1. INTRODUCTION

An oldest oral disease of human beings, dental caries is a multifactorial disease that has been found to be a significant concern for almost all human populations. It is a widespread disease affecting peoples of all ages and an important health problem in India and the rest of the world. The World Health Organization (WHO) estimated that out of 1116985.088 world population ('000), the dental caries alone caused 88.4702682495117 deaths in WHO member states. Oral diseases caused about 27538 deaths in India, wherein dental caries alone accounted for 16653 deaths (WHO, 2008). Hence, now a day’s dental caries prevalence increase is predicted to be a pending public health global crisis (Bagramian et al., 2009). Though the dental caries have been described as a “dietobacterial” disease, the biofilm forming cariogenic bacteria are implicated as main causatives. This infectious microbial disease causes teeth’s calcified tissue localized dissolution and destruction (Loesche, 1986). They also cause bad breath and foul taste. In highly progressed cases, infection can spread from the tooth to surrounding soft tissues which may lead, to an edentulous mouth (Baelum et al., 1997).

Dental plaque is an example of a biofilm which plays not only a primary role in the pathogenesis of disease but also harbors bacteria having markedly different characteristics than those of identical cells growing in liquid broth (Sohaibani and Murugan, 2012). Xie et al. (2008) defined these dental plaques as “the diverse microbial community found on the tooth surface embedded in a polymer matrix of bacterial and host origin”. It is accepted that bacteria’s ability to form and maintain biofilms plays a key role in their extraordinary adaptation to survive at “feast and famine”, and also in adjusting their needs to accommodate highly diverse environments. Hence many and perhaps most species of bacteria prefer this biofilm mode of growth since they enjoy a number of advantages including protection from environmental factors for instance, host defence mechanisms, potentially toxic environmental substances like lethal chemicals or antibiotics (Socransky and Haffajee, 2000), etc. The structure of the biofilm and the physiological attributes of biofilm organisms confer them an inherent resistance to antimicrobial agents (Donlan and Costerton, 2002). Hence, dental biofilm treatment with antibiotics or other biocides is usually ineffective at eradicating them. Further, the effective prevention of caries would be possible only when the current mechanical
methods including tooth brushing, inter dental cleaning along with professional scaling procedures should remove all plaque, reduce plaque levels below the threshold level for disease, and alter plaque pathogenicity (Addy, 1986), which is unachievable one. Although a number of control measures are in place for oral biofilm, most of them seem to be almost ineffective due to the increased resistance conferred by sessile cells (Sandasi et al., 2011). In several cases, the countless abundant commensals prevents the pathogenic organisms increase thereby halts the ecological balance shift towards pathogenic state. It is more important know the commensal bacteria biology like the oral streptococci to understand the microbial ecologic factors that support a healthy flora and thereby prevent the transition to a pathogenic flora (Nikitkova et al., 2013). The highly active broad spectrum antibiotics affects more the beneficial commensals thus causes oral microbiota imbalance. In addition, these chemotherapeutic agents are known for their unwanted side effects. These drawbacks justify further research and development into natural antibacterial agents that are safe for the host and specific for oral pathogens (Singh et al., 2007).

However, dental caries are preventable. Regular removal of dental plaque and food deposits from the teeth plays an important role factor in their prevention. Although the toothbrushes and toothpastes are widely used, natural methods of tooth cleaning using chewing sticks selected and prepared from the twigs, stems or roots from a variety of plant species is practised in many countries and cultures. Selected clinical studies have shown that chewing sticks, when properly used, can be as efficient as toothbrushes in removing dental plaque due to the combined effect of mechanical cleaning, enhanced salivation, and the antimicrobial substances leaching out into the oral cavity. Recent WHO year 2000 consensus report on oral hygiene also recommended and encouraged the use of these sticks as an effective tool for oral hygiene research (Wu et al., 2011). Since time immemorial plant wealth is greatly exploited for its therapeutic potential and various oral ailments curing medicinal efficacy. Number of plants like Azadirachta indica L, Salvadoras persica L. etc., are reported as a source for chewing sticks or as a tooth cleaning powder (Hebbar et al., 2004) in India and documented evidence for their use in dental health has been found even in ancient Ayurvedic texts (Telles et al., 2009). All kinds of these plants were shown to have medicinal and anti-cariogenic properties, which would have been explored for management and even to cure problems such as oral cavity
deformities, plaques and infections in ancient India (Venugopal et al., 1998). Though numbers of reports are available on the antibacterial activities of the selected Indian medicinal plants, available literature on their anticariogenic activity is very limited.

