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General Observations:
The STZ induced diabetic rats showed 15-20% mortality during the experimental period of 60 days whereas, no mortality was observed in the normal, normal treated and diabetic treated group rats. No visible side effects and variations in animal behavior (respiratory distress, abnormal locomotion and cathelepsy) are observed in normal rats treated with Commiphora mukul gum resin (EtCMGR), indicating the non-toxic nature of EtCMGR administration. Diabetic (D) group showed the characteristic signs of diabetes such as polyuria, polydipsia, polyphagia and failure to gain body weight. No significant variation was observed in intake of food and water among two groups (N & NT). Administration of (EtCMGR) suspension resulted in rectification of clinical signs of diabetes within 15 days of treatment in DT groups.

Body Weight:
Mean body weights of four experimental groups at 15 day intervals are summarized in table 1. No significant variation in initial weight was observed between two groups, N and NT, whereas, D and DT groups showed a slight but significantly lesser initial weight compared to N and NT groups. During the experimental period, D group showed a gradual decrease in body weight whereas, N, NT and DT groups showed a trend of gradual increase in body weight (Fig 1).

In spite of increased food consumption (Hyperphagia), decrease in body weight of diabetic rats is possible due to defect in glucose metabolism and increase in muscle wasting. Increased breakdown of proteins for providing substrate for gluconeogenesis and the absence of anabolic effect of insulin are responsible for muscle wasting and loss of weight under diabetic conditions (Swanston-Flat et al., 1990). Though DT rats showed a gradual increase in body weight at the end of experimental period, the body weight of DT group was significantly lower (24 %) than N group but significantly higher (52 %) than D group. Thus, C. mukul administration for 60 days partially prevented the weight loss in DT group compared to D group, and this may be due to improvement in insulin secretion and insulin action.
Table 1: Effect of *Commiphora mukul* treatment on body weight (grams) in STZ induced diabetic rats during the experimental period

<table>
<thead>
<tr>
<th>Groups</th>
<th>Initial 0 days</th>
<th>15 days</th>
<th>30 days</th>
<th>45 days</th>
<th>60 days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>182.54±1.99&lt;sup&gt;a&lt;/sup&gt;</td>
<td>207.00±2.11&lt;sup&gt;c&lt;/sup&gt;</td>
<td>232.21±1.25&lt;sup&gt;c&lt;/sup&gt;</td>
<td>255.01±4.00&lt;sup&gt;c&lt;/sup&gt;</td>
<td>282.00±1.99&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Normal Treated</td>
<td>184.34±2.31&lt;sup&gt;a&lt;/sup&gt;</td>
<td>207.45±1.13&lt;sup&gt;c&lt;/sup&gt;</td>
<td>229.71±2.05&lt;sup&gt;c&lt;/sup&gt;</td>
<td>252.19±1.87&lt;sup&gt;c&lt;/sup&gt;</td>
<td>276.56±1.47&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Diabetic</td>
<td>168.92±1.87&lt;sup&gt;b&lt;/sup&gt;</td>
<td>161.53±2.74&lt;sup&gt;a&lt;/sup&gt;</td>
<td>149.32±2.11&lt;sup&gt;a&lt;/sup&gt;</td>
<td>143.84±1.87&lt;sup&gt;a&lt;/sup&gt;</td>
<td>134.63±1.63&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Diabetic Treated</td>
<td>166.27±1.33&lt;sup&gt;b&lt;/sup&gt;</td>
<td>175.34±1.13&lt;sup&gt;b&lt;/sup&gt;</td>
<td>185.63±2.57&lt;sup&gt;b&lt;/sup&gt;</td>
<td>197.24±2.95&lt;sup&gt;b&lt;/sup&gt;</td>
<td>214.62±2.45&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Values are given as mean ± SE from eight rats in each group. Values of four Experimental groups not sharing a common superscript letter at respective days differ significantly at P<0.05 (Duncan's multiple range test).
Figure 1: Body weight of Normal (N), Normal treated (NT), Diabetic (D) and Diabetic treated (DT) rats during the experimental period.
Plasma glucose and plasma insulin level:

The data presented in table 2 and figure 2 indicates the fasting plasma glucose and insulin levels of 4 experimental groups during the experimental period. During the 60 days of experimental period the N and NT rats remained persistently euglycemic. The initial plasma glucose levels in D and DT group were 5-fold greater than to N and NT groups. During the experimental period, in D group the plasma glucose level was further elevated to 5.2, 5.1, 5.2, 5.6, and 5.5-fold whereas in the DT group the elevated glucose level gradually decreased to 5.3, 3.0, 2.0, 1.4, and 1.2-fold at 0, 15, 30, 45, and 60 days intervals compared to corresponding normal group. In D group, the plasma glucose level increased gradually during the experimental period from 388.31 ±4.63 to 425.24 ± 3.72 mg/dl. The percent increase in plasma glucose level of D group was 1, 4, 7, and 10 % at 15, 30, 45, and 60 days intervals compared to initial level whereas in C. mukul treated diabetic group (DT), a significant anti-hyperglycemic effect was evident from the 15 day onwards and the decrease in the glucose was maximum and reached near normal values by 60 days of treatment. The percent decrease in plasma glucose level of DT group was 42, 60, 73 and 76 % at 15, 30, 45, and 60 days of C. mukul treatment respectively. Thus, the present study indicates that chronic treatment of diabetic rats for 2 months with C. mukul decreased the plasma glucose level to the near normal values. The plasma glucose level of NT and DT groups during the experimental period clearly indicate that C. mukul does not exhibit hypoglycemic activity; instead, it shows anti-hyperglycemic effect.

Initial plasma insulin levels of D and DT groups were around 4 folds lower than N and NT groups. N rats showed no significant variation in insulin levels during experimental period. By the end of the experimental period, D rat plasma insulin levels decreased by 5.7 % i.e., 12.30±1.60 to 11.60±0.65 μ units/ml. Diabetic treated rats showed a significant increase in insulin concentration and percent increase was 29, 53, 89, and 128 % at 15, 30, 45, and 60 days respectively compared to initial level. Thus at the end of experimental period, the insulin level of DT rats was 152 % more than D group.
Table 2: Effect of Commiphora mukul treatment on fasting plasma Glucose (mg/dl) and Insulin (Units/ml) levels in STZ induced diabetic rats during experimental period.

<table>
<thead>
<tr>
<th></th>
<th>Plasma</th>
<th>0 Days</th>
<th>15 Days</th>
<th>30 Days</th>
<th>45 Days</th>
<th>60 Days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>Glucose</td>
<td>74.51±1.10a</td>
<td>75.93±0.77a</td>
<td>76.82±0.97a</td>
<td>73.14±0.91a</td>
<td>76.80±1.59a</td>
</tr>
<tr>
<td></td>
<td>Insulin</td>
<td>42.00±0.65a</td>
<td>41.80±0.71a</td>
<td>43.60±0.98a</td>
<td>45.50±1.70a</td>
<td>46.00±1.25a</td>
</tr>
<tr>
<td>Normal treated</td>
<td>Glucose</td>
<td>75.10±0.91a</td>
<td>73.52±1.10a</td>
<td>78.23±0.81a</td>
<td>75.60±1.09a</td>
<td>74.33±1.32a</td>
</tr>
<tr>
<td></td>
<td>Insulin</td>
<td>41.10±0.39a</td>
<td>43.20±0.76a</td>
<td>42.30±0.75a</td>
<td>43.60±0.59a</td>
<td>44.70±1.22a</td>
</tr>
<tr>
<td>Diabetic</td>
<td>Glucose</td>
<td>388.31±4.63b</td>
<td>391.2±32.30b</td>
<td>402.51±1.72c</td>
<td>414.60±2.63c</td>
<td>425.24±3.72b</td>
</tr>
<tr>
<td></td>
<td>Insulin</td>
<td>12.30±1.06b</td>
<td>11.85±0.58b</td>
<td>11.30±0.46b</td>
<td>12.00±0.49b</td>
<td>11.60±0.65b</td>
</tr>
<tr>
<td>Diabetic treated</td>
<td>Glucose</td>
<td>397.64±4.69b</td>
<td>229.11±11.59c</td>
<td>157.57±4.90b</td>
<td>106.52±3.66b</td>
<td>95.30±1.48a</td>
</tr>
<tr>
<td></td>
<td>Insulin</td>
<td>12.80±0.51b</td>
<td>16.50±0.56c</td>
<td>19.60±0.65c</td>
<td>24.20±0.59c</td>
<td>29.30±0.93c</td>
</tr>
</tbody>
</table>

Values are given as mean ± SE from eight rats in each group. Values of four experimental groups not sharing a common superscript letter at respective days differ significantly at P<0.05 (Duncan’s multiple range test).
Fig. 2a: Fasting plasma Glucose levels of Normal (N), Normal treated (NT), Diabetic (D), and Diabetic treated (DT) rats during the experimental period.

