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Pharmacology

Comparative Histopathological Effects of Metformin and Glibenclamide on Kidney in Alloxan Induced Diabetic Albino Rats

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Original Research Article

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Abstract: Diabetes mellitus is a systemic metabolic disorder characterised by elevated blood glucose levels due to absolute or relative deficiency of insulin secretion from pancreatic β-cells. Clinically this disease is associated with a number of chronic complications including nephropathy, neuropathy, retinopathy, cardiovascular diseases and is responsible for causing impairment in kidney functions. Hence in this study we evaluated the histopathological effect of Metformin and Glibenclamide on kidney in Alloxan induced diabetic Albino rats.The present study was conducted on 24 experimental animals which were divided into four groups of with 6 rats in each group and were treated accordingly Group 1:Healthy control (HC) rats, Group 2:Diabetic control (DC) rats,Group 3:Diabetes mellitus (DM) + Metformin(M) rats, Group 4: Diabetes mellitus (DM)+ Glibenclamide(G) rats. After 28 days of treatment rats were sacrificed and histopathological study of kidney was done.Blood glucose and body weight was also determined.The results showed that both Metformin and Glibenclamide reversed Alloxan induced diabetic histopathological changes in kidney in Albino rats. The present Results demonstrate that normoglycemia with metformin and glibenclamide ameliorates diabetic induced histopathological lesions in kidney
Keywords: Diabetes, histopathology, Kidney, Metformin, Glibenclamide.

INTRODUCTION

Diabetes mellitus is a systemic metabolic disorder characterised by elevated blood glucose levels due to absolute or relative deficiency of insulin secretion from pancreatic β-cells. The individual with diabetes has a 25-fold increased risk of blindness, a 20-fold increase in the risk of renal failure, a 20-fold increase in the risk of amputation as a result of gangrene and a 2 to 6 fold increased risk of coronary heart disease and ischemic brain damage. Renal impairment is one of the serious and common diabetic complications**.** Elevated serum levels of urea and creatinine are significant markers of renal damage.

The major goal in the treatment of diabetes has been to keep both short-term and long term glucose levels within acceptable limits, thereby reducing the risk of long term complications

Histopathological studies have revealed that the alloxan-induced diabetic rats, display feathery degeneration, thickening of glomeruli, inflammatory cells and severe congestion in kidney tissue. Whereas metforminand glibenclamide treated albino rats showed protection from alloxan- induced changes in kidney.

AIM AND OBJECTIVES

To Compare the Histopathological effects of Metformin and Glibenclamide on kidney in Alloxan-Induced Diabetic Albino Rats.

MATERIALS AND METHODS

The present study is based on the findings carried out on a total of 24 albino rats weighing between 120-160gm. The rats were procured from the Central Animal House, Department of Pharmacology, Govt. Medical College, and Jammu. The study was conducted after getting clearance from Institutional Animal Ethics Committee (IAEC).

24 experimental animals were divided into four groups of 6 rats each and each group was administered drugs as follows:-

Group 1: Healthy control (HC) rats served as controls and were administered only Normal saline (0.5ml/day) orally.

Group 2: Diabetic control (DC) rats were induced with diabetes using alloxan (150mg/kg intraperitonially) and were not given any form of treatment throughout the study.

Group 3: Diabetes mellitus (DM) + Metformin (M) rats were induced with diabetes by alloxan and treated with standard drug metformin 500mg/kg orally for 28 days.

Group 4: Diabetes mellitus (DM) + Glibenclamide(G) rats were induced with diabetes by alloxan and treated with glibenclamide 10mg/kg for 28 days orally**.**

The animals were kept in clean plastic cages in a well ventilated room and were maintained at room temperature of (25 ± 2^0c). Rice husk was used as bedding material. All animals were fed with rat feed and water ad-libutum throughout the experimental period. Their cages were cleaned of waste daily.

