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Abstract:

The present work was carried out to study the therapeutic effect of cod liver oil, flaxseed oil and zinc for enhancing fertility of rats with injured tests, serum hormonal levels, lipid profile, blood antioxidants parameters and fatty acids in liver of rats injured with cyclophosphamide were determined. In the experiment, thirty five rats were divided randomly into seven groups (n=5). The first group was the negative control fed on basal diet, the other groups were injected with single dose of cyclophosphamide (100 mg\ kg BW) to induce testicular injury, the second group was positive control. Other groups 3,4,5 and 6 treated with cod liver oil, flaxseed oil, mixed oils, zinc (0.5 mg/kg) and mixed oils with zinc. The results revealed that cod liver oil, flaxseed oil, mixed oils, zinc and mixed oils with zinc increased significantly sexual hormones, specially testosterone (T), follicle stimulating hormone level (FSH) and luteinizing hormone(LH). On the other hand, elevated antioxidant enzymes response by increasing significantly total antioxidant and superoxide dismutase (SOD). The results showed that cod liver oil, flaxseed oil, mixed oils, zinc and mixed oils with zinc improved liver function when compared with positive control. The results found that polly unsaturated fatty acids in rats liver treated with cod liver oil, flaxseed oil and zinc had high content of docosahexaenoic and eicosapentaenoic acids which leads to fertility enhancement.

Keywords: Oils, zinc, fertility, rats.

Introduction:

Polyunsaturated fatty acids omega-3, mainly the long chain eicosapentaenoic (EPA) and docosahexaenoic (DHA) fatty acids, have been widely used as adjuvants in therapy. They are mainly found in deep cold water fishes with a high fat content, as well as in the oil extracted of them (Roynette *et al.*, 2004; Berquin *et al.*, 2008).

Omega-3 fatty acids are (PUFAs), human do not have the required metabolic pathways to synthesize the precursor FA (α-linolenic acid). It is a good for the production of the longer bioactive omega-3. Therefore, these long chain polyunsaturated fatty acids intaken either from plant foods or by direct intake of docosahexaenoic and eicosapentaenoic from industrial products or sea foods (Seo *et al.*, 2005). Poly unsaturated fatty acids, FA essential for man as linoleic acid (C18:2n-6; LA) and α-linolenic acid (C18:3n-3; LNA) the precursors of PUFA n-6 and n-3 series (Lands *et al.*, 1992; Zelenka *et al.*, 2008). Animals cannot synthesize n-6 or n-3 fatty acids de novo because of a lack of the appropriate fatty acid desaturase enzymes. The n-6 PUFA and the n-3 PUFA therefore need to be provided in the diet as these PUFAs are essential for numerous processes including growth, reproduction, vision, and brain development (Gurr *et al.*, 2000).

Testosterone is a steroid hormone that originates cholesterol and is the main androgen hormone in men with an important anabolic role. Around 95% of it secreted by Leydig cells located in testicles and 5% by the cortex of the adrenal glands (Kadi *et al.*, 2000). Also testosterone can be administered through injections, adhesives, gels, topically, pills or implants (Margo, 2006; Hsu *et al.*, 2011). Testosterone levels gradually decrease with orderly age and decline of it can cause morbidity and a substantial reduction in the quality of life (Zhao *et al.*, 2013).

Sperm cells contain high ratio of PUFA especially (C22:6 n-3) is a lot of in sperm cells (Rooke *et al.*, 2001). Blood serum phospholipids and in other cell membranes has low content of PUFA (C22:6 n-3) than normal spermatozoa (Lenzi *et al.*, 1996).

A stresses on sexual and reproductive hormone system, including the impact of heat stress and injectable hormones, which decline the amount of testosterone and spermatogenesis (Lue *et al.*, 2000 ; Ghasem *et al.*, 2016).

Intake of an appropriate n-3/n-6 PUFA ratio in the diet of rats increased sperm characteristics and enhanced the structure integrity of testis and sperm, thereby improving reproductive performance, which may be related to changes in hormone metabolism (Yan *et al.*, 2013).

High proportions of long chain polyunsaturated fatty acids in the phospholipid fraction of spermatozoa. High PUFA proportion of the rabbit sperm is necessity antioxidant order to maintain specific membrane properties (fluidity, flexibility, etc) (Mourvaki *et al.*, 2010). It is known that the fatty acid composition of sperm membranes, especially their unsaturated components, determine their biophysical characteristics such as fluidity and flexibility as appropriate for their specific functions, including sperm motility and fertilizing capacity (Khatibjoo *et al.*, 2011).

Increase n-3/n-6 PUFA ratio, sperm density and motility were increased, and the sperm deformity rate tended to decrease (Amin *et al.*, 2011).

