

# Comparison of the Nutrition on Different Fats on Lipid Profile, and Some Biochemical Parameters and their Relationship with Liver and Kidney Function in Male Rats

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## Abstract

This study was conducted on forty-nine healthy male albino males rats, from 1/11/2019 to 12/30/2019 for two months in the animal house of the College of Veterinary Medicine - University of Tikrit, their ages ranged from 3-4 months and weights of 250-350 g current study aimed to determine the effect of nutrition on the different types of fat in the levels of blood lipids, kidney function, and liver enzymes. The results showed a significant increase with a level of 0.05 ( $p \leq$ ) in blood sugar concentration in the two groups of animal fat and vegetable oil group with a concentration of 8% g / kg of diet, and the two olive oil groups did not show significant differences compared to the control group. Levels of cholesterol, triglycerides, low-density lipoproteins, and very low-density lipoproteins in a group that was given fat animal by 8% g / kg of diet and a significant decrease in the group of olive oil by 4% g / kg of diet, significant increase ( $p \leq 0.05$ ) in the concentration of high-density lipoproteins in the two groups that have been treated with Olives oil by: 4% and 8% g / kg, respectively. There were no significant differences in the rest of the other study treatments compared to the control group, and a significant increase of ( $p \leq 0.05$ ) in the concentrations of urea, creatinine and uric acid in the animal fat group by 8% g/kg of the diet, and a significant decrease in the group that was treated with olive oil. As for the liver enzymes activity ALT, AST, and ALP there was a significant increase ( $p \leq 0.05$ ) in the group that was treated as animal fat by 4% g / kg of diet, while we did not observe significant differences in the group that was treated with olive oil by 4% g / Kg of diet compared to a control group.

**Keywords:** Biochemical markers, Liver function, kidney function.

## Introduction

Fats are one of the essential and main substances in food, as well as fats are one of the most important nutrients for humans, as metabolism generates many biologically active molecules [1] Fats are of great importance in our daily diet because they are an important source of energy stored in the body because they are play main role in the synthesis of cellular membranes as well as in the manufacture and synthesis of steroid hormones, and has a role in the manufacture of fat-soluble vitamins, and in the regulation of vital body functions, as well as in the manufacture and formation of bile acids important in the emulsification of fats [2] Whereas one gram of fat produce 9 kilograms / caloric energy twice as much as protein and carbohydrates, as the excess of fat is stored in fatty tissue and in the liver until it is needed [3]. humanity faces many health problems regarding the fat content of food due to

its association with common diseases as a result of this relationship, the cholesterol content of food has become an important topic after the recommendations made by many studies to reduce fat consumption in food [4], Fats differ in some characteristics depending on the source of the fat, and some physical characteristics of fats are an important factor in distinguishing between one fat and another depending on its composition and content in fatty acids [5]. Fats are classified according to their source to animal fats, which is the food supply obtained from animal sources and includes Cow and sheep fat, and milk fat from cows and sheep, are all saturated fats that are solid at room temperature [6]. Free beef fat is considered a complex fat consisting of saturated fatty acids such as short, medium and long chain fatty acids [7]. Free fat is considered a healthy source of edible fats with many beneficial properties. It is also considered a

powerful antioxidant because it contains carotenoids and some vitamins dissolved in fats such as A, E, and D, in addition to that they contain minerals such as calcium and magnesium all give high oxidative efficacy against many diseases [8].

