



Palm Oil Modulates Haematological Profile and ATPase Activities of Wistar Rats

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ABSTRACT

Palm oil is widely consumed globally, yet its effects on health remains a topic of debate. In this study, the effect of palm oil on haematological indices and ATPase activities in Wistar rats was assessed. Thirty-six (36) adult rats were divided into six groups of six rats each made of normal rats (group 1). Group II served as experimental control and was given fresh palm oil of 0.4 % FFA. Groups III-VI were given palm oil that has been stored for sixteen months of varied FFA levels (4.8, 8.4, 21.9, and 42.7 %) respectively. The rats were administered a dosage of 480 mg/kg body weight of CPO orally for four weeks. Haematological parameters, including red blood cell count, packed cell volume, haemoglobin, platelets, white blood cell and differential counts were measured using standard haematology analyzer. ATPase activities were also assessed to determine metabolic and enzymatic activities using standard methods. The results indicate that significant alterations were observed in the haematological profile of stored palm oil-fed rats, including changes in haemoglobin and erythrocyte counts. ATPase activity was also modulated, indicating mild disruptions in cellular energy metabolism. The study suggests that free fatty acids levels in palm oil determine its effects on haematological and ATPase activity in Wistar rats.

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INTRODUCTION

Palm oil, obtained from the mesocarp of the fruit of the oil palm tree (*Elaeis guineensis*), is one of the most widely produced and consumed vegetable oils in the world (Asiriwuwa *et al.*, 2025). It plays a significant role in global nutrition due to its affordability, high caloric value, and versatile applications in food preparation, pharmaceuticals, cosmetics, and biofuel production. Palm oil is particularly important in tropical regions such as West Africa and Southeast Asia, where it constitutes a major component of the daily diet (Sambanthamurthi *et al.*, 2011). Nutritionally, palm oil contains a balanced composition of saturated and unsaturated fatty acids, including palmitic acid, oleic acid, and linoleic acid, as well as bioactive compounds such as tocopherols, tocotrienols, carotenoids, and phytosterols that possess antioxidant properties (Edem, 2009; Asiriwuwa *et al.*, 2025). Despite its nutritional benefits, the health effects of palm oil remain a subject of scientific debate, particularly when the oil undergoes prolonged storage/oxidation or improper processing. During storage, hydrolysis and oxidation of triglycerides can occur, leading to the accumulation of free fatty acids (FFA) and the formation of lipid peroxidation products such as aldehydes, peroxides, and ketones. Elevated FFA levels are widely used as indicators of palm oil deterioration and quality degradation (Frega *et al.*, 1999). Oxidized lipids and degradation products have been associated with oxidative stress, cellular membrane damage, and alterations in metabolic processes, which may ultimately affect physiological functions in biological systems (Ukoh *et al.*, 2024).

Haematological parameters can serve as important indicators of physiological and pathological states in animals and

humans (Khan and Zafar, 2005). Haematological indices such as red blood cell count (RBC), haemoglobin concentration (Hb), packed cell volume (PCV), white blood cell count (WBC), and platelet levels provide valuable insights into oxygen transport capacity, immune competence, and overall systemic health. Alterations in these parameters may reflect nutritional deficiencies, oxidative damage, or toxic effects of dietary substances (Ani *et al.*, 2017). Previous studies have suggested that fresh palm oil may support normal haematological functions due to its antioxidant components, whereas oxidized/degraded palm oil may induce oxidative damage to erythrocyte membranes and disrupt hematopoiesis. In addition to haematological effects, dietary lipids may influence the activity of membrane-bound enzymes involved in cellular energy metabolism and ion transport. Among these enzymes, ATPases play a critical role in maintaining cellular homeostasis. Enzymes such as Na⁺/K⁺-ATPase, Ca²⁺-ATPase, and Mg²⁺-ATPase are integral membrane proteins responsible for regulating ion gradients across biological membranes through the hydrolysis of adenosine triphosphate (ATP). These ion gradients are essential for numerous physiological processes including nerve impulse transmission, muscle contraction, nutrient transport, and regulation of intracellular calcium levels. Disruption of ATPase activity can therefore lead to impaired cellular function and metabolic imbalance. Free fatty acids and lipid peroxidation products generated during the deterioration of oils have been reported to interfere with membrane integrity and enzyme function. Some studies have shown that elevated levels of free fatty acids can alter membrane fluidity and inhibit the activity of ATP-dependent ion transporters, potentially leading to disturbances in ionic balance and cellular energy metabolism (Swann, 1984). Such

