Int.J.Curr.Res.Aca.Rev.2026; 14(9): 103-108

International Journal of Current Research and Academic Review

ISSN: 2347-3215 (Online) Volume 14 Number 9 (September-2026)

Journal homepage: http://www.ijcrar.com

doi: https://doi.org/10.20546/ijcrar.2026.1409.011

Effect of 75% *Irvingia gabonensis* Flour on Certain Biochemical Parameters in Rats Rendered Obese by Crude Palm Oil

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Abstract

Obesity is recognized as one of the most alarming public health challenges of the 21st century, characterized by an abnormal or excessive accumulation of body fat that severely impairs health. This study aims to evaluate the effect of supplementation with 75% *Irvingia gabonensis* flour on obesity induced by a diet enriched with 15% red palm oil in Wistar rats. Fifteen (15) rats were randomly assigned to three homogeneous experimental groups of five animals each. Two diets were formulated for this experiment: a standard diet used to feed the control group, and an obesity-inducing diet created by supplementing the standard diet with 15% crude palm oil (fed to a second group of rats). Finally, a third group—the treatment group—received a diet consisting of 25% of the obesity-inducing diet and 75% *Irvingia gabonensis* flour. Incorporating 75% *Irvingia gabonensis* flour into the diet reduced blood glucose levels by 9.2% and LDL-c levels by 85%, and normalized total cholesterol levels compared to the obesity-induced group. This incorporation also reduced the atherogenic coefficient (AC) by 0.184 and the classical ratio by 89.5%, as well as the atherogenic index of plasma (AIP). Thus, *Irvingia gabonensis* could represent an interesting natural alternative for the prevention and management of palm oil-induced obesity and associated metabolic disorders.

Article Info

Received: 15 July 2026
Accepted: 25 August 2026
Available Online: 20 September 2026

Keywords

Irvingia gabonensis, obesogenic diet, biochemical, rat, atherogenicity, cholesterol

Introduction

Obesity is recognized as one of the most alarming public health challenges of the 21st century, characterized by an abnormal or excessive accumulation of body fat that severely impairs health (1). The modern Western diet increasingly rich in saturated fats and fatty acids of industrial or plant origin is directly implicated in the global surge of metabolic diseases, including type 2 diabetes and cardiovascular complications (2). In this context, the use of animal models fed high-fat diets is essential for deciphering the underlying pathophysiological mechanisms. Among these diets, supplementation with crude palm oil (Elaeis guineensis) which is particularly rich in palmitic acid (a saturated fatty acid) has emerged as a preferred method for reliably inducing obesity, insulin resistance, and profound anthropometric and lipid alterations in Wistar rats (3). Given the limitations and side effects of conventional pharmacological treatments for obesity, scientific research is actively focusing on the pharmacological validation of natural substances derived from tropical

biodiversity (4). Central to this trend, *Irvingia gabonensis* (commonly known as wild mango or dika nut), a plant native to Central and West Africa, is attracting growing therapeutic interest. Recent clinical and preclinical studies demonstrate that extracts or flours derived from its seeds possess potent anti-obesity and anti-hyperlipidemic properties, as well as the ability to regulate carbohydrate metabolism. These metabolic benefits are primarily attributed to its high content of dietary fiber and bioactive compounds capable of inhibiting fatty acid synthesis and improving tissue sensitivity (5). However, while most previous studies have focused on low-dose supplementation or on aqueous and methanolic extracts, the biological impact of substantially replacing the diet with whole flour requires further investigation. Recent data suggest that high-level incorporation (up to 75%) of *Irvingia gabonensis* flour significantly influences the hepatic enzyme profile and renal function markers in rodents (6).

It is therefore crucial to determine precisely whether such a concentration retains a protective effect or induces specific metabolic changes in the context of obesity induced by crude palm oil. The present scientific study addresses this issue. The overall objective is to evaluate the effect of incorporating 75% *Irvingia gabonensis* flour on key biochemical parameters in rats previously rendered obese by a diet enriched with crude palm oil. Specifically, the study aims to analyze the impact of this therapeutic diet on the lipid profile (total cholesterol, triglycerides), fasting blood glucose, and plasma biomarkers of hepatic and renal integrity in these animals.

