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Some Biochemical Alterations Following Diabetes Induction In Albino Wistar Rats

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Abstract: ***Diabetes mellitus is a chronic disease associated with carbohydrate, protein and lipid metabolic disorders which can result to several complications within the biological system, and consequently, huge financial burden. This study aims at assessing the alterations in some biochemical parameters using diabetic animal model. Eighteen albino wistar rats (140g-200g) were divided into two groups of nine rats each. Diabetes was induced in group II by a single intraperitoneal dose of alloxan (150mg/kg of body weight) while group I (control) was given normal saline. Fasting blood samples were collected for biochemical analysis, and body weights measured pre- and post- alloxan administration. The results showed significant decrease and increase in body weights of diabetic and control groups respectively. The diabetic rats also showed significant elevations of plasma glucose, glycated hemoglobin, alanine aminotransaminase, aspartate aminotransaminase, gamma glutamyl transferase, urea, creatinine, total cholesterol, triglyceride, low density lipoprotein cholesterol, very low density lipoprotein cholesterol as well as atherogenic index of plasma, and a significant decrease in high density lipoprotein cholesterol when compared to the control. The study shows that diabetes mellitus causes alterations in some biochemical parameters in the biological system which may lead to various adverse consequences.***

Keywords: ***Diabetes mellitus, Alloxan, Lipid profile, Atherogenic index of plasma.***

I. INTRODUCTION

Diabetes mellitus is a metabolic disorder affecting the entire human race with accompanying life threatening complications (Tielmans et al., 2007). It is characterized by hyperglycemia which could result from deficiency of insulin production, action or both (Rasineni et al., 2010). The resultant persistent hyperglycemia causes certain alterations or abnormalities in carbohydrate, protein and fat metabolism in addition to long-term complications affecting the kidneys, eyes, nerves, heart and blood vessels (American Diabetes Association, 2008). The total number of people with diabetes mellitus worldwide was estimated at 150 million, and this figure is likely to double by the year 2025 (King et al., 1995), and consequently placing a huge burden on the healthcare system. Diabetes mellitus results in multi-system consequences associated with biochemical changes which present with disturbances in the metabolism of carbohydrates, fats and proteins. The present study was therefore designed to assess the changes in some of the biochemical parameters associated with diabetes.

II. MATERIALS AND METHODS

EXPERIMENTAL ANIMALS

Eighteen albino wistar rats weighing between 140g and 200g were used for this study. They were fed a standard poultry diet (Vital Feeds, Jos) and water ad libitum, allowed

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an adaptation period of two weeks, and were handled according to the institutional and international guidelines for the care and use of laboratory animals.

EXPERIMENTAL DESIGN

The rats were randomly assigned into two groups of nine animals each, groups I and II are the control and diabetic group respectively. The body weights were measured, and baseline blood samples collected from both groups before commencement of the study. After 28 days, body weights were also measured as well as blood samples collected for biochemical analyses.

INDUCTION OF DIABETES

Diabetes was induced in group II by a single intraperitoneal injection of 150mg/kg body weight of alloxan monohydrate (Sigma Aldrich) dissolved in normal saline after an overnight fast while group I was given normal saline.

BLOOD SAMPLE COLLECTION

Blood samples were collected from each group after an overnight fast before (pre) and after (post) induction of diabetes. The blood samples were dispensed into fluoride oxalate, EDTA and plain tubes for plasma glucose, glycated hemoglobin and other serum biochemical analyses respectively.