The effects of cod liver oil on serum lipids, cholesterol, lipoproteins, atherogenesis and thrombogenesis (Saynor *et al.*, 1984 & Ross, 1986). Cod liver oil influences lipid metabolism in several mothed that may be of value in the treatment and prevention of cardiovascular disease and inflammatory disorders (Simopoulos, 1989).

Flaxseed (*Linum usitatissiumum*), also known as linseed, flaxseed oil contains high levels of α-linolenic acid (αLA) omega-3 and may be utilized as a feedstuff rich in energy essential long chain FA is a precursor for the production of eicosapentaenoic acid (EPA), a precursor to the production of docosahexaenoic acid (DPA). Health benefits are associated with diets containing αLA, 9. This essential fatty acid is linoleic acid (LA), a precursor for arachidonic acid (AA) known to produce pro-inflammatory agents (Greiner *et al.*, 1997).

Flaxseed oil is a good source of omega 3 fatty acids (linolenic acid). (Singh *et al.*, 2011).

Zinc is a high concentration in male sex organs like prostate, testicles and in the spermatozoa, its important role in reproduction is undeniable (Oliveira *et al.*, 2004).

Zinc and organs involved in anatomical development and normal function of male reproductive, enhances spermatogenesis by actively participating in the maturation of spermatozoa and the preservation of germinative epithelia. Therefore, low zinc intake levels might incur delay of testicle development and cessation of spermatogenesis (Oliveira *et al.*, 2004 ; Yunsang and Wanxi 2011).

Cyclophosphamide is medication for treatment tumor it has many side effects, including reproductive toxicity (Howell, and Shalet 1998). Studies have shown that cyclophosphamide reduces luteinizing hormones, FSH, testosterone and spermatogenesis (Johari *et al.*, 2011).

We conclude that cod liver oil, flaxseed oil and zinc may protect male reproductive tract from toxic effects of cyclophosphamide and has beneficial effects on serum hormonal levels, lipid profile, some blood antioxidants parameters and total lipids in liver of rats.

Materials and Methods:

Materials:

Oils: cod liver oil and flaxseed oil were purchased from Bader Company, Cairo, Egypt.

Cyclophosphamide: (Medica AG Pharmaceutical, Frankfurt, Germany). Zinc sulfate: (Merc Pharmaceutical, Egypt) .

Animals: Thirty five of healthy adult male albino rats, that initially weighed approximately (115+5g) were used in this study. The animals were obtained from National Research Center, Cairo, Egypt. The animals were allocated in plastic cages. Rats had fed on basal diet for seven days before beginning the experiment. (Table1) according to (NRC, 1995 and Reeves *et al.*, 1993). The vitamin and mineral mixture had the prepared according to (Campbell, 1963).

Table (1): Composition of the basal diet.

Ingredients	g/kg diet
Casein	200
Corn starch	497
Sucrose	100
Vitamin mixture	020
Mineral mixture	100
Corn oil	050

Cellulose	030
Methionine	003

Induction of testicular:

Rats were injected (i.p) with cyclophosphamide (100mg/kg BW) induce testicular injury (**Zhang and Yang, 2006**).

Methods:

Experimental design:

Thirty five adult male albino rats were divided randomly into seven groups (n=5). Group (1) Negative control (ve-) rats fed on basal diet only. other groups injected with a single dose of cyclophosphamide (100 mg\ kg BW) to induce testicular injury,

Group (2) Positive control (ve+).

Group (3) received cod liver oil (0.5 mg/kg)

Group (4) received flaxseed oil (0.5 mg/kg)

Group (5) received mixed oils (0.5 mg/kg)

Group (6) received zinc sulfate (0.5 mg/kg) **Dissanayake *et al.* (2009**)

Group (7) received mixed oils with zinc (0.5 mg/kg).

At the end of experiment 4 weeks animals were deprived of food overnight and sacrificed by decapitation. Blood was collected in tubes, without anticoagulant for serum separation.

Body weight are measured three times every week during four weeks. Daily changes in body weight as percentages were recorded, body weight percentage was determined as the following:

$$\text{Change in body weights (\%)} = \frac{\text{Final weight} - \text{Initial weight} \times 100}{\text{Initial weight}}$$

At the end of experiment food efficiency ratio was calculated as following:

$$\text{FER} = \frac{\text{Body weight gain (gm)}}{\text{Food intake (gm)}}$$

Serum hormonal analysis:

Testosterone (T), follicle stimulating hormone (FSH) and luteinizing hormone (LH) were analyzed using the method of **Maruyama (1987**).

