



International Journal of Pharmacology

ISSN 1811-7775

Research Article

Protective Effect of Rutaecarpine in Streptozotocin-Induced Diabetes Cataracts Through the Inhibition of Aldose Reductase Activity in Male Albino Rats

Zhaoli Chen, Xiaoyu Zhang, Ning Liu and Ting Zhang

Department of Ophthalmology, Qingdao Municipal Hospital (Group), No. 1, Jiaozhou Road, 266011 Qingdao, China

Abstract

Background and Objective: Diabetes mellitus (DM) is a major metabolic disease which affect the millions of people worldwide. The DM induces the several associate diseases like cataract, renal and hepatic failure. The main enzyme aldose reductase during the polyol pathway serves an important part in the formation of cataracts in diabetic conditions. Rutaecarpine (RT), which is an indolopyridoquinazoline alkaloid and principal compound of evodia shows therapeutic activity against several chronic illnesses. It is proven to be effective in the treatment of diabetes mellitus via altering oxidative stress and inflammation. The goal of current research was to scrutinize the pharmacological action of rutaecarpine in streptozotocin-induced diabetic cataracts in rodents.

Materials and Methods: All the animals were divided into five groups and streptozotocin-induced diabetic rats were supplemented with different doses of rutaecarpine. The formation of cataracts was evaluated using the scoring technique by observer and lens digital picture. Other biochemical parameters were also assessed such as blood, insulin, blood glucose, body weight, sorbitol content, protein carbonyl content, mRNA and expression of aldose reductase in each group of rats. We also measured the kinetics properties of aldose reductase.

Results: Administration of rutaecarpine alters all biochemical parameters compared to diabetic cataract rats. Rutaecarpine significantly stuck the formation of cataracts in the rats in a dose-dependent manner. The kinetic properties of aldose reductase i.e., V_{max} , K_m and K_i had also attenuated in diabetic cataract rats as well as mRNA and protein expression. **Conclusion:** Collectively, we can say that rutaecarpine has the potential to reduce the progression of cataracts in a diabetic case via the reduction of aldose reductase.

Key words: Rutaecarpine, diabetic cataracts, albino rats, aldose reductase

Citation: Chen, Z., X. Zhang, N. Liu and T. Zhang, 2023. Protective effect of rutaecarpine in streptozotocin-induced diabetes cataracts through the inhibition of aldose reductase activity in male albino rats. *Int. J. Pharmacol.*, 19: 147-156.

Corresponding Author: Ting Zhang, Department of Ophthalmology, Qingdao Municipal Hospital (Group), No. 1, Jiaozhou Road, 266011 Qingdao, China

Copyright: © 2023 Zhaoli Chen *et al.* This is an open access article distributed under the terms of the creative commons attribution License, which permits unrestricted use, distribution and reproduction in any medium, provided the original author and source are credited.

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Diabetes mellitus is considered a metabolic disease that impacts millions of populations all over the world. Diabetes arises because of inadequate levels of insulin and resistance to insulin¹. The major reason behind blindness in diabetic patients is cataracts, especially in developing countries.

The most common complication of DM is cataracts, their progression and appearance based on the diabetes duration and hyperglycemia severity. The best treatment for cataracts is surgery but after surgery, it causes vision deterioration. So, some non-surgical approaches should be available to stop the progression and formation of the cataract^{2,3}.

Several mechanisms are responsible for the formation of cataracts in the diabetic case including polyol pathway activation, higher oxidative stress and an elevated level of glycation (non-enzymatic) of the lens. One study reported the different types of drugs for diabetic cataracts such as ranirestat, fidarestat, zenarestat, ranirestat, epalrestat, tolrestat and sorbinil, as aldose reductase inhibitors^{4,5}. Although various aldose reductase inhibitors have been known, they all have unacceptable side effects and show little pharmacokinetic activity. Hence, improvement in conventional aldose reductase inhibitors is in need with few side effects or not for the preventive and therapeutic effects in diabetic cataracts⁶.

So, an enzyme named aldose reductase during the polyol pathway serves an important role during hyperglycemia. A researcher has shown that in the ocular lens, aldose reductase converted the glucose into sorbitol and accumulation of sorbitol produces eye swelling because of advanced osmotic pressure in an eye. It may be a pharmacological target in the management of cataracts induced by diabetes⁷.

For the management of diabetic cataracts, natural product identification or isolated compound from plant sources are in great demand. As in Chinese medicine, rutaecarpine (RT) is also termed Wu Zhu Yu and obtained from plant-derived from *Evodia rutaecarpa* (Juss) Benth (dried fruit)⁸. Rutaecarpine has been shown to have a variety of pharmacological actions, for example, suppression of atherosclerosis, analgesic, vasodilatory effects, antidiabetic, anti-inflammatory and inotropic activity on the heart. It also inhibits adipogenesis/lipogenesis, which lowers lipid buildup in adipocytes. Diabetic cataract progression is also caused by oxidative stress⁹. So, rutaecarpine shows valuable activity in this case also. It was also reported that rutaecarpine shows pharmacological activity against the complication

of diabetes¹⁰. Although anti-inflammatory and anti-oxidative potentials of rutaecarpine are reported in literature but the underlying mechanism for diabetic cataracts is not examined yet.

The aim of the present study was to scrutinize the effect of rutaecarpine against streptozotocin-induced diabetic cataract in animals via inhibition of aldose reductase enzyme.

MATERIALS AND METHODS

The study was carried out March, 2022.

Rats and diabetes induction: All of the studies were carried out in compliance with the Laboratory Animal Welfare and Ethics Committee of Qingdao Municipal Hospital (Group). All the norms and regulations were followed as mentioned in the guidelines (Approval No. QDSL-2022-0104). All the rodents aged 5 weeks old, were procured from the departmental animal house.