BIOCHEMICAL ANALYSES

Fasting plasma glucose (FPG), serum Urea, Creatinine, Alanine aminotrasaminase (ALT), Aspartate aminotrasaminase (AST), Total cholesterol (TC), High density lipoprotein cholesterol (HDL-C) and Triglyceride (TG) were determined using enzymatic colorimetric assay kits (Randox, Northern Ireland), Low density lipoprotein cholesterol (LDL-C) and Very low density lipoprotein cholesterol (VLDL-C) were calculated using Friedewald's formula (Friedewald et al., 1972), Atherogenic index of plasma (AIP) was calculated as the logarithmically transformed ratio of molar concentrations of TG to HDL-C as stated by Dobiásová and Frohlich (2001). Glycated hemoglobin (HbA1c) was measured in whole blood using micro-column chromatography as described by Bissé and Abragam (1985), and gamma glutamyl transferase was determined using Mindray BS-120 auto analyzer according to Gendler and Kaplan (1984).

STATISTICAL ANALYSIS

This was performed using the Statistical Package for Social Sciences (SPSS) version 20.0. All values were reported as mean ± standard deviation, and statistical significance set at $p<0.05$.

III. RESULTS

Group	Weight(g)
I (pre)	167.9 ± 6.2[a]
(post)	178.9 ± 5.6[b]
II (pre)	171.1 ± 5.9[a]
(post)	164.4 ± 6.5[b]

NOTE: Values are expressed as mean ± SEM. a, b within column signifies that means with different letters differs significantly at $p<0.05$ while means with the same letters does not differ significantly at $p>0.05$.

Table 1: Effect of diabetes on the body weights of experimental animals pre- and post-alloxan administration

Table 1 shows that the control and diabetic groups showed a significant ($p<0.05$) decrease and increase in their body weights respectively. However, prior to alloxan administration, the body weights of both groups did not differ significantly ($p>0.05$).

Group	FPG(mmol/l)	HbA1c(%)
I(pre)	5.0 ± 0.1[a]	3.9 ± 0.1[a]
(post)	4.9 ± 0.1[a]	3.8 ± 0.1[a]
II(pre)	4.9 ± 0.1[a]	4.0 ± 0.1[a]
(post)	13.1 ± 0.3[b]	8.1 ± 0.1[b]

NOTE: Values are expressed as mean ± SEM. a, b within column signifies that means with different letters differs significantly at $p<0.05$ while means with the same letters does not differ significantly at $p>0.05$.

Table 2: Effect of diabetes on the FPG and HbA1c of control and diabetic groups pre- and post- alloxan administration

In Table 2, the diabetic group showed significant ($p<0.05$) increase in FPG and HbA1c levels when compared to the control at post alloxan administration but no significant ($p>0.05$) difference before alloxan administration.

Group	ALT(iu/l)	AST(iu/l)	GGT(iu/l)	Urea(mmol/l)	Creat(µmol/l)
I (pre)	11.1 ± 0.8[a]	17.1 ± 1.0[a]	0.8 ± 0.2[a]	3.7 ± 0.4[a]	96.4 ± 1.9[a]
(post)	11.6 ± 1.0[a]	16.6 ± 0.8[a]	0.9 ± 0.1[a]	3.8 ± 0.4[a]	96.9 ± 1.6[a]
II (pre)	11.9 ± 0.9[a]	17.1 ± 1.0[a]	0.9 ± 0.1[a]	3.6 ± 0.3[a]	97.0 ± 1.9[a]
(post)	20.3 ± 0.7[b]	24.2 ± 0.7[b]	4.4 ± 0.2[b]	9.7 ± 0.1[b]	127.7 ± 1.6[b]

NOTE: Values are expressed as mean ± SEM. a, b within column signifies that means with different letters differs significantly at $p<0.05$ while means with the same letters does not differ significantly at $p>0.05$.

Table 3 Effect of diabetes on Liver enzymes and Kidney function of control and diabetic groups pre- and post- alloxan administration

In Table 3, the levels of ALT, AST, GGT, Urea and Creatinine were significantly ($p<0.05$) elevated after alloxan administration in the diabetic group, however, there were no significant differences in these values between the two groups before (pre-) alloxan administration.

Group	TC (mmol/l)	HDL-C (mmol/l)	LDL-C (mmol/l)	VLDL-C (mmol/l)	TG (mmol/l)	AIP
I (pre)	2.3 ± 0.1[a]	1.2 ± 0.2[a]	0.6 ± 0.1[a]	0.5 ± 0.1[a]	0.9 ± 0.1[a]	-0.1 ± 0.0[a]
(post)	2.4 ± 0.1[a]	1.3 ± 0.1[a]	0.5 ± 0.0[a]	0.5 ± 0.0[a]	1.0 ± 0.1[a]	-0.1 ± 0.0[a]
II (pre)	2.3 ± 0.1[a]	1.2 ± 0.1[a]	0.6 ± 0.1[a]	0.5 ± 0.0[a]	1.0 ± 0.2[a]	-0.1 ± 0.0[a]
(post)	3.8 ± 0.1[b]	0.6 ± 0.0[b]	2.2 ± 0.1[b]	1.1 ± 0.1[b]	2.4 ± 0.1[b]	0.6 ± 0.2[b]

NOTE: Values are expressed as mean ± SEM. a, b within column signifies that means with different letters differs significantly at p<0.05 while means with the same letters does not differ significantly at p>0.05.

Table 4: Effect of diabetes on the Lipid profile and Atherogenic Index of plasma of control and diabetic groups pre- and post- alloxan administration.

The mean levels of TC, TG, LDL-C, VLDL-C and AIP were significantly increased (p<0.05) whereas the HDL-C level decreased significantly (p<0.05) in the diabetic group after alloxan administration, however, the baseline values in both groups did not differ significantly (p>0.05).

IV. DISCUSSION

Severe hyperglycemia occurs when rats are injected with alloxan (Helal et al., 2015). Diabetes induction by alloxan resulted in loss of body weight which could be as a result of the catabolic effect of diabetes on protein metabolism resulting in tissue and muscle wasting as well as breakdown. This result agrees with the works of Oyedepo (2012) and Enwenighi et al. (2015). The marked increase in glucose levels of diabetic rats may be as a result of damage of the pancreatic beta cells by alloxan, and consequently reducing insulin secretion. In the same context, some studies demonstrated that alloxan caused selective destruction of insulin secreting cells, thereby inducing hyperglycemia (Mir et al. 2006; Muthulingan, 2012). The elevated glycated hemoglobin, a marker of glycemic control in diabetic group may be explained by the persistent or uncontrolled hyperglycemia which has been reported to cause glycosylation of certain proteins like hemoglobin. This result agrees with the works of Daisy and Rajathi (2009) and Neeraj (2012).

The liver being an insulin-dependent tissue plays a vital role in glucose and lipid homeostasis, and as such is involved in diabetes (Seifter, 1982). The elevations in the serum levels of liver enzymes in this study is indicative of hepatocellular damage, and consequently the release of these enzymes from the cytoplasm into the circulation. The findings of this study is consistent with those of Erukainure et al. (2013) and Nwanjo (2007) but disagrees with that of Abubakar et al. (2009) who reported insignificant changes in the enzyme levels.

Diabetes is known to adversely affect the glomerulus, and the significant elevation in serum urea and creatinine levels recorded in this study is indicative of impaired renal function in the diabetic rats. This agrees with the reports of Eze (2012) and Neeraj (2012). The increase in TC, LDL-C, TG and AIP values as well as decreased HDL-C level which are risk factors of cardiovascular disease could be attributed to the action of hormone sensitive lipase which promotes lipolysis thus, increasing free fatty acids and triglyceride levels in the circulation. The free fatty acids are broken down to acetyl co-A, and channeled to the synthesis of cholesterol, thereby increasing blood cholesterol level (Oyedepo, 2012). These findings correspond with the reports of Erukainure et al. (2013), Obi et al. (2015) and Obi-Ezeani et al. (2016).

In conclusion, the findings of this study show that diabetes mellitus enhances the development of certain biochemical derangements which may be associated with some complications of diabetes.

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