REVIEW ARTICLE

Metformin Contributed with Lactic Acidosis in The White Male Rats

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ABSTRACT

Metformin treatment associated with development lactic acidosis (MALA) is a clinical problem. Recently, not found any drug to decrease or prevent MALA. The present study is designed to evaluate the advantage and disadvantages of metformin drugs in white male rats. A sample of 30 white male rats were randomly divided into three groups each group contain ten rats.: Group one administrated distal water to kept as control group for two months, Group two administrated metformin at dose 250 mg/kg for two months, and Group three administrated metformin at dose 500 mg/kg for two months. After the end of the experiment, two months, the rats were sacrificed to obtain the blood and tissues for analysis. The results show no significant change ($p > 0.05$) in the final weight of rats and the weight of the kidney and liver relative to the bodyweight as well as, the results show no significant change ($p < 0.05$) in the levels of urea and creatinine in the serum of rats treated with metformin drug. Also, the results appear no significant change ($p > 0.05$) in the liver enzymes include aspartate aminotransferase, alanine aminotransferase, alkaline aminotransferase, gamma-glutamyltransferase and total bilirubin in the metformin-treated groups relative to controls. In conclusion, the present study recorded not found harmful effects in the liver and kidney after taking metformin against diabetes except lactic acidosis state after using a drug for a long time.

Keywords: Kidney, Liver, MALA, Metformin, Rats

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INTRODUCTION

Metformin an oral medication regularly used to treat patients with type two diabetes and is suggested, in conjunction with a way of life adjustment, (for example, eat less and weight control).[1] It is a first-line oral treatment in the ongoing rules of the American Diabetes Association and European Association of the Study of Diabetes.[2] It has been accounted for that MET keep up concentrated glucose control and decline the danger of diabetes-related end focuses and demise,[3] MET treatment in T2D is related with less hypoglycaemic assaults when contrasted and insulin and sulfonylureas. The decrease in cardiovascular mortality by MET contrasted, and other oral diabetic operator has been affirmed in excess of 30 clinical preliminaries.[4] Ongoing clinical preliminaries recommend that MET may have remedial potential in different conditions, including diabetic nephropathy, cardiovascular infections, polycystic ovary ailment and treatment of tumor. In spite of its clinical helpfulness, little is thought about the tissue harmfulness of MET.[5]

Be that as it may, clinicians are terrified of one uncommon symptom of metformin treatment; the lactic acidosis (LA) caused most presumably by mitochondrial brokenness. The causality of an event of LA with metformin treatment is questionable. The danger of metformin-related LA (MALA) ranges from 2 to 9/100 000 patients.[5,6]

Be that as it may, unconstrained announcing of LA is growing.[5-7] MALA conveys half mortality which is high.7 While different investigations have announced 26 to 30% mortality.[5,7,8]

Sulphonylureas (SUs, for example, "glimepiride and glibenclamide (GB), are generally utilized oral antidiabetic drugs which empower insulin emission by hindering an ATP-subordinate potassium channel (KATP channel) on plasma film of pancreatic β-cells.[6] In spite of the overall utilization of SUs, loss of β-cell mass and work, and hypoglycemic scene have raised concern concerning its utilization.[7] Studies have demonstrated that SUs may instigate apoptosis in β-cell lines and rat islets,[8] and its treatment disappointment is normal in long haul treatment.[9] In any case, some confirmation has proposed that interminable utilization of SUs may prompt endoplasmic reticulum (ER) worry in β-cells, which at long last causes weariness of β-cell work,[10] and the decrease in β-cell work causes the dynamic weakening of glycemic control. The utilization of SUs for the treatment of T2D may subsequently have an injurious impact on different tissues in the body.[11]

In perspective of the over, this investigation was intended to get a complete review of the dangerous impact of MET in the liver and kidney of male rats.

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MATERIALS AND METHODS

Chemicals

Metformin hydrochloride was purchased from the Merck Company, United Kingdom.

