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### Consuming Fat Supplemented Diet with Different Vegetable Oils Impacts the Serum Lipid Profiles and Body Weights of Male Rats

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#### Abstract

This research was conducted in order to evaluate the influence of consuming diet supplemented with different types of fixed oils (Olive oil, Soybean oil and Coconut oil) on the lipid profiles and body weights of healthy albino rats and linking these effects to their chemical compositions thus reaching out to recommendation about the healthiest type of fats among the studied oils to be used in healthy nutrition programs. The lipid contents of the fixed oils were determined by Gas Chromatography (GC) method, the vitamin contents were determined by High Performance Liquid Chromatography (HPLC) and the proximate analyses of the oils were also evaluated by different assays. Twenty-four healthy male albino rats were divided into four groups as follows: Olive oil supplemented diet, Soybean oil supplemented diet, Coconut oil supplemented diet and oil-free supplemented diet control group. The dieting continued for 28 days, at the end of which, the serum levels of total cholesterol, Low Density Lipoproteins (LDL), High Density Lipoproteins (HDL), Triglycerides (TG) and Free Fatty Acid (FFA) were measured spectrophotometrically and the rats body weights were monitored on daily basis. The results showed that olive oil, the richest studied oil in unsaturated fatty acids (84.16 %), had favorable effect on the rats' lipid profiles, serum free fatty acid levels and rats body weights. In conclusion, olive oil supplemented diets are considered healthy diets and beneficial to decrease the risk of cardiovascular diseases.

Keywords: Olive oil; Soybean oil; Coconut oil; Fatty acids; Cholesterol

#### Introduction

Cardiovascular diseases (CVD) are the number one cause of death worldwide, responsible for the deaths of around eighteen million people every year, representing thirty-one percent of all global deaths. (Mendis *et al.* 2011) (World Health Organization 2017). Development of atherogenesis and coronary heart diseases have many risk factors including hyperlipidemia (Prasad *et al.* 2016). The American Heart Association Scientific Advisory reported that diets enriched with Monounsaturated Fatty Acid (MUFA) tend to raise the levels of High Density Lipoproteins (HDL) and lower that of triacylglycerol (Kris-Etherton 1999; Lichtenstein *et al.* 2006). Also, several epidemiological and clinical studies indicated that omega-3 Polyunsaturated Fatty Acid (PUFA) could prevent coronary heart diseases through different mechanism as antiarrhythmic, hypolipidemic and antithrombotic roles (Esrey *et al.* 1996; Howard *et al.* 2006; Rabrenovic 2011). A further evidence is that Mediterranean regions show significantly lower incidence of CVD, this could be attributed to their dietary habits (Martínez-González and Sánchez-Villegas 2004). Long-term intake of olive oil, a fundamental component of the Mediterranean diet, that contains high concentrations of MUFA, was proved to reduce the risk of developing hypertension (Alonso and Martínez-González 2004; Estruch *et al.* 2006). Accordingly, approaches for the prevention and treatment of coronary heart diseases should include population dietary modification (Rajaram *et al.* 2001; Hu and Willett 2002).

to reduce the risk of developing hypertension (Alonso and Martínez-González 2004; Estruch *et al.* 2006). Accordingly, approaches for the prevention and treatment of coronary heart diseases should include population dietary modification (Rajaram *et al.* 2001; Hu and Willett 2002). Balanced healthy diet is defined as having a variety of food that provides important nutrients for health maintenance and prevention of a number of serious conditions including mainly CVD. A healthy diet should contain adequate proportion of carbohydrates, fats, and proteins "macronutrients" along with the recommended daily allowances of vitamins, minerals "micronutrients". The recommended daily fat intake for healthy adults should be within 15 to 30 % of total caloric intake (World Health Organization 2008).

