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Effect of the ketogenic diet on weight, liver and kidney function

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1. Abstract

This research was performed at the laboratories of the Food Science Department and the animal facility of the College of Veterinary Medicine at Tikrit University, from January 10, 2025, until November 15, 2025. The objective was to examine the impact of a ketogenic diet on the biochemical properties of laboratory white rats subjected to diets comprising fats from diverse sources, including rice oil (T2), animal fats (T3), hydrogenated vegetable oils (T4), ghee (T5), and olive oil (T7), over periods of 28 days and 21 days, respectively. The results indicated a substantial difference ($p < 0.05$) in the initial weights of the rats, which ranged from 284 to 326.25 g. The animals' weights diminished markedly by the conclusion of the experiment, with the greatest weight increase in treatment T2 recorded at 267.50 g, in contrast to 380.00 g in the control group at T1. The findings indicated a notable elevation in ALT enzyme concentration, peaking in treatment T6, which involved coconut oil supplementation, whereas treatment T3, comprising animal fats, had the lowest levels. The findings indicated a notable elevation in ALP enzyme levels across all treatments relative to the control group. Treatment T4, involving the administration of hydrogenated oils, produced the highest concentration, whereas treatment T2, utilizing rice oil, resulted in the lowest concentration. The findings indicated a notable elevation in ALP enzyme levels across all treatments relative to the control group. The findings demonstrate that administering different oils to rats influenced renal function. All treatments demonstrated a statistically significant elevation in creatinine concentration ($p < 0.05$) relative to the control sample T1, which recorded a value of 0.53 g/dl. Conversely, creatinine levels in treatment T2 considerably diminished to 0.47 mg/dl, representing the lowest figure among all treatments. Treatment T4 exhibited the highest creatinine level across the regimens. The findings indicated a considerable increase in urea levels in treatments T2, T5, T6, and T7, although treatments T3 and T4 exhibited a decrease, with levels of 27.67 and 28.33 g/dl, respectively. Treatment T2 produced the highest urea concentration, whilst treatment T3 exhibited the lowest, in comparison to the control treatment T1. The table indicates a substantial reduction in uric acid levels across all treatments T2, T3, T4, T5, T6, and T7, with values of 4.17, 3.73, 3.43, 3.17, 2.73, and 7.13, respectively, in contrast to the control therapy T1, which recorded a level of 5.30.

Keywords: ketogenic diet, weight, liver and kidney function

I. Introduction

Obesity was once considered a health problem associated with high-income countries, but it has now become a global health phenomenon affecting developing countries despite their lower socioeconomic levels(Daran *et al.*, 2023; Sethi & Ford, 2022). Recently, there has been significant interest in studying the impact of different dietary patterns on human health, particularly those patterns that lead to significant changes in metabolism and immune system function. The ketogenic diet (KD) is one of the most scientifically studied dietary patterns, due to its reliance on drastically reducing carbohydrates and increasing fat consumption, which forces the body to rely on ketone bodies as its primary energy source instead of glucose. This diet was initially used for therapeutic purposes, especially in conditions such as drug-resistant epilepsy. Its applications have since expanded to include obesity, type 2 diabetes, and some neurological and immune disorders (Malinowska & Żendzian-Piotrowska, 2024)

The ketogenic diet is a nutritional plan characterized by a high fat content, moderate to low protein intake, and very low carbohydrate levels. This diet induces elevated synthesis of ketone bodies from fat catabolism for energy generation(Kosinski & Jornayvaz, 2017). Certain research indicate that the ketogenic diet possesses therapeutic

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advantages for many ailments. It has been suggested as an adjunctive treatment for polycystic ovary syndrome, acne, cancer, and respiratory distress(Alessandro *et al.*, 2015). It is advantageous as anticonvulsant therapy to diminish the incidence of seizures in individuals with epilepsy (Rho, 2017). The ketogenic diet may aid in lowering HbA1C levels in individuals with type 2 diabetes, stabilizing mood in those with bipolar illness, and decreasing cholesterol levels in obese patients(Chrysafi *et al.*, 2024; Zhou *et al.*, 2022). The current study aims to determine the effect of the ketogenic diet on liver and kidney function and weight.