Determination of some antioxidant parameters:

Testis was homogenized for biochemical analysis of oxidant and antioxidant enzymes including superoxide dismutase (SOD) which determined according to (Dechatelet *et al.*, 1974). Determination of malondialdehyde (MDA) by the method described by Stocks and Donnandy (1971). Total Antioxidants (TA) was determined according to Beutler (1984).

Biochemical analysis:

Uric acid, Urea and Creatinine were determined in the serum according to Patton & Crouch, 1977; Han *et al.*, 1984 and Bartels *et al.*, 1972 respectively. Aspartate amino transferase (AST), Alanine amino transferase (ALT) and Alkaline phosphatase (ALP) were determined by the method described by (Reitman and Frankel, 1957 and Kind and King, 1954) respectively. Triglycerides, serum total cholesterol, high density lipoprotein and low density lipoprotein were assayed by Roeschlau *et al.* (1974) and Fossati and Prencipel (1982), respectively.

Determination of fatty acids pattern in liver:

Determination of fatty acids pattern in total lipids of liver were analyzed by using Gas Liquid Chromatography (GLC) as described by Vogel (1975).

Statistical analysis:

Statistical analysis for collected data was done following the procedure outline by Gomez and Gomez (1984). The treatment means were compared using the least significant difference test (LSD) at 5% level of probability as outline by Waller and Duncan (1969).

Results and Discussion:

Effects of cod liver oil, flaxseed oil, zinc and their mixture on BWG , FI and FER of experimental rats

Data in Table (2) showed that the body weight gain (BWG) of all rat groups treated with cod liver oil, flaxseed oil, zinc ,mixed oil and mixed oil with zinc increased significantly at the end the experiment when compared with positive control.

Among the treated groups, the highest weight gain was noticed in the group received mixed oils with zinc, cod liver oil and zinc group which reached 66.8, 61.2 and 55.00 respectively. On the other hand, data in Table (2) showed that food efficiency ratio was increased significantly in all group treated when compared with positive control .

Nederrgaard *et al.* (1983), Alling *et al.* (1974), Privett and Cortesi (1972) and Mak *et al.* (1983), showed that reduced weight gain and general deterioration of conditions in rats fed excess in the form of highly unsaturated plant oils.

These results are accordance with that of Estienne *et al.* (2008), they found that weight gain tended to increase as the ratio of n-3/n-6 PUFAs increased.

Table (2): Effects of cod liver oil, flaxseed oil, zinc and their mixture on BWG, FI and FER of experimental rats.

Groups	BWG (g)	FI (g/d)	FER
Negative control	73.6±2.11[a]	15.78± 2.20[a]	0. 078±0.003[a]
Positive control	34.4±8.11[d]	14.24± 0.20[c]	0. 040±0.005[c]
cod liver oil	61.2± 6.11[b]	15.22± 0.23[ab]	0.067±0.005[b]
flaxseed oil	54.6±9.13[c]	15.19±2.32[ab]	0.060±0.002[b]
Mixed oils	52.8±2.17[c]	14.72±0.21[c]	0.060±0.003[b]
Zinc	55.00±9.17[bc]	14.88±0.92[bc]	0.061±0.003[b]
Mixed oils with zinc	66.8±4.9[ab]	14.88±0.01[bc]	0.04±0.005[b]

Mean values in each column having different superscript are significant $p < 0.05$.

Effects of cod liver oil, flaxseed oil, zinc and their mixture on sexual hormones levels of experimental rats.

Data in Table (3) revealed a significant elevation in sexual hormones specially testosterone (T), follicle stimulating hormone level (FSH) and luteinizing hormone(LH) in rat groups treated with cod liver oil, flaxseed oil, mixture oils, zinc and their combination when compared with positive control.

Concerning testosterone (T), data presented in Table (3) revealed that an increase significantly was observed in testosterone (T) levels, in rats treated with mixed oils with zinc then zinc group, mixed oil, cod liver oil and flaxseed oil which their values were 25.81, 22.5, 21.01, 20.12 and 19.33 mg/mL when compared with positive control which was 16.03 mg/mL.

As evident from Table (3), data showed an increase significantly in follicle stimulating hormone level (FSH) presented in rats groups treated with mixed oils with zinc followed by zinc group then cod liver oil their values were 130.32, 125.59 and 118.98 mg/mL respectively when compared with positive control which was 90.17mg/mL.

On the other hand, the results in Table (3) showed that significantly increase in luteinizing hormone level (LH) which was noticed in rats receiving mixed oil with zinc followed by zinc group followed by cod liver oil values were 4.00, 3.8 and 2.92 respectively, compared to positive control which recorded1.8 mg/ml.

The results suggested that mixed oils with zinc, cod liver oil and zinc improvement sexual hormones, specially testosterone (T), follicle stimulating hormone level (FSH) and luteinizing hormone (LH). These results are accordance with that of (Hartoma *et al*., 1977 and Tikkiwal *et al*., 1987), who stated that oral zinc supplementation improves sperm concentration.

