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Antimicrobial Activity of Mint Extract on Periodontopathic Bacteria-An in-Vitro Study

Chanchal Katariya*, Sankari Malaiappan

Department of Periodontics, Saveetha Institute of Medical and Technical Sciences, (SIMATS)

Saveetha University, India

ABSTRACT

Aim: The aim of the current study is to assess the antimicrobial activity of aqueous and ethanol mint extract against anaerobic and aerobic bacteria found in subgingival plaque of chronic periodontitis patients.

Materials and method: Fresh mint leaves were collected, washed, dried and then finely powdered. Aqueous extract was prepared using 50mg of mint powder, mixed with 100ml of distilled water. The mixture was boiled till it reached 10ml and was filtered using filter paper. Ethanol extract was prepared using 50mg of mint powder and was mixed with 50ml of ethanol. The mixture was left for 24 hours with periodic shaking and was filtered. The prepared extracts were compared with chlorhexidine in aerobic and anaerobic microbial culture.

Results: Chlorhexidine showed maximum zone of inhibition against both aerobic and anaerobic bacteria of 23mm and 20mm respectively. Followed by ethanolic extract of mint with zone of inhibition against both aerobic and anaerobic bacteria of 17mm and 16mm respectively. Zone of inhibition against aerobic and anaerobic bacteria is 11mm and 9mm respectively.

Conclusion: These results supports a notion that mint extract can be used for treatment of periodontal diseases.

Key words: Chlorhexidine, Anaerobic bacteria,

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Corresponding author: Chanchal Katariya
e-mail ✉ : chanchalkatariya0@gmail.com
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INTRODUCTION

Dental plaque is the major causative factor for alleviation of two most common dental problems, dental caries and periodontitis [1]. Investigational studies have proven that dental plaque is responsible for the commencement and advancement of periodontal diseases and it has been harmonized with amount of dental plaque and development of the disease [2]. Dental plaque is a biofilm of microorganisms that grows on surfaces of teeth, prosthesis, implants and dental restorations. It is a sticky colourless deposit at first, but when it forms tartar, it is often brown or pale yellow. The colonizing bacteria that make up the biofilm consortium is one of the major reasons for periodontitis [3]. Periodontal disease is a complex infectious disease resulting from interplay of bacterial infection and host response to bacterial challenge, and the disease is modified by environmental, acquired risk factors and genetic susceptibility. The universal need of using alternate prevention and treatment approaches for oral diseases that are circumspect, efficacious, and economical comes from the increase in disease incidence, resistance of pathogenic bacteria to presently used antibiotics and chemotherapeutics, opportunistic infections in immunocompromised individuals. In spite of assorted means being monetarily available, these chemicals can alter oral micro biota and have unappealing side effects such as vomiting, diarrhoea, and tooth staining [4].

Herbal medicine has become one of the major leading therapeutic approaches for oral diseases. Mentha also known as mint is a genus of plants in the family Lamiaceae. Mentha is a member of the tribe Mentheae in the subfamily Nepetoideae. The tribe contains about 65 genera, and relationships within it remain obscure [5]. It was cultivated by the ancient Egyptians and documented in the Icelandic pharmacopoeia of the thirteenth century. It is widely grown in temperate areas of the world, particularly in Europe, North America and North Africa but nowadays cultivated throughout all regions of the world. The medicinal parts are the essential oil extracted from the aerial parts of the flowering plant, the dried leaves, the fresh flowering plant and the whole plant. Mint leaves yield approximately 0.1–1.0% volatile oil which is composed primarily of Menthol (29–48%) and methane (20–31%) [6]. Mint oil is classified as a carminative (prevents and relieves intestinal gas) [7]. Mint herbs have been allegedly to hold diverse biological effects, including antioxidant, anti-inflammatory, anticancer, and

antimicrobial activities [8]. The Mentha piperita were found a rich source of phytochemical compounds like diterpenes, steroids, tannin, flavonoids, cardial glycosides, alkaloids, phenols, coumarin, and saponin.

The null hypothesis of the study is that aqueous mint extract does not show any antimicrobial activity against periodontopathic bacteria. The aim of the current study is to assess the antimicrobial activity of aqueous and ethanol mint extract against anaerobic and aerobic bacteria found in subgingival plaque of chronic periodontitis patients.

MATERIAL AND METHOD

Preparation of Mint Powder

Fresh mint leaves were first washed under running tap water, then sterilized distilled water, then dried at room temperature in the dark before being ground to powder with an electric blender. The plants' leaves were air dried at room temperature for two weeks before being ground into a coarse powder of 100gms.

Preparation of Mint Extract

Aqueous extract

50mg of mint powder was mixed with 100ml of distilled water. The mixture was boiled till it reached 10ml and was filtered using filter paper.

Ethanol extract

50mg of mint powder was mixed with 50ml of ethanol. The mixture was left for 24 hours with periodic shaking and was filtered.