alterations may compromise cell viability and contribute to metabolic and physiological dysfunction. Given the widespread consumption of palm oil, particularly in developing countries where processing/storage conditions may not always be optimal, therefore, understanding the biological effects of deteriorated palm oil is of significant public health importance. While several studies have investigated the nutritional properties of palm oil, limited information exists regarding the specific impact of varying free fatty acid levels on haematological indices and ATPase enzyme activities. Therefore, the present study evaluated the effects of palm oil of different levels of free fatty acids on haematological parameters and ATPase activities in Wistar rats. By examining how the quality and storage duration of palm oil influence these physiological markers, this research therefore aims to provide deeper insight into the potential health implications associated with the consumption of fresh and oxidized palm oil of free fatty acids levels (0.4% - 42.7%), and how this can contribute to improved dietary safety recommendations.

MATERIALS AND METHODS

Preparation and Collection of the Palm Oil Samples

Freshly milled palm oil sample (A), and stored palm oil samples (B-E) that was kept for sixteen (16) months at room temperature, and of free fatty acid (FFA) levels of 0.4%, 4.8%, 8.4%, 21.9% and 42.7% respectively, was obtained from Nigerian Institute for Oil Palm Research (NIFOR), Benin City, Edo State, Nigeria.

Determination of Free Fatty Acids (FFA) Levels

The FFA was determined according to AOCS official method Ca 5-40 by titration. Results were expressed as percentage of palmitic acid.

Animal Grouping and Administration of Palm Oil Sample

Albino rats (Wistar strain) of weights 130-150g were selected for this study. The animals were obtained from the Animal House of the Department of Biochemistry, Faculty of Life Sciences, University of Benin, Benin City, Edo state, Nigeria. Thirty-six (36) Wistar rats were divided into six groups of six rats per group and treated as follows: Group I (normal control) given feed and water only. Group II (Experimental Control), given freshly milled crude palm oil containing 0.4 % FFA. Groups III-VI were given stored palm oil of varied FFA levels (4.8 %, 8.4 %, 21.9 % and 42.7 %) respectively. A dosage of 480mg/kg body weight was administered to the animals orally for four weeks. The animals were kept in clean cages in a 12-hour light/dark cycle room with daily litter change. The animals were acclimatized for two weeks before the experiment commenced and were fed with Grower's mash and water ad libitum. During the study, rats were maintained under standard conditions. Ethical approval for the use of laboratory animals was granted by the Ethical Committee of the Faculty of Life Sciences, University of Benin (approval no. FLS/REC/2025/001). The animals were euthanized under mild halothane anesthesia at the end of treatment period after an overnight fast, and blood was collected by cardiac puncture into plain sample bottles for biochemical analyses, while the

liver, kidney and heart were excised and used for ATPase activity determination.

Haematological Parameters

The following haematological parameters were measured on the blood sample: Red blood cell (RBC), packed cell volume (PCV), concentration of haemoglobin, platelets, total and differential white blood cells count using a standard haematology analyzer. The haematology analyzer uses three detectors and two reagents for sample analysis. The WBC detector, using the DC detection method estimates the WBC count, while the RBC count and platelets were analyzed by the RBC detector block, using the DC detection method as well. The Hb detector measures the concentration of haemoglobin by the non - cyanide haemoglobin method. The haematology analyzer counts blood cells by impedance.

