The implications of sunflower seeds and flax seeds on plasma lipids profile and fatty acids profile-IL-6 correlation in laying hens: A comparative study

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Abstract- **The present study was done to examine the ascendency of incorporating plant source of Omega-6 and omega-3 fatty acids into laying hens diet, on the plasma lipids profile and plasma fatty acids profile-IL-6 correlation. Seventy five bird (Hisex-breed), 20 week old, were brought from animal production research center (Kuku), they were halved into two groups, group (A) was signed as control group, subjected to a diet based on corn, group (B) was under diet contains 10% flax seeds and group (C), was under basic diet supplemented with 10% sunflower seeds, the three formulae were designed to sustain the requirements of laying hens as recommended by (NRC).**

The trial duration was eight weeks, blood samples were collected once per month (week 4 and 8), in EDTA coated vials, immediately placed into iced-container, centrifuged at 3000rpm/20 min, samples were separated in aliquot, and stored at -20°C and -80°C until analysis.

Saturated fatty acids and Arachidonic acid levels were significantly ($p<0.05$), high in control group (A), while the summation of omega-6 fatty acids was significantly ($p<0.01$) high in sunflower seeds treated group. The control group (A), revealed significant high concentration of plasma cholesterol, triglycerides and LDL-Cholesterol, while the HDL-Cholesterol level was enhanced by the addition of sunflower seeds and flax seeds. Adding 10% of sunflower seeds and 10% of flax seeds; to the basic diet of laying hens subsequently reduced the level of plasma IL-6 significantly compared to the control group (A).

***Index Terms*— Omega-6, Omega-3, Arachidonic Acid, HDL-Cholesterol, IL-6-Laying hens**

Abbreviations: LDL: Low Density Lipoprotein, HDL: High Density Lipoprotein, EFA: Essential Fatty Acids, PUFA: Poly Unsaturated Fatty Acids, NFE: Nitrogen Free Extract, FID: Flame Ionization Detector, LA: Linoleic Acid, AA: Arachidonic Acid, LC-PUFA: long chain-PUFA, GLA: Gamma Linolenic Acid, SCD: Stearoyl Co-A Desaturase.

I. Introduction

The unsaturated fatty acids are classified into three main families; n-3, n-6 and n-9, due to which carbon atom from the methyl end where the first double bond is attached.

Due to lack of enzymes essential for desaturation at carbon atoms 3 and 6, the n-3 and n-6 families cannot be produced in the body. Thus, the parent fatty acids in these families (linolenic acid [18:3 n-3] and linoleic acid [18:2 n-6], respectively) are essential and can only be derived from the diet.

Essential fatty acids (EFA) and their derivatives played the major role in lipid metabolism, platelet functions, immune system, inflammatory response, and epidermal functions, it is important to assure optimal EFA intakes (Sardesai, 1992). Dietary essential fatty acids (EFA), linoleic acid [18: 2(n-6), LA] and alpha-linolenic acid [18:3(n-3), ALA] are converted to long-chain polyunsaturated fatty acids (LCPUFAs) by desaturase and chain-elongation enzyme systems (Holman, 1998). Phospholipids, cholesterols, saturated fatty acids and monounsaturated fatty acids can be synthesized de novo within the human body. Because mammals cannot introduce a double bond beyond the delta-9 position in the fatty acid chain,

however, linoleic (n-6) and linolenic acid (n-3) must be ingested in the diet (Wayne; Fenton and Michael, 2000). Each of these EFA is in turn the substrate for further desaturation and elongation.

The ω-3 and ω-6 PUFAs play an important role in health and treatment of diseases. They can act as antibacterial agents (Ward and Singh, 2005)., (Guedes; Amaro and Malcata, 2011); (Huang and Ebersole, 2010), anti-inflammatory agents (Pulz and Gross, 2004).,(Schmitz and Ecker, 2008), antioxidants (Plaza; Herrero; Cifuentes and Ibanez, 2009) in the treatment of cardiovascular diseases (Mozaffarian and Wu, 2011), and cancer cell proliferation (Field and Schley, 2004., Das; Zuniga and Ojima, 2009), Such properties are indicative of the potential for PUFAs as nutraceuticals and as pharmaceuticals. The (ω-3/ω-6) ratio can significantly influence the body's metabolic function (Candela; Lopez and Kohen, 2011).

