

Sodium butyrate arrests pancreato-hepatic synchronous uric acid and lipid dysmetabolism in high fat diet fed Wistar rats

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ARTICLE INFO

Keywords:

Pancreas

Short chain fatty acids

NAFLD

Uric acid

High fat diet

ABSTRACT

High fat diet (HFD) is a risk factor for metabolic syndrome which is characterized by overt glucose dysmetabolism and tissue derangement. The liver and pancreas are important metabolic tissues with anatomical proximity sharing splanchnic and mesenteric circulation but it is unclear whether, there is an associated metabolic status between the two organs in health and disease. Uric acid (UA) hypersecretion and ectopic lipid accumulation are characteristic pathophysiology of an array of non-communicable diseases. Sodium butyrate (BUT) is reputed for therapeutic roles in metabolic derangement. Therefore, the present study investigated synchrony in hepatic and pancreatic UA and lipid metabolic status in HFD-induced glucose dysregulation and probed the beneficial effects of BUT. Twenty-four female Wistar rats were treated with normal rat chow and distilled water (*po*) or sodium butyrate (200 mg/kg; *po*) or high fat diet and distilled water (*po*) or high fat diet and sodium butyrate. Results showed that HFD increased plasma, pancreatic and hepatic triglyceride, triglyceride-glucose index, malondialdehyde, uric acid (UA), lactate dehydrogenase but reduced glucose-6-phosphate dehydrogenase. Histological analysis revealed hepatic and pancreatic architectural derangement and cellular degeneration in HFD-fed animals. However, BUT reversed the HFD-induced systemic, pancreatic and hepatic synchronous dysmetabolism with evidence of improved histology. HFD-induced lipid and UA alterations were synchronous in the pancreas and liver. BUT elicits beneficial effects on systemic and tissue HFD-induced deleterious metabolic changes which were synchronized in pancreas and liver of rats.

1. Introduction

Globally, there is a growing incidence in the spectrum of diet-based diseases like diabetes, insulin resistance, obesity, cardiovascular diseases, hypertension etc. and diets rich in fat is reputed to play a major role in their pathogenesis [1,2]. Chronic consumption of high-fat diet creates a distortion in the body's metabolic milieu [1,3,4]. Furthermore, a high-fat diet feeding is accompanied by an increase in the circulating triglycerides [5,6] which by extension, results in oxidative stress [7,8]. Circulating free fatty acids penetrate the liver and are either esterified into triglycerides for storage, modified into lipoproteins or used as energy sources [9]. However, excess influx of free fatty acids into the liver

promotes hepatocyte lipid accumulation resulting in non-alcoholic fatty liver disease (NAFLD) [10] marked by increased aspartate aminotransferase (AST), alanine aminotransferase (ALT) concentrations etc. [11].

The pancreas also suffers the consequence of excess circulating free fatty acids and triglycerides which accumulate as fats in the pancreas termed non-alcoholic fatty pancreatic disease (NAFPD) [12,13]. This pancreatic condition has been reportedly associated with hyperglycemia and a decline in glucose-mediated insulin secretion via molecular mechanisms that involve a metabolic crosstalk with fatty liver [14,15]. Both the liver and pancreas are involved in the regulation of the acid-base balance of the body, pertinently via their alkaline secretions [16–18]. However, a decline in their functional capacity following the

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<https://doi.org/10.1016/j.bioph.2020.110994>

Received 20 May 2020; Received in revised form 24 October 2020; Accepted 1 November 2020

Available online 13 November 2020

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accumulation of fat would consequently increase metabolic acidosis which further exacerbates their injury [16]. This metabolic dysregulation resulting in insulin resistance/impaired glucose regulation is a major connecting link among metabolic disorder as in NAFLD and NAFPD [13,19]. Report have been demonstrated that NAFPD could be an early sign of ectopic fat accumulation and an advance demonstration of metabolic disorder than NAFLD [20], suggesting that NAFPD precedes NAFLD or could sometimes co-existing metabolic disorder [19].

Interestingly high fat diet has been reported to induce elevated tissue uric acid content [21]. Also, hyperuricemia has been associated with the etiology of NAFLD, insulin resistance and pancreatic β -cell dysfunction [22]. It is noteworthy to state that NAFPD may result in speedy development of type 2 diabetes mellitus [15], through the surplus TG accumulation in the pancreatic β -cells causing diminished β -cells' number, function and finally dysregulated insulin secretion [23]. However, the connection between elevated tissue uric acid and NAFPD has not been fully elucidated. Furthermore, the metabolic link between pancreatic adipocytes and islets has not been widely investigated [24,14].

The gut microbiota, involved in the production of sodium butyrate, has recently emerged as a major player in the regulation of metabolic homeostasis in health and disease [25–27]. Sodium butyrate has been reported to mitigate the development of metabolic syndrome through its anti-inflammatory, anti-oxidative, enhanced insulin sensitivity, lipid-lowering effects and amelioration of hepatic steatosis [28–32]. Recently we demonstrated that butyrate averts uric acid-associated cardiac tissue damage in high-fat fed rats [21]. However, the role of butyrate in the regulation of the pancreato-hepatic metabolic crosstalk in high-fat fed rats has not been established. Hence, we set out to investigate the impact of chronic high-fat diet feeding on hepatic and pancreatic uric acid level and lipid metabolism as well as to probe the putative modulatory role of sodium butyrate on the target conditions.