Unfortunately, many people were scared away from using vegetable oils during the low-fat craze of the last decades, as they believed in a myth that these oils are unhealthy food and hyperlipidemia is caused by increased fats proportion in diet, but actually hyperlipidemia was found then to be mainly attributed to the type of fats supplied rather than the amount (German and Dillard 2004). Scientific evidence has provided reliable and significant results approving the association between diets high in SFA and increased serum cholesterol level which ultimately leads to building up of fatty plaque on the walls of blood vessels, atherosclerosis and consequently heart attacks and strokes (Siri-Tarino *et al.* 2010). Other studies proved that atherosclerosis and CVD risks increase with the elevation of serum cholesterol as well as with the the type of fats. Also, it is well known that the dietary SFA and cholesterol

raise total blood cholesterol and low density lipoprotein (LDL) cholesterol levels, while PUFA lowers total blood cholesterol and LDL cholesterol levels (Pietinen et al. 1997; Xu et al. 2006). The (World Health Organization 2008) confirmed that replacing SFA with PUFA decreases LDL cholesterol concentration and the total/HDL cholesterol ratio. Thus, it is recommended to reduce the SFA intake to less than 10% of calories (Zimmerman and Snow 2012).

In keeping with this evidence, we decided to evaluate the impact of three vegetable oils namely, olive oil, soybean oil and coconut oil on the lipid profiles of healthy albino rats aiming to identify the healthiest type among the selected oils. The intention for using these oils in particular is the variability of the degree of saturation of their fatty acid content. This variability allows further confirmation to relate the degree of fatty acid saturation to the effect on lipid profile and consequently the incidence of CVD.

#### **Material and Methods**

Material and Methods
Material for phytochemical investigations

Olive oil (Olea europaea L., Family Oleaceae), soybean oil (Glycine max L. Merr., Family Fabaceae) and coconut oil (Cocos nucifera L., Family Arecaceae) were purchased from local Egyptian market. All chemical reagents and solvents were analytical grade (E-Merck, Darmstadt, Germany).
Authentic vitamins (A, B3, B6, B12, C and E) were kindly supplied by the Agricultural Research Center, Food Technology Research Institute, Giza, Egypt. Authentic Fatty acids and Hydrocarbons standards were kindly supplied by National Research Center, Giza, Egypt.

#### **Determination of vitamins by HPLC**

Determination of vitamins by HPLC The vitamin content of the three oils was assayed using an Agilent 1260 infinity HPLC Series (Agilent, USA), equipped with Quaternary Pump, a Kinetex XB-C18 Column 100 mm x 4.6mm (Phenomenex, USA), operated at 35 °C. The separation is achieved using a binary linear elution gradient with (A) 25 mM NaH<sub>2</sub>PO<sub>4</sub> PH=2.5 (B), Methanol (v/v). The injected volume was 20 µl. Detection: VWD detector set at 254 nm for ascorbic acid, 280 nm for vitamins A and E and 220 nm for vitamins B. Quantitation was achieved at by an internal standard method (Evangelisti *et al.* 1997). Vitamin C content was determined according to (Romeu-Nadal *et al.* 2006), fat-soluble vitamins (A determined according to (Romeu-Nadal *et al.* 2006), fat-soluble vitamins (A and E) were determined according to (Beyer and Jesnsen 1989) method while vitamins B was determined as described by (Batifoulier *et al.* 2005)

#### **Determination of mineral content**

The mineral content of the three studied oils was determined according to A.O.A.C guidelines (AOAC 1995). Oil samples were separately wet

digested in an Advanced Microwave Digestion System ETHOS 1 with concentrated sulfuric acid and digestion catalysts (a mixture of copper sulfate and anhydrous sodium sulfate, 1:10), then the digested solutions were measured using an Atomic Absorption Spectrometer (Inductively Coupled Plasma ICP-AES Spectrometer, iCAP 6000 series, Thermo Scientific).

#### Determination of amino acids content

The amino acids content of the three studied oils was analyzed on an Automatic Amino Acid Analyzer AAA400 INGOS Ltd. Czech Republic, adopting the method of (Cosmos and Simon-Sarkadi 2002).