Fig. 2b: Fasting plasma insulin levels of Normal (N), Normal treated (NT), Diabetic (D), and Diabetic treated (DT) during the experimental period.
But this increase in plasma insulin levels does not reach normal values. The NT rats showed a gradual decrease in plasma insulin levels during experimental period at 45 and 60 days when compared to corresponding data of N rats. Thus *C. mukul* treatment has given partial protection against STZ induced depletion in plasma insulin levels. The diabetic rats showed severe hyperglycemia and hypoinsulinemia. Streptozotocin (2-deoxy-2-(3-(methyl-3-nitrosoureido)-D-glucopyranose) (STZ) is a nitrosourea derivative often used to induce diabetes mellitus in experimental animals through its toxic effect on pancreatic β-cells (Ruiz et al., 1994). It enters the β-cell via a glucose transporter (GLUT-2) and causes alkylations of DNA leading to generation of reactive oxygen and nitrogen species. As a result of the streptozotocin action, β-cells undergo destruction by necrosis (Szkudelski, 2001). Thus STZ diabetic rats show depletion in plasma insulin level resulting in hyperglycemia. Insulin plays a central role in regulation of glucose homeostasis, when circulating levels of insulin decreases in D rats resulted in hyperglycemia. The gradual decrease in plasma insulin levels during experimental period in D rats may propagate the β-cells damage due to hyperglycemia induced oxidative stress.

The possible explanation for the anti-hyperglycemic activity of *C. mukul* observed in DT rats may be due to the presence of a variety of pharmacologically important phytochemicals present in the resin of *C. mukul* which might have stimulated the insulin secretion or protected the intact functional β-cells from further deterioration or might have caused regeneration of damaged β-cells of pancreas. Thus it is probably due to fact that pancreas contains stable (quiescent) cells which have a capacity of regeneration (Govin et al., 1986., Kuman et al., 1992). In addition phytochemicals like flavonoids and alkaloids in EtCMGR might have protected the intact β-cells function from hyperglycemic induced oxidative stress on pancreas. It was also claimed that antioxidants cause possible beneficial effect in preventing type I diabetes by stopping the oxidative damage on pancreas and increasing the insulin secretion (Bravi et al., 2006). This possibility was strengthened that insulin concentration were also increased by antioxidant treatment (Ibrahim, 2008).
Brain - lipids:

Lipids are important constituents of the brain, not only because of the large surface-to-volume ratio of neurons-neurons but also contain a higher proportion of lipid than other cells because lipid is the main constituent of neuronal cell membrane (Greenwood, 2001). They play an important role in structural and functional activity. The central nervous system has the second greatest concentration of lipids, immediately after adipose tissue. It comprises 50-60% of dry weight of brain. These brain lipids contain a very high amount (35%) of long-chain polyunsaturated fatty acids (LCPUFA). The alterations in brain lipids lead to changes in physiological properties of membranes, in enzyme activities, receptors, transport, and cellular interactions and activities of membrane bound proteins (Bourre et al., 1989, Clandinin et al., 1997). These physiological changes are accompanied by learning and behavioral deficits in rats and mice (Yamamoto et al., 1988, Nakashima et al., 1993).

Total lipids constitute phospholipids, glycolipids, cholesterol, and triglycerides. The tissue lipids profile of N, NT, D and DT rats at the end of experimental period are presented in table 3. Diabetic group showed a significant decrease in TL (59%) content when compared with normal group. *C. mukul* treatment for 60 days caused a significant increase in TL (63%) content in DT rats when compared to D group. But this significant increase does not reach the normal values. Thus DT rats showed a still significantly lesser TL (34%) content in brain when compared to N group whereas *C. Mukul* treatment for 60 days to N rats showed a slight but not significant decrease in TL content when compared to N rats.

Lipids are major fuels for tissues such as skeletal muscle, heart muscle, adipose tissue, and liver but not of normal brain. Blood-derived glucose normally accounts for >90% of the fuel utilized by the brain (Siesjo, 1978; Lund-Anderson, 1979; Sokoloff, 1981; Pardridge, 1983). In type I and type II diabetes mellitus, whole-body glucose utilization is impaired. Some tissues, such as heart and skeletal muscle, can adapt to diabetes by increasing the ratio of fatty acids to glucose oxidation (Randle et al., 1963). The brain can also adapt to some extent. Thus, Ruderman et al., (1974) showed that cerebral metabolism is altered in experimental type I diabetes as
in starvation, the brain slowly decreases its utilization of glucose leading to insufficient energy production from glucose oxidation and to a lowered cytosolic acetyl-CoA concentration for fatty acid synthesis and remodeling. As a result the brain uses alternative fuels including ketone bodies and fatty acids. However, fatty acids are in short supply owing to limited transport across the BBB. This death of substrate leads to loss of endogenous fatty acids both by increased oxidation and by decreased synthesis. In addition earlier studies also indicate enhanced LPO in the brain of STZ (Montilla, 2005) and alloxan (Rizwan, 2005) in diabetic rat models. Fatty acid loss leads to changes in membrane composition and, indirectly, to neurotransmitter dysfunction. Prolongation of the metabolic perturbations leads to neuronal degeneration and abnormal brain function.

The decreased total lipid content of brain of D rats may be due to increased catabolism of lipids and/or enhanced LPO due to oxidative stress observed under diabetic conditions. C. mukul treatment for 60 days showed a partial recovery from diabetic induced decrease in TL content of DT rat’s brain. This may be due to its protective effect against diabetic induced alterations in lipid metabolism.

Anionic phospholipids contribute to the charge characteristics of cellular membranes and provide a proper microenvironment for many membrane-bound enzymes (Sun and Sun, 1985). Phospholipids and glycolipids are essential components of myelin, microsomal and mitochondrial fractions of brain. Cholesterol is an important constituent of brain and is used for membrane synthesis and for many other activities by cells throughout the body. Brain is the site of high lipid turnover approximately 25% of the total amount of cholesterol present in body is localized in brain and most of it is local synthesis (Dietschy, 2004). Thus there is a highly efficient recycling of cholesterol in the brain with minimal losses to the circulation and there is a profound loss of lecithin, cephalin, sphingomyelin, cholesterol and cerebroside in demyelinating nerve disease (Bjorkhem and Meaney, 2004). The adrenergic stimulation of the central or sympathetic nervous system might increase the mobilization of free fatty acids from peripheral depots. At the same time, there may be increased synthesis of triglycerides and decreased ability to mobilize triglycerides from brain tissues (Othman, 2005).
Table 3: Effect of *Commiphora mukul* treatment on lipid profile in brain of STZ induced diabetic rats.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Normal</th>
<th>Normal treated</th>
<th>Diabetic</th>
<th>Diabetic treated</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Total lipids</strong></td>
<td>330.65±14.70&lt;sup&gt;d&lt;/sup&gt;</td>
<td>300.21±6.27&lt;sup&gt;c&lt;/sup&gt;</td>
<td>135.16±15.76&lt;sup&gt;a&lt;/sup&gt;</td>
<td>220.00±6.53&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>(mg/g tissue)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Phospholipids</strong></td>
<td>64.80±1.42&lt;sup&gt;c&lt;/sup&gt;</td>
<td>61.76±1.29&lt;sup&gt;c&lt;/sup&gt;</td>
<td>46.21±2.06&lt;sup&gt;a&lt;/sup&gt;</td>
<td>51.69±1.53&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>(mg/g tissue)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Triglycerides</strong></td>
<td>8.44±0.22&lt;sup&gt;a&lt;/sup&gt;</td>
<td>7.63±0.32&lt;sup&gt;b&lt;/sup&gt;</td>
<td>11.28±0.24&lt;sup&gt;d&lt;/sup&gt;</td>
<td>8.84±0.44&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>(mg/g tissue)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Cholesterol</strong></td>
<td>32.52±0.97&lt;sup&gt;d&lt;/sup&gt;</td>
<td>29.21±0.36&lt;sup&gt;c&lt;/sup&gt;</td>
<td>24.42±1.39&lt;sup&gt;a&lt;/sup&gt;</td>
<td>28.31±0.80&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>(mg/g tissue)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Glycolipids</strong></td>
<td>3.49±0.22&lt;sup&gt;c&lt;/sup&gt;</td>
<td>4.39±0.27&lt;sup&gt;d&lt;/sup&gt;</td>
<td>0.96±0.11&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.06±0.22&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>(mg/g tissue)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Values are given as mean ± SE from eight rats in each group. Values of four experimental groups not sharing a common superscript letter differ significantly at P<0.05 (Duncan's multiple range test).
Figure 3: Percent change in Total lipids, Phospholipids, Triglycerides, Cholesterol, and Glycolipid in the brain of D and DT rats compared with N rats.
increase in PL, GL, and cholesterol (12 %, 114 % and 16 %) and significant decrease in TG (22 %) in DT rats when compared to D group. But this significant increase PL, GL, and cholesterol does not reach the normal values whereas TG levels are normalized. Thus PL, GL, and cholesterol levels (20 %, 41 % and 13 %) in DT rats were still significantly lesser (25%, 40% and 14%) when compared to N group whereas C. mukul treatment for 60 days to N rats showed a slight increase in GL (54 %) and decrease in cholesterol and PL (10 % and 5 %) and no significant change in TG levels when compared to N rats.

The decreased PL, GL, and cholesterol and increased TG content of D rats compared to N rats indicate the presence of lipid damage or oxidation. In STZ induced diabetic rat brain alterations may be responsible for demyelization and nerve degeneration. Our reports of decreased PL and cholesterol content in diabetic rats are in agreement with earlier studies (Malaisse et al., 2006) suggesting that membrane function may be altered and in addition to physicochemical alterations, the specific activities of several membrane bound enzymes (AchE, Mg$^{2+}$ - ATPase, and Na$^+$, K$^+$ - ATPase) are significantly decreased and play a role in the development of diabetic complications in brain. The decrease in membrane fluidity of diabetic brain could be due to the peroxidation of membrane phospholipids through free radicals, which is generated by persistent hyperglycemia. Similar reports were also observed with regard to the effect of morphine sulphate on brain (Othman, 2005).