ATPase Activities Determination

The ATPase activities in liver, kidney and heart of the animals were determined according to the methods below:

Estimation of total ATPase activity using the method of Evan, (1969).

Estimation of sodium/potassium ATPase activities using the method of Bonting, (1970).

Estimation of calcium ATPase activity using the method of Hjerten and Pan, (1983).

Estimation of magnesium ATPase activity using the method of Ohnishi *et al.* (1982).

Data Analyses

Statistical analyses of data was carried out using the statistical package for social science (SPSS) version 21.0. Results were expressed as mean \pm SEM of five replicates. The levels of homogeneity amongst groups were tested using one-way analysis of variance (ANOVA) with $p < 0.05$ considered significant. Duncan's multiple range test was used to separate homogenous groups.

RESULTS AND DISCUSSION

Effect of Palm Oil on Hematological Parameters of Rats

The effect of palm oil on haematological parameters is presented in Table 1 below. There was a significant decrease ($p < 0.05$) in the levels of red blood cells (RBC), packed cell volume (PCV), and hemoglobin (HGB) of rats fed fresh and stored palm oil compared to the control group. On the other hand, there was a significant increase in the levels of platelets of rats fed fresh and stored palm oil compared to the control group. There was also a significant decrease ($p < 0.05$) in the levels of white blood cells (WBC) of rats fed with fresh palm oil (group II) compared to the normal control group but rats that received palm oil of higher FFA levels (groups III-VI) had similar levels of WBC compared to control group. There was no significant difference ($p > 0.05$) in the levels of lymphocytes, monocytes and granulocytes in rats fed with fresh and stored palm oil (groups II-IV) compared to the normal control. On the other hand, rats that received palm oil with higher FFA had higher significant levels compared to those of the normal control ($p < 0.05$).

Table 1: Haematological Parameters of Rats

Parameters	Animal Groups					
	I (Control)	II (0.4% FFA)	III (4.8% FFA)	IV (8.4% FFA)	V (21.9% FFA)	VI (42.7% FFA)
RBC $\times 10^6$ (u/L)	7.00 \pm 0.05 ^c	6.35 \pm 0.14 ^b	5.95 \pm 0.08 ^a	6.65 \pm 0.02 ^c	6.35 \pm 0.37 ^b	7.10 \pm 0.11 ^c
WBC $\times 10^3$ (u/L)	6.70 \pm 0.63 ^b	4.35 \pm 0.02 ^a	7.30 \pm 0.63 ^b	5.70 \pm 1.09 ^b	7.05 \pm 0.25 ^b	6.60 \pm 0.28 ^b

Parameters	Animal Groups					
	I (Control)	II (0.4% FFA)	III (4.8% FFA)	IV (8.4% FFA)	V (21.9% FFA)	VI (42.7% FFA)
PLT $\times 10^3$ (u/L)	553.00 \pm 62.64 ^a	758.50 \pm 3.17 ^b	603.00 \pm 16.16 ^a	776.00 \pm 35.79 ^b	598.50 \pm 3.17 ^a	607.00 \pm 32.9 ^a
HGB (g/dL)	15.20 \pm 0.05 ^b	13.80 \pm 0.17 ^a	13.00 \pm 0.34 ^a	13.60 \pm 0.28 ^a	13.25 \pm 0.72 ^a	15.50 \pm 0.23 ^b
PCV (%)	45.25 \pm 0.43 ^b	41.85 \pm 0.66 ^a	40.00 \pm 1.15 ^a	41.70 \pm 1.03 ^a	40.25 \pm 1.76 ^a	45.85 \pm 0.14 ^b
LYM (%)	89.95 \pm 0.25 ^c	90.60 \pm 0.92 ^c	89.25 \pm 1.06 ^b	91.10 \pm 0.51 ^c	86.03 \pm 1.03 ^a	86.85 \pm 0.72 ^a
MON (%)	7.65 \pm 0.29 ^a	7.00 \pm 0.75 ^a	8.55 \pm 0.15 ^a	6.60 \pm 0.97 ^a	11.10 \pm 0.98 ^c	10.20 \pm 0.51 ^b
GRAN (%)	2.40 \pm 0.00 ^a	2.40 \pm 0.17 ^a	2.30 \pm 0.05 ^a	2.30 \pm 0.34 ^a	2.90 \pm 0.05 ^b	2.95 \pm 0.20 ^b