The animals were weighed and injected alloxan 150mg/kg dissolved in distilled water using insulin syringe via intraperitoneal route**.** Diabetes mellitus was confirmed after 75 hours of alloxan injection by testing the blood glucose levels using glucometer and glucose test strip. Animals with blood glucose level of 250mg/dl and above were considered diabetic and were given metformin and glibenclamide orally for 28 days after dissolving these drugs in distilled water. Albino rats of all groups were sacrificed after 28 days by keeping them in an inverted glass jar containing a large piece of cotton soaked in chloroform, so that the process can occur without pain and discomfort as recommended by Laboratory Animals Information Service Centre**.**

OBSERVATIONS

Blood glucose and body weight of albino rats of 4 different groups were observed on zero, 7,14 and 28 day of experimental study shown below

Table-1: Blood glucose estimation

Group	Day zero (Drug started orally)	Day 7	Day 14	Day 21	Day 28
Healthy control	88	86	89	88	90
Diabetic control	260	275	305	312	325
Diabetes mellitus+Metformin	262	225	200	162	106
Diabetes mellitus+Glibenclamide	255	234	208	135	114

Table-2: Mean body weight estimation

Group	Day zero (Drug started orally)	Day 7	Day 14	Day 21	Day 28
Healthy control	150	162	187	210	230
Diabetic control	156	154	140	132	125
Diabetes mellitus + Metformin	156	150	138	127	120
Diabetes mellitus + Glibenclamide	160	164	168	171	177

MICROSCOPIC OBSERVATION

Group 1 (Healthy control)

Histologically, kidney of control rats of Group 1 contained numerous nephrons consisting of the renal corpuscle, the proximal convoluted tubule, thin and thick limbs of loop of Henle and distal convoluted tubules. The renal corpuscle contained a tuft of capillaries, the glomerulus surrounded by Bowman's capsule. In between glomerulus and Bowman's capsule, there was urinary space. The proximal convoluted tubule was lined by simple cuboidal epithelium with brush border, where as the distal convoluted tubule was lined by simple cubiodal epithelium (Fig. A).

Group 2 (Diabetic control)

Kidneys of Alloxan induced diabetic rats of Group 2 showed glomerular alterations. The glomerular size was expanded and congested in diabetic rats resulting in reduction in Bowman's space (Fig. B). Glomeruli were infiltrated by inflammatory cells. Degenerated tubules were observed in cortex. In some tubules, the cells had separated from the basement membrane and collected in the center. Many tubules had completely sloughed off epithelium. Cells at other places were separated from one another also and were shed into the lumina of tubules. The lumina contain the cellular debris.

Group 3 (Diabetic control+Metformin)

Kidney of Alloxan induced diabetic rats treated with metformin showed normal sized glomerulus with normal Bowman's space and mild degeneration of renal tubules (Fig.C).

Group 4 (Diabetic control + Glibenclamide)

Kidney of Alloxan induced diabetic rats treated with glibenclamide showed normal architecture of the cortex and medulla. In the cortex, the renal corpuscles appeared normal with mild congestion in the glomeruli. Renal tubular degeneration was also seen (Fig D).

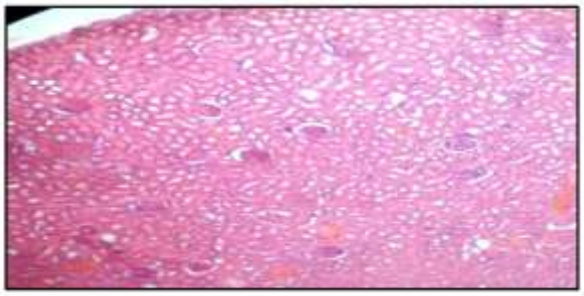

Fig-A: Photomicrograph of the longitudinal section of kidney of Healthy control group 1 rat showing normal architecture of cortex containing glomeruli, PCTs, DCTs and interstitial tissue containing blood vessel. Haematoxyline and Eosin 100X

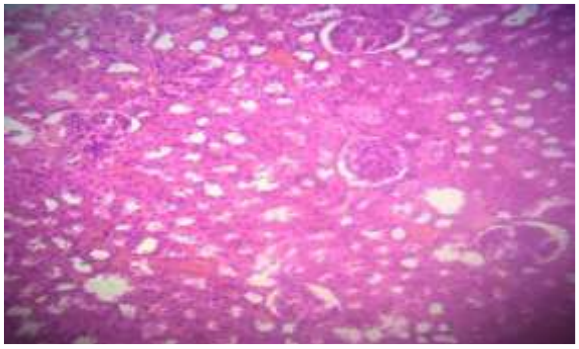

Fig-B: Photomicrograph of the longitudinal section of kidney of Diabetic control group 2 rat showing congested and expanded glomerulus with decreased Bowman's space, degenerated renal tubules .Haematoxyline and Eosin 100X