Animals

Adult male rats weighting between 200–230 g were breeds in the animal house/faculty of Science/Kufa University animals were kept in ventilated cages at room temperature (28–30ºC) and maintained on standard laboratory chow and water *ad libitum*.

Experimental design

Thirty male rats (*Rattus norvegicus*) were kept for one week for acclimatization before the experiment and randomly divided into three groups each group have ten rats: Group one administrated distal water to kept as control group for two months, Group two administrated metformin at dose 250 mg/kg for two months, and Group three administrated metformin at dose 500 mg/kg for two months. Drugs were prepared with distal water and given daily to the animals by oral gavage for two months.

Blood collecting

Blood collecting after the last dose of metformin by heart puncture, each rat was anesthetized by the mixture of 0.1 ml of xylazine and 0.5 mL of ketamine,[14] blood kept in gel tube for 15 minutes in room temperature after this blood was separated by centrifuging at 3000 rpm for 10 minutes, the sera obtained were kept in deep freeze for biochemical analysis.

Biochemical analysis

Urea and creatinine were analyzed by methods of (15), (16) respectively, liver enzymes levels AST, ALT were determined by methods of (17), (18), while ALP was measured by method of (19) and ϒ-GG was determined by method of (20), finally, total bilirubin was analyzed by method of (21)

Histopathology of tissues

"Tissues fixed in 10% formalin and Bouin's solution was dehydrated in 95% ethanol and then cleared in xylene before embedded in paraffin. Micro sections (about 4 µm) were prepared and stained with hematoxylin and eosin (H&E) dye, and were examined under a light microscope by a Histopathologist who was ignorant of the treatment groups.[22]

Statistical analysis

All values were expressed as the mean ± S.E. of ten animals per group. Data were analyzed using one-way ANOVA using SPSS (10.0). Values were considered statistically significant at $p < 0.05$ ".[23]

RESULTS

Administration of MET at doses of 250 and 500 mg/kg body weight did not significantly ($p > 0.05$) affect the weight-gain and relative weights of liver and kidney of the rats (Table 1). Also, there were no significant ($p > 0.05$) differences in the levels of serum creatinine and urea in MET treated rats (Table 2). MET treatment did not produce significant ($p > 0.05$) alteration in the activities of serum aspartate aminotransferase, serum alanine aminotransferase serum alkaline phosphatase, ϒ-glutamyl transferase and total bilirubin relative to controls Table 3).

Table 1. Effect of metformin hydrochloride (Met) on the body weights and relative weights of liver and kidney of rats after two months of treatment.

Groups	*Initial weight (g)*	*Final weight (g)*	*Relative liver weight (%)*	*Relative kidney weight (%)*
Control	202.3 ± 6.3	229.4		± 4.2
				3.3 ± 0.5
				0.8 ± 0.2
Met 250 mg/kg	200.1 ± 7.1	209.2		± 5.2
				3.5 ± 0.3
				0.7 ± 0.1
Met 500 mg/kg	209.2 ± 6.8	210.3		± 2.4
				3.2 ± 0.4
				0.9 ± 0.4

Values are the Mean ± SE, n = 10, $p < 0.05$

Table 2: Effect of metformin hydrochloride (Met) on some biochemical parameters of rats after two months of treatment.

Groups	*Creatinine (µmol/L)*	*Urea (µmol/L)*
Control	2.9 ± 0.2	9.5 ± 2.3
Met 250 mg/kg	3.2 ± 0.1	10.4 ± 1.4
Met 500 mg/kg	3.3 ± 0.3	10.5 ± 0.4

Values are the Mean±SE, n = 10, $p < 0.05$

Table 3: Effect of metformin hydrochloride (Met) on live enzyme levels and total bilirubin of rats after two months of treatment.

