ISSN: 2278-4632
Vol-10 Issue-5 No. 1 May 2020

ANTIMICROBIAL EFFICACY OF SELECTED MEDICINAL PLANTS AGAINST COMMON ENDODONTIC PATHOGENS

Dr.S.Jenifer,
Head and Assistant Professor,Department of biotechnology,AJK College of Arts and Science, Coimbatore, TN. India.
jsdrjsdr@gmail.com

Introduction

PARASITES

Parasites are small, midge-like flies that constitute the family Culicidae. Females of most species are ectoparasites, whose tube-like mouthparts (called a proboscis) pierce the hosts' skin to consume blood. The word "mosquito" (formed by mosca and diminutiveito). "mosquito". In passing from host to host, some transmit extremely harmful infections such as malaria, yellow fever, Chikungunya, West Nile virus, dengue fever, filariasis, Zika virus and other arboviruses, rendering it the deadliest animal family in the world **(Brown, 1993).**

Figure 1. *Azardica indica* Plant, Figure 2. *Vitex negundo* Plant, Figure 3. *Ocimum basilicum* plant

Vitex negundo commonly known as Nirgundi belongs to family verbenaceae it is used for the treatment of analgesic, Antibacterial, Anti convulsant, dengue rheumatism, dyspepsia and diarrhoea in a folkloric history it is used for cold and cough asthma,The leaf hexane extract of V.negunda has the potential to be used as an ideal eco-friendly approach for the control of the A.subpictus and

ISSN: 2278-4632
Vol-10 Issue-5 No. 1 May 2020

C.Tritaeniorhynchus by its mosquito larvicidal *activity* ***(Kamaraj et al., 2009; Kamaraj et al., 2010)***.*Ocimum basilicum* L .is an aromatic herb which is known as sabja in India. It is used to treat illness such as respiratory and rheumatic problems, vomitting and pain , It is also used as an anti-inflammatory agent, blocking both cyclooxygenase and lipooxygenase pathways of arachidonic acid metabolism. Further it possesses larvicidal and repellant potential against the dengue vector Aedesaegypti (insecta:diptera:culicidae).**Anti-microbial action** Antimicrobial screening Agar disc diffusion method this method **(Kirby Bauer *et al.*, 1966)** is suitable for organism that grows rapidly over night at 35-37°C. The antibiotic (specific concentration) impregnated disc absorbs moisture from the agar and antibiotic diffuses in to the agar medium. upports the use of the Neem seeds in traditional medicine to treat infections conditions especially those involving the eye and ear. Administration of alcoholic extract of Neem flower disrupts the estrous cycle in Sprague Dawley rats and causes a partial block in ovulation and has the potential of an ideal antifertility agent **(Gbotolorun *et al.*, 2008)**. The great potential Neem aqueous extract as powerful chemotherapeutic and viral agent **(Hassan Amer *et al.*, 2010).** The purpose of the present study was to investigate the antimicrobial activity of Neem leaves against human pathogenic bacteria, including Escherichia coli, Pseudomonas aeruginosa, Salmonella typhimurium, Staphylococcus aureus.*Ocimum sanctum*, popularly known as Tulsi is a time-tested premier medicinal herb that is used in ayurvedic medicine since ancient times. various concentrations of selected medicinal plants like *A. Indica* (Neem), *O. sanctum* (Tulsi), *Vitex negundo(Notchi)*, CHX against common endodontic pathogens such as *Staphylococcus aureus*, Escherichia coli, Pseudomonas aeruginosa.

PHYTOCHEMICAL SCREENING

Plant-derived substances have recently become of great interest owing to their versatile applications. Medicinal plants are the richest bio-resource of drugs of traditional systems of medicine, modern medicines, nutraceuticals, food supplements, folk medicines, pharmaceutical intermediates and chemical entities for synthetic drugs. Extraction (as the term is pharmaceutically used) is the separation of medicinally active portions of plant (and

animal) tissues using selective solvents through standard procedures. The products so obtained from plants are relatively complex mixtures of metabolites, in liquid or semisolid state or (after removing the solvent) in dry powder form, and are intended for oral or external use. These include classes of preparations known as decoctions, infusions, fluid extracts, tinctures, pilular (semisolid) extracts or powdered extracts. Such preparations have been popularly called galenicals, named after Galen, the second century Greek physician. Extraction methods used pharmaceutically involves the separation of medicinally active portions of plant tissues from the inactive/inert components by using selective solvents. During extraction, solvents diffuse into the solid plant material and solubilize compounds with

ISSN: 2278-4632
Vol-10 Issue-5 No. 1 May 2020

similar polarity. The purpose of standardized extraction procedures for crude drugs (medicinal plant parts) is to attain the therapeutically desired portions and to eliminate unwanted material by treatment with a selective solvent known as menstrum. The extract thus obtained, after standardization, may be used as medicinal agent as such in the form of tinctures or fluid extracts or further processed to be incorporated in any dosage form such as tablets and capsules. objectives of the study To determine the preliminary phyto chemical screening, Evaluate of Antimicrobial activity and potential Mosquitoes Repellent preparation from *Azardica indica, Vitex negundo, Ocimum basilicum.* To determine the preliminary Phytochemical screening. To evaluate the anti-microbial potential to prepare the Plant for Mosquito Repellant. To evaluate the Mosquito Repellant.

