GENERAL SUMMARY
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- Aldose reductase catalyzes the conversion of glucose to sorbitol and causes the accumulation of sorbitol in various tissues under the condition of hyperglycemia. The accumulated intracellular sorbitol causes development of diabetic complications such as cataract, neuropathy, and nephropathy. It has been reported that inhibitors of aldose reductase reduce the tissue sorbitol content in diabetic animals and are useful as therapeutic agents for diabetic complications.

- Nigerloxin, a bioactive molecule obtained from Aspergillus niger by solid state fermentation has previously been shown to inhibit soybean lipoxygenase and rat lens aldose reductase in vitro. There are no extensive studies which support the aldose reductase inhibition and free radical scavenging activity of nigerloxin. The present study was intended to establish the aldose reductase inhibitory potential and antioxidant activity in vivo of this fungal metabolite nigerloxin in diabetic animal model with the following objectives. It was also further envisaged to evaluate the protective effect of nigerloxin if any, on the eye lens and kidney of induced diabetic animals.

- The antioxidant potential of nigerloxin in vitro was studied by different assay procedures including, phosphomolybdenum method, DPPH', ABTS'⁺ and FRAP, and compared with one of the well known natural antioxidant, curcumin. The fungal metabolite nigerloxin was found to be an effective antioxidant in in vitro assays including, phosphomolybdenum method, DPPH', ABTS'⁺ and FRAP. The ferric reducing potency of nigerloxin as demonstrated by FRAP assay method was even found to be superior to that of the natural antioxidant curcumin. The antioxidant potency of nigerloxin may be attributed to its electron donating nature.

- Oxidative stress plays a key role in the progression of diabetes and its complications. The beneficial influence of the fungal metabolite nigerloxin, a new aldose reductase inhibitor and a free-radical scavenger was investigated on oxidative stress in streptozotocin induced diabetic rats. Groups of diabetic rats
were administered with nigerloxin for 30 days at a oral dose of 25 mg and 100 mg/kg body weight/day.

- Diabetic rats showed significantly increased lipid peroxide levels in blood and liver, which was accompanied by lowered concentrations of antioxidant molecules and activities of antioxidant enzymes in blood and liver. Administration of nigerloxin for 30 days at a daily dose of 100 mg/kg body weight to diabetic rats significantly decreased plasma and liver lipid peroxides, elevated the nonenzymatic antioxidants − ascorbic acid, reduced glutathione, and total thiols and elevated the activities of antioxidant enzymes in blood and liver.

- Nigerloxin showed a tendency to counter lipid abnormalities in diabetic animals, while fasting glucose and body weight were unaffected by nigerloxin treatment. Thus, this animal study has indicated the beneficial influence of nigerloxin on oxidative stress associated with diabetes which may have an implication in delaying or ameliorating the secondary complications.

- The role of osmotic and oxidative stress has been implicated in the pathogenesis of diabetic cataract. In the present study, the beneficial influence of fungal metabolite nigerloxin was investigated on diabetes induced alteration in the eye lens of streptozotocin administered rats. Groups of diabetic rats were orally administered nigerloxin (100 mg/kg/day) for 30 days.

- Activities of lens polyol pathway enzymes – aldose reductase and sorbitol dehydrogenase, lipid peroxides and advanced glycation end products were increased in the eye lens of diabetic animals. Glutathione and activities of antioxidant enzymes – superoxide dismutase, glutathione-S-transferase and glutathione peroxidase were decreased in the eye lens of diabetic animals.

- Administration of nigerloxin significantly decreased lens lipid peroxides and advanced glycation end products in diabetic rats. Increase in lens aldose reductase and sorbitol dehydrogenase activities was countered by nigerloxin treatment. Lens
glutathione and antioxidant enzyme activities were significantly elevated in nigerloxin treated diabetic rats.

