Effect of different high-fat diets on lipid profile in albino rats

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Abstract:
High-fat diets and sedentary lifestyles are the main causes of obesity, which is a major risk factor for chronic illnesses like heart disease, high blood pressure, type 2 diabetes, chronic kidney disease, and several types of cancer. This comparative study examines how male albino rats' lipid profiles are affected by high-fat diets made from widely eaten vegetable oils. A total of 48 male albino rats weighing between 100 and 150 grams were randomly assigned to five groups of eight. As the control group, Group 1 was given a diet that contained 11% energy from 5% soybean oil. Rats in Group 2 were given a diet that contained 30% crude palm oil and 2.5% soybean oil (CPO group). Rats in Group 3 were given a diet that contained 30% refined palm oil and 2.5% soybean oil (RPO group). Rats in groups 2–5 were fed a diet consisting of 2.5% soybean oil and 30% groundnut oil (GO group) and Group 5 rats were fed a diet consisting of 2.5% soybean oil and 30% shea butter (SHB group). Blood samples were taken at the end of the 8-week feeding protocol in order to evaluate lipid profiles, and the atherogenic index and coronary risk index were also computed. The results indicated that there were no significant differences ($p < 0.05$) in the levels of total cholesterol, triglycerides, low-density lipoprotein cholesterol, or high-density lipoprotein cholesterol between the rats fed high-fat diets and the control group. In conclusion, the high-fat meals assessed in this study did not negatively impact the rats' lipid profiles and fared similarly to soybean oil.

Keywords: **High-fat diets, Lipid profile, Albino rats, Vegetable oils, Hypercholesterolemia.**

1. Introduction

Dietary habits significantly influence human health, with high-fat diets being one of the primary contributors to chronic conditions like cancer, heart disease, type 2 diabetes, and obesity [1][2][3]. The composition and quantity of dietary fats directly impact lipid metabolism, lipid peroxidation, and the development of metabolic syndromes, making this a critical area of study in nutritional science and public health. Fats are an essential macronutrient required for energy storage, hormone synthesis and the absorption of fat-soluble vitamins, although dyslipidemia—a significant risk factor for cardiovascular disease—has been connected to excessive ingestion, particularly of saturated and trans fats [1]. Triglycerides, low-density lipoprotein cholesterol, high-density lipoprotein cholesterol, and total cholesterol are examples of lipid profile metrics, are widely used markers for assessing cardiovascular risk. Elevated levels of LDL-C, often referred to as "bad cholesterol," along with reduced levels of HDL-C, which is considered "good cholesterol," increase the likelihood of plaque formation in blood vessels, leading to atherosclerosis [1]. Triglycerides, another component of the lipid profile, are energy-storage molecules that, when excessively elevated, contribute to endothelial dysfunction and cardiovascular complications [1]. Thus, understanding how dietary fat influences these parameters is vital for addressing the growing prevalence of lipid-associated disorders.

Vegetable oils are a primary origin of dietary fat in many regions and vary significantly in their composition of saturated, monounsaturated, and polyunsaturated fatty acids. Oils such as crude palm oil (CPO), refined palm olein (RPO), groundnut oil (GO), and shea butter (SHB) are commonly consumed globally and have distinct effects on lipid metabolism. For example, studies have shown that palm oil, which is abundant in saturated fatty acids, can increase LDL-C levels, while oils high in monounsaturated fatty acids, such as groundnut oil, may have a more favorable impact on the lipid profile by reducing LDL-C and increasing HDL [1][3][4]. Shea butter, although less studied, contains stearic acid, a saturated fat that is believed to have a neutral impact on lipid profiles [5]. In cooking, these oils are frequently used, frying, and food processing, making their effects on health outcomes an important subject of investigation.

The connection between cardiovascular health and dietary fats extends beyond lipid metabolism. High-fat diets are also implicated in oxidative stress, where excess fat intake leads to disturbance of the equilibrium between pro-oxidants and antioxidants due to the generation of reactive oxygen species in the body [6][7]. This imbalance contributes to lipid peroxidation, cellular damage, and the progression of chronic diseases. Antioxidant defenses, including enzymes like superoxide dismutase and catalase, play a crucial role in mitigating oxidative damage, and their activity can be modulated by dietary fat composition [8][9]. Hence, evaluating the effect of different diets heavy in fat on these parameters is crucial for understanding their broader health implications.