Throughout the ages, humans have relied on the nature for their basic needs including food, housing, clothing, means of transportation, fertilizers, flavors and fragrances, and not the least medicines (Diallo et al., 1999). Nature has been a source of medicinal agents since time immemorial. During the past thousands of years, these medicinal plants are used as traditional treatment for numerous human diseases in many parts of the world. The use of medicinal plants as traditional medicines is well known in rural areas of many developing countries (Gupta et al., 2005). Medicinal plants are staging a comeback, their renaissance is happening throughout the world. The medicinal plant products today symbolize safety in contrast to their synthetic counterparts that are regarded as unsafe both to human and environment. However, the earlier eyeless reliance on synthetics came to an end, and people are now returning to the naturals with a hope of safety and security (Alavijeh et al., 2012).

The significant role of natural products in both drug discovery and chemical biology are a well-known one. If truth be told, numerous current drugs either mimic natural molecules or have structures that are fully or in part derived from natural motifs. Barks, stems, leaves, flowers and fruits of plants, various animal tissues and microorganisms serve as a source of these natural antimicrobials (Rathish and Sumitra, 2007). Certain important chemical intermediates essential for modern drug manufacturing could also be obtained from plants. Apart from the stable market available for plant-derived drug worldwide, plants still remain an essential source for new drugs. No doubt, antibiotics are one of the best weapons in fighting bacterial infections and have well promoted the health-related quality of human life as long as their introduction. However, during these days, these health benefits are under peril since several frequently used antibiotics became less and less effective against certain illnesses, display toxic reactions (Sarkar et al., 2003) and resistance development against them. This resistance development due to their indiscriminate use now becomes a global concern (Westh et al., 2004). This has forced scientists to search for new antibacterial substances from various sources. The screening of plant extracts and plant products for antibacterial activity has
shown that higher plants represent a potential source of novel antibiotic prototypes (Afolayan, 2003). It is clear that the unlimited sources of active ingredients found in the plant kingdom are extremely useful in much troublesome diseases management. Furthermore, the active components of herbal remedies act synergistic with the number of other appear to be substances (Westh et al., 2004).

The oldest form of healthcare known to human beings, the herbal medicines, now becomes the primary source of knowledge for modern medicine. The basic molecular and active structures of synthetic fields are derived mostly from the rich natural sources. Traditional medicinal plants possess a vast array of substances that can be used to treat chronic and infectious diseases (Jiang et al., 2006). Apart, these natural products are known to be chemically balanced, effective and least injurious with none or much reduced side effects as compared to synthetic medicines. Besides, interest in medicinal plants escalates enormously due to herbal products use as natural cosmetics general public self-medication, and scientific investigations on their biological effects in human beings. Further than this plant’s pharmaceutical approach, there is a trend of supplementing the diet with herbal product primarily with an eye on life quality improvement and prevention of elderly people diseases (Maffei, 2003). Furthermore, this plant-derived drugs interest revival is mainly due to the current widespread belief “green medicine” is “safe and more dependable than the costly synthetic drugs, many of which have adverse side effects” (Rathish and Sumitra, 2007).

An enormous knowledge on the utilization of plants to treat various illnesses might have accumulated in areas where the plants use nevertheless is of high importance (Diallo et al., 1999). Among ancient civilizations, India has been known to be rich repository of medicinal plants. Plants used, especially in Ayurveda can provide biologically active molecules and lead structures for the development of modified derivatives with enhanced activity and/or reduced toxicity. The medicinal plant as such and their crude extract are used as medicaments (Joy et al., 2001). Widespread practice of these traditional medicinal systems has to be continued owing to many reasons. The increasing population, drugs insufficient supply, too expensive cost of treatments, side effects of several allopathic drugs and resistance development to current drug have led to increased emphasis on our reliance on plant materials as medicine source for a wide
variety of human ailments. Regardless of the vast influences and our dependence on modern medicine and synthetic drugs remarkable advances, a sizeable section of the world population yet likes plant derived drugs. Thus, the scientific study on traditional medicines, drugs having bioprospecting origin and systematic conservation of the concerned medicinal plants are of enormous significance (Joy et al., 2001). Many of the plant metabolites are constitutive, existing in healthy plants in their biologically active forms, but others occur as inactive precursors and are activated in response to tissue damage or pathogen attack (Osbourne, 1996). The array of metabolites produced by plants is daunting, with wide ranging chemical, physical and biological activities. They constitute a source of bioactive substances and presently scientific interest on them has increased due to the quest for new drugs of plant origin. However, the isolation and identification of the active principles and elucidation of their mechanism of action is of paramount importance. Hence, works on traditional medicine mixtures and their single active compounds have incredible importance. Modern science and technology have an essential role to play in these processes. “Inventorisation of herbal drugs used in traditional and modern medicines for a country like India, appears to be a stupendous task, where a number of well-established indigenous or traditional systems, including Ayurveda, Unani, Siddha, Homoeopathy, Tibetan, Amchi, Yoga and Naturopathy are practised along with modern medicine for the management of total health care system” (Joy et al., 2001). Plants are the core of these thousands of years old sophisticated traditional medicine systems and provides new remedies to mankind continuously. Unlike our ancestors who made new discoveries on the healing power of plants through trial and error, now we can apply more scientific approaches to understand the medicinal properties of plants and develop new products. Currently used various approaches to discover drugs from nature include

