Available online at www.ijpcr.com
International Journal of Pharmaceutical and Clinical Research 2015; 7(2): 151-153

ISSN- 0975 1556

Research Article

Comparative Evaluation of Antibacterial Properties of Different Extracts of *Juglans regia* (Walnut) & *Erethia laevis* (Ajaan Vruksh) Against Salivary Micro Flora

Mutha M.[1,*], Deshpande R.R.[1, 3], Shep S.[1], Deshpande N.R.[2], Torne R.[2]

[1]*Dr. D. Y. Patil Dental College and Hospital, Pimpri, Pune-18, Maharashtra, India*
[2]*Dr. T. R. Ingle Research Laboratory, Department of Chemistry, S. P. College, Pune – 30, Maharashtra, India*
[3]*Deenanath Mangeshkar Hospital, Pune-4, Maharashtra, India*

Available Online: 26th February, 2015

ABSTRACT

Objective: evaluation of antimicrobial properties of different concentration of extracts of Juglans regia and Erethia. Laevis extracts against human salivary micro flora.

Method: microbial inhibition assay was prepared using the agar 'well-diffusion' method. Sterile 8.0mm diameter of well were impregnated with the extract of different concentrations.

Results: This study compares antimicrobial activities of "Juglans regia and Erethia Laevis". The zone of inhibition is measured by excluding the diameter of the well. These zones of inhibition are directly proportional to the concentration.

Conclusion: The ingredients derived from Erethia laevis & Juglans regia plants used in this study are herbal, they are eco-friendly and do not produce any side effects as well as are effective and economical, as compared to the synthetic drugs. The study also confirmed the antimicrobial potentials of the plant at different concentrations and can be used as preventive and therapeutic measure in dentistry.

Keywords: Erethia laevis, Juglans regia, Medicinal Plants, Antimicrobial

INTRODUCTION

Caries is a more gradual disease process, with demineralization and remineralization occurring over time. Thus, the process is not a step function where surfaces or teeth show transition, instantly, from sound to cavitations. Neither visual examination nor various radiographic methods can adequately describe the process. During the timeframe from 1968 till today, significant advances have been made with respect to our understanding of the molecular basis of caries and our ability to measure earliest enamel demineralization changes and thus caries progression[1,2]. Thus, efforts need to be made for the primary prevention of dental caries initiation, rather than its treatment, throughout the life.

Herbal medicine is still the mainstay of about 75 - 80% of the world population, mainly in the developing countries, for primary health care[3]. This is primarily because of the general belief that herbal drugs are without any side effects besides being cheap and locally available[4]. According to the World Health Organization (WHO), the use of herbal remedies throughout the world exceeds that of the conventional drugs by two to three times[5]. Natural products of higher plants may provide a new source of antimicrobial agents with possibly primordial prevention type of mechanism of action.

Moreover, chemicals like chlorhexidine and amine fluorides have undesirable side-effects such as staining of teeth and restorations increase in calculus deposition and imbalance of the oral and intestinal flora, thus leading to vomiting and diarrhea. These drawbacks justify the search for new effective anticariogenic compounds that could be employed in caries prevention[6,7].

In this sense, efforts have been made to evaluate the antimicrobial properties of extracts of Erethia laevis and Juglans regia against human salivary micro flora.

MATERIALS AND METHODS

The ethical approval was taken from the ethical committee of the institute.

Inclusion criteria: Patients of 6 – 12 years old in mixed dentition age group with moderate caries (DMFT=3-4) (modified WHO criteria 2003) having good general health[8].

Exclusion criteria: Patients with history of antibiotic and oral drug therapy, chemical anti-plaque agents prior to six months of study initiation, physically and mentally handicapped patients were excluded from the study.

Plant extracts: Plant materials used in this study were procured from the local market of Pune, Maharashtra, India. The plant materials of J. regia were authenticated at Agharkar Research Institute, Pune, India with authentication number AHMA S/B –14319. The plant material of Erethia laevis was Authenticated at Botanical

**Author for Correspondence*

Survey of India, Pune, Maharashtra. Its voucher number is BSI/WC / Tech / 2006 /185.

Table: 1 Mean value of Zones of inhibition of Juglans regia extract at different concentrations.

Concentration of the J. regia extract(µg/ml)	Mean
100	8.75
150	10.5
200	10.25
250	10.75
Control	18.50

Table: 2 Mean values of Zones of inhibition of Erethia laevis extract at different concentrations

Concentration of E. laevis (µg/ml)	Mean
100	00
200	0.4
400	4.8
800	6.7
Control	18.50

Preparation of extracts

Air shade dried powdered bark material (10g) of Juglans regia was extracted using acetone (50ml), by soaking it for 24 hours at room temperature. The solvent was evaporated under reduced pressure to obtain crude acetone extract (6.6 %). Air shade dried and pulverized material (60.0 g) of E.laevis was charged with methanol (360 ml) at room temperature for eighteen hours. The solvent was recovered in vacuum under reduced pressure to yield crude methanol extract (5.83 %).

Consent: Written informed consent was taken from the parents or guardian of the subjects after explaining the procedure to them.