Albino rats are frequently used as models in nutritional and biomedical research due to their metabolic and physiological similarities to humans [10][11]. This study investigates the effects of high-fat diets from commonly consumed vegetable oils—CPO, RPO, GO, and SHB—on the lipid profiles and cardiovascular risk indices of male albino rats. Soybean oil, a polyunsaturated fat source known for its cardioprotective properties, was used as the control diet. The lipid profile (TC, TG, LDL-C, and HDL-C) and calculated indices such as the atherogenic index (AI) and coronary risk index (CRI) were analyzed to evaluate the potential impact of these oils on cardiovascular health.

By comparing the lipid-modulating effects of these oils, this study aims to provide insights into how dietary fat type influences health outcomes, particularly regarding lipid metabolism and cardiovascular risk. The findings have important implications for dietary recommendations and public health policies aimed at decreasing the burden of lipid-associated diseases.

II. Material and Method

I. Reagent and Equipment

Among the reagents utilized were phosphate buffer (10 mM, pH 7.2), sodium hydroxide (NaOH), sodium chloride (NaCl), hydrochloric acid (HCl), monopotassium phosphate (KH_2PO_4), and dipotassium phosphate (K_2HPO_4), chloroform, and ethanol, were acquired from Sigma Aldrich and were of analytical quality, Johannesburg, South Africa. All kits for lipid profile assays were obtained from Randox Laboratories, Crumlin, United Kingdom. The equipment used included an oven (Genlab), water bath (Clifton water bath), analytical balance (Sartorius, Germany), pH meter (Hanna Instruments), magnetic stirrer (Genlab), beakers, conical flasks, syringes and needles (5 mL & 10 mL), EDTA bottles, plane bottles, 1X Phosphate-buffered saline (PBS), benchtop centrifuge (Pec Medical, USA), microcentrifuge, microcentrifuge tubes, fluorescence microplate reader (able to read excitation in the 530-570 nm range and emission in the 590-600 nm range), 50 μL to 300 μL adjustable multichannel micropipette with disposable tips, and 10 μL to 1000 μL adjustable single-channel micropipettes, 96-well microtiter plates, tubes for diluting glycerol standards and a plate reader with a 540 nm filter. The vegetable oils used in the study (soybean oil, crude palm oil, refined palm olein, groundnut oil, and shea butter) were purchased from commercial and local markets in Ekiti State, Nigeria.

II. Sample Preparation

The animals were starved for a whole night at the conclusion of the trial, and then anesthetized using chloroform. Blood samples were collected from the abdominal aorta using sterile syringes and transferred into EDTA tubes for plasma separation and plain tubes for serum extraction. The blood that was taken was centrifuged at 5000 g for 5 minutes at 4°C to separate serum and plasma. Organs, including the liver, kidney, heart, and brain, were removed and then cleaned with ice-cold PBS, dried off, and weighed.

III. Proximate Analysis

The experimental diets' proximate composition was ascertained using the established procedures of the Association of Official Analytical Chemists (AOAC) official methods of analysis, 20th edition, 2016. This included moisture content by oven drying, ash content by incineration in a muffle furnace, crude protein using the Kjeldahl method, crude fat by Soxhlet extraction, and carbohydrate by difference.

IV. Diet Formulation

Experimental diets were prepared by incorporating different vegetable oils (soybean oil, crude palm oil, refined palm olein, groundnut oil, and shea butter) into standard rat pellets to provide 55% of energy from fat for Groups 2-5, while the control group received a diet with 5% lipid from soybean oil (11% energy). A 2.5% soybean oil supplement was included to all high-fat meals to guarantee sufficient consumption of essential fatty acids. The formulated diets were produced every week to maintain freshness and stored in airtight containers to prevent rancidity.

Control Group: Diet that includes 5% fat, such as soybean oil, contributing 11% energy from fat.

CPO Group: High-fat (HF) diet contributing 55% of energy is generated by fat, formulated as 2.5% soybean oil + 30% crude palm oil (CPO).

RPO Group: High-fat (HF) diet contributing 55% of energy is generated by fat, formulated as 2.5% soybean oil + 30% refined palm olein (RPO).