Cerolini *et al*., 2000; Safarinejad & Safarinejad, 2012, showed that the degree of lipid composition, specially omega-3 and omega-6 are indicator to affect sperm quantity.

These results are accordance with that of Martin *et al.* (1994). They recorded that the concentrations FSH, LH, and T, increased by increasing the ratio of omega-3/omega-6 Polly unsaturated fatty acids. LH stimulates the interstitial cells located in the testes to produce testosterone, and FSH plays a good role in spermatogenesis.

These results are accordance with that of (Demark *et al.* 2008). They found that flaxseed oil improvement of prostate diseases.

According to Ashok and Sekhon (2011) semen zinc level had a significant effect over sperm morphology. Zanaty *et al*. (2004) showed a direct relationship between viscosity of semen and semen zinc level.

Weak sperm mobility and production was related to lower level of zinc in semen plasma in infertile males (Zhou *et al.*, 2015).

Increasing the ratio omega-3 /omega-6 polly unsaturated fatty acids, due to increased sperm quality and motility, (Amin *et al.*, 2011). Excessive n-3 PUFA supplementation decreased the sperm density and motility in the experiment, which indicated the importance of the n-6/n-3 PUFA ratio in sperm quality (Al-Daraji *et al.*, 2010).

These results are accordance with that of (Veldhuis *et al.*, 2009),who founded that by increasing the ratio of n-3/n-6 PUFAs, the concentrations of GnRH, FSH, LH, and T increased.

The results suggested that intake of cod liver oils specially mixed oils are a necessary for enhancing fertility of rats.

Table (3): Effects of cod liver oil, flaxseed oil, zinc and their mixture on fertility hormones level of experimental rats.

Hormones level	**T ng/mL**	**FSH ng/mL**	**LH ng/mL**
Negative control	29.2±1.7 [a]	150.38±6.75[a]	5.9±0.76[a]
Positive control	16.03±2.01[d]	90.17±7.55[d]	1.8±0.16[d]
cod liver oil	20.12±3.01[b]	118.98±0.65[c]	2.92±0.6[c]
flaxseed oil	19.33±14.81[c]	114.42±0.80[c]	2.54±0.5[c]
Mixed oils	21.01±14.10[b]	116.60±0.09[c]	2.47±0.37[c]
Zinc	22.5±1.3[b]	125.59±0.80[b]	3.8±0.6[b]
Mixed oils with zinc	25.81± 19.64[ab]	130.32 ± 19.64[b]	4.00±0.4[b]

Mean values in each column having different superscript are significant $p < 0.05$

T: testosterone. FSH: Follicle Stimulating Hormone level LH: Luteinizing hormone

Effects of cod liver oil, flaxseed oil, zinc and their mixture on antioxidant parameters of experimental rats.

Data in Table (4) revealed a significant elevation in total antioxidants (TA), superoxide dismutase (SOD), and a decrease significantly in malondialdehyde (MDA) were observed in rats groups treated with cod liver oil, flaxseed oil, zinc and their combination oil when compared with positive control.

Concerning total antioxidants (TA), data presents in Table (4) revealed that an increase significantly on total antioxidants (TA) observed in rats treated with mixed oils with zinc, followed by mixed oils, zinc, cod liver oil and flaxseed oil values were 4.02, 3.94, 3.69, 3.32, and 2.58 mmol/l respectively in comparing with positive control was 1.67 mmol/l.

Data presented in Table (4) revealed an increase significantly in superoxide dismutase (SOD).

As evident from Table (4), showed that a significant decrease in malondialdehyde (MDA) which observed in rats treated with mixed oils with zinc was 9.21, mixed oils was 10.11, flaxseed oil was11.37 and cod liver oil was 12.02 mmol/L when compared with positive control was 18.35 mmol/L.

This increase in sperm population, motility and viability of experimental group in comparison to control group cause polyunsaturated fatty acids, these productive effects are reflected may be by the decrease of malonaldehyde level and increase in total anti-oxidants capacity. Antioxidants can interfere with the oxidation process by reacting with free radicals, chelating catalytic metals, acting as oxygen scavengers (Jan *et al*., 2010) .

