Investigational
Medicinal Chemistry & Pharmacology

Research Article Open Access

Effect of a phytochemical-enriched functional food formulation from *Ipomoea batatas*, *Daucus carota* subsp. sativus, and *Gnetum africanum* on metabolic syndrome in Wistar Rats

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Abstract

Background: Metabolic syndrome is a major public health concern, often exacerbated by patients' difficulty in adhering to restrictive diets. This study proposes an innovative dietary approach by developing low-glycemic-index biscuits specifically formulated to support the management of this condition.

Methods: The phytochemical-enriched formulations were produced using composite flours of sweet potato (*Ipomoea batatas* (L.)) (60 - 65%), carrot (*Daucus carota* subsp. *sativus*) (15 - 30%), and *Gnetum africanum* Welw. (okok) leaves (10 - 20%), with proportions optimized using Minitab software. Among nine initial formulations, including a wheat-based control formulation, one was selected based on its superior nutritional and sensory properties. The biological efficacy of formulations was evaluated *in vivo* by feeding *Wistar* rats with biscuits for 28 days. Parameters associated with metabolic syndrome were assessed.

Results: It appears from the study that revealed that our formulation had significantly higher contents of ash, fiber, and lipids compared to the control formulation. The same formulation also presents significant health benefits, even if not preferred in sensory evaluation. Supplementation of that flour at 10% and 20% in the diet was able to significantly reduce food intake ($p < 0.05$), lower LDL cholesterol, and increase HDL cholesterol levels, and significantly decrease triglyceride levels. Additionally, an anti-inflammatory effect has been observed. Both doses significantly reduced postprandial glycemic peaks.

Conclusion: These findings suggest that the formulated biscuits have strong potential for consumer acceptance and provide high energy value. Their nutritional composition and observed biological effects highlight their potential as a functional dietary strategy for the management of metabolic syndrome.

Keywords: *Daucus carota* subsp *Sativus*; Formulation; Functional food; *Gnetum africanum; Ipomoea batatas* and Metabolic syndrome.

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Citation on this article: Teugwa Mofor C; Koumagueyeng A, Zofou D, Tekou FA, Dzoyem JP. Effect of a phytochemical-enriched functional food formulation from Ipomoea batatas, Daucus carota subsp. sativus, and Gnetum africanum on metabolic syndrome in Wistar Rats. Investigational Medicinal Chemistry and Pharmacology (2026) 9(2):138; Doi: https://dx.doi.org/10.31183/imcp.2026.00138

Invest. Med. Chem. Pharmacol. (IMCP) ISSN: 2617-0019 (Print)/ 2617-0027 (Online); *© The Author(s). 2026 Open Access This article is available at https://investchempharma.com/*