GO Group: High-fat (HF) diet contributing 55% of energy is generated by fat, formulated as 2.5% soybean oil + 30% groundnut oil (GO).

SHB Group: High-fat (HF) diet contributing 55% of energy is generated by fat, formulated as 2.5% soybean oil + 30% shea butter (SHB).

V. Experimental Animal

Forty-eight (48) male albino rats with weights ranging from 100–150 g were employed. The animals were obtained from the animal colony of the Federal Polytechnic, Ado-Ekiti, Nigeria. Ethical approval was obtained from the Ethical Committee of Federal University Oye-Ekiti. The study was conducted in strict compliance with the institution's ethical guidelines for the care and use of laboratory animals. Additionally, all procedures adhered to internationally recognized ethical standards, including the Declaration of Helsinki and the ARRIVE guidelines. They were housed in the Experimental Animal Holding Facility, Department of Biochemistry, Federal University, Oye-Ekiti, under typical circumstances, which include a 12-hour light/dark cycle, a temperature of 22–25°C, and a relative humidity of 50–60%. Before starting the dietary manipulations, the rats were allowed unlimited access to water and regular pellet meal for a week to acclimate them.

VI. Preparation of Serum and Tissue Homogenate

The rats were anesthetized with chloroform after fasting for the entire duration of the experiment. The abdominal aorta was used to draw blood, which was then put into ordinary serum tubes. Prior to centrifugation at 5000 g for five minutes at 4°C, the blood samples in the plain tubes were left to clot at room temperature. After careful decantation, the resultant supernatant (serum) was kept at -20°C until biochemical assays were carried out.

After being meticulously removed, the liver, kidney, heart, and brain were rinsed twice with ice-cold phosphate-buffered saline (PBS, pH 7.4) and blotted dry with filter paper instead. To preserve enzyme activity and avoid protein denaturation, tissue samples were homogenized in ice-cold PBS using a mechanical homogenizer. After centrifuging the homogenates at 10,000 g for 15 minutes at 4°C, the supernatants were gathered for further biochemical tests, including evaluations of lipid and enzyme activity. The homogenates were stored at -20°C to preserve their integrity until further use.

VII. Biochemical Assays

Standard techniques were used to determine the lipid profile. Commercial assay kits from Randox Laboratories were used to measure serum total cholesterol, triglycerides, high-density lipoprotein cholesterol (HDL-C), and low-density lipoprotein cholesterol (LDL-C). The values of the lipid profile were used to compute the atherogenic and coronary risk indices. To guarantee the accuracy and precision of the findings, every analysis was performed in triplicate.

VIII. Statistical Analysis

Mean ± standard error of mean (SEM) was used to present the data. One-way analysis of variance was used to estimate group mean differences, and the Tukey test was used for all pairwise comparisons. Statistical significance is assumed at 95% confidence level. All statistics were carried out using GraphPad Prism version 6.

III. Results

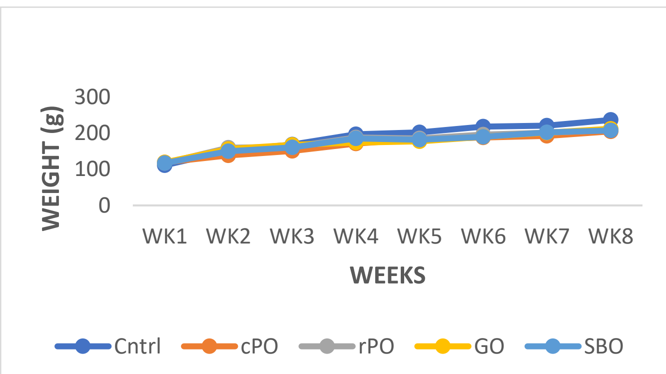

Fig. 1. Effect of different high-fat diet on changes in body weight (CPO, crude palm oil; RPO, refined palm olein; GO, groundnut oil; SHB, shea butter).

Throughout the study, no deleterious effects or mortality were observed in any of the experimental groups. Food intake remained consistent across all treatment groups. The impact of feeding different high-fat diets on body weight changes is illustrated in Fig. 1. Body weight increased steadily each week within each group. There were no significant differences in body weight changes among the high-fat diet groups compared to the control group fed with soybean oil.