N-6 PUFA, also known as linoleic acid (LA) is thought to be pro-inflammatory and it can be converted into arachidonic acid (AA), (Myers and Allen, 2012). AA is the major substrate for eicosanoids production, which plays an important role in regulating inflammatory and immune responses (Calder, 2006).

There is one omega-6, however, called gamma linolenic acid (GLA) with an impressive set of disease-fighting powers. New research reveals this nutrient's power to combat chronic inflammation, eczema, dermatitis, asthma, rheumatoid arthritis, atherosclerosis, diabetes, obesity even cancer(Belch and Hill, 2002); (Surette: Stull and Lindemann, 2008); (Boyce; Schirmer and Phinney, 2007) and (Das, 2005).

GLA also shows promise in lowering low-density lipoprotein (LDL) and triglyceride levels, while increasing high-density lipoprotein (HDL) concentration (Guivernau; Meza; Barja and Roman, 1994).

Sunflower oil has gained importance due to increased content of oleic and linoleic acid that may help diminishing the cholesterol leading to reduction in heart diseases (Chowdhury *et al*. 2007). Also, cholesterol content and cancer risk is controlled by the presence of high content of phytosterols in sunflower seeds, *i.e*., approximately 280 mg per 100 gm (Phillips *et al*. 2005). Tocopherols in sunflower oil protect body from inflammation and tumors by neutralizing free radicals and avoiding oxidative injury to cells, thus helpful in diseases like rheumatoid arthritis and bronchial asthma. Vitamin E has a positive effect on coronary system of the body and hence reduces stroke and atherosclerosis (Dutta, 2003 and Singh *et al*. 2005).

The essential parent N-3 PUFA, linolenic acid is present in very high concentrations in flaxseed `oil (55%) but occurs also in other vegetable fats, especially rapeseed and soybean oil. Important dietary sources for the very long-chain n-3 PUFA eicosapentaenoic acid (20:5 n-3) and docosahexaenoic acid (22:6 n-3) are fish, particularly oily fish such as salmon, **(**Ratnayake and, Galli, 2009**).**

There is an important amount of scientific data as related to the beneficial and protective effects of n-3 omega PUFAs and their effects against inflammation, cancer and heart diseases. The association between major and bipolar depression, schizophrenia, pregnancy quality, osteoporosis, renal failure, **(**Ugur and Chris, 2010**).**

Besides the anti-inflammatory effect of n-3 omega PUFAs, the other beneficial and protective effect of these fatty acids is on the cardiovascular system. Tran's fatty acids, cholesterol and saturated fats are mainly responsible for atherosclerosis. On the contrary, n-3 omega PUFAs are beneficial in reducing cholesterol and thus the risk of myocardial infarction, **(**Zyriax and Windler, 2000**).**

II. Materials and Methods

The experiment was held in the Veterinary Research Institute (VRI), from January to march 2014. The duration of the experiment was eight weeks.

Four, full wire cages were made, each cage was (2X 1.5X 1 meter), and the capacity of each cage was 15 birds. The cages were placed at an open poultry house.

Seventy five laying hens (Hisex) breed, 20 week old, obtained from animal production research center (kuku), were utilized in this study. The birds were divided into three groups, 25 birds per group.

Experimental Diets: The diets were formulated to meet the requirements of egg production according to the directions of the national research council**,** (NRC, 1994). Three formulae of diets were prepared, 10% of flaxseeds and 10% of sunflower seeds were inserted into the experimental groups.

The supplementary sources were subjected to proximate analysis, to determine its content of protein, fat, fiber, N.F.E and energy.