2. Materials and methods

2.1. Animals

All experimental protocols were conducted according to the guidelines for the use and care of experimental animals and it was approved by the College Ethical Review Committee, College of Medicine and Health Sciences, Afe Babalola University, Nigeria in accordance with guidelines of the National Institutes of Health Guide (NIH) for the Care and Use of Laboratory Animals and every effort was made to minimize both the number of animals used and their suffering. Twenty-four (24) adult female rats with weights ranging between 135–150 g were used for the study. Rats had unrestricted access to standard rat chow and tap water. After one week of acclimatization, the animals were randomly assigned to 4 groups of $n = 6$ each. Rats were maintained under standard environmental conditions of temperature, relative humidity, and 12 -h dark/light cycle. Body weight was also monitored for all the groups.

2.2. Treatment

Control group (with diet containing 8% energy as fat) received distilled water (vehicle; *po*) daily. HFD group received high fat rich diet (containing 60 % energy as fat). BUT group received sodium butyrate (200 mg/kg body weight) daily, HFD + BUT group received a combination of high fat with sodium butyrate daily. The treatment lasted for 6 weeks.

2.3. Sample preparation

Finally, after 6 weeks of treatment, the rats were anesthetized with pentobarbital sodium. Blood was collected by cardiac puncture into heparinized bottles, accordingly. Blood collected in heparinized bottle was centrifuged at 3000 rpm for 5 min. Plasma was stored frozen until needed for biochemical assay.

2.4. Oral glucose tolerance test (OGTT) and insulin resistance (IR)

Glucose challenge test was performed 48 h before the end of the experiment. The rats were kept in fasting condition for 12-h overnight. Blood sample was collected by tail cut method before glucose load and blood glucose level assessed by a glucometer (ONETOUCH®-LifeScan, Inc., Milpitas, CA, USA), it was denoted as 0th time point. Glucose (2 g/kg *bw*) was administered orally (*po*) and blood glucose was assessed subsequently after 30, 60, 90- and 120-min. Glucose tolerance was expressed as a function of the area under the OGTT curve (AUC) and postload glucose responses. The IR was determined using the homeostasis model assessment for insulin resistance ($\text{HOMA-IR} = \text{fasting glucose (mmol/l)} \times \text{fasting insulin } (\mu\text{U/l})/22.5$).

2.5. Biochemical assays

The plasma levels of corticosterone were estimated using ELISA kits (Elabscience Biotechnology Co., Ltd., Wuhan, China). Insulin was also determined using ELISA kit (Ray Biotechnology, Cruz, Canada). The biological markers of dyslipidemia including total cholesterol (TC), triglyceride (TG), high-density lipoprotein-cholesterol (HDL-C) were estimated by standardized enzymatic colorimetric methods using assay kit obtained from Fortress Diagnostics Ltd. Antrim, UK. The atherogenic indices (TC/HDL-C and TG/HDL-C ratios) were estimated.

2.6. Histopathology of rat's pancreas and liver

The pancreas and hepatic tissues were excised and fixed in 10 % formalin solution immediately after removal. The specimens were dehydrated in ascending grades of alcohol, cleared in xylene and embedded in paraffin. Serial sections were cut, stained appropriately and examined by the light microscope for evaluation of histopathological changes. The same pathologist, who had no knowledge of the experimental data, reviewed the histological slides.

2.7. Statistical analysis

All experimental data were expressed as means $\pm$ SEM. Statistical significance for measured variables was determined by one-way analysis of variance (ANOVA) for the comparison of the mean values of variables among the groups. Bonferroni's test was used to identify the significance of pair wise comparison of mean values among the groups. Statistically significant differences were accepted at $p < 0.05$. Statistical group analysis was performed with SPSS statistical software.

3. Results

3.1. Effects of sodium butyrate on body weight, triglyceride and total cholesterol in high fat diet-fed female rats

Despite unaffected body weight in butyrate-treated and high fat diet-fed rats, there were comparable significant increases in plasma, pancreatic and hepatic triglyceride when compared to the control group, which were significantly reduced when concomitantly treated with sodium butyrate. Besides, high fat diet significantly increased plasma but not pancreatic and hepatic total cholesterol when compared to the control group, which was normalized by treatment with BUT (Table 1).

3.2. Sodium butyrate restores glucose homeostasis in high fat diet-fed female rats

High fat diet significantly increased fasting blood glucose despite increase in plasma insulin when compared with control group. In addition, the rats fed with high fat diet showed impaired pancreatic beta-cell function when compared with control group. This implied that high fat diet impaired glucose uptake leading to hyperglycemia and

Table 1

Body weight and lipid profiles.

	CON	BUT	HFD	HFD + BUT
Body Weight change (g)	19.6 ± 8.0	15.2 ± 5.3	16.9 ± 4.4	12.9 ± 5.7
Triglyceride (mg/dl)				
Plasma	61.9 ± 2.7	63.2 ± 2.4	77.8 ± 2.8*	61.5 ± 3.3 [#]
Pancreatic	73.6 ± 2.9	76.6 ± 3.7	97.5 ± 1.8*	74.1 ± 2.5 [#]
Hepatic	67.5 ± 3.1	59.2 ± 2.8	84.1 ± 2.5*	72.1 ± 1.7 [#]
Total Cholesterol (mg/dl)				
Plasma	122.5 ± 2.3	122.1 ± 3.8	154.1 ± 1.8*	129.1 ± 2.8 [#]
Pancreatic	86.3 ± 3.2	86.4 ± 5.4	85.4 ± 5.4	80.0 ± 1.0
Hepatic	88.6 ± 2.3	85.6 ± 2.9	84.6 ± 4.1	86.8 ± 3.8

Values are expressed as mean ± SEM of 6 rats per group; *p < 0.05 vs control, [#]p < 0.05 vs HFD group. Data were analyzed by one-way ANOVA and Bonferroni's *Post hoc* analysis; CON; control, BUT; sodium butyrate, HFD; high fat diet.

compensatory hyperinsulinemia that subsequently distressed pancreatic beta-cell, thus causing impaired beta-cell function, which has been previously associated with prediabetes and type 2 diabetes in humans and experimental rodents. Nevertheless, concurrent treatment of high fat diet-fed rats with sodium butyrate restored glucose homeostasis with corresponding enhancement of pancreatic beta-cell function (Fig. 1A–C).

