

The Effects of Varieties Sources of Omega-3 Fatty Acids on Diabetes in Rats

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ABSTRACT

The aim of this study was to investigate the effects of varieties sources of omega-3 on diabetic rats. Fifty six male albino rats were divided into 7 groups: first group was fed on normal basal diet (maintained at negative control group), group 2 diabetic group fed on 60% fructose, group 3 fed on 60% fructose + 8% fish oil, group 4 fed on 60% fructose + 8% flaxseed oil, group 5 fed on 60% fructose + 4% corn oil + 4% fish oil, group 6 fed on 60% fructose + 4% corn oil + 4% flaxseed oil and group 7 fed on 60% fructose + 4% fish oil + 4% flaxseed oil. Fatty acid composition of investigated oils showed that the corn oil contained high levels of omega-6 fatty acid (LA: 56.95%), flaxseed oil contained high level of short chain omega-3 fatty acid (ALA: 56.31%), meanwhile fish oil only has high concentration of long chain omega-3 fatty acid 39.20% (23.98% of EPA and 15.22% of DHA). After 8 weeks feeding of fish oil, flaxseed oil, corn oil + fish oil, corn oil + flaxseed oil and fish oil + flaxseed oil showed reducing levels of glucose by 49.09%, 44.0%, 43.4%, 43.6% and 44.9% respectively. Data revealed significant decrease ($P < 0.05\%$) was observed of TC, TG, LDL and VLDL of all treatment. Urea and creatinine in diabetic rats was increased, in contrast the glutathione reduced was decrease compared to the control group. Fish oil showed the highest decrease in urea and creatinine and the highest increase in glutathione levels and insulin compared to other sources of omega-3. Omega-3 fatty acids have potential effect to protect pancreas from up normality changes which induced in diabetic disease. The data suggested that omega-3 fatty acid and fish oil especially may be effective in the prevention of diabetic disease.

Keywords: Diabetes Mellitus; Omega-3; Fish Oil; Flaxseed Oil; Corn Oil; Insulin; Antioxidant Enzyme

1. Introduction

Diabetes mellitus is considered as a common, growing, serious, costly and potentially preventable public health problem. In 2030, the number of people with diabetes is estimated to increase from 117 million in 2000 to 366 million [1]. The prevalence rate is 8.9% - 12.3% in human population [2]. Diabetes is a disease of carbohydrate metabolism, but it is a general metabolic disorder of all three of the energy nutrients fat, carbohydrate and protein [3]. The most common problem encountered in diabetes mellitus is atherosclerotic cardiovascular disease [4], seen frequently associated with typical dyslipidaemia, including hypertriglyceridaemia. Hypercholesterolemia low levels of high density lipoprotein cholesterol (HDL-c) and high level of low density lipoprotein cholesterol (LDL-c) [5].

On the other hand there is low prevalence of diabetes in Greenland [6] and Alaskan Eskimos [7] as well in people living in the Faeroe Islands [8], populations known for a very high intake of omega-3 fatty acids. The prevalence of diabetes mellitus in Alaska has found that

15.7/1000 subject was lower than over United States vat of 24.7/1000 and 8.8/1000 among Alaskan Eskimos [9]. The lower prevalence of diabetes in this population due to their higher intake of omega-3 rich fish and marine mammals [10]. Average daily consumption of omega-3 among about 3 - 4 g/d compared to with 0.2 g/d by the American population [11]. The prevalence of diabetes in Egypt in 2008 was 4.07%. The global prevalence of diabetic in the year 2010 among adults has been estimated to be 6.4%. It is estimated that by the year 2030, Egypt will have at least 8.6 million adults with diabetes [12].

