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RESEARCH ARTICLE

Comparative study of the antibacterial activity of *Azadirachta indica* A. Juss. and *Piper betle* L. against *Streptococcus mutans*, an oral pathogenic bacterium

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Abstract

The present study aimed to evaluate and compare the antibacterial activity of *Azadirachta indica* A. Juss. (Neem) and *Piper betle* L. (Betel) against *Streptococcus mutans*, a key oral pathogen associated with dental caries. Fresh and dried leaf and stem samples of both plants were collected, shade-dried, powdered and subjected to solvent extraction using methanol and distilled water. The antibacterial potential was assessed using the agar well diffusion method, with gentamicin serving as the positive control and plain solvents as negative controls. The results demonstrated that all plant extracts exhibited varying degrees of antibacterial activity. Notably, fresh methanol extract of *A. indica* stem showed the highest zone of inhibition (10.5 mm), followed by the leaf extract of *P. betle* leaf (8 mm). In general, dried extracts were less effective than fresh ones and methanol proved to be a more efficient solvent than water for extracting bioactive compounds. These findings underscored the potential of these traditionally used medicinal plants as natural alternatives for oral hygiene management and supported the development of plant-based mouthwashes or dental formulations. Further in vivo studies and clinical trials were recommended to validate these *in vitro* findings and to identify the active constituents responsible for the antibacterial effects.

Keywords: antibacterial activity; *Azadirachta indica* A Juss; methanol extracts; *Piper betle* L; *Streptococcus mutans*

Introduction

Oral health was an integral component of overall health and well-being, as the condition of the oral cavity often reflected and influenced general health. According to the World Health Organization (WHO), oral diseases were major public health problems due to their high prevalence and impact on quality of life and they shared common risk factors with other noncommunicable diseases. Several health issues such as nutritional deficiencies (like vitamin C or B_{12} deficiency) and systemic diseases (such as diabetes, anaemia and HIV) often manifested early signs in the oral cavity, including gum bleeding, oral ulcers, dry mouth and infections (1). Various factors like a person's nutritional status, smoking, alcoholism and hygiene among others, were linked to a wide range of diseases. Oral hygiene played a central role in preventing oral diseases and maintaining oral health. The best way of maintaining good oral hygiene for good health of oral tissue was by "plaque control" since plaque was a main factor responsible for dental and gingival disease (2).

The adhesion of microbial pathogens to the tooth surface was a primary event in the formation of dental plaque and the progression to tooth decay. Over 750 species of bacteria inhabited the human oral cavity, forming a complex and dynamic microbial community. A significant number of these microorganisms were implicated in the development of oral diseases such as dental caries, periodontal disease and endodontic infections. Understanding the composition and role of the oral microbiota was essential for advancing oral health strategies. The human oral microbiome comprised over 700 bacterial species, highlighting the diversity and clinical significance of these microbial populations in oral health and disease (3).

The development of dental caries involved acidogenic and aciduric Gram-positive bacteria, primarily the *Streptococci mutans, Lactobacilli* and *Actinomycetes*, which metabolized sucrose to organic acids that dissolved the calcium phosphate in teeth, causing decalcification and eventual decay (4). Treating and preventing oral diseases was usually safe and effective. But problems like antibiotic resistance, infections in people with weak immune systems and high treatment costs in developing countries remained common challenges. Moreover, allopathic medicines were too expensive and capital intensive for a developing country like India and had only limited success in the

prevention of periodontal disease and in the treatment of a variety of oral diseases.

Hence the research for alternative products continued and plant extracts used in traditional medicine were considered as good alternative to western medicine (5). Medicinal plants had been used as traditional treatments for numerous human diseases for thousands of years and in many parts of the world, continue to serve as the primary source of medicine, especially in rural areas of developing countries (4). Many herbal medicines proved to be efficacious and their beneficial effects were supported by scientific evidence. Herbs offered potential alternatives to current treatments for oral health problems. However, there was still limited information regarding their effects on oral tissues, mechanisms of action and potential side effects. Herbal products had comparatively fewer side effects and were safer to use than conventional medicines particularly in forms like pastes, cream, ointments, gargles or mouthwash. There was an urgent need for evidence - based research on herbal medicine to evaluate their efficiency and safety in the treatment of oral diseases (6). An antimicrobial was an agent that killed microorganism or inhibited their growth. Agents that killed microbes were called microbicidal, while those that merely inhibited their growth were called biostatic (7). Antibiotics helped extend life expectancy by improving outcomes of bacterial infections. They played a pivotal role in achieving major advances in medicine and surgery. Antibiotics reduced the morbidity and mortality caused by foodborne and other poverty-related infections (8).