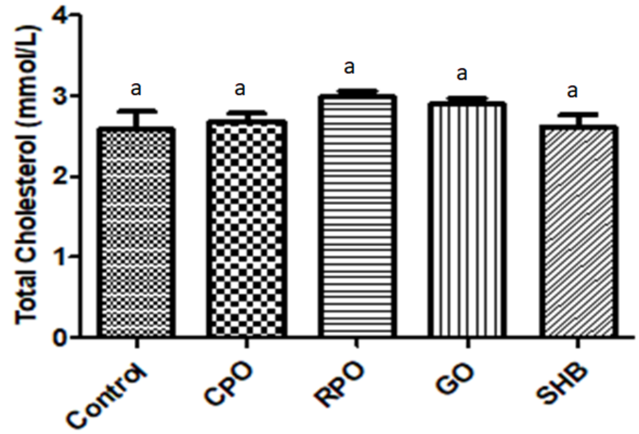

Fig. 2. Effect of different high-fat diet on total cholesterol (CPO, crude palm oil; RPO, refined palm olein; GO, groundnut oil; SHB, shea butter).

Fig. 2 illustrates total cholesterol levels (mmol/L) for groups subjected to various dietary treatments, including the control (soybean oil), crude palm oil (CPO), refined palm olein (RPO), groundnut oil (GO), and shea butter (SHB). Across all groups, there is no statistically significant difference in total cholesterol levels, as denoted by the shared label "a." This indicates that under the given experimental conditions, the different high-fat diets do not substantially alter total cholesterol levels when compared to the control. Despite minor numerical differences, none of the tested diets stand out as having a distinct impact on total cholesterol concentration.

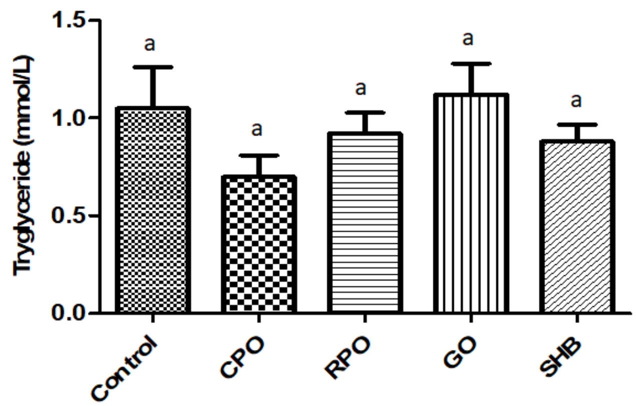

Fig. 3. Effect of different high-fat diet on triglycerides (CPO, crude palm oil; RPO, refined palm olein; GO, groundnut oil; SHB, shea butter).

The effect of feeding different high-fat diets on triglyceride levels is illustrated in Fig. 3. Lower triglyceride levels in the human body are associated with a reduced risk of cardiovascular and other severe diseases. In comparison to the control (soybean oil), crude palm oil (CPO) exhibited the lowest triglyceride concentration, followed by shea butter (SHB) and refined palm olein (RPO), all of which were lower than the control. Groundnut oil (GO), however, showed a higher concentration than the control. Although there were some variations, there was no significant difference in the effect of these high-fat diets on triglyceride levels when compared to the soybean oil control.

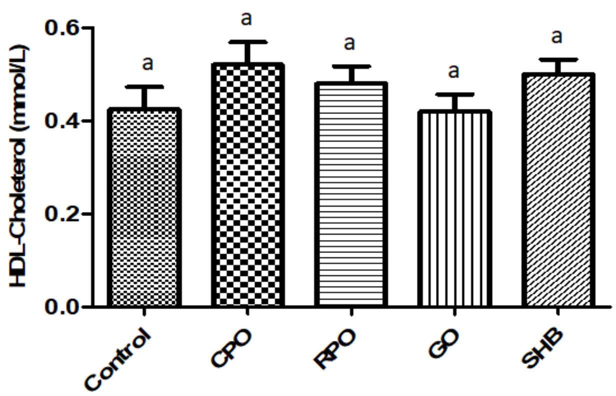

Fig. 4. Effects of *Waltheria indica* leaf diet on the liver lipid profile of experimental rats

The effect of different high-fat diets on high-density lipoprotein cholesterol (HDL-C) levels in the experimental groups, including the control (soybean oil), crude palm oil (CPO), refined palm olein (RPO), groundnut oil (GO), and shea butter (SHB) is illustrated in Fig. 4. Across all groups, HDL-C levels were not significantly different (denoted by the common letter "a"), indicating that feeding the rats with high-fat diets from different sources did not lead to substantial variations in HDL-C concentration when compared to the control. This suggests that the different vegetable oils have comparable impacts on maintaining HDL-C levels in the study model, which is considered a favorable component for cardiovascular health.