3.3. Sodium butyrate attenuates elevated triglyceride-glucose index in high fat diet-fed female rats

High fat diet significantly increased the plasma, pancreatic and hepatic triglyceride-glucose indices when compared to the control group, which were attenuated by concomitant treatment with sodium butyrate (Fig. 1D–F).

3.4. Sodium butyrate preserves pancreatic and hepatic tissue cytoarchitecture and minimizes cellular infiltration in high fat diet-fed female rats

The histology of pancreatic tissue using Gordon special stain revealed normal pancreatic cells with no cellular degeneration in control rat, normal pancreatic cells with no degeneration in sodium butyrate-treated rat, scanty pancreatic cells with fatty degeneration high fat diet-fed rats and improved pancreatic cells with mild or no fatty degeneration in HFD + BUT-treated rats (Fig. 2). Similarly, the histology of hepatic tissue using H and E stain showed hepatic tissue with preserved architecture, normal hepatocytes and central veins in control rat, hepatic tissue with preserved architecture, normal hepatocytes and central veins in sodium butyrate-treated rat, disrupted hepatic tissue with severe cellular infiltration and degenerated central veins in high fat-treated rat and hepatic tissue with preserved architecture and mild cellular infiltration in HFD + BUT-treated rats (Fig. 3).

3.5. Sodium butyrate reduces plasma, pancreatic and hepatic lipid peroxidation in high fat diet-fed female rats

High fat diet led to a significant increase in plasma, pancreatic and hepatic MDA, which is a biomarker of lipid peroxidation and concomitant treatment with BUT significantly reduced the excessive lipid peroxidation observed in high fat diet-fed rats (Fig. 4A–C).

3.6. Sodium butyrate improves glutathione- and G6PD-dependent antioxidants in high fat diet-fed female rats

High fat diet significantly decreased plasma and pancreatic but not hepatic GSH when compared with control group (Fig. 4D–F). In addition, pancreatic and hepatic but not plasma G6PD activity decreased significantly compared to the control group (Fig. 5A–C). However, treatment with sodium butyrate significantly restored the GSH- and G6PD-dependent antioxidant capacity in high fat diet-fed rats.

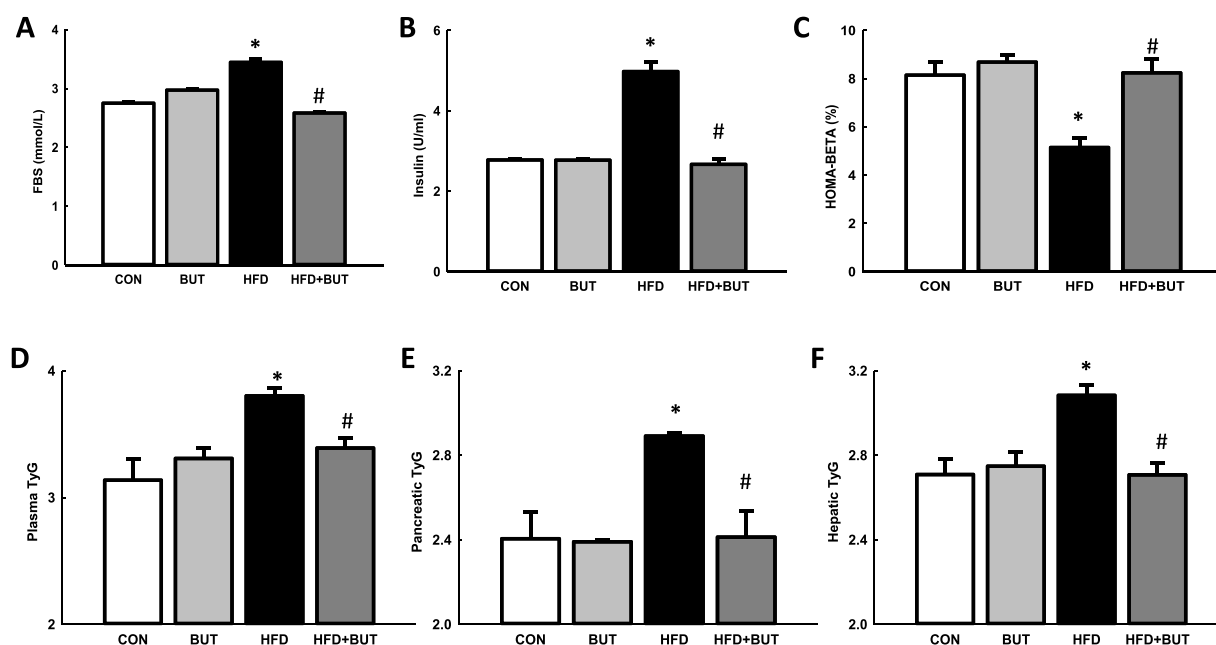


Fig. 1. Sodium butyrate improves glucose homeostasis and reduces triglyceride-glucose index (TyG) in female rats following high fat diet for 6 weeks. BUT improved fasting blood glucose (A), fasting insulin level (B), and HOMA-β (C). BUT also reduces plasma (D), pancreas (E) and hepatic (F) TyG. Values are expressed as mean ± SEM of 6 rats per group; *p < 0.05 vs control, [#]p < 0.05 vs HFD group. Data were analyzed by one-way ANOVA and Bonferroni's *post hoc* analysis; CON, control; HFD, high fat diet; BUT, sodium butyrate.