❖ **Management:**

Each group received its diet from day one. Drinking system contained two tanks for each cage, the tanks were cleaned, and the water was changed twice daily. Birds received 24 hour light/day throughout the experiment.

Three (ml) of blood was collected from twenty bird of each group, the blood was taken using a three (ml) syringe, and received into EDTA coated vials, and immediately were kept in iced container, the samples were centrifuged at 3000 **rpm** for 20 minutes, and plasma was separated in aliquot and transferred into plane vials. Plasma samples were stored at -20 and -80°C until analysis.

❖ **Fatty Acids Analysis:**

Lipids were extracted in chloroform-methanol (2:1 v/v), according to the method of, **(**Folch, *et al*, 1957**).**

Methyl esters of the lipid extract were prepared according to, **(**Wang; *et al*, 2000**).**

Table (1): proximate analysis of supplementary sources:

	D.M%	Moisture%	Protein %	Fat %	Fiber%	Ash%	N.F.E%	Energy%
Flaxseeds Seeds	**96.55**	**3.45**	**27.84**	**35.69**	**10.78**	**4.67**	**17.57**	**3932.0**
Sunflower seeds	**98**	**2**	**26.00**	**36.49**	**11**	**6**	**18.5**	**3054.07**

Table (2-1): Diets composition:

Group Raw Materials%	Group A	Group B	Group C
Corn %	70	60	59.0
Wheat hull %	0	4.1	5.4
Groundnut cake %	14.3	10	10
Concentrate %	5	5	5
Calcium Carbonate%	10	10	10
Salt (Nacl) %	0.125	0.25	0.125
Methionine %	0.34	0.34	0.31
Lysine %	0.15	0.15	0.11
Mycofix %	0.1	0.1	0.1
Flax seeds%	----	10	-----
Sunflower seeds%	----	-----	10
Premix	0.1	0.1	0.1

**Supplied per kilogram of diets: Vitamin A, 5000 IU; Vitamin D ,500 IU; Vitmin E, 5 IU; Vitamin K, 1 IU; Vitamin B , 1.5 mg, Vitamin B , 2.5mg, 1 2 Ca-pantothenate, 2.5mg, niacin acid, 10 mg; pyridoxine,3mg; biotin, 0.1mg; folic acid, 0.25mg; Vitamin B , 0.005mg. Supplied per kilogram 12 b of diets: MnSO. 7H O100mg, FeSO. 7H O, 220mg; ZnSO . 7H O, 150mg; CuSO . 7H O, 20mg; KI, 2mg; Na SeO, 0.4 mg.*

- ***A**=Control group, **B**=10% flaxseeds supplemented group, **C**= 10% Sunflower seeds supplemented group.*

Table (2-2): Nutritional values calculated:

Parameter	Group A	Group B	Group C
ME. Kcal/Kg	**2729**	**2832**	**2821**
C.P%	**17.25**	**18.13**	**18.24**
E.E%	**5**	**6.4**	**6.2**
C.F%	**4.2**	**4.3**	**4.1**
Available phosphorus%	**0.52**	**0.65**	**0.63**
Calcium%	**3.9**	**3.9**	**3.9**

Table (3): Fatty acids profile of control and experimental group diets

Fatty acid	Control group (A)	Flaxseeds supplemented group (B)	Sunflower supplemented group (C)
(g/100 g total fatty acids)			
SFA	33.0187	11.4265	9.9754
MFA	24.1065	16.22	34.6841
PUFA	42.8749	72.3778	55.3437[A]
C18:3 (*n*-3)	1.6975	49.1068	10.5616
Σ*n*-3	1.6975	49.1068	10.5616
C18:2 (*n*-6)	23	11.6735	31.0756
C20:4 (*n*-6)	10.9769	3.4817	6.5908
Σ*n*-6	33.9769	15.1552	37.6664
PUFA/SFA	1.3	6.33	5.6
Σ*n*-3/Σ*n*-6	0.05	3.24	0.3

***SFA**= Saturated fatty acids, **MFA**=Mono unsaturated fatty acids, **PUFA**=Poly unsaturated fatty acids, **C18:3 (n-3)** =Linolenic acid (omega-3), **C18:2 (n-6)** =Linoleic acid (omega-6),* **C20:4 (*n*-6) =** Arachidonic acid (omega-6).