A quantity of 65 mg kg⁻¹ streptozotocin was mixed in a buffer solution of citrate of pH = 4.5 and single i.v., the injection was used for the diabetes induction after 7 days of acclimatization¹¹. A vehicle injection was given to age-matched control rats in the same amount (citrate buffer). After two days of STZ injection, a higher level of non-fasting plasma glucose levels (>350 mg dL⁻¹) validated the induction of diabetes. Streptozotocin-treated rats were also given tap water D-glucose (5%), which accelerated cataracts production in less time.

Dosage regimen: Rutaecarpine was mixed in 0.5% of carboxymethylcellulose (CMC) for the treatment of diabetic cataract. After the induction of diabetes, administration of RT was started immediately. All the male albino rats were treated with RT except the negative group and water *ad libitum*. Each rat's water intake was measured on daily basis, on this data, we also calculated the daily dose of RT (mean). Fifty four male albino rats were divided into 5 classes where n = 9.

Group I : Control groups

Group II : Diabetic cataracts (DC) group induced by STZ

Group III : DC rats were treated with RT at a dose level of 10 mg kg⁻¹ (DC+RT10)

Group IV : DC rats were treated with RT at a dose level of 20 mg kg⁻¹ (DC+RT20)

Group V : DC rats were treated with RT at a dose level of 30 mg kg⁻¹ (DC+RT30)

Table 1: Classification of opacification in the lens based on observer score

Score	Characteristics
0	Clear
1	Peripheral vesicles and opacities
2	Central opacities
3	Diffused opacities
4	Mature cataract
5	Hypermaturation cataract

Assessment of body weight, plasma insulin and blood glucose:

Body weight from each group was measured on weekly basis. Method of glucose oxidase was utilized to estimate the levels of blood glucose and in this glucose convert into gluconic acid (Invitrogen™, Glucose Detection Kit, Thermo Fisher Scientific Inc)¹² and concentration of plasma insulin (ab200011, Abcam)¹³ were measured using a marketed obtainable enzyme kit as per specification by the manufacturer.

Assessment of cataracts: We used the digicam (canon IXUS 185 digital camera system) that has a non-reflecting illuminator that gives images with high resolution of the eyes lens. Based on the score obtained by an observer and high resolution of the image clicked by digicam of the lenses used to determine the severity of diabetes cataracts¹⁴. As per the classification of opacification in the lens, lenses status was scored for the observer-based scoring method (Table 1).

The average right and left lens scores were used to calculate the cataract of each animal. The center portion of the eyes had an opaque area and this signifies the harm to the vision and covers 30% of the diameter of the eyeball, was analyzed for quantitative image analysis. The opacity was determined by:

$$\text{Opacity (\%)} = \frac{\text{Number of pixels in the opaque area}}{\text{Total number of pixels in the lens's chosen central region}}$$

Each animal's right and left lens opacities were combined and averaged.

Rutaecarpine content in the lens tissue homogenate: The lenses were first homogenized in 1% HClO₄ (perchloric acid) before being neutralized with 2 M K₂CrO₄ (potassium carbonate). At 25°C, centrifugation of homogenates at 5230 g for 15 min. The HPLC was used to determine the amount of rutaecarpine in the supernatants¹⁵. Measurement of content of rutaecarpine in lense tissue notes as nmol g⁻¹ wet tissue.

Sorbitol content: All group rats' lense was taken to evaluate the content of sorbitol. As previously stated, the sorbitol content of the lenses was determined. Tissues were

homogenized in 16% perchloric acid and 2 M potassium carbonate was used to neutralize it, in a nutshell and centrifuge at 25°C at 5500 g for 10 min¹⁶. The content of sorbitol was determined with the help of the marketed available kit (EnzyChrom™ Sorbitol Assay Kit, Bioassay System, Hayward, CA, USA) as per the specification of the manufacturer. The sorbitol concentration in the lens was measured in micromoles per gram of wet tissue (mol g⁻¹ wet tissue).

Carbonyl content of protein in lens homogenate: The 1 mM EDTA (ethylenediaminetetraacetic acid) of pH = 6.7 was used along with Cold phosphate buffer (2 mL) with 1 m to homogenize lenses, t 4°C, centrifugation was done on homogenates at 8700 g 15 min¹⁷. Determination of protein carbonyl content was done by the marketed available kit and following the manufacturer's instructions (Protein Carbonyl Content Assay Kit, Abcam plc, Cambridge, UK). The amount of protein carbonyl in the lens was measured in terms of nmol/mg protein.

Glutathione content: The content of glutathione was measured with the help of the previously reported procedure of Ellman's reaction in the lens tissue of rats. In short, the glutathione sulfhydryl group reacted with the chemical DTNB. Estimation of absorbance was taken at 412 nm spectrophotometrically which gives a quantitative measurement of -SH groups¹⁸.

Aldose reductase kinetics studies: Measurement of Aldose reductase in rats' lenses was carried out by the reported method. The kinetic studies (K_m, V_{max} and K_i) of the aldose reductase inhibition were noted as per the Cheng-Prusoff equation¹⁹.

mRNA expression level of aldose reductase: Lens tissue homogenate was used to extract the RNA by the use of oligo primers. Aldose reductase study using the primers.

Qualitative RT-PCR of lens tissue: Whole RNA was extracted out from the lens tissue homogenate using Trizol reagent as per mentioned instruction. Conversion of cDNA from RNA with a kit. The AR study was done with the help of RT-PCR on a PCR instrument. For the, was used. Aldose reductase level mRNA expression was assessed with the help of a 2^{-ΔΔCT} procedure²⁰. An appropriate primer for gene expression information of AR is GAPDH as an internal reference, (forward) 5-GACACTTGGA GCTGCTTTT-3 and (reverse) 5-CTGGTTCACC GAAGATGGT-3). Aldose reductase-(forward) 5-ACCTCCAGCTTCTGTCACTG-3 and (reverse) 5-TCATCCATGGCGTGTCACTG-3.