Background

Globalization has profoundly transformed lifestyles worldwide, particularly dietary habits, which play a crucial role in the prevention and development of various chronic diseases. Among these, metabolic syndrome (MetS), also referred to as "syndrome X," represents a cluster of interrelated metabolic abnormalities, including abdominal obesity, dyslipidemia, type 2 diabetes, and cardiovascular diseases [1]. This condition is increasingly prevalent, and its incidence rises with age [2]. Several organizations, including the World Health Organization (WHO), the International Diabetes Federation (IDF), and the National Cholesterol Education Program Adult Treatment Panel III (NCEP ATP III), have proposed diagnostic criteria for MetS [3]. According to the IDF [4]. MetS is defined by central obesity (waist circumference ≥94 cm in men and ≥80 cm in women), combined with at least two of the following factors: fasting plasma glucose ≥100 mg/dL or type 2 diabetes, blood pressure ≥130/85 mmHg, triglycerides ≥150 mg/dL, and HDL cholesterol ≤40 mg/dL in men or ≤50 mg/dL in women. The global prevalence of metabolic syndrome has reached alarming levels. It is estimated to affect up to 86.3% of certain populations [5], with substantial regional variations: approximately 86% in the United States [6], 39.6% to 80% in Europe 57.7% to 91.9% in Asia [7], and 60.4% to 71.7% in Sub-Saharan Africa [8]. The prevalence is particularly high among individuals with diabetes [9]. In Cameroon, a study conducted in Yaoundé in 2018 reported that the most affected age group (30.22%) was between 25 and 34 years, with a higher prevalence among women [10]. This rising trend, especially in developing countries, is largely attributed to sedentary lifestyles and poor dietary habits [11]. These findings highlight the urgent need to develop innovative nutritional strategies for the prevention and management of metabolic syndrome. In this context, the development of functional foods based on natural ingredients rich in bioactive compounds represents a promising approach. For instance, the incorporation of sweet potato flour into bakery products has been shown to improve nutritional quality, health benefits, and consumer acceptability [12]. Sweet potatoes are rich in phenolic acids, flavonols, flavanones, and anthocyanidins, which contribute to their antidiabetic effects through multi-target mechanisms [13]. Similarly, formulations enriched with carrot flour (up to 15%) exhibit increased dietary fiber content [14]. The combination of sweet potato, carrot, and okok (*Gnetum africanum* Welw.) flours is particularly relevant due to their complementary nutritional profiles and potential synergistic bioactive properties. Given the increasing prevalence of metabolic syndrome and the challenges associated with adherence to restrictive diets, this study proposes an innovative dietary alternative based on functional biscuits with a low glycemic index. This work focuses on evaluating the potential effects of biscuits formulated from composite flours of sweet potato, carrot, and okok on key components of metabolic syndrome. We hypothesize that the nutritional and bioactive properties of these natural ingredients may positively influence metabolic parameters associated with the syndrome. The main objective of this study is to contribute to improved dietary management of individuals with metabolic syndrome. Specifically, this study aims to: (i) optimize the formulation, (ii) determine their nutritional composition and energy value, and (iii) assess their effects on anthropometric and metabolic parameters, including body weight, abdominal circumference, lipid profile, blood glucose levels, and inflammatory markers in *Wistar* rats. The outcomes of this research are significantly important for both scientific advancement and public health. The validation of a non-restrictive dietary strategy could open new perspectives for the development of functional foods based on locally available and nutrient-rich resources. Ultimately, this study contributes to a better understanding of the relationship between diet and metabolic diseases, thereby supporting the development of more effective prevention and management strategies.

Methods

Study design

This study was conducted in two main phases: (i) preparation and physicochemical characterization of formulation produced from composite flours, and (ii) in vivo evaluation of their effects on metabolic syndrome parameters using Wistar rats.

Plant material and raw ingredients

The plant materials used in this study included sweet potato (*Ipomoea batatas*), carrot (*Daucus carota* L.), and okok leaves (*Gnetum africanum*). These raw materials were sourced from local markets and selected based on their freshness and quality. All samples were thoroughly washed with clean water prior to processing to remove impurities. The plant materials used in this study were purchased from a local market and authenticated based on their morphological characteristics using standard botanical references.

Preparation of flours

Sweet potatoes and carrots were peeled, sliced into thin pieces, and dried under controlled conditions until constant weight was achieved. The dried samples were then ground into fine flours using a mechanical grinder and sieved to obtain uniform particle size. Okok leaves were sorted, washed, and dried before being finely ground into powder. All flours were stored in airtight containers at room temperature until further use. The formulation was prepared using composite flours of sweet potato (60–65%), carrot (15–30%), and okok (10–20%). The proportions were optimized using Minitab statistical software based on predefined criteria. A total of nine formulations were developed, including a control biscuit made from wheat flour (E0). Among the composite formulations, E3 was selected as the optimal formulation based on its nutritional composition and sensory acceptability. The ingredients were mixed to form a homogeneous dough, shaped, and baked at controlled temperature until golden brown. After baking, the formulation was cooled at room temperature and stored in airtight packaging.