Very few studies are available on the protective effect of plants on altered lipid components in diabetic animals. Many plants are known to exhibit hypolipidemic activity in experimentally induced diabetes, cholesterol fed and triton induced hyperlipidemia animals. Semecarpus anacardium extract (Sharma et al., 1995) and Terminalia belerica (Shaila et al., 1995) exhibited hypocholesterolemic action in hypercholesterolaemic rabbits. Similarly ethanolic root extract of Plumbago Zeylanica, alone and in combination with vitamin E, significantly reduced serum total cholesterol, LDL-Cholesterol and triglycerides levels in experimentally induced hyperlipidemic rabbits (Ram, 1996). Guggulipid, an alkaloid of Commiphora mukul (Dalvi et al., 1994), dried flowers of Adenocalymma alliaceum (Srinivasan and
Srinivasan, 1995), *Trigonella graceum* (Sharma et al., 1990) are reported to possess hypolipidemic activity in hypercholesterolaemic rats and in type 1 diabetic animals.

The loss and ineffective utilization of glucose in diabetes leads to breakdown of fat and protein. The increased mobilization and utilization of fat which meets the energy requirement results in hyperlipidemia especially an increase in non-esterified fatty acids, TG, and cholesterol levels of plasma. Diabetes has been associates with an increased risk of developing premature atherosclerosis due to increase in TG and LDL levels and decrease in HDL levels (Surekha et al., 2005). Thus dyslipidemia is a well known characteristic feature of diabetes due to altered lipid metabolism in many tissues.

**Acetylcholine & Acetylcholine esterase:**
Generation of oxidative stress and consequent lipid peroxidation was reported in the brain of diabetic induced rats. It is expected that activities of membrane-bound proteins and enzymes (AchE and ATPase) and other membrane-associated phenomenon would be disturbed by peroxidative membrane damage. There are evidences suggesting that diabetes is associated with abnormalities of neurotransmitter turnover. AchE in brain is chiefly localized in neurons and belongs to a family of hydrolases. It hydrolyses acetylcholine and is used as a marker for cholinergic neural function. It is known to be involved in synaptogenesis and regulates cholinergic nerve and neuromuscular transmission. AchE is known to be anchored to the membrane through covalent linkage to phosphoinositoldiacylglycerol (Low et al., 1986). This enzyme like protein was shown to mediate cytoarchitectural changes that support the neurotogenesis (Appa Rao, 2008). Further, AchE is known to have many non-classical functions. There is a growing body of evidence for morphogenic role of AchE. During early development, AchE expression is tightly correlated with neurite outgrowth, in addition AchE role in cell survival and growth (Appleyard, 1992). Thus, there is a high correlation between cholinergic function, AchE activity and cognition. Accordingly, estimation of AchE activity provides an important correlate of cholinergic activity and cognitive function.
Acetylcholine long been known to be present in the brain and its synthesis and metabolism are well established. Ever since the discovery of Ach as a neurotransmitter, its function in health and dysfunction in disease has been increasingly recognized. Further, pharmacological manipulation of cholinergic function has been found useful in the treatment of CNS disorder like Alzheimer's and Parkinson's disease. Thus, assessing cholinergic function is considered as an important tool in neuroscience research.

In the present study we examined the concentration of Ach and the activity of AchE in the brain of four experimental groups and the data is presented in table 4 and figure 4. Diabetic group showed a significant decrease in the activity of AchE (8 %) and increase in Ach (29 %) content when compared with N group. C. mukul treatment for 60 days resulted in a significant increase in the activity of AchE (40 %) and significant decrease in Ach (48 %) content in DT rats when compared to D group. The significantly low concentration of neurotransmitter Ach in DT rats compared to D rats reached the normal values, whereas the activity of AchE (29 %) of DT rats has increased beyond the level of N rats. Treatment with C. mukul for 60 days in N rats showed a significant higher activity of AchE (68 %) and slight decrease in Ach (36 %) content when compared to N group. Thus, the present study indicates the protective effect of C. mukul treatment in maintaining the normal level of neurotransmitter i.e., Ach even under STZ induced diabetic state.

The changes in cholinesterase activity might reflect impairment in biosynthesis, degradation or insertion into plasma membrane. Our reports of increased concentration of Ach and decreased activity of AchE in diabetic rats are in agreement with earlier studies (Szutowicz et al., 1994, Dash et al., 1991, Makar et al., 1995). Enhanced oxidative stress reflected by increased lipid peroxidation in D rats brain may be the reason for observed inhibition of AchE activity and consequent increase in the Ach level in the present study. Accumulation of Ach at synaptic regions could lead to desensitization of the cholinergic receptors on the postsynaptic membrane rendering it ineffective to the transmitter action. Abnormal effects of AchE activity have been reported in various diseases including diabetics.
Table 4: Effect of *Commiphora mukul* treatment on acetylcholine content and activity of acetylcholine esterase in brain of STZ induced diabetic rats.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Normal</th>
<th>Normal treated</th>
<th>Diabetic</th>
<th>Diabetic treated</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acetylcholine esterase</td>
<td>0.296±0.019&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.498±0.026&lt;sup&gt;d&lt;/sup&gt;</td>
<td>0.274±0.009&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.382±0.023&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>(μmoles of Ach hydrolysed/min/mg protein)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Acetylcholine</td>
<td>3.59±0.17&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.28±0.11&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.39±0.18&lt;sup&gt;c&lt;/sup&gt;</td>
<td>2.80±0.18&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>(μmoles of Ach/g tissue)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Values are given as mean ± SE from eight rats in each group. Values of four experimental groups not sharing a common superscript letter differ significantly at P<0.05 (Duncan’s multiple range test).
Figure 4: Percent change in the Acetylcholine content and activity of Acetylcholine esterase in brain of D and DT rats compared to N rats.
Several Studies have reported decreased activity of choline esterases during the diabetes leading to accumulation of Ach, cholinergic hyperactivity, convulsion, and status epileptics (Ragoobirsingh et al., 1992, Rizvi et al., 2001, Dave, 2002). In addition to regulating blood glucose levels, insulin plays a neuromodulatory role in the CNS (Kwok et al., 1985, Figlewicz and Szot, 1991). Thus uncontrolled type II diabetes, in addition to interfering with cerebral energy metabolism, may also compromise neurotransmitter function, uncontrolled diabetes in animals models leads to altered neurotransmitter metabolism (McCall, 1992), and abnormal levels of several neuropeptides (McCall, 1992). Insulin signaling abnormalities could be the underlying mechanism affecting the outcome of Alzheimer’s disease; insulin resistance and disorder degradation of amyloid seem to link diabetes mellitus with Alzheimer’s disease.

The increased activity of acetylcholine esterase by *C. mukul* treatment observed both in DT and NT rats may be due to increased plasma insulin concentration and decreased blood glucose levels in DT rats compared to D rats and increased insulin sensitivity in NT rats. Similar to our studies decreased AchE activity in brain and heart can be reversed by insulin administration (Gumieniczek et al., 2002).

The decreased activities of AchE and BchE by LPO were reversed when diabetic rats were treated with N-benzoyl-D-phenyl alanine and metaformin (Natarajan, 2007). Anti-diabetic drugs could be potentially useful in treating Alzheimer’s disease: PPAR gamma agonists, by improving insulin sensitivity, decreasing inflammation and improving cerebral energy metabolism; intranasal insulin, by restoring brain insulin levels in Alzheimer’s disease.

Thus restoration of Ach concentration and increased AchE activity in DT rats may be due to improved plasma insulin level by *C. mukul* treatment.
Proteins and Transaminases:
Concentration of tissue proteins are affected by alterations in dietary proteins and also by hormonal factors. Insulin has been shown to stimulate tissue protein synthesis (Jepson et al., 1988). Although the brain is an insulin-independent tissue, owing to the limited penetration of pancreatic insulin, several reports indicate the presence of insulin-sensitive areas and insulin binding sites (LeRoith et al., 1988; Azam et al., 1990) and the possibility of insulin playing a role in brain metabolism and function. Investigators have reported that protein synthesis declines in liver and muscle of diabetic rats (McNurlan and Garlick, 1980; Millward et al., 1976), and that insulin treatment stimulated protein synthesis in muscle (Jefferson 1980, Jepson et al., 1988; Pain and Garlic, 1974). Measurements of tissue transaminases are useful to assess the tissue functions. Both GPT and GOT are important enzymes which are located in the cytoplasm and mitochondria of brain cells (Benjamin, 1978, Ringer and Dabieh, 1979). Their activities are related to protein metabolism and the maintenance of amino acid homeostasis and might be an indicator of mitochondrial injury (Cohen, 1970). Further, enhanced proteinglycation and oxidative stress under hyperglycemic conditions indicates altered protein metabolism in diabetes. In order to understand the altered protein and amino acid metabolism in the brain under diabetic condition and to assess the therapeutic efficacy of C. mukul, total protein content and activities of transaminases (GOT and GPT) measured in the four experimental groups and the data presented in the table 5 and figure 5.