Groups	*AST (U/l)*	*ALT (U/l)*	*ALP (U/l)*	*YGT (U/l)*	*Total bilirubin (mmol/L)*
Control	30.1±7.2	37.9±1.2	77.5±2.1	36.1±0.5	330.1±7.9
Met 250 mg/kg	29.2 ± 6.6	36.2 ± 0.1	78.4 ± 1.5	37.1 ± 2.1	333.2 ± 8.6
Met 500 mg/kg	27.1 ± 5.4	37.3 ± 0.2	76.5 ± 1.4	38.1 ± 0.3	337.1 ± 5.8

Values are the Mean ± SE, n = 10, $p < 0.05$

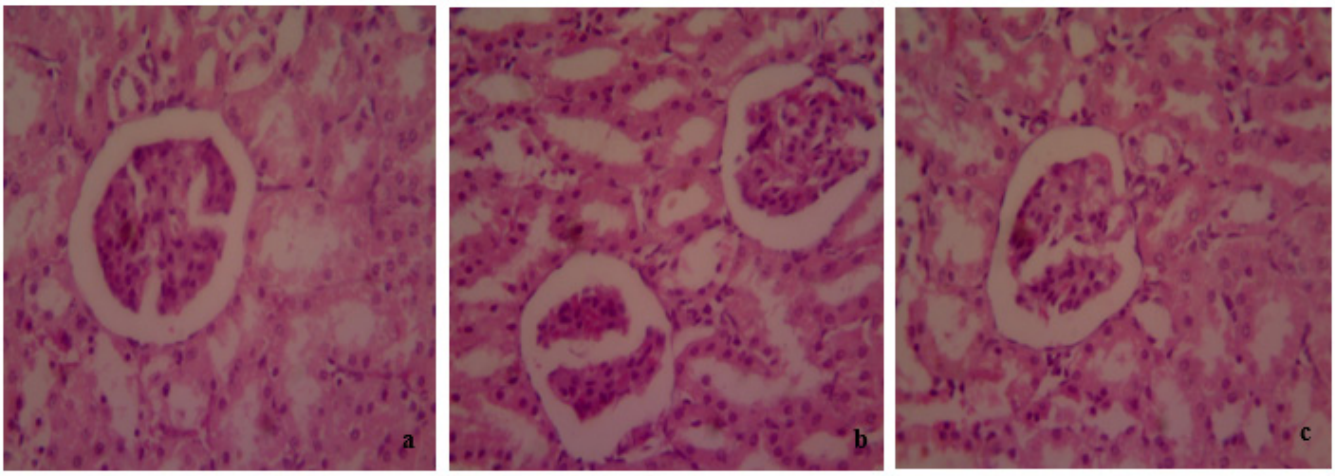

Figure 1: Kidney section of rat (a) in control group. (b) rats group treated with metformin at dose 250 mg/kg. (c) rats group treated with metformin at dose 500 mg/kg show normal histologic architecture with normal proximal convoluted tubules lined by cuboidal epithelium (H&E 400X).

