Abstract

Derivatives of plant origin have long been known to possess biological activity. Many traditional cultures remain mostly dependent on plants for their food and medicine, and often consider them both in the same context. The putative efficacy of medicinal herbs relies on empirical or anecdotal data and tradition of use, which frequently cannot satisfy the requirements of evidence-based medicine. Thus, establishing the pharmacological basis for the actions of medicinal herbs is a constant challenge. Even when scientific data are available, it is often difficult to determine whether a consistent herbal preparation was used in various studies. Often, several herbs are mixed to achieve a pharmacological effect, thereby making it extremely difficult to attribute the effect to a particular herb. Recognition of these confounding factors by researchers will aid in generating useful data that can lead to a better understanding of the proper role of medicinal herbs in health.

Free radicals have been implicated in the causation of several diseases such as liver cirrhosis, atherosclerosis, cancer, diabetes, etc and compounds that can scavenge free radicals have great potential in ameliorating these disease processes (Wilson, 1988). Natural antioxidants strengthen the endogenous antioxidant defenses from Reactive Oxygen Species (ROS) and restore the optimal balance by neutralizing the reactive species. They are gaining immense importance by virtue of their critical role in disease prevention.

We have selected the leaves of *Lagerstroemia speciosa* and the bark of *Mangifera indica* for their biological screening. The leaves of *L. speciosa* (Lythraceae), a south east Asian tree more commonly known as Banaba, have been traditionally consumed in various forms by Philippinos for treatment of diabetes and kidney related diseases. *M. indica* (Anacardiaceae) has large potential to treat various ailments among tribal communities inhabited in the remote regions of Indian subcontinent. Both the plants are the source of bioactive compounds with potential health-promoting activity. They are reported to have high amount of polyphenols and tannins in their extracts. Very few pharmacological studies had been reported so far in the selected extracts/crude isolates. In this thesis, the selected extracts and crude tannins and polyphenols were screened for their phytochemical, antioxidant and pharmacological screening including possible mechanism of action.

This study was conducted to document the new ethno-medico, physico-chemical and phytochemical information and traditional use of medicinal plants against

inflammatory, renal, gastrointestinal and liver, hyperglycaemic and hyperlipidemic disorders and thus to conserve the rapidly disappearing traditional knowledge system.

The physicochemical parameters and elemental analysis of the plant materials were carried out using flame photometry and atomic absorption spectrophotometer (AAS). Different fractions were collected by successive solvent extraction. The phytochemical tests and quantitative estimation of phytoconstituents were carried out in the selected plant part. HPTLC finger printing and HPLC of the polyphenols and tannins were done to identify the various polyphenolic compounds. The IR, Mass and NMR spectra were recorded and the compound is now being subjected to C^{13}–NMR study and the structural elucidation is now being attempted. The GC-MS of the blossom oil was carried out for both the plants and compounds were identified and compared with the library spectra (on-line) as well as with reference MS-spectral data.

In vitro antioxidant activity of successive solvent extracts were carried out and significant antioxidant activity was exhibited by these extracts. The protective effect of extracts, polyphenols and tannins of two plants on hydrogen peroxide induced haemolysis, lipid peroxidation and degradation of membrane proteins were also carried out. The oxidative haemolysis of rat erythrocytes by hydrogen peroxide was inhibited in a dose dependent manner by the extracts and polyphenols.

Acute toxicity studies of the selected solvent extracts were carried out using mice and LD_{50} concentration was determined. Our pharmacological screening was established in *in vitro* and *in vivo* anti-inflammatory activity of the total ethyl acetate and ethanol extracts of *L. speciosa* and *M. indica*. Carrageenan induced acute inflammation and formalin induced chronic paw edema in mice were studied. The total ethanol extracts has a significant anti-inflammatory effect against carrageenan and formalin induced paw edema in a dose dependent manner. Total ethanol extract of both plants could scavenge the superoxide anion, might inhibit the recruitment of PMNs and thereby reduce inflammation significantly. Ethyl acetate and alcohol extracts showed significant anti-nociceptive effects, which has central and peripheral anti-nociceptive activity, which may be partially mediated by opioid receptors as these receptors play an important role in pain sensation.

Diuretic activities of these plants were compared with two types of diuretics (furosemide and mannitol). The ethyl acetate fraction did not increase urinary excretion when compared with ethanol extracts.