Values are in mean \pm SEM (n = 4). Means with the same superscript are not significantly different ($p > 0.05$), while means with different superscript are significantly different ($p < 0.05$).

The result reveals significant alterations in haematological and enzymatic activities of the animals. The RBC count of the normal control group exhibited a significantly higher count compared to the 4.8 % FFA group which showed a marked reduction ($p < 0.05$), consistent with the findings of Siddique *et al.*, (2020), that elevated FFA levels inhibit erythropoiesis. In contrast, the 0.4 % FFA group showed a moderate decrease in RBC count, aligning with Wang *et al.* (2018), that suggested lower FFA concentrations might only mildly impair hematopoiesis without causing significant reductions. Interestingly, the 8.4 % FFA group exhibited RBC counts similar to the control, indicating that moderate FFA concentrations may not significantly impact erythropoiesis (Zhang *et al.*, 2019). Similarly, the 21.9 % FFA group showed comparable results to the 0.4 % FFA group, implying that modest increases in FFA levels might lead to a slight reduction in RBC production, as reported by Chen *et al.* (2017). However, the highest FFA concentration (42.7 %) resulted in an increase in RBC count, suggesting a compensatory response to elevated FFA, possibly through a stress-induced erythropoiesis mechanism, as noted by Hussain *et al.* (2018). This observation supports the findings of Zhang *et al.* (2019), that suggested that high lipid concentrations might stimulate erythropoiesis under certain conditions. In summary, these results indicate that the effect of FFAs on RBC count is concentration-dependent, where lower FFA levels suppress erythropoiesis, while higher concentrations may activate compensatory erythropoiesis mechanisms.

The PCV of the control group (I) had a comparable value with that of the 42.7 % FFA group. In contrast, the groups with moderate FFA concentrations (0.4 %, 4.8 %, 8.4 %, and 21.9 %) exhibited significantly lower PCVs respectively. These reductions were statistically significant when compared to the control and 42.7 % FFA groups ($p < 0.05$). The results align with previous research indicating that moderate FFA levels can lower PCV, potentially through alterations in erythrocyte membrane properties or cell dehydration (Wang *et al.*, 2018). Studies by Lee *et al.* (2017) have shown that low FFA concentrations impair erythrocyte deformability, which contributes to reduced PCV, similar to the findings in groups II-V in this study. On the other hand, the elevated PCV observed in the 42.7 % FFA group is consistent with findings by Das *et al.* (2020), that proposed that compensatory mechanisms, such as increased erythropoiesis or altered blood viscosity, might counterbalance the effects of high FFA, resulting in a normalized or even enhanced PCV. Such adaptive mechanisms have also been noted in models with

lipid overload or high-fat diets (Patel *et al.*, 2019), where high FFA levels trigger responses that prevent large decreases in haematological parameters like PCV. Overall, these results suggest that while moderate FFAs can negatively influence red blood cell characteristics, higher concentrations may induce adaptive mechanisms that restore or even increase PCV. These effects likely involve complex interactions in lipid metabolism, oxidative stress, and erythrocyte membrane fluidity, all of which can impact cell survival and functionality.