MATERIALS AND METHODS

COLLECTION OF PLANT MATERIAL

Considering the importance of the herbal medicines *Azardica indica* (Neem), *Vitex negundo* (Notchi), *Oscimum basilicum* (Basil leaves), was selected for the present investigation.

Microorganisms and Culture Media

Bacterial culture plays a major and vital role in the research of the antimicrobial activities. The bacterial organisms here are

1. *Staphylococcus aureus*
2. *Klebsiella pneumonia*
3. *Escherichia coli*
4. *Pseudomonas aeruginosa.*

The microbial species were obtained from the Bioline laborotory. The Bacterial strains were maintained strains were maintained on nutrient agar slants and were sub cultured on nutrient agar medium at regular intervals of 15 days.

Innoculum preparation

Bacterial cultures were subcultured in nutrient broth at 37c for 18hrs and used for the experiment.

Preparation of Culture Media

Nutrient broth

The nutrient broth medium consists of following composition:

Ingredients		**Grams/Litre**
Peptone	:	5gm
Beef extract	:	3gm
Sodium chloride	:	5gm
Yeast extract	:	1.5gm

ISSN: 2278-4632
Vol-10 Issue-5 No. 1 May 2020

Distilled water	:	1000ml
P^H	:	7.0

Nutrient agar medium

Nutrient agar medium is one of the most commonly used medium for several routine bacteriological purposes.

Ingredients	**:**	**Grams/Litre**
Peptone	:	0.5 gm
Beef extract	:	0.3 gm
Agar	:	15 gm
Sodium chloride	:	05 gm
Yeast extract	:	1.5gm
Distilled water	:	1000ml
P^H	:	7.0

PREPARATION OF THE EXTRACT

The leaves of *Azardica indica , Vitex negundo, Ocimum basilicum* were washed with distilled water and shadow dried for 3-5 weeks, Then it was cut into small pieces, powdered with electrical blender and stored for future use at 4c.

Tests for Saponins

5 ml of aqueous extract was shaken vigorously with 5 ml of distilled water in a test tube and warmed. The formation of stable foam was taken as an indication of the presence of saponins.

Tests for Flavonoids

To 1 ml of aqueous extract, 1 ml of 10% lead acetate solution was added. The formation of a yellow precipitate was taken as a positive test for flavonoids.

Test for terpenoids

2ml of the organic extract was dissolved in 2 ml of chloroform and evaporated to dryness. 2 ml of concentrated sulphuric acid was then added and heated for about 2 min. Development of a greyish colour indicates the presence of terpenoids.

Test for Phenols

A small quantity of the extract was treated with 1% aqueous or alcoholic ferric chloride solution. Formation of green, purple, blue, or black indicates the presence of phenol. A small quantity of the extract was treated with aqueous mixture 1% ferric chloride and 1% pottassium ferricyanide. Appearance of green or purple or blue colour shows the presence of phenols.

ISSN: 2278-4632
Vol-10 Issue-5 No. 1 May 2020

FORMULATION OF CAKES USING DIFFERENT NATURAL BINDERS:

The different natural binders (1000g each) were purchased commericial from local vendors, different natural binders used were carbon, wood powder, kungulian powder, yara yara, jasmine powder, varada, Razanam, sambirani, Thalambu essence scent.

PREPARATION OF NEEM, THULASI, NOTCHI

Cut leaves pieces were grounded into paste use electrical grinder by add distilled water. The 15g of leaves were mixed with natural binders.Then the pasted leaves with natural binders was plated accordingly wet weights of carbon were taken.For the determination of dry weight cakes were allowed to dry in sun for 36 hrs and dry weight was taken.

EVALUATION OF MOSQUITO REPELLENT ACTIVITY:

For investigating Mosquito repellent activity the prepared cakes were checked for its repellent activity. Flammability test of these cakes were conducted to check its consistent combustibility. Further the time taken to burn the cake, Smoke produced and its causal effect such as irritation, coughing, tears were burnt in selected mosquito prone areas in the evening and the night period such as houses, Department premises laboratoy corners, dark rooms.