- ***A**=Control group, **B**=10% flaxseeds supplemented group, **C**= 10% Sunflower seeds supplemented group.*

❖ **Gas Chromatograph Analysis:**

Fatty acid composition was determined using (Shimadzu-2010) gas chromatograph, fitted with Flame ionization detector (FID). Separation of fatty acids was achieved using DB-WAX column, serial number (us6551263 H), of 0.25um film thickness, 30 meter length and 0.25 mm inner diameter.

Fatty acids methyl esters were identified by comparison of retention times with standards, and expressed as percentage of methyl esters.

III. Determination of lipids profile

Cholesterol was measured using commercial kits (Spectrum, Cairo, Egypt), using spectrophotometric method described by, and **(**Ellefson; *et al*, 1976**)** and **(**Roesclau; *et al*, 1974**).** The intensity of the colored complex is proportional to the cholesterol concentration. The absorbance was measured at a wave length 500 nm.

Triglycerides were measured using commercial kits (Spectrum, Cairo, Egypt), using spectrophotometric method described by, and **(**Bucolo and David, 1973**).** The intensity of the colored complex is proportional to the triglycerides concentration. The absorbance was measured at a wave length 500nm.

HDL-Cholesterol was measured using kits (Spectrum, Cairo, Egypt). The method used is a spectrophotometric method; described by **(**Lopez-Virella, 1977**).** The optical density was read against the blank at 546nm.

LDL-Cholesterol was measured using kits (Bio-system, reagent and instruments, Barcelona, Spain). The method used is a spectrophotometric method described by, **(**Assmann; *et al*, 1984**).** The optical density of the sample was read against the blank at 500 nm.

For the quantitative measurement of chicken Interleukin-6 (IL-6), a sandwich enzyme-linked immunosorbent assay (ELISA) kit (Assay Company), was used. The principle of the test is that the IL-6 is measured using a sandwich monoclonal antibodies in a biotin system ELISA.

Results are calculated by using curve-fitting of standards of known concentration. The IL-6 amount in each sample is determined by interpolating from the absorbance value (Y axis) to IL-6 concentration (X axis) using the standard curve equation.

IV. Statistical Analysis

The data were analyzed by using Statistics-10 program designed for Windows. Differences between obtained values were carried out by analysis of variance (ANOVA) the LCD test was used for determining the significance level of at least $p<0.05$.

V. Results

Saturated fatty acids level was significantly ($p<0.05$), high in control group (A), compared to the treated groups, on the other hand group (C), showed significant ($p<0.05$), higher level of plasma saturated fatty acids compared to group (B).

The highest plasma concentration of poly unsaturated fatty acids, was recorded by group (B), followed by group (C), the difference was significant at ($p<0.05$), compared to the control group (A).

The total detected omega-6 fatty acids, was significantly high ($p<0.05$), in plasma of group (C), compared to group (A) and (B).

Flaxseeds supplemented group (B), showed significant ($p<0.01$), low plasma concentration of omega-6 fatty acids compared to the control group (A).

The summation of the detected omega-3 fatty acids, was significantly ($p<0.01$), high in flaxseeds supplemented group (B), compared to group (A) and (C).

The control group (A), recorded significant ($p<0.05$), high concentration of plasma Arachidonic acid compared to the treated groups.

Arachidonic acid was not detected at all in the plasma of group (B), which received 10% flaxseeds.

The highest concentration of plasma Linoleic acid, was observed in group (C), followed by group (A), the difference was significant at ($p<0.05$).

Flaxseeds treated group (B), recorded the highest level of plasma Linolenic acid, compared to group (A), followed by group (C), the difference was significant at ($p<0.01$).