Proximate composition analysis

The nutritional composition of the formulation was determined using standard analytical methods. The following parameters were analyzed: Moisture content Ash content Crude protein Lipid content Crude fiber Carbohydrate content (by difference) Energy value was calculated based on standard conversion factors.

Sensory evaluation

Sensory analysis was conducted to assess the acceptability of the different biscuit formulations. A panel of evaluators assessed attributes such as color, texture, taste, aroma and overall acceptability using a hedonic scale.

Experimental animals

Male *Wistar* rats were used for the in vivo study. The animals were housed under standard laboratory conditions, with controlled temperature, humidity, and a 12-hour light/dark cycle. They had free access to water and standard laboratory feed throughout the experimental period.

Ethical consideration

Animal experiments were performed in this study according to the guidelines set for the care and use of laboratory animals and with the rules formulated under the Animal Welfare Act by the United States Department of Agriculture (USDA) and by adopting ARRIVE guidelines, and in agreement with the Ethics and Animal Experimentation Committee of the University of Dschang.

Experimental design and diets

The rats were randomly divided into four groups of five animals each: Control group receiving standard diet Negative control group (metabolic disorder model, if applicable) Test groups receiving diets supplemented with our formulation at 10% and 20% incorporation levels The feeding trial lasted for 28 days.

Measurement of parameters

Food Intake and body weight

Food intake was recorded daily, and body weight was measured weekly throughout the experimental period.

Anthropometric parameters

Abdominal circumference was measured to assess central obesity.

Biochemical analysis

At the end of the experiment, blood samples were collected for biochemical analysis. The following parameters were determined: Total cholesterol LDL cholesterol HDL cholesterol Triglycerides Blood glucose the atherogenic index was calculated using standard formulas.

Organ weights and adipose tissue

After sacrifice, organs such as liver and kidneys were excised and weighed. Adipose tissue mass was also measured.

Inflammatory marker

An inflammatory marker was assessed to evaluate the anti-inflammatory effect of the formulated biscuits.

Postprandial glycemic response

Postprandial blood glucose levels were measured at specific time intervals after biscuit consumption to evaluate glycemic response.

Statistical analysis

Data were expressed as mean ± standard deviation. Statistical analysis was performed using appropriate software (e.g., SPSS or Minitab). Differences between groups were assessed using analysis of variance (ANOVA), followed by post hoc tests where applicable. A p-value < 0.05 was considered statistically significant.

Results

Proximate composition of biscuits

The proximate composition of the different biscuit formulations is grouped in Table 1. It revealed significant variations depending on the ingredients used. The optimized formulation (E3) exhibited significantly higher ash, crude fiber, and lipid contents compared to the control biscuit (E0) ($p < 0.05$). These differences reflect the contribution of okok leaves and carrot flour, both known for their richness in minerals and dietary fiber. Moisture content remained within acceptable limits for all formulations, ensuring good shelf stability. Carbohydrate content was relatively higher in the control biscuit, likely due to the predominance of refined wheat flour. The calculated energy values indicated that all biscuit formulations provide substantial caloric intake, with E3 showing a slightly enhanced nutritional profile due to its composite nature.

Sensory evaluation

The Sensory profile of the reference biscuit and the sweet potato-based biscuit is presented in Figure 1. Sensory analysis showed that the control biscuit (E0) was initially preferred by panelists in terms of taste, texture, and overall acceptability. However, the composite formulation, particularly formulation E3, achieved satisfactory acceptability scores. Although slightly lower than the control, these scores remained within an acceptable range, suggesting good potential for consumer acceptance. The incorporation of okok leaf powder slightly influenced color and flavor, which may explain the observed differences in preference.

Effect on food intake and body weight

Figure 2 presents the effect of food intake on body weight. A significant reduction in food intake was observed in rats fed with the E3 biscuit at a 20% incorporation level compared to the control group ($p < 0.05$) Body weight evolution showed a moderated weight gain in treated groups, suggesting a potential regulatory effect of the formulated biscuits on energy intake and metabolism.