Saliva collection: The subjects were told to rinse with water; saliva was allowed to accumulate in the floor of the mouth for approximately two minutes and by asking the subject to spit in funnel, saliva (3ml) was collected in vial. 10 samples were collected in the early morning time. These salivary samples were diluted (3:1) in a sterile vial containing 1ml of normal saline and were used to inoculate on the agar plates.

Antimicrobial Assay: The microbial inhibition assay was prepared using the agar 'well-diffusion' method. Sterile 8.0mm diameter of well were impregnated with the extract of different concentrations. Adequate amount of Muller Hinton Agar were dispensed into sterile plates and allow solidifying under aseptic conditions. The test samples of saliva (0.1ml) were inoculated with a sterile spreader on the surface of solid Muller Hinton Agar medium in plates. After the media was solidified; a well was made in the plates with the help of a cup-borer (8.0mm). The well was filled with different concentrations of the extract and plates were incubated at 37 ± 0°C for 24 hours after incubation, the plates were observed for zones of inhibition of growth and the diameters of these zones were measured in millimeters by using bacterial inhibition zone reading scale.

RESULTS

The results of the anti-microbial assay of the acetone extract of J. regia showed average zones of inhibition (mm) as in reported in TABLE: 1. A dose dependant evaluation of extract on a salivary micro flora was analyzed and recorded. It was noted that zone diameter is increased from 100 µg to 250 µg and remain steady. The crude extract can be enhanced in activity upon further work. The experiment was performed with various concentrations of the E. laevis. The results are depicted in TABLE: 2. It showed that all the concentrations have marked activity against the tested microorganisms. Results of test samples are reported after twenty four hours and indicate its dose dependent activity. It appears that zone of inhibition increases at 400 µg/ ml from K to M. Extract M reveals maximum zone of inhibition at 400 µg/ ml, as compared to K and L extract. The results watched carefully materialized that there is very slight increase in activity at 800 µg/ ml.

DISCUSSION

Emergence of multidrug-resistant strains, significant side effects of existing antibacterial drugs and their limited options are unsolved problems even today. The search for alternative, superior medicines leads us to the promising source of natural products. Plants contain chemical substances that take part in the metabolic activities thereby helping to fight the bacterial infections. Alternatives to available antibiotics for disease management are increasingly felt due to the increase in the resistance of bacterial isolates. Awareness for misuse of antibiotics and also the potential risk of using synthetic form of phytochemicals have been reported. This has necessitated the requirement of second and third line drugs. From this study, it was evident that the extract of E. Laevis and J. regia has antimicrobial activity. This antimicrobial activity may be compared with other 'synthetic' antimicrobial agents. It may have fewer side effects as it falls in the category of natural medicine. These plant extract can be formulated in the form of a dentifrices, mouth washes, gum paints or as an intracanal medicament where an antimicrobial agent is required. The leaves of E. Laevis and bark of J. regia have been conclusive in demonstrating antimicrobial action. It may be interesting to obtain other active ingredients from the same plant or from different parts like stem, fruits etc. to assay its active ingredient and other properties and compared against each other. Natural product of higher plants may provide a new source of antimicrobial agents with possibly primordial prevention type of mechanism of Action [1] [9]. The scientific approach has confirmed the antimicrobial potential of the plant extract thus adding weight to its use as a preventive remedy for various microbial diseases of hard tissues in the oral cavity in traditional medicine. The study provides a lead molecule which can be further developed against dental caries.

CONCLUSIONS

Significant advances have been made with respect to our understanding of the molecular basis of caries and our ability to measure earliest enamel demineralization changes and thus, caries progression. Thus, efforts need to be made for the primary prevention of dental caries initiation, rather than its treatment, throughout the life. The resistance in many dental pathogens to currently used antibiotic drugs is ever increasing. The ingredients derived from Erethia laevis & Juglans regia plants used in this study are herbal, they are ecofriendly and do not produce any side effects as well as are effective and economical, as compared to the synthetic drugs. The study also confirmed the antimicrobial potentials of the plant, thus supporting its folklore application as a preventive remedy for various microbial diseases of hard and soft tissues in the oral cavity.

REFERENCES

1. Hamill F.A. Traditional herbal drugs of southern Uganda: Part III: Isolation and methods for physical characterization of bioactive alkanols from Rubus apetalus. J Ethnopharmacol. 2003;87(1): 15-19
2. Katz B.P. Huntington E. Statistical issues for combining multiple caries diagnostics for demonstrating caries efficacy. J Dent Res. 2004; 83(C): C109 - C112
3. Kamboj VP. Herbal Medicine. Current Sci 2000; 78: 35-9.
4. Gupta LM and Raina R. Side effects of some medicinal plants. Current Sci. 1988; 75: 897-900.
5. Evans M. A guide to herbal remedies. 1994 Orient Paperbacks
6. Park K.M, You J.s, Lee HY, Back NI, Hwang JK. Kuwanon G. J Ethnopharmacol. 2003; 84: 181-5.
7. Chung Jy, Choo Jh, Lee MH, Hwang JK. Phytomedicine 2006; 13: 261-6.
8. Vieira A, Marazita M and Goldstein T. J Dent Res. 2008; 87: 435
9. Motsei M, Lindsey L., Van Staden J, Jaeger K. J Ethnopharmacol. 2003; 86 (2-3): 235-241