Table (4): Plasma Fatty acids profile of control and experimental groups

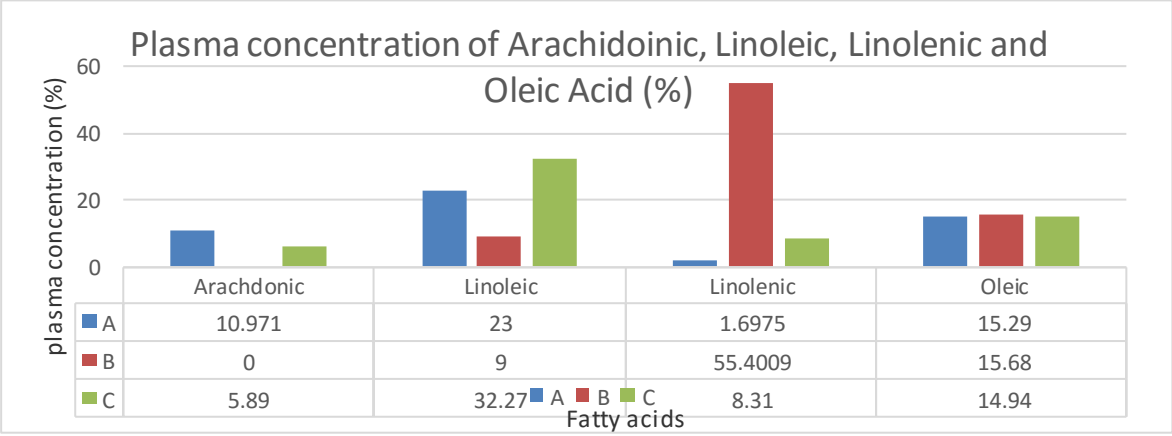
Fatty acid	Control group (A)	Flax seeds supplemented group (B)	Sunflower seeds supplemented group (C)
(ml/100 ml total fatty acids)			
SFA	33.01^{A}±1.6840	6.68^{B}±1.6840	10.11^{B}±1.6840
MUFA	24.1^{B}±2.1320	25^{B}.94±2.1320	33.68^{A}±2.1320
PUFA	42.87^{B}±2.3130	67.4^{A}±2.3130	55.34^{AB}±2.3130
C18:3 (*n*-3)	1.69^{C}±1.7127	55.4^{A}±1.7127	8.31^{B}±1.7127
Σ*n*-3	1.695^{C}±1.8347	58.4^{A}±1.8347	8.31^{B}±1.8347
C18:2 (*n*-6)	23^{B}±1.1862	9^{C}±1.1862	32.27^{A}±1.1862
C20:4 (*n*-6)	10.97^{A}±0.5346	0^{C}±0.5346	5.89^{B}±0.5346
Σ*n*-6	$33.^{97AB}$±1.1808	9^{C}±1.1808	38.16^{A}±1.1808
Σ*n*-3/Σ*n*-6	0.05^{A}±0.6359	10.1±0.6359	0.3^{A}±0.6359

Data are means ± standard error. Means in the same raw followed by the same letters are not significantly different at (p<0.05).

***SFA**= Saturated fatty acids, **MUFA**=Mono unsaturated fatty acids, **PUFA**=Poly unsaturated fatty acids, **C18:3 (n-3)** =Linolenic acid (omega-3), **C18:2 (n-6)** =Linoleic acid (omega-6),* **C20:4 (*n*-6) =** Arachidonic acid (omega-6).

- ***A**=Control group, **B**=10% sunflower seeds supplemented group, **C**= 10% Sunflower seeds supplemented group.*

- ***Figure (1):***



Week four showed that, the control group (A); recorded significant ($p<0.01$) high level of plasma cholesterol compared to group (B) and (C), while there was no significant different concentration of plasma cholesterol between the latest groups, While, a significant ($p<0.01$) low concentration of plasma cholesterol in group (B) and (C) compared to the control group (A) was recorded by the end of week 8.

There was no significant different levels of plasma triglycerides observed between all groups at the 4th week, while by the end of week 8, a significant ($p<0.01$) high concentration of plasma triglycerides was noticed in the control group (A); compared to group (C) and (B).

