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## *Introduction*

## 1.0 INTRODUCTION

Many medicinal plants, traditionally used for thousands of years, are present in a group of herbal preparations of the Indian traditional health care system (Ayurveda) and proposed for their interesting multilevel activities. Amongst the medicinal plants used in Ayurvedic preparations for their therapeutic action, some have been thoroughly investigated and some are still to be explored (Sushma *et al.*, 2013). The use of plants for therapeutic purposes is getting increasingly popular as they are believed to be beneficial and free of side effect. However, the rationale for the use of medicinal plants has twelled largely on long term clinical experience with little or no scientific data on their efficacy and safety (Mbiantcha *et al.*, 2013).

Medicinal plants are of great importance to the health of individuals and communities in general. The medicinal value of plants lies in some chemical substances that produce a definite physiological action on the human body. The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids and phenolic compounds (Mir *et al.*, 2013). Medicinal herbs are important sources for the enormous physiological activities in humans and therapeutic remedies of various ailments (Maobe *et al.*, 2013).

Plants produce a wide array of organic compounds, usually secondary metabolites, which in addition to imparting characteristic odour, flavour and pigment properties, sometimes exhibit antimicrobial action. The extraction and possible subsequent therapeutic application of these biologically active phytochemicals is not a recent development. Clinical trials are being carried out with many plant-derived antimicrobials for human use (Kamalakannan *et al.*, 2012).

The drug resistance of human and animal pathogens is one of the best documented cases of biological evolution and is a serious problem both in developed and developing countries. The daily consumption of more than one ton

of antibiotics in some countries has resulted in resistance to bacterial populations, thus causing a serious public health problem. In face of this scenario, the search for substances from natural sources, including plants, has been gaining importance in the pharmaceutical companies (Hossain *et al.*, 2012).

The increasing incidence of deep venous thrombosis and its closely related pulmonary embolism during the post-operative and post traumatic management of the patients has led the investigators to examine the aetiology and prevention of the procedure. Cardiovascular diseases remain the cause of mortality and morbidity, especially in the developed world. But even in developing countries the incidence of cardiovascular diseases is still increasing (Jesonbabu *et al.*, 2012). Thrombolytics are used to dissolve the fibrin of blood clots which are potentially life-threatening, especially those in the arteries of the heart and lungs. It is also used against the clots formed in shunts during kidney dialysis and multiple pulmonary emboli. All available thrombolytic agents still have significant shortcomings, including the need for large doses to be maximally effective, limited fibrin specificity and bleeding tendency. Due to the shortcomings of the available thrombolytic drugs, attempts are underway to develop improved recombinant variants of these drugs (Uddin *et al.*, 2013).

Heparin and Aspirin are only moderately efficient for acceleration of lysis and prevention of reocclusion, but are safe. More selective thrombin inhibitors and antiplatelet agents are more potent, but their safety remains to be confirmed. Continued investigation in this area will provide new insights and promote progress towards the development of the ideal thrombolytic therapy, characterized by maximized stable coronary arterial thrombolysis with minimal bleeding (Ansari *et al.*, 2012).

The free radicals are the culprit for not only in support of aging but also many age-related diseases. Free radical damage within cells has been linked to a range of disorders including cancer, arthritis, atherosclerosis, Alzheimer's disease, and diabetes. There has been some evidence to suggest that free radicals and some

reactive nitrogen species trigger and increase cell death mechanisms within the body such as apoptosis and in extreme cases necrosis. Targeted antioxidants may lead to better medicinal effects such as mitochondria-targeted ubiquinone, for example, may prevent damage to the liver caused by excessive alcohol. Some reviews suggest that antioxidant could reduce side effects and increase survival times (Ali *et al.*, 2013).