Immunohistochemistry study of lens tissue: Lens was removed from the rats and fixed with formalin. Paraffin-embedded lenses were cut into 4 μm sections. For rehydration, graded alcohol and xylene were utilized. Endogenous peroxide activity was inhibited using 0.3% hydrogen peroxide. About 2% BSA was used to incubate for half an hour which inhibited nonspecific binding sites. Section of the lens was incubated with primary anti-aldose reductase antibody at RT for 12 hrs, followed by 60 min with fluorescein isothiocyanate-conjugated secondary antibody. The staining intensity was estimated after viewing the slices under a fluorescent microscope²¹.

Statistical analysis: The results of the experiments are reported as Mean $\pm$ SEM. The ANOVA was followed by Dunnett's test to examine differences between the control and treated groups. The statistical significance level was considered at $p < 0.05$.

RESULTS

Body weights, plasma glucose levels and serum insulin:

The therapeutic impact of rutaecarpine was measured in streptozotocin-induced diabetic cataracts in male albino rats through the aldose reductase inhibition activity. After the treatment with RT, there was more weight gain in rat's bodies than in the diabetic rats. The gain in body weight of rats at a

dose level of 10, 20 and 30 mg of RT was 223 ± 4.4 , 257 ± 5.8 and 265 ± 6.8 , respectively whereas, diabetic rats show body weight of 206 ± 3.4 . There was a significant increase was found in the blood glucose after supplementation of RT in a dose-dependent manner in diabetic rats. A similar trend was observed for the level of plasma insulin after supplementation of RT in STZ rats than in group II (Table 2).

Score and opacity of lens cataract: Throughout the experimental period, there were no symptoms of cataracts were seen in the lenses of control rats, whereas, 79% of cataracts were found in the lenses after 2 weeks of induction diabetes in negative groups. On the other hand, RT (10), RT (20) and RT (30) groups rats showed cataracts at a percent of 72, 64 and 59, respectively. It was also observed that with time the progression of cataracts in the diabetic group produced hyper mature cataracts in the lenses. Occurrence of hypermature cataracts in RT (10), RT (20) and RT (30) in 20% of lenses, 12% of lenses and 8% of lenses, respectively. Figure 1a summarized the qualitative analysis of the results. Opacity measurement in the central region of the eye and scoring method was summarized in Fig. 1b. Outcomes suggested the cataract progression was the same in diabetic rats and DC+RT rats until 3 weeks. After diabetic induction, there was a delay in cataract progression at 6 weeks in DC+RT rats in a dose-dependent pattern.

Table 2: Impact of rutaecarpine on the rat's body weight, blood glucose and insulin in the experimental animals

Experimental group	Body weight (g)		Blood glucose (mmol L ⁻¹)		Blood insulin (microunit mL ⁻¹)	
	Initial	Final	Initial	Final	Initial	Final
Group I	181 $\pm$ 3.2	304 $\pm$ 6.8	5.1 $\pm$ 0.4	4.9 $\pm$ 0.5	15.1 $\pm$ 0.4	16.2 $\pm$ 0.5
Group II	170 $\pm$ 3.8	206 $\pm$ 3.4*	22.7 $\pm$ 1.2*	25.0 $\pm$ 1.2*	4.9 $\pm$ 0.3*	4.6 $\pm$ 0.2*
Group III	182 $\pm$ 4.9	223 $\pm$ 4.4 ^a	22.7 $\pm$ 1.1	21.1 $\pm$ 0.8 ^a	4.3 $\pm$ 0.3	6.3 $\pm$ 0.3 ^a
Group IV	179 $\pm$ 3.5	257 $\pm$ 5.8 ^a	23.7 $\pm$ 1.1	18.3 $\pm$ 1.3 ^a	5.3 $\pm$ 0.3	8.4 $\pm$ 0.2 ^a
Group V	181 $\pm$ 4.1	265 $\pm$ 6.8 ^a	22.3 $\pm$ 1.3	14.8 $\pm$ 1.2 ^a	4.5 $\pm$ 0.2	10.7 $\pm$ 0.2 ^a

Outcomes were represented as the Mean $\pm$ SD where n = 9, data analysis was performed using ANOVA followed by Dunnett-Test, where * $p < 0.05$ (group II vs group I) and ^a $p < 0.05$ (groups III, IV and V vs group II)

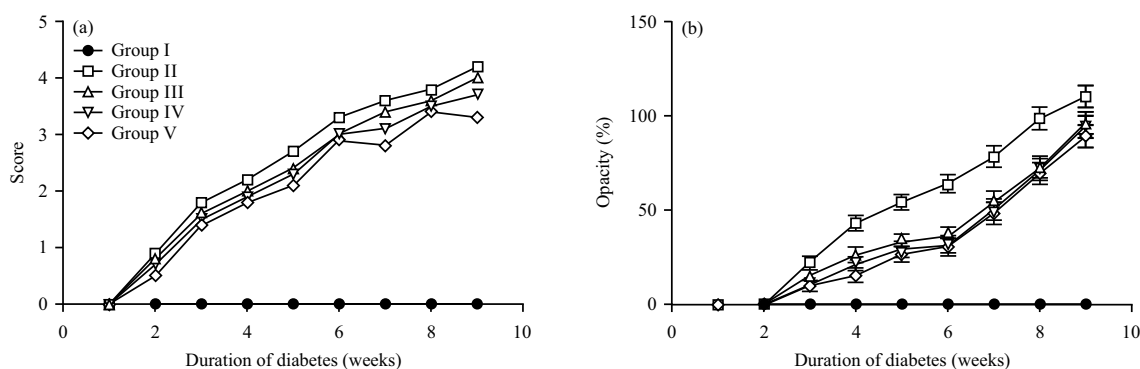


Fig. 1: Impact of rutaecarpine on the rat's lens in the experimental group, (a) Score and (b) Opacity