# Determination of lipid content by GC For unsaponifiable matters

**For unsaponifiable matters** Unsaponifable matters were separated using HP- Hewlett Packard GC-system, series 6890 equipped with Flame Ionization Detector (FID). A capillary column (HP-5 5% phenyl methyl siloxane, 30m x 320 μm, film thickness 0.25 μm) was used in the separation. The injector port temperature was set 240°C (split mode) and the detector cell at 280°C. The flow rate of the carrier gas, N2, was 20 ml/min, for H2 20ml/min and for air 200ml/min. The column temperature was 80°C for 1 min and then increased to 280°C by the rate of 8°C/min, with maximum column temperature 325°C then isothermally for a total run time of 20 minutes for a total run time of 20 minutes.

#### For fatty acid methyl esters

For fatty acid methyl esters Fatty acid methyl esters were separated by the same GLC apparatus as unsaponifable matters. A capillary column (HP-5 5% phenyl methyl siloxane, 30m x 320  $\mu$ m, film thickness 0.25  $\mu$ m) was used in the separation. The injector port temperature was set 250°C (splitless mode) and the detector cell at 280 °C. The flow rate of the carrier gas, Hydrogen flow 30 ml/min, air flow 300 ml /min and N2 was 10 ml/min. The column temperature was 70°C for 1 min and then increased to 220°C by the rate of 4°C/min, then isothermally for a total run time of 20 minutes.

### **Experimental animals**

Twenty-four Sprague-Dawley male albino rats, weighting  $200 \pm 20$  gm obtained from the animal house of MSA University were used for this study. Prior to the initiation of the studies, the animals were randomized and assigned This to the initiation of the studies, the animals were randomized and assigned to treatment groups. Four rats were housed per cage (size  $26 \times 41$  cm) and placed in the experimental room for acclimatization a week before the experiment. The animals were fed with standard laboratory diet and with tap water ad libitum, and kept under hygienic conditions and in an air-conditioned animal room at  $23\pm1$  °C with a 12 h light/dark cycle.

#### **Experimental design**

Rats were randomly allocated into four groups of six animals each. Fed according to Table 1.

Group 1: Normal control group and had no oil in the feed Group 2 (Soybean oil): rats received the soybean oil supplemented diet 30% for  $2\bar{8}$  days

Group 3 (Coconut oil): rats received the coconut oil supplemented diet 30% for  $2\bar{8}$  days

Group 4 (Olive oil): rats received the olive oil supplemented diet 30% for 28 davs

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Table 1: Dietary	composition of	of olive oil,	soybean oil	and coconut oil	supplemented feed

Diet	Control	Olive oil group	Soybean oil	Coconut
composition	group		group	oil group
Corn	69	39	39	39
Fish meal	5	5	5	5
Groundnut	10	10	10	10
Bone meal	5	5	5	5
Palm oil	5	5	5	5
Soya bean oil			30	
Coconut oil				30
Olive oil		30		
Vitamin	1	1	1	1
Corn starch	5	5	5	5
Total	100	100	100	100

#### Blood samples and biochemical analysis **Preparation of blood samples**

At the end of the study, rats were fasted overnight, anesthetized with thiopental sodium (50 mg/kg) (Vogler *et al.* 2006) and blood samples were collected (5 ml per rat). Blood samples were centrifuged at 3000 rpm for 15 min after 30 minutes of collection and stored at  $-80^{\circ}$ C until analyzed. Serum Total cholesterol (TC), LDL, HDL, TG and FFA were determined using colorimetric assay kits.

#### **Biochemical analysis**

Analysis of serum was carried out for measuring TC, LDL and HDL levels using colorimetric assay kit (Biochain, USA), TG using enzymatic assay kit (XpressBio life science products, USA) and FFA using Abnova colorimetric assay kit

#### **Statistical analyses**

All data were expressed as mean  $\pm$  SD and analyzed using Prism program version 6. For all parameters, comparisons among groups were

carried out using one-way analysis of variance (ANOVA) followed by Bonferroni's multiple comparisons test. All P values reported are two-tailed and P<0.05 was considered significance.

#### **Ethics Statement**

Animal care and handling was performed in conformance with approved protocols of MSA University and Egyptian Community guidelines for animal care.