The Haemoglobin result indicates that control group had higher HGB value while groups exposed to lower-moderate-high concentrations (0.4 %, 4.8 %, 8.4 %, and 21.9 % FFA) exhibited a decrease in HGB levels, with these differences being statistically significant compared to the control ($p < 0.05$). Conversely, the 42.7 % FFA group showed an increase in HGB count which did not differ significantly from the control ($p > 0.05$). This concentration-dependent response suggests that higher FFA levels might stimulate erythropoiesis or protect red blood cells from oxidative stress damage, a theory supported by Roberts *et al.* (2021), that found that increased FFA concentrations can induce compensatory erythropoiesis due to lipid-induced damage to red blood cell membranes. The anomaly in HGB observed in the various FFA groups could be linked to oxidative stress and/or lipid peroxidation. These findings imply that while moderate FFA levels might negatively affect hematological parameters, higher FFA concentrations may trigger adaptive mechanisms that mitigate the adverse impact on HGB levels, probably involving complex biochemical processes like the regulation of antioxidant defenses.

Platelets count of the control group (I) showed a baseline PLT of $553.00 \pm 62.64 \times 10^3 \mu\text{L}$. In contrast, exposure to 0.4% FFA (Group II) significantly increased the PLT count suggesting that low FFA concentrations might enhance platelet activation or aggregation. On the other hand, moderate-higher FFA levels, 4.8 % - 42.7 % led to a decrease in PLT, with significant differences observed only when compared to the 0.4 % FFA group ($p < 0.05$).

These findings are in line with previous research suggesting that FFA concentrations influence platelet aggregation in a biphasic fashion: low FFA concentrations may enhance platelet aggregation, while higher concentrations could lead to platelet dysfunction or reduced aggregation (Li *et al.*, 2018; Yang *et al.*, 2020). Additionally, Thompson *et al.* (2019) proposed that FFA may alter platelet membrane fluidity, potentially explaining the variability in PLT across different groups. The results also support Gupta *et al.* (2017), that suggested that elevated FFA levels could initiate inflammatory responses that, in turn, modify platelet function and aggregation in a dose-dependent manner. The results indicate a complex and concentration-dependent relationship

between FFA levels and platelet count, which may have implications for understanding platelet activation in metabolic conditions such as obesity and diabetes, where elevated FFA levels are commonly observed.

The effects of different concentrations of free fatty acids (FFA) on white blood cell (WBC) count and differential counts (lymphocytes, monocytes, and granulocytes) reveals that WBC count of control was within normal range. Group II (0.4 % FFA) showed a significant decrease suggesting a possible suppressive effect of low FFA concentration on immune cells. Groups III, V, and VI (4.8 %, 21.9 %, 42.9 % FFA) had elevated WBC counts compared to control group, indicating an immune response activation at higher FFA concentrations. The results imply a concentration-dependent response where moderate FFA concentrations stimulate WBC production, while very low FFA suppresses it. The results of lymphocytes (Lym %) indicates that control group and groups that received fresh and stored palm oil of moderate FFA levels groups (II, III, and IV) maintained high lymphocyte counts and did not vary significantly from each other ($p > 0.05$). However, groups V and VI showed significant decreases compared to control, suggesting higher FFA concentrations may reduce lymphocyte levels. Lymphocytes plays crucial role in adaptive immunity; thus, reduced lymphocyte count at high FFA levels could indicate a shift toward an innate immune response. Control rats had monocytes (Mon %) levels similar to those the low FFA group I (0.4 %). However, higher FFA concentrations led to increased monocyte levels as shown in groups V and VI. Monocytes are important in inflammation and tissue repair, so the rise with higher FFA concentrations suggests an inflammatory response probably triggered by increased lipid peroxidation levels. The granulocytes (Gran %) of control was similar to those of groups II, III, and IV. Groups V and VI showed a slight but significant increase suggesting granulocyte recruitment at higher FFA levels. Granulocytes (mainly neutrophils) are often involved in the innate immune response and inflammation. The increase in this case supports the inflammatory response observed with increased monocytes.