Table (4): Effects of cod liver oil, flaxseed oil, zinc and their mixed oils on antioxidant parameters of experimental rats

Parameters / Groups	Total Antioxidants (mmol/l)	SOD (U/Ml)	MDA (nmol/L)
Negative control	4.55±0.22[a]	54.55±4.1[a]	6.47±0.76[d]
Positive control	1.67±0.15[c]	22.33±2.01[c]	18.35±1.16[a]
cod liver oil	3.32±0.15[b]	45.60±3.04[b]	12.02±0.96[b]
flaxseed oil	2.58±0.80[b]	40.62±0.06[b]	11.37±0.52[c]
Mixed oils	3.94±0.06[ab]	48.90±1.10[ab]	10.11±0.37[c]
Zinc	3.69±1.80[ab]	46.33±0.08[ab]	13.21±0.96[b]

Mixed oils with zinc	4.02 ± 0.06^{ab}	49.90 ± 1.10^{a}	9.21 ± 0.36^{c}

Mean values in each column having different superscript are significant p < SOD=Superoxide dismutase.

Effects of cod liver oil, flaxseed oil, zinc and their mixture on lipid profile of rats.

An increase significantly in serum triglyceride, total cholesterol and LDL-cholesterol levels were observed in positive control than rats treated with mixed oils with zinc , mixed oils, zinc, flaxseed oil and cod liver oil, the results recorded that in the Table (5) a significant decrease of serum total cholesterol levels in rats groups receiving mixed oil with zinc was (89.00 ± 6.5 mg/dl), followed by mixed oil was (96.68 ± 4. 28 mg/dl) than those of positive control group was (158.19 ± 27.12 mg/dl).

Concerning triglyceride, data presents in Table (5) revealed that the groups treated with mixed oil was (84.8 ± 8.08 mg/dl), cod liver oil was (88.27 ± 13.03 mg/dl) and flaxseed oil was (89.9 ± 24.34 mg/dl) showing significant decrease in triglyceride levels comparing with positive control which recorded (190.80 ± 14.82 mg/dl).

Results in Table (5) indicated that all the test groups receiving cod liver oil, flaxseed oil and mixed oils revealed significant decreases in LDL-cholesterol levels in comparing with the positive control, the highest reduction was mixed oils with zinc which recorded (35.86 ± 4.9 mg/dl) followed by zinc which recorded (43.78 ± 0. 37 mg/dl) than positive control which recorded (91.4 ± 23.43 mg/dl).

On the other hand, the results in Table(5) showed that increase significantly in HDL-cholesterol was noticed in rats treated with zinc which reached (37.97 ± 2.75 mg/dl), followed by cod liver oil which reached (36.4 ± 2.66 mg/dl) than positive control which reached (28.58 ± 2.54 mg/dl).

It is advised by giving cod liver oil, flaxseed oil and mixed oils to the patients with hyperlipidemia or those exposed to atherosclerosis.

Results in Table (5) indicated that all the test groups receiving cod liver oil, flaxseed oil, zinc, mixed oils and mixed oils with zinc showed decrease in atherogenic indexes than those of positive control group.

Flaxseed oil used in treatment of respiratory and digestive system diseases. Also, flaxseed added to bakery products enhance their taste and dietetic qualities. On the other hand flaxseed oil is very rich in polyunsaturated fatty acids (Hunter, 1990; Fataneh *et al*., 2012). These results are accordance with that of Cunnane *et al*. (1994); Hu *et al*. (1999); Heller *et al*. (2010); Ganorkar, Jain (2013), who recorded that flaxseed oil contained Alpha-linolenic acid (ALA) which reduces the cholesterol level and improves cellular lipid metabolism.

These results are accordance with that of Fukumitsu *et al*. (2008), who found that flaxseeds has an effect on lipid profiles in mice, it decreases hypercholesterolemia and hyperlipidemia.

These results are accordance with that of Giada (2010) who found that flaxseed used in treatment many diseases such as atherosclerosis, diabetes mellitus and cancer. These results are accordance with that of Zhao *et al*. (2007) who recorded that flaxseeds has treatment cardiovascular diseases and which improve the lipid profile. Due to the components presence of flaxseed it has physiologically beneficial effects on one's health, important part of a balanced nutrition, bioactive foods and functional foods. (Brzezinsk and Debi, 1999; Hasler *et al*., 2000; Thompson, 1993).

These results are accordance with that of Bhathena *et al*. (2002), who recorded that the consumption of diets with flax oil reduced serum cholesterol and triglycerides in rats with normal lipid profiles.

Table (5): Effects of cod liver oil, flaxseed oil, zinc and their mixed oils on lipid parameters of experimental rats.