Streptococcus mutans was a gram-positive bacterium that resided in the human mouth and more specifically, in the multispecies biofilms on the surface of teeth. *S. mutans* was the major cariogenic organism - the result of their ability to produce the large quantities of glucans as well as acids, exceeding the salivary buffering capacities, which gave the bacteria an advantage to outcompete non-cariogenic species at low pH environments. They spread into other sites of oral mucosa and gained access into deeper tissues, causing dissolution of enamel and dentin which resulted in cavitations within the tooth (9).

Piper betle Linn. was commonly known as betel or pan. It belonged to the family *Piperaceae*. Betel was a perennial, dioecious, creeper probably a native of Malaysia, cultivated in India for its leaves used for chewing. Stems were semi-woody, climbed by short adventitious roots; leaves were 5-20 cm long and broadly ovate, slightly cordate and often unequal at the base, shortly acuminate, acute, entire with often an undulate margin, glabrous, yellowish or bright green, shining on both sides; petiole was stout 2-2.5 cm long, pendulous; fruits were rarely produced, often sunken in the fleshy spikes, forming nodule like structures. In India, the male plant was usually cultivated, which did not produce fruit. Betel vine was propagated only vegetatively by cuttings taken from healthy vines at least two years old. These were obtained from vines of the previous year's growth, trimmed into sections of 30 - 45 cm length. Each cutting contained 3-5 nodes and was planted in such a manner that two nodes were buried in the solid and one or more nodes remained above ground and pointed towards the standards, onto which they would eventually trail. Betel chewing was traditionally considered beneficial and was regarded as a source of dietary calcium. The leaves contained vitamin B (nicotinic acid), ascorbic acid and carotene (10). The leaves were used in a number of traditional remedies such as for treatment of stomach ailments, infections and as a general tonic. Some research suggested that betel leaves had the ability to boost the immune system. In the Indian sub-continent, a small bundle of betel known as 'pan-supari' was offered to guests as courtesy (11). The juice obtained from the leaves was useful for certain eye diseases (12).

The *Piper betel* L. leaf was described to have contained compounds piperol-A, piperol-B and methylpiper. The betle leaves contained starch, sugars, diastases and essential oils composed of terpinen-4-ol, safrole, allyl pyrocate, cholo monoacetate, eugenol, eugenylacetate, hydroxyl chavicol and the betel oil contained cadinene, carvacrol, allylcatechol, chavicol, p-cymene, caryophyllene, chavibetol, cineole, estragol as the key components. Phytochemical analysis on leaves revealed the presence of alkaloids, tannins, carbohydrates, aminoacids and steroidal components (13). Studies on *Piper betle* L. reported that it contained important medicinal properties like anti-cancerous, anti-malarial, anti-filarial, anti-bacterial, antifungal, insecticidal, antioxidant, wound healing activity, oral hygiene and anti-asthmatic effect (14).

Azadirachta indica A. Juss., which was commonly known as neem, belonged to the family of *Meliaceae*. Neem had been used in Ayurvedic medicine for over 4000 years, with documented references dating back to 4500 years ago. Besides that, the plant was regarded as village dispensary in India. Neem was called by a variety of names by different ethnicity; in Tamil it was called *Vembu*, Hindi-*Nim* and English-*Lilac*. Neem was native to East Indian and Burma and grew abundantly in south East Asia and West Africa; it was cultivated in Pakistan, Peninsular Malaysia, Singapore, Philippines etc. The neem tree was about 12-18 m in height with a circumference of up to 1.8-2.4 m. Neem was a flowering plant which produced flowers on 3-5 years of age in which the flowers were 4-7 mm in length and 6-10 mm in width. The flower had a jasmine like odour and white in colour. The leaves were dark green in colour up to 30 cm in length and had 3 lobed stigma and seeded drupes. The fruit of neem was about 2 cm long with white kernels and when mature it was able to produce 50 kg of fruit yearly. The branches of neem were dense and up to 10 cm in length and has dark brown bark. Furthermore, the neem tree was able to adapt very dry condition which is up to 1200 m from mean sea level with minimal rainfall of 458 mm per year. Besides that, the plants can grew well in a calcareous soil with pH up to 8.5 (15).