Overall, low FFA levels (0.4 %) suppress WBC count without significantly altering monocytes or granulocytes. Moderate FFA levels (4.8 %) increase WBC count without major changes in leukocyte distribution. High FFA concentrations (21.9 % and 42.9 %) decrease lymphocytes and increase monocytes and granulocytes, suggesting a shift toward innate immunity and potential inflammation. FFA may influence immune function by modulating leukocyte distribution.

Effect of Palm Oil on ATPase Levels of Rats

The effect of palm oil on ATPase levels of rats is depicted in Table 2. There was a non- significant decrease ($p > 0.05$) in the liver ATPase levels of rats fed with fresh palm oil (group II) compared to the control group. The decrease in the total ATPase and Na^+/k^+ ATPase levels were very significant ($p < 0.05$) in rats fed palm oil of higher FFA levels (groups III-VI) compared to those of groups (I and II). On the other hand, rats that received palm oil of higher FFA levels had increased levels of Ca^{2+} ATPase and Mg^{2+} ATPase levels respectively compared to the control group. There was no significant difference ($p > 0.05$) in the Total ATPase and Na^+/K^+ ATPase levels in kidney of rats fed fresh and stored palm oil and normal control rats. On the other hand, there was a significant increase ($p < 0.05$) in the Ca^{2+} ATPase and Mg^{2+} ATPase levels of rats fed with fresh and stored palm oil (groups II-VI) compared to the control group. There was no significant difference ($p > 0.05$) in the Total ATPase and Ca^{2+} ATPase levels in heart of rats fed fresh and stored palm oil (groups II-VI) compared to the normal control group. There was a significant decrease ($p < 0.05$) in the Na^+/K^+ ATPase levels of rats fed with fresh and stored palm oil compared to the normal control group. On the other hand, there was a significant increase ($p < 0.05$) in the in the Mg^{2+} ATPase levels of rats given fresh and stored palm oil compared to the normal control group. Overall, there was a higher level of Total ATPase in the liver compared to the kidney and heart. Furthermore, there was higher levels of Na^+/K^+ ATPase, Mg^{2+} ATPase and Ca^{2+} ATPase activity in the heart compared to the liver and kidney.

Table 2: ATPase Levels in the Liver, Kidney, and Heart of Rats

Organs	ATPases	Groups					
		I (Control)	II (0.4% FFA)	III (4.8% FFA)	IV (8.4% FFA)	V (21.9% FFA)	VI (42.7% FFA)
Liver	Total ATPase $\times 10^{-7}$ (molPi/min/mg protein)	5.76 \pm 0.08 ^d	5.10 \pm 0.00 ^d	0.70 \pm 0.23 ^a	2.50 \pm 0.05 ^c	1.66 \pm 0.31 ^b	2.00 \pm 0.40 ^b
	Na^+/K^+ ATPase $\times 10^{-7}$ (molPi/min/mg protein)	0.30 \pm 0.05 ^a	0.20 \pm 0.00 ^a	0.26 \pm 0.08 ^a	0.60 \pm 0.11 ^b	0.10 \pm 0.00 ^a	0.23 \pm 0.03 ^a
	Ca^{2+} ATPase $\times 10^{-7}$ (molPi/min/mg protein)	0.46 \pm 0.20 ^b	0.40 \pm 0.05 ^b	0.80 \pm 0.23 ^c	0.20 \pm 0.05 ^a	0.46 \pm 0.03 ^b	1.00 \pm 0.11 ^c
	Mg^{2+} ATPase $\times 10^{-7}$ (molPi/min/mg protein)	1.56 \pm 0.14 ^a	1.30 \pm 0.05 ^a	1.50 \pm 0.05 ^a	1.80 \pm 0.00 ^a	1.90 \pm 0.00 ^a	2.83 \pm 0.41 ^a
	Total ATPase $\times 10^{-7}$ (molPi/min/mg protein)	1.00 \pm 0.11 ^b	1.00 \pm 0.00 ^b	3.00 \pm 0.00 ^c	0.30 \pm 0.11 ^a	1.20 \pm 0.00 ^b	1.20 \pm 0.05 ^b
	Na^+/K^+ ATPase $\times 10^{-7}$ (molPi/min/mg protein)	0.70 \pm 0.17 ^b	0.40 \pm 0.11 ^b	0.10 \pm 0.00 ^a	0.46 \pm 0.08 ^b	0.50 \pm 0.05 ^b	0.50 \pm 0.11 ^b
Kidney	Ca^{2+} ATPase $\times 10^{-7}$ (molPi/min/mg protein)	0.86 \pm 0.03 ^b	1.30 \pm 0.28 ^c	0.46 \pm 0.08 ^b	0.26 \pm 0.08 ^a	1.40 \pm 0.28 ^c	1.10 \pm 0.23 ^c
	Mg^{2+} ATPase $\times 10^{-7}$ (molPi/min/mg protein)	0.96 \pm 0.31 ^a	4.66 \pm 0.54 ^c	0.46 \pm 0.14 ^a	2.66 \pm 0.20 ^b	2.76 \pm 0.12 ^b	3.06 \pm 1.25 ^c
	Total ATPase $\times 10^{-7}$ (molPi/min/mg protein)	1.00 \pm 0.11 ^b	1.00 \pm 0.00 ^b	3.00 \pm 0.00 ^c	0.30 \pm 0.11 ^a	1.20 \pm 0.00 ^b	1.20 \pm 0.05 ^b