#### **Results**

#### Nutritional profiles of the oils

The fat soluble vitamins (A and E) clearly predominates the water soluble ones as (vitamin C and B). Soybean oil was found to have the highest concentration of vitamin A amongst the studied oils, while coconut oil had the highest concentration of vitamin E. The studied oils exhibited relatively high amounts of sodium, moderate amounts of potassium and low amounts of calcium. Soybean oil contains the highest concentration of sodium, coconut oil contains the highest concentration of calcium while olive oil contains the highest potassium proportion among the studied oils. As for the amino acids content, only coconut oil was found to contain most of the analyzed amino acids on contrast to the other two oils. Arginine was the predominant amino acid in the oils of soybean and coconut (27.77 and 26.96 mg/100ml) respectively. The nutritional profiles of the oils were presented in Table 2. **Table 2:** Nutritional profiles of olive oil, soybean oil and coconut oil

Reference	Olive Oil	Soybean Oil	Coconut Oil		
Vitamin content (µg/100ml)					
Niacin (B3)	4.45	Traces	Traces		
Pyridoxine (B6)	3.358	1.282	Traces		
Cobalamin (B12)	3.316	3.627	5.710		
Ascorbic acid	ND	4.244	3.706		
β-Carotene (A)	86.92	109.80	20.63		
α-Tocopherol (E)	1.12	5.52	228.40		
Mineral content (mg/10	00ml)				
Sodium	968.00	992.40	974.40		
Potassium	52.60	48.00	41.60		
Calcium	0.64	15.40	21.80		
Magnesium	18.20	18.60	21.20		
Iron	8.43	2.70	8.01		
Manganese	0.02	0.05	0.01		
Zinc	0.40	1.00	0.20		
Amino acids content (mg/100ml)					
Aspartic acid	ND	ND	0.46		
Theronine	ND	ND	0.68		
Seronine	ND	ND	6.07		

Glutamic acid	ND	2.57	4.48
Proline	ND	ND	ND
Glycine	ND	ND	ND
Alanine	0.95	ND	0.09
Valine	ND	ND	0.05
Methionine	ND	ND	ND
Isoleucine	ND	ND	ND
Leucine	ND	ND	0.28
Tyrosine	ND	ND	ND
Phenylalanine	ND	ND	0.22
Histadine	ND	ND	0.17
Lysine	5.14	2.95	4.37
Arginine	ND	27.77	26.96

ND: not detected

#### Lipid profiles of the oils

Following the saponification of the studied oils according to the British Pharmacopeia 2016. The unsaponifiable fractions were separated and analyzed using GC. While the saponifiable fractions, were separately used to prepare the fatty acid methyl esters of the corresponding oils adopting the method described by Ichihara and Fukubayashi (2010) and then analyzed by GC. As shown in table (3), twenty standard hydrocarbons (C11-C30) were assessed in each oil, along with five sterols namely, cholesterol, campasterol, stigmasterol,  $\beta$ -sitosterol and  $\alpha$ -amyrine. C27 n-Heptacosane was the most abundant hydrocarbon in all three oils. Cholesterol and  $\beta$ -sitosterol were not detected in coconut oil, while, campesterol and stigmasterol were detected in all studied oils.

Table 3: Results of GLC analysis of the identified unsaponifiable matter of olive of	il,
sovbean oil and coconut oil	

Reference	Relative percentage		
	Olive oil	Soybean oil	Coconut oil
C11 n-Undecane	1.0563	1.2675	ND
C12 n-Dodecane	0.0116	0.0627	ND
C13 n-Tridecane	0.4283	0.9834	ND
C14 n-Tetradecane	0.3239	0.863	0.0911
C15 n-Pentadecane	0.0829	2.9711	0.2758
C16 n-Hexadecane	1.1353	ND	0.7656
C17 n-Heptadecane	1.1341	0.2431	2.5571
C18 n-Octadecane	1.1398	5.9754	2.9365
C19 n-Nonadecane	4.6776	17.2111	3.7815
C20 n-Icosane	0.625	0.1543	5.3902
C21 n-Henicosane	15.7537	16.4123	7.3084
C22 n-Docosane	6.9490	ND	7.8579
C23 n-Tricosane	ND	ND	9.0166
C24 n-Tetracosane	ND	0.9615	7.8800
C25 n-Pentacosane	1.1349	3.3761	3.2816