Effect on anthropometric parameters

Figure 3 presents the effect of the formulation on abdominal circumference. Rats receiving the E3 biscuit exhibited a reduction in abdominal circumference compared to control groups. This effect was more pronounced at the 20% incorporation level, indicating a potential impact on central obesity.

Effect on lipid profile

Table 2 shows effect of consumption of the formulation on lipid profile. The consumption of E3 formulation significantly improved lipid parameters. At a 20% incorporation level, a significant decrease in LDL cholesterol and an increase in HDL cholesterol were observed ($p < 0.05$). Additionally, triglyceride levels were significantly reduced at the 10% incorporation level. These findings suggest a beneficial effect of the composite formulation on lipid metabolism and cardiovascular risk factors.

Atherogenic index

The atherogenic index is presented on Table 3. It appears to be a reduction in rats fed with the E3 biscuit, particularly at the higher incorporation level. This reduction indicates a lower risk of cardiovascular complications.

Organ weights and adipose tissue

Organ weights and adipose tissue are grouped in Figure 4. A decrease in adipose tissue mass was observed in treated groups compared to controls, reflecting reduced fat accumulation. Liver and kidney weights did not show any significant pathological variations, suggesting the absence of toxic effects associated with biscuit consumption.

Inflammatory marker

A significant reduction in the inflammatory marker was observed in rats receiving the E3 biscuit at 20% incorporation ($p < 0.05$), indicating a potential anti-inflammatory effect. Results are presented in Figure 5.

Postprandial glycemic response

Variation of blood glucose level during oral glucose tolerance test is presented in Figure 6. The postprandial glycemic response was significantly attenuated in rats fed with the composite formulation. Both 10% and 20% incorporation levels resulted in reduced glycemic peaks compared to the control group, highlighting the low glycemic index potential of the formulated biscuits.

Discussion

The present study aimed to develop functional formulation based on locally available plant resources and to evaluate their effects on key parameters of metabolic syndrome. The findings demonstrate that the optimized formulation (E3), composed of sweet potato, carrot, and okok flours, provides significant nutritional and metabolic benefits. The higher ash and dietary fiber contents observed in the E3 formulation are consistent with the known composition of *Gnetum africanum* and carrot, which are rich in minerals and non-digestible carbohydrates. Dietary fiber plays a central role in metabolic regulation by delaying gastric emptying, reducing glucose absorption, and improving insulin sensitivity [15:16]. These mechanisms may explain the significant reduction in postprandial glycemic response observed in rats fed with the composite biscuits. Similar findings have been reported in studies evaluating fiber-enriched foods, which demonstrated improved glycemic control and reduced risk of type 2 diabetes [17]. Although the control biscuit (E0) showed higher sensory acceptability, the E3 formulation maintained satisfactory organoleptic properties. This observation is consistent with previous studies indicating that the incorporation of plant-based flours can slightly alter sensory characteristics while significantly enhancing nutritional quality [16]. The challenge in functional food development lies in achieving an optimal balance between health benefits and consumer acceptability. The reduction in food intake observed at higher incorporation levels (20%) suggests a satiety -enhancing effect, likely due to the high fiber content of the composite formulation. Dietary fiber is known to promote satiety through increased gastric distension and modulation of appetite-regulating hormones [17]. This effect may explain the moderate weight gain and reduced abdominal circumference in treated groups, highlighting the potential role of these formulations in the management of obesity, a core component of metabolic syndrome [18]. The improvement in lipid profile, characterized by decreased LDL cholesterol, increased HDL cholesterol, and reduced triglyceride levels, is particularly noteworthy. These effects may be attributed to the presence of bioactive compounds such as polyphenols, flavonoids, and carotenoids found in sweet potato and carrot. These compounds have been widely reported to exert hypolipidemic and antioxidant effects, thereby reducing cardiovascular risk [19,13]. Similar improvements in lipid metabolism have been observed in studies involving plant-based functional foods rich in antioxidants [20]. Furthermore, the reduction in the atherogenic index reinforces the cardioprotective potential of the formulated biscuits. The decrease in adipose tissue mass observed in treated groups supports the hypothesis that these functional foods may limit fat accumulation and improve lipid metabolism. This is in agreement with findings from previous studies showing that diets rich in plant bioactive compounds can modulate lipid storage and energy balance [11]. Importantly, no significant adverse effects were observed on liver and kidney weights, suggesting that the formulated biscuits are safe under the experimental conditions. This finding is essential for the development of functional foods intended for long-term consumption and is consistent with studies reporting the safety of plant-based dietary interventions [8]. The significant reduction in inflammatory markers at higher incorporation levels indicates a potential anti-inflammatory effect. Chronic low-grade inflammation is a hallmark of metabolic syndrome and contributes to the development of insulin resistance and cardiovascular diseases [21]. The observed anti-inflammatory effect may be attributed to the synergistic action of polyphenols and other bioactive compounds present in the composite flours, which are known to modulate inflammatory pathways [22]. Overall, the combined improvements in glycemic control, lipid profile, inflammation, and body composition suggest that the formulated biscuits could serve as an effective dietary strategy for the prevention and management of metabolic syndrome. These findings support the growing body of evidence highlighting the role of functional foods in addressing non-communicable diseases. However, this study has some limitations. The use of an animal model may not fully reflect human metabolic responses, and further clinical trials are required to validate these findings in human populations. In addition, long-term studies would be necessary to assess the sustainability and safety of regular consumption. Despite these limitations, this study provides strong evidence supporting the potential of locally sourced functional foods as cost-effective and accessible strategies to combat metabolic syndrome. It also underscores the importance of integrating traditional plant resources into modern nutritional interventions.