Outcomes were represented as the Mean $\pm$ SD where n = 9, data analysis was performed using ANOVA followed by Dunnett-Test, where * $p < 0.05$ (group II vs group I) and ^a $p < 0.05$ (groups III, IV and V vs group II)

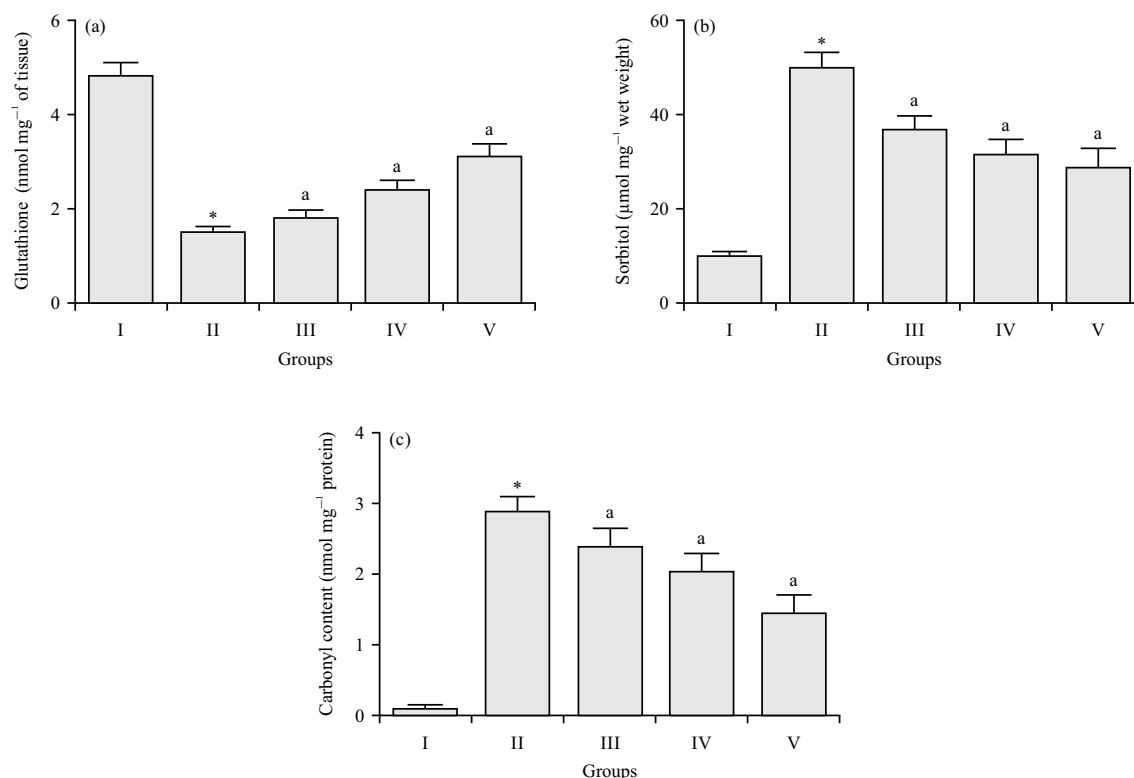


Fig. 2(a-c): Impact of rutaecarpine on the rat's lens homogenate in the experimental group, (a) Glutathione level, (b) Sorbitol and (c) Protein carbonyl content

Outcomes were represented as the Mean $\pm$ SD where n = 9, data analysis was performed using ANOVA followed by Dunnett-Test, where *p<0.05 (group II vs group I) and ^ap<0.05 (groups III, IV and V vs group II)

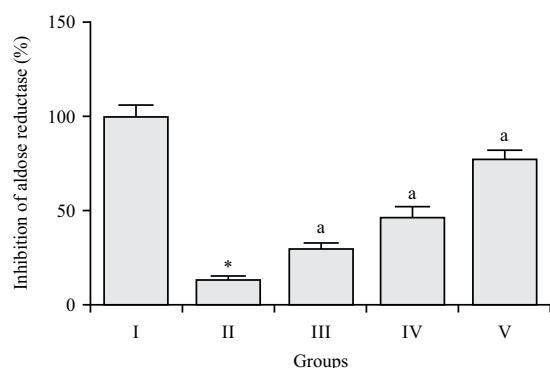


Fig. 3: Impact of rutaecarpine on the rats lens (inhibition of aldose reductase) in the experimental group

Outcomes were represented as the Mean $\pm$ SD where n = 9. Data analysis was performed using ANOVA followed by Dunnett-Test, where *p<0.05 (group II vs group I) and ^ap<0.05 (groups III, IV and V vs group II)

Glutathione level in lens: Treatment with RT significantly elevated the level of glutathione in the diabetic animal's lens in comparison with control rats. It was substantially elevated after the treatment with RT in a dose-dependent manner (Fig. 2a).

Content of sorbitol in lens tissue: After diabetes induction, there was a significant elevation in the level of sorbitol in 1st week in the diabetic group's lenses. This elevated level was maintained till 9 weeks after diabetes induction. Significant differences were detected in the sorbitol content among the group II and group V rats (Fig. 2b).

Content of protein carbonyl in lens tissue: A similar trend was observed for protein carbonyl, diabetic rats show higher content of PC in a time-dependent pattern whereas significant reduction was observed in group V rats as summarized in Fig. 2c.

Aldose reductase activity and kinetic property: Percentage inhibition indicates the term activity of AR (aldose reductase). The activity of enzyme aldose reductase was considerably reduced after supplementation of RT than in diabetic rats. The enzyme inhibition percentage was found to be 99.3, 12.2, 29.3, 46.4 and 76.8% for normal control, diabetic rats and tested drugs at different dose levels (Fig. 3).