The control group (A), showed significant ($p<0.05$), high concentration of plasma LDL, compared to group (B), while the difference was not significant compared to group (C), this difference became pronounced at week 8, and it was significant at ($p<0.05$).

The levels of plasma HDL; at week **4** was not significant between the treated groups, while by the end of the **8th** week, The flaxseeds supplemented group, continued at the top with significant ($p<0.05$), high concentration plasma **HDL**, compared to group (A) and (C).

Group (B) and (C), at the fourth week, showed a significant ($p<0.01$) and ($p<0.003$) low concentration of IL-6, respectively, compared to the control group (A). By the end of week **8**, a significant ($p<0.01$) low concentration of plasma IL-6, in group (B), compared to group (A) and (C).

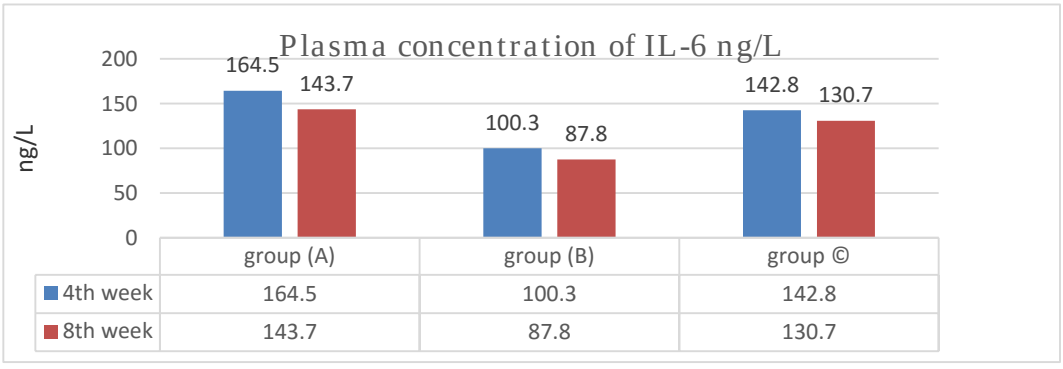
Group (C), showed significant ($p<0.01$) low level of IL-6, compared to the control group (A).

Table (5): Plasma lipids profile:

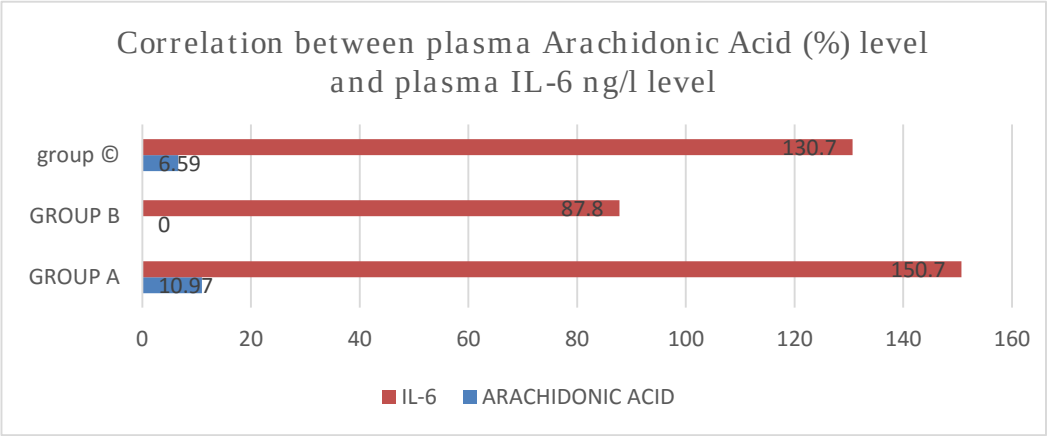
Parameter	Cholesterol mg/dl		Triglycerides mg/dl		HDL-Cholesterol mg/dl		LDL-Cholesterol mg/dl	
Group	**4th week**	**8th week**	**4th week**	**8th week**	**4th week**	**8th week**	**4th week**	**8th week**
Control (A)	219.20^{A} ±8.05	221.08^{A}± 8.17	419.13^{A}± 24.15	402.18^{A}± 16.66	11.726^{B}± 0.68	10.970^{C}± 0.4987	78.740^{A}± 5.81	80.920^{A}± 4.81
Group (B)	191.53^{B} ±8.05	168.00^{B}± 8.17	366.47^{A}± 24.15	311.64^{B}± 16.66	13.540^{A}± 0.68	14.160^{A}± 0.49	64.054^{B}± 5.81	62.900^{B}± 4.81
Group (C)	187.13^{B} ±8.0536	161.08^{B}± 8.1765	396.79^{A}± 25.007	317.82^{B}± 16.669	12.706^{AB} ±0.6835	12.676^{B}± 0.4987	67.074^{AB} ±5.8106	65.320^{B}± 4.8108