- Examination of rat eyes indicated that nigerloxin delayed cataractogenesis in diabetic rats. The results suggest the beneficial countering of polyol pathway enzymes and potentiation of antioxidant defense system by nigerloxin in diabetic animals, implicating its potential in ameliorating diabetic cataract.

- Since nigerloxin was previously shown to inhibit aldose reductase activity and improved antioxidant defense system in the eye lens of diabetic rats, the beneficial influence of nigerloxin was also investigated in galactose induced juvenile cataract in experimental animals. Cataract was induced in Wistar rats by feeding 30% galactose in diet. Groups of galactose fed rats were orally administered with nigerloxin (25 and 100 mg/kg body weight/day) for 24 days.

- Lens aldose reductase activity was increased significantly in galactose fed animals. Lens lipid peroxides and advanced glycation end products were also significantly increased. Antioxidant molecule – reduced glutathione, total thiols and activities of antioxidant enzymes – superoxide dismutase and glutathione peroxidase were decreased in the lens of galactose fed animals.

- Oral administration of nigerloxin once a day for 24 days at a dose of 100 mg/kg body weight significantly decreased lens lipid peroxides and advanced glycation end products in galactose fed rats. Lens aldose reductase activity was reduced and lens antioxidant molecules and antioxidant enzyme activities were elevated significantly by nigerloxin administration. The results suggest that alteration in polyol pathway and antioxidant defense system were countered by nigerloxin in the lens of galactose fed animals, suggesting the potential of nigerloxin in ameliorating the development of juvenile cataract in experimental animals.

- Elevated polyol pathway enzyme activities and oxidative stress play an important role in the development and progression of diabetic nephropathy. Here, we investigated the beneficial influence of the fungal metabolite nigerloxin, a potent
aldose reductase inhibitor and free radical scavenger in the kidney of streptozotocin-induced diabetic rats. A group of diabetic rats was orally administered with nigerloxin for 30 days (100 mg/kg).

- Diabetic rats showed increased lipid peroxides, advanced glycation end products, elevated activities of polyol pathway enzymes and lowered antioxidant defense system in kidney. Administration of nigerloxin decreased kidney lipid peroxides and advanced glycation end products. Activities of polyol pathway enzymes were reduced while activities of all antioxidant enzymes, glutathione and ascorbic acid were elevated in the kidney of nigerloxin treated diabetic rats.

- These results indicated the beneficial influence of nigerloxin on polyol pathway and oxidative stress associated with diabetes which are implicated in ameliorating the development of diabetic nephropathy.

- We also investigated the antioxidant potential of nigerloxin, in another experimental model of nephropathy. Experimental kidney toxicity was induced in male Wistar rats by administration of gentamicin at a dose of 80 mg/kg, i.p., for a period of 8 days. Groups of rats were orally administered with nigerloxin for 8 days at a dose of 25 mg and 100 mg/kg body weight/day along with gentamicin.

- Gentamicin induced increase in lipid peroxides, decrease in glutathione, total thiols and activities of antioxidant enzymes — catalase, glutathione peroxidase, super oxide dismutase, glutathione reductase and glutathione-S-transferase in the kidney. Blood creatinine and urea concentrations were significantly increased.

- Blood creatinine and urea levels were reduced by nigerloxin treatment. Nigerloxin treated rats also exhibited elevated activities of superoxide dismutase, glutathione peroxidase, glutathione reductase, catalase and glutathione-S-transferase in kidney. Glutathione and total thiols in kidney were elevated in nigerloxin treated rats. Thus, nigerloxin ameliorated oxidative stress induced by gentamicin in the renal tissue.
Thus, these animal studies have verified the aldose reductase inhibitory potential of the fungal metabolite nigerloxin in \textit{in vivo} situation in both eye lens and kidney tissue. The free radical scavenging and antioxidant properties of this novel fungal metabolite \textit{in vivo} were also evidenced in appropriate animal models. Such a beneficial effect of this compound on aldose reductase pathway and oxidative stress associated with diabetic complications can be implicated in ameliorating the development of diabetic nephropathy and cataract.