Volume 64, Issue 3, 2020

Journal of Scientific Research

Institute of Science,

Banaras Hindu University, Varanasi, India.

National Conference on Frontiers in Biotechnology & Bioengineering (NCFBB 2020), JNTU Hyderabad, India

Role of *Aloe vera* Gel Against Neuro Degeneration, Biochemical and Behavioral Alterations in STZ Induced Diabetic Rats

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***Abstract*: Diabetes is a major source of morbidity, mortality, and emotional concern. This study was to assess the relationship between Diabetes-induced reduction in sciatic nerve's changes. Oxidative stress plays a major role in progression and development of Diabetes-Mellitus (DM) and its impediments like neuropathy. Decreased glucose-levels, water-intake and gain of body-weight by *Aloe vera* gel indicate its anti-diabetic activity. Behavioral studies brought about after treating with *Aloe vera* extract (AVE) showed significant improvement in motor coordination and thermal sensitivity in diabetic-treated group when compared with the diabetic-control group. The staining of these Diabetic Rat`s sciatic nerve showed the distorted morphology of the axons, there by indicating axonal degeneration, and swelling of nerve fibre. In the AVE treated diabetic group, there was improved structure and nerve regeneration as was seen in the normal control Rats. Therefore, it is confirmed that AVE is a strong antioxidant against diabetes mellitus (DM) disorder and also its role in neuroprotection by attenuating the behavioral and morphological changes in streptozotocin (STZ) induced Diabetic Rat. Further studies are needed to know its exact mechanism of action on neurodegeneration mechanisms in diabetic Rat nerves.**

***Index Terms*: *Aloe vera*, Antioxidant, Diabetes, Morbidity, Neurodegeneration, Neuropathy, Sciatic nerve, Streptozotocin.**

I. INTRODUCTION

Diabetes is said to be one of the oldest diseases known to man (Patlak, 2002). It's a disease in which blood glucose concentration in the body are elevated due to the improper functioning of beta cells of pancreas. The blood glucose values are higher than normal in individuals suffering from DM. Diabetes is a major, tough, prevailing and pricey public health problem of the world.

Expansion in the overall diabetes prevalence rates largely reflects an increase in risk factors for type 2 DM. The menace of having diabetic neuropathy rises with age and duration of diabetes (Zhang *et al*., 2010). Many authors agreed that hyperglycemia causes tissue-damage through mechanisms like increase of sugar flux (Glucose & other) through the Polyol-pathway, enhanced formation of advanced-glycation-end products (AGEs) intracellularly, raised expression of the receptors for AGEs and its inducing ligands, PKC activation, and overwrought of the hexosamine-pathway (Brownlee, 2001 and Giacco *et al*., 2010). Oxidative stress of cells & tissues results due to the increased generation of ROS (Gumieniczek *et al*., 2002) and lead to lipid-peroxidation (Baynes, 1991) thereby causing membrane leakage. The toxic effects of hyperglycaemia results to these complications (Oyibo *et al*., 2002 and Romanovsky *et al*., 2010). *Aloe vera* is indigenous to Rajasthan, Andhra Pradesh, Gujarat, Maharashtra, UK, Himachal Pradesh, and Tamil Nadu. There are approximately 500 species of the genus Aloe belongs to Liliaceae family having healing property. The leaf pulp extract showed decreased glycaemic activity in diabetic (I & II) - rats (Okyar *et al*., 2001 and Rajasekaran *et al*., 2004) and diabetes- induced oxidative stress (Giugliano *et al*., 1996). The potentially active compounds like phytosterols, lophenol, cycloartenol and alkylated derivatives of them were present (Tanaka *et al*., 2006). The design of this study to know the result of extract of *Aloe vera* (AVE) on diabetic-neuropathy in streptozotocin infected experimental diabetic (wistar) rats.

II. MATERIALS AND METHODS

A. Experimental Animals and Induction of Type 2 diabetes:

A population of 20 male Albino wistar Rats (Rattus norvegicus) weighing 250±50g, were taken for the study. The maintenance of animals under standard (laboratory) conditions with standard pellets and water (*ad libitum)*. All the acclimatized animals were

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DOI: 10.37398/JSR.2020.640314

Selection and Peer-review under responsibility of the Program Chairs.

maintained following the guidelines of CPCSEA (CPCSEA No:383/01/a/CPCSEA). Diabetes was induced with streptozotocin (STZ) in overnight fasted Rats.

B. Plant Extract Preparation:

The plant gel was extracted from *Aloe vera* leaves. After washing and sterilizing it, the thick outer epidermal layer was carefully removed using a sterilized knife. The thick viscous jelly layer within, was homogenized subjected to extraction using 95% ethanol (72 hours). The evaporated residue stored in small dry pre-sterilized small containers at -20°C till using further. The dosing schedule followed was once per day.

C. Experimental Design:

1. The animals of control (CN) group received normal water.
2. The animals of diabetic control (DC) group received STZ 60mg/kg body weight (*i.p*).
3. The animals of diabetic + AVE group received the extract of *Aloe vera* plant (AVE) (300 mg/kg B.W/ml/day) orally from Day 1 till the end of the experiment after induction of diabetes.