STZ induced diabetic rats showed a significant decrease in protein content (28 %) with significantly increased activities of GOT (67 %) and GPT (24 %) in brain when compared with N group. C. mukul treatment for 60 days in DT rats resulted in a significant increase in protein content (18 %) with significantly decreased activities of GOT (27 %) and GPT (15 %) in brain compared to diabetic group. But this change in protein content and transaminases in DT rat does not reach the normal values. Thus, C. mukul treatment for 60 days has given a partial protection from altered protein and amino acid metabolism in the brain of diabetic rats. However NT rats showed no significant variation in protein content and activities of GPT and GOT when compared to N rats.
Table 5: Effect of *Commiphora mukul* treatment on protein content and activities of transaminases in brain of STZ induced diabetic rats.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Normal</th>
<th>Normal treated</th>
<th>Diabetic</th>
<th>Diabetic treated</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein (mg/g tissue)</td>
<td>28.11±0.79&lt;sup&gt;d&lt;/sup&gt;</td>
<td>22.57±0.52&lt;sup&gt;b&lt;/sup&gt;</td>
<td>20.26±0.62&lt;sup&gt;a&lt;/sup&gt;</td>
<td>23.88±0.70&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>GPT (µg of Pyruvate formed/min/mg protein)</td>
<td>3.58±0.22&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.50±0.16&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.45±0.07&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.79±0.13&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>GOT (µg of Pyruvate formed/min/mg protein)</td>
<td>2.89±0.20&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.01±0.11&lt;sup&gt;b&lt;/sup&gt;</td>
<td>4.83±0.15&lt;sup&gt;d&lt;/sup&gt;</td>
<td>3.53±0.11&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Values are given as mean ± SE from eight rats in each group. Values not sharing a common superscript letter differ significantly at P<0.05 (Duncan's multiple range test).
Figure 5: Percent change in the activities of transaminases in the brain of D and DT rats compared to N rats.
Our reports of decrease in protein content and increase in extent of GPT and GOT activities are supported by earlier studies in diabetic brain (Lemberg et al., 2007). This may be due to increased protein turnover and also increased catabolism of amino acid in the brain of diabetic rats due to enhanced protein oxidation and proteinglycation observed under hyperglycemic conditions. Enhanced non-enzymatic glycation of proteins in diabetes may decrease the half life of proteins thus contributing to the enhanced protein degradation (Vlassara and Palace, 2002). Inefficient utilization of glucose in insulin deficiency state leads to enhanced breakdown of protein thereby providing substrates for ketone bodies by decreasing the utilization of glucose in brain of type I diabetes (Ruderman et al., 1974). Increased aminotransferase activities might participate in the enhanced synthesis of excitatory amino acid neurotransmitters in the nervous system (Santos et al., 1987).

These transaminase reactions are reversible, but the equilibrium of the GOT and GPT reactions favour formation of asparate and alanine respectively (Moss and Henderson, 1999). The observation that glutamate oxaloacetate transaminase (GOT) is present in rat cerebral homogenate at about the same concentration as in liver suggests its importance in the brain amino acid pool homeostasis (Streecher, 1970). The GOT activity is found in both mitochondrial and soluble fractions of the brain. The enzyme in mitochondria is latent and can be activated by various procedures that disrupt the mitochondrial structure. The critical role of mitochondrial dysfunction in the pathogenesis of age-related nerve cell degeneration has been suggested in several occasions (Beal et al., 1993; Schulz et al., 1996). Increased GOT activity may be connected with an increased transport of NADH from the cytosol to mitochondria (Netopilova et al., 2001). Matthews et al (2000) reported that both the enzymes degrade glutamate; though only GPT was able to reduce toxic (500 µM) levels of glutamate into the physiologic (<20 µM) range. The excitotoxic effect of glutamate is believed to be the cause of several neurodegenerative processes (Mahy et al., 1999) and several enzymes with the capacity to degrade glutamate have been suggested as possible neuro-protectants (Matthews et al., 2000). Thus, enhanced
transaminase activities in D rats indicate an adaptive mechanism to decrease the glutamate level in the brain.

The partial rectification of decreased protein content and enhanced transaminases activities observed in DT rats by *C. mukul* treatment may be due to its antihyperglycemic activity with enhanced insulin secretion. Thus *C. mukul* treatment to diabetic rats showed improved brain energy metabolism.

**Oxidative biomarkers:**
Oxidative stress has been implicated in the development and complications of diabetes. Hyperglycemia and insulin resistance or insufficiency in diabetes can cause oxidative stress by excessive reactive oxygen species and can increase damage and alter antioxidant status in nerve cells. Oxidative stress is a relative overload of oxidants which leads to the destruction of both were renal and vascular cells of the central nervous system. High oxidative stress can lead to micro vascular cerebral diseases, e.g. stroke, cerebral hemorrhage, and brain infraction (Kannel and McGee, 1979; Paton and Passa, 1983). The reason for high risk of cerebral diseases, inspite the fact that brain consumes 20% of oxygen in the body, is that it has low concentration of antioxidants and high content of unsaturated fatty acids and catecholamines that are easily oxidized, making the brain more vulnerable to oxidative damage than any other organ in the body. Moreover, brain has a high ratio of membrane surface area of cytoplasmic ratio, extended axonal morphology prone to injury, and neuronal cells are non-replicating. The central nervous system is vulnerable to oxidative stress; especially when a toxicant can modify the physiological balance between anti- and pro-oxidant mechanisms (Gonthier et al., 2004). The development of diabetes associated complications in nervous system was attributed to the increased glucose concentration. Thus hyperglycemic is the most important factor in the onset and progress of diabetic complications mainly by oxidative stress through excessive production of ROS. The oxidized or nitrosylated products of free radical attack have decreased biological activity leading to loss of energy metabolism, cell signaling, transport and other major functions.
These altered products are also targeted for degradation further decreasing cellular functions. Accumulation of such injury ultimately leads the cell to die through necrotic or apoptotic mechanisms.

Hyperglycemia induced tissue damage favors some particular subset of cell types which are involved in diabetic complications like capillary endothelial cells of retina, mesengial cells of glomerulus's, neurons and Schwann cells in peripheral nerves. This is because most cells are able to reduce the transport of glucose inside the cell when they are exposed to hyperglycemia so that their internal glucose concentration stays constant. In contrast, the cells damaged by hyperglycemia are those that cannot do this efficiently (Kaiser et al., 1993, Heilig et al., 1995), as a result a hyperglycemic condition leads to enhanced glucose concentration inside the cells. This increased intracellular glucose load leads to auto-oxidation of glucose, generation of free radicals, elevated LPO, increased polyol pathway flux, increased formation of AGE's, activation of protein kinase C and increased hexosamine pathway flux, thus ultimately enhanced oxidative stress.

The extent of oxidative stress in STZ diabetic rats and to evaluate the beneficial effect of *C. mukul* against diabetes induced oxidative stress, extent of LPO, protein glycation, and activities of Sorbitol dehydrogenase and XOD are assessed in brain of four experimental groups.

**Lipid peroxidation:**
Lipid peroxidation is a free radical-related process, which is potentially harmful because its uncontrolled, self-enhancing process causes disruption of membranes, lipids and other cell components. ROS can increase the permeability of blood brain barrier, inhibit the mitochondrial respiration and can lead to a geometrically progressing lipid peroxidation (Gilman et al., 1993). Free radicals generated in the brain are also reported to influence gene expression, subsequently effecting apoptosis and neuronal death (Gligun et al., 2000). In the diabetes mellitus abnormal increased levels of lipid, lipoprotein and lipid peroxides in plasma may be due to the abnormal lipid metabolism (Suckling and Jackson, 1993).
If there is an imbalance between the levels of free radicals and antioxidant enzymes the amount of free radicals would increase and provoke LPO. ROS especially superoxide anion radicals, hydroxyl radicals and alkyl peroxyl radical are potent initiators of lipid peroxidation. MDA, the end product of the LPO of membrane lipids currently considered to be basic markers of oxidative stress, accumulates when lipid peroxidation increases.

Extent of LPO in brain was studied in four experimental groups and presented in table 6 and figure 6. Normal rats treated with C. mukul showed no significant changes in extent of LPO when compared to N rats. STZ induced diabetic rats (78 %) showed a significant increase in the extent of LPO in brain compared to N rats. C. mukul treatment resulted a significant decrease in LPO in DT rats (29 %) when compared to D group. However, LPO of DT group (26 %) is still significantly greater than N rats. Thus, C. mukul treatment has given a partial protection against enhanced LPO observed under STZ induced diabetic conditions.

The present data revealed that the brain of diabetic rats showed a consistently raised level of MDA, the biomarkers of oxidative damage of lipid-rich membrane. An increase in MDA in the brain of the rats would have profound effect as it is indicative of increased chance of tissue damage. Our reports of increase in extent of LPO in brain tissue of diabetic rats is in agreement with earlier studies (Leelavinothan Pari et al., 2007; Kumar et al., 1993; Manisha Rastogi et al., 2008) suggesting that the increase in extent of LPO plays a role in the development of diabetic complications in brain. Similar reports were also observed in arsenic induced toxicity in mice brain (Rao and Avani, 2004) and aluminium induced changes in rat brain (Kaur et al., 2003). The enhanced peroxidation of membrane lipids in STZ diabetes may result in loss of membrane integrity and function which is further reflected in the present study by decreased activity of membrane bound enzyme like AchE. The observed significant decrease in LPO of DT group compared to D group indicate that C. mukul treatment might have given protection against free radical mediated damage by its antioxidant potential and/or by its glycemic control and
quenching the free radicals. Guggulipid, the resin of *C. mukul* was reported to decline LPO in hypercholestermic humans (Singh et al., 1994) and Cu$^{+2}$ mediated in vitro lipid peroxides (Ramesh chader, 2003). Till the date, several chemical components such as diterpenes, sterols, steroids, esters and higher alcohols have been identified in *C. mukul* (Zhu et al., 2001; Wang et al., 2004). The active lipid lowering agent, a standardized fraction from ethyl acetate extract of guggul gum containing guggulsterone mixed with some other steroids, diterpenes, esters and higher alcohols named as guggulipid and guggulsterone have been demonstrated to reduce the risk of cardiac events and improves cardiac function in experimental and clinical studies (Ulbricht et al., 2005; Gaur et al., 1997; Sheela et al., 1995; Nityanand et al., 1989; Szapary et al., 2003). The protective action of guggulsterone is due to antioxidant property because it inhibits the generation of oxygen free radicals (Chander et al., 2002). Guggulsterone also reported to have protection against oxidative modifications of lipid and protein components of LDL induced by Cu$^{+2}$ *invitro* (Singh et al., 1977). A standard alcoholic fraction from guggulipid containing E-guggulsterone is already studied and prove for its protection against free radical damage in the skin and scavenging of superoxide anions ($O_2^-$) and hydroxyl radicals (OH$^-$) in non-enzymatic test systems (McCord et al., 1969).