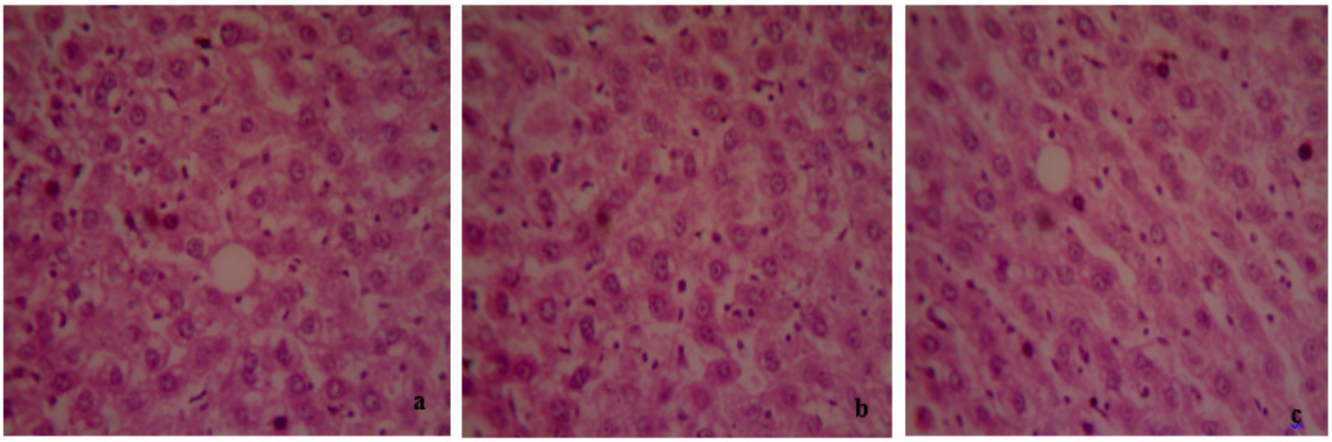

Figure 2: Liver section of male rats in (a)control group. (b) rats group treated with metformin at dose 250 mg/kg. (c) rats group treated with metformin at dose 500 mg/kg show normal central vein and normal hepatocytes architecture (H&E 400X).

DISCUSSION

The correct part of "metformin in the lactic acidosis isn't comprehended. The Cochrane gathering[22] near results investigation of metformin mediation versus the traditional approach (COSMIC) study,[23] and the United Kingdom planned diabetes ponder[24] have questioned the presence of lactic acidosis within sight of metformin and henceforth the term metformin-prompted lactic acidosis has along these lines been changed to MALA. Metformin lessens pyruvate dehydrogenase movement, and mitochondrial transport of decreasing operators, and "in this way improves anaerobic digestion. This move to anaerobic digestion, within sight of lessened insulin, builds a generation of antecedents of the Kreb's cycle.[25]

In the present examination, metformin-treated gatherings did not demonstrate any noteworthy weight change, which might be because of the metformin initiated diminish in calorie admission and weight reduction.[26]

Metformin bolstered rats did not produce any huge change in serum creatinine, blood urea, and kidney histopathological scores, which feature that both metformin did not influence kidney solidness.[27]

The liver assumes a critical part in managing glucose levels in various physiological and neurotic states, for example, diabetes.[28]

Also, numerous medications and mixes are used and detoxified in the liver, so it is liable to potential harm from a huge cluster of remedial and natural synthetic compounds.[29] An immense number of proof demonstrates that diabetes is related with an extensive variety of liver variations from the norm, including anomalous glycogen statement, nonalcoholic greasy liver ailment (NAFLD), fibrosis, cirrhosis, hepatocellular carcinomas, irregular lifted liver chemicals, intense liver malady and viral hepatitis.[30]

ALT and AST proteins are in charge of the generation of ketone bodies from amino acids and, in this manner, deliver high centralization of glucose level. As these blend treatments diminish the ALT and AST level, in this manner they diminish the blood glucose level.[31]

Plasma levels of AST and ALT are as a matter of first importance markers in evaluating liver wounds. Proof has demonstrated that in interminable hepatitis and cirrhosis, serum AST levels are higher than ALT; this may reflect hepatic cell rot with discharging of mitochondrial AST.[32] Metformin

just at the measurement of 100 mg/kg could diminish AST levels in diabetic rats. The advantageous impacts of metformin on liver catalysts action in diabetic rats have been accounted for in a few investigations.[33]

Histopathological evaluation of liver tissues affirmed the film balancing out and cytoprotective capability of metformin at measurements of 500 mg/kg against the dangerous indications instigated by xenobiotic. Serum bilirubin is a standout amongst the most delicate tests utilized in the analysis of hepatic ailments. It offers valuable data on how well the liver is working.[34]

Treatment with metformin at three dosages (250 and 500 mg/kg) had not altogether modified aggregate bilirubin levels when thought about inside the gatherings and demonstrating the viability of the metformin in support of ordinary utilitarian status of the liver.[35]

CONCLUSION

Throughout the examination, there exists a subordinate measurement variety in hepatoprotection, and this article firmly underscores that high-dosage metformin might be a potential restorative competitor against any xenobiotic with no fundamental danger on renal tissues. Moreover, this examination may fill in as a basic instrument to incite an enthusiasm for screening metformin against different metabolic issue. however, utilized metformin for significant lot related with lactic acidosis in the body so can't utilized for long time.

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