Rat's sciatic nerves were dissected out (Mizisin, 2004) on day 15 and further tests were followed.

D. Chemicals Used:

Streptozotocin (Sigma-Aldrich, USA), other solvents were obtained from Himedia, India.

E. Behavioral Studies:

1) Rotarod test:

The level of coordination, balance, physical condition and motor activity of rats on rotarod was noted (Hutter-Saunders *et al*., 2012). Time was noted and obtained results were analyzed by statistical software program (SPSS.21).

2) Hot plate test:

Thermal nociceptive response was assessed by the hot plate test (Gunn *et al*., 2011).

F. Biochemical Estimation:

Protein content of sciatic nerve was measured (Lowry *et al*., 1951). The Na^+ - K^+ ATPase enzyme's activity measured by Kaplay (1978) and Taussky & Shorr (1953). Acetyl choline was estimated by Hestrin (1949) method.

G. Histopathological Studies:

The H&E stained formalin (10% v/v) stored sciatic nerves were observed using light microscope (Lillie *et al*., 1976).

H. Statistical Analysis:

The obtained results were expressed by using Mean ± SEM, one-way ANOVA and t-test.

III. Results and Discussion

A. Behavioral Tests:

The Diabetic rats exhibited decrease ($p<0.05$) in endurance time on rotarod as compared with CT group. The simultaneous treatment with AVE showed reversal ($p<0.05$) in the latency of fall during rotarod test compared to that of diseased group. As indicated in Fig.1. the results of AVE alone groups were found to be nearer to that of control group. The AVE administered diabetic rats showed reversal ($p<0.05$) of thermo nociceptive pain response when compared to the diabetic animals. The AVE received diabetic group of Rats performed comparatively well than the diabetic control group thus indicating the effectiveness of AVE. The diabetic control Rats showed decreased motor coordination on Rotarod as there was no strength for effective grip on the rotating cylindrical rod, due to their improper ability to stay on and leading to their fall off the rod that could be accredited to neural damage leading to loss of sensitivity and also loss of strength to respond. However, thermal sensitivity was instantaneous in the treated group. They started licking their paws immediately and were trying to jump out of the instrument.

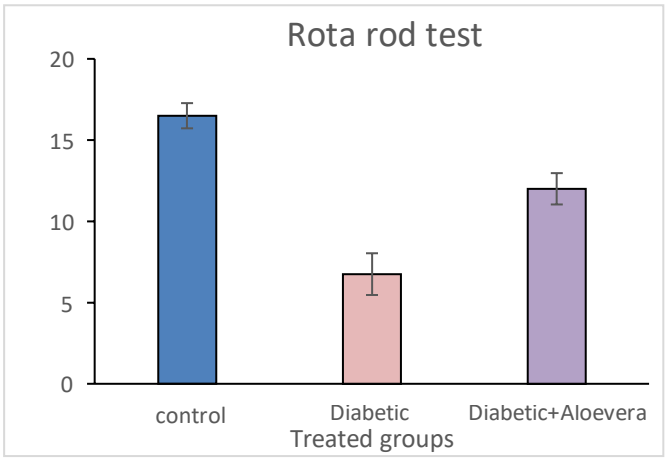

Figure 1: Protective effect of AVE on the motor coordination of diabetic rats.

Figure 2. Protective effect of AVE on thermal nociceptive pain in diabetic rats

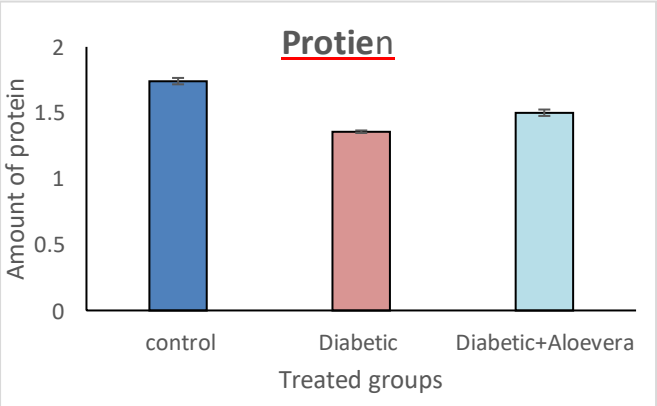

Figure 3. Protective effect of AVE on amount of Protein in diabetic Rats.

B. Effect of Biochemical Estimation:

The treated diabetic rats with AVE (p<0.05) attenuated the protein, Na^+-K^+ ATPase and Ach values of sciatic nerve tissue. The biochemistry of diabetic group shows that the activity of Na+-K+ ATPase, protein amount, acetylcholine, are decreased compare to control group, and rats received with *Aloe* gel increases biochemical changes (Baynes, 1991 & Brownlee, 2001).