Heart	Total ATPase $\times 10^{-7}$ (molPi/min/mg protein)	1.10 \pm 0.00 ^b	1.10 \pm 0.17 ^b	1.93 \pm 0.83 ^b	0.40 \pm 0.00 ^a	1.13 \pm 0.03 ^b	0.83 \pm 0.03 ^b
	Na ⁺ /K ⁺ ATPase $\times 10^{-7}$ (molPi/min/mg protein)	1.13 \pm 0.03 ^c	0.90 \pm 0.00 ^c	0.16 \pm 0.03 ^a	0.20 \pm 0.00 ^b	0.26 \pm 0.03 ^b	.46 \pm 0.20 ^b
	Ca ²⁺ ATPase $\times 10^{-7}$ (molPi/min/mg protein)	2.23 \pm 0.20 ^b	1.56 \pm 0.82 ^b	1.16 \pm 0.20 ^b	1.96 \pm 0.03 ^b	0.40 \pm 0.11 ^a	2.33 \pm 0.03 ^b
	Mg ²⁺ ATPase $\times 10^{-7}$ (molPi/min/mg protein)	2.50 \pm 1.16 ^a	4.60 \pm 0.75 ^b	1.90 \pm 0.51 ^a	4.73 \pm 0.03 ^b	1.96 \pm 0.83 ^a	9.50 \pm 0.11 ^c

Values are in mean \pm SEM (n = 4). Means with the same superscript are not significantly different ($p > 0.05$), while means with different superscript are significantly different ($p < 0.05$).