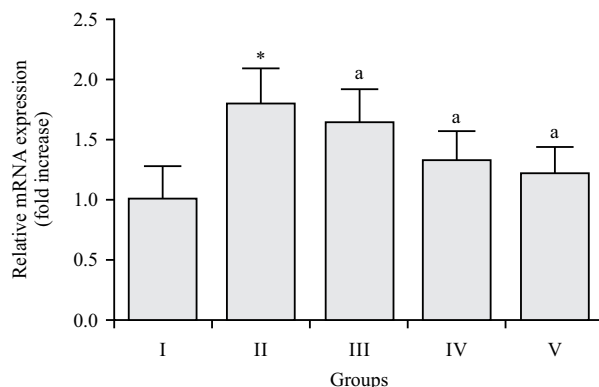


Fig. 4: Impact of rutaecarpine on the rat's lens (relative mRNA expression) in the experimental group

Outcomes were represented as the Mean $\pm$ SD where n = 9. Data analysis was performed using ANOVA followed by Dunnett-Test, where *p<0.05 (group II vs group I) and ^ap<0.05 (groups III, IV and V vs group II)

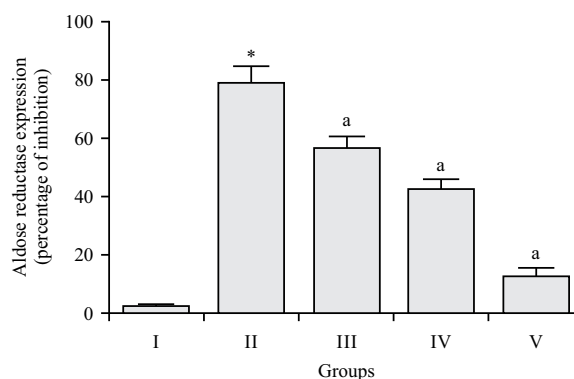


Fig. 5: Impact of rutaecarpine on the rat's lens (aldose reductase expression) in the experimental group

Outcomes were represented as the Mean $\pm$ SD where n = 9. Data analysis was performed using ANOVA followed by Dunnett-Test, where *p<0.05 (group II vs group I) and ^ap<0.05 (groups III, IV and V vs group II)

Table 3: Effect of rutaecarpine on kinetic parameter of aldose reductase

Group	V_{max}	$K_m \times 10^{-3} \text{ mM}$	K_i
Group I	0.14534 ± 0.0001	0.43785 ± 0.0027	0.00000 ± 0.0000
Group II	$0.43765 \pm 0.0003^*$	$3.09859 \pm 0.0325^*$	$2.04562 \pm 0.0301^*$
Group III	0.24321 ± 0.0008^a	2.67541 ± 0.0145^a	1.67392 ± 0.0137^a
Group IV	0.21783 ± 0.0007^a	1.93281 ± 0.0231^a	1.15482 ± 0.0251^a
Group V	0.15739 ± 0.0052^a	0.93273 ± 0.0021^a	1.15482 ± 0.0251^a

Outcomes were represented as the Mean $\pm$ SD where n = 9, data analysis was performed using ANOVA followed by Dunnett-Test, where *p<0.05 (group II vs group I) and ^ap<0.05 (groups III, IV and V vs group II)

Following treatment with RT, the kinetic properties of enzyme aldose reductase for example maximum velocity of the reaction (V_{max}), Michaelis constant (K_m) and inhibitor constant (K_i) were measured. The kinetic parameters of AR were measured in all group rats and lower values were observed in lens tissue homogenate of diabetic animals than in the control. Following RT administration in a dose-dependent pattern, the V_{max} of AR was 0.24321 ± 0.0008 , 0.21783 ± 0.0007 and 0.15739 ± 0.0052 , respectively, whereas, K_m of AR was observed as 2.67541×10^{-3} , 1.93281×10^{-3} mM and 0.93273×10^{-3} mM for group III, group IV and group V, respectively and shown in

Table 3, the inhibitor constant, (K_i) of AR was 1.67392 ± 0.0137 , 1.15482 ± 0.0251 and 1.15482 ± 0.0251 , respectively which is significant.

Gene expression: In the group II rats that have a diabetic cataract, aldose reductase mRNA expression was significantly enhanced and increased by 1.4-fold in comparison to control groups. Although a significant reduction was observed in the levels of mRNA expression of aldose reductase after treatment with RT in a dose-dependent manner (Fig. 4). It was revealed to be reduced by 17.4, 22.8 and 49% following RT administration, respectively.

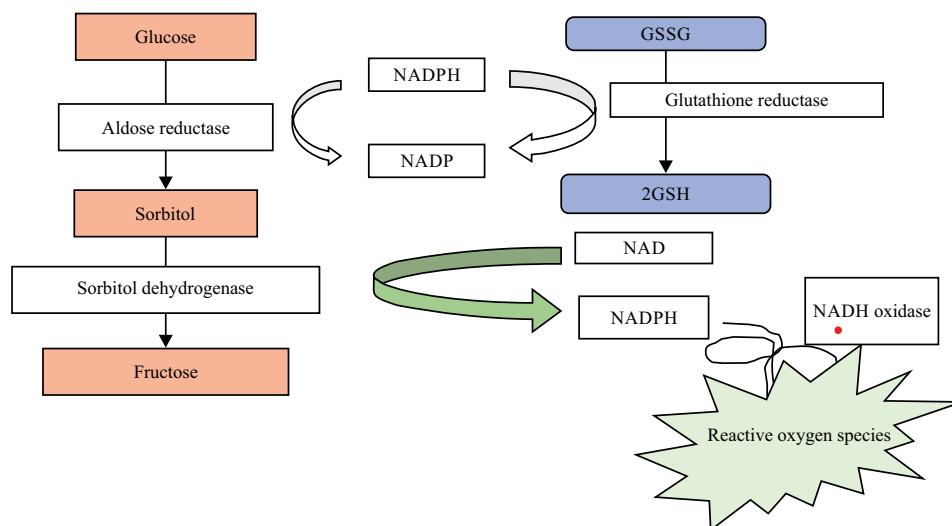


Fig. 6: Mechanism of aldose reductase during oxidative stress induced by diabetes

Immunohistochemical study: Reduction in the protein expression of aldose reductase was confirmed by an immunohistochemical study by 18.34% (group V), 42.46% (group IV) and 56.39% (group III), respectively which was significant and summarized in Fig. 5. Figure 6 showed the proposed mechanism.

