• Chronic exposure of fluoridated ground-water causes health problem not only to human beings, but also to domestic animals in the form of fluorosis. Regardless of the circumstantial evidences on the involvement of fluoride in the etiology and pathogenesis of fluorosis, several lines of evidences including the observations of this study strongly advocate the involvement of fluoride as aggravating or risk factor in the progression and pathogenesis of diabetes.

• The effect of high plasma fluoride levels appeared to be transient, while, upon chronic exposures affected glucose metabolism when plasma fluoride concentrations exceeded 0.1 mg/l. The in vivo effect was appeared to be due to inhibition of insulin secretion rather than insulin-receptor interaction. In fluoride endemic zones, diabetic individuals were shown to consume and retain excess fluoride than healthy individuals, due to increased water intake and decreased renal clearance. Therefore it was hypothesized to determine the toxic assessments in diabetic population as their kidney function puts them at higher risk for the potentially detrimental effects of long-term fluoride exposure. In addition prophylactic treatments by phytoextracts were hypothesized to be beneficial to address the fluoride toxicity in diabetics, thereby this study was conducted.

• In this study, diabetic mice (STZ induced) were exposed to 270ppm fluoridated water (600ppm NaF) ad libitum for 30-days and later supplemented with Ginseng (Panax Ginseng) and Banaba (Lagerstroemia speciosa) individually and together in variable doses (50, 150 and 250 mg/kg b.w dose) for 15-days to check their ameliorative efficacy in mitigating the actions of fluoride in diabetics.

• The differential toxicity observed in different functional tissues may be related to the toxico-dynamics of fluoride, although physiological and biochemical constraints associated with diabetes appears to be directly related to influence the susceptibility of different organ system(s) to fluoride toxicity. Kidney and brain tissues were found to be more affected due to fluoride accumulation and showed different pattern of changes. Differences
in the retention of fluoride in soft tissues viz., liver, kidney, testis and discrete brain tissues like cortex, cerebellum and hippocampus could be anticipated on the basis of their physiological differences of the functional system.

- Phytoextracts viz., ginseng and banaba, having free radical scavenging abilities showed significant anti-hyperglycemic effect and were helpful to recover the altered glycosylated hemoglobin, hepatic glycogen, serum urea, creatinine and tissue fluoride levels. Among the dose regimens tested, 50 and 250mg/kg b.w doses of GE and BLE supplementation showed insignificant changes in biochemical markers studied throughout the experiment. This could be due to the antagonistic behavior of the biomolecules present in the phytoextracts which offered hindrance to the protective role of the plant extracts. Thereafter, 15-day length of GE and BLE exposure showed maximum recovery.

- High fluoride uptake in mice was found to be diabetogenic and further aggravated the complications in STZ induced diabetic mice by altering the enzymes connected to carbohydrate metabolism. Serum thyroid hormones were assessed by using Bayer Centaue autoanalyser (Thyrocare laboratories, Bombay) and results obtained showed that FT$_3$ was decreased upon fluoride exposure in diabetic mice. GE and BLE exposure individually or in combination at a dose of 150mg/kgbw/day for 15-days exhibited protective effects by normalising the carbohydrate metabolising enzymes viz. HK, PGI, PK, G-6-Pase, LDH, G-6-PDH and TCA cycle enzymes.

- In the present study, fluoride exposure caused a significant accumulation of fluoride in liver, kidney and discrete brain regions resulting in the generation of excessive free radicals and thereby consequential alterations in MDA levels along with xanthine oxidase, catalase, superoxide dismutase and glutathione system viz., glutathione peroxidase, glutathione transferase, and total reduced glutathione levels examined. Co-supplementation of GE and BLE at a dose of 150mg/kgbw was found effective for the protection against oxidative stress and in bringing attenuation of lipid peroxidation, thereby normalizing the homeostatic antioxidant system.
• The assessments were made on the similarities and dissimilarities between fluoride-induced toxicity in STZ induced diabetic mice and the results confirm an association between fluoride toxicity and free radical mediated oxidative stress that already existed in diabetics. Among the other mechanisms, increased oxidative stress, protein oxidation, aminotransferase activities (ALT and AST) and decreased levels of protein thiols, total proteins, lipid profile indices and DNA fragmentation as found in the present study are suggestive of macromolecular alterations in functional tissues studied viz., liver, kidney and discrete brain regions. The findings of this study divulge that GE and BLE co-supplementation at a dose of 150mg/kgbw/day for 15-days elicited the beneficial efficacy by normalising the serum lipid levels, protein oxidation and amino acid metabolism observed in fluoride toxicated diabetic mice.