There are three types of omega-3 in foods. The short chain is alpha linolenic acid ALA: 18:3n-3 (Cis-9,12,5, octadecatrienoic acid), and long chain is eicosapentaenoic acid EPA: 20:5n-3 (Cis-5,8,11,14,17-eicosapentaenoic acid) and docosahexaenoic acid DHA: 22:6n-3 (Cis-4,7,10,13,16,19 docosahexaenoic acid). The major sources of ALA are vegetable oils (such as canola and soybean) with other sources being flaxseed oil. Flaxseed is alternative to plant source of omega-3 fatty acid. It is one of the richest plant sources of omega-3 fatty acid alpha-linolenic (ALA, C18:3n-3) [13]. The bioavailabil-

ity of ALA is greater in oil than in milled seed flaxseed and has greater bioavailability in milled seed than in whole seed [14]. A total intake of ALA 1.5 - 3 g/day appears too beneficial. While fish and fish oil is major sources of EPA and DHA. British National Foundation recommends females and males have an intake of DHA/EPA of 1.1 and 1.4 g/day, respectively [15]. An increased intake of omega-3 fatty acids might reduce the risk of developing diabetes has been tested in mice, where supplementation with fish oil inhibited hyperglycemia and pancreatic insulinitis in streptozotocin-induced diabetes. Also omega-3 fatty acid can reduce insulin resistance in skeletal muscle [16]. ALA is considered to be nutritionally essential because of its specific function as precursor of the long-chain n-3 PUFA [17]. The higher consumption of ALA was associated with higher plasma insulin and improves glucose used and efficiency [18]. EPA and DHA act as hypolipidemics exert prophylactic effects on cardiovascular disease, protect against insulin resistance and obesity in rodent fed high diet and insulin response to glucose in healthy human [19,20]. Polyunsaturated fatty acid (EPA and DHA) play a central role in normal development and functioning of the brain and central system [21]. EPA and DHA also plus a major role in regulating inflammation via substances called prostaglandins. EPA and DHA produced anti-inflammatory prostaglandin.

The relationship of trace elements with coronary artery disease and diabetes may be best examined by comparing population that have larger in dietary consumption of minerals and more pronounced differences in the prevalence of diabetes and coronary artery disease. Zinc is required for normal immune function and taste acuity and enhances the *in vitro* effectiveness of insulin [22].

The aim of the present study was to investigate the effects of fish oil and flaxseed oil and mixed from fish oil and flaxseed oil as the different sources of omega-3 fatty acids on serum glucose, lipid profile (including total cholesterol, HDL-c, LDL-c, Triacylglycerids), kidney function (serum urea and creatinine), antioxidant enzyme (glutathione reduced), insulin and serum zinc in diabetes rats by high fructose diet.

2. Materials and Methods

2.1. Materials

Fish oil (omega-3 plus) was purchased from Sedico Pharmaceutical Company 6-October City-Egypt. Flaxseed oil was purchased from Tanta of oils and flaxseed Company. Corn oil was purchased from local market. All of kits were purchased from Bio-diagnostic 29 Tahreer St., Dokki, Giza, Egypt.

Fatty acid analysis of oils was determined using the method of [23]. Fatty acid was converted to fatty acid

methyl ester according to the method by [24]. Trace GC Mass Ultra thermo was used to separate fatty acids on 10 m capillary Colum (0.1 nm ID × 0.2 μm). The carrier gas used was helium set at a flow rate of 0.5 ml/min and split less. Time 1min. The injection temperature was 250°C and the FID detector was 350°C. Initial oven temperature was 40°C and rose to 230°C and holding for 20 min. Identification of compounds was based on molecular weight of fatty acids.

2.2. Experimental Design

Fifty six male albino rats weighing between 100 - 120 grams were obtained from animal house of Ophthalmic Institute Academic Research Giza, Egypt. The animal housed individually in stainless steel cages under controlled condition at constant temperature (22°C) and lighting 12 h. light-dark cycle) and given free access to food and water at all time. The rats were divided randomly into seven groups—eight rats in each group and fed the following diets for 8 weeks:

Group 1: Rats were fed on standard diet mentioned as normal control (negative control group).

Standard diet was prepared according to [25]. It contained 14% casein, 2% cellulose, 5% salt mixture, 1% vitamin mixture, 0.25% choline, 0.3% DL-methionine, 8% corn oil and remaining percentage was starch.

Group 2: Rats were fed on fructose rich diet 60% as a diabetic control group (positive control group). Fructose rich diet was prepared according to [26]. It contained 600 gram fructose for each 1 kg diet (it is similar to standard diet but containing 60% fructose).

Group 3: Rats were fed on fructose rich diet + 8% fish oil (omega-3 plus).

Group 4: Rats were fed on fructose rich diet + 8% flaxseed oil.

Group 5: Rats were fed on fructose rich diet + 4% corn oil + 4% fish oil.

