SUMMARY AND CONCLUSION

Type 2 diabetes mellitus (DM) is the most common form of diabetes and is usually caused by life-style factors and also related to insufficient insulin production and resistance of target tissues to insulin. Hyperglycemia can be handled initially with oral synthetic agents and insulin therapy. However, these synthetic agents produce some serious side effects and are relatively expensive for developing countries. One of the current trends in the management of diabetes is to decrease post-prandial hyperglycemia. This can be achieved through the inhibition of carbohydrate hydrolyzing enzymes such as alpha-amylase and alpha-glucosidase. Alpha-amylase and glucosidase inhibitors from medicinal plants can be used for developing new target drugs for the treatment of diabetes, obesity and hyperlipidemia.

Study on the plants and plant parts for alpha-amylase and alpha-glucosidase inhibition as well as antioxidant potential becomes important in choosing good candidature plants for preclinical studies on antidiabetic property. Hence the present study on “Isolation and Characterization of Antidiabetic and Antioxidant Principles from Saraca indica and Polyalthia longifolia bark” was performed with the following objectives:

Objectives of the Study

1. Analysis of phytochemicals and inorganic elements in the bark of Saraca indica and Polyalthia longifolia
2. Assessment of alpha-amylase inhibitory activity and antioxidant potential of various solvent extracts of the barks of the two selected medicinal plants
3. Investigation of in vitro alpha-amylase and alpha-glucosidase inhibitory activities of the potent extracts and mechanism of inhibition
4. Evaluation of the antidiabetic and antioxidant properties of the potent extracts in diabetic rats
5. Isolation and characterization of selected active components with pharmacological potential from the bark of *Saraca indica* and *Polyalthia longifolia*

The study was conducted in four phases and the results are summarized below.

In **Phase I**, screening of phytochemicals in *Saraca indica* and *Polyalthia longifolia* bark revealed the presence of carbohydrates, phenols, tannins, flavonoids, terpenoids, steroids, except saponins and alkaloids. The barks of the chosen medicinal plants were found to contain inorganic elements that are essential for the management of diabetes mellitus.

**Phase II** involved the studies on alpha-amylase, alpha-glucosidase inhibition and antioxidant potential of *Saraca indica* and *Polyalthia longifolia* bark. Among the various solvents used for the extraction of *Saraca indica* and *Polyalthia longifolia* barks, highest percentage yield of extract was obtained using ethanol. Total antioxidant potential and percentage inhibition of alpha-amylase and alpha glucosidase were found to be highest with ethanolic extract of both the plants denoted as SIE and PLE compared to other solvent extracts. Ethanolic and hexane extracts exhibited increased free radicals scavenging activity and inhibition of *in vitro* lipid peroxidation. Results of Dixon plot, Cornish and Bowden plot revealed that the mechanism of alpha-amylase inhibition was 'mixed' by SIE and 'competitive' by PLE whereas the mechanism of alpha-glucosidase inhibition was 'competitive' by both SIE and PLE. MTT assay revealed that the extracts SIE and PLE at higher concentration (1 mg/mL) were moderately cytotoxic in Vero and L6 cell-lines.

In **Phase III**, the antidiabetic and antioxidant properties of the ethanolic extracts of *Saraca indica* and *Polyalthia longifolia* bark in Niacinamide-Streptozotocin (NIA-STZ) induced diabetic rats were evaluated. Acute toxicity studies revealed that the extracts were non-toxic upto a higher dose of 2000 mg/kg b.w. Diabetic rats treated with SIE 400 mg/kg b.w and PLE 400 mg/kg b.w. showed significant decrease (p<0.001) in the levels of blood glucose monitored at weekly interval. Total cholesterol, triglyceride, low density lipoprotein (LDL), very low density lipoprotein (VLDL), urea, uric acid, creatinine levels, enzyme markers of hepatic injury namely SGOT, SGPT, ALP and marker of cardiac injury namely LDH, glucose-6-phosphatase, thio-barbituric acid reactive substance
(TBARS) levels were decreased, compared to NIA-STZ induced diabetic rats. Body weight, hemoglobin, red blood cells, white blood cells and their functional indices, total protein, high density lipoprotein (HDL), activities of hepatic enzymic-antioxidants namely CAT, SOD, GPx, GR and non-enzymic antioxidants including vitamin C, vitamin E, GSH, hepatic enzymes of carbohydrate metabolism namely hexokinase, glucose-6-phosphate dehydrogenase, succinate dehydrogenase and malate dehydrogenase activities were increased compared to NIA-STZ induced diabetic rats. The effect was on par with that of diabetic rats treated with the oral diabetic drug, glibenclamide.

The results of histopathological analysis revealed that damage caused in the liver, pancreas and kidney in the diabetic rats were reversed on standard drug glibenclamide / SIE / PLE extracts treatment. This indicated the regeneration of tissues. The high dose (400 mg/kg b.w.) of SIE and PLE treated diabetic rats showed protection that was similar to the action of glibenclamide.

**Phase IV** involved isolation and characterization of selected antidiabetic and antioxidant principles from ethanolic extracts of the barks of *Saraca indica* and *Polyalthia longifolia*. Twelve compounds were identified under the class of phenols, lignin glycosides and terpenoids in the HPLC fractions by using QTOF-MS, H\(^1\)NMR and FT-IR.

The H\(^1\)NMR and FT-IR spectral analysis confirmed three compounds in the ethanolic extract of the bark of *Saraca indica* and the bark of *Polyalthia longifolia*. It could be suggested that these compounds may be responsible for their antidiabetic and antioxidant potential. The ethanolic extracts of the barks of *Saraca indica* and *Polyalthia longifolia* with antidiabetic potential could be exploited in the formulation of drug. The barks of both the plants are equally effective in terms of antioxidant and antidiabetic potential, but they are expected to act through different mechanisms.
Future recommendations

1. The extracts can be tested in various animal models of diabetes

2. Synthetic compounds derived by modifications of the identified compounds may be produced and tested *in silico* on various targets to obtain antidiabetic drugs

3. Cell culture protocol could be developed for large scale development of the compounds