Microbial Culture

Nutrient agar was used as culture media. 14 grams of nutrient agar is mixed in 500 ml of distilled water and heated till the agar is completely dissolved. The mixture is autoclaved for 15 minutes for 1210C. After autoclaving the mixture is poured in culture plates using micropipette and the mixture is allowed to set. For aerobic bacteria, strain of streptococcus mutants was used. For anaerobic bacteria, subgingival plaque was collected from chronic periodontitis patient and was incubated for 24 hours and cultured. Three wells were prepared in each culture plate for chlorhexidine, aqueous mint extract and ethanolic mnt extract. The extracts' MIC was determined using the agar well diffusion method. Every solution was applied ascetically into the wells of nutrient agar plates that had already been seeded with standardized bacterial isolate inoculum. The plates were incubated for 24 hours at 370°C. The extract concentration with the clearest zone of inhibition was chosen as the lowest concentration (MIC).

RESULTS

The agar well diffusion method was used to screen the antimicrobial activity of the curry leaves extract at a concentration of 50 microliter, and the zone of inhibition was measured in millimetres. Chlorhexidine showed maximum zone of inhibition against both aerobic and anaerobic bacteria of 23mm and 20mm respectively. Followed by ethanol extract of mint with zone of inhibition against both aerobic and anaerobic bacteria of 17mm and 16mm respectively. Zone of inhibition against aerobic and anaerobic bacteria is 11mm and 9mm respectively (Figures 1 and 2), (Table1).

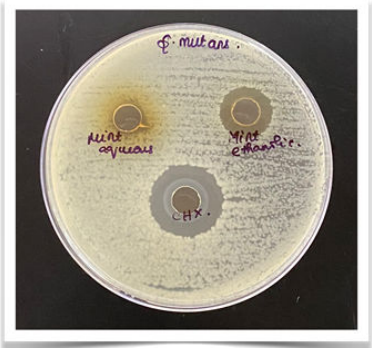

Figure 1: Shows zone of inhibition of chlorhexidine, aqueous mint extract and ethanolic mint extract against aerobic bacteria.

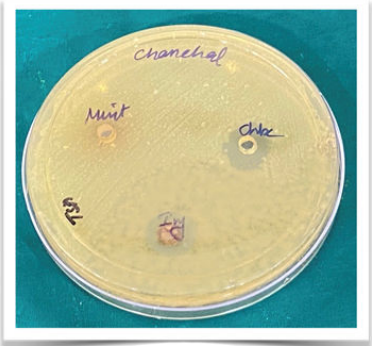

Figure 2: Shows zone of inhibition of chlorhexidine, aqueous mint extract and ethanolic mint extract against anaerobic bacteria.

Table 1: Depicts the zone of inhibition of specific extract against aerobic and anaerobic bacteria.

Extract	Aerobic extract (MM)	Anaerobic bacteria (MM)
Chlorhexidine	23	20
Ethanolic mint	17	16
Aqueous Mint	11	9

DISCUSSION

Plants are the foundation of a complex traditional medicine scheme, and natural products are excellent sources of new drug development leads. It has also been used as an appetite stimulant, a therapy for gastrointestinal infections, and to reduce blood sugar in diabetics, in addition to these properties. It has also been reported to be used to treat some forms of cancer and viral infections. The antimicrobial potency of plants is believed to be due to tannins, saponins, phenolic compounds, essential oils and flavonoids. These compounds are known to be biologically active and therefore aid the antimicrobial activities of the plants. These secondary metabolites exert antimicrobial activity through different mechanisms. 5-a-stigmasta-7, 25-dien-3-b-ol, elasterol, and lanosterol are active constituents that may be responsible for its antibacterial activity [9]. Herbalists consider it as an astringent, antiseptic, antipruritic, antispasmodic, anticatarrhal, antimicrobial, rubefacient, stimulant and emmenagogue [10].

In the present study, even though chlorhexidine showed maximum antibacterial effect, the results were comparable to mint extracts. Antibacterial activity of aqueous extracts of common Mints was tested against multidrug-resistant bacteria, suggesting that their extracts can be used against multidrug-resistant bacteria capable of causing nosocomial and community-acquired infections [11]. A study found that all of the peppermint oils tested strongly inhibited plant pathogenic microorganisms, but only moderately inhibited human pathogens. The antimicrobial activity of these oils was discovered to be attributable to menthol using the bio autography assay [12]. The essential oils from mint species have antimicrobial activity against several clinical isolates studied, according to the current research, and thus can be a good source of natural antimicrobial agent [13]. Chlorhexidine mouthwash in reducing microbial plaque is better than mint mouthwash but regarding feeling flavour, no burning sensation and less staining, mint mouthwash had better efficacy [13].

According to Suree and Pana, the bacterial strains' varying degrees of sensitivity may be attributed to the bacteria's inherent tolerance and the presence and combinations of phytocompounds present in the extracts [14,15]. The future scope of this study is to use mint extract as irrigant and local drug delivery as adjunct periodontal therapy.

CONCLUSIONS

Plants are considered to have medicinal properties. Plant secondary metabolites were discovered to be a source of a variety of phytochemicals that could be used as direct intermediates in the development of new drugs. It is important to encourage the use of herbs in dentistry in order to reduce the number of side effects associated with the use of synthetic medication. In the future, it will be critical to scientifically isolate the active component of mint leaves and conduct numerous animal and clinical trials to determine the exact mechanism of mint leaves' antimicrobial activity. Not only the plant's leaves and bark, but also its seeds, flowers, and other parts are being studied for their medicinal properties.

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