The human body has several mechanisms to counteract oxidative stress by producing antioxidants which are either naturally produced in body, or externally supplied through foods and /or supplements. Endogenous and exogenous antioxidants act as free radical scavengers and therefore can enhance the immune defense and lower the risk of cancer and degenerative diseases (Shinde *et al.*, 2012). Antioxidant nutrients have the ability to scavenge free radicals in the system and neutralize them before they do any damage to body cells. Most plants have protective biochemical functions of naturally occurring antioxidants in the cells. Many secondary compounds and enzymes of higher plants have been demonstrated with *in vitro* experiments to protect against oxidative damage by inhibiting or quenching free radicals and reactive oxygen species (Panda *et al.*, 2012).

The antioxidants play a vital role in delaying, intercepting or preventing oxidative reactions catalyzed by free radical. However, there have been concerns about synthetic antioxidants such as butylated hydroxy anisole (BHA) and butylated hydroxyl toluene (BHT) because of their possible activity as promoters of carcinogenesis. Hence, strong limitations have been placed on their use and there is a trend to replace them with naturally occurring antioxidants. Moreover, these synthetic antioxidants also show low solubility and moderate antioxidant activity. Therefore, search for natural antioxidant has greatly been increased in the recent scenario (Kumar *et al.*, 2012).

Toxicity studies play an important role in identification and isolation of new compounds from crude extracts. Bioactive compounds are often toxic to shrimp larvae (*Artemia salina*). Therefore, Brine shrimp lethality assay is in use to monitor

lethality of different chemicals to shrimp larvae (Solanki *et al.*, 2013). The crustacean *Artemia salina* Leach (brine shrimp) is an invertebrate that has been widely used for studies of ecotoxicology, as well as of general toxicology of chemicals and natural compounds. *A. salina* cysts are easily available commercially and inexpensive. Hence this assay may be useful in situations where rapidity and low cost make it practical to test large number of samples for preliminary toxicity screening (Otang *et al.*, 2013).

Docking is a method of molecular modeling, which predicts the preferred orientation of one molecule to a second when bound to each other to form a stable complex. Molecular docking can be defined as an optimization problem, which would describe the “best-fit” orientation of a ligand that binds to a particular protein of interest and is used to predict the structure of intermolecular complex formed between two or more molecules (Onkara *et al.*, 2013).

The concept of docking is important in the study of various properties associated with protein-ligand interactions such as binding energy, geometry complementarity, electron distribution, hydrogen bond donor acceptor properties, hydrophobicity and polarizability. Elucidation of ligand binding mechanisms is the necessary step to obtain more selective and potent drugs for this new potential target (Sindhu *et al.*, 2011).

Ligand molecules may act as an antagonist of receptor and are also used as drug targets. Several medicinal plants that have been used for a long time are the new source candidates of therapeutic molecules. Therefore, the medicinal plants and their compounds may act as ligands. These plant-derived ligands have shown their potentials for the receptor inhibition and they may be less toxic as they have been used by people for generations. Therefore, medicinal plants have enormous potential for mining the ligand molecules (Chakraborty *et al.*, 2012).

*Piper betle* Linn. (Piperaceae), a dioecious, perennial creeper, climbing by many short adventitious rootlets, widely cultivated in hotter and damper parts of

the country is widespread in damp forests and is cultivated in India and other countries in South-East Asia, such as Vietnam and China. Betel leaves were reported to have high antioxidant effects, antidiabetic, Antibacterial Effect and Pro-apoptotic effect (Dwivedi *et al.*, 2011).

Therefore, the present study is focused on with the following objectives:

- To detect the phytoconstituents of the plant samples qualitatively and quantitatively
- To assess the antioxidant property by estimating the enzymic and non-enzymic antioxidants in the selected plant samples.
- To determine the thrombolytic activity by % clot lysis.
- To screen the biosafety of the plant samples using brine shrimp lethality bioassay.
- To analyze the active components present in *Piper betle L.* by HPTLC technique.
- To assess the bioavailability of hydrochavicol using *in silico* studies.