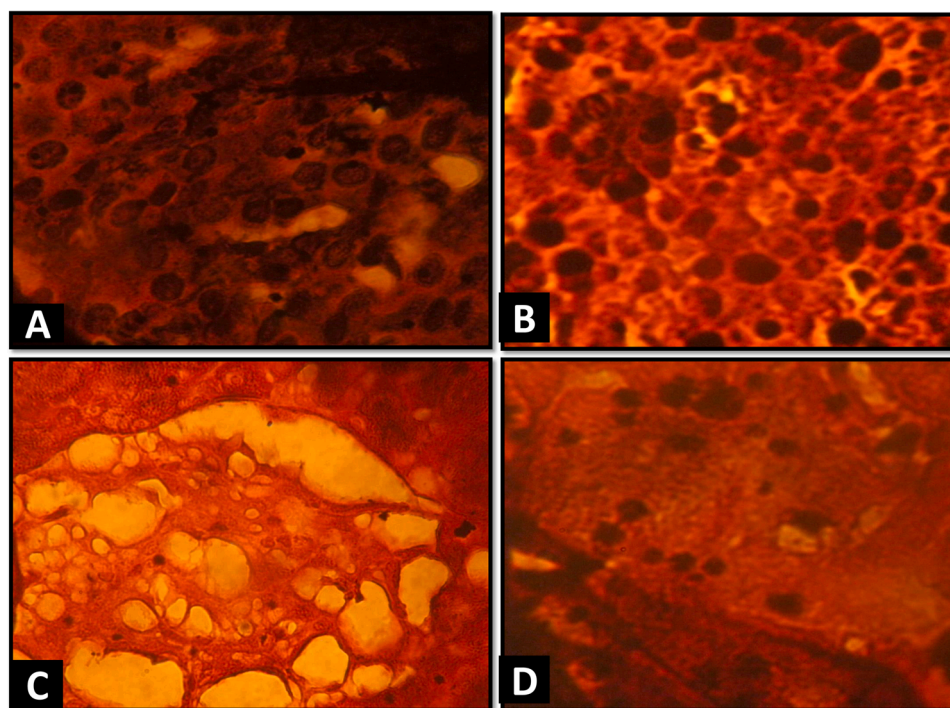


Fig. 2. Photomicrographs of Gordon's staining of pancreas showed normal pancreatic cells with no cellular degeneration in control rat (A), normal pancreatic cells with no degeneration in sodium butyrate-treated (B), pancreas with fatty degeneration and scanty pancreatic cells in high fat-treated (C) and improved pancreas in high fat plus sodium butyrate-treated (D) rats ($\times 200$, transverse section).

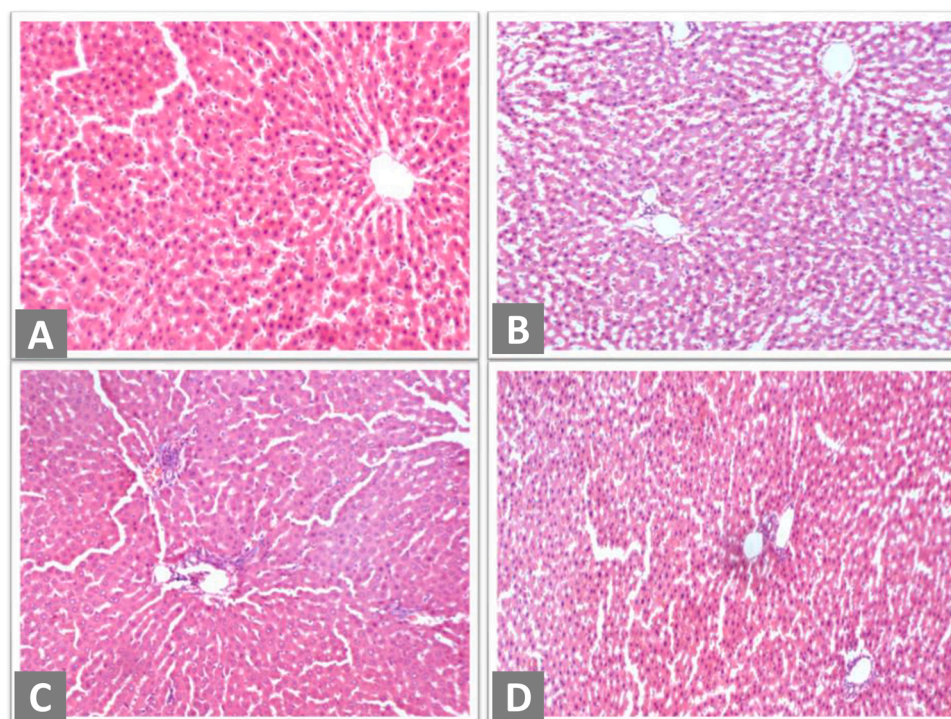


Fig. 3. Photomicrographs of H & E paraffin staining of liver showed hepatic tissue with preserved architecture, normal hepatocytes and central veins in control rat (A), hepatic tissue with preserved architecture, normal hepatocytes and central veins in sodium butyrate-treated rat (B), disrupted hepatic tissue with severe cellular infiltration and degenerated central veins in high fat-treated rat (C) and hepatic tissue with preserved architecture and mild cellular infiltration in high fat plus sodium butyrate-treated (D) rats ($\times 100$, transverse section).

3.7. Sodium butyrate attenuates elevated tissue injury markers in high fat diet-fed female rats

Lactate dehydrogenase (Fig. 5D–F) but not AST and ALT activities (Table 2) significantly increased in the plasma, pancreas and liver of the rats fed with high fat diet compared to the control group. Nevertheless,

the alteration was attenuated by treatment with sodium butyrate.