Group 6: Rats were fed on fructose rich diet + 4% corn + 4% flaxseed oil.

Group 7: Rats were fed on fructose rich diet + 4% fish oil + 4% flaxseed oil.

Corn oil was added to fish oil and flaxseed oil to reduce metallic or fishy flavor and bitter taste of oils.

Each rat was weight at the initial and the end of experimental and food intake also was recorded daily. At the end of experiment (8 weeks), rats were killed after an overnight fast and blood of each rat was collected in two tubes. The first tube was containing sodium fluoride to preserve glucose to determined of glucose. The blood in the second tube was centrifuged at 3000 rpm for 20 minutes to obtain the serum, which is kept at -20°C until analysis.

Serum glucose, serum total cholesterol, serum HDL-c,

serum LDL-c and serum triacylglycerol were determined according to [27-31] respectively. Serum urea and serum creatinine were determined according to [32,33] respectively. Serum glutathione reduced (GSH) and serum zinc were determined by [34,35].

2.3. Histopathological Assessment

At necropsy, the pancreas fixed in 10% buffered formalin until analysis. Tissue slices were routinely processed for paraffin embedding and sections were prepared and stained with hematoxylin and eosin. Histopathological assessment was performed by light microscope, X-400 on all tissues of the control group and treatment.

2.4. Statistical Method

Analysis of the data was of preventative variables in form mean \pm SD buy SPSS windows version 17.0 according to Snedecor and Cochran [36]. A NOVA (analysis of variance) used for comparison of *P* value (probability).

3. Results and Discussion

Data in **Table 1** show fatty acid composition of investigated oils sample. Results showed that corn oil contained high levels of omega-6 fatty acid (56.95%) and low level of short chain omega-3 (0.94%). Corn oil does not contain any long chain PUFA (EPA and DHA). These results are in agreement with Petersen *et al.* [37]. Who reported that omega-6 fatty acid (linoleic acid) was the major fatty acid in corn oil (55.9%).

Flaxseed oil contained low level of SFA (7.38%) and omega-6 (10.77%) while it contained high level of short chain omega-3 fatty acid (ALA: 56.31%). Flaxseed oil does not contain long chain n-3 fatty acid (EPA and DHA). These results are in line with those [38-41].

Fish oil only has high concentration of long chain omega-3 fatty acid 39.20% (23.98% of EPA and 15.22% of DHA). These results are agreement with [42,43] who mentioned that the long chain polyunsaturated fatty acid was found mainly in fish and fish oil. Fish and fish oil are the only major dietary source for human of omega-3 long chain fatty acid [44].

Data in **Table 1** also shows fatty acids analysis of the mixed of corn and fish oils; corn and flaxseed oils; fish and flaxseed oils; mixed of corn and flaxseed oils contained lower level of ALA (28.625%) than in flaxseed oil (56.31%); and higher level of omega-6 (33.86%) than flaxseed oil (10.77%).

Mixed of corn and fish oils contained lower level of short chain omega-3 ALA (0.47%) than corn oil (0.94%), and moderately level (20.07%) of long chain omega-3. Mixed of fish and flaxseed oils contained moderately level of ALA (28.155%) and enrichment in long chain omega-3 (EPA and DHA (19.60%). These results are accordance with data reported by Campioli *et al.*, [44] who reported that use mixed oil (flaxseed and fish oil) enrichment in EPA and DHA is preferable to the one containing of ALA.

Data in **Table 2** demonstrated that there are no significant different ($P \leq 0.05$ %) between seven studies groups in initial body weight (mean ranged between 113.33 ± 2.58^a to 115 ± 5.48^a gram).

Data presented in **Table 2** showed that there are no significant different ($P \leq 0.05\%$) in food intake in diabetic group, diabetic rats + 8% flaxseed oil and diabetic rats + 4% corn + 4% flaxseed oils compared to control group. Meanwhile the lowest value of food intake was observed in group 3 (fed on 8% fish oil), group 5 (fed on 4% corn + 4% fish oils) and group 7 (fed on 4% fish + 4% flaxseed oils). These may be due to decrease appetite of food because fishy flavor or metallic flavor of fish oil.

Table 1. Relative percentage of fatty acid composition in investigated oils.