**Protein glycation:**

Protein oxidation and glycation are post-translational modification that are implicated in the pathological development of many age related diseases process and diabetic complications. Chronic hyperglycaemia contributes to tissue and organ damage- retina, kidney and nerves by promoting the formation of advanced glycation end products (AGE). The AGE accumulation both in intra and extracellular proteins plays an essential role in the pathogenesis of diabetic complications by production of cross links on extracellular matrix proteins, by interaction with specific cellular receptors and by modification of nucleic acids. In diabetes, these reactions are greatly accelerated and are important in the pathogenesis of diabetic complications. One of the consequences of hyperglycemia is the excessive non-enzymatic glycation of proteins known as Millard reaction and leads to their structural and functional changes, resulting in complications of the diabetes.
The extent of protein glycation in brain was measured in four experimental groups and presented in the table 6 and figure 6. Normal rats treated with *C. mukul* resin showed a no significant change in extent of protein glycation when compare to N rats. Diabetic and DT groups showed a significant increase in extent of protein glycation (101 % and 32 %) when compared with N group. Thus a significant decrease in protein glycation was observed in DT group compare to D group which indicated the protective role of *C. mukul* against hyperglycaemia induced proteinglycation. However, the extent of proteinglycation in the DT rats is still significantly greater than normal values. Hence *C. mukul* treatment has given a partial protection against hyperglycemia induced protein glycation in DT rats.

Glycation initiated changes in brain proteins play an important role in the development of diabetes related pathological changes (Vlassara et al., 1983). Brain is an insulin independent tissue and leads to high exposure to glucose. Enhanced glycosylation by elevated glucose concentration may induce the formation of oxygen derived free radicals through protein glycosylation, which releases early and late glycosylation end products contributing to enhancement of oxidative stress. Both protein glycosylation and protein oxidation are biochemical alterations occurring in diabetes (Resm et al., 2001) and implicated in the pathological development of many age related disease process. In addition the enhanced protein glycation in D rats may also due to enhanced operation of polyol pathway leading to the production of fructose from sorbitol. Fructose is reported as a potent glycosylating agent than glucose. Protein glycation was shown to alter the protein conformation and function and decrease its half-life period and subjected to fragmentation. This protein fragmentation is inhibited by anti oxidants confirming that tissue damage associated with diabetes has an oxidative origin. Antioxidant like vitamin E has been shown to block glycation of protein (Jainapalmer, 1997). These modifications of intracellular proteins including, most important proteins are involved in the regulation of gene transcription (Giardinol et al., 1994; Shinohara et al., 1998). These AGE precursors can diffuse out of the cell and modifies the extra cellular matrix molecules near by
(McLellan et al., 1994) which change signaling between the matrix and the cell and causes cellular dysfunction (Charonis et al., 1990). AGEs increase in the brain during normal aging. Interestingly, experimental evidence supports the premise that AGEs are further increased in the brain in the presence of vascular or Alzheimer's dementia (Bar et al., 2003; Girones et al., 2004). Further, in diabetes and Alzheimer's disease, evidence suggests that CML (Crboxymethyl-Lysine) accumulation is greater than that seen in Alzheimer's disease alone (Girones et al., 2004). Further, multiple studies suggest that AGEs are directly neurotoxic to cultured neurons (Chen et al., 1994; Takeuchi et al., 2000). AGEs and their precursors (methylglyoxal and glyoxal) may increased the aggregation and cytotoxicity of intracellular amyloid-beta carboxy-terminal fragments (Woltjer et al., 2003). Taken together, these considerations underscore the premise that AGEs may be central to the exacerbation of dementia and enhanced prediction of stroke. The enhanced protein glycation of D rats compare to N rats indicates the presence of increased rate of non enzymatic glycation in D group. Our reports of increase in extent of protein glycation in diabetic rat brain are in agreement with those by earlier studies (Vlassara et al., 1983; Brownlee et al., 1988; Das et al., 2004) suggesting that the increase in extent of protein glycation plays a role in the development of diabetic complications in brain.

Beneficial effect of C. mukul treatment was observed in the DT rats by decreasing the extent of protein glycation compared to D rats. This declined protei glycation may be due to its antihyperglycemic activity and/or by its antioxidant potential. Similar our observations, Dearlove et al., (2008) reported that the extract of Culinary Herbs and Species showed inhibitory action against fructose-mediated protein glycation which was correlated to their phenolic content. Ethanolic extract of Lawsonia inermis and gallic acid are reported to have protective activity against protein oxidation and glycation. Even though C. mukul resin was not reported to contain phenols. The other phytochemicals of resin might have shown inhibitory activity against protein glycation. Thus, C. mukul resin treatment may prevent or delay the progress of diabetic complications in the brain due to protein glycation.
Sorbitol dehydrogenase:
The development of diabetic associated complications in the nervous system was found to be directly attributed to the increased glucose concentration as well as increased polyol pathway activity in brain of diabetic patients. Under normal conditions, there is little flux through the polyol pathway. However, under conditions of diabetic hyperglycemia, the flux through the pathway increases, as there is an overflow of glucose from the normal metabolic pathways such as glycolysis. Glucose uptake in peripheral nerves is not dependent on insulin. Therefore, high blood glucose levels in diabetes lead to high nerve glucose concentration and enhanced operation of polyol pathway. It consists of two successive dehydrogenases, namely aldose reductase (alditol: NADP⁺ 1-oxidoreductase) and sorbitol dehydrogenase (L-iditol:NAD⁺ 5-oxidoreductase). Conversion of glucose to sorbitol by aldose reductase requires NADPH and form NADP⁺ and thereby competes with other NADPH requiring reactions. Conversion of sorbitol to fructose by sorbitol dehydrogenase is coupled to reduction of NAD⁺ to NADH and this competes with glycolysis at the glyceraldehydes step for NAD⁺ (Gonzalez et al., 1986).

Hyperglycemia in non-insulin dependent cells activate AR enzyme which leads to intensive metabolism of glucose into sorbitol and then to fructose. It reduces the proportion of NADPH/NADP and increases the proportion of NADH/NAD. The trouble in the oxidation of NADH in the respiratory chain is indicated as "hyperglycemic pseudo hypoxia" and leads to increasing the quantity of ROS in the cells. Enhanced activity of AR under diabetic state depletes its cofactor NADPH which is also required for the glutathione reductase to regenerate glutathione (GSH), causing substantial decrease in the cellular antioxidant capacity. Previous researchers revealed an accumulation of sorbitol and fructose in neural tissues of diabetic animals (Amano et al., 2002). Schwann cells of diabetic rats show abundantly expressed AR which results in the development of both axonopathy and sensory neuropathy due to accumulation of sorbitol (Calcutt et al., 2004). The accumulated sorbitol, in nervous tissue of diabetic animals, can increase the cellular osmolarity, resulting in water retention; cell oedaema and increases in cytosolic Na⁺.
concentration may contribute to the etiology of diabetic neuropathy (Nattrass, 1986). In animal models, treatment with AR inhibitors (ARI) was shown to be effective in preventing the development of various diabetic complications, including cataract, neuropathy, and nephropathy. Although there has been extensively research on aldose reductase, the role of SD has been overlooked.

The activity of SD in brain was measured in four experimental groups and presented in table 6 and figure 6. STZ induced diabetic rats showed a significant increase in the activity of SD reaching about (100 %) of the normal values. *C. mukul* treatment for 60 days in DT rats results in a significant decrease in activity of SD (45 %) in brain when compared to D group. But this change in SD activity in DT rats (10 %) does not reach the normal values still it is higher than N rats. Thus, *C. mukul* treatment for 60 days has given a partial recovery from enhanced SD activity observed in the brain of STZ induced diabetic rats. However, N rats treated with *C. mukul* have showed a significant decrease in SD (20 %) activity compared to N rats.