Neem showed its therapeutic role in health management due to its rich source of various types of ingredients. The most important active constituent was azadiractin and the others were nimbolinin, nimbin, nimbidin, nimbidol, sodium nimbinate and quercetin. Leaves contained bioactive compounds like nimbin, nimbinane, 6-desacetyl nimbinene, nimbandiol, nimbolide, ascorbic acid, 7-deacetyl 7-benzoylazadiradione, 7-deacetyl 1-7-benzoylgedunin, 17-hydroxy azadiradione and nimbiol. Quercetine, sitosterol and Polyphenolic flavonoids, were purified from neem fresh leaves and were known to have anti-bacterial and antifungal properties. Seeds held valuable constituents including gedunin and azadirachtin (16). The neem oils were extracted from the pulp of the fruits, which were used in the manufacture of margosa soap and several skin ointments.

The oilcake, obtained from the seeds, was used as a fertilizer and manure. Almost all the compounds were used to repel insects and to preserve woollens. A decoction of leaves was an antiseptic and was used to wash ulcers and wounds. An extract of the leaves was used in the manufacture of tooth pastes and soaps. The seed oil was applied as an antiseptic. The gum, bark, leaves and seeds were applied in snake bite (17). From centuries, millions had cleaned their teeth with neem twigs and taken neem tea as a tonic. The tree had relieved many different pains, fevers, infections and other complaints so that it was called the "village pharmacy". Neem had antibacterial, antiviral, anti-inflammatory, antioxidant, anti-carcinogenic and anti-snake venom activity. Neem was employed to treat skin diseases, digestive disorders and also to prevent sexually transmitted diseases (18).

Materials and Methods

Plant extract preparation

The various plant parts used for antibacterial activity were listed as follows: *Piper betle* Linn. -Leaves. *A. indica* A. Juss. - Stems and Leaves. Plant materials were collected from Thrissur, Kerala and taxonomically identified (19). The collected plants were washed with water, disinfected with ethanol 5 % and rinsed with distilled water and finally dried in shade for 72 hr. The dried plant material of each plant species was grounded into fine powder. The powders were collected separately, in air tight containers, labelled and stored at room temperature for dried extract preparation.

Extraction by mortar and pestle

Ten gram each of plant materials were weighed separately and crushed using mortar and pestle with 10 mL of each solvent (methanol and distilled water). Each extract was poured into test tubes, labelled and the mouth of each tube was covered to prevent evaporation.

Cold extraction

One gram of each powdered sample was taken in separate test tubes and 10 mL of each solvent was added. Then each test tube was labelled and covered properly. The test tubes were placed in shaker (Rotek, Cochin) at 350 rpm for 7 days. A solid residue was deposited at the bottom of the test tubes. It was then collected and stored at room temperature for further use.

Test microorganism

The common oral pathogenic bacteria *S. mutans* was used in this study. The culture was procured from Microbial Type Culture Collection and Gene Bank (MTCC) Chandigarh.

Solvents

The solvents used were 10 % methanol and distilled water.

Standard

The antibiotic gentamicin (50 mg mL^{-1}) (Genta LC, Life Cure Biotech, New Delhi) was used as the standard.

Culture media

The media used for antibacterial test was Mueller Hinton Agar (MHA) medium (Nutri Select® Plus, Germany). The culture media was prepared by dissolving 19.5 g of MHA in 500 mL of distilled water and was allowed to boil without clumps. Then it was transferred to conical flask and plugged with cotton plug for sterilization.

Inoculation of bacteria

The broth was kept inside the laminar airflow (LAF) chamber and was inoculated using a sterile inoculating loop.

Antibacterial assay by agar well diffusion method

The antibacterial agents present in the plant extract were allowed to diffuse out into the medium and interact in plates with freshly seeded test organisms. The resulting zones of inhibition were uniformly circular, as there was a confluent lawn of growth. The diameter of zone of inhibition was measured.