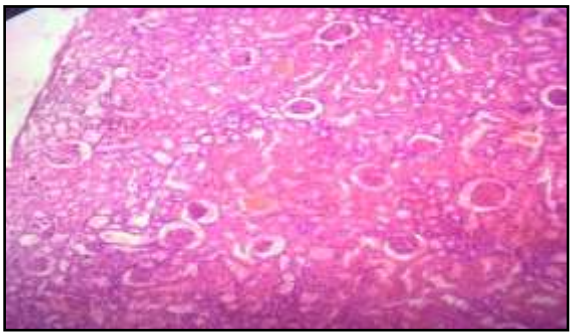

Fig-C: Photomicrograph of the longitudinal section of kidney of Diabetic mellitus+metformin group rat showing normal sized glomeruli with normal Bowman's space, degeneration of renal tubules. Haematoxyline and Eosin 100X

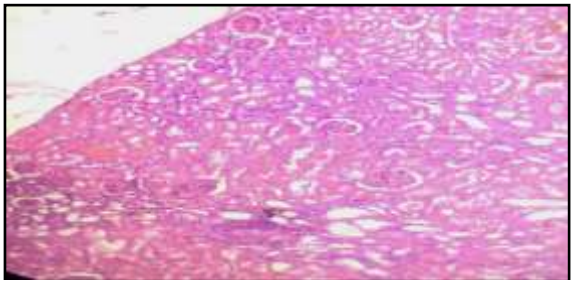

Fig-D: Photomicrograph of the longitudinal section of kidney of Diabetic mellitus+glibenclamide group 4 rat showing normal size glomeruli with normal Bowman's space, tubular degeneration.Haematoxyline and Eosin 100X

DISCUSSION

Diabetes mellitus is a systemic metabolic disorder characterized by elevated blood glucose levels due to absolute or relative deficiency of insulin secretion from pancreatic β-cells.

Increase in blood glucose level leads to structural and functional changes in target organs of diabetic patients. In the present study alloxan monohydrate, a toxic glucose analogue was used for induction of diabetes in albino rats.

EFFECT ON BODY WEIGHT

In the present study a decrease in body weight was observed in albino rats after induction of Diabetes using alloxan. This is in agreement with previous studies done by Kumar S *et al*. [1] and Devi YA *et al*. [2].

Also in the present studytreatment of alloxan induced diabetic rats with metformin 500mg/kg body weight orally for 28 days showed the significant reduction in the body weightwhen compared to healthy control group. Similar findings were observed by Afzal M *et al*. [2] in their study**.**

In the present study we also observed that after treatment with glibenclamide in oral dose of 10mg/kg body weight for 28 days there was significant increase in body weight when compared to diabetic control group.This is in accordance with previous studies done by Yassin MM *et al.* [3],Kumar S *et al*. [1]and Devi YA *et al.* [2].

EFFECT ON BLOOD SUGAR

In the present study we observed that treatment of diabetic induced rats with metformin showed significant decrease in blood glucose level. Similar results were observed by Kianifard D *et al.* [4] and Khadre SEM *et al*. [5] which is in accordance with present study.

In present study we also observed that treatment of Diabetic induced rats with glibenclamide for 28 days showed significant decrease in blood glucose level. The present study is in agreement with Kumar S *et al*. [1] and Sunil C *et al*. [6].

MICROSCOPIC CHANGES IN KIDNEY

In the present study we observed that kidney of alloxan induced diabetic rats showed expanded glomerular size with hypercellularity, sloughing of epithelial cells of proximal convoluted tubule, degenerated renal tubules, and cellular debris in the lumen of tubules.The present study is in agreement with Sunil C *et al.*[6]. Similar study was conducted byKhadre SEM *et al.* [5]who observed similar findings except for cellular debris. Hence our study is in partial agreement with Khadre SEM *et al.* [5].

In the present study daily administration metformin (500mg/kg) for 28 days in alloxan induced diabetic ratsshowed normal sized glomerulus with normal bowman's space and mild degeneration of renal tubules. The present study is in agreement with. Nasri H and Rafieian-Kopaei M [7]and Emam HT[8].

In present study daily administration glibenclamide (10mg/kg) for 28 days reversed diabetic induced changes which was revealed by normal appearance of the renal corpuscle with mild congestion in the glomeruli and degeneration of tubules at certain areas. A similar study done by Mahood AKS [2] revealed that after treatment with glibenclamide kidney tissue of alloxan induced diabetic albino rat showed healing features, which resembles that of normal kidney. The present study is in agreement with Mahood AKS [2].

CONCLUSION

In conclusion the present Results demonstrate that normoglycemia with metformin and glibenclamide ameliorates diabetic induced histopathological lesions in kidney. Thus the frequent biochemical and laboratory analysis is important to check the occurrence of complications during the course of treatment.

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