The current increase in the activity of SD could be contributed to increased availability of sorbitol; substrates of SD of diabetic rats compared to N rats indicate the enhanced activity of AR enzyme in these animals. Thus reflecting the enhanced operation of polyol pathway in the brain of STZ diabetic rats. The increased brain SD activity, shown in STZ diabetic animals, might be considered as an enzymatic adaptation which facilitates the degradation of the reported accumulated sorbitol to fructose. Nerve fructose levels may also increase and may initiates the glycation process in brain, as fructose promotes glycation more efficiently than glucose. More over, a novel monosaccharide phosphate, fructose-3-phosphate was identified in the nervous system of diabetic animals. Fructose-3-phosphate was reported as a potent protein glycosylating agent and is enzyme in activator. Enhanced SD activity was reported in diabetic brain (Obrosova et al., 1999, Ibrahim, 2008) of experimentally induced diabetic animals. Over expression of SD stimulates ROS generation in high glucose exposed retinal pericytes, and subsequently potentiates the cytopathic effects of glucose (Amano et al., 2002).
Table 6: Effect of *Commiphora mukul* on STZ induced diabetic alterations in rat brain - LPO, Protein glycation, activities of SD and XO.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Normal</th>
<th>Normal treatment</th>
<th>Diabetic</th>
<th>Diabetic treated</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lipid peroxidation (nmoles of MDA formed/mg protein)</td>
<td>0.190±0.022&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.164±0.009&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.338±0.021&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.240±0.018&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Protein glycation (nmoles of Fructose/mg protein)</td>
<td>30.86±0.47&lt;sup&gt;a&lt;/sup&gt;</td>
<td>34.25±0.61&lt;sup&gt;b&lt;/sup&gt;</td>
<td>62.01±1.35&lt;sup&gt;d&lt;/sup&gt;</td>
<td>40.79±1.07&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Sorbitol dehydrogenase (nmoles of NADH consumed/min/mg protein)</td>
<td>0.106±0.010&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.134±0.004&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.214±0.011&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.116±0.006&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Xanthine oxidase (μmoles of uric acid formed/min/mg protein)</td>
<td>24.52±1.27&lt;sup&gt;a&lt;/sup&gt;</td>
<td>19.28±1.334&lt;sup&gt;b&lt;/sup&gt;</td>
<td>36.07±1.36&lt;sup&gt;d&lt;/sup&gt;</td>
<td>29.33±1.09&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Values are given as mean ± SE from eight rats in each group. Values of four experimental groups not sharing a common superscript letter differ significantly at P<0.05 (Duncan’s multiple range test).
Figure 6: Percent change in the Lipid peroxidation, Protein glycation, activities of Sorbitol dehydrogenase and Xanthine oxidase in the brain of D and DT rats compared to N rats.

![Graph showing percent change in LPO, Proteglycation, SD, and XO for N vs D and N vs DT.]

- LPO: 77 vs 26
- Proteglycation: 100 vs 32
- SD: 102 vs 10
- XO: 50 vs 20

N Vs D
N Vs DT
Where as studies of Kicic and Palmer indicated that STZ induced diabetes increased testicular SD gene expression with no alternation the gene expression of kidney and brain. Further, insulin treatment did not attenuate the enhanced testicular SD gene expression. Interestingly, *C. mukul* administration could attenuate significantly the activity of SD in the brain of DT group. Elevated brain GSH levels shown in DT and NT rats by *C. mukul* administration may also responsible for the decreased SD activity compared to corresponding controls. Increased intracellular GSH level was reported to be one of the inhibitory factors for SD activity (Bergmeyer et al., 1988). Studies of O,Brien et al., (1982) indicate that human brain SD was enhanced by flavonoid quercetin. This flavonoid derivative has been shown to inhibit AR in human brain and lens (Varma et al., 1975; Varma & Kinoshita, 1976). Aqueous extract of the plant *Scoparia dulcis*, which is rich in flavonoids and diterpenoids, was also reported to inhibit hepatic SD. Thus the present study provides an evidence for new molecular mechanism concerning with antidiabetogenic and neuroprotective efficiencies of *C. mukul* via its inhibitory action in the brain polyol pathway, a main contributing factor for diabetes induced brain damage.

**Xanthine oxidase:**

When neuronal cells are under oxidative stress, excessive ROS are produced that may induce neuronal death. The reactive oxygen species can lead to cell injury through cell membrane lipid destruction and can diffuse intracellularly and cause mitochondria enzyme damage and cleavage of DNA. Xanthine oxidase and its requisite substrate would be present in high concentrations in diabetic tissue and consequently would result in oxygen free radical generation. Xanthine oxidase has been proposed to be a major source of ROS in diabetes mellitus (Butler et al., 2000). Xanthine oxidase acts on xanthine and hypoxanthine with the resultant production of oxygen free radicals.

Elevation of XO activity in tissue especially in brain acts as a biomarker of oxidative stress. XO activity in the brain were studied in four experimental groups and presented in table 6 and figure 6. Diabetic rats showed a significant increase in
the activity of XO (50%) when compared to normal group. *C. mukul* treatment for 60 days in DT rats resulted in a significant decrease (18%) in the activity of XO in brain when compared to D group. But this change in XO activity in DT rats does not reached the normal values. Thus, *C. mukul* treatment for 60 days has given a partial recovery from enhanced activity of XO in the brain under STZ induced diabetic complications. However N rats treated with *C. mukul* showed a significant increase (21%) in XO activity compared to N rats.

The most significant observation of the present study was higher percent increase in XO activity in STZ diabetic animals. Oxygen-free radicals are generated by a variety of sources at the cellular level. An important source is XO which is formed from xanthine dehydrogenase (XD) either reversibly (via oxidation or blockage of its thiol groups) or irreversibly (via limited proteolysis) in pathological conditions (Hile and Nishino, 1995). In normal tissue, XD, which is the native form of the enzyme, catalyzes the conversion of hypoxanthine to xanthine and uric acid through the utilization of NAD⁺ as its electron acceptor. This reaction does not lead to the production of oxygen-free radicals or the generation of hydrogen peroxide (H₂O₂). However, XO is also able to catalyze the reduction of oxygen in pathological states leading to the formation of superoxide (O₂⁻) and H₂O₂ where the reaction is considered to play a crucial role in the proposed mechanism of oxidative injury (Corte and Stirpe, 1972). Enhancement of ROS has been reported to elicit translocation of cytosolic bax to mitochondria and to activate bax to induce the release cytochrome c, produced via the highly activated cytochrome oxidase reaction, shown in the rat brain, from mitochondria. Then, cytochrome c could stimulate (Cai and Jones, 1998) caspases and leading to apoptotic cell death (Choi et al., 2003), the decreased DNA and RNA content of brain tissue of STZ diabetic rats indicates enhanced DNA and RNA damage under diabetic conditions leading to increased catabolism of purine nucleotides. The enhanced activity of XO activity is a matter of concern since XO is recognized as one of the extra-mitochondrial sites of super oxide (O₂⁻) generation. Superoxide is generated through increased XO seen in diabetic mice, and may be involved in the pathogenesis of diabetic complications.
(Matsumoto et al., 2003). It has been shown that an important source of increased ROS in the STZ-induced diabetic rat is XO (Parks et al., 1988). Additionally, XO has been implicated in free radical production during diabetes based on the observed decrease in oxidative stress in diabetic patients during treatment with the XO inhibitors such as allopurinol (Desco et al., 2002). \( \text{O}_2^- \) generated extra mitochondrially should be eliminated in the first instance by SOD of the post mitochondrial fraction. The enhanced XO activity in further compounded by observed decrease in SOD activity in D rats.

The increased XO activity of diabetic rats compared to normal rats indicates that the diabetic state resulted in hyper-induction of XO activity due to the presence of DNA damage or oxidation in D group by STZ induced oxidative stress. Our reports of increase in extent of XO was supported by earlier studies in rat brain (Ozkan et al., 2006 & 2007), suggesting that the increase in extent of XO may play an important role in contributing free radical mediated damage and development of diabetic complications in brain. Treatment to NIDDM patients the XO inhibitor allopurinol reduces the level of oxidized lipids in plasma and improves the blood flow. Experimental data suggests that the role of XO is tissue dependent (Valko et al., 2007). The observed significant increase in XO of DT group compared to D group indicate that C. mukul treatment have provided a protection against free radical mediated DNA damage and further development of diabetic complications in brain.

**Antioxidant system:**

In the present study the observed enhanced oxidative stress markers in the brain of STZ diabetic rats indicate enhanced oxidative stress in the brain of diabetic rats. C. mukul treatment for 60 days has decreased the intensity of oxidative stress markers in diabetic treated rats. Further to understand the protective role of C. mukul administration against diabetic induced oxidative stress in the brain, the antioxidant status-GSH and the activity of antioxidant enzymes are analyzed in the four experimental groups.
'Under normal conditions the generation of free radicals or ROS in the brain, as in other tissue is maintained at extremely low levels. Under physiological conditions, a wide range of antioxidants defense protect the body against the adverse effects of free radical production in vivo. Antioxidants can interfere with the oxidation process by reacting with free radical, chelating, and catalytic metals and also by acting as oxygen scavengers (Buyukokuroglu et al., 2001). However, antioxidant supplements or foods containing antioxidants may be used to help the human body reduce oxidative damage. The most commonly used antioxidants are BHA, BHT, propyl gallate and terta-butyl-hydroquinone (Gulcin et al., 2003). However, they have been suspected of being responsible for liver damage and carcinogenesis in laboratory animals. Therefore, the development and utilization of more effective antioxidants are desired.

In diabetes, oxidative stress has been found to be mainly due to an increased production of oxygen free radicals and a sharp reduction of antioxidant defenses. Hyperglycemia the main symptom of diabetes not only increases the production of ROS but also affect antioxidant reactions catalyzed by ROS scavenging enzymes. Hence therapeutic agents with both hypoglycemic and anti-oxidative properties would be useful as anti-diabetic agents. The present study was undertaken to examine the possible antioxidant activity of C. mukul treatment in STZ induced diabetic rats.