Any oil product extracted from plants is called Vegetable fat, and possesses many fruits, whether seeds or pulp, on different amounts of oil, and the most important plants are sunflower, soybeans, peanuts, olive oil, palm oil and other plants, and vegetable oils are one of the most important plant ingredients. Which are mainly included in food [9], as well as vegetable oils are a good source of energy and of great economic importance in addition to being consumed largely in the prevailing diet [10]. It is characterized by its containment of unsaturated fatty acids that the body cannot synthesis. Among the most important vegetable oils, olive oil is one of the healthiest nutritional oils [11]. Those who are deprived of the best types of oils, and the health-promoting component of a healthy diet, and he noted its effects on the pathogenic genes involved in diseases such as heart disease, cancer and neuropathy through the chemical components of olive oil Such as biophenol and oleic acid, and vitamin E, as well as containing phenolic compounds and monounsaturated fatty acid and unsaturated fatty acids, which act as antioxidants [12]. In addition to the above, many recent studies have confirmed the ability of monounsaturated fatty acids to limit the cardiovascular system through their ability to reduce lipid profiles, inhibit the oxidation of low-density lipoprotein LDL-C and activate antioxidants in the body [13], and sunflower oil is considered one of the most important oilseed crops in the world [14]. It is also considered one of the important oils in human nutrition and is yellow in color and light in density [15] and contains polyunsaturated fatty acids, which is linoleic acid (48-74%) and monounsaturated fatty acids, which are oleic acid (14-40)., %) And linolenic acid and contains saturated fatty acids in a small percentage represented by palmitic acid and stearic acid, and that the consumption of these fats is beneficial to health and thus reduces the risk of developing atherosclerosis and heart disease by lowering the level of cholesterol in the blood [16].

## **Materials and Method:**

### **Experimental animals:**

Albino males rats were used in this study. The ages of the animals ranged from (3-4) months and their weights ranged from (250-300) grams. The animals were obtained from animal house of the College of Veterinary Medicine at the University of Tikrit, and placed in plastic cages with mesh metal covers prepared for this purpose. The animals were subjected to laboratory conditions of light, temperature of 25 C°, and the degree of humidity ranged between 10-30% throughout the study period. the animals were subjected to a two-week introductory period for the purpose of acclimatization to the place and leeches before starting the treatment after ensuring her safety from diseases.

### **Experiment design**

In this study, 49 white male rats were used, and they were divided randomly into seven groups, and each group consisted of seven animals of similar weights as follows:

\* The first group (C), the control group, was given throughout the trial period only a standard diet.

\* The second group, A1, this group was given the feed treated with animal fat (4% g / kg) of the diet throughout the experiment period.

The third group, A2, is the group of animals that were given animal fat (8% g / kg) from the diet and water throughout the experiment period.

\* The fourth group, P1, a group of animals that were given vegetable oil (4% g / kg) of the diet throughout the experiment period.

\* Fifth total, P2, the group of animals that were given vegetable oil at a ratio of (8% g / kg) throughout the experiment period.

\*The sixth group, O1, is a group of animals that were given olive oil (4% g / kg) of the diet throughout the experiment period.

\*The seventh group, O2, the group of animals that were given olive oil (8% g / kg) throughout the experiment period.

### Preparing of food processing:

The components of the concentrated feed mixture were obtained from the College of Veterinary Medicine, University of Tikrit, and its components and the chemical composition of the diet used in feeding rats were in accordance with the nutritional and physiological requirements established by it [17]. Seven types of diets were used in this study, which include the standard diet especially for the control group and three types of diets. Added to it 4% for each kilogram of a diet, animal fat, vegetable oil, olive oil, and the other three diets add 8% to each kilogram of rations from animal fat, vegetable oil and olive oil, respectively.

The feed mixture for rats was modified by adding wheat flour to it to facilitate the formation of the food pieces, then the food used in feeding rats was made by adding the required weight of fat to the flour, then adding the flour and mixing it well, then it was added to the other ingredients and distributed well to obtain a uniform distribution of the fat, Then forming the food after mixing it with a little distilled water and placing it in the feed machine for the purpose of obtaining food in the form of fingers with a diameter of 1.5 cm and a length of 1-2 cm, then drying the food at room temperature for two days away from sunlight to avoid dust and insects, and then Complete the drying in an electric oven at a temperature of 50 ° C and store the dried food in a cool dry place in nylon bags, and number the bags according to each group [18].

### Blood sample collecting:

After the end of the 60-day trial period, blood was drawn once every 14 days, with four withdrawals, as blood was drawn from the vein located in the eyehole according to [19] and blood samples were collected in plastic tubes with tight covers and left for a period of 20 minutes at laboratory temperature, after that, the serum is separated from the blood plasma by a centrifuge for a period of ten minutes at a speed of 3000 rpm, after which the serum is kept in freezing at a temperature of -20 ° C until the laboratory tests are carried out.