The autoclaved media were transferred aseptically into each sterilized petri plate. The plates were left at room temperature in LAF for solidification. Agar wells were prepared with the help of sterilized micropipettes. Wells were labelled as MS, WS and +ve for methanol sample, water sample and gentamicin, respectively.

The culture suspensions from the nutrient broth were swabbed on the media using streak plate method. Various plant extracts were carefully added to the wells in the labelled petri plate and the antibiotic disc (gentamicin) with a disc potential 30 µg was placed in the well for comparison of antibacterial activity. Each plate was wrapped with cling film. Plates were incubated at 37 ºC for 24 hr. The diameter of zone of inhibition of the tested microorganism by various extracts was measured and compared with those observed with standard antibiotic disc and the results were recorded simultaneously.

Statistical analysis

The experimental data obtained from the agar well diffusion method were statistically analysed to evaluate the antibacterial activity of various plant extracts. The primary dependent variable was the zone of inhibition (mm) formed against *S. mutans*. Data were expressed as mean ± standard deviation (SD) from triplicate determinations. A $p < 0.05$ was considered statistically significant. All statistical analyses were carried out using SPSS version 20 (20).

Results and Discussion

The present study evaluated the antibacterial activity of methanol and aqueous extracts of *Piper betle* leaves and *A. indica* (leaves and stem), using both fresh and dry plant materials, against *S. mutans,* a key oral pathogenic bacterium. The antibacterial potential was assessed using the agar well diffusion method and the efficacy was determined by measuring the diameter of the zone of inhibition (in mm) (Fig. 1A-F).

The results (Table 1) indicated that all fresh plant extracts exhibited measurable antibacterial activity, with methanolic extracts showing greater inhibition than aqueous extracts. The fresh methanol extract of *A. indica* stem exhibited the highest antibacterial activity with a mean inhibition zone of 10.5 mm, followed closely by *Piper betle* leaf extract (8 mm). In contrast, aqueous extracts produced relatively smaller inhibition zones, ranging from 4.5 mm to 7 mm, highlighting lower solubility or extraction efficiency of antibacterial compounds in water (21).

The dry samples also exhibited antibacterial activity, but to a lesser extent compared to fresh extracts (Table 2). This decline could be attributed to possible degradation of active

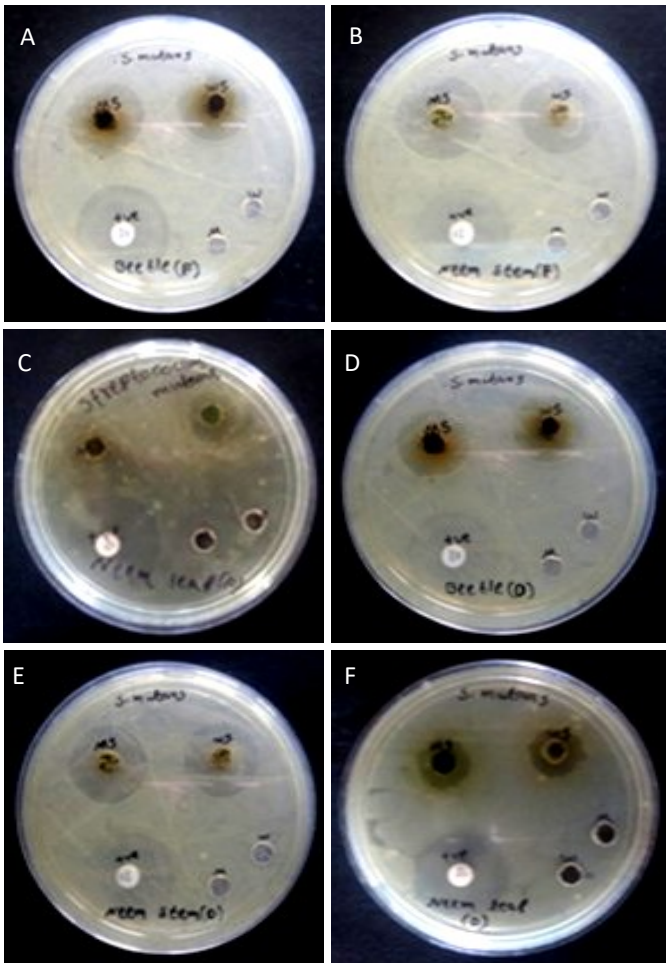

Fig. 1. (A-F) Plates showing zone of inhibition against *S. mutans* by plant extracts. A) *P. betle* leaf (Fresh); B) *A. indica* stem (Fresh); C) *A. indica* leaf (Fresh); D) *P. betle* leaf (Dry); E) *A. indica* stem (Dry) and F) *A. indica* Leaf (Dry).