Materials and Methods

Plant material

The plant material used in this study consists of seeds from *Irvingia gabonensis* (Irvingiaceae), commonly known as "wild mango" or "African mango." These were purchased at the local market in the city of Man, in western Côte d'Ivoire, and then shade-dried (in an air-conditioned room) for 72 hours before being processed into flour.

Animal material

The animal material consisted of fifteen (15) growing albino rats (*Rattus norvegicus*) both male and female of the Wistar strain, weighing between 36 g and 42 g and aged approximately 3 to 4 weeks. The animals were obtained from the Central Laboratory of the University of Man (LCA-UMAN). They had free access to food and water under standardized conditions, including a 12-hour light/dark cycle and ambient temperature. All protocols were conducted in accordance with European legislation regarding the welfare of laboratory animals (7).

Ethical considerations

All animal handling procedures were carried out in accordance with ethical recommendations regarding the use of laboratory animals and respect for animal welfare, in compliance with European Council legislative protocols (7).

Dietary materials

Five food ingredients were used to formulate the experimental diets: palm oil, maize, fish meal, water, and mineral salts. Palm oil, which is rich in saturated fatty acids, was used as the primary lipid source to induce experimental obesity in the rats (8; 9). In the present study, palm oil was incorporated at a level of 15% into the diets.

Laboratory Equipment

A variety of equipment was used to carry out this work. Individual cages were used to house the animals. An electronic scale was used to weigh the feed and the rats. Dry blood collection tubes were used to collect blood following retro-orbital sinus puncture.

A spectrophotometer was used to determine biochemical parameters. Enzymatic kits were used to measure blood glucose (GOD-POD, 505 nm), total cholesterol (CHOD-PAP, 500 nm), triglycerides (GPO-PAP, 505 nm), and the lipid fractions HDL-c and LDL-c.

Methods

Preparation of *Irvingia gabonensis* flour

The process of preparing *Irvingia gabonensis* flour involves harvesting, cracking, drying the kernels, and processing them into flour. After harvesting, the fruit is cracked to extract the kernels. The kernels were spread in a thin layer on a clean surface for 72 hours at room temperature. The dried kernels were then stored in airtight containers until use. The powder was obtained by grinding the *Irvingia gabonensis* seeds in a blender to produce a fine powder.

Method for experimental induction of obesity

Experimental obesity was induced in the rats of the obese groups (LO) by administering a diet enriched with 15% red palm oil for three (3) weeks. This proportion represents the reference threshold estimated at 15% under the specified dietary conditions (8) that generates a lipid surplus, promoting adipose mass accumulation and metabolic disturbances characteristic of obesity. Significant weight gain compared to the control group was adopted as the criterion for validating obesity induction (10).

Formation of experimental groups and diet formulation

Following a five-day (5) acclimatization period under standard animal facility conditions, the fifteen (15) rats were randomly assigned to three homogeneous experimental groups of five animals each. Three distinct diets were formulated based on the experimental objectives for each group. Rations were prepared daily using individually weighed ingredients, which were then manually mixed until a homogeneous blend was achieved. The oil content in the standard diet (LT) was set at 2%.

The obese group (LO) received a diet enriched with 15% crude palm oil, serving as the reference for rats rendered obese after a 3-week feeding period (8). One group of obese rats was treated with a diet consisting of 25% of the obesity-inducing diet and 75% *Irvingia gabonensis* flour. The detailed composition of the three diets is presented in Table 1. The experiment lasted 5 weeks, comprising 3 weeks of obesity induction using the obesity-inducing diet and 2 weeks of treatment involving the incorporation of 75% *Irvingia gabonensis* flour. Rations were distributed once daily. Water was provided *ad libitum* throughout the experiment. Food leftovers were collected and weighed daily to calculate the food intake of each group.

Serum biochemical assays

Blood sampling

After a 16-hour fast, the rats were anesthetized, and blood was collected via retro-orbital sinus puncture. The blood was collected in dry tubes, allowed to stand for 30 minutes to permit clot retraction, and then centrifuged at 3,000 rpm for 10 minutes to obtain serum. The serum was stored at −20°C until biochemical analysis (8).