3.8. Sodium butyrate normalizes uric acid content in high fat diet-fed female rats

High fat diet led to a comparable significant increase in plasma,

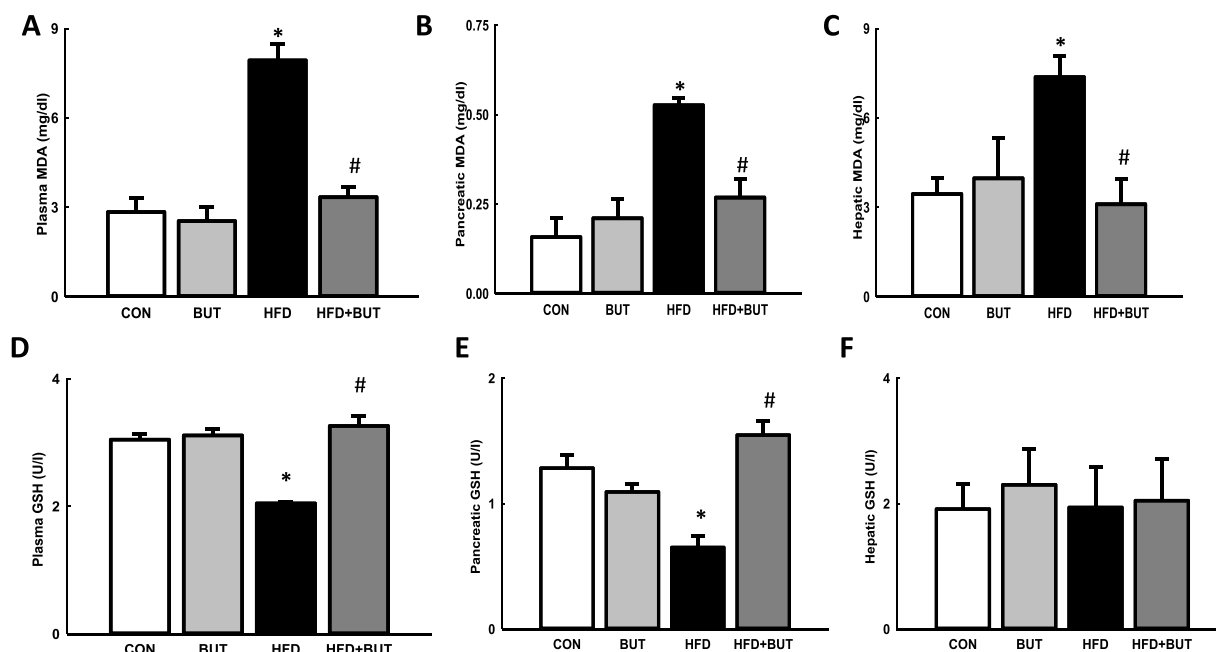


Fig. 4. Sodium butyrate attenuates lipid peroxidation marker and improves glutathione in female rats following high-fat diet for 6 weeks. Plasma (A), pancreatic (B) and hepatic (C) MDAs were attenuated by BUT. BUT also restores plasma (D), pancreatic (E) and hepatic (F) GSH. Values are expressed as mean $\pm$ SEM of 6 rats per group; * $p < 0.05$ vs control, # $p < 0.05$ vs HFD group. Data were analyzed by one-way ANOVA and Bonferroni's post hoc analysis; CON, control; HFD, high fat diet; BUT, sodium butyrate.