### Biochemical tests

1- Determination of serum glucose concentration: The concentration of glucose in the blood serum was

determined using the France Biolabo Analysis Kit, an enzymatic method in which glucose is oxidized.

2- Determination of serum cholesterol concentration: The determination of serum cholesterol was carried out using a ready-made kit from the Spanish company Bio Systems [21].

3- Triglyceride concentration measurement: The measurement of TG concentration in rat blood serum was determined by the enzymatic method by using a ready-made kit from the Spanish company (Bio Systems) [22].

4- Determination of the concentration of high-density lipoproteins of cholesterol in serum: The concentration of high-density lipoprotein (HDL-C) for cholesterol in the blood serum of rats was determined by using a ready-made kit from the Spanish company Bio System [23].

5- 5- Determination of the concentration of low-density lipoproteins of cholesterol: The concentration of C-LDL has been estimated in light of the following equation:

$$\text{LDL-c(mg/dl)} = \text{Total cholesterol} - (\text{HDL-C} + \text{VLDL-C})$$

6- determination the concentration of very low-density lipoproteins for cholesterol: The value of VLDL-C was calculated according to the following method:

$$\text{VLDL-C concentration (mg/dl)} = (\text{Triglycerides}/5) \text{ according to [22].}$$

7. Urea-Creatinine Concentration Measurement: The concentration of creatinine and urea in the blood serum of the experimental animals was measured using the Analysis Kit manufactured by France, Biolabo [25].

8. Determination of uric acid concentration: The concentration of uric acid in the blood serum was measured using the BIOLABO SA, France test kit.

9. Determination of the activity of the enzyme alanine amino transferase transporter enzyme activity and aspartate amine transporter enzyme: the enzymatic methods were used to measure the activity of AST and ALT in the blood serum by using of a ready-made analysis kit from the French company BIOLABO.

10. Determination of the effectiveness of alkaline phosphatase enzyme: it was measured according to the colorimetric method shown in the analysis kit of the French-made Bio Merieux company

- Statistical analysis: The results were statistically analyzed using the One-way ANOVA system to extract the significant differences between more than two averages from the experiment groups with emphasis on these differences by measurement the Standard Error (SE), where the statistical analyzes were performed with significant differences ( $P \leq 0.01$ ),  $P \leq 0.05$ ) [26].

## Results and Discussion

### Comparison of the effect of nutrition on different lipids in blood sugar, cholesterol, triglyceride, HDL-C, LDL-C and VLDL-C concentrations:

Table (1) shows a comparison of the effect of nutrition on different types of fats in the concentrations of blood sugar, cholesterol, triglycerides, HDL-C, LDL-C and VLDL-C for the study treatments, where the highest concentration of blood sugar level was recorded in group A2 that was treated with animal fat at (8% g / kg of diet). The reason is that following a diet containing saturated fats can reduce the activity of intracellular enzymes associated with fatty acid synthesis and thus reduce the ability Intracellular use of insulin [27]. The O2 group that was treated with olive oil at a rate (4% g / kg of the diet) recorded a significant decrease with the level of  $P (\leq 0.05)$ . The reason is due to the nature of the active ingredients and their concentrations in olive oil, the oil contains high concentrations of polyphenols, especially compounds Olirubin, in addition to that, monounsaturated fatty acid, especially Oleic acid, works to improve blood sugar

concentrations by reducing insulin resistance in cells and preventing blood lipid disorders that accompany a decrease in the concentration of glucose in the blood. Also, a significant increase was observed, with a P-value of 0.05 level, in the concentrations of cholesterol and triglycerides, LDL-C and VLDL-C, in the A2 group that were treated with animal fat by (8% g / kg of diet).