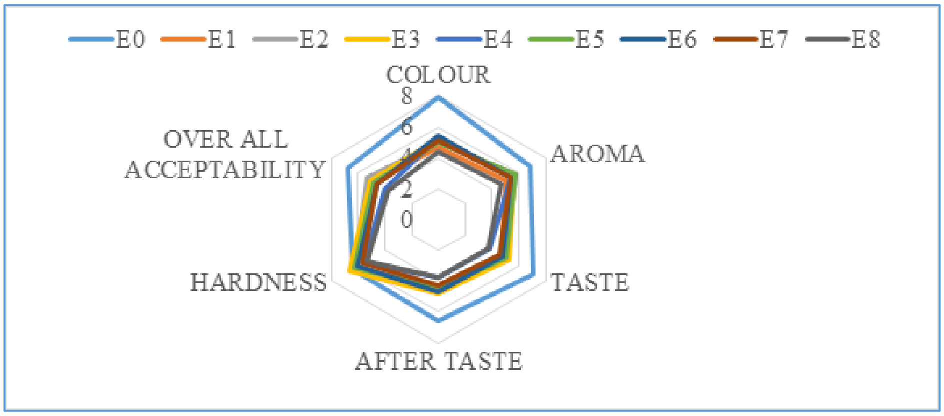

Figure 1. Sensory profile of the reference biscuit and the sweet the sweated biscuit. E0: reference biscuit, E1-E8: formulated biscuit at different percentage.