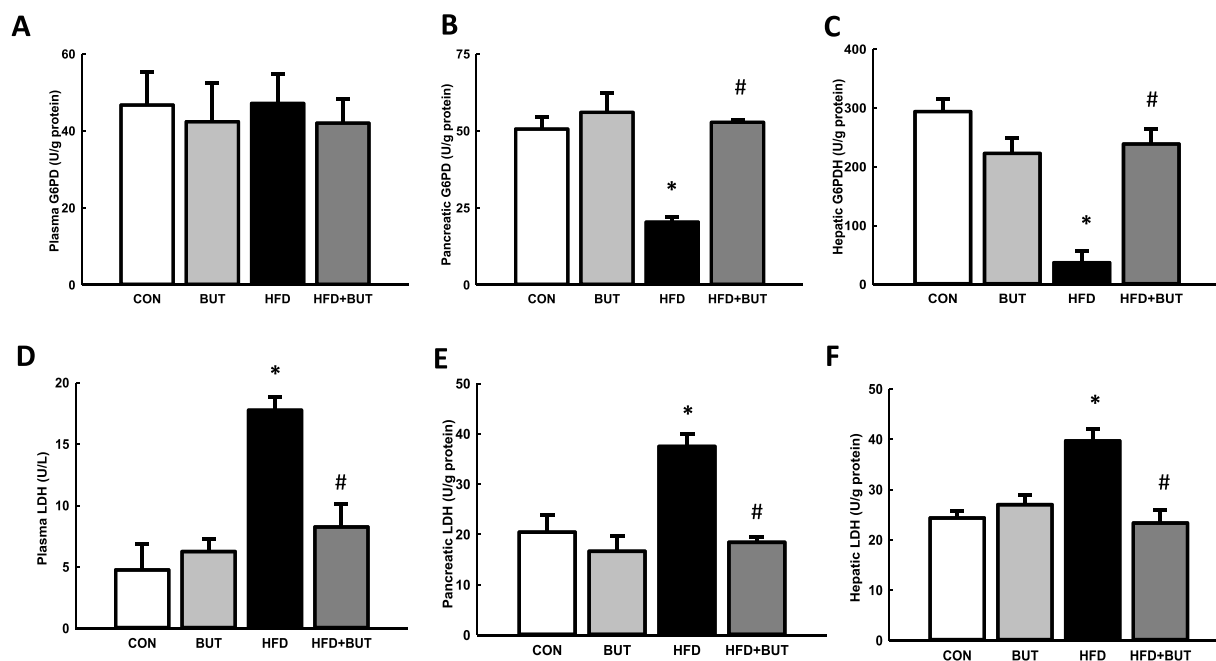


Fig. 5. Sodium butyrate reversed altered glucose 6 phosphate dehydrogenase (G6PD) and ameliorates lactate dehydrogenase in female rats following high-fat diet for 6 weeks. BUT increased plasma (A), pancreatic (B) and hepatic (C) G6PD. BUT also reduced plasma (D), pancreatic (E) and hepatic (F) LDH. Values are expressed as mean $\pm$ SEM of 6 rats per group; * $p < 0.05$ vs control, # $p < 0.05$ vs HFD group. Data were analyzed by one-way ANOVA and Bonferroni's post hoc analysis; CON, control; HFD, high fat diet; BUT, sodium butyrate.

pancreatic and hepatic uric acid concentration when compared with control group. However, treatment with sodium butyrate significantly blotted this alteration in high fat diet-fed rats (Fig. 6).

4. Discussion

Crosstalk from one organ to another may elicit contrary or similar

outcomes in the recipient organ. The import of this in the etiology of disease is that complications may ensue when one organ communicates with another by pathological signals which endanger the function of the recipient. The results of this study showed that HFD as expected elicited overt lipid dysregulation with elevated circulating TG and triglyceride-glucose index (TyG). This was accompanied by synchronous hepatic and pancreatic elevated TG accumulation, lipid peroxidation, UA, LDH and

Table 2
AST and ALT values.

	CON	BUT	HFD	HFD + BUT
AST (U/l)				
Plasma	164.5 ± 28.3	155.8 ± 37.5	160.5 ± 21.3	168.6 ± 16.8
Pancreatic	249.2 ± 89.8	256.2 ± 42.4	277.5 ± 85.3	263.9 ± 47.1
Hepatic	19.7 ± 2.6	17.3 ± 0.8	17.8 ± 0.2	17.2 ± 0.5
ALT (U/l)				
Plasma	44.1 ± 4.7	40.4 ± 2.9	44.5 ± 2.0	44.5 ± 3.8
Pancreatic	136.1 ± 24.5	133.0 ± 11.9	130.8 ± 27.1	144.7 ± 13.0
Hepatic	11.8 ± 0.2	16.3 ± 4.5	11.6 ± 1.2	11.0 ± 0.5

Values are expressed as mean ± SEM of 6 rats per group; **p* < 0.05 vs control, #*p* < 0.05 vs HFD group. Data were analyzed by one-way ANOVA and Bonferroni's *Post hoc* analysis; CON; control, BUT; sodium butyrate, HFD; high fat diet.

reduced G6PD. These intramural alterations are agreeably pathological and pathological metabolic synchrony in the pancreas and liver, two pivotal organs in energy homeostasis can establish a prediabetic status or cause overt diabetes mellitus type 2 (T2DM) in predisposed individuals. However, findings here show that short chain fatty acids (SCFAs) possess the potential to arrest synchronous pancreato-hepatic metabolic anomalies initiated by HFD in animals as it ameliorated lipid and UA dysmetabolism in the liver and pancreas aside resolving impaired circulating TG status and TyG.

Several studies using HFD model have shown consistently that chronic consumption of fat-rich diet is a risk factor for obesity, T2DM, and obesity-related diseases [21,33]. It is already conceded by researchers that chronic HFD increases lipid overflow in the circulation due to overshoot of adipose storage capacity [34]. This agreeably leads to ectopic lipid deposition in which the skeletal muscle receives the greatest impact causing insulin resistance (IR). TyG is an acceptable index of IR because both lipid and glucose disturbances are involved. The present study shows that HFD caused elevated circulating TG and TyG (Table 1 and Fig. 1D). Among the multiple mechanisms by which intramyocellular lipids engender IR is direct activation of toll-like receptor type 4 in tissue macrophages to stimulate inflammatory responses

that leads to deleterious post-translational modification of molecules involved in insulin signaling and disruption of the rate limiting step of insulin signaling, phosphorylation of insulin receptor substrate at serine instead of tyrosine amino acid residue due to events activated by fatty acid metabolites [35]. The established skeletal muscle IR causes glucose intolerance and increases circulating blood glucose which are important signals for both pancreatic and hepatic glucoregulatory functions.

Pancreatic and hepatic tissues are glucose sensitive. In hyperglycemic milieu, the glucose sensing system involving but not limited to insulin-independent glucose transporters present in liver and pancreas are crucial causing elevated intracellular glucose and diabetic-related signaling. Besides, HFD-engendered TG accumulation in the pancreas and liver is also a risk factor for metabolic complications like non-alcoholic fatty liver disease (NAFLD). Therefore, in HFD, there is plausibility that high circulating glucose and TG stemming from established peripheral (skeletal muscle and adipose tissue) IR and lipid spill over from adipose tissue can feed diabetic signals (altered free fatty acid and glucose metabolism) to glucoregulatory organs and importantly lead to tissue diabetic memory [34,36–38]. The results here showed that there was TG accumulation in the liver and pancreas of HFD-fed rats. Hepatic and pancreatic β -cell malfunction in T2DM is linked to oxidative stress and inflammation [39,40] which are also characteristics of lipotoxicity [35]. In tissues, accumulation lipid metabolites increase the signaling of mitogen activated protein kinase (MAPK) and nuclear factor kappa B (NF- κ B) which causes expression of inflammatory and oxidant molecules [35]. Accompanying TG accumulation, this study also found elevated UA (Fig. 6 B&C), LDH (Fig. 5 E&F) and reduced G6PD (Fig. 7) in the pancreas and liver. Production of UA is associated with oxidant effects via the activities of xanthine oxidase [41]. UA can also be a danger signal for the nucleotide-binding domain and leucine-rich repeat-containing family, pyrin domain containing 3 (NLRP3) inflammasome, an inflammation-inducing cytosolic molecule which responds to urate crystals or cholesterol crystals as damage molecular patterns. Deficiency of NLRP3 was beneficial to myocardial function subsequent to ischemia–reperfusion injury [42]. In addition, nuclear factor