1. Ethno-botanical: Ethnic and traditional medicine
2. Random screening: Bioassay guided routes
3. Chemotaxonomic: Screening of relatives

Traditional healing systems around the world that utilize herbal remedies are an important source for the discovery of new antibiotics (Okpekon et al., 2004). Traditional medical practices are an important part of the primary healthcare system in the developing world (Sheldon et al., 1997). Traditional knowledge about medicinal plants
are continually guiding towards the search for novel treatments. They have given clues for valuable drugs discovery in the face of the advanced modern high throughput drug discovery and screening techniques (Buenz et al., 2004). It also facilitates pharmacological studies leading to synthesis of a more potent drug with reduced toxicity (Ebana et al., 1991; Manna and Abalaka, 2000). Traditionally used medicinal plants have recently attracted the attention of the pharmaceutical and scientific communities. This has involved the isolation and identification of metabolites produced by plants and their use as active principles in medicinal preparations (Taylor et al., 2001).

In developing countries, low income people such as farmers, people of small isolated villages and native communities use folk medicine for the treatment of common infections (Rojas et al., 2006). Hence, herbal medicines assume pronounced significance in the primary healthcare of individuals and communities in many developing countries (Ghosh, 2003). WHO estimates more than 80% of the world’s populations rely on such traditional plant-based systems of medicine for primary health care (Calixto, 2005). So over three-quarters of the world population relies mainly on plants and plant extracts for their health care. Plant drugs also constitute about 25% of the total drugs sold in developed countries like United States. At the same time, their contribution is as much as 80% in fast developing countries including India and China. Thus, the economic importance of medicinal plants is much more to country like India than to rest of the world. These countries provide two third of the plants used in modern system of medicine. Of the 2-50,000 higher plant species on earth, more than 80,000 are medicinal (Alavijeh et al., 2012).

India with its diverse agro-climatic conditions and regional topography has been considered as the treasure house or botanical garden of plant genetic resources. It is recognized as one of the world’s top twelve mega diversity countries (Prakash, 1998). It is a varietal emporium of medicinal plants and is one of the richest countries in the world with regard to medicinal plants genetic resources. This one is rich in all the three levels of biodiversity viz. species diversity, genetic diversity and habitat diversity (Chanda and Kaneria, 2011). It is also one of the supreme emporiums of ethno botanical wealth and harbours 53 millions of tribal population formed as 550 ethnic communities and it have a century old heritage of medicinal plants and herbal medicines for alleviating ailments and
promotion of health and happiness (Rasingam et al., 2012). Considering the current rate of deforestation with the concurrent loss of biodiversity, the knowledge and experience of the traditional herbalists needs to be documented accurately (Grierson and Afolayan, 1999).

Ethno-botany tries to study the relationship between humans and plants. During the last century, it has evolved into disciplines that look at the people-plant relationship in a multidisciplinary manner, such as ecology, economic botany, pharmacology, public health and other disciplines as needed (Balick, 1996). A traditional primary health care practice of indigenous people pertaining to human health is termed ethno-medicine. It is concerned with the cultural interpretations of health, disease, illness and also addresses the health care seeking process and healing practices. On other words, it is an area of research that deals with medicines derived from plants, animals; minerals and etc were used in the treatment of various diseases based on folklore and herbal charms (Maheswari, 1988). This is the mother of all other systems of medicine such as Ayurveda, Siddha, Unani, Nature cure and also modern medicine. The ethnobiological scientific knowledge reciprocally relates human beings with their environment. Sustainable utilization of the locally available resources and the conservation methods they adopt are the basic concerns of ethnobiology (Rasingam et al., 2012). These ethnic peoples have high knowledge on the plants and their medicinal values (Ghosh, 2003). Tribal population provides considerable information about the use of many plants or plant parts as medicine. This knowledge transmitted through particular cultural and traditional information exchange mechanisms, for example, maintained and transmitted orally through elders or specialists like breeders, healers, etc and often to only a select few people within a community (Hansen et al., 2003). Thus the ethno botanical survey can bring out many different clues for the development of drugs to treat human diseases (Ghosh, 2003).