The data presented in table 7 and figure 7 revealed the brain GSH content and the activity of GSH independent antioxidant enzymes i.e. SOD and CAT and GSH dependent enzymes i.e. GR, GPx, and GST.

**Glutathione and Glutathione reductase:**

STZ induced diabetic rats showed a significant decrease in GSH content (38 %) with significantly decreased activity of GR (24 %) in brain when compared with normal group. C. mukul treatment for 60 days in DT rats results in a significantly increase in GSH content (20 %) and activity of GR (25 %) in brain when compared to D group.
But this significant increase in DT rats did not reach to normal values and showed a still significantly lowered GSH content (25%) and activity of GR (5%) compared to normal rats. Thus *C. mukul* treatment for 60 days has given a partial recovery in GSH content and activity of GR in the brain against STZ induced alterations. However N rats treated with *C. mukul* have showed a significant increase in GR content (5%) and with no alterations in GSH activity when compared to N rats.

Glutathione is the most abundant intracellular antioxidant, the dysregulation of which is widely implicated in the disease states. There is *in vitro* and clinical evidence that abnormal glutathione status is involved in β-cell dysfunction and in the pathogenesis of long-term complications of diabetes. Studies in the role of oxidative stress in diabetes rely heavily on the ability to measure glutathione. It is found ubiquitously in animal cells present at milli molar concentrations predominantly in the reduced state. Some 20% or intracellular glutathione is located in the mitochondrial where it helps to protect against ROS produced as byproducts of electron transport chain. Glutathione plays an important role in detoxification and it offers a nucleophillic thiol which is important for detoxification of electrophillic metabolites and metabolically produced oxidized agents. Glutathione is essential for maintaining the reducing capacity of cells, the loss of capacity may cause the cell to undergo apoptosis.

Glutathione is abundant in dietary sources with fruits and vegetables contributing to 50% of intake, though most of the body’s requirements are synthesized by denovo from its constituent amino acids. It is predominantly synthesized in hepatocytes from which it is exported to peripheral tissues (Deneke and Fanburg, 1989). Glutathione has a variety of crucial physiological roles. First and foremost, it is the central member of a complex antioxidant system protecting the cell from oxidative stress (Pompella et al., 2003). It is a cofactor for GPx which is defense mechanism against peroxides, preventing the accumulation of ROS and cellular injury. GSSG is formed as a product of detoxification. GSH is regenerated by the action of GR, an NADPH dependent enzyme; thus normally about 99% of intracellular exits in the reduced from (GSH). GSH is also a cofactor for the enzyme
dehydroascorbate reductase, which recycles dehydroascorbate back to reduced ascorbic acid (Vit C) and therefore maintains Vit C and E in their reduced forms by the reversible oxidation of its sulfhydral groups (Reed, 1990). Secondly, GSH serves as a cofactor in conjugation reactions mediated by GST which protect macromolecules against toxic xenobiotics such as drugs and carcinogens. The adducts formed are then actively secreted from the cell. Whilst this is beneficial in ridding the cell of toxins, it depletes the cellular GSH pool. Thirdly, GSH modulates the redox status of thiol groups in signaling proteins, through which it can influence a variety of cell functions like transcription, gene expression, cell proliferation, and programmed cell death. GSH itself is known to translocate into the nucleus and may participate directly in the regulation of gene transcription. Glutathione reductase catalyses the reduction of GSSG to GSH and maintains the intracellular pool of GSH. Glutathione reductase is highly active in kidney, liver, and other tissues and it regenerates GSH by thiol transfer reactions. This is a flavor enzyme and contains a cysteine moiety that under goes reduction and oxidation during catalytic cycle.

Thus depleted GSH content in diabetic rats might have influenced all these important functions of GSH. The significant decrease in GSH level in brain during diabetes is probably due to increased production of free radicals or increased utilization of GSH or decreased denovo synthesis and/or decreased regeneration of GSH due to decreased activity of GR by oxidative stress in diabetes. The high flux of glucose through the activated polyol pathway in diabetic brain may consume NADPH, which results in decreased level of reduced GSH, and consequent altered in the activities of all GSH-dependent enzymes like GR; GPx and GST. Our reports of decreased GSH & GR content in diabetic rats was supported by earlier studies (Montilla et al., 2005, Leelavinothan et al., 2007, Ibrahim, 2008) suggesting that the decreased GSH are sensitive and potentially relevant indicators of neurotoxicity and plays a role in the development of diabetic complications in brain. Similar reports were observed in effect of alcohol consumption in brain (Das et al., 2007) and also in diabetic heart (Gumieniczek et al., 2002).
Moreover, the brain mitochondria of diabetic rats show consistently reduced ATP levels, as a result of the prominent changes in activity of the mitochondrial respiratory chain enzymes in such rats. ATP content is closely related to denovo GSH synthesis and maintenance of membrane potential; thus, a reduction in ATP would produce an irreversible drop in membrane potential. The mitochondrial pool of GSH is considered vital for cell survival: after severe mitochondrial GSH depletion, cell death occurs; this, however, is delayed by antioxidant treatment (Han et al., 1997, 2003). The high degree of oxidative stress in brain and renal tissue coincides with biochemical signs typical of diabetes, such as high levels of glycemia as well as decreased plasma insulin levels. In the present study, DT group showed a partial recovery in GSH and GR levels when compared to normal rats indicating the protective role of C. mukul against diabetic induced oxidative stress. Interest has developed in the potential therapeutic modifications of glutathione status in the treatment of diabetes. Most of them contain a wide scale of antioxidants with potent scavenging activity for ROS.

**Glutathione peroxidase:**

Glutathione peroxidases (GSH-Px) are selenoenzymes which catalyses the reduction of hydrogen peroxide to water whereas the organic hydrogen peroxides are reduced to alcohol at the expense of GSH. Glutathione peroxidase resides in the cytosol and mitochondrial matrix. It acts as an enzyme protecting hemoglobin from oxidative destruction by H\(_2\)O\(_2\) and also acts as a "contraction cofactor" of mitochondria i.e. as a compound preventing loss of contractibility of mitochondria under special conditions. It catalyses the reduction of H\(_2\)O\(_2\) and organic hydroperoxides including those derived from saturated lipids to alcohol. It protects the biomembranes from oxidative attack. It prevents lipid peroxidation by scavenging H\(_2\)O\(_2\) and slowing down H\(_2\)O\(_2\) dependent free radical attack on the lipids. Non-selenium dependent glutathione peroxidase is also found in kidney, testis, adrenal, brain and fat in addition to the selenium glutathione peroxidase.
Diabetic rats showed a significant decrease in activity of GPx (23 %) when compared with normal group. Treatment with C. mukul for 60 days in DT rats results in significant increase in the activity of GPx (12 %) in brain when compared to D group. But this significant increase in DT rats does not reached to normal values and showed a still significantly lesser in GPx activity (15 %) than N rats. Thus C. mukul treatment for 60 days has given a partial recovery in GPx activity in the brain against STZ induced alterations. However N rats treated with C. mukul has showed a partial recovery in GPx activity when compared to normal rats.

Our reports of decreased GPx in diabetic rat brain was supported by earlier studies (Makar et al., 1995; Pedro Montilla et al., 2005, Leelavinothan et al., 2007, Ibrahim, 2008) and similar reports were observed in effect of alcohol consumption in brain (Das et al., 2005) and in chronic stress induced changes in rat brain (Bhattacharya et al., 2000) suggesting that the decreased activity of GPx indicated accumulation of peroxides and causes effect to biomolecules, and further correlates with the levels of GSH & GR. Reduced activity of GPx may result from radical inactivation and glycation of the enzyme (Hodgson and Fridovich, 1975). The decreased activity GR resulted in decreased production of GSH, which is essential for GPx activity for detoxification of peroxides. Glucose-6-phosphate DH produces NADPH which is a substrate for regenerating GSH from its oxidized from GSSG.

**Glutathione-S-transferase:**

GST is multifunctional protein found in many tissues and shows a broad specificity for organic hydroperoxides but not for H₂O₂ (Bruce et al., 1982). GST is thought to play a physiological role in initiating the detoxification of potential alkylating agents (Booth et al., 1961; Boyland and Chasseaud, 1969; Wood, 1970) including pharmacologically active compounds. This enzyme catalyses the reaction of such compounds with the –SH group of GSH, thereby neutralizing their electrophillic sites and rendering the products more water-soluble. GST have important role in the CNS, including a line of defense in astrocytes against potentially toxic xenobiotics and in protecting the myelin sheath (Cooper and Meiser, 1993).
Diabetic rats showed a significant increase in GST activity (45%) when compared with N rats. Treatment with *C. mukul* for 60 days in DT rats resulted in significant decrease (21%) in the GST activity in brain when compared to D group. But this significant increase in DT rats does not reach to normal values and showed a still significantly higher GST activity (14%) than N rats. Thus *C. mukul* treatment for 60 days has given a partial recovery in GST activity in the brain against STZ induced depletion. However, N rats treated with *C. mukul* have showed no significant change in GST activity when compared to N rats.

Our reports of increased GST in diabetic rats was supported by earlier studies (Bharadwaj et al., 2008) suggesting that the increased activity of GST in D group indicates enhanced utilization of GSH for detoxification of toxic products. The enhanced activity of GST may result in the involvement of deleterious oxidative changes due to accumulation of toxic products and acts as an indicator of neurotoxicity.