DISCUSSION

Effective pharmacological substances are those which delay the cataract progression and might be assessed carefully *in vivo*. The current investigation explains the effect of RT which interrupts the progression in diabetic rats that have cataracts. Diabetic rats showed higher levels of protein carbonyl and sorbitol in the lenses in comparison with control rats. RT reduced the higher content of protein carbonyl and sorbitol in the lenses of diabetic rats. These outcomes beautifully protect the oxidative destruction to lens tissue proteins which are partially dedicated to an anti-cataract property of RC^{22,23}.

The RT supplementation improves insulin responsiveness and oral rutaecarpine supplementation raises gut hormone levels, enhancing insulin's effect on the level of blood glucose^{24,25}. Literature cited about the Improved oxidative stress causes elevated LPO and reduced levels of glutathione, enzymatic antioxidant parameters such as GPx, catalase and SOD in diabetes patients. This is consistent with Zhang *et al.*²⁶. prior findings showed a similar change in diabetic cataracts. Aldose reductase has been linked to retinopathy, cataract, neuropathy, microangiopathy and corneal epitheliopathy, among other diabetes problems. Aldose reductase inhibition can avoid all of these diabetes consequences^{27,28}.

Hyperglycemia can also cause peripheral neuropathy, sorbitol buildup and a variety of eye diseases. During diabetic circumstances, sorbitol-lowering medications inhibit the formation of cataracts²⁹. Furthermore, RT supplementation slowed the onset of diabetic cataracts by lowering glucose levels. In diabetic rats, RT therapy effectively decreased aldose reductase activity.

Diabetes patients have high amounts of glucose present in the eye's aqueous humor and it can reach the lens via insulin-independent, facilitated and passive transport routes³⁰. Aldose reductase is a key enzyme of the polyol pathway which transforms sorbitol from glucose and amasses since the lens is not allowed to diffuse it out passively³¹. As sorbitol builds up in the lens, it causes osmotic stress, which causes morphological changes such as swelling and hydration. Furthermore, activation of pathways related to polyol may affect the decrease in the ratio of NADPH/NADP and impair the ratio of Gpx and GR, lowering the reduced decrease in the ratio of GSH and oxidized glutathione and causing oxidative stress³².

Increased amounts of reactive oxygen species cause cellular proteins and lipids to be damaged. Reactive oxygen species in the lens can cause soluble proteins to precipitate and is mainly responsible for the refraction of the light via oxidation of polyunsaturated fatty acid and sulfhydryl (-SH) groups³³. Previous report suggest that polyol pathway play a crucial role in the formation of cataract during initiation of diabetes condition³⁴. It is well known that higher quantity of protein carbonyls (marker of reactive oxygen driven protein oxidation) and sorbitol (a marker of polyol pathway) observed in the diabetic lenses³⁵. Therefore, the boosted level of sorbitol observed during the 1st week following diabetes induction,

but the rise in protein carbonyl level was modest. Despite the quick rise in sorbitol levels in the first week, cataracts developed throughout the diabetic period and full cataracts took several weeks to occur.

These findings support the widely held belief that cataract formation is complicated and involves numerous phases, including oxidative processes. Rutaecarpine inhibited the formation of cataracts while also reducing the increased amounts of sorbitol in the lens and preventing the accumulation of protein carbonyls. As a result, rutaecarpine shows little activity against the active polyol pathway under our test settings. Hyperglycemia can produce reactive oxygen species through a variety of processes, including raised an increase in advanced glycation end-products as well as activation of the polyol pathway, overproduction of mitochondrial superoxide and altered activity of protein kinase C activity^{36,37}. Rutaecarpine can quench ROS while also increasing the expression of the antioxidant enzymes³⁸. The anti-cataract activity of rutaecarpine appears to be effective because of the reduction in oxidative destruction in proteins of the lens and the exact mode of action is unknown. To understand the precise mechanisms, more research is needed.

In streptozotocin-induced diabetic rats, higher intake of water, reduced body weight and higher value of plasma glucose are the frequent signs. In our work, animals who have diabetes develop bradycardia but not hypertension. The RT did not affect these symptoms. The RT has revealed the reduction in levels of blood glucose levels in diabetic rodents induced by STZ in earlier research. Effect of RT seen on level blood glucose levels and that could be attributable to the severity of hyperglycemia^{39,40}. To reduce variability and speed of the development of diabetic cataracts, researchers used a combination of streptozotocin therapy and feeding of D-glucose to generate severe hyperglycemia. As prior investigations show that rats that received streptozotocin didn't have an extra quantity of D-glucose. In clinical investigations, a varied dose of rutaecarpine (5-2000 mg/day) was investigated for metabolic diseases to validate its therapeutic range⁴¹. The rutaecarpine dosing range for a human weighing 60 kg is 10-100 $\mu\text{g kg}^{-1}$ i.v.¹⁰. As a result, the actual quantity of rutaecarpine used in this investigation at a dose level of 30 mg/kg/day was found to be effective for pharmacological usage.

CONCLUSION

In short, the results of present study showed that rutaecarpine has potential against diabetes cataracts induced by STZ in rats via reduced expression of aldose reductase.

Further research have to be carried out to scrutinize the underlying mechanism and exact pharmacological action of the rutaecarpine.