Number reports are available on the usage of traditional plants and natural products for the oral disease treatment. Many traditional medicinal plant-derived medicines have been recorded in pharmacopoeias as agent treating infections, and a number of them have been recently examined for their usefulness against oral microbial pathogens. Several traditional medicinal plants extracts or phytochemicals were revealed
to inhibit oral pathogen growth, reduce dental plaque development, influence the bacterial adhesion to surfaces and reduce oral disease symptoms.

Hence, more studies are available on the evaluation of potential application of traditional medicinal plants in the prevention or treatment of oral diseases. A number of studies have examined the plant extracts and their products activity on specific oral pathogens while others have focused on the products ability to reduce the microbial pathogens adhesion to the tooth surface, the primary event in the dental plaque formation and the progression to tooth decay and periodontal diseases (Palombo, 2009). Dentistry in particular has many reports on the use of these natural products treating oral diseases. Recently, a number of compounds isolated from natural products have been investigated for their efficacy against oral microbial pathogens (Pandit et al., 2011). The vast array of studies available had shown the way for the acceptance of traditional medicine and natural products as an alternative form of oral health care (Prabu et al., 2006) particularly their potential as a source for anticaries agent development. The natural phytochemicals isolated from herbal medicines could offer effective alternatives to antibiotics and represent a promising approach to the prevention and treatment of dental caries and other oral infections (Yim et al., 2013).

This is the right time to look into the rich traditional knowledge and herbal medicine heritage of India. There is an imperative need to inventory and record all ethno-biological information among the diverse ethnic communities before the traditional cultures are completely lost (Lenin Bapuji and Venkat ratnam, 2009). Ethno-botany is not new to India because of its rich ethnic diversity. In South India, the ethno-botanical effort was started by Janaki Ammal 1956 and 1984. After that ethno-botanical studies among various tribal communities were investigated in Tamilnadu such as the Jatapus (Rama Rao et al., 1996), Paliyar (Ignacimuthu et al., 1998), Kanikar (Ignacimuthu et al., 1998), Todas (Ignacimuthu et al., 1998), Gonds; Koysd ; Konda reddis; Valmikis; Koyas; Chenchus; Lambadis; Jatapus; Savaras; Bagatas; Kammaras; Khondas; Nukadoras; Porjas; Kotas; Rajan et al., 2002), Kattunayakas (Balakrishnan et al., 2003), Apatani (Kala, 2005), Chellipale (Udayan et al., 2005), Malasars (Pandikumar et al., 2007), Malamalasars (Yesodharan, 2007), Irulas (Ramachandran and Nair, 1981; Ramachandran and Manian, 1991), Malayalis (Dwarakan and Ansari, 1992; Viswanathan, 1997;
Rengalakshmi, 2005). Kolli hills, Namakkal district, Tamilnadu is a ‘Naturalists Heaven’ a treasure trove of medicinal plants, and the native home of traditional hill country and people. Ever since Kolli hills have always been famous for its medicinal plants. An extensive range of medicinal plants and herbs used in Ayurvedic, Siddha and Unani are natured, cultivated, gathered and sent from here (Elavarasi and Saravanan, 2012). The available literature on the ethanobotanical wealth of Kolli hills is scarce. In the present study, an attempt has been made to conduct the ethno-botanical survey on malayalies tribal community living in Kolli hills with special references to their knowledge between oral diseases and medicinal plants.

1.1 OBJECTIVES OF THIS STUDY

The rationale of this study is to identify oral hygiene maintaining traditional medicinal plants used by tribal peoples living in medicinal plant rich Kolli hills through ethno botanical survey, determination of their anticaries and antibiofilm activity, bioguided fractionation of highly inhibitory plant extracts, isolation then identification of the responsible bioactive compound as well as virtual screening for their bioactivity and interaction with potential targets.

1) To isolate, identify and determine the antibiotic susceptibility pattern of the biofilm forming bacteria from clinical specimens suffering from dental caries.
2) To assess the adherence and biofilm forming ability of the isolates employing cultural as well as molecular methods.
3) To extract, estimate and characterise exopolysaccharides (EPS) produced by the isolates
4) To screen the traditional medicinal plants for their cariogenic bacterial growth and biofilm inhibitory potential
5) To extract, isolate and characterise the bioactive antimicrobial compounds from the efficient plant *Mimosops elengi* L.
6) To determine the mode of action of phytochemical active compounds on biofilm specific events using *in vitro* and *in silico* methods
7) To formulate and develop an effective antibiofilm caries inhibiting product