Type of oil	Total SFA*		Total MUFA**	Omega-6			Omega-3		Total n-3
	Myristic acid	Palmitic acid	Oleic acid	LA 18:2n-6	AA 20:4n-6	ALA 18:3n-3	EPA 20:5n-3	DHA 22:6n-3	
Corn oil	-	15.52	26.59	56.95	-	0.94	-	-	0.94
Flaxseed oil	-	7.38	25.54	10.77	-	56.31	-	-	56.31
Fish oil	11.88	21.73	27.19	-	-	-	23.98	15.22	39.2
Mixture of corn and flaxseed oils	-	11.45	26.065	33.86	-	28.625	-	-	28.625
Mixture of corn and fish oils	5.94	28.625	26.89	28.475	-	0.47	11.99	7.61	20.07
Mixture of fish and flaxseed oils	5.94	14.555	26.365	5.385	-	28.155	11.99	7.61	47.755

**Total saturated fatty acids; *Total monounsaturated fatty acids.

Table 2. Mean value \pm SD initial body weight, body weight gain, food intake and feed efficiency of experimental animal.

Groups	Initial body weight Means \pm SD	Final body weight Means \pm SD	Body weight gain Means \pm SD	Food intake daily]Means \pm SD	Feed efficiency ratio (FER) Means \pm SD
1) Control	115 \pm 5.48 ^a	160.33 \pm 8.68 ^a	45.33 \pm 6.74 ^a	9.97 \pm 0.077 ^a	4.54 \pm 0.60 ^a
2) Diabetic	115 \pm 5.48 ^a	125.67 \pm 5.12 ^c	10.67 \pm 0.52 ^b	10.05 \pm 0.52 ^a	1.06 \pm 0.04 ^b
3) Diabetic fish oil	115 \pm 5.48 ^a	149.79 \pm 24.7 ^{ab}	34.67 \pm 80.02 ^a	8.0 \pm 1.13 ^b	4.65 \pm 2.9 ^a
4) Diabetic + flaxseed oil	115 \pm 5.48 ^a	155 \pm 10.48 ^a	40.0 \pm 8.94 ^a	10.16 \pm 0.52 ^a	3.95 \pm 0.94 ^a
5) Diabetic+ corn oil + 4% fish oil	113.33 \pm 2.58 ^a	152.14 \pm 9.34 ^{ab}	45.33 \pm 6.74 ^a	8.13 \pm 0.43 ^b	5.74 \pm 0.92 ^a
6) Diabetic + corn oil + flaxseed	113.33 \pm 2.58 ^a	150.17 \pm 17.33 ^{ab}	37.83 \pm 15.44 ^a	10.05 \pm 0.40 ^a	4.79 \pm 2.24 ^a
7) Diabetic + fish oil + flaxseed oil	113.33 \pm 2.58 ^a	149.33 \pm 12.8 ^{ab}	36.67 \pm 6.74 ^a	8.09 \pm 0.65 ^b	3.66 \pm 1.22 ^a

Data in a columns with different superscript letters are statistically different ($P \leq 0.05$).

The present study are in a contrary with Sandgruber and Buetter, [45] who reported that the odor profiles of the fish oil were described with attributes fatty, fish like, geranium leaf like, grassy-green, malty, metallic, oily, rancid, rancid-oil like.

There is significant decrease ($P \leq 0.05\%$) in body weight gain in diabetic group compared to control group. Meanwhile there are no significant different in body weight gain and food efficiency ratio between different sources of omega-3.

Data in **Table 3**, illustrated that a significant increase $P \leq 0.05$ in plasma glucose, total cholesterol (TC), total triacylglycerol (TG), low density lipoprotein (LDL) and very low density lipoprotein (VLDL) (238.33 ± 4.50 , 128 ± 8.94 , 209.33 ± 30.15 , 52.0 ± 5.58 and 41.87 ± 6.30 mg/dl respectively of diabetic group compared to control group 152.33 ± 32.62 , 84.33 ± 11.32 , 153.67 ± 20.59 ; 24.30 ± 1.55 and 30.73 ± 4.11 mg/dl respectively. Similar results obtained by Yador *et al.*, [46] and Petersen *et al.*, [37] who reported that the administration of fructose (66% from diet) lead to hyperglycridaemia. After 8 weeks of administration of fish oil, flaxseed oil, corn oil + fish oil, corn oil + flaxseeds oil and flaxseed oil + fish oil reduce level of glucose by 49.09%, 44.0%, 43.4%, 43.6% and 44.9% respectively. From these results it could be noticed that consume mainly monounsaturated fatty acid (corn oil) and short chain polyunsaturated fatty acid (flaxseed oil) and long chain polyunsaturated (fish oil) was beneficial for diabetic disease. The different oil and EPA, DHA in fish oil decrease glucose. Our results approved by Shariati *et al.*, [47] who reported that fish oil diet and corn oil diet had an inhibitory effect on increase plasma glucose in diabetic rat by 46.8% and 40.7%. Also, Djousse *et al.*, [48] found that a higher consumption of ALA was associated with higher plasma insulin and improve glucose use and efficiency.