SIGNIFICANCE STATEMENT

This study discovers the anticataract effect of rutaecarpine against streptozotocin induced diabetes that can be beneficial for cataract patients. This study will help the researcher to uncover the critical area of cataract occur during the diabetes that many researchers were not able to explore. Thus, a new therapy for cataract may be arrived at.

ACKNOWLEDGMENT

The authors would like to thanks to the hospital for the support and motivation in writing this manuscript.

REFERENCES

1. Farzaei, F., M.R. Morovati, F. Farjadmand and M.H. Farzaei, 2017. A mechanistic review on medicinal plants used for diabetes mellitus in traditional Persian medicine. *J. Evidence-Based Complementary Altern. Med.*, 22: 944-955.
2. Kiziltoprak, H., K. Tekin, M. Inanc and Y.S. Goker, 2019. Cataract in diabetes mellitus. *World J. Diabetes*, 10: 140-153.
3. Barman, S. and K. Srinivasan, 2019. Zinc supplementation ameliorates diabetic cataract through modulation of crystallin proteins and polyol pathway in experimental rats. *Biol. Trace Elem. Res.*, 187: 212-223.
4. Pandurangan, M. and D.H. Kim, 2016. Therapeutic potential of cyanobacteria against streptozotocin-induced diabetic rats. *3 Biotech*, Vol. 6. 10.1007/s13205-016-0411-0.
5. Dunlop, M., 2000. Aldose reductase and the role of the polyol pathway in diabetic nephropathy. *Kidney Int.*, 58: S3-S12.
6. Muthuraman, P. and K. Srikumar, 2009. A comparative study on the effect of homobrassinolide and gibberellic acid on lipid peroxidation and antioxidant status in normal and diabetic rats. *J. Enzyme Inhibit. Med. Chem.*, 24: 1122-1127.
7. Xie, H., Q. Tong, Z. Xiang, C. Zhou, L.S. Wan and J. Chen, 2022. Demethylbellidifolin, a potential aldose reductase inhibitor ameliorates diabetic nephropathy by regulating the polyol pathway. *Phytomed. Plus*, Vol. 2. 10.1016/j.phyplu.2021.100152.
8. Huang, X.F., Y.H. Dong, J.H. Wang, H.M. Ke, G.Q. Song and D.F. Xu, 2020. Novel PDE5 inhibitors derived from rutaecarpine for the treatment of Alzheimer's disease. *Bioorg. Med. Chem. Lett.*, Vol. 30. 10.1016/j.bmcl.2020.127097.
9. Tian, K.M., J.J. Li and S.W. Xu, 2019. Rutaecarpine: A promising cardiovascular protective alkaloid from *Evodia rutaecarpa* (Wu Zhu Yu). *Pharm. Res.*, 141: 541-550.