II. Materials and methods

Preparation of Laboratory Animals:

Modification of Experimental Setting Based on Research Healthy, disease-free laboratory animals were obtained from the College of Veterinary Medicine, Tikrit University. Thirty-five adult male albino rats, aged 8-9 weeks and weighing between 210-220 grams, were collected and randomly divided into seven groups of five animals each, with similar weights. The animals were housed in plastic cages with a wood shavings floor, which were replaced four times a week. The animals were fed regularly using a commercially prepared diet according to the (Council *et al.*, 1995). The standard ketogenic diet for rats consisted of three rations: the first, at a concentration of 30%, included 150 grams of regular mix, 500 grams of flour, 300 grams of animal fats (including lard, coconut oil, hydrogenated oil, and rice oil), and 50 grams of multivitamins. The second 45% feed included (100g regular mix, 400g flour, 450g animal fat, coconut oil, hydrogenated oil and rice oil, 50g multivitamin). The third 65% feed included (100g regular mix, 250g flour, 600g animal fat, coconut oil, hydrogenated oil and rice oil, 50g multivitamin).

Experimental Design: The animals were divided into seven groups, including:

T1: Control Treatment

T2: Rice Oil-Based Rats

T3: Animal Fat-Based Rats

T4: Hydrogenated Oil-Based Rats

T5: Free-Fat-Based Rats

T6: Coconut Oil-Based Rats

T7: Olive Oil-Based Rats

Assessment of biochemical test

The level of Alanine Aminotransferase (ALT) ، Aspartate Aminotransferase (AST) و Alkaline phosphatase (ALP) was estimated using a ready-made analytical kit manufactured by the French company BIOLABO. The operating principle of this kit is based on the enzymatic method.

Statistical analysis:

The data were statistically analyzed through the experimental system within the ready-made statistical program (SAS, 2012) and using the CRD completely randomized design system, as the means were chosen according to Duncan's (Dancan, 1955) multiple range test to determine the significance of the differences between the means of the factors affecting the studied traits at the level of (0.05).

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III. Result

The results show a significant difference ($p < 0.05$) in the weights of the rats at the initial weight stage, which ranged from 284 to 326.25 g. The weights of the animals decreased significantly at the end of the experiment, with the weights gained in treatments T2, T3, T5, T6, and T7 being 267.50, 297.50, 281.25, 292.25, and 265.00 g respectively, while they increased significantly in treatment 4T, reaching 356.25 g when compared with the weights of the animals in the control group, which were 380.00 g at T1. As shown in Table (1)

Table (1). The effect of feeding various oils according to the program based on the ketogenic diet and its effect on the weight rates of rats after 28 days.

Treatments	Initial weight in grams	Final weight in grams	Weight difference
T1	326.25 ± 19.08 ab	380.00 ± 15.41 ab	53.75 ± 5.54 ac
T2	284.25 ± 14.04 c	267.50 ± 11.63 bb	-16.75 ± 3.83 bc
T3	316.25 ± 36.82 a	297.50 ± 9.24 bc	-8.00 ± 3.13 cc
T4	310.00 ± 7.36 ac	356.25 ± 12.80 ab	53.75 ± 11.43 ab
T5	291.25 ± 11.25 c	281.25 ± 9.65 bc	-10.00 ± 2.04 cc
T6	305.00 ± 22.07 ba	292.25 ± 21.33 ba	-12.25 ± 1.31 cc
T7	291.25 ± 19.51 cb	265.00 ± 9.269 bc	-26.25 ± 4.06 bc

The results in Table (2) show that the AST enzyme showed a significant decrease ($p < 0.05$) in treatments T2, T4, T5, T6, and T7, reaching 73.33, 79.33, 66.00, 194.33, and 87.00 IU/L, respectively, compared to the control group T1, where the enzyme level was 107.67 IU/L. Treatment T3 had the highest level compared to the control group, reaching 131.00 IU/L, while the enzyme level was lowest in treatment T5, in contrast to treatment T3, which had the highest enzyme level. The results showed that the ALT enzyme exhibited a significant increase in concentration, with average increases in the treatments reaching 72.00, 40.33, 44.67, 44.33, 60.33, and 45.33 IU/L respectively, compared to 33.33 IU/L for the control treatment T1. Observing the results of the treatments, it was found that treatment T6, which included feeding with coconut oil, had the highest increase, while treatment T3, which consisted of feeding with animal fats, had the lowest. The results in the table also show a significant increase