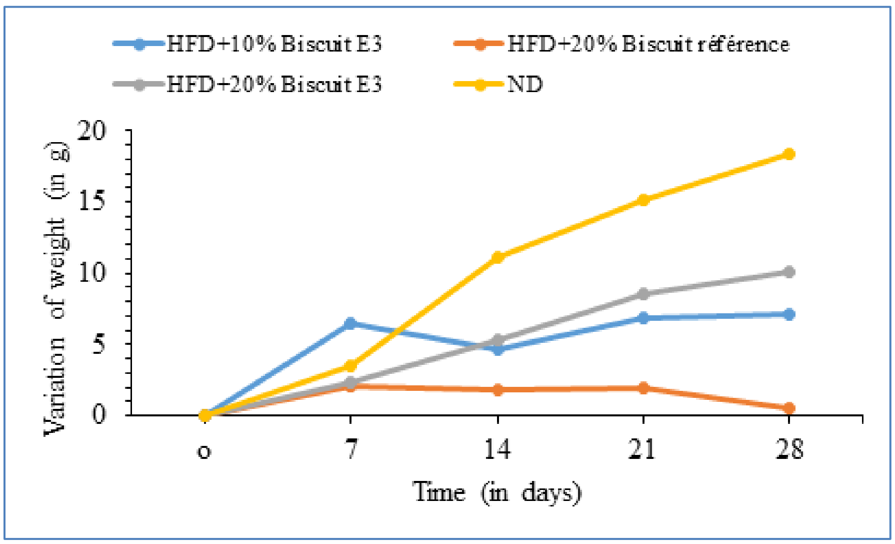

Figure 2. Effect of food intake on body weight.
ND: normal diet HFD: high fat diet

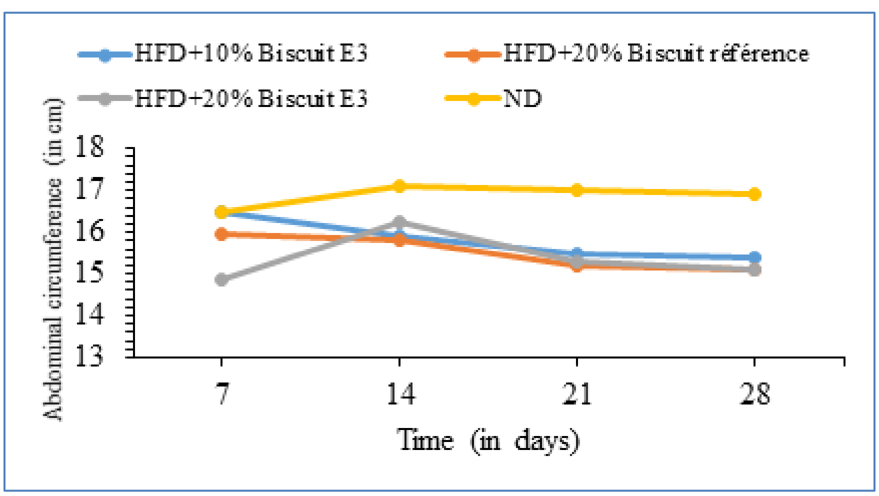

Figure 3. Abdominal circumference of animal.
ND: normal diet HFD: high fat diet

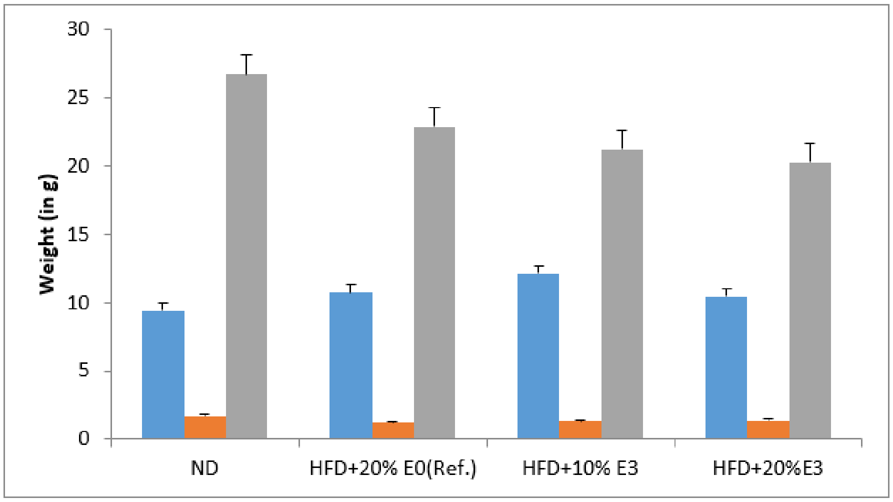

Figure 4. Effect of biscuit on organ weights and adipose tissue (liver, kidney and fats respectively).
ND: Normal Diet, HFD: High-Fat Diet