The present study, exhibited increase in HDL of different source of omega-3. The vast majority of studies conducted of treated with n-3 fatty acids have demon-

strated 5% - 10% increases in HDL cholesterol levels [49]. The mechanisms for this effect are not known. Should further studies confirm this finding, the increase in HDL during treatment with n-3 fatty acids might be another antiatherogenic mechanism of these compound. The mechanisms for the effect of omega-3 fatty acid in increase HDL are not known.

Meanwhile there are decreases in total cholesterol; rate of decrease was 39.06%, 25%, 33.98% and 33.07% and 11.46% respectively of fish oil, flaxseed oil, corn oil + fish oil, corn oil + flaxseed oil and fish oil + flaxseed oil when compared to the diabetic group. Cholesterol lowering effect of flaxseed oil was lower than fish oil. In comparison of cholesterol lowering effect between fish oil (containing EPA, DHA: 39.2%) and flaxseed oil (containing ALA: 56.31%) a significant lower of total cholesterol was observed in fish oil group (39.06%) but flaxseed oil was (25%). These explained by the fact dietary flaxseed has a benefit on serum cholesterol clearance like fish oil, which may be due to increase omega-3 long chain polyunsaturated fatty acid in rats by a conversion of ALA in flaxseed oil to EPA and DHA [50].

The results in **Table 3** also showed that decrease in serum triacylglycerol, LDL and VLDL of all treatments. The triglyceride-lowering effect of omega-3 fatty acids has been mainly ascribed to reduce hepatic synthetic of VLDL. Omega-3 fatty acid suppressed hepatic lipogenesis and reduced circulating TG level [51].

This results approved by Caterina *et al.*, [52] reported that n-3 fatty acids reduced plasma TG. N-3 fatty acid suppressed hepatic lipogenesis and reduced circulating triglyceride levels. The main effect of dietary n-3 fatty acids on plasma lipids and lipoproteins in general is reduction in plasma triglycerides by 20% - 50% in healthy subjects and even more in subjects with hypertriglyceridemia, including diabetic patients. Morise *et al.*, [53] observed that a higher ALA intake decrease cholesterol content in liver, however, increase in the feces. The reduced hepatic cholesterol content was accounted for

higher cholesterol secretion into bile thus leading to a depletion of into intrahepatic pool of cholesterol [54].

Data in **Table 4** illustrated that kidney function of diabetic rats fed on different sources of omega-3. It is noticed that increase in urea (53.50 ± 7.12 mg/dl) and creatinine (1.13 ± 0.05 mg/dl) in diabetic group compared to control group (33.67 ± 9.17 mg/dl and 0.77 ± 0.05 mg/dl). These result are agreement with Niewoehner *et al.*, [22] reported urea were elevated in diabetic subjects. Treatment with fish oil, flaxseed, corn oil + fish oil, corn oil + flaxseed oil and fish oil + flaxseed oil decrease urea and creatinine in diabetic rats. The reduction in urea and creatinine of fish oil was more than other treatments. These result me be due to the containing fish oil on polyunsaturated EPA and DHA.

Data of glutathione reduced in **Table 4** show decrease in of glutathione in diabetic rats (1.39 ± 0.03 mg/dl) compared to control group (2.88 ± 0.05 mg/dl). Treated with different source of omega-3 was increased of glutathione level. Fish oil revealed highest increased in glutathione levels compared to other omega-3 and no significant difference ($P \leq 0.05\%$) between other sources of

omega-3. These results may be due contained fish oil DHA and EPA with high concentration. Similar results were obtained by with Garrel *et al.*, [55] who reported that feeding the adequate ALA diet or enriched EPA and DHA diet increased enzyme antioxidant.