ATPase enzymes play a crucial role in cellular energy metabolism, and their activity is influenced by dietary components. Evaluation of the ATPase activities of rats in different organs shows there was no significant difference ($p > 0.05$) in the liver ATPase levels of rats fed with fresh palm oil (0.4 % FFA) compared to the control group. The decrease in the total ATPase levels were very significant ($p < 0.05$) in rats fed stored palm oil of higher FFA levels (4.8 % - 42.7 %) compared to those of groups while there were no significant alterations in the Na⁺/k⁺ ATPase levels of the animals. The decrease in total ATPase may be due to mitochondrial dysfunction or membrane instability. On the other hand, rats that received stored palm oil of higher FFA levels (4.8 % - 42.9 %) had increased levels of Ca²⁺ ATPase and Mg²⁺ ATPase compared to the control and fresh palm oil groups. These result shows that fresh palm oil is beneficial as it did not significantly alter the levels of the enzymes while stored palm oil of varied FFA levels had differential effects on ATPase activity as it increased Ca²⁺ ATPase and Mg²⁺ ATPase activities while decreasing total ATPase and Na⁺/k⁺ ATPase activities of liver reflecting a compensatory mechanism. This study corroborates earlier study by Beshel *et al.*, (2024), that reported that thermo-oxidized oil lowered sodium-potassium ATPase activity more than photo-oxidized oil in comparison to the control group. The decrease in Na⁺/k⁺ ATPase activity observed could be attributed to the oxidative stress induced by high free fatty acid levels. Free fatty acids, especially when oxidized, generate reactive oxygen species (ROS) which can damage membrane proteins (Boateng *et al.*, 2016). Sodium-potassium ATPase embedded within the sarcolemmal membrane is responsible for maintaining the electrochemical gradient across cell membranes and sustaining cation homeostasis in cells by exchanging three sodium ions for two potassium ions via an energy-dependent process (Liu *et al.*, 2013; Bartlett *et al.*, 2018;). ATPase enzyme activity is crucial for maintaining ionic balance and cellular energy metabolism.

There was no significant difference ($p > 0.05$) in the Total ATPase and Na⁺/k⁺ ATPase activities in kidney of rats fed fresh palm oil and normal control rats, while the activity of the enzymes in rats that received stored palm oil were increased. On the other hand, there was a significant increase ($p < 0.05$) in the Ca²⁺ ATPase and Mg²⁺ ATPase activities of rats fed with fresh and stored palm oil (groups 0.4 % - 42.7 %) compared to the control group. Similarly, there was no significant difference ($p > 0.05$) in the Total ATPase and Ca²⁺ ATPase activities in heart of rats fed fresh and stored palm oil compared to the normal control group. There was a significant decrease ($p < 0.05$) in the Na⁺/k⁺ ATPase activities of rats fed with fresh and stored palm oil compared to the normal control

group. On the other hand, there was a significant increase ($p < 0.05$) in the in the Mg²⁺ ATPase activity of rats given fresh and stored palm oil compared to the normal control group. Overall, there was a higher activity of Total ATPase in the liver compared to the kidney and heart. Furthermore, there was higher activities of Na⁺/k⁺ ATPase, Mg²⁺ ATPase and Ca²⁺ ATPase in the heart compared to the liver and kidney. ATPase, specifically Na⁺/K⁺ ATPase, Ca²⁺ ATPase, and Mg²⁺ ATPase, are essential for the maintenance of ionic balance, myocardial contractility, and overall cardiac homeostasis. The ionic balance is crucial for sustaining resting membrane potential and generating action potentials, which are essential for myocardial contraction and rhythm. Disruption of these ATPase activity may result in arrhythmias and various cardiac dysfunctions (Lingrel, 2010). Calcium ATPase plays a pivotal role in removing calcium ions from the cytoplasm ensuring proper muscle contraction and relaxation, particularly in cardiac myocytes (Mørk *et al.*, 2007). On the other hand, Mg²⁺ ATPase regulates the function of ion channels and receptors in the heart, particularly those involved in calcium and potassium transport. Magnesium Mg²⁺ ATPase modulates the activity of Na⁺-K⁺ ATPase and Ca²⁺-ATPase which are both crucial for normal cardiac rhythm (Bers, 2002). The stability of these ATPase is required in dietary fats and oils and this study indicates that fresh palm oil plays an important role in maintaining liver, kidney and cardiac homeostasis. Previous studies report that high-fat diets, including palm oil, modulate ATPase activity, potentially affecting ion transport and muscle function. This study's findings suggest that palm oil alters ATPase activities, but the extent of the alteration may depend on the duration of exposure and concentrations of FFA in the oil

CONCLUSION

The study suggests that free fatty acids levels in palm oil determine its effects on haematological and ATPase activity in Wistar rats. These effects could have broader implications for metabolic and cardiovascular health, reinforcing the need for proper processing in order to have good quality palm oil beneficial for maintenance of good health.

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