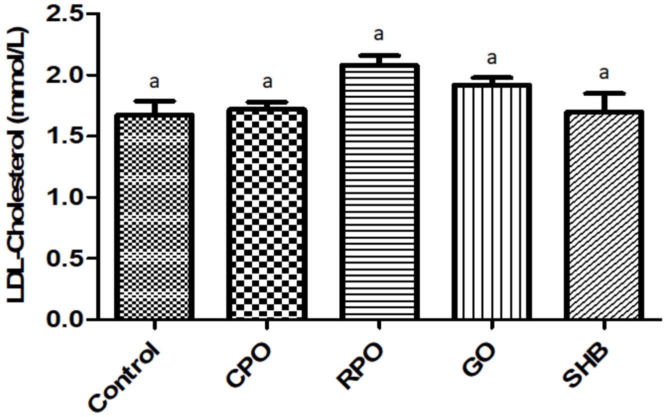

Fig. 5. Effect of different high-fat diet on low density lipoprotein cholesterol (CPO, crude palm oil; RPO, refined palm olein; GO, groundnut oil; SHB, shea butter).

The graph (Fig. 5) shows the levels of low-density lipoprotein cholesterol (LDL-C) across the groups fed different high-fat diets, including the control (soybean oil), crude palm oil (CPO), refined palm olein (RPO), groundnut oil (GO), and shea butter (SHB). No significant differences in LDL-C levels were observed among the groups (as denoted by the common letter "a"), indicating that these high-fat diets did not lead to significant changes in LDL-C concentrations compared to the control group. This suggests that the consumption of these fats, have comparable effects on LDL-C levels, which is typically associated with cardiovascular risk when elevated.

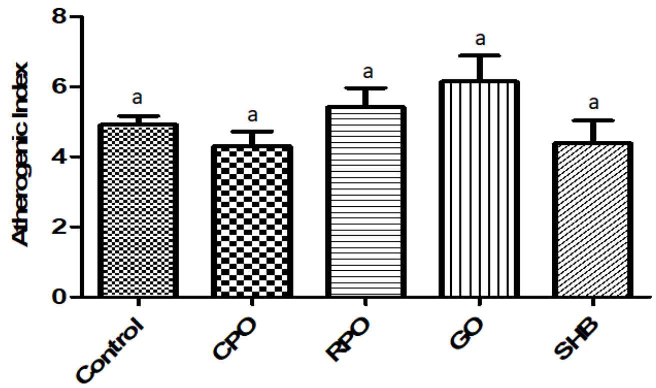

Fig. 6. Effect of different high-fat diet on atherogenic index (CPO, crude palm oil; RPO, refined palm olein; GO, groundnut oil; SHB, shea butter).

The graph (Fig. 6) presents the atherogenic index values for groups fed different high-fat diets: the control (soybean oil), crude palm oil (CPO), refined palm olein (RPO), groundnut oil (GO), and shea butter (SHB). There was no significant difference in the atherogenic index across all the groups, as indicated by the shared letter "a". This suggests that none of the diets produced a marked increase or decrease in the atherogenic index compared to the control group. The similar values across these groups imply that the high-fat diets tested do not substantially influence the overall risk of cardiovascular disease based on this index, under the conditions of this study.

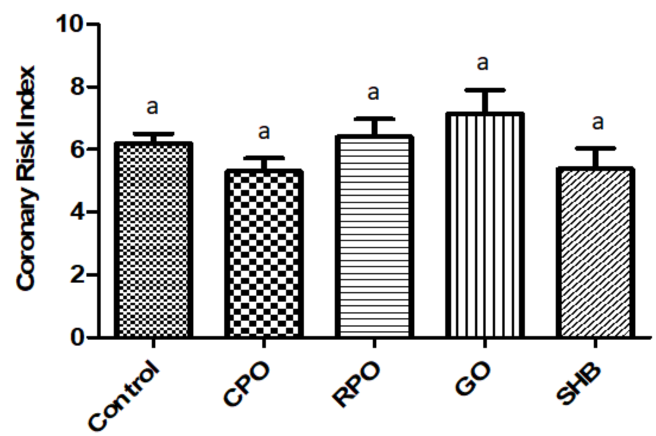

Fig. 7. Effect of different high-fat diet on coronary risk index (CPO, crude palm oil; RPO, refined palm olein; GO, groundnut oil; SHB, shea butter).

