrier gas with a flow rate of 1.4 mL/min. Injector temperature was set at 250°C and each sample of 1 μL quantity was injected with the split mode ratio of (50:1) with the detector temperature 260°C. The mass detection was performed with an electron impact (EI) ion source mode at 70 eV in the mass scan range of 50–550m/z. Two standard libraries NIST & Wily were used for fatty acids comparison and identification.

Characterization of sterols, tocopherols and hydrocarbons by GC-MS

The sterols, tocopherols and hydrocarbons were determined by GC-MS according to the reported method [Ramadan *et al*., 2006]. Separation of these components was achieved on an HP-5MS (5% phenyl methylsiloxane) capillary column (30 m, 0.25 mm i.d, 0.25 μm film thickness, Agilent Technologies, Palo Alto, CA, USA). The oven temperature started from 100°C with and continued to rise up to 295°C with a ramp rate of 10°C/min. The final temperature was then held for 20 min. Helium was used as a carrier gas with a flow rate of 10.2 mL/min. An aliquot of 2 μL was injected in a splitless mode of injector. The identification of unsaponifiable components was carried out by spectral matching with NIST and Wily libraries installed within GC-MS software.

Statistical analysis

The data obtained was analyzed statistically using ANOVA and Tukey's HSD multiple range test at $p<0.05$ was applied for multiple sample comparison analysis. SPSS version 18.0 (SPSS Inc., Chicago, IL, USA) software was used for statistical evaluation.

RESULTS AND DISCUSSION

The DD samples of canola and sunflower oil were examined for their physico-chemical characterization and the results of the moisture content; FFAs, PV, SV and IV are given in Table 1. All samples of DD obtained appeared as dark red and liquid at room temperature. Moisture content in the DD was determined and found to be almost similar to the reported values [Carmona *et al*., 2010]. Comparatively, higher percentage of FFAs was observed in DDCLO-2 (54.4%) as compared to DDCLO-1 (36.2%) and DDCLO-3 (35.7% of canola oil). In the study reported by Güçlü-Üstündağ & Temelli [2007], the FFA content was found to be 50.4% which is relatively similar to the FFA value of DDCLO-2. Likewise, for DDSFO-1 and DDSFO-2 the FFA values were found to be 40.9% and 39.8%, respectively. The SV of DDCLO-1 was 175.6 mg/g of oil and that of DDCLO-3 was 175.8 mg/g of oil, which was relatively similar and significantly higher ($p<0.05$) than DDCLO-2 (164.2 mg/g of oil), DDSFO-1 (140.2 mg/g of oil) and DDSFO-2 (152.4 mg/g of oil). In the study reported by Ramamurthi *et al*. [1991] on DDCLO, the saponification value was 199.6 (mg/g of oil) which is higher than in our samples of canola distillate. Whereas, saponification results of the sunflower distillate sample are in good agreement with the reported studies [Carmona *et al*., 2010; Pagani & Baltanás, 2010]. The higher saponification value of the DDCLO-l and DDCLO-3 and lower FFA content indicated the presence of high amount of triacylglyceride compared to DDCLO-2 [Khatoon *et al*., 2010]. The PV is a measure of the amount of peroxides and hydroperoxides formed in the sample of oils and fats as a result of oxidation. The PV determination is most commonly used for the measurement of oxidative rancidity of oils and fats [Babalola & Apata, 2011]. The PVs of DDCLO-l, DDCLO-3 and DDSFO-1 were relatively similar, *i.e.* 7.0 mEq/kg of oil, 6.9 mEq/kg of oil and 7.1 mEq/kg of oil although, significantly lower than that of DDSFO-2 (8.0 mEq/kg) and DDCLO-2 (8.9 mEq/kg of oil), which suggested the presence of a high concentration of deteriorating components like hydroperoxide in DDSFO-2 as well as in DDCLO-2. In this study, the PV of each analyzed DD sample was less than 10 mEq of O_2/kg of oil and comparable to values reported for soybean oil DD (7.6 mEq/kg) [Khatoon *et al*., 2010]. Another parameter used to characterize oils and fats is the iodine value which is a measure of unsaturation in oil and fats. The IV of DDSFO samples (126.2–127.1 g of I_2/100g) was significantly higher ($p<0.05$) than that of DDCLO (99.8–107.5 g of I_2/100g). The high iodine value revealed that the high percentage of unsaturated fatty acids was present in the distillate sample. In the present work, a relatively lower value of iodine (127.1 g of I_2/100g) was estimated from the sunflower distillate sample when compared to the results (136.8 g of I_2/100g) reported by Pagani & Baltanás [2010].