The nephroprotective activity results showed the renal SOD, CAT, GPx and GSH level significantly reduced in the cisplatin treated group. The kidney marker like serum creatinine and urea were found to be decreased more significantly by the administration of ethanol extract of the plants. When compared with the two plants, the EtOAc extract of *L. speciosa* significantly prevents the toxicity of cisplatin where as *M. indica* showed maximum activity in EtOH extract treatment study. The results of the nephroprotective investigation indicate that ethanol extract of *M. indica* rendered significant protection against gentamicin induced nephrotoxicity in a dose dependent manner. Pretreatment of rats with *L. speciosa* and *M. indica* extract dose dependently inhibited the increased level of all hepatic marker enzymes in serum, indicating the liver protective activity of these plants.

Both the plant extracts exhibited significant anti ulcer activity in ethanol and cold stress induced models. The treated group can withstand the stress and reduced the formation of gastric acid induce lesions in the stomach.

The tissue with less developed antioxidant defense mechanism such as the heart is highly susceptible to injury by anthracycline-induced oxygen radicals. The treatment with polyphenols and tannin caused a significant restoration of the antioxidant enzymes such as CAT, SOD, GPx and G6PDH, where activities of these enzymes were increased in the heart tissues of DOX treated group as compared to normal group. Polyphenols and tannins have significantly restored the levels of above antioxidant enzymes towards normal, indicating the beneficial antioxidant potential of the above fractions. The biochemical changes support the histopathological changes, where DOX in chronic administration has produced its characteristic morphological changes in the myocardium.

The anti-diabetic potency of tannins and polyphenols using streptozotocin (STZ) induced diabetic model and compared their antidiabetic activity with their *in vivo* antioxidant potential for their ability to scavenge the formed free radical in diabetic rats. Long-term administration of tannins and polyphenols significantly decreased the serum glucose level and the polyphenols showed better activity than the tannins.

Disturbances in fatty acid metabolism, i.e. elevation of free fatty acids (FFAs), are regarded as one of the major determinants in the pathogenesis of insulin resistance, (Boden *et al.*, 2005) which is the charecteristic feature of type 2 diabetes mellitus and is frequently associated with obesity. In the liver, elevated FFA may contribute to hyperglycaemia by antagonizing the effects of insulin on endogenous glucose production.

Long-term treatment with tannins and polyphenols reduced the free fatty acid level significantly in our study.

Liver marker enzymes like ALP, ALT, AST and bilirubin were increased in diabetic control group and our treatments reverse the toxicity produced by streptozotocin (STZ). Liver glycogen level was significantly less in diabetic group and the excess amount of glucose is not stored in the liver. SOD, CAT and GSH were decreased significantly in the pancreas of diabetic rats. Whereas treatment with polyphenols and tannins, reversed the STZ induced free radical damage. Histopathological examination of kidney and pancreas showed the toxicity in cell morphology in control diabetic animals and it was reversed by the drug treatment. The morphology and beta cell count has been changed in the pancreatic islet by the long-term treatment with the plant constituents.

Glutamate dehydrogenase (GDH) is a homo hexameric mitochondrial matrix enzyme that catalyses the reversible oxidative deamination of glutamate to α-ketoglutarate plus free ammonia using either NAD or NADP as a co-factor (Colman, 1991). In diabetic condition, the increase in the Vmax of GDH observed during conversion of α-ketoglutarate to glutamate, indicates an increase in glutamate content in brain.

In the high fat diet (HFD) fed diabetic studies, both polyphenols and tannins reduced the serum glucose level. Administration tannins reduced the body weight significantly and may be due to the anti obese property of *L. speciosa*. A variety of derangements in metabolic and regulatory mechanisms, due to insulin deficiency are responsible for the observed accumulation of lipids (Rajalingam *et al.*, 1993). The impairment of insulin secretion results in the enhanced metabolism of lipids from the adipose tissue to the plasma.

The ^{14}C glucose uptake in the rat gastrocnemius muscle by *in vitro* model showed the presence of insulin the glucose uptake significantly increased when the normal animals were prior treated with tannins and polyphenols.

From the above all studies, it can be concluded that both the plant extracts and the isolated fractions possesses potent pharmacological activity based on its active constituents along with their antioxidant potential. Administration of the extracts and its isolates reduced the organ toxicity and restored the enzyme levels to normal. Both the plant phytoconstituents reduced the secondary complications of diabetes mellitus. In future, the study has to be extended with the combination of these two plants and its isolated fractions for their selected pharmacological studies and its mechanism of action.