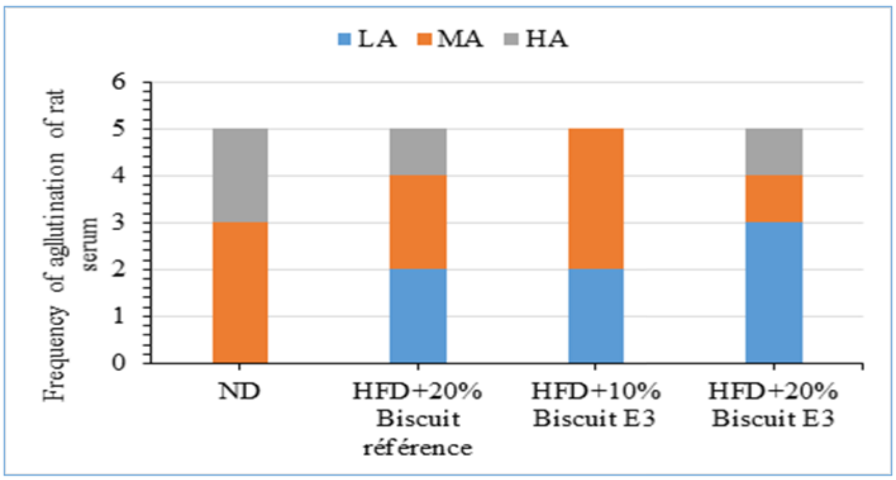

Figure 5. Effect of biscuits on inflammatory markers.
ND: Normal Diet, HFD: High-Fat Diet

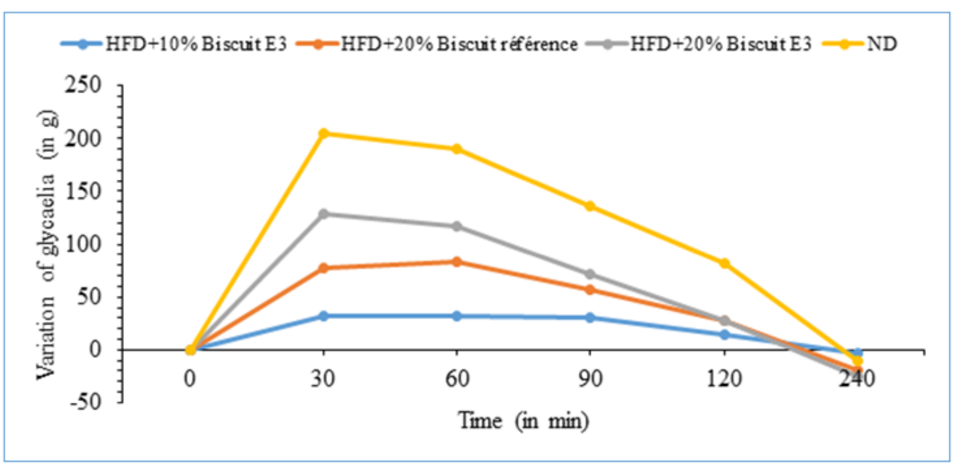

Figure 6. Variation of glycaemia during oral glucose tolerance test (OGTT).
ND: Normal Diet, HFD: High-Fat Diet

Table 1. Proximate composition of biscuits.