Table 1. Zone of inhibition of fresh plant extracts against *S. mutans*

Sample	Solvent	Mean Zone of Inhibition (mm)
P. betle leaf (Fresh)	Methanol	8.0
P. betle leaf (Fresh)	Water	7.0
A. indica leaf (Fresh)	Methanol	5.0
A. indica leaf (Fresh)	Water	4.5
A. indica stem (Fresh)	Methanol	10.5
A. indica stem (Fresh)	Water	9.0

Table 2. Zone of inhibition of dry plant extracts against *S. mutans*

Sample	Solvent	Mean zone of inhibition (mm)
P. betle Leaf (Dry)	Methanol	7.0
P. betle Leaf (Dry)	Water	6.5
A. indica Leaf (Dry)	Methanol	4.0
A. indica Leaf (Dry)	Water	3.0
A. indica Stem (Dry)	Methanol	10.0
A. indica Stem (Dry)	Water	9.0

phytochemicals during drying or reduced bioavailability (22). The dry methanol extract of *A. indica* stem maintained substantial activity with a zone of 10 mm, indicating the relative stability of its active compounds. *Piper betle* leaf extracts also showed consistent activity (7 mm with methanol), though slightly lower than fresh samples (Fig. 2).

The two-way ANOVA results revealed that both plant type and extraction method (fresh *vs*. dry) had statistically significant effects ($p < 0.05$) on antibacterial activity (Table 3). The interaction between solvent type and plant material was also significant, suggesting that methanol enhanced the extraction of antibacterial compounds more effectively than water, particularly in fresh samples. Among all combinations, the fresh methanolic extract of *A. indica* stem was more effective.

Statistical analysis using two-Way ANOVA also showed that the type of plant extract had a significant impact on antibacterial activity ($p < 0.001$). However, neither the sample type (fresh *vs*. dry) nor the interaction between plant and sample type had statistically significant effects ($p > 0.05$) (Table 4). These results confirmed that plant selection was a critical factor in determining antibacterial efficacy, while the difference between

Table 3. Two-way ANOVA – Effect of plant type and sample type on antibacterial activity

Source of variation	Sum of squares	df	*F*-value	*p*-value
Plant type	60.667	2	63.304	0.000
Sample type (Fresh/Dry)	1.687	1	3.522	0.110
Plant × Sample Interaction	0.500	2	0.522	0.618
Residual	2.875	6		

A significant effect was observed for plant type ($p < 0.001$), indicating that this variable differed significantly among the three plant types. In contrast the effects of sample type and the interaction between plant type and sample type were not statistically significant ($p > 0.05$).

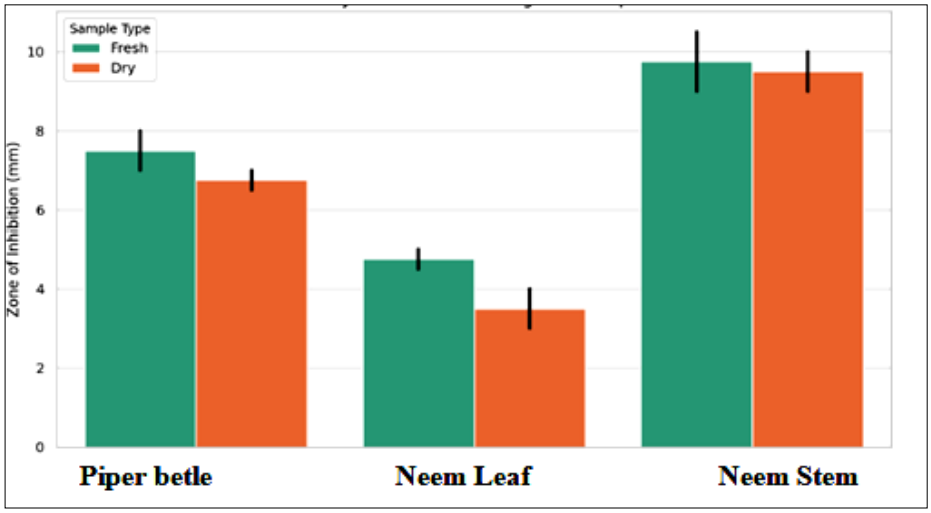

Fig. 2. Comparative antibacterial activity of fresh and dry extracts of different plants against *S. mutans*.