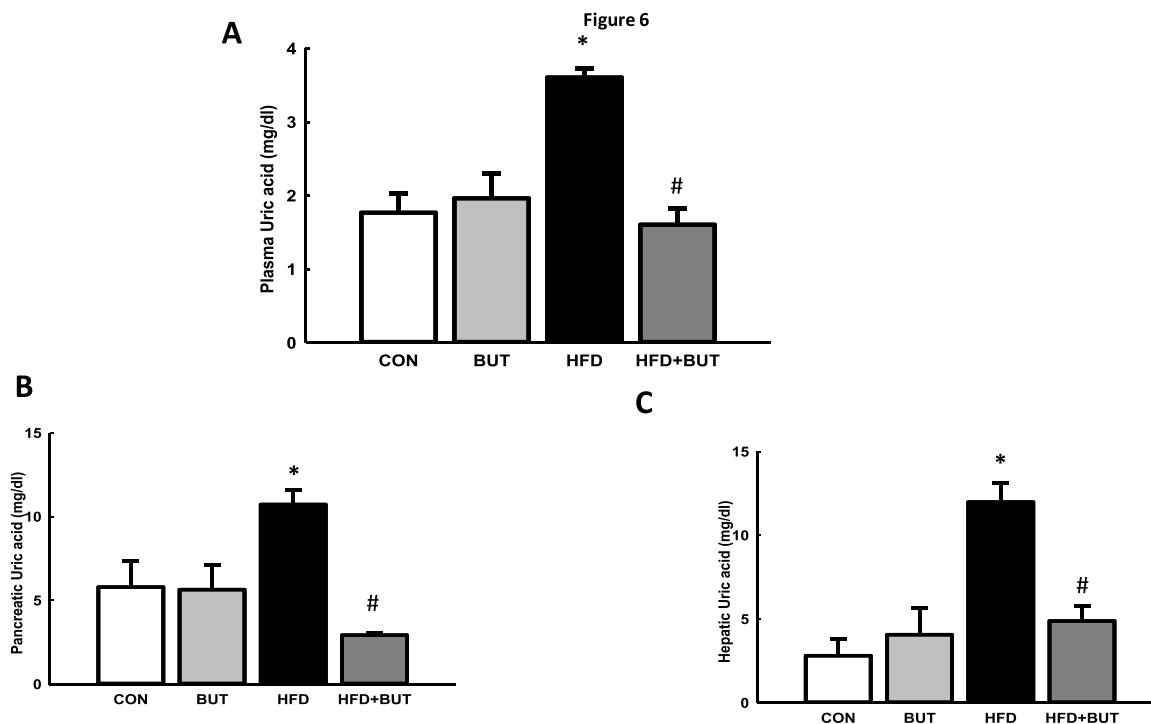


Fig. 6. Sodium butyrate ameliorates plasma (A), pancreatic (B) and hepatic (C) uric acid levels in female rats following high-fat diet for 6 weeks. Values are expressed as mean ± SEM of 6 rats per group; **p* < 0.05 vs control, #*p* < 0.05 vs HFD group. Data were analyzed by one-way ANOVA and Bonferroni's *post hoc* analysis; CON, control; HFD, high fat diet; BUT, sodium butyrate.

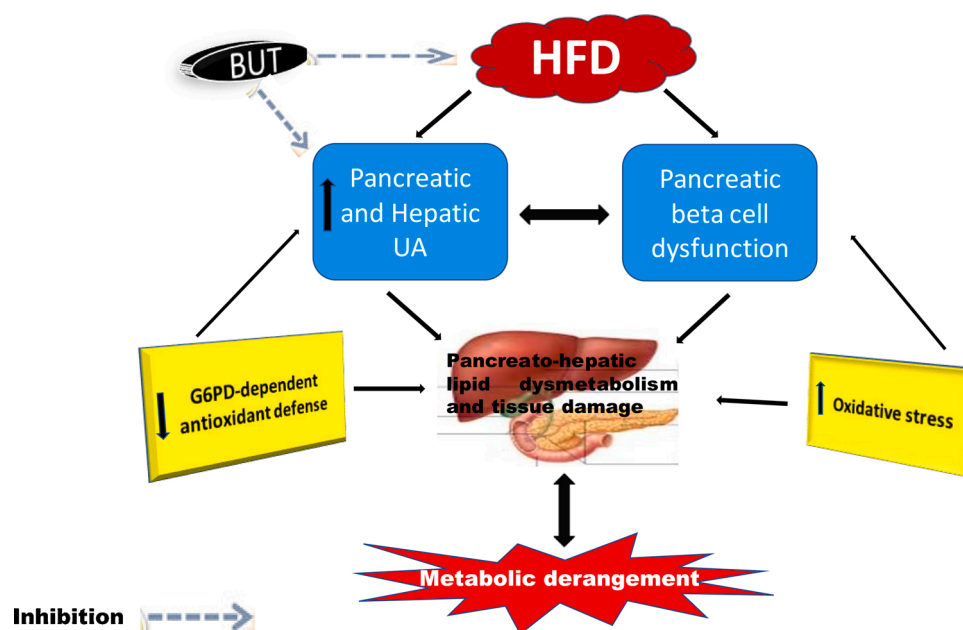


Fig. 7. Schematic diagram illustrating the likely pathway through which sodium butyrate arrests pancreato-hepatic uric acid and lipid dysmetabolism. HFD: High fat diet, BUT: sodium butyrate, UA: Uric acid.

erythroid 2-related factor 2 (Nrf2) is important in cellular protection against inflammation and oxidative stress and is triggered by oxidant molecules. There is plausibility that Nrf2 signaling is triggered by exposure to HFD here as a result of oxidant fat-related molecules as shown in a previous study [43].