The graph (Fig. 7) illustrates the coronary risk index values for groups consuming different high-fat diets, including the control (soybean oil), crude palm oil (CPO), refined palm olein (RPO), groundnut oil (GO), and shea butter (SHB). The index values show no significant differences across all groups, as evidenced by the shared designation "a" above each bar. This suggests that the various high-fat diets tested do not exhibit significant impacts on coronary risk compared to the control diet under the given experimental conditions. While slight variations exist, no diet appears to pose an elevated risk or confer substantial protection in terms of the coronary risk index.

IV. Results and Discussions

I. EFFECT OF HIGH-FAT DIETS ON BODY WEIGHT CHANGES

The impact of giving albino rats various high-fat diets over the course of eight weeks is seen in Fig. 1. Body weight increased gradually in all groups, with no discernible differences between the high-fat diet group and the soybean oil-consuming control group. This suggests that, given the circumstances of this study, the type of dietary fat may not have a significant impact on weight gain. Consistent food intake across all groups further supports the conclusion that the observed weight changes were not driven by differences in appetite or consumption patterns.

Previous studies have reported mixed outcomes on the effect of high-fat diets on weight. For instance, Patel *et al.* [12] found that diets rich in saturated fats tend to promote greater weight gain compared to unsaturated fats. However, our findings align with research by Shiraev *et al*. [13], who demonstrated that variations in dietary fat sources might have minimal impact on body weight if caloric intake is maintained. This suggests that calorie content, rather than fat type, may play a more prominent role in influencing body weight.

The steady weight gain observed in this study highlights the potential risks associated with prolonged high-fat diet consumption, irrespective of fat type. While no deleterious effects were recorded during the experimental period, sustained weight gain can predispose individuals to obesity and metabolic syndrome over time. These results highlight the significance of calorie moderation and dietary balance in preventing weight-related health issues.

II. TOTAL CHOLESTEROL LEVELS

Fig. 2 presents the effect of various high-fat diets on total cholesterol levels. Crude palm oil (CPO) and shea butter (SHB) exhibited cholesterol levels comparable to the control group (soybean oil), while refined palm olein (RPO) and groundnut oil (GO) showed slightly higher levels. Despite these variations, no statistically significant differences were observed across the groups. This suggests that the tested high-fat diets do not significantly elevate total cholesterol within the experimental timeframe.

Similar results have been published by Yu *et al*. [14], who observed that unrefined fats like crude palm oil tend to have a neutral effect on cholesterol due to their bioactive compounds, such as tocotrienols. In contrast, diets high in refined fats or saturated fatty acids, like RPO, may slightly elevate cholesterol levels [15]. However, the absence of significant differences in our study could reflect species-specific metabolic responses or the duration of dietary exposure.

Maintaining healthy total cholesterol levels is critical for cardiovascular health. The findings indicate that certain fats, like CPO and SHB, may be better dietary options for individuals at risk of hypercholesterolemia. Nonetheless, caution is warranted with fats like RPO, which could potentially elevate cholesterol levels if consumed over extended periods.

III. TRIGLYCERIDE LEVELS

Fig. 3 shows the effect of different high-fat diets on triglyceride levels. CPO and SHB resulted in the lowest triglyceride concentrations, followed by RPO, with GO showing the highest levels among the test groups. However, these differences were not statistically significant compared to the control. Lower triglyceride levels in CPO and SHB diets may be attributed to their bioactive components, which are known to modulate lipid metabolism.

Research by Ash *et al.* [16], Scaccini *et al*. [17], and Sampurna *et al*. [18] support this observation, noting that unrefined oils with high antioxidant content can help lower triglyceride levels. Conversely, oils like groundnut oil, which are rich in monounsaturated fats, have been associated with higher triglyceride levels in some circumstances [19]. The lack of significant differences in this study might reflect a complex interplay of dietary composition and metabolic adaptability.