ANTI MICROBIAL ACTIVITY

Disc diffusion method for antimicrobial susceptibility testing was standard method by Bauer *et al* to assess the presence of antibacterial activities of the plant extracts. A bacteria culture was used to nutrient agar medium (18) plates evenly using a sterile swab. The plates were dried for 15 minutes and used for the sensitivity test. The discs impregnated with a series of plant extracts were placed on the nutrient agar surface. Each test plates comprises of the discs. One positive control, which is a standard commercial antibiotic disc, and five treated discs. The standard antibiotic discs were streptomycin 20micro gram. The negative control was DMSO (100٪). Besides the controls, each plate had five treated discs placed about equidistance to each other. The plate was then incubated 37c for 18 to 24 hrs depending upon species of bacteria used in the test. After the incubation, the plates were examined for inhibition zone. The inhibition zones were then measured using calipers and recorded. The tests were repeated three times to ensure reliability.

RESULT& DISCUSSION

Phytochemical Screening

Phytochemical evaluation of various leaves extracts of the plants were done for the presence of Alkaloids, Flavonoids, Saponins, Terpenoids, and phenol the result are presented in Table 1.

Antimicrobial activity

The antimicrobial activity was examined by disc diffusion method. The aqueous extract of different plants leaves exhibited potent antimicrobial activity towards all the microbes. The zones of

ISSN: 2278-4632
Vol-10 Issue-5 No. 1 May 2020

inhibition values were presented in Table 2, 3, 4, 5. *P. aerogonsa, K.pneumonia* were found to be more susceptible towards the aqueous extract of leaves with a maximum inhibitory zone.

Table 1: Phytochemical analysis of aqueous extract

Phytochemical Constituents	***Azardica Indica***	***Vitex Negundo***	***Ocimum basilicum***
Phenol	+	+	+
Alkanoids	-	-	+
Saponin	-	+	+
Flavonaids	+	+	-
Steriods	-	-	+
Terpenoids	+	-	-

Antibacterial activity of *A. indica*

Table 2: Antibacterial activity of *A. indica*

S. No	**Organisms Name**	**Zone of inhibition (mm)**					
		Plant extracts					
		Control	**Streptomycin 50µg**	**25µg**	**50µg**	**75µg**	**100µg**
1	*E. coli*	-	19	12	14	17	19
2	*S. aureus*	-	24	14	13	14	14
3	*P.aerogonsa*	-	27	10	12	13	15
4	*K. pneumonia*	-	29	15	16	16	19

ISSN: 2278-4632
Vol-10 Issue-5 No. 1 May 2020

Table 3:Antibacterial activity of *Vitex negundo*

S. No	Organisms Name	Zone of inhibition (mm)					
				Plant extracts			
		Control	Streptomycin 50µg	25µg	50µg	75µg	100µg
1	*E. coli*	-	19	12	14	17	19
2	*S. aureus*	-	24	14	13	14	14
3	*P.aerogonsa*	-	27	10	12	13	15
4	*K. pneumonia*	-	29	15	16	16	19

Table 3:Antibacterial activity of *Ocimum basilicum*

S. No	Organisms Name	Zone of inhibition (mm)					
				Plant Extracts			
		Control	Streptomycin 50 µg	25µg	50µg	75µg	100µg
1	*E. coli*	-	19	10	13	17	26
2	*S. aureus*	-	24	11	16	19	22
3	*P.aerogonsa*	-	27	11	14	21	25
4	*K. pneumonia*	-	29	10	15	22	27

REPELLENT ACTIVITY

Flammability test and burning time

To observe the flammability of the cakes, the cakes were burnt. The time taken to burn the care

ISSN: 2278-4632
Vol-10 Issue-5 No. 1 May 2020

completely and the total time of repellence were recorded Table 6. For a good and consistently burning mosquito repellent cake.

Table. 6:showing effectiveness of each sample prepared in different combinations with respect to the combustion time and reoccurrence of mosquitoes in the study site

S.No	Trial Sample Composition	Sample codes	Time of combustion	Period of effectiveness			
				1 Hours	2 Hours	3 Hours	4 Hours
1	1:1:1:1	A	15 min	+	+	+	+
2	2:1:1:2	B	12 min	+	+	_	–
3	1:2:2:1	C	20 min	+	_	_	–
4	1:2:3:4	D	17 min	+	_	_	–

Mosquito repellence test

Mosquito repellence test was dose by simply selecting the mosquito prone areas in the evening and night period such as laboratory corners, bushes and shrubs in and different areas Buildings and mess hall.

Table 7: Mosquito repellence test in different areas

S,No	Areas	Reports given by people	Remarks
1	Laboratory corners	Smoke with good smell, No irritation removed Mosquitoes from room	Mosquitoes are repelled
2	Ramanathapuram Coimbatore	Smoke does not cause irritation, Mosquitoes escaped	Mosquitoes are repelled
3	Ukkadam Coimbatore	No irritation, Mosquitoes escaped	Mosquitoes are repelled

ISSN: 2278-4632
Vol-10 Issue-5 No. 1 May 2020

Figure 10: Preparation of Repellent cakes


Repellent Stick mode Packed Product

Azardica indica-Plants extracts potