

Comparative Effects of Antidiabetic Drug, Metformin and Deferoxamine, on Serum Lipids, Serum Ferritin and Endocrine Indicators of Diabetes Mellitus Complications in Streptozotocin Diabetic Rats

ABSTRACT

Aims: Increasing evidence has indicated that iron overload not only increases risks of insulin resistance and diabetes, but also causes cardiovascular diseases in non-diabetic and diabetic subjects. The present study compared the effects of metformin (met) and deferoxamine (DFX) on the serum lipids, serum ferritin and endocrine indicators of diabetes mellitus complications in streptozotocin experimental diabetes in rats.

Study design: Experimental diabetes was induced in overnight fasted rats by a single dose i.p each of nicotinamide and, 15 min after, STZ followed by administration of the antidiabetic drugs, met (os, 250 mg/kg b.wt) and DFX (i.p, 150 mg/kg b.wt), daily for 14 days. Blood and histological samples were collected and prepared for biochemical and histopathological analysis of indicators of cytotoxic side effects. Results were analysed statistically by Student t-test and analysis of variance (ANOVA).

Results: Both drugs caused progressively increased hypoglycaemic effect with repeated doses. However, Metformin showed markedly higher potency in hypoglycaemic activity than DFX. STZ diabetes caused hyperlipidaemic effect with respect to lipid profile parameters except HDL and treatment with antidiabetic drugs metformin and deferoxamine reversed the hyperlipidaemic effect. On the other hand, STZ caused hypolipidaemic effect with respect to HDL but the antidiabetic drugs reversed it. However, DFX is more potent than metformin in reversing the effect of STZ diabetes on HDL. STZ diabetes induced elevation of serum ferritin while inducing reduction in serum insulin level. Metformin treatment reversed the adverse effects of STZ on insulin secretion only, whereas, DFX significantly reversed the adverse side effects of STZ on both serum ferritin and serum insulin. However, metformin-treatment and DFX-treatment exhibited comparable potency in their effects on insulin secretion.

Conclusion: Evidence from histological study of the pancreas suggests that both metformin and DFX were sufficiently biologically significant to effectively reverse the disruptive effects on the pancreatic exocrine tissue of diabetic rats.

Keywords: Streptozotocin, serum ferritin, type 2 diabetes, dyslipidaemia.

1. INTRODUCTION

Diabetes mellitus (DM) is a major degenerative disease in the world today afflicting many lives both in the developed and developing countries [1]. Increasing incidence in the developing countries, especially in the younger age group, affecting mainly the people in the productive years of their lives is also of great concern [2]. The chronic hyperglycaemia of diabetes is associated with long-term damage, dysfunction, and failure of different organs, especially the eyes, kidneys, nerves, heart, and blood vessels [3], [4].

Type 2 diabetes (T2DM) is the most common form and comprises of 90% of people with diabetes around the world [5]. Type 2 diabetes is characterized by the combination of peripheral insulin resistance and inadequate insulin secretion by pancreatic beta cells [6], [7]. Diabetes is associated with profound alterations in plasma lipid and lipoprotein profile and with an increased risk of coronary heart disease [8]. Increased triglyceride (TG) and reduced high density lipoprotein cholesterol (HDL-C) levels are the key characteristics of dyslipidaemia in type 2 diabetes [8].

Epidemiological studies have reported a strong association between elevated serum ferritin concentrations and increased risk for diabetes [9], [10]. Excess iron impairs pancreatic β cell function and causes β cell apoptosis [10], [11]. Increasing evidence has indicated that iron overload not only increases risks of insulin resistance and diabetes, but also causes cardiovascular diseases in non-diabetic and diabetic subjects [12], [13]. Epidemiologic studies in overt iron overload states have shown that the incidence of cardiac disease is increased and that treatment with iron chelation improves cardiovascular outcome [14], [15].

The management of diabetes mellitus is considered a global problem and a successful treatment is yet to be discovered [16]. Metformin is considered a cornerstone in the treatment of diabetes and is the most frequently prescribed first line therapy for individuals with type 2 diabetes [17]. It is one of a few antihyperglycaemic agents associated with improvements in cardiovascular morbidity and mortality which is a major cause of death in patients with type 2 diabetes [18]. Metformin was approved by the Food and Drug Administration (FDA) for use in the United States in 1995 [19]. Metformin major effect is to decrease hepatic glucose output [20]. In addition, metformin decreases glucose absorption in the small intestine, increases insulin-mediated glucose utilization in peripheral tissues (such as muscle and liver), and has an antilipolytic effect that lowers serum free fatty acid concentrations, thereby reducing substrate availability for gluconeogenesis [20], [21].

Deferoxamine is the most common iron chelator in clinical use [22], [23]. The capacity of deferoxamine to chelate iron (Fe) and mediate its excretion in Fe-overloaded patients is well documented [24]. It has been approved for use in the USA since the late 1970's [25], [26]. Deferoxamine acts by binding free iron in the bloodstream and enhancing its elimination in the urine [27]. Deferoxamine works in treating iron toxicity by binding trivalent (ferric) iron (for which it has a strong affinity), forming ferrioxamine, a stable complex which is eliminated via the kidneys [28]. By removing excess iron, the agent reduces the damage done to various organs and tissues [27].

The social and financial burden of diabetes is mainly due to the complications. Because of this, a growing number of researches have focused on diabetes and its complications, with the aim to expand our knowledge about pathogenic and pathophysiological mechanisms, preventive strategies and potential novel therapies. This study was carried out to compare the effect of metformin on serum lipids, serum ferritin and endocrine indicators of DM with that of deferoxamine which act by a different mechanism in streptozotocin-induced diabetic rats to determine their effectiveness in treating/preventing diabetes and its complications and as an effort in the search for more effective therapeutic initiatives for type 2 DM.

2. MATERIALS AND METHODS

2.1 Drugs and Chemicals:

Streptozotocin (STZ), deferoxamine (DFX), nicotinamide (NA), citric acid, trisodium citrate, sodium hydroxide, sodium chloride, chloroforms, and formaldehyde were purchased from Zayo-Sigma Aldrich Chemicals Ltd, St Louis, MO, USA; metformin HCl (Diabetmim, Hovid Pharmaceutical, Pune Ma Harashtra, India) while insulin and ferritin ELISA kits were purchased from Syntiron Bioresearch Inc., Carlsbad, California, USA).

2.2 Experimental Animals:

Thirty five male Wistar strain albino rats weighing 172-233 g were used in this study. The rats were obtained from the Animal House Unit of the Federal College of Veterinary and Medical Laboratory Technology (FCVMLT), Vom. The rats were housed in clean metallic cages, kept in a well ventilated room and allowed to acclimatize to the laboratory condition of the Nigerian Institute for Trypanosomiasis Research (NITR) Vm, animal experiment room at $25\pm 2^{\circ}\text{C}$ with 12 hour

64 light/dark cycle for two weeks before being used. Animals were fed a standard animal pellet obtained from Dagwom farm of National Veterinary Research
65 Institute (NVRI), Vom, and had free access to water *ad libitum*. All animals were carefully monitored and all the experimental protocol with the animals was in
66 accordance with the internationally accepted principles for laboratory animal use and the experimental protocols were duly approved by the Institutional Ethical
67 Committee of NITR.

68 **2.3 Experimental Induction of Type 2 Diabetes in Rats:**

69 Diabetes was induced in overnight fasted rats by a single intraperitoneal (i.p) injection of freshly prepared streptozotocin (35 mg/kg b.wt) dissolved in 0.1M
70 citrate buffer (pH 4.5), 15 min after the intraperitoneal administration of nicotinamide (110 mg/kg), dissolved in normal physiological saline. The control (group
71 A) animals were injected i.p with the equivalent volume of the citrate buffer solution [29], [30].

72 Diabetes was confirmed by the symptoms of polydipsia, polyuria, glycosuria and elevated fasting blood glucose concentration, 48 hours and then one week
73 after STZ injection. Diabetes was developed and stabilized in STZ treated rats over a period of 7 days [31]. Following the criteria of previous workers [32], [33],
74 rats with stable glycosuria and blood glucose levels of more than 11 mmol/L (>11 mmol/L) were considered to be diabetic and, therefore, used for the study.

75 76 **2.4 Animal Treatment**

77 Following induction of diabetes, animals were randomly and evenly distributed into four groups A, B, C and D of six rats each (n=6) in clean metallic standard
78 rat cages. Animals in group A (normal control) and those in group B (diabetic control) were given only water *ad libitum*. While those in group C (diabetic) were
79 administered orally with antidiabetic drug metformin (Met) at a dose of 250 mg/kg body weight once daily for 14 days by forceful gavage. Rats in group D (also
80 diabetic) were administered intraperitoneally with iron-chelating drug deferoxamine (DFX) at a dose of 150 mg/kg body weight once daily for 14 days. The
81 dosages of drugs used in this study were pre determined in a pilot study. Baseline fasting blood glucose levels of each rat for all groups were measured before
82 diabetes induction and at the time of grouping of the animals. Blood glucose levels were estimated just before drug administration on day 0, 5, 10 and 14.

83 **2.5 Monitoring of Blood Glucose Level during Treatment**

84 All blood samples for monitoring of blood glucose level *in situ* were taken from the tail vein of the rats using 24 gauge needles at intervals of 0, 5, 10, and 15
85 days. Blood glucose level was determined by the glucose oxidase method using reactive strips and a single touch glucometer (Accu-Chek Active, Roche
86 Diagnostics, Mannheim, Germany). Results were initially recorded in mg/dl and then converted to mmol/l by dividing values in mg/dl by a factor of 18

87 **2.6 Blood Collection and Preparation**

88 After overnight fast, the animals were sacrificed on the 15th day under mild chloroform anaesthesia and blood was obtained via cardiac puncture. Blood sample
89 was transferred into plain centrifuge tube and allowed to clot at room temperature. It was then centrifuged within 1 hour of collection at 4000x g for 10 min on a
90 digital centrifuge (Biofuge 200, Bosch Medical Systems, Corinth, TX, USA) to separate the serum from the clot. The resultant serum sample were stored frozen
91 at -20°C. Prior to assay, frozen sera were completely thawed and well mixed and all reagents were allowed to attain room temperature.

92 **2.7 Histopathological Examination**

93 After sacrificing the animals on the 15th day post treatment, the pancreas of two animals from each group were excised and immediately fixed in 10% neutral
94 buffered formalin solution after washing with normal saline. The resultant fixed tissue samples were used for histopathological examination in the
95 Histopathology Laboratory of FCVMLT and Diagnostic Center of NVRI, Vom, using the routine procedures developed in the respective laboratories. The tissues
96 were washed, dehydrated with alcohol and cleared with xylene. Serial sections of 4 µm thickness were cut using a rotary microtome (ERM-200P, Erma, Tokyo,
97 Japan). The sections were deparaffinised with xylene and hydrated in descending grades of alcohol. The slides were then transferred to haematoxylin for 10
98 min, followed by rinsing with water. These were examined and later counterstained with eosin, rinsed with water, dehydrated with ascending grades of alcohol,
99 cleared with xylene and mounted. The slides were observed using a light microscope.

100 2.8 Biochemical Analysis of Rat Serum Sample

101 Serum triglyceride (TG), total cholesterol (TC), and high density lipoprotein cholesterol (HDL-c) levels were measured colorimetrically by enzymatic point
102 methods using Roche-Hitachi kit via Automatic Analyzer (HITACHI 902, Roche Diagnostics, Mannheim, Germany) according to the procedure described in the
103 manufacturers' operation manual. Serum insulin and ferritin levels were estimated by enzyme linked immunosorbent assay (ELISA) technique using ELISA kits
104 (Syntron Bioresearch Inc., Carlsbad, California, USA).

105 2.9 Statistical Analysis

106 All statistical analysis was performed with SPSS software version 16.0. Data are expressed as the means ± standard error of mean (S.E.M). Student's t-
107 test and analysis of variance (ANOVA) were employed in comparing means of continuous variables as appropriate. Differences were considered statistically
108 significant if $p < 0.05$.

109 3. RESULTS

110 The results on the effect of metformin and deferroxamine on the fasting blood glucose levels of diabetic rats are summarised on Figure 1. The diabetic control
111 animals (group B) exhibited gradually increased hyperglycaemia. There was a significant ($p < 0.05$) elevation in fasting blood glucose level in the diabetic control
112 (group B) when the values for Days 0, 5, 10 and 15 were compared to the corresponding values in the normal control rats. Oral administration of metformin
113 (250 mg/kg/day) significantly ($p < 0.05$) reduced the elevated levels of blood glucose on the 5th, 10th and 15th day of treatment compared to the corresponding
114 values in the untreated diabetic control ($p < 0.05$). On the other hand, intraperitoneal administration of deferroxamine (150 mg/kg/day) caused significantly
115 ($p < 0.05$) reduced blood glucose level on the 10th day and 15th day of treatment only compared to the diabetic control. This would suggest that the onset of
116 induced hypoglycaemic effect occurred 5 days earlier in rats given oral metformin than in those given intraperitoneal DFX. Both drugs caused progressively
117 increased hypoglycaemic effect with repeated doses. As at Day 15, the % reduction in blood glucose level induced by met (35.58%) was significantly ($p < 0.05$)
118 higher than that (16.6%) induced by DFX, an indication that metformin was more potent than DFX at the experimental condition. Similarly, from Day 5, the
119 blood glucose level of diabetic rats given DFX (group D) were consistently and significantly ($p < 0.05$) higher than those given met (group C). This is another
120 indication that metformin is more potent than DFX. Normal control rats (group A) did not exhibit any significant alterations in blood glucose levels during the
121 experiment.

122 The results on the comparative effects of metformin and deferroxamine on the various lipid risk factors are summarised on Table 1. There was a significant
123 ($p < 0.05$) decrease in the level of serum high density lipoprotein (HDL) and a significant ($p < 0.05$) increase in the levels of total cholesterol, triglycerides and low
124 density lipoprotein (LDL) in diabetic control rats (group B) compared to normal control rats (Group A), suggesting that experimental diabetes in rats caused a

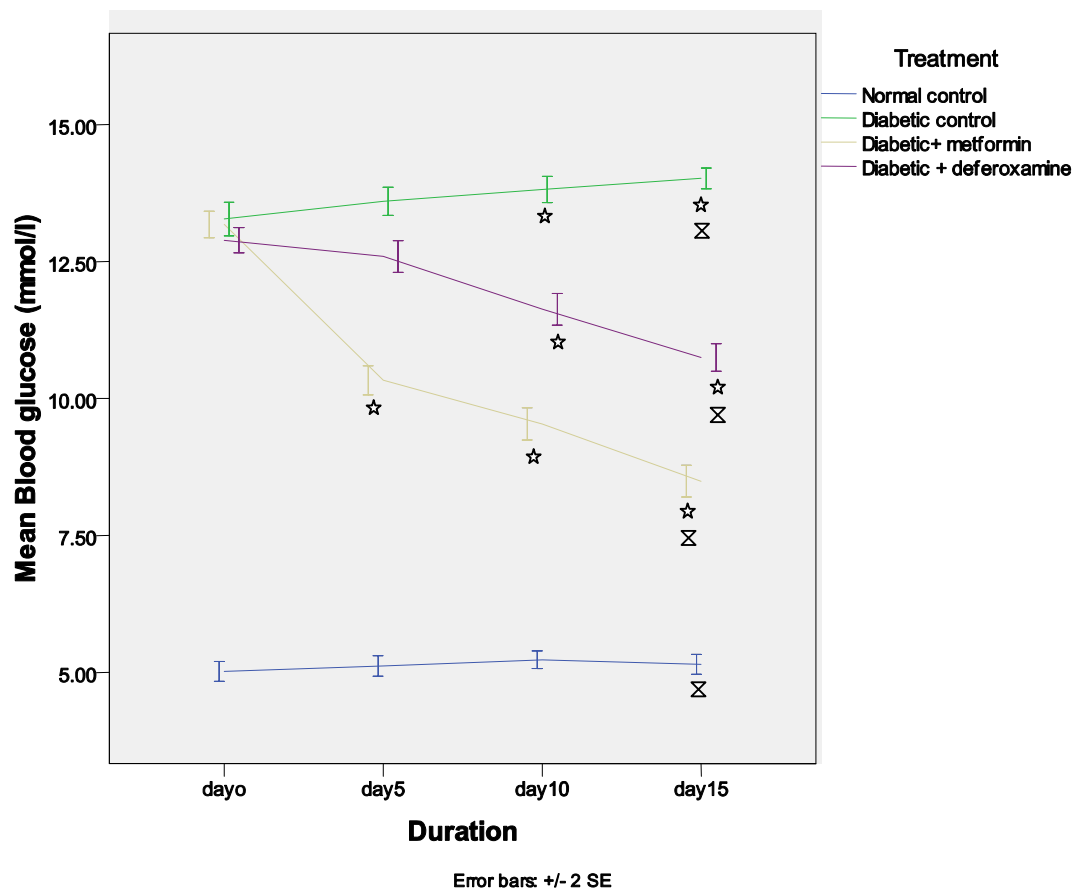
125 reduction in serum HDL but elevation of serum total cholesterol, triglyceride and LDL levels. Administration of metformin to STZ-NA induced diabetic rats
126 (group C) led to a significant reduction ($p < 0.05$) in the level of triglycerides, total cholesterol, and LDL and significant increase ($p < 0.05$) in the level of HDL,
127 compared to diabetic control rats (group B). Similarly, deferroxamine treated rats (Group D) showed significant ($p < 0.05$) reduction in the levels of triglycerides,
128 total cholesterol and LDL and increased the HDL significantly ($p < 0.05$) compared to diabetic control rats after 15 day of treatment. These observations indicate
129 that STZ diabetes caused hyperlipidaemic effect with respect to lipid profile parameters except HDL and that treatment with antidiabetic drugs metformin and
130 deferroxamine reversed the hyperlipidaemic effect. On the other hand, STZ caused hypolipidaemic effect with respect to HDL but the antidiabetic drugs
131 reversed it. The mean serum HDL level in DFX-treated rats was significantly ($p < 0.05$) higher than that of metformin-treated rats, suggesting that DFX is more
132 potent than metformin in reversing the effect of STZ diabetes on HDL. Mean serum total cholesterol, triglyceride and LDL levels were higher in metformin
133 treated rats than in DFX- treated animals but the difference were not statistically significant.

134 The comparative effects of metformin and deferroxamine on serum insulin and ferritin levels are summarised in Table 2. The mean serum ferritin level was
135 significantly higher ($p < 0.05$) while serum insulin level was significantly lower ($p < 0.05$) in the diabetic control (group B) than in the normal control (group A),
136 suggesting that STZ diabetes induced elevation of serum ferritin while inducing reduction in serum insulin level probably by causing cytotoxic side effects on the
137 pancreatic tissue.

138 The serum ferritin level was lower while the serum insulin level was higher in STZ diabetic rats treated with metformin (group C) than in the diabetic control
139 controls (group B) but only in the case of serum insulin was the difference statistically significant ($p < 0.05$). This is an indication that metformin treatment
140 reversed the adverse effects of STZ on insulin secretion only. Whereas, DFX significantly reversed the adverse side effects of STZ on both serum ferritin and
141 serum insulin as evidenced by the significantly ($p < 0.05$) lower level of ferritin and significantly ($p < 0.05$) higher level of insulin in DFX treated rats (group D)
142 compared to diabetic controls (group B). The mean serum ferritin level of DFX-treated diabetic rats was significantly lower than those of metformin-treated
143 diabetic rats, an indication that deferroxamine is more potent than metformin in reducing serum ferritin. On the other hand, there is no significant difference
144 ($p > 0.05$) between the mean serum insulin levels of metformin treated diabetic rats and those of DFX-treated diabetic rats, suggesting comparable potency. This
145 observation was corroborated by histopathological findings on the pancreas tissue (see plate 1). Evidence suggests that they were sufficiently biologically
146 significant to effectively reverse the disruptive effects on the pancreatic exocrine tissue. Compared to the diabetic control which showed degeneration of the
147 pancreatic beta cells of the Islet of Langerhans (Plate 1B), pancreatic tissues of the rats given metformin (Plate 1 C) and deferroxamine (Plate 1 D) showed
148 normal –appearing pancreatic lobular structure and preserved endocrine Islet of Langerhans.

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151

152 **Figure 1: Effect of Metformin and Deferoxamine on Blood Glucose Level (Mmol/L) during 14 Days Treatment in Diabetic Rats**

153 ☆ Statistically significant at $p < 0.05$ compared with day 0 of same treatment group

154 ⌘ Bars with this superscripts differ from each other statistically at $p < 0.05$ when compared between treatment groups at day 15

Table 1: Mean Serum Lipid Profiles of Streptozotocin Diabetic Rats following 14-Day Treatment with Metformin and Deferoxamine

Group	Treatment	Serum lipid concentration (mmol/L)			
		Total Cholesterol	Triglyceride	HDL	LDL
A	Normal control	1.10± 0.06	0.75± 0.06	0.60± 0.02	0.23± 0.07
B	Diabetic control	1.48± 0.05 ^a	1.50± 0.19 ^a	0.42± 0.02 ^a	0.55± 0.13 ^a
C	Diabetic + Met	1.22± 0.05 ^b	0.93± 0.05 ^b	0.49± 0.02 ^{ab}	0.30± 0.05 ^b
D	Diabetic + DFX	1.18± 0.04 ^b	0.82± 0.05 ^b	0.56± 0.02 ^{bc}	0.26± 0.04 ^b

Tabulated Values are means ± S.E.M for six rats in each group; Statistical significance between means was assessed using one-way analysis of variance followed by Duncan's test as a post-analysis of variance test

^aStatistically significant difference at p<0.05 compared to the normal control (group A)

^bStatistically significant difference at p<0.05 compared to the diabetic control (group B)

^cStatistically significant difference at p<0.05 compared to the diabetic + met treated (group C)

Abbreviations: HDL=high density lipoprotein; LDL= low density lipoprotein; Met= metformin; DFX= deferoxamine.

180 **Table 2: Mean Serum Insulin and Ferritin Levels in Streptozotocin Diabetic Rats Following 15-Day Treatment with Antidiabetic Drug**
181 **Metformin and Iron Chelating Drug Deferoxamine**

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Group	Treatment	Serum Insulin (μ u/ml)	Serum Ferritin (ng/ml)
A	Normal control	1.42 $\pm$ 0.18	1.21 $\pm$ 0.01
B	Diabetic control	0.85 $\pm$ 0.04 ^a	1.25 $\pm$ 0.01 ^a
C	Diabetic + Met	1.33 $\pm$ 0.08 ^b	1.24 $\pm$ 0.01 ^a
D	Diabetic + DFX	1.38 $\pm$ 0.19 ^b	1.20 $\pm$ 0.01 ^{bc}

183 Tabulated values are means $\pm$ S.E.M for six rats in each group; Statistical significance between means was assessed using one-way analysis of variance
184 followed by Duncans test as a post-analysis of variance test

185 ^aStatistically significant difference at p<0.05 compared to the normal control (group A)

186 ^bStatistically significant difference at p<0.05 compared to the diabetic control (group B)

187 **Abbreviations: Met= metformin; DFX= deferoxamine**

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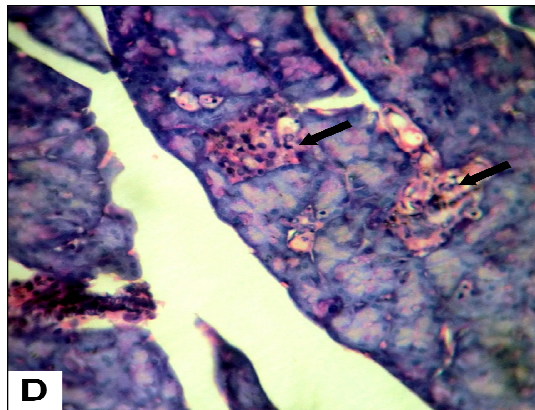
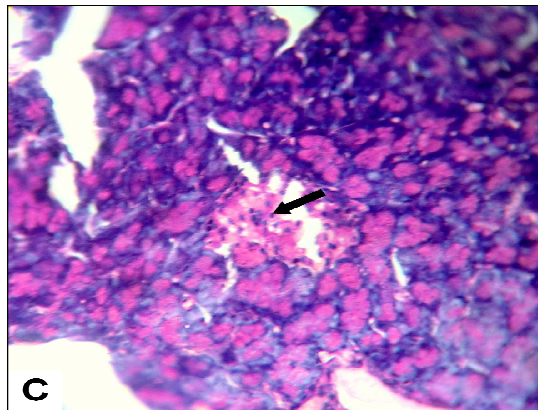
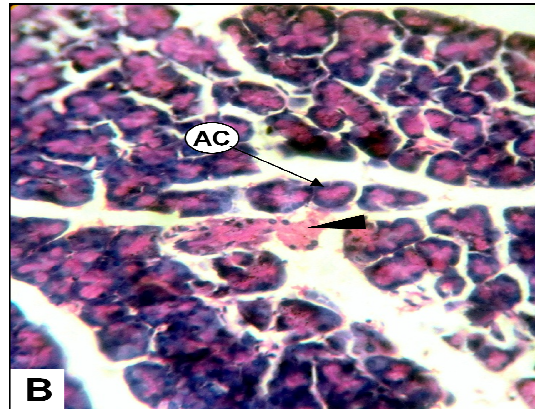
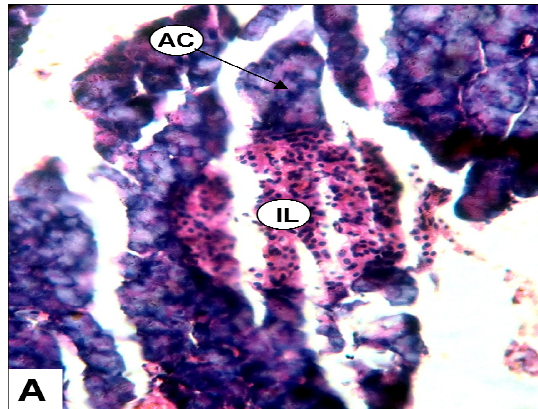


Plate 1: Histopathological Changes in Pancreatic Tissue of Control and Experimental Rats.

A. Normal control (Group A) shows normal exocrine acini (AC) and endocrine Islet of Langerhans (IL); **B. Diabetic control (Group B)** shows a severely reduced number of Islets as well as depleted Islet (arrow head); the Islet cells have been replaced by an eosinophilic acellular substance; **C. Metformin treated (Group C)** shows normal-appearing pancreatic lobule with preserved Islets (arrow); **D. deferoxamine treated (group D)** shows a normal pancreatic lobule with increased population of preserved Islets (arrows)

H & E Stain; A and C (X10), B and D (X30)

196 **4. DISCUSSION**

197 Streptozotocin (STZ)-nicotinamide (NA) type 2 model shares a number of features with human type 2 diabetes both histologically and metabolically and is
198 characterized by moderate stable hyperglycaemia. Hence, STZ-NA induced diabetes model was used in the present study [34], [35]. In the present study,
199 streptozotocin injection caused β cells degeneration in rats, therefore, release of insulin by the pancreas was decreased resulting in hyperglycaemia. This
200 confirms induction of experimental diabetes in rats. This is similar to the findings of several earlier researchers [36], [37].

201 The results indicate that metformin has antidiabetic potentials as has been established by several studies [4]. This is evident in the rapid reduction in blood
202 glucose levels that were noticed once administration of the drug commenced as well as the alleviation of all symptoms of diabetes that were initially noticed
203 before commencement of treatment. Metformin (given o.s) showed markedly higher potency than deferoxamine (given i.p) in hypoglycaemic activity (earlier
204 time onset, higher % reduction); despite the fact that i.p is faster route than o.s, yet metformin acted earlier. The blood glucose lowering effect of metformin is
205 dependent on presence of insulin [21], therefore, according to the results of the present work, it seems that administering STZ-NA in rats was not able to
206 degenerate all pancreatic β -cells and so did not lead to a complete lack of β -cell population as was obvious from the histological findings.

207 The fundamental mechanism underlying hyperglycaemia in diabetes mellitus involves the over production of glucose and or decreased utilization of glucose by
208 the tissues [21], [38]. The mechanisms of metformin-mediated improvement of insulin sensitivity have remained obscure, however, multiple pathways of action
209 have been proposed, including a decrease of hepatic glucose production, an increase of peripheral glucose utilization and a reduction of intestinal glucose
210 absorption [4], [39]. Metformin also increases low-affinity and high-affinity receptors of insulin, and improves insulin resistance [40].

211 Administration of deferoxamine to diabetic rats caused significant reduction of blood glucose and serum ferritin concentration in the present study. The
212 observed effect of deferoxamine on blood glucose and serum ferritin level may be due to improved utilization of glucose by peripheral tissue through increase in
213 insulin receptor activity and signaling in hepatocytes is in line with the findings of Dongiovanni *et al.* [41], who investigated the effect of iron depletion by
214 deferoxamine on insulin signaling and glucose uptake in HepG2 hepatocytes and in rat liver and reported that deferoxamine induced the constitutive glucose
215 transporter Glut1 and the insulin receptor. Use of phlebotomy or iron-chelation therapy to reduce ferritin levels was associated with improved glucose tolerance
216 in patients with hereditary hemochromatosis (HH), healthy blood donors, patients with metabolic syndrome, and patients with T2DM [42], [43]. Epidemiological
217 study showed that even at 'normal' levels, iron exerted detrimental effects on pancreatic beta-cell function, and that these effects were reversible with dietary
218 restriction or iron-chelation therapy [11].

219 In this study, lipid profile alterations produced by STZ-NA administration to experimental rats is in agreement with the findings of other researchers who did
220 report significant changes in lipid abnormalities in type 2 diabetes [4], [30]. The beneficial improvement in lipid profile following metformin administration
221 obtained in the present study seem to corroborate previous findings that this drug does ameliorate dyslipidaemia [39], [44]. Lipid abnormalities are due to
222 resistance to insulin and hyperglycaemia [45], [46]. Improvement in insulin secretion in the islets of Langerhans by administration of metformin and
223 deferoxamine to diabetic rats which was evident in this study may have led to improvement in the serum lipids levels [37], [47]. It has been documented that
224 metformin exerts direct effects on hepatic glucose and lipid metabolism and suppresses lipogenic enzymes, particularly acetyl-CoA carboxylase (ACC) activity
225 via an 5'AMP activated protein kinase (AMPK)-dependent pathway, thus leading to decreased lipogenesis but increased fatty acid oxidation [20], [39].
226 Epidemiologic studies in overt iron overload states such as transfusional iron overload and hemochromatosis have shown that the incidence of cardiac disease
227 is increased and that treatment with iron chelation improves cardiovascular outcome [14], [15].

229 **5. CONCLUSION**

230 The intraperitoneal injection of STZ-NAD induced type 2 diabetes and caused hyperlipidaemic effect with respect to serum TC and TG profile, but
231 hypolipidaemic effect with respect to HDL-C in experimental rats. Treatment with either antidiabetic drug metformin or deferoxamine, reversed the
232 hyperlipidaemic effect on serum TC and TG and the hypolipidaemic effect on HDL-C.

233
234 Deferoxamine was more potent than metformin in reducing serum ferritin and in mitigation of serum HDL in diabetic rats while metformin showed markedly
235 higher potency in hypoglycaemic activity (earlier time onset, higher % reduction). If epidemiological studies confirm an aetiologic role of iron overload in human
236 subjects, chelating iron using deferoxamine may be useful in treating diabetes in future. In addition, iron chelating drug may be a potential drug for ameliorating
237 diabetic complications.

238 **COMPETING INTERESTS**

239 Authors have declared no competing interests exit.

240 **CONSENT**

241 Informed consent was used in the recruitment of the participants and confidentiality was maintained in accordance with standard medical practice

242 **ETHICAL APPROVAL**

243 Ethical approval was given by the Ethics Committees of Jos University Teaching Hospital, Plateau Specialist Hospital and ECWA Evangel Hospital Jos.

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