Triglycerides are a key marker of cardiovascular health, with elevated levels linked to a higher chance of heart disease. The trends observed in this study suggest that certain fats, particularly unrefined ones, might offer protective effects by minimizing triglyceride accumulation. This underscores the potential benefits of incorporating such fats into heart-healthy diets.

IV. LOW-DENSITY LIPOPROTEIN CHOLESTEROL (LDL-C)

Fig. 4 depicts the impact of different high-fat diets on LDL-C levels. RPO exhibited the highest LDL-C levels, while CPO and SHB showed the lowest, comparable to the control. Elevated LDL-C, often referred to as "bad cholesterol," is a substantial risk factor for cardiovascular disease and atherosclerosis. The relatively lower LDL-C levels in CPO and SHB diets may reflect the influence of their unsaponifiable fractions, such as tocopherols and phytosterols.

Studies by Brousseau *et al*. [20] corroborate these findings, highlighting the role of dietary fats rich in bioactive compounds in lowering LDL-C levels. On the other hand, refined fats like RPO, which lack these beneficial components, have been shown to increase LDL-C concentrations [21]. These differences underscore the importance of fat processing methods and composition in determining lipid outcomes.

Lower LDL-C levels are critical for reducing cardiovascular risk. These findings reinforce dietary recommendations to prioritize unrefined fats over refined ones to minimize the adverse effects on LDL-C and associated health risks.

V. HIGH-DENSITY LIPOPROTEIN CHOLESTEROL (HDL-C)

The effect of different high-fat diets on HDL-C levels is shown in Fig. 5. CPO led to the highest HDL-C levels, followed by SHB, RPO, and GO. HDL-C, commonly known as "good cholesterol," plays a protective role in cardiovascular health by facilitating cholesterol transport back to the liver for excretion. The variations observed suggest that certain fats may enhance HDL-C levels more effectively.

Research by Basu *et al*. [22] aligns with these findings, indicating that plant-based oils with high unsaturated fat content can significantly elevate HDL-C levels. The role of bioactive compounds in unrefined fats, such as tocotrienols in

CPO, has been particularly noted for its HDL-C-boosting properties. These trends highlight the potential of specific dietary fats in improving lipid profiles and reducing cardiovascular risk.

Higher HDL-C levels are associated with reduced risk of coronary artery disease. The findings suggest that incorporating fats like CPO and SHB into the diet could have beneficial effects on cardiovascular health, emphasizing the role of fat type in dietary planning.

VI. ATHEROGENIC INDEX (AI) AND CORONARY RISK INDEX (CRI)

The impact of high-fat meals on the coronary risk index (CRI) and atherogenic index (AI) is seen in Figs. 6 and 7, respectively. CPO and SHB exhibited the lowest values for both indices, while RPO and GO were higher but not significantly different from the control. Lower indices indicate reduced cardiovascular risk and suggest that unrefined fats may provide cardioprotective benefits.

Similar trends have been reported by Ajebli *et al*. [23], who found that unrefined plant-based fats lower AI and CRI due to their favorable lipid-modulating effects. Conversely, refined fats tend to elevate these indices, contributing to increased cardiovascular risk. The neutral impact observed in this study may reflect the short duration of dietary exposure.

AI and CRI are critical markers of cardiovascular risk, with lower values indicating better heart health. These findings reinforce the importance of dietary fat selection in mitigating cardiovascular disease, emphasizing the benefits of unrefined over refined fats.

VII. CONCLUSION

This study concludes by highlighting the intricate interactions between dietary fat content and metabolic health outcomes, as well as the varying effects of diverse high-fat diets on body weight, lipid profiles, and cardiovascular risk markers. The results indicate that while all tested fats led to weight gain, crude palm oil and shea butter showed relatively favorable effects on lipid profiles, potentially due to their bioactive components. Conversely, refined palm olein posed a higher risk for elevated LDL-C and cardiovascular indices. These findings underscore the importance of dietary fat selection, not merely for weight management but also for mitigating cardiovascular risk. This evidence supports targeted dietary recommendations prioritizing fats with healthier profiles and calls for further research into their long-term effects on human health to guide public health strategies and individualized nutrition planning.

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