Nutrition on saturated fats raises the level of cholesterol and triglycerides, and since animal fat contains a high percentage of saturated fatty acids and these acids are Lauric Palmitic and Meristic, therefore their consumption led to an increase in harmful fats in the serum, as an increase in the proportion of fats leads to an increase Chylomicron production in the intestine and when it degrades causes the release of fatty acids. Therefore, liver cells will receive large quantities of fatty acids, causing an increase in the liberation of triglycerides, as the increase in TG is caused by an increase in VLDL and chylomicron, which leads to hypertriglyceridemia resulting from increased LDL. C and low HDL-C [28,29]. The time for feeding on olive oil leads to a reduction in the lipid profile because it contains squalene, which works to remove free radicals and thus reduces LDL oxidation, or it may be due to its containment of phenolic compounds that reduce or inhibit the reactions of the lipid peroxidation chain reaction.

The reason is due to the inhibition of the Triglyceride Lipase enzyme responsible for the fragmentation of triglycerides, and consequently, an increase in the metabolism of fats, and that nutrition on olive oil leads to an increase in high-density lipoproteins. Its antioxidant activity increases the activity of Catalase and Superoxide dismutase (SOD) in the liver [30].

**Table (1) Comparison of the effect of nutrition on different lipid types in blood sugar concentrations and lipid profiles throughout the study period.**

Treatments/features	control	Animal fat4%	Animal fat8%	Vegetable oil P1	Vegetable oil P2	Olive oil O1	Olive oil
Glucose	87.45±17.7 B	103.70 ±17.94 a	108.00±23.44 a	97.65 ±14.23 ab	106.77 ±12.93 a	90.37±5.19 b	91.24 ±7.58 b
Cholesterol	96.93± 3.11 Cd	107.05 ±20.59 bc	122.30 ±20.98 a	101.90 ±18.10 bc	111.09 17.20± B	89.16 ±7.60 d	98.48±12.04 c

**Cont... Table (1) Comparison of the effect of nutrition on different lipid types in blood sugar concentrations and lipid profiles throughout the study period.**

Triglyceride	69.05±5.75 c	83.65±5.75 b	103.55±34.40 a	82.00 ±14.11 b	84.41 ±18.14 b	78.11±7.37 bc	80.38 ±9.32 b
HDL-C	34.500 ±2.94 B	34.650 ±4.24 B	35.850 ±7.10 b	34.500±2.81 b	37.182 2.15 ± ab	38.316 2.49± a	39.810 ±3.65 a
LDL-C	52.17±4.73 B	54.26 ±15.25 B	65.57±10.11 a	51.53 ±15.14 b	56.98 ±14.48 B	35.31±7.00 d	42.70 ±10.69 C
VLDL-C	13.790 ±0.99 C	16.640 ±2.83 B	20.520 ±6.80 a	16.32 ±2.72 b	16.65 ±3.44 B	15.537 ±1.37 bc	16.162 ±1.85 B

**A comparison of the effect of nutrition on different types of fats in the concentrations of urea, creatinine and uric acid:**

Table (2) shows the effect of nutrition on different types of fats in the concentrations of urea, creatinine and uric acid. The results of current study showed a significant increase in the levels of urea, creatinine and uric acid in a group that was fed on animal fat by (8% g / kg of the diet) and a significant decrease in the group that were given olive oil by (4% g / kg of the diet). The excretion of urea is a result of the metabolism of proteins and amino acids, and that the estimation of the concentration of urea is an important and diagnostic function of disorders and diseases of the kidneys such as urinary tract infections, stones in the kidneys and deficiencies in their functions [31] The results of our current study agree with [32] when feeding on saturated fats leads to raising the level of cholesterol and triglycerides, thus leading to oxidative stress, free radical production, and oxidation of proteins and amino acids. This process results in an increase in the production of urea, and that feeding on unsaturated fats, especially olive oil, reduces the level of

urea because it contains polyphenols and maintains its normal level in the body, as well as the effectiveness of antioxidants and vitamin E that prevent oxidative effect to renal cells and glomeruli. Creatinine is one of the least variable compounds in concentration levels as it is one of the best and most accurate tests for indicating the safety of the kidneys is to estimate the concentration of creatinine in the blood serum. The results of our current study agree with [33], the reason of the decrease in the concentration of the Cyclooxygenase-2 enzyme, which leads to the production of Prostaglandins, and this leads to a decrease in the renal blood flow, impair the kidney function and increase the levels of creatinine in the blood.