Serum zinc level was significant lower in diabetic rats 166.3 ± 3.14 μ g/dl as compared to control group (362.3 ± 6.71). These results approved by [56,57] reported zinc deficiency occurs in subject and animals diabetic compared to control group. While treated with different sources of omega-3 caused improved in zinc levels of diabetic rats. Fish oil was heights effect in improved zinc levels compared to the other sources of omega-3.

Data of insulin levels are shown in **Table 4**. Plasma insulin levels in diabetic rate were markedly decrease compared to control group. These results approved by Mahmud *et al.*, [57] reported that insulin levels were decrease in diabetic rat. All of oils (omega-3) increased the insulin compared to the diabetic group. Meanwhile, fish oil feeding show the highest increased of insulin levels (105.37 ± 2.44) in the rats compared to the diabetic group. The reason for this is not clear, however, it

Table 3. Effect of omega-3 fatty acid on glucose, TC, TG, LDL and VLDL in diabetic rats.

Group	Serum glucose (mg/dl)	Total cholesterol (mg/dl)	Triacylglycerid (mg/dl)	HDL (mg/dl)	LDL (mg/dl)	VLDL (mg/dl)
1) Control	152.33 ± 32.62^b	84.33 ± 11.32^b	153.67 ± 20.59^b	36.67 ± 4.50^b	24.30 ± 1.55^c	30.73 ± 4.11^b
2) Diabetic	238.33 ± 4.50^a	128.0 ± 8.94^a	209.33 ± 30.15^a	28.0 ± 6.26^c	52.0 ± 5.58^a	41.87 ± 6.30^a
3) Diabetic + 8% fish oil	105.0 ± 11.62^c	78.0 ± 4.73^c	103.33 ± 10.67^c	44.67 ± 3.39^a	38.0 ± 3.09^b	20.67 ± 2.13^c
4) Diabetic + 8% flaxseed oil	117.0 ± 13.42^c	96.0 ± 22.41^b	103.0 ± 6.75^c	41.0 ± 2.37^{ab}	36.0 ± 4.43^{bc}	20.60 ± 1.35^c
5) Diabetic + 4% corn oil + 4% fish oil	103.33 ± 7.22^c	84.50 ± 8.91^b	129.67 ± 26.56^b	41.50 ± 4.59^{ab}	30.0 ± 2.28^d	25.93 ± 5.25^b
6) Diabetic + 4% corn oil + 4% flaxseed	104.0 ± 2.37^c	85.67 ± 16.62^{bc}	138.33 ± 7.17^b	36.67 ± 1.86^b	32.0 ± 6.32^{cd}	27.67 ± 1.43^b
7) Diabetic + 4% fish oil + 4% flaxseed oil	107.67 ± 10.46^c	113.33 ± 12.24^a	147.0 ± 26.87^b	38.0 ± 3.03^b	39.33 ± 3.61^b	29.40 ± 5.37^b

Data in a column with different superscript letters are statistically different ($P \leq 0.05$).

Table 4. Effect of omega-3 fatty acid on kidney function, zinc and insulin in diabetic rats.

Groups	Urea (mg/dl)	Creatinine (mg/dl)	Glutathione (mg/dl)	Zinc (μ g/dl)	Insulin (UI/ml)
1) Control	33.67 ± 9.17^d	0.77 ± 0.05^{dc}	2.88 ± 0.05^{ab}	362.3 ± 6.71^a	108.68 ± 1.92^a
2) Diabetic	53.50 ± 7.12^a	1.13 ± 0.05^a	1.39 ± 0.03^b	166.3 ± 3.14^f	61.27 ± 1.44^e
3) Diabetic + fish oil	37.33 ± 4.93^{cd}	0.73 ± 0.05^e	6.46 ± 9.4^a	299.33 ± 5.82^b	105.37 ± 2.44^b
4) Diabetic + flaxseed oil	40.33 ± 1.37^{bcd}	0.90 ± 0.09^{bc}	2.53 ± 0.07^{ab}	247.67 ± 8.31^c	102.05 ± 2.30^{dc}
5) Diabetic + corn oil + fish oil	42.67 ± 2.58^{bc}	0.83 ± 0.05^d	2.47 ± 0.05^{ab}	242.67 ± 2.87^c	103.67 ± 2.87^{bc}
6) Diabetic + corn oil + flaxseed oil	45.50 ± 3.27^b	0.82 ± 0.10^d	2.29 ± 0.06^{ab}	210.67 ± 2.73^e	103.90 ± 1.46^{bc}
7) Diabetic + fish oil + Flaxseed oil	42.83 ± 6.82^{bc}	0.97 ± 0.05^b	2.50 ± 0.04^d	229.33 ± 3.14^d	100.63 ± 2.76^d