Data are means ± standard error. Means in the same columns followed by the same letters are not significantly different at ($p<0.05$).

- **Figure (2):**



- **Figure (3):**



VI. Discussion

Compared to the first month, the second one recorded significant low levels of plasma cholesterol in treated groups compared to the control group., this result confirms what was reported by (Basmacioglu, 2003), they found that, inclusion of fish oil and flax oil in diet, decreased serum cholesterol concentration of laying hens, the same result was reported by (Van Elswyk; Hargis; Williams, and Hargis., 1994), who demonstrated that dietary fish oil supplementation at 3% inclusion level, resulted in decreased serum cholesterol concentration in hens.

The current study results are corroborated also by which was reported by (Shivani and Sunil., 2013), that sunflower ethanolic extract, had lowering effect on plasma total cholesterol in diabetic rats, compared to control diabetic rats, and normalize effect compared to the normal control rats, also our results are supported by what was reported by (Hazim; *et al*., 2010), who mentioned that inclusion of flax oil reduced plasma cholesterol of laying quail, another supportive result was stated by (Wei-meng; *et al*., 2012), who reported that total cholesterol levels were significantly low in ducks received diets included sunflower oil.

Another findings confirm our current result, what was reported by (Sayed, 2013), that serum cholesterol concentration, was reduced in hens; received menhaden oil.

Newman; Bryden; Fleck; Ashes; Buttemer; Storlien, and Downing, (2002), showed that dietary PUFA, reduced plasma cholesterol in broiler chickens, when compared to groups fed saturated fatty acids SFA. Omega-3 fatty acids, suppress the synthesis of triglycerides and Apo lipoprotein B, increase the removal of VLDL, by peripheral tissue or the liver, and increase the excretion of bile in feces (Leaf and Webber., 1988), which can also reduce the serum concentration of cholesterol and triglycerides.

The results of the current study are in agreement with (Wei-meng; *et al*., 2012), who reported that, the inclusion of fish oil (omega-3 source), and sunflower oil (omega-6 source), had significantly reduced the serum triglycerides level, these results could be attributed to the high content of PUFA in diets, which had been reported as triglycerides and cholesterol reduction factor. The diet poly unsaturated fatty acids content of the current study confirms these results.

Another result corroborate the current findings, is what was reported by (Tejsawi; *et al*., 2013), that, supplementing rats with flax oil had reduction effect on triglycerides level.

The triglycerides result, is confirmed by our plasma fatty acids profile result, which is supported by what was reported by (Donald and Jump., 2002), that PUFA, induces several genes encoding proteins involved in fatty acid transport and binding, fatty acyl CoA formation, and oxidation (mitochondrial, peroxisomal and microsomal). This treatment also represses many genes encoding enzymes involved in de novo lipogenesis. Enhanced fatty acid oxidation coupled with inhibition of de novo lipogenesis (DNL) likely contributes to the decline triglyceride secretion (as VLDL) from rodent liver as well as the hypolipidemic effect of n3-PUFA. This mechanism shifts hepatic lipid metabolism from lipid synthesis and storage to oxidation.