Bromatological Characteristics	Samples	
	E0 (Ref.)	E3
Dry matter (%)	99.41 ± 0.12^{b}	99.50 ± 0.13^{a}
Moisture content (%)	2.28 ± 0.12^{a}	0.49 ± 0.13^{b}
Ash content (%)	3.10 ± 0.02^{b}	5.32 ± 0.02^{a}
Crude protein (%)	11.85 ± 0.01^{a}	8.26 ± 0.01^{b}
Crude fiber (%)	3.49 ± 0.00^{b}	7.32 ± 0.01^{a}
Lipid (%)	20.67 ± 0.43^{a}	23.05 ± 1.27^{a}
Carbohydrate (%)	62.09 ± 0.33^{a}	62.87 ± 1.36^{a}
Energy Value (Kcal/100 g)	481.79 ± 2.26^{a}	491.97 ± 5.93^{a}

Values are presented as mean ± standard deviation (SD). Different superscript letters (a, b) within the same row indicate statistically significant differences between formulations (E0 vs. E3) at $p < 0.05$. E0: control biscuit (wheat flour-based); E3: optimized composite biscuit.

Table 2. Lipid profile of animals

Parameters	TAG (mg/dL)	Cholest (mg/dL)	HDL- c (mg/dL)	LDL- c (mg/dL)
ND	205.78 ± 11·17^{a}	197.49 ± 40·34^{a}	53.70 ± 22.25^{a}	117.18 ± 36.37^{b}
HFD+ 20% reference Biscuit	206.70 ± 8.75^{a}	162.79 ± 8.94^{a}	48.91 ± 32·38^{a}	72.54 ± 24.95^{ab}
HFD+ 10% Biscuit E3	202. 901 ± 4.86^{a}	186.78 ± 20·17^{a}	45.75 ± 32.90^{a}	100.44 ± 15.28^{ab}
HFD+20% Biscuit E3	205.96 ± 4.26^{a}	178.31 ± 24.53^{a}	58.99 ± 39.42^{a}	78.12 ± 23.34^{a}

Values are expressed as mean ± SD. Different superscript letters (a, b) within the same column denote significant differences among experimental groups ($p< 0.05$). ND: normal diet (healthy control); HFD: high-fat diet (metabolic syndrome model); HFD + 20% reference biscuit: HFD supplemented with 20% control wheat biscuit; HFD + 10% Biscuit E3: HFD supplemented with 10% optimized composite biscuit; HFD + 20% Biscuit E3: HFD supplemented with 20% optimized composite biscuit.

Conclusion

This study demonstrated that the optimized composite biscuit formulation significantly improved metabolic parameters associated with metabolic syndrome in *Wistar* rats. The formulation reduced postprandial glycemic response, improved lipid profile, decreased abdominal circumference, and exhibited anti-inflammatory effects without observable toxicity. These findings support the potential use of plant-based functional foods as complementary dietary strategies for the management of metabolic syndrome. Further studies involving long-term evaluation and clinical trials in humans are recommended to validate these effects.

Abbreviations

AI: Atherogenic Index
ANOVA: Analysis of Variance
BW: Body Weight
CHO: Total Cholesterol
DM: Diabetes Mellitus
GI: Glycemic Index
HDL: High-Density Lipoprotein
HFD: High-Fat Diet
IDF: International Diabetes Federation
LDL: Low-Density Lipoprotein
MetS: Metabolic Syndrome
NCEP: National Cholesterol Education Program
OGTT: Oral Glucose Tolerance Test
TAG: Triacylglycerols
WHO: World Health Organization

Authors' Contribution

AK, DF, and FAT conducted experiments, analyzed the data, and drafted the manuscript. JPD designed the study, supervised the research, and revised the manuscript. CTM conceived the study, supervised the experiments, and approved the final version. All authors read and approved of the final manuscript.

Acknowledgments

The authors thank the Department of Biochemistry, University of Dschang, for providing laboratory facilities and technical support.

Conflict of interest

The authors declare no conflict of interest

Article history:

Received: 02 April 2026
Received in revised form: 28 May 2026
Accepted: 12 June 2026
Available online: 12 June 2026

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