Table 4. Two-way ANOVA – Effect of solvent and sample type on antibacterial activity.

Source	Sum of squares	df	*F*-value	*p*-value
Solvent	2.521	1	0.328	0.583
Sample type (fresh/dry)	1.688	1	0.220	0.652
Solvent × Sample type	0.021	1	0.003	0.960
Residual	61.500	8		

Two-way ANOVA results examined the effects of solvent, sample type (fresh vs. dry) and their interaction on antibacterial activity. None of the main effects or interactions were statistically significant, as indicated by the *p*- > 0.05. This suggests that neither the type of solvent nor the sample condition had a significant impact on antibacterial activity.

fresh and dry forms was less pronounced.

The antibacterial zones of herbal extracts were compared to the standard antibiotic gentamicin (11-12 mm). Though slightly less than the standard, the performance of *A. indica* stem and *Piper betle* leaves, especially in methanol approached the efficacy of gentamicin. This highlighted the potential of these plant extracts as natural alternatives for managing oral pathogens.

The higher antibacterial activity in *Piper betle* was attributed to the presence of phenols (eugenol, chavicol) and terpenes, while *A. indica* contained azadirachtin, nimbidin and quercetin - all known for their antimicrobial properties. The traditional use of both plants in oral care, such as chewing betel leaves or brushing teeth with neem twigs, aligned with these findings.

Conclusion

Plants played a vital role in human health and numerous studies revealed the medicinal and pharmaceutical values of many species. The present study demonstrated the antibacterial effect of two plants: *Piper betle* Linn. and *Azhadiracta indica* Juss. (22). The plants were collected, identified and their methanol and water extracts were prepared using various plant parts. Oral pathogens were treated with these plant extracts and standard antibiotics to compare of their sensitivity patterns. Analysis using the disc diffusion method revealed that the fresh methanol extract of *A. indica* stem exhibited the maximum zone of inhibition against *S. mutans*, indicating strong antibacterial activity. It was evident from the present study that fresh extracts were more active than dry samples. It was also observed that methanol extracts showed greater antibacterial effects than aqueous extracts. The present investigation, along with previous studies, supported the use of plant parts inhibiting bacterial growth. Therefore, the preparation of mouth rinses, toothpastes and other oral hygiene products based on these natural extracts could become more effective and would likely have fewer side effects. However, additional *in vivo* studies and clinical trials would be needed to further evaluate the active compounds and their potential applications.

Authors' contributions

JP developed the research idea, conducted the experiments, supervised the study and contributed to the writing, editing and final approval of the manuscript. MX assisted in experimental design, provided critical inputs during data interpretation and contributed to the review, editing and refinement of manuscript. KJ initiated the study and participated in its design and coordination. S edited and revised drafts in collaboration with other authors and contributed technical expertise in specific methods. All authors read and approved the final version of the manuscript.

Compliance with ethical standards

Conflict of interest: The authors declare that they have no conflicts of interest.