The control group (A), showed significant ($p<0.05$), high concentration of plasma LDL, compared to the treated groups, and flaxseeds supplemented group, recorded significant ($p<0.05$), high concentration plasma HDL, compared to group (A) and (C), by the end of the trial period.

The current study revealed significant low level of plasma LDL-Cholesterol concentration in the treated group (B), compared to the control one (A), Flax seeds supplementation had a contrary effect on plasma HDL-Cholesterol, which was significantly enhanced, an auxiliary findings to our result was reported by (Simopoulos, 1991), who studied the effect of dietary omega-3 fatty acids on factors and mechanisms involved in the development of inflammation, atherosclerosis and immune diseases; the observed a reduction in LDL-cholesterol, triglycerides and an increase in levels of HDL-cholesterol, also our current observation is accommodated with the findings of (Mandaescus, and Mocana, 2005); when they fed hyperlipidemic patients diets supplemented by flaxseeds, the patients showed significant reduction in total cholesterol, triglycerides and LDL-cholesterol.

These results also could be justified by the modulation of dietary fat composition which affects serum lipid concentrations, replacing SFA and *Trans* fatty acids (TFA) with *cis*-PUFA lowers serum LDL-cholesterol concentration.

Group B, which received 10% flax seeds, showed significant low concentration of plasma IL-6, compared to group C and the control group A, this result is corroborated with what was mentioned by (Ashraf, 2007), that PUFAs, suppress pro inflammatory cytokines such as interleukins, and

tumor necrosis factor (TNF) and thus function as endogenous anti-inflammatory molecules.

Also the potency of omega-3 in particular, to give rise to relatively anti-inflammatory eicosanoids and much more inhibitors of innate immune activation (Lee, and Hwang. 2006), could be a justification of the current result.

The high concentration of IL-6, in group C, which was recorded at the first month could be attributed to the inflammatory eicosanoids produced from Arachidonic acid, which derived from linoleic acid.

By the end of the trial, group C, which was supplemented by 10% sunflower seeds, showed significant low level of IL-6 compared to control group, the possible anti-inflammatory effect of omega-6 fatty acids (Das., 2007), could be an explanation of this result, the mechanism involved in this reduction is within the metabolic pathway of linoleic acid, gamma linoleic acid, which derived from linoleic acid, reduces inflammation by activating the powerful peroxisome proliferator-activated receptors (PPAR), which are intracellular receptors that modulate cell metabolism and response to inflammation, also GLA, has impeding the ability of arachidonic acid to convert to detrimental inflammatory molecules (Hontecillas; O'Shea; Einerhand; Diguardo, and Bassaganya-Riera., (2009); Blech, and Hill., 2000), also the plasma arachidonic acid concentration in the control group A, justify the high level of plasma IL-6 in the plasma of group A compared to group C, the concentration was 10 and 6% of the total detected fatty acids respectively.

Since Arachidonic acid; is the precursor of the inflammatory agents such as PGE2, cytokines, interleukin-1 (IL-1), interleukin-2 (IL-2), interleukin-6 (IL-6), Our current result of plasma fatty acids profile is synergetic to the results of IL-6, in different groups, the Arachidonic acid in control group by the end of week 8 was 10% of total detected fatty acids, will group C, recorded 6% and the mentioned acid was not detected in group B, which received 10% flax seeds as a source of omega-3 fatty acids, which alter the pro-inflammatory cytokine genes expression, perhaps by altering the intracellular signaling mechanisms that lead to activation of pro-inflammatory cytokine genes. This might occur through inhibition of activation of transcriptional factors, such as NF-κβ, which regulate activation of tumor necrosis factor-alpha (TNF-α), interleukin-1beta (IL-1β), and interleukin-6 (IL-6). The NF-κβ is activated by phosphorylation, often by protein kinase C, and subsequent dissociation of its inhibitory subunit (Alwi; Santoso, and Suyono. 2007).

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