Groups	TC mg/dl	TG mg/dl	HDL-c mg/dl	LDL-c mg/dl	VLDLc
Negative control	88.21±14.33[bc]	78.6±9.62[d]	38.6±3.11[a]	33.9±13.43[c]	15.7±1.9[d]
Positive control	158.19±27.12[a]	190.80±14.82[a]	28.58±2.54[c]	91.4±23.43[a]	38.16±0.7[a]
cod liver oil	105.6±16.54[b]	88.27±13.03[cd]	36.4±2.66[a]	51.6±11.37[b]	17.6±2.8[cd]

flaxseed oil	117.14±27.6 5^{maleb}	89.9±24.34 ^{cd}	33.60±1.9 3^{b}	65.58 ±12.36^{b}	17.96±0. 5^{cd}
Mixed oils	96.68±4. 28^{bc}	84.8±8.08^{cd}	32.02±2.7 6^{b}	47.7 ±1.76^{b}	16.96±5. 7^{cd}
Zinc	103.27±4. 54^{b}	107.6±5.25 ^{b}	37.97±2.7 5^{a}	43.78±0. 37^{b}	21.52±7. 0^{b}
Mixed oils with zinc	89.00±6.5^{b}	90.7±3.456 ^{cd}	35.00±4.1 2^{ab}	35.86±4.9^{c}	18.14±4. 2^{cd}

Mean values in each column having different superscript are significant $p < 0.05$.

Effects of cod liver oil, flaxseed oil, zinc and their mixture on liver enzymes of experimental rats.

Table (6) showed significant decrease in AST, ALT, ALP in groups treated with cod liver oil, flaxseed oil, zinc, mixed oil with zinc and mixed oil when compared with positive control.

Data showed a significant decrease in AST in groups treated with mixed oils reached to 103.82 (Iµ/L) followed by cod liver oil, mixed oils with zinc and flaxseed oil values were 116.86,118.08 and 125.08 Iµ/L, respectively when compared with positive control 191.57.

Therefore, it is possible to suggest that these oil specially mixed oils with zinc and mixed oils are safe and might improve liver function.

Effect of polyunsaturated omega-3 fatty acids (PUFA) found in marine cod liver oil, especially eicosapentaenoic acid (20:5 3; EPA) and docosahexaenoic acid (22:6 3; DHA), proved their desirable. Also, they are extremely prone to anti oxidation. Which are biologically very active and which can promote many effects on the cell function and viability (Esterbauer, 1985; Benedetti & Comporti, 1987). It is also possible to suggest that cod liver oil, flaxseed oil and zinc might directly improve the liver function specially mixed oils with zinc.

Table (6): Effects of cod liver oil, flaxseed oil, zinc and their mixed oils on liver function of experimental rats.

Groups	AST (Iμ/L)	ALT (Iμ/L)	ALP (Iμ/L)
Negative control	80.80±2.11[e]	60. 21±1.78[b]	93. 26±3.97[d]
Positive control	191.57±5.58[a]	144.86±3. 27[a]	164.39±6.49[a]
cod liver oil	116.86±4.75[cd]	74.76±3. 13[c]	119.41±3.65[bc]
flaxseed oil	125.08±4.28[c]	72. 03±3.43[c]	124.74±5.649[b]
Mixed oils	103.82±3.02[d]	80. 75±2.14[d]	128.89±12.622[b]
Zinc	141.20±1.48[b]	65. 83±3.43[b]	129.41±2.07[bf]
Mixed oils with zinc	118.08±4.28[cd]	52.40±9.17[bc]	114.74±5.649[bc]

Mean values in each column having different superscript are significant $p < 0.05$.

Means with the same letter are insignificantly different.

Effects of cod liver oil, flaxseed oil, zinc and their mixture on uric acid, urea and creatinine levels of experimental rats.

The results showed that significant decreases in uric acid, creatinine and urea this observed in rats treated with cod liver oil, flaxseed oil, zinc, mixed oils and mixed oils with zinc when compared with positive control. Concerning uric acid data present in Table (7) showed a significant decrease in rats treated with mixed oils with zinc had 1.5 mg/dl, followed by zinc group, mixed oils, cod liver oil values were 1.65, 1.72 and 1.79 mg/dl, when compared with positive control 3.20mg/dl.

Concerning creatinine level, data presented in Table (7) revealed significant decrease in rats receiving mixed oils with zinc , zinc, mixed oils, flaxseed oil and cod liver oil compared to positive control. The results showed that cod liver oil, flaxseed oil and zinc were the best effective in kidney function specially mixed oils with zinc.

Data in Table (7) revealed a significant decrease in urea levels of rats groups treated with mixed oils with zinc which reached 13.1mg/dl, followed by mixed oils which reached 16.21mg/dl, when compared to positive control which reached 39.93mg/dl. These results of treatment effects of cod liver oil, flaxseed oil, zinc, mixed oils with zinc and mixed oils on some renal function represented in creatinine, uric acid, urea. It is also possible to suggest that these mixed oils with zinc might directly improve the structural and function alintegrities of cells of the blood, kidney.

Table (7): Effects of cod liver oil, flaxseed oil, zinc and their mixture on uric acid, urea and creatinine of experimental rats.