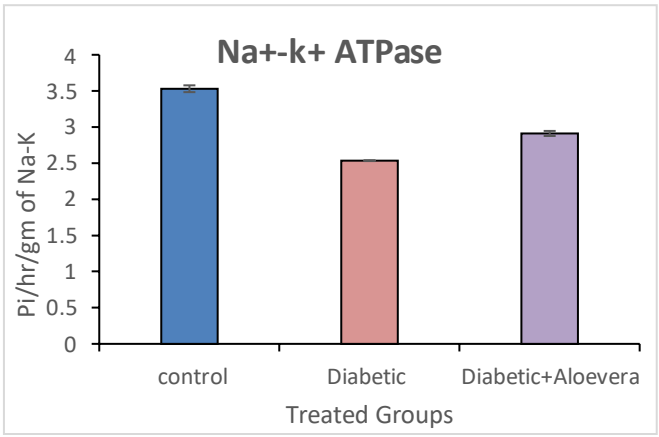

Figure 4. Protective effect of AVE on amount of Na+-k+ ATPase activity in diabetic rats.

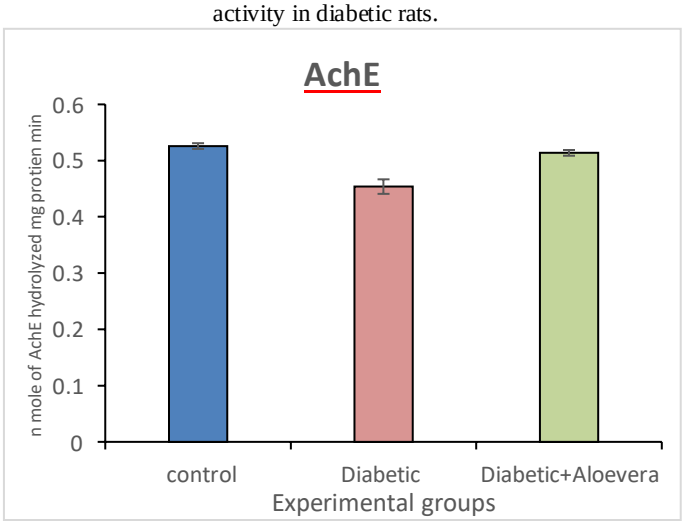

Figure 5**.** Protective effect of AVE on acetyl choline content in diabetic rats.

C. Histopathological Studies:

Histological study of the Rat's sciatic nerve (T.S) with H & E stain (Hematoxylin and eosin) (10X), showed the defensive effect of AVE shown in figure 6 below. Sciatic nerve from the NC rat (A), the connective tissue surrounding the nerve fascicle, perineurium, varied sizes of axons with myelin sheath and separated by an endoneurial connective tissue. However, in Diabetic rat (B) group SN showed cell shrinkage, de-myelination, cell necrosis & degeneration surrounded by "thin perineurium". Treatment of diabetic rats with *Aloe vera* (C) enabled the appearance of axons, myelin spaces & perineurium in normal. Histopathological evidence in this study clearly showed the occurrence of neuron degenerative changes due to diabetic neuropathy (Romanovsky *et. al*., 2010).

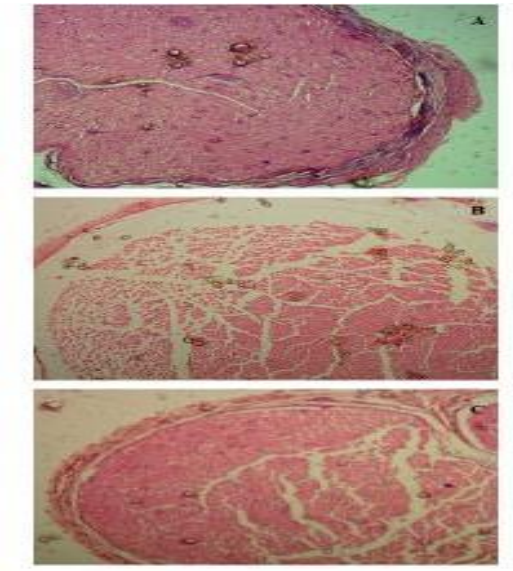

Figure 6. Effect of A*loe vera* gel extract (AVE) on sciatic nerve in diabetic rats

CONCLUSION

The prevalence of diabetes is rising worldwide due to over population, aging, urbanization, unhealthy lifestyle and the rise in number of obese individuals. Therapies like physical exercises, allopathic and natural drugs have the tendency to reduce the impact of (oxidative) stress and can be fruitful in combating diabetes associated damages. *Aloe vera* has confirmed its potential anti-oxidant nature in the current study ameliorating the oxidative-stress caused DM and also its role in neuroprotection by depreciating the behavioral, biochemical and morphological changes in streptozotocin-infected diabetic Rat. Therefore, it is confirmed that AVE is a potent antioxidative agent against diabetes neuropathy and neurodegeneration in diabetic rat nerves.

ACKNOWLEDGMENT

The funding & facilities were provided from UGC DSA I SAP II program and neuroscience lab, department of Zoology, Osmania

university.

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