Fatty acid composition

The fatty acids esterified to the glycerol backbone present in the DD were analyzed by GC-MS in the form of methyl esters, the results of fatty acid composition are presented in Table 2 as a mean with their standard deviation. Fatty acids profile of DD samples comprised of a number of fatty acids which involved palmitic acid (16:0), palmitoleic acid (16:1), stearic acid (18:0), oleic acid (18:1), linoleic acid (18:2), linolenic acid (18:3), arachidic acid (C20:0), eicosenoic acid (C20:1), eicosadienoic acid (C20:2) and docosanoic acid (C22:0).

The oleic acid (18:1) was the main monounsaturated fatty acid in all samples of DDCLO and its content ranged from 49.3 to 51.2%. Oleic acid (C18:1) is involved in the reduction of LDL cholesterol levels [Aftab *et al*., 2010]. Among polyunsaturated fatty acids linoleic acid (18:2) was significantly ($p<0.05$) higher in all samples of DDSFO compared to linolenic acid (18:3) which was high in all the samples of DDCLO. Linoleic acid (18:2) content of our analyzed DDSFO samples (52.8–53.3%) was higher than that reported by Ghosh & Bhattacharyya [1996]. Linoleic acid (18:2) plays one of the most imperative roles in the prevention of diverse heart and vascular diseases as it is a key component in most of human food [Bello *et al*., 2011]. Eicosenoic acid (C20:1) was found only in DDCLO sample with an estimated range from 2.5–3.3%. Palmitic acid (6.9–10.4%) was the most copious saturated fatty acid in all samples of DD relative to the stearic acid (1.8–6.3%) and docosanoic acids (0.7%–3.1%). FAMEs are high value products and could be used as a fuel in the form of biodiesel [Jiang *et al*., 2006].

Sterols, tocopherols and hydrocarbons composition

GC-MS analysis of unsaponifiable extracts for all DD samples established a profile of sterol compounds mainly β-sitosterol, brassicasterol, stigmasterol, campesterol and cycloartenol were identified and presented in Table 3. β-Sitosterol has been exposed to display effective anticancer and antipyretic properties [Castelli *et al*., 2006], and found as the main constituent in DD. The percentage content changes during processing with the possibility to achieve the highest value covering the range of β-sitosterol (8.4–10.4%) followed by campesterol (1.8–5.4%), brassicasterol (2.1–2.7%), stigmasterol (1.5–2.1%) and lanosterol (0.4–1.0%) in overall samples of canola and sunflower distillate. Whereas brassicasterol and lanosterol were found only in DDCLO samples and these components were not detected in DDSFO samples. The high percentage of campesterol was found in all samples of DDCLO relative to DDSFO samples and our results are in good agreement with the study reported by Verleyen *et al*. [2001]. Among tocopherols, the maximum concentration of γ-tocopherol (1.7–3.9%) was found in all samples of DDCLO analyzed relative to α–tocopherol (3.8–4.6%) which was copious in DDSFO and the obtained results for tocopherols were quite similar to literature data [Güçlü-Üstündağ & Temelli, 2007].