Groups	Uric acid mg/dl	Creatinine mg/dl	Urea mg/dl
Negative control	1.53± 0.78[e]	0.8± 0.018[e]	13.88±4.72[e]
Positive control	3.20±0.52[a]	2.62±0.47[a]	39.93±3.20[a]
cod liver oil	1.79±0.72[de]	1.87±0.32[b]	24.14±3. 29[c]
Flaxseed oil	1.82±0.57[d]	1.83±0.25[b]	30.17±3.59[b]
Mixed oils	1.72±0.93[de]	1.72±0.15[b]	16.21±3.43[c]
Zinc	1.65±0.14[de]	1.65±0.13[b]	29.90±2.38[b]
Mixed oils with zinc	1.5±0.3[de]	0.7±0.02[e]	13.1±1.54[c]

Mea n values in each column having different superscript are significant $p < 0.05$.

Fatty acids in liver total lipids of rats treated with cod liver oil, flaxseed oil, zinc and their mixture.

The main fatty acids identified in liver of rats treated with cod liver oil, flaxseed oil, zinc. Mixed oils and mixed oils with zinc, are presented in Table (8). The results showed that fatty acids in liver of rats group treated with cod liver oil have a higher content of docosahexaenoic acid which reached 3.1 followed by rats treated with zinc which reached 2.9 followed by rats treated with flaxseed oil which reached 2.8 then rats treated with mixed oils with zinc which reached to 2.78. Also, fatty acids in liver of rats treated with cod liver oil have a higher content of eicosapentaenoic acid which reached 2.45 followed by rats treated with flaxseed oil which reached 2.39 then rats treated with zinc which reached 2.35.

On the other hand, arachidonic acid was a higher content in liver of rats negative control which reached 2.2, followed by rats treated with mixed oils with zinc which reached 2.15 then rats treated with zinc which reached 2.05. The results showed that, fatty acids in liver of rats treated with mixed oil with zinc have higher content of linolenic acid n-3, followed by rats treated with mixed oils then rats treated with cod liver oil. On the other hand, fatty acids in liver rats treated with zinc have higher content of n-6 PUFA linoleic acid

The results suggest that Polly unsaturated fatty acids in liver of rats treated with cod liver oil, flaxseed oil and zinc have higher content of docosahexaenoic and eicosapentaenoic, because chemical composition of cod liver oil, flaxseed oil shows high content of Polly unsaturated fatty acid specially docosahexaenoic and eicosapentaenoic, leads to fertility enhancement.

Flax has a regulating effect on liver metabolism. Verified that the supplementation of flax oil not only diminishes serum cholesterol levels but also increases the proportion of -linolenic acid (C18:3 -3) in the liver of rats with a dose-dependent response (Melo *et al*., 2012).

Table (8): Fatty acids in total lipids in liver of rats treated with cod liver oil, flaxseed oil, zinc and their mixed oil.

	Negative control	Positive control	cod liver oil	flaxseed oil	Mixed oils	Zinc	Mixed oils+ zinc
S. Fatty Acids Undecanoic acid	0.18±0.02	0.35±0.3	0.11±0.01	0.18±0.01	0.11±0.007	0.12±0.04	0.08±0.01
Lauric acid	0.14±0.22	0.07±0.008	0.03±0.005	0.17±0.02	0.04±0.005	0.05±0.005	0.03±0.004
Tridecanoic acid	0.18±0.02	0.18±0.03	0.14±0.01	0.21±0.03	0.10±0.007	0.11±0.02	0.09±0.006
Myristic acid	0.19±0.02	0.28±0.06	0.6±0.08	0.33±0.01	0.19±0.007	0.4±0.08	0.19±0.02
Pentadecanoic acid	6.9±0.5	8.98±0.07	6.5±0.6	7.8±0.7	6.2±0.5	6.5±0.4	6.8±0.62
Palmitic acid	24.0±2.4	26.9±2.3	21.0±1.2	24.2±2.1	22.3±2.5	23.0±1.2	22.1±1.7