Ethical issues: None

References

1. World Health Organization. Oral health. [Internet]. Geneva: World Health Organization; [cited 2025 Mar 17]. Available from: https://www.who.int/news-room/fact-sheets/detail/oral-health
2. Sharda A, Sharda J. Factors influencing choice of oral hygiene products used among the population of Udaipur, India. Int J Dent Clin. 2010;2(2):7–13.
3. Dewhirst FE, Chen T, Izard J, Paster BJ, Tanner AC, Yu WH, et al. The human oral microbiome. J Bacteriol. 2010;192(19):5002–17. https://doi.org/10.1128/jb.00542-10
4. Palombo EA. Traditional medicinal plant extracts and natural products with activity against oral bacteria: Potential application in the prevention and treatment of oral diseases. Evid Based Complement Alternat Med. 2011;2011:680354. https://doi.org/10.1093/ecam/nep067
5. Nagi R, Devi BY, Rakesh N, Reddy SS, Patil DJ. Clinical implications of prescribing nonsteroidal anti-inflammatory drugs in oral health care- A review. Oral Surg Oral Med Oral Pathol Oral Radiol. 2015;119(3):264–71. https://doi.org/10.1016/j.oooo.2014.12.002
6. Vijayakumar S, Yabesh JM, Prabhu S, Manikandan R, Muralidharan B. Quantitative ethnomedicinal study of plants used in the Nelliyampathy hills of Kerala, India. J Ethnopharmacol. 2015;161:238–54. https://doi.org/10.1016/j.jep.2014.12.006
7. Juhász J, Kertész-Farkas A, Szabó D, Pongor S. Emergence of collective territorial defense in bacterial communities: Horizontal gene transfer can stabilize microbiomes. PLoS One. 2014;9(4):e95511. https://doi.org/10.1371/journal.pone.0095511
8. Ventola CL. The antibiotic resistance crisis: part 1: causes and threats. Pharm Ther. 2015;40(4):277–83. Available from: https://pmc.ncbi.nlm.nih.gov/articles/PMC4378521/
9. Metwalli KH. Dental plaque microbiology. In: Hassan AAF, editor. Oral microbiology. InTech; 2013. https://doi.org/10.5772/53944
10. Pandey AK, Das S, Kumar P. Various methods for minimizing post-harvest losses of betelvine leaves (*Piper betel* L.). Int J Curr Microbiol Appl Sci. 2018;7:1037–43.
11. Fazal F, Mane PP, Rai MP, Thilakchand KR, Bhat HP, Kamble PS, et al. The phytochemistry, traditional uses and pharmacology of *Piper betel* Linn. (betel leaf): A pan-Asiatic medicinal plant. Chin J Integr Med. 2014;20(10):777–85. https://doi.org/10.1007/s11655-013-1334-1
12. Bhalerao SA, Prabhu DV. Aluminium toxicity in plants- A review. J Appl Chem. 2013;2(3):447–74.
13. Dwivedi V, Tripathi S. Review study on potential activity of *Piper betle*. J Pharmacogn Phytochem. 2014;3(4):93–98.
14. Ranjith D, Nisha AR, Nair SN, Litty M, Rahman M, Juliet S. Evaluation of analgesic and anti-inflammatory activity of herbal formulation used for mastitis in animals. Int J Appl Sci Eng. 2018;6(1):37–48.

https://doi.org/10.30954/2322-0465.1.2018.4

15. Narwal SS, Tauro P, Bisla SS. Neem in sustainable agriculture. Jodhpur: Scientific Publishers; 2003.
16. Nishan M, Subramanian P. Toxicity activity of *Azadirachta indica* and *Murraya koenigii* against *Aedes albopictus* larvae in laboratory conditions. Mortality. 2014;1(1):1–6.
17. Pandey BP. Economic botany. New Delhi: S. Chand Publishing; 1999.
18. Kumar VS, Navaratnam V. Neem (*Azadirachta indica*): Prehistory to contemporary medicinal uses to humankind. Asian Pac J Trop Biomed. 2013;3(7):505–14. https://doi.org/10.1016/S2221-1691(13)60105-7
19. Sasidharan N. Illustrated manual on tree flora of Kerala supplemented with computer-aided identification. Peechi: Kerala Forest Research Institute; 2006. p. 698. Available from: https://docs.kfri.res.in/xxxxKFRI-RR/KFRI-RR282.pdf
20. Keselman HJ, Rogan JC. A comparison of the modified-Tukey and Scheffe methods of multiple comparisons for pairwise contrasts. J Am Stat Assoc. 1978;73(361):47–52. https://doi.org/10.2307/2286529
21. Nalina T, Rahim ZH. The crude aqueous extract of *Piper betle* L. and its antibacterial effect towards *Streptococcus mutans*. Am J Biotechnol Biochem. 2007;3(1):10–15. https://doi.org/10.3844/ajbbsp.2007.10.15
22. Shukla R, Satish V, Singh VK, Kumar S, Gupta S, Gavani U. Antibacterial activity of fresh leaves of *Piper betle* Linn. Pharma Res. 2009;1:110.

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