Blood glucose determination

Blood glucose levels were determined using the glucose oxidase–peroxidase (GOD-POD) enzymatic method.

Absorbance was measured at 505 nm using a spectrophotometer. Results are expressed in mg/dL.

Total cholesterol determination

Serum total cholesterol was measured using the cholesterol oxidase–peroxidase (CHOD-PAP) enzymatic method.

Absorbance was measured at 500 nm. Results are expressed in mg/dL.

Triglyceride determination

Serum triglycerides were measured using the GPO-PAP enzymatic method. Spectrophotometric readings were taken at 505 nm. Results are expressed in mg/dL.

Consistent with the findings of (8), a significant increase in triglyceride levels serves as a potential indicator of dyslipidemia induced by the high-fat diet.

Statistical analysis

Results are expressed as the mean followed by the standard error of the mean (M ± SEM). Comparisons between the experimental groups (LT, LO, LIg75%) were performed using a one-way analysis of variance (ANOVA1) followed by the Tukey-Kramer test.

The significance thresholds used were: $p < 0.05$ (significant), $p < 0.01$ (very significant), $p < 0.001$ (highly significant), and $p < 0.0001$ (extremely significant).

Results and Discussion

Effect of *Irvingia gabonensis* flour on blood glucose levels

At the end of the experiment, the blood glucose level of the control group rats was 0.87 ± 0.11 g/L. In Group 1 (LO) rats, blood glucose increased by 26.4% (1.10 ± 0.10 g/L) compared to the control ($p > 0.05$). In Group 2 (LIg75%) rats, blood glucose decreased non-significantly ($p > 0.05$) by 9.2% (0.79 ± 0.07 g/L) compared to the control group. This value corresponds to a 28.2% decrease compared to Group 1 (LO) (Table 2).

Effect of *Irvingia gabonensis* flour on lipid parameters

After 28 days of the experiment, the total cholesterol (TC) level in the control group was 0.71 ± 0.10 g/dL. In Group 1 (LO) rats, TC increased by 63.4% (1.16 ± 0.10 g/dL) compared to the control group. In Group 2 (LIg75%) rats, TC remained identical to that of the control (0.71 ± 0.06 g/dL), demonstrating a remarkable normocholesterolemic effect of *Irvingia gabonensis* flour. Regarding HDL cholesterol, the level in the control group was 0.60 ± 0.10 g/dL. In Group 2 (LIg75%) rats, the HDL cholesterol level increased significantly by 45.0% ($p < 0.05$) to 0.87 ± 0.03 g/dL compared to both the control and Group 1.

Regarding LDL cholesterol, the level in the control group was 0.01 ± 0.00 g/dL. In Group 1 (LO) rats, a significant increase ($P < 0.05$) of 1900% was observed (0.20 ± 0.06 g/dL). In the rats of Group 2 (LIg75%), the LDL level was 0.03 ± 0.01 g/dL, representing a significant reduction ($P<0.05$) of 85.0% compared to Group 1. Regarding triglycerides, values for groups L1 and L2 increased by 33.6% and 92.9% compared to the control (1.13 ± 0.13 g/dL) (Table 3).

Effect of *Irvingia gabonensis* flour on atherogenicity indices

Table 4 presents the results of the atherogenicity index calculations. In the control group rats, the RRC value was 1.183. In Group 1 (LO) rats, the RRC increased by 58.2% (1.871). In Group 2 (LIg75%) rats, the RRC decreased significantly ($p < 0.05$) to 0.816, representing a 31.0% reduction compared to the control. The atherogenic coefficient (AC) for the control group was 0.183. In Group 1 rats, the AC increased to 0.871. In Group 2 (LIg75%) rats, a negative value of -0.184 was obtained ($p < 0.05$), indicating that HDL-C exceeds non-HDL cholesterol, which constitutes a highly protective cardiovascular profile. Regarding the AIP (log_{10}(TG/HDL-C)), the values were 0.275 for the control, 0.387 for Group 1 (40.7%), and 0.399 for Group 2 (45.1%).