**Superoxide dismutase and Catalase:**
The GSH independent antioxidant enzymes in brain are superoxide dismutase (SOD) and Catalase (CAT).

The primary catalytic cellular defense that protects cells and tissues against potentially destructive reactions of O$_2^*$ and their derivatives is the Cu/Zn-SOD. SOD scavenges the superoxide anion to form H$_2$O$_2$, thereby diminishing the toxic effects caused by this radical. Wohaieb and Godin (1987) suggested that the reactive oxygen free radicals could inactivate and reduce the brain SOD activity. Recent studies demonstrated that a hyperglycemia-induced process of overproduction of O$_2^*$ by the mitochondrial electron-transport chain appear to be the initial and key event in the activation of all other pathways involved in the pathogenesis of diabetes-related formation of AGE's, activation of protein kinase C and nuclear factor kB.

CAT is heme protein which catalyses the reduction of hydrogen peroxide and protects the tissues from highly reactive hydroxyl radicals (Chance et al., 1952). H$_2$O$_2$ is considered a key metabolite because of its relative stability, its diffusion and its involvement in cell signaling cascade (Fridovich, 1995; Khan and Wilson, 1995; Pantopoulso et al., 1997; Sujuki et al., 1997). CAT or GPx was the primary enzyme in
the removal of \( \mathrm{H}_2\mathrm{O}_2 \) (Scott et al., 1991), CAT decomposes \( \mathrm{H}_2\mathrm{O}_2 \) without generation of free radicals by minimizing one electron transfer (Sebastain et al., 1997 & Corroehr et al., 1986), whereas GPx needs GSH for decomposition. CAT resulted in protection against \( \mathrm{H}_2\mathrm{O}_2 \) mediated LPO. The \( \mathrm{O}_2^- \) has known to inactivate CAT.

STZ induced diabetic rats showed a significant decrease in the activity of SOD (42 %) and significant increase in the activity of CAT (87 %) when compared to normal group. \textit{C. mukul} treatment for 60 days results in a significant increase in the activity of SOD (45 %) and significant decrease in the activity of CAT (21 %) in the brain when compared to D group. These alterations in SOD and CAT activities by \textit{C. mukul} treatment in DT rats restored these enzymes activities to normal level. The observed decreased SOD activity could result from inactivation by \( \mathrm{H}_2\mathrm{O}_2 \) or glycation of enzymes (Vlassara et al., 1983). Thus \textit{C. mukul} treatment for 60 days has given a better protection against STZ diabetic induced alterations in SOD and CAT activities. However N rats treated with \textit{C. mukul} has showed a significant change in SOD activity and significant decrease in the activity of SOD (25 %) and significant decrease in the activity of CAT (28 %) when compared to N rats.

In diabetes the autoxidation of glucose results in formation of which inactivates SOD (Fajans, 1995). Therefore the accumulation of may be one of the explanations for decreased activity of SOD in D rats. It has been observed that SOD can be rapidly induced in some conditions, when cells or organisms are exposed to oxidative stress (Michiels et al., 1994). The highest SOD activity was reported in RBCs at the onset of diabetes but subsequent decrease was observed in its activity with the duration of diabetes (Domingues et al., 1998). The lowered activity of SOD in D rats may be with longer disease duration, even though SOD induction may be seen at early stages.

Our reports of decreased SOD (Leelavinothan et al., 2004, Rizwan et al., 2005, Montilla et al., 2005, Ibrahim, 2008) and enhanced CAT (Makar et al., 1995) in diabetic rats was supported by earlier studies. Hodson and Fridovich (1975) and Pigleot et al (1990) have reported the partial inactivation of these enzymes activities
by hydroxyl radicals and hydrogen peroxide. The decreased activity of SOD could also be due to their decreased protein expression levels in the diabetic conditions as reported recently in liver (Sindhu et al., 2004).

The decreased activity of SOD in D rats may result in accumulation of O$_2$ and OH. The enhanced SOD activity in DT rats may protect CAT and GPx against inactivation by O$_2$ anions as these anions have been shown to inactivate CAT and GPx. The increase of CAT activity suggests a compensatory response to oxidative stress due to an increase in endogenous H$_2$O$_2$ production. The increased H$_2$O$_2$ is related to drastic loss of GSH which is required to activate GPx, the H$_2$O$_2$ detoxifying enzyme. The significant increase in CAT activity of brain of diabetic rats indicates a compensatory adaptive mechanism against oxidative stress to maintain homeostasis. However the observed enhancement in the oxidative markers i.e., LPO, Protein glycation, activities of XD and SD in the brain of D rats indicate that the enhanced CAT activity is not sufficient to overcome the oxidative stress.

However chronic or excessive increase in oxidant production can adversely affect the cellular physiology and functions. Further more, the physiological products of H$_2$O$_2$ promotes cell cycle progression in response to growth factors, excessive production of H$_2$O$_2$ does not accelerate DNA synthesis but, paradoxically may induce cell cycle arrest by oxidants induced degradation of cdc25c (Savitskey and Finkel, 2002). Chronic or excessive oxidative stress may interfere with normal functioning of tissues affected by diabetic hyperglycemia by increasing blood flow and disturbing hemodynamics in the retina and decreasing neural conductivity in peripheral nerves (Hounsam et al., 2001). These defects may in part be due to altered activities of signaling pathways that regulate differential functions of these tissues.

The restoration of SOD and CAT activities to normal levels by C. mukul in DT rats revealed the operation of efficient defense against the first line of oxidative stress in these animals. Commiphora species [Commiphora caudate (CC) and Commiphora varpubescens (CP)] are reported to possess efficient antioxidant activity
Table 7: Effect of Commiphora mukul on STZ induced diabetic alterations in rat brain GSH content, and activities of Antioxidant enzymes.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Normal</th>
<th>Normal treated</th>
<th>Diabetic</th>
<th>Diabetic treated</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glutathione (μg of GSH/g tissue)</td>
<td>7.41±0.58&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.99±0.34&lt;sup&gt;c,a&lt;/sup&gt;</td>
<td>4.58±0.70&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.52±0.34&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Glutathione reductase (μmoles of NADPH oxidized/min/mg protein)</td>
<td>31.41±1.65&lt;sup&gt;a&lt;/sup&gt;</td>
<td>32.88±1.17&lt;sup&gt;a&lt;/sup&gt;</td>
<td>23.88±0.75&lt;sup&gt;b&lt;/sup&gt;</td>
<td>29.94±0.66&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Glutathione peroxidase (μg of GSH consumed/min/mg protein)</td>
<td>12.27±0.29&lt;sup&gt;c&lt;/sup&gt;</td>
<td>12.10±0.33&lt;sup&gt;c&lt;/sup&gt;</td>
<td>9.28±0.36&lt;sup&gt;b&lt;/sup&gt;</td>
<td>10.45±0.25&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Glutathione-s-transferase (μmoles of GSH-CDNB conjugate formed/min/mg protein)</td>
<td>0.150±0.007&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.158±0.005&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.218±0.009&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.172±0.009&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Superoxide dismutase (U/min/mg protein)</td>
<td>8.97±0.07&lt;sup&gt;d&lt;/sup&gt;</td>
<td>8.70±0.23&lt;sup&gt;b&lt;/sup&gt;</td>
<td>5.19±0.13&lt;sup&gt;a&lt;/sup&gt;</td>
<td>7.53±0.19&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Catalase (mmoles of H&lt;sub&gt;2&lt;/sub&gt;O&lt;sub&gt;2&lt;/sub&gt; decomposed/min/mg protein)</td>
<td>2.62±0.24&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1.88±0.14&lt;sup&gt;d&lt;/sup&gt;</td>
<td>4.91±0.20&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.86±0.19&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
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Values are given as mean ± SE from eight rats in each group. Values of experimental groups not sharing a common superscript letter differ significantly at P<0.05 (Duncan’s multiple range test).
Figure 7: Percent change in the GSH content, activities of Catalase, glutathione peroxidase, glutathione reductase, glutathione-s-transferase, superoxide dismutase and catalase in the brain of D and DT rats compared to N rats.
(Deepa et al., 2009). Studying Ramesh Chander et al., (2002 & 2003) revealed the cardio protective and antioxidant activities of synthetic guggulsterone and guggulsterone obtained from gum resin of C. mukul, due to inhibiting the generation of reactive oxygen species (ROS).

In conclusion our results demonstrated the STZ diabetes induces a deleterious brain damage which is in part, amenable to attenuation by C. mukul treatment. The neuro protective efficiency of C. mukul can be correlated directly to its improving effect on hyperglycemia, hypoinsulinemia and its anti-oxidant reduced scavenging properties. Moreover the attenuating effect of C. mukul on the active polyol pathway and protein glycation shown in diabetic rat brain may also play a role in its neuro protective efficiency. Many other plants like Scoparia dulcis (Leelavinothan and Muniappan, 2004), Herparidin, a citrus bioflavonoid (Ibrahim, 2008), Trigonella (Siddiqui et al., 2005), and supplementation of Nicotinamidie (Ibrahim and Rizk, 2008) and alpha-lipoic acid (James et al., 1999) also possesses potent antioxidant activity and exerts a beneficial protective effect on the brain of STZ induced diabetes.

Thus, the present study showed that C. mukul resin possesses potent antioxidant activity, which may be directly or indirectly responsible for its hypoglycemic and β-cell regeneration capacity. Further studies are in progress in identifying the active components in C. mukul and their role in controlling diabetes.