10. Jia, S. and C. Hu, 2010. Pharmacological effects of rutaecarpine as a cardiovascular protective agent. *Molecules*, 15: 1873-1881.
11. Vadivelan, R., R.G. Krishnan and R. Kannan, 2019. Antidiabetic potential of *Asparagus racemosus* Willd leaf extracts through inhibition of α -amylase and α -glucosidase. *J. Tradit. Complementary Med.*, 9: 1-4.
12. Wang, Z., Y. Yang, X. Xiang, Y. Zhu, J. Men and M. He, 2010. Estimation of the normal range of blood glucose in rats. *J. Hyg. Res.*, 39: 133-137.
13. Gandhi, G.R. and P. Sasikumar, 2012. Antidiabetic effect of *Merremia emarginata* Burm. F. in streptozotocin induced diabetic rats. *Asian Pac. J. Trop. Biomed.*, 2: 281-286.
14. Kametaka, S., T. Kasahara, M. Ueo, M. Takenaka and M. Saito *et al.*, 2008. A novel high resolution *in vivo* digital imaging system for the evaluation of experimental cataract in diabetic rats. *J. Pharmacol. Sci.*, 106: 144-151.
15. Rex, J., T. Knowles, X. Zhao, J. Lemp, C. Maissa and S.S. Perry, 2018. Elemental composition at silicone hydrogel contact lens surfaces. *Eye Contact Lens: Sci. Clin. Pract.*, 44: S221-S226.
16. Ashizawa, N., M. Yoshida, Y. Sugiyama, N. Akaike and S. Ohbayashi *et al.*, 1997. Effects of a novel potent aldose reductase inhibitor, GP-1447, on aldose reductase activity *in vitro* and on diabetic neuropathy and cataract formation in rats. *Jpn. J. Pharmacol.*, 73: 133-144.
17. Mori, A., O. Saigo, K. Sakamoto, T. Nakahara and K. Ishii, 2010. Hyperglycemia impairs acetylcholine-induced vasodilation of retinal arterioles through polyol pathway-independent mechanisms in rats. *J. Pharmacol. Sci.*, 112: 336-342.
18. Konsue, N., S. Yimthiang and W. Kwanhian, 2018. Effect of fermentation conditions of noni (*Morinda citrifolia* L.) juice on glutathione content and lipid oxidation in vero cells. *Int. Food Res. J.*, 25: 1534-1540.
19. Daniellou, R., H. Zheng and D.R.J. Palmer, 2006. Kinetics of the reaction catalyzed by inositol dehydrogenase from *Bacillus subtilis* and inhibition by fluorinated substrate analogs. *Can. J. Chem.*, 84: 522-527.
20. Soliman, M.M., M. Abdo Nassan and T.A. Ismail, 2016. *Origanum majoranum* extract modulates gene expression, hepatic and renal changes in a rat model of type 2 diabetes. *Iran. J. Pharm. Res.*, 15: 45-54.
21. Evilsizor, M.N., H.F. Ray-Jones, J. Lifshitz and J. Ziebell, 2015. Primer for immunohistochemistry on cryosectioned rat brain tissue: Example staining for microglia and neurons. *J. Vis. Exp.*, Vol. 99. 10.3791/52293.
22. Tanimoto, T., H. Fukuda, H. Sato and J. Kawamura, 1983. Sorbitol pathway in lenses of normal and diabetic rabbits. *Chem. Pharm. Bull.*, 31: 204-208.
23. Higashi, K., A. Mori, K. Sakamoto, K. Ishii and T. Nakahara, 2019. Probucol slows the progression of cataracts in streptozotocin-induced hyperglycemic rats. *Pharmacology*, 103: 212-219.
24. González-Ortiz, M., R. Medina-Santillán, E. Martínez-Abundis and C.R. von Drateln, 2001. Effect of glycine on insulin secretion and action in healthy first-degree relatives of type 2 diabetes mellitus patients. *Hormone Metab. Res.*, 33: 358-360.
25. Gannon, M.C., J.A. Nuttall and F.Q. Nuttall, 2002. The metabolic response to ingested glycine. *Am. J. Clin. Nutr.*, 76: 1302-1307.
26. Zhang, S., F.Y. Chai, H. Yan, Y. Guo and J.J. Harding, 2008. Effects of N-acetylcysteine and glutathione ethyl ester drops on streptozotocin-induced diabetic cataract in rats. *Mol. Vision*, 14: 862-870.
27. Scotti, L., M.B. Fernandes, E. Muramatsu, K.F. Pasqualoto and V.D.P. Emerenciano *et al.*, 2011. Self-organizing maps and VolSurf approach to predict aldose reductase inhibition by flavonoid compounds. *Braz. J. Pharmacogn.*, 21: 170-180.
28. Reddy, G.B., A. Satyanarayana, N. Balakrishna, R. Ayyagari, M. Padma, K. Viswanath and J.M. Petrash, 2008. Erythrocyte aldose reductase activity and sorbitol levels in diabetic retinopathy. *Mol. Vision*, 14: 593-601.
29. Mirsky, N., R. Cohen, A. Eliaz and A. Dovrat, 2016. Featured article: Inhibition of diabetic cataract by glucose tolerance factor extracted from yeast. *Exp. Biol. Med.*, 241: 817-829.
30. Alvarado-Vásquez, N., P. Zamudio, E. Cerón, B. Vanda, E. Zenteno and G. Carvajal-Sandoval, 2003. Effect of glycine in streptozotocin-induced diabetic rats. *Comp. Biochem. Physiol. C: Toxicol. Pharmacol.*, 134: 521-527.
31. Li, W., Y. Zhang and N. Shao, 2019. Protective effect of glycine in streptozotocin-induced diabetic cataract through aldose reductase inhibitory activity. *Biomed. Pharmacother.*, Vol. 114. 10.1016/j.biopha.2019.108794.
32. Lu, Q., M. Hao, W. Wu, N. Zhang and A.T. Isaac *et al.*, 2018. Antidiabetic cataract effects of GbE, rutin and quercetin are mediated by the inhibition of oxidative stress and polyol pathway. *Acta Biochim. Pol.*, 65: 35-41.
33. Gong, X., Q. Zhang and S. Tan, 2013. Inhibitory effect of r-Hirudin variant III on streptozotocin-induced diabetic cataracts in rats. *Sci. World J.*, Vol. 2013. 10.1155/2013/630651.
34. Shi, Q., H. Yan, M.Y. Li and J.J. Harding, 2009. Effect of a combination of carnosine and aspirin eye drops on streptozotocin-induced diabetic cataract in rats. *Mol. Vision*, 15: 2129-2138.
35. Valavala, V.K., R.K. Vangipurapu, V.R. Banam, U.M.R. Pulukurthi and N.R. Turlapati, 2011. Effect of mustard (*Brassica juncea*) leaf extract on streptozotocin-induced diabetic cataract in Wistar rats. *J. Food Biochem.*, 35: 109-124.

36. Gondi, M., S.A. Basha, P.V. Salimath and U.J.S.P. Rao, 2017. Supplementation of mango (*Mangifera indica* L.) peel in diet ameliorates cataract in streptozotocin-induced diabetic rats. *J. Food Biochem.*, Vol. 41. 10.1111/jfbc.12300.
37. Ma, Z., J. Liu, J. Li, H. Jiang and J. Kong, 2020. Klotho ameliorates the onset and progression of cataract via suppressing oxidative stress and inflammation in the lens in streptozotocin-induced diabetic rats. *Int. Immunopharmacol.*, Vol. 85. 10.1016/j.intimp.2020.106582.
38. Xu, Y.L., Y. Zhao, Y.N. Sui and X. Lei, 2018. Protective effect of *Pterocarpus marsupium* bark extracts against cataract through the inhibition of aldose reductase activity in streptozotocin-induced diabetic male albino rats. *3 Biotech*, Vol. 8. 10.1007/s13205-018-1210-6.
39. Bahmani, F., S.Z. Bathaie, S.J. Aldavood and A. Ghahghaei, 2016. Inhibitory effect of crocin(s) on lens α -crystallin glycation and aggregation, results in the decrease of the risk of diabetic cataract. *Molecules*, Vol. 21. 10.3390/molecules21020143.
40. Grama, C.N., P. Suryanarayana, M.A. Patil, G. Raghu, N. Balakrishna, M.N.V.R. Kumar and G.B. Reddy, 2013. Efficacy of biodegradable curcumin nanoparticles in delaying cataract in diabetic rat model. *PLoS ONE*, Vol. 8. 10.1371/journal.pone.0078217.
41. Surbala, L., C.B. Singh, R.V. Devi and O.J. Singh, 2020. Rutaecarpine exhibits anti-diabetic potential in high fat diet-multiple low dose streptozotocin induced type 2 diabetic mice and *in vitro* by modulating hepatic glucose homeostasis. *J. Pharmacol. Sci.*, 143: 307-314.