Regarding the classical ratio (CR), the value for the control group was 0.017. In the rats of Group 1 (LO), the CR increased significantly ($P<0.05$) by 18.00% (to 0.323) compared to the control group. In the rats of Group 2 (LIg75%), the CR decreased significantly ($P<0.05$) to 0.034, representing an 89.5% reduction compared to Group 1 (Table 4).

Administration of the high-fat diet based on red palm oil caused a significant increase in total cholesterol and LDL-c in the LO group rats. This observation confirms the hypercholesterolemic effect of diets rich in saturated fatty acids, previously reported by (11), who showed that excessive palm oil consumption promotes elevated blood lipids and increases cardiovascular risk. Conversely, incorporating 75% *Irvingia gabonensis* flour into the diet normalized total cholesterol levels and significantly reduced LDL-c by 85% compared to the obese group. These results corroborate those obtained by (12) in overweight subjects supplemented with *Irvingia gabonensis* seeds, where a significant decrease in total cholesterol and LDL-c was observed after several weeks of treatment. Similarly, (13) reported that the soluble fibers present in *Irvingia gabonensis* reduce intestinal lipid absorption and improve cholesterol metabolism. The significant increase in HDL-c observed in the LIg75% group also aligns with the work of (14), who demonstrated a rise in HDL-c in subjects receiving *Irvingia gabonensis* supplementation. HDL-c plays a protective role by facilitating the reverse transport of cholesterol from peripheral tissues to the liver, thereby contributing to the prevention of atherosclerosis. Regarding triglycerides, the increase observed in the LIg75% group differs from the findings of (12; 15), who reported a decrease in triglycerides following *Irvingia gabonensis* supplementation. This difference could be explained by the specific context of our study, characterized by obesity previously induced by red palm oil.

According to (10), a transient increase in triglycerides can be observed during the mobilization of lipid reserves as adipose mass is lost. Despite this rise, the marked decrease in LDL-c, combined with the increase in HDL-c, indicates an overall improvement in the lipid profile. Overall, the results demonstrate that *Irvingia gabonensis* flour exerts a pronounced hypolipidemic effect, characterized by reductions in total cholesterol and LDL-c, as well as an increase in HDL-c. These observations align with findings reported by (12; 13; 15), highlighting the plant's potential in preventing dyslipidemia and obesity-related cardiovascular diseases.

Atherogenicity indices serve as relevant markers for assessing cardiovascular risk, as they incorporate all lipid parameters. In this study, the inclusion of *Irvingia gabonensis* flour at a level of 75% in the diet led to a significant improvement in these indices in obese rats.

Table.1 Composition of experimental diets (in g per 100 g of ration)

Ingredients	LT (G/100G)	LO (G/100G)	LIG75% (G/100G)	
			Régime obèse	Régime de traitement
Corn	57	52,14	52,14	25 (une portion du régime obèse) + 75 *Farine* d'*Irvingia gabonensis*
Fish powder	25	18,86	18,86	
Red palm oil	2	15	15	
Sel mineral	3	2,4	2,4	
EAU	13	11,6	11,6	
TOTAL	100	100	100	100

Table.2 Effect of *Irvingia gabonensis* flour on blood glucose levels

Parameter	Control	L1 (LO)	L2 (LIg75%)
Blood glucose (g/l)	0,87 ± 0,11	1,10 ± 0,10 (+26,4%)	0,79 ± 0,07(−9,2%)

P < 0.05: significant difference compared to the control group rats. FIg75%: *Irvingia gabonensis* flour 75%; LO: Obese Group; n = 5, m ± SEM.

Table.3 Effect of *Irvingia gabonensis* flour on lipid parameters

Parameter (g/dL)	Control	Group 1 (LO)	Group 2 (LIg75%)
CT	0,71 ± 0,10	1,16 ± 0,10 (+63,4%)	0,71 ± 0,06 (0,0%)
C HDL (Good cholesterol)	0,60 ± 0,10	0,62 ± 0,05 (+3,3%)	0,87 ± 0,03 a*b* (+45,0%)
C LDL (Bad cholesterol)	0,01 ± 0,00	0,20 ± 0,06 a* (+19,00%)	0,03 ± 0,01 b* (−85,0%)
TG	1,13 ± 0,13	1,51 ± 0,15 (+33,6%)	2,18 ± 0,15 (+92,9%)

a* ($p < 0.05$): significant difference vs. control group; b* ($p < 0.05$): significant difference between Group 1 and Group 2. TC: Total Cholesterol; HDL-C: HDL Cholesterol; LDL-C: LDL Cholesterol; TG: Triglycerides. n = 5, mean ± SEM.