• Prophylactic supplementation of phytoextracts viz., GE and BLE were found to be effective in decreasing the extent of oxidative stress and normalizing the antioxidant enzymes examined. Similar trends of amelioration was observed with regard to the parameters studied viz., PCO, -SH groups, transaminases, carbohydrate metabolizing enzymes. From the findings it can be said that antioxidant supplementation plays an important role in the prevention of fluoride induced oxidative stress in diabetics and bolstering the cellular antioxidant defense system.

• The histological impairments observed in liver, kidney and discrete brain regions in toxicated mice corroborate with the enhanced oxidative insult. Supplementation of GE and BLE at dose 150mg/kgbw showed almost normal hepatocytes and kupffer cells thereby reducing hepatic alterations. In renal tissues, normal glomerular size, GBM and mesangial matrix and further absence of KW nodules and tubular dilations were noticed upon supplementation of GE and BLE at a dose of 150mg/kgbw. In GE and BLE supplemented group, appreciable improvement was observed in the organization of cerebral layers and reducing the vacuolar spaces among the neuronal cells. GE and BLE supplementation proved advantageous as the molecular, granular and Purkinje cell layers were found to be intact and
significant improvement was observed in distribution and structural abnormality of hippocampal neurons. In testis, changes were encountered in the germ cells due to their low antioxidant ability and more polyunsaturated fatty acid content, while the supplementation of GE and BLE resulted in regenerating the necrotic germinal epithelium, spermatogenic and interstitial cells. In bone marrow cells phyto extracts paved way to alleviate the micronuclei formation and chromosomal damages due to fluoride exposure, thus enlightening the protective role of ginseng and banaba co-exposure against genotoxicity.

- The findings of the study divulge that both phytoextracts exhibits significant fertility potential by normalizing the diminished sperm density, viability, motility and the altered testicular biochemical variables of fluoride toxicated diabetic mice, thereby, it can be postulated that GE and BLE co-supplementation at a dose of 150mg/kgbw/day for 15-days elicit the beneficial efficacy by normalizing reproductive functions in fluoride toxicated diabetic mice.

- In the present study, phytoextracts ginseng and banaba was found to exhibit their potential antioxidant ability in curbing oxidative stress and normalizing the mitochondrial oxidative phosphorylation. The pharmacological actions of ginseng extract are attributed to ginsenosides, which are tetracyclic triterpenoid saponin glycosides of the saponins family of steroids and are known to possess the ability to scavenge free radicals. Similarly, the pentacyclic tri-terpenoids of corosolic acid of banaba extract is shown to be unique in its antioxidant and free radical scavenging activity. In addition, the presence of carboxyl hydrogen in corosolic acid may be responsible for its free radical scavenging activity. Consistent with earlier reports on the antioxidative abilities of these phytoextracts, the present study findings also confer the antioxidant ability of above extracts and found a reduction in oxidative stress thereby reduced toxic burden and related impairments in fluoride toxicated diabetic mice connected to the functional systems. It can be postulated from the results that the synergy among the extracts of GE and BLE enhances the tri-terpenoid bioavailability or efficacy by forming a
chelate complex which could be responsible for the synergistic actions, thereby removes free toxic radicals from the cells and in turn inhibits oxidative damage. Thereby the antioxidant efficacy of GE and BLE might have reduced the oxidative stress and reduced the impairments in general and toxic burden in particular. The co-supplementation of GE and BLE at a dose of 150mg/kgbw (for 15days), therefore, could simultaneously improve the overall ability of scavenging free radicals from the cellular components, offered a synergistic action to provide protection against fluoride toxicity in diabetics.

- In conclusion, the above results support the hypothesis that diabetic population is highly vulnerable to the toxic effects of fluoride and its effects were noticed on the functioning of hepatic, renal and neuronal systems, raising the possibility that higher doses of fluoride may have a greater effect in diabetics. An area of research in this context is the investigation as to whether there is a synergism of fluoride-induced oxidative stress with the already increased levels of oxidative stress and ROS production that are evident in diabetics. An environment of oxidative stress in diabetic population compounded with further insult from fluoride toxicity may lead to more detrimental consequences, which may inevitably affect the ability of functional organs. It therefore is important not only to further elucidate the effects of fluoride on performance and cellular functioning, but also to develop practical counter measures, such as prophylactic antioxidant therapy, suitable dose, their adverse effects etc. These interventions, as well as others, may protect against the deleterious effects associated with fluoride toxicity and general health of population. Therefore, this study recommends that intake of ginseng and banaba will be useful for diabetic patients residing in fluoride endemic zones. However further studies are warranted on the dose regimens required for the human population.