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in ALP enzyme values for treatments T2, T3, T4, T5, T6, and T7, reaching 104.00, 178.33, 197.67, 159.33, 125.67, and 107.00 IU/L, respectively, compared to the enzyme concentration in the control group T1, which was 126.33 IU/L. Treatment T4, which involved feeding hydrogenated oils, yielded the highest concentration, while treatment T2, which involved feeding rice oil, showed the lowest concentration when compared between the samples.

Table (2). Effect of feeding on various oils according to the program based on the ketogenic diet and its effect on liver enzymes IU/L for rats after 28 days.

Treatments	ALP U/L	AST U/L	ALT U/L
T1	3.71 ± 125.33 d	3.38 ± 107.67 b	0.88 ± 33.33 b
T2	6.00 ± 104.00 e	5.24 ± 73.33 de	5.51 ± 72.00 a
T3	4.10 ± 178.33 b	4.93 ± 131.00 a	2.19 ± 40.33 b
T4	7.22 ± 197.67 a	6.23 ± 79.33 cde	3.28 ± 44.67 b
T5	5.24 ± 159.33 c	5.51 ± 66.00 cd	3.76 ± 44.33 b
T6	4.37 ± 125.67 d	3.33 ± 94.33 bc	6.06 ± 60.33 a
T7	4.93 ± 107.00 e	5.69 ± 87.00 cd	3.18 ± 45.33 b

The results in Table (3) indicate that feeding rats a diet of various oils for 28 days had an effect on kidney function. All treatments showed a significant increase in creatinine concentration ($p < 0.05$). Treatments T3, T4, T5, T6, and T7 had creatinine values of 0.60, 0.73, 0.76, 0.75, and 0.70 g/dl, respectively, compared to the control group T1, which had a value of 0.53 g/dl. Creatinine values decreased significantly in treatment T2 to 0.47 mg/dl, the lowest value among all treatments. Treatment T4 recorded the highest creatinine value compared to the other treatments. The results showed that urea values increased significantly in treatments T2, T5, T6 and T7, reaching 53.67, 32.00, 43.33 and 31.00 g/dl respectively, compared to the control sample T1, which was at 29.00 g/dl. Urea values decreased in treatments T3 and T4, reaching 27.67 and 28.33 g/dl respectively. The results showed that treatment T2 gave the highest urea value, while treatment T3 had the lowest value among all treatments compared to the control treatment T1. The results in the table also showed a significant decrease in uric acid values in all treatments T2, T3, T4, T5, T6 and T7, reaching 4.17, 3.73, 3.43, 3.17, 2.73 and 7.13 respectively, compared to the control treatment T1, which reached 5.30.

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Table (3). The effect of feeding various oils according to the program based on the ketogenic diet and its effect on kidney function in rats after 28 days.

Treatments	URIC mg/dl	CRE (IU/L)	UREA (µmol/L)
T1	0.21 ± 5.30 a	0.09 ± 0.53 ab	0.58 ± 29.00 b
T2	0.65 ± 4.17 ab	0.07 ± 0.47 b	1.45 ± 35.67 a
T3	0.29 ± 3.73 ab	0.06 ± 0.60 ab	0.33 ± 27.67 b
T4	1.03 ± 3.43 ab	0.03 ± 0.73 a	0.67 ± 28.33 b
T5	0.92 ± 3.17 b	0.03 ± 0.67 ab	3.00 ± 32.00 ab
T6	0.20 ± 2.73 b	0.07 ± 0.57 ab	1.33 ± 34.33 a
T7	0.38 ± 4.13 ab	0.06 ± 0.70 a	1.00 ± 31.00 ab