Gunawan *et al*. [2008] described that in DD, 30–40% of nonpolar lipid fraction (NPLF) are HCs which included four main groups that are aliphatic, steroidal, sesquiterpene and triterpene (squalene) HCs. The major aliphatic hydrocarbons HCs in vegetable oils DD are from C_{12} to C_{35} chain length and have immaterial applications in human life after

TABLE 3. Compounds identify in non-saponifiable fraction of canola and sunflower deodorizer distillates by GC-MS .

Components	DDCLO–1 (%)	DDCLO–2 (%)	DDCLO–3 (%)	DDSFO–1 (%)	DDSFO–2 (%)
1,13-Tetradecadiene ($C_{14}H_{26}$)	0.85±0.01^{b}	0.87±0.02^{b}	0.82±0.01^{a}	nd	nd
Tricosane ($C_{23}H_{48}$)	0.22±0.01^{b}	0.23±0.01^{b}	0.14±0.01^{a}	0.59±0.01^{c}	2.45±0.01^{d}
Tetracosane($C_{24}H_{50}$)	0.12±0.01^{a}	0.14±0.01^{b}	0.12±0.01^{a}	nd	nd
Heptacosane ($C_{27}H_{56}$)	1.39±0.01^{c}	0.89±0.02^{a}	1.07±0.01^{b}	2.05±0.01^{d}	1.04±0.01^{b}
Octacosane ($C_{28}H_{58}$)	0.56±0.01^{c}	0.52±0.01^{b}	0.41±0.01^{a}	2.55±0.03^{d}	3.02±0.01^{e}
Nonacosane ($C_{29}H_{60}$)	6.30±0.02^{d}	5.79±0.04^{c}	6.32±0.02^{d}	5.58±0.05^{b}	4.12±0.01^{a}
Tetratriacontane ($C_{34}H_{70}$)	nd	nd	nd	0.66±0.01^{b}	0.55±0.01^{a}
Hexatriacontane ($C_{36}H_{74}$)	0.46±0.01^{b}	0.59±0.01^{c}	0.41±0.02^{a}	0.61±0.01^{c}	0.95±0.01^{d}
Squalene ($C_{30}H_{50}$)	3.49±0.01^{c}	3.29±0.04^{b}	2.97±0.05^{a}	4.51±0.03^{e}	4.33±0.12^{d}
α-Tocopherol ($C_{28}H_{48}O_2$)	1.89±0.06^{a}	2.05±0.04^{b}	1.95±0.03^{ab}	3.83±0.01^{c}	4.61±0.07^{d}
γ-Tocopherol ($C_{29}H_{50}O_2$)	3.39±0.07^{d}	1.72±0.01^{a}	3.85±0.02^{e}	2.68±0.05^{c}	1.95±0.01^{b}
Brassicasterol ($C_{28}H_{46}O$)	2.13±0.02^{a}	2.35±0.05^{b}	2.74±0.05^{c}	nd	nd
Campesterol ($C_{28}H_{48}O$)	5.19±0.14^{d}	5.39±0.23^{d}	4.19±0.05^{c}	1.78±0.01^{a}	2.06±0.02^{b}
Stigmasterol ($C_{29}H_{48}O$)	1.49±0.01^{a}	1.79±0.01^{b}	1.92±0.01^{c}	2.11±0.10^{d}	1.71±0.01^{b}
β-Sitosterol ($C_{29}H_{50}O$)	8.39±0.25^{a}	8.51±0.05^{a}	9.81±0.12^{b}	9.97±0.07^{b}	10.40±0.10^{c}
Lanosterol ($C_{30}H_{50}O$)	0.96±0.01^{c}	0.62±0.01^{b}	0.45±0.01^{a}	nd	nd
4,22-Stigmastadiene-3-one ($C_{29}H_{46}O$)	nd	nd	nd	1.16± 0.01^{a}	1.47±0.01^{b}
Stigmast-4-en-3-one ($C_{29}H_{48}O$)	5.09±0.05^{a}	5.59±0.03^{b}	5.12±0.10^{a}	6.86±0.06^{c}	5.83±0.11^{d}
Stigmastan-3,5-diene ($C_{29}H_{48}$)	2.83±0.11^{b}	2.52±0.36^{ab}	2.12±0.01^{a}	2.53±0.06^{ab}	2.15±0.31^{a}

nd: not detected, Values are presented as means ± SD of three replicates. Values within the same rows followed by the different superscript are significantly different at 95% significance ($p < 0.05$).