C26 n-Hexacosane	1.388	14.9123	3.051
C27 n-Heptacosane	35.8514	18.3413	11.9952
C28 n-Octacosane	4.5681	0.3912	8.5935
C29 n-Nonacosane	5.1979	ND	4.4054
C30 n-Triacotane	3.747	6.7214	17.105
Cholesterol	3.1967	3.8923	ND
Campesterol	2.1837	2.2746	2.4451
Stigmasterol	2.8044	0.9882	0.4488
β-Sitosterol	3.4595	1.7364	ND
α –Amyrine	2.7817	ND	0.4994

ND: not detected

According to the results shown in table (4), the unsaturated fatty acids predominates the fatty acid fractions of olive and soybean oils with relative percentages of (84.16 % and 62.74%) respectively. On the other hand, the saturated fatty acids represented the majority of fatty acids of coconut oil (95.19 %). Olive oil contains the highest percentage of mono-unsaturated fatty acid (MUFA) as oleic acid (30.23%) and poly-unsaturated fatty acids (PUFA); linoleic acid (7.74%), linolenic acid (1.27%) and arachidonic acid (42.95%). Soybean follows olive oil in this percentage as it mainly contains linoleic acid (33.03%) followed by oleic acid and linolenic acid.

Table 4: Results of GLC analysis of the identified fatty acids of olive oil, soybean oil and

coconut oll				
Reference	Relative percentage			
	Olive oil	Soybean oil	Coconut oil	
Caprylic acid C8 (0)	ND	3.47744	7.49276	
Capric acid C10 (0)	2.23489	ND	5.87549	
Lauric acid C12 (0)	1.96350	1.92572	52.53590	
Tridecanoic C13 (0)	1.82106	ND	ND	
Myristic acid C14 (0)	3.97467	ND	19.79576	
Palmitic acid C16 (0)	3.23581	4.72993	7.09982	
Stearic acid C18 (0)	2.23573	14.97243	2.25345	
Behenic acid C22 (0)	ND	12.14963	ND	
Oleic acid C18 (1) ω9	30.23044	21.20814	4.23181	
Linoleic acid C18(2) ω6	7.73774	33.03382	0.57229	
Linolenic acid C18(3) ω3	1.26674	8.50289	ND	
Arachidonic acid C20(4) ω6	42.94693	ND	ND	
Percentage of unsaturated fatty acids	84.16	62.74	4.81	
Percentage of saturated fatty acids	15.84	37.26	95.19	

ND: not detected

### Effect of olive oil, soybean oil and coconut oil supplemented feed on serum lipid profiles

As shown in table 5 and figure 1, the mean serum level of total cholesterol was significantly increased in three different oil groups compared to the control group, P value was < 0.001. The mean serum level of HDL-

cholesterol was significantly raised in olive oil group compared to control group and the other two oils groups (P value < 0.001).

On the other hand, the mean serum level of LDL-cholesterol and TG were significantly reduced in the olive oil group compared to control group and the other two oils groups (P value < 0.001).

 Table 5: Effect of olive oil, soybean oil and coconut oil supplemented feed on serum lipid profiles

Groups	Total cholesterol	HDL-C	LDL-C	TG (mg/dL)
	(mg/dL)	(mg/dL)	(mg/dL)	
Control	$58.2 \pm 4.49$	$34 \pm 0.89$	$32.3 \pm 1.37$	$47.3 \pm 3.26$
Olive oil	66.8 ± 4.71 <sup>a</sup>	$35.8 \pm 0.75$ <sup><i>a</i></sup>	$24.5 \pm 1.05$ <sup><i>a</i></sup>	59 ± 2.82 <sup><i>a</i></sup>
Soybean oil	69.2 ± 3.13 <sup><i>a</i></sup>	$34.2 \pm 0.75$ <sup>b</sup>	$28.3 \pm 0.81$ ab	79.3 ± 6.12 ab
Coconut oil	$66.8 \pm 3.54^{\ a}$	32.7 ± 1.51 <sup>b</sup>	$28.3 \pm 0.81$ ab	73.1 ± 5.26 <sup>ab</sup>

Results were expressed as mean  $\pm$  SD and analyzed using one-way ANOVA followed by Bonferroni's post hoc test, <sup>*a*</sup> significant from control at P < 0.001, <sup>*b*</sup> significant from olive oil group at P < 0.001.