IV. Discussion

These results are consistent with (Daniel *et al.*, 2014) in an experimental study on mice subjected to a high-fat diet, who observed negative effects on weight and metabolism through modification of the intestinal flora composition. Therefore, based on the above results and the studies that agree with them, it is confirmed that following the ketogenic diet has negatively affected the growth rates of laboratory rats fed this diet, and that supplementation through oral administration has mitigated these effects by modifying the intestinal environment, although the effect is partial. It can also be argued that the reason for this is that the ketogenic diet, characterized by a sharp decrease in carbohydrates and a rise in fats, may have resulted in reduced glucose-dependent energy production and altered gut microbiota composition and balance, potentially leading to decreased growth. However, it may have also contributed to a positive improvement in the gut microbiota balance affected by the ketogenic diet, resulting in increased production of certain short-chain fatty acids (SCFAs) such as butyrate, which may support nutrient absorption and promote growth. A study by (Jornayvaz *et al.*, 2010) showed that prolonged ketogenic feeding in rats caused hepatocyte hyperplasia and fat accumulation. The results of a study by (Al-Muzafar & Amin, 2017)also supported the negative effects of the ketogenic diet on liver tissue by increasing markers of inflammation and liver damage.

The results were consistent with (Daniel *et al.*, 2014; Kolb *et al.*, 2021)found, that relying on the ketogenic diet can cause an increase in liver enzymes due to the stimulation of lipolysis and the accumulation of toxic ketone bodies. The results were also consistent with(Qu *et al.*, 2025), who conducted a clinical trial on twenty samples who followed the ketogenic diet and observed a decrease in liver enzymes after 20 days of the trial. The reason for the decrease in liver enzymes when following the ketogenic diet may be due to MT2 receptors, which are a

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type of melatonin hormone receptor found in the body's cells. These receptors belong to the protein-associated receptor family, and when melatonin hormone binds to these receptors, it causes important physiological effects, including regulating the biological clock and regulating the action of hormones, including insulin and leptin. It also affects glucose metabolism, in addition to its effect on metabolism and neurological functions. This explains the appearance of some neurological symptoms in people who follow the ketogenic diet(You *et al.*, 2024).

The increase in some results can be explained by impaired kidney function due to the metabolic stress associated with the ketogenic diet, which in turn leads to the accumulation of protein metabolism products such as urea. These results are consistent with (Arsyad *et al.*, 2020) indicated, demonstrating that prolonged ketogenic diets lead to increased blood urea concentrations due to the increased use of amino acids as an energy source. A study by (Chen *et al.*, 2021) also supported these observations, and this finding aligns with the results of(Sun *et al.*, 2020), who showed that diets high in fat and protein can cause damage to the renal tubules and increased creatinine accumulation. This increase reflects a decrease in glomerular filtration rate and kidney dysfunction resulting from metabolic stress and increased ketone body concentration.

The decrease could be due to a change in the gut microbiota, which contributes to lowering urea concentrations by modifying nitrogen metabolism and promoting gut microbial balance. This, in turn, positively impacts renal excretion efficiency. Ketones, specifically beta-hydroxybutyrate (BHB), one of the main ketone bodies produced by the liver from fats during fasting, prolonged exercise, or following a ketogenic diet, are key. BHB inhibits the activity of histone deacetylases (HDACs), vital proteins that regulate gene expression by removing the acetyl group. Their primary benefit lies in the precise control of cellular functions in kidney tissue, enhancing the expression of genes that respond to oxidative stress, including Foxo3a and Mt2.52. This provides protection against oxidative stress in human kidney cells and various animal models. Studies also show that following a ketogenic diet or BHB therapy can activate the red blood cell nuclear transcription factor 2 (Nrf2) pathway, which is a key pathway for detoxification and oxidative stress(Milder *et al.*, 2010).

Conclusion

The study observed a significant decrease in the weight of rats on the ketogenic diet in all samples except the group fed hydrogenated oils. The results showed significant differences in liver enzymes. ALT levels increased in the treatments fed rice and coconut oil, while AST and ALP levels increased significantly in the animal fat-based diet and decreased significantly in the other treatments. Kidney function was not significantly affected in most treatments.

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