Besides initiating ROS production through elevated UA production, insulin resistance and inflammatory responses, hepatic lipid accumulation dampens mitochondrial functions by increasing lipid peroxidation and production of malondialdehyde and 4-hydroxynonenal which inhibits mitochondrial enzymes like cytochrome c oxidase, oxidizes cardiolipin and impairs mitochondrial respiration. Hepatic ROS production induced by obesity also decreases mitochondrial biogenesis by reduction in PPAR- γ coactivator 1 α , nuclear respiratory factor 1 and mitochondrial transcriptional factor A mRNA [44]. Furthermore, LDH activities have also been linked to oxidant dysregulation especially when it favors lactate production [21]. It is therefore plausible that the suppression of G6PD, the rate limiting molecule in the pentose phosphate pathway that fuels the glutathione antioxidant system realized here is due to overwhelming oxidant effect of elevated intracellular lipids and UA. In addition, this study also showed evidence of increased lipid peroxidation (Fig. 4A–C) which is known to cause damage to membrane and cellular microstructure. As a plausible consequence to the discussed pancreatic and hepatic events in HFD, histological analysis here revealed impairment of cellular architecture and cellular degeneration in the liver and pancreas (Figs. 2&3). It should therefore be noted that the metabolic alterations related to TG accumulation, UA production and G6PD-dependent antioxidants in the liver can be a mirrored pancreatic metabolic status or vice versa especially in HFD due to comparable reception and handling of diabetic signals from peripheral tissues.

Effective treatment for metabolic syndrome is still a major concern for medical and paramedical scientists. However, a handy success has been made towards describing preventive measures. In the last decade, several scientific research endeavors have identified the beneficial effects of SCFAs and role of intestinal dysbiosis in the etiology of various diseases [45]. The present study investigated whether the reputedly therapeutic SCFA, butyrate could arrest pancreatic and hepatic synchronous events in HFD-fed rats. First, it was found here that treatment of HFD-fed rats with BUT prevented TG disturbances (Table 1) in the circulation and maintained TyG (Fig. 2). This shows that BUT can

potentially obliterate the progression of peripheral IR. A study from our laboratory showed that BUT ameliorated IR (HOMA-IR) and prevented cardiac derangements in HFD-fed female rats [21]. Increased SCFA is associated with release of glucagon-like peptide 1 (which promotes insulin action). In adipocyte, SCFAs are also known to prevent lipolysis and modulate inflammation [46,47]. These actions of SCFAs can predictably prevent peripheral IR. In addition, G-protein coupled receptor (GPR41) activation in peripheral tissues by SCFAs is the likely mechanism by which they affect adipose tissue and skeletal muscle metabolism [48,49]. It is therefore not surprising that BUT ameliorated pancreatic and hepatic lipid accumulation (Table 1) and UA dysmetabolism (Fig. 8) leading to amelioration of lipid peroxidation (Fig. 5) and improved histology (Figs. 3&4) in this study. Based on the result here, at least with the amelioration of lipid peroxidation, there is plausibility that BUT had improved Nrf2 signaling leading to expression of antioxidant target genes and cellular protection. It is hence suggested that BUT be studied regarding Nrf2 activation [43]. These beneficial effects of BUT in HFD-fed rats can be safely attributed to hepatic and pancreatic withdrawal of diabetic signals due to metabolically ailing peripheral tissues. In support of this, a previous study showed that the suppressive effect of HFD on butyrate formation by gut microbiome and increased hepatic fat accumulation were ameliorated by addition of BUT-producing dietary fiber feed [33].

In conclusion, the present study added noteworthy findings on deleterious effects of HFD to literature. These findings pointed out HFD-induced synchronous pancreatic and hepatic elevated lipid accumulation and UA levels which were accompanied by architectural and integrity deficiency in tissue micro structure. In addition to existing body of knowledge, BUT, a SCFA is shown here to avert uric acid and lipid dysmetabolism in the liver and pancreas of HFD-fed rats. Thus, BUT protects the liver and pancreas against dangerous diabetic signals and might prevent or prolong the onset of overt diabetes or obesity-related disease in HFD.

Statement on the contributions of authors

Adeyanju O, Olaniyi K, Olatunji L and Soladoye A conceived and designed the experiments. Adeyanju O, Badejogbin C and Soetan O conducted the experiments. Michael O and Oniyide A contributed in

statistical and data analyses. Adeyanju O, Areola D and Dibia C wrote the manuscript and Adeyanju O revised and edited the same. All authors read and approved the manuscript for publication.

Compliance with ethical standards

The investigation was conducted in accordance with the National Institute of Health Guide for the Care and Use of Laboratory Animals and was approved by the University ethical review committee, with protocol identification number NHREC29/10/2019, and every effort was made to minimize both the number of animals used and their sufferings.

Funding information

This study was not funded by any grant body but the authors involved.

Transparency document

The [Transparency document](#) associated with this article can be found in the online version.

Declaration of Competing Interest

The authors report no declarations of interest.

Acknowledgements

The authors acknowledge the technical support of Mrs. Olaiya Oluranti of Department of Physiology, Afe Babalola University and Mr. Adebowale of Bridge Biotech is also appreciated for the laboratory analysis.

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