Data in a column with different superscript letters are statistically different ($P \leq 0.05$).

may be related to the upregulation of insulin synthesis (by β cells) and its subsequent transport to the extracellular milieu. This speculation is based on the fact fish oil component, particularly EPA and DHA significantly increase plasma membrane fluidity [58]. Increase insulin levels is due to the stimulatory effects of omega-3, thereby potentiating the existing β cells of the islets langerhan's in diabetic rats [59]. Mori *et al.*, [60] reported that Both EPA and DHA significantly increase fasting insulin by 18% ($P \leq 0.035$) and 27% ($P \leq 0.001$) respectively. Some studies suggested that some of oils may improve useful against insulin resistance and type 2 diabetes mellitus [61].

Histopathological Assessment of Pancreas

The structure of the pancreas of the normal control and diabetic rats are shown in **Figures 1** and **2**. Pancreas of normal control rats showed normal islets, whereas that of diabetic animals showed atrophy of islets langerhen's hyperplasia of B cells of islets of longerhen's. However, pancreas of rats from group 3 to 7 revealed no histopathological changes (**Figures 3-7**). This results agreement with Riccillo *et al.*, [62] and Verma *et al.*, [63] reported the type-2 diabetic induce markedly abnormal change in rat islets. Omega-3 protected the pancreatic β -cells may be by decrease oxidative stress and preserving pancreatic

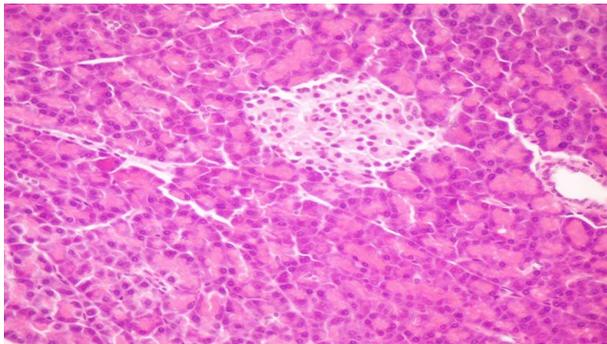


Figure 1. Pancreas of control group 1 showing no histopathological change.

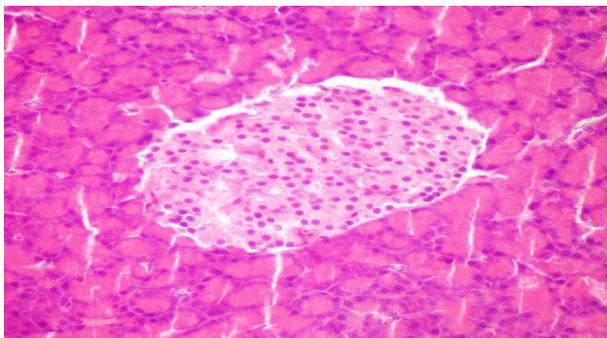


Figure 2. Pancreas of diabetic rat (group 2) showing hyperplasia of B cells of islets of langerhan's.

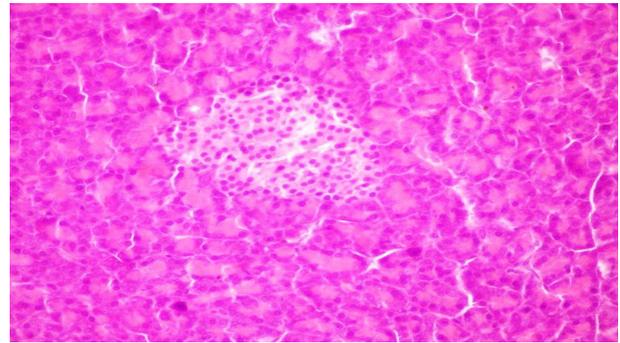


Figure 3. Pancreas of rats from group 3 fed on 8% fish oil showing no histopathological changes.