Fig 1. Effect of olive oil, soybean oil and coconut oil supplemented feed on serum lipid profiles

Results were expressed as mean  $\pm$  SD and analyzed using one-way ANOVA followed by Bonferroni's post hoc test, a: significant from control at P < 0.001, b: significant from olive oil group at P < 0.001.

### Effect of olive oil, soybean oil and coconut oil supplemented feed on serum FFA levels

The mean serum level of FFA was significantly increased in three different oil groups compared to the control group, P value was < 0.001.

On the other hand, the mean serum level of FFA was significantly reduced in the olive oil group compared to the other two oils groups (P value < 0.001). The results were presented in Table 6 and figure 2.

 Table 6: Effect of olive oil, soybean oil and coconut oil supplemented feed on serum free fatty acids levels

Groups	Free fatty acid (mg/dL)
Control	$14.9 \pm 0.96$
Olive oil	$18.02 \pm 0.99$ <sup>a</sup>
Soybean oil	$21.9 \pm 1.16^{ab}$
Coconut oil	$31.7 \pm 2.02^{\ ab}$

Results were expressed as mean  $\pm$  SD and analyzed using one-way ANOVA followed by Bonferroni's post hoc test, <sup>*a*</sup> significant from control at P < 0.001, <sup>*b*</sup> significant from olive oil group at P < 0.001.

The effect of different oils on serum free fatty acids level



Fig 2. Effect of olive oil, soybean oil and coconut oil supplemented feed on serum free fatty acids levels

Results were expressed as mean  $\pm$  SD and analyzed using one-way ANOVA followed by Bonferroni's post hoc test, a: significant from control at P < 0.001, b: significant from olive oil group at P < 0.001.

## Effect of olive oil, soybean oil and coconut oil supplemented feed on body weight in rats

The % $\Delta$  weight was significantly increased in coconut oil group compared to the control group, P value was < 0.01. The % $\Delta$  weight was significantly reduced in olive oil group compared to control, soybean oil and coconut oil groups (P value < 0.001). The results were presented in Table 7 and figure 3.

Groups	Initial weight (gm)	Final weight (gm)	%Δ weight (gm)		
Control	$152 \pm 11.02$	$178 \pm 10.3$	$16.9 \pm 3.44$		
Olive oil	$153 \pm 5.82$	$171 \pm 16.8$	$12.4 \pm 3.2^{a}$		
Soybean oil	$155 \pm 13.5$	$188\pm20.52$	$20.7 \pm 5.42^b$		
Coconut oil	$145 \pm 11.3$	$181 \pm 20.66$	$24.8\pm8.25^{ab}$		

 Table 7: Effect of olive oil, soybean oil and coconut oil supplemented feed on body weight in rats

Results were expressed as mean  $\pm$  SD and analyzed using one-way ANOVA followed by Bonferroni's post hoc test, <sup>*a*</sup> significant from control at P < 0.01, <sup>*b*</sup> significant from olive oil group at P < 0.001.

#### The effect of different oils on body weights in rats



Fig 3. Effect of olive oil, soybean oil and coconut oil supplemented feed on body weight in rats

#### Discussion

The results of the current study that tested the consumption of olive oil, soybean oil and coconut oil confirmed the variability of the chemical composition of the three oils which was further reflected on the lipid profiles, free fatty acids serum levels and body weights of the experimental rats. Our results showed that serum analysis of olive oil supplemented-feed rats had the highest HDL levels, significantly different from the control group. Similarly, previous studies related the elevation of HDL levels to the increased MUFA intake (as oleic acid in olive oil) (Aguilera *et al.* 2005). The present study supports this hypothesis as olive oil (rich in MUFA) showed better results than the other two oils. In addition, olive oil had a lowering effect on LDL levels; this finding was in accordance with (Lastra *et al.* 2001), this may be also attributed to its content of MUFA. (Kien *et al.* 2013). Basically, unsaturated fatty acids lower serum LDL by different mechanisms including redistribution of cholesterol between plasma and tissue pools (Matson and Grundy 1985), up-regulation of the LDL receptor (Fernandez and McNamara 1989) or increasing LDL apoB-100 fractional catabolism (Turner *et al.* 1981). Accordingly, it can be concluded that, the positive effect of olive oil