Uric acid is the final product of the purine metabolism processes in the body as well as acts as an antioxidant in some liquids. It is an inhibitor of lipid peroxidation and converts some metal ions such as copper and iron into weakly reactive forms capable of capturing hydroxyl radicals [34]. Excessive uric acid concentration is due to an increase oxidative stress from cardiovascular disease.

**Table (2) shows the effect of nutrition on different types of fats in the concentrations of kidney function.**

Treatments/ features	control	Animal fat4%	Animal fat8%	Vegetable oil P1	Vegetable oil P2	Olive oil O1	Olive oil
urea	33.65 ± 5.35 Ab	32.65 ± 5.59 B	37.75± 4.55 a	34.10± 9.21 ab	30.14 ± 4.71 B	31.84± 2.71 B	33.91± 3.49 Ab
Creatinine	0.67± 0.08 C	0.79± 0.15 B	0.85± 0.17 a	0.77± 0.07 B	0.78± 0.09 B	0.67± 0.08 C	0.66± 0.09 C
Uric acid	1.57± 0.16 D	1.95± 0.31 C	2.71±0.57 a	2.13± 0.23 Bc	2.21± 0.26 B	1.96± 0.23 C	2.11± 0.27 Bc

**The effect of nutrition on different types of fats in the concentrations of ALT, AST, and ALP liver enzymes:**

Table (3) shows the effect of nutrition on different types of fats in the concentrations of liver enzymes. The results of our current study showed a significant increase in the level of AST and ALP concentrations in a group that was given animal fat by 8% g / kg of diet and a significant decrease in the group that were given olive oil by 4% g / kg of diet compared to the other treatments groups of this study.

No differences in ALT concentration were observed in all study treatments. The liver plays an important role in the metabolism of foodstuffs such as fats, carbohydrates and proteins and works on producing bile salts that are important in the metabolism of fats [36] that feeding on olive oil works to reduce the concentration of ALP and AST because it contains antioxidants that work to sweep free radicals, or the reason may be attributed to the activation of neurotransmitters, especially acetylcholine which is located in the meeting areas between nerve cells - nerve cells or nerve cells - muscle, so all vital activities in the body are activated. And that feeding on saturated fats leads to an increase in the concentration of

AST. The reason is that the continuation of feeding on saturated fats leads to atherosclerosis and thus increases the concentration of AST in the blood serum as a result of the occurrence of necrosis in the body tissues and damage to the liver causing the release of liver enzymes into the bloodstream through the hepatic channels. Or the reason is due to an increase in the base enzyme phosphatase that occurs in the case of hepatic obstruction, as this enzyme is secreted in the liver, and with all of it, the change in the concentration of enzymes in the serum is attributed to a weakness in liver function. These enzymes appear in the serum as a result of their release in the liver when hepatic tissue is destroyed when the liver suffers from chronic diseases as in acute hepatitis.

The activity of liver enzymes in the tissues is greater than their activity in the blood serum. Or the reason is the increase in the concentration of the levels of ALP and AST, which is due to the cumulative over-nutrition of the fats and vegetable oils in the expression of the enzyme Glucose-6-phosphate dehydrogenase which increases the level of oxidative stress in the liver and thus causes damage. Animal also contains many alkaline components such as phospholipids that activate the saponification process, which affects the increase of liver enzymes [37,38].

**Table (3) the effect of nutrition on different types of fats in liver enzyme concentrations.**

Treatments/features	control	Animal fat4%	Animal fat8%	Vegetable oil P1	Vegetable oil P2	Olive oil O1	Olive oil
AST	26.70 ±3.51 d	31.40 ±4.46 Cd	45.55 ±6.55 a	35.70 ± 10.24 bc	43.73 3.990 ± a	27.74 ±6.90 d	37.19 ±13.35 B
ALT	19.35 ±3.32 A	21.40 ±10.26 A	23.35±11.98 a	24.45±7.69 a	22.95±5.93 a	21.53±7.28 a	26.10±8.75 a
ALP	70.25 ±6.53 c	111.55 ±27.63 A	113.15 ±14.94 a	90.95 ± 15.17 b	105.18 ± 23.16 Ab	77.89 ± 7.29 c	100.43 ± 27.57 Ab

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