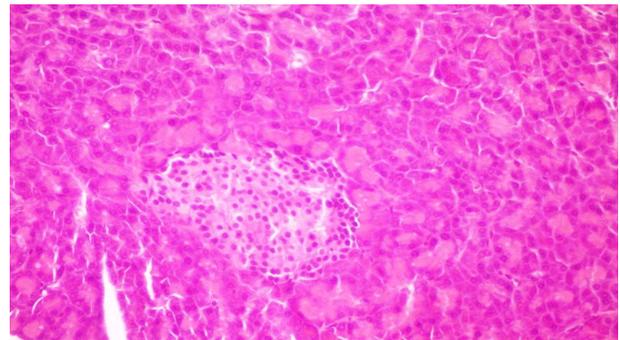


Figure 4. Pancreas of rats from group 4 fed on 8% flaxseed oil showing no histopathological changes.

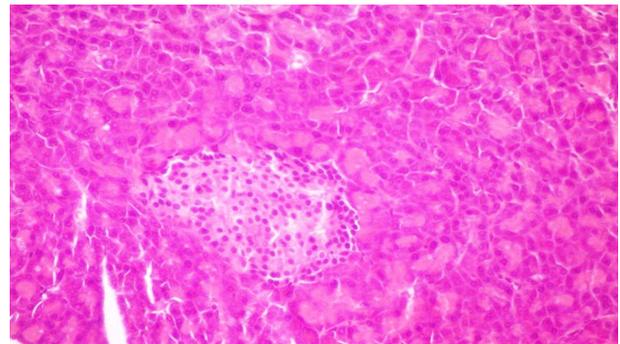


Figure 5. Pancreas of rats from group 5 fed on 4% corn oil + 4% fish oil showing no histopathological changes.

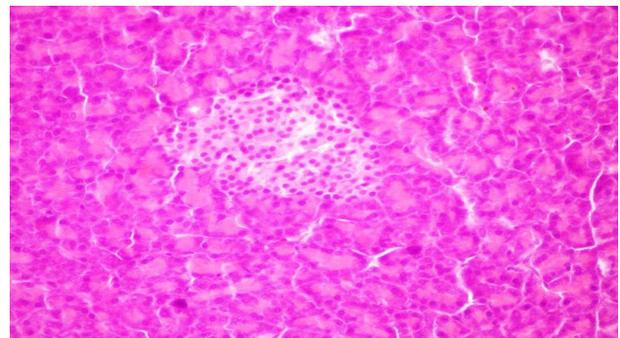


Figure 6. Pancreas of rats from group 6 fed on 4% corn oil + 4% flaxseed oil showing no histopathological changes.

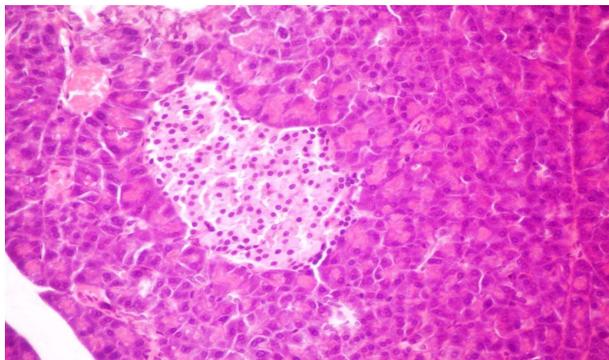


Figure 7. Pancreas of rats from group 7 fed on 4% fish oil + 4% flaxseed oil showing no histopathological changes (X-400).

β -cells integrity [52].

4. Conclusion

In view of these data, it is possible to the omega-3 fatty acid especially Long chain polyunsaturated fatty acid (EPA, DHA) in fish oil have antidiabetic effect. Fish oil were more effective of reduction glucose, TC, TG, LDL, Creatinine and Urea more than other sources of omega-3 fatty acids in diabetic rats. Omega-3 fatty acids have potential effect to protect pancreas from up normality changes which induced in diabetic disease. The data suggested that, omega-3 fatty acid specifically long chain (EPA and DHA in fish oil) may be beneficial in diabetic disease.

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