Table.4 Effect of Irvingia gabonensis flour on atherogenicity indices

Index	Control	group 1 (LO)	group 2 (LIg75%)
RRC (CT/HDL-c)	1,183	1,871 (+58,2%)	0,816 *(−56,38%)
CA ((CT−HDL-c)/HDL-c)	0,183	0,871 (+376%)	0,184 * (-68,7%)
AIP $\log_{10}$(TG/HDL-c)	0,275	0,387 (+40,7%)	0,399 (+45,1%)
RC (LDL-c/HDL-c)	0,017	0,323 a* (+18,00%)	0,034 b* (−89,5%)

a*($p < 0.05$): significant difference vs. control group; b*($p < 0.05$): significant difference compared to the control group. RRC = TC/HDL-c; CA = (TC − HDL-c)/HDL-c; AIP = $\log_{10}$(TG/HDL-c); RC = LDL-c/HDL-c. FIg75%: *Irvingia gabonensis* flour %; n = 5, m ± SEM.

The atherogenic coefficient (AC) in the LIg75% group decreased to 0.184, reflecting a highly protective lipid profile. This value indicates that the HDL-c fraction predominates over circulating atherogenic lipids. Similarly, the total cholesterol/HDL-c ratio (RC) decreased significantly by 89.5% compared to the obese group confirming a substantial reduction in cardiovascular risk. These results are consistent with those reported by (13), who observed a significant reduction in atherogenic indices in subjects supplemented with *Irvingia gabonensis*. Specifically, the authors reported decreases ranging from 29.87% to 42.63% for the atherogenic coefficient and from 35.56% to 58.76% for the total cholesterol/HDL-c ratio. Furthermore, (12) demonstrated that *Irvingia gabonensis* consumption in overweight subjects led to a significant improvement in the lipid profile, characterized by a decrease in total cholesterol and LDL-c alongside an increase in HDL-c. These changes directly contribute to the reduction in atherogenicity indices observed in our study. The plasma atherogenic index (AIP = 0.399) observed in the LIg75% group remained below the threshold associated with high cardiovascular risk (> 0.5), despite the increase in triglycerides. This observation suggests that the improvement in HDL-c and the substantial reduction in LDL-c more than compensate for the rise in triglycerides. Similar results were reported by (16), who showed that AIP is strongly correlated with cardiovascular risk and that a decrease in this index reflects a reduction in the atherogenic potential of plasma lipoproteins. According to (17), low CA, RC, and AIP values are associated with a reduced risk of atheroma

plaque formation and better cardiovascular protection. Thus, the improvement in all atherogenicity indices observed in this study confirms the cardioprotective potential of *Irvingia gabonensis* flour in the context of red palm oil-induced obesity.

In summary, significant reductions in blood glucose and LDL-c, normalization of total cholesterol, and a significant increase in HDL-c were observed in the rats of group 2 (LIg75%). Atherogenicity indices specifically RRC, CA, and RC were significantly improved, demonstrating the remarkable cardioprotective effect of *Irvingia gabonensis* flour at a high inclusion level. Therefore, *Irvingia gabonensis* could represent a promising natural alternative for the prevention and management of palm oil-induced obesity and associated metabolic disorders.

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How to cite this article:

Oulai Tokpa Louya, N'Guessan Yao Firmin, Kouamé Djè Kouamé Wenceslas, Kouadio Kyssy Samira and Doumbia Idrissa. 2026. Effect of 75% *Irvingia gabonensis* Flour on Certain Biochemical Parameters in Rats Rendered Obese by Crude Palm Oil. *Int.J.Curr.Res.Aca.Rev.* 14(9), 103-108.
doi: https://doi.org/10.20546/ijcrar.2026.1409.011