Accordingly, it can be concluded that, the positive effect of olive oil on lipid profile is due to the predomination of UFA (84.16 %) over the SFA fractions. This attribution is supported by the results of several studies (German and Dillard 2004; Lefevre *et al.* 2005; Fernandez and West 2005 and World Health Organization 2008). Moreover, olive oil was found to have the least elevating effect on TGs among the other oils. Previous studies had shown that the positive effect of olive oil on lipid profile is not only restricted to MUFA and PUFA percentages; but also to the synergistic role of minor constituents (Al Juhaimi *et al.* 2017). Therefore, in this study, we examined the vitamins, minerals and amino acids contents of the three oils.

Coconut oil, rich in SFA, showed a similar effect to that of olive oil on the serum cholesterol levels,

This can be justified by the assumption of the biochemical link between the tocopherol levels (found in a significantly higher proportion in coconut oil 228.40 µg/100ml than olive oil 1.12 µg/100ml) and the degree of unsaturation in vegetable oils (Kamal Eldin and Anderson 1997). Another justification to this result, may be related to its high content of Medium Chain Fatty Acids (MCFA) (approximately 66%); MCFA are absorbed and transported through the portal vein to the liver where they are rapidly oxidized, generating energy (Schumacher *et al.* 2016). According to (Liu *et al.* 2017), MCFA reduce serum cholesterol by regulating the metabolism of bile acid.

MCFA reduce serum cholesterol by regulating the metabolism of bile acid.
Our study proved that coconut oil has the least favorable effect on HDL; this finding is in agreement with (Paz *et al.* 2010). However, (Feranil *et al.* 2011) reported positive association between coconut oil intake and the HDL levels.

Soybean oil showed a decreased LDL and an increased triglycerides serum levels more than the control group but less than olive oil fed group. The content of PUFA (linoleic acid 33.03%) in soybean may be the reason for this, as there is a relationship between PUFA and increasing the sensitivity and number of LDL receptors (Mustad *et al.* 1996). However, the difference

between the effects of the olive and soybean oils can be attributed to the presence of a relatively lower total unsaturation percentage in soybean oil than olive oil. Our results revealed that the serum levels of free fatty acids were significantly reduced in the olive oil group compared to the other two oils supplemented-feed groups. Where free fatty acids are intermediary metabolites that are formed from the hydrolysis of triglycerides by lipoprotein lipase and hormone-sensitive lipase, so that could explain its effect on reducing the TG in blood as well.

Also, this research verified that olive oil supplemented diet was superior in its effect on the rats' body weight where the difference in the body weights before and after dieting on olive oil was  $(12.4 \pm 3.2)$  that was even lower than that of the control group  $(16.9 \pm 3.44)$  and much lower than the other two oils supplemented groups.

#### Conclusion

The results reported herein confirmed that unsaturated fatty acids are primarily recommended for daily intake. Olive oil, the richest studied oil in UFA, showed favorable effect on lipid profiles, therefore, it is regarded as a strong evidence for assuming this recommendation. Olive oil has the least elevating effect on total cholesterol among the other oils. Olive oil significantly decreases LDL cholesterol, and triglycerides. On the other hand, olive oil significantly elevates HDL which is the good cholesterol exerting the cardio-protective effects. Olive oil supplemented diet significantly reduced the serum levels of free fatty acids and did not increase the rats body weight. Hence, olive oil supplemented diets could be of clinical benefit to individuals at risk of cardiovascular disease.

#### **Conflict of interests**

The authors declare no conflicts of interest associated with this publication.

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