Stearic acid	4.1±0.45	6.4±0.7	3.1±0.15	4.8±0.8	2.9±0.19	3.9±0.15	4.4±0.21
MUFA Myristoleic acid	0.8±0.135	1.5±0.14	1.19±0.029	1.1±0.006	1.65±0.13	1.19±0.029	1.62±0.167
Pentadecenoic acid	1.1±0.07	0.65±0.05	0.9±0.18	1.02±0.009	1.7±0.1	0.9±0.18	0.84±0.08
Oleic acid	27.3±2.1	33.9±1.8	19.1±0.9	23.5±1.8	21.6±3.2	25.1±0.9	23.8±1.04
n-6 PUFA Linoleic acid	20.2±0.7	13.5±0.7	18.6±0.87	17.5±1.05	19.4±0.41	20.6±0.87	19.2±0.59
Eicosadienoic acid	3.2±0.21	5.78±0.43	2.75±0.16	3.1±0.3	2.82±0.07	2.65±0.16	2.695±0.05
Arachidonic acid	2.2±0.02	0.42±0.02	1.05±0.014	1.9±0.01	1.04±0.031	2.05±0.014	2.15±0.021
Docosapentaenoic acid	0.11±0.07	1.05±0.004	-	0.106±0.07	-	-	-
n-3 PUFAs Linolenic acid	0.29±0.014	0.19±0.09	0.92±0.08	0.12±0.01	1.5±0.12	0.92±0.08	1.9±0.18
Eicosapentaenoic acid	0.54±0.041	0.41±0.03	2.45±0.32	2.39±0.003	2.2±0.1	2.35±0.32	1.9±0.15
Docosahexaenoic acid	1.0±0.08	0.52±0.05	3.1±0.1	2.8±0.06	2.07±0.04	2.9±0.1	2.78±0.1

Conclusion:

Intake of flaxseed oil, cod liver oil and zinc and mixed oils are a necessary for enhancing fertility of rats. However, further works are needed to better understand the mechanisms to take advantage of flaxseed oil, cod liver oil in food preparation and food industry.

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التأثير العلاجي لزيت كبد الحوت وزيت بذره الكتان والزنك في تعزيز الخصوبة فى الفئران المصابة في الخصية

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الملخص :

أجريت الدراسة بهدف اكتشاف التأثير العلاجي لزيت كبد الحوت وزيت بذره الكتان وخليطهما والزنك لتعزيز الخصوبة في الفئران التي تم إصابتها في الخصية بتأثير سيكلوفسفاميد, والواضح أثره في بعض هرمونات الدم، ودهون الدم، وبعض مضادة للأكسدة في الدم وأيضا الدهون الكلية في الكبد. في التجربة البيولوجية، تم تقسيم خمسه وثلاثون فأرا عشوائيا إلى سبع مجموعات متساوية، عدد كل مجموعه ٥ فئران. المجموعة الأولى (الكنترول السالب)، ثم تم حقن المجاميع الباقيه بسيكلوفوسفاميد (١٠٠ ملغ/كجم من وزن الجسم) لإصابة الخصية، المجموعة الثانية (الكنترول الموجب) وتم حقنها بسيكلوفوسفاميد فقط، باقي المجاميع حقنت بسيكلوفوسفاميد وتم علاجها بزيت كبد الحوت، زيت بذره الكتان، خليط من زيت كبد الحوت وزيت بذره الكتان، الزنك، خليط من زيت كبد الحوت وزيت بذره الكتان والزنك (٠.٥ ملغ/كلغ). وكشفت النتائج أن الزيوت المختلطة مع الزنك، زيت كبد الجوت، الزنك، زيت بذرة الكتان، حققت ارتفاع في مستوي الهرمونات الجنسية لدى الذكور، وخاصة هرمون تستوستيرون (T)، وهرمون (FSH) وهرمون (LH). أشارت النتائج ان زيت كبد الحوت، زيت بذور الكتان والزنك ابرز فعالية كبيره في رفع استجابة الانزيمات المضادة للأكسدة من خلال زيادة كبيرة في مستوي (SOD)، وكانت أفضل النتائج للزيوت المختلطة مع الزنك والزيوت المختلطة. وأظهرت النتائج أن زيت كبد الحوت، زيت بذور الكتان، الزيوت المختلطة، الزنك، الزيوت مختلطة مع الزنك كان له افضل تأثير علي وظائف الكبد مقارنة بالكنترول الموجب. وأشارت النتائج أن الأحماض الدهنية العديدة الغير مشبعة في دهون كبد الفئران التي تم علاجها بزيت كبد الحوت، زيت بذور الكتان، الزنك اظهرت ارتفاع في محتوى الدوكوساهكسانويك وايكوسابنتانويك، ويرجع ذلك لان التركيب الكيميائي للزيت كبد الحوت، زيت بذور الكتان يحتوي علي نسبة عالية من الاحماض الدهنية العديدة الغير مشبعة خصيصا الدوكوساهيكسانويك وايكوسابنتانويك، مما يؤدي الي تعزيز

الخصوبة. وأوصت الدراسة أن تناول زيت بذور الكتان، زيت كبد الحوت والزنك لهم دور كبير في تعزيز الخصوبة لدي الذكور. وعليه توصى الدراسة الي مزيد من الأعمال البحثيه من أجل فهم أفضل الاليات للاستفادة من تلك الزيوت في مجال اعداد الأطعمة والتصنيع الغذائي.