TABLE 4. Comparative results of sterols, tocopherols, and hydrocarbons present in deodorizer distillates of canola and sunflower oil.

Samples	Total sterols (%)	Total tocopherols (%)	Total hydrocarbons (%)
DDCLO-1	18.16±0.22^{b}	5.28±0.05^{b}	13.35±0.02^{b}
DDCLO-2	18.66±0.27^{c}	3.77±0.04^{a}	12.31±0.07^{a}
DDCLO-3	19.15±0.09^{d}	5.79±0.05^{c}	12.30±0.04^{a}
DDSFO-1	13.86±0.14^{a}	6.51±0.06^{d}	16.56±0.03^{c}
DDSFO-2	14.18±0.09^{a}	6.56±0.07^{d}	16.47±0.14^{c}

Values are presented as means ± SD of 3 measurements calculated within each sample after summation. Values within the same column followed by the different superscript are significantly different at 95% significance ($p < 0.05$).

being substituted by economical petroleum HCs [Kasim *et al*., 2009].

Squalene was found the main and important component of hydrocarbons family which was comparatively higher (4.3–4.5%) in DDSFO samples than in the DDCLO samples (3.0–3.5%). León Camacho *et al*. [2004] suggested that stigmasta-3, 5-diene is a steroidal hydrocarbon produced after the dehydration of β-sitosterol. Our samples contained stigmasta-3, 5-diene in the range from 2.1–2.8% which indicated the loss of β-sitosterol due to high temperature of deodorizer. Two components *i.e.* 4, 22-stigmastadiene-3-one and stigmast-4-en-3-one, could be the dehydrogenation products of stigmasterol and β-sitosterol during deodorization process. Stigmast-4-en-3-one was found at maximum of 5.1–6.9% in all samples of DD which was proportional to 4, 22-stigmastadiene-3-on (1.2–1.5%) that was only present in DDSFO samples [Naz *et al*., 2012]. Table 4 displayed the results of total sterols, total tocopherols and total hydrocarbons in each DD sample analyzed. Total sterols in DDCLO samples were significantly high in concentration (18.2–19.2%) than their concentration in DDSFO (13.9–14.2%). Total tocopherols and hydrocarbons estimated were significantly greater in DDSFO samples as compared to DDCLO and this is already mentioned in the literature [Carmona *et al*, 2010; Güçlü-Üstündağ & Temelli, 2007]. The small variations in the composition of each sample may be due to the sample preparation or might be affected by deodorization condition.

CONCLUSIONS

Use of industrial waste and byproducts for advantageous purpose is a basic requirement of the industry to fulfill with environmental and economic issues. Furthermore, replacement of natural products for synthetic materials has gained worldwide consideration in different areas like synthetic tocopherols BHA and TBHQ are now substituted with natural extract of tocopherols. Therefore, to find potential use of DD, characterization of its main component is very essential. In this study, chemical characterization of sunflower and canola deodorizer distillate samples was carried out. Results of the present study indicated that due to the presence of natural valuable components such as sterols, squalene, tocopherols and fatty acid in significant amount the DD could be used in various applications. The saponifiable part of these deodorizer distillates is a potential candidate for biodiesel production due to high FFA contents while the unsaponifiable part could be used in cosmetic industry because

it is a concentrated source of natural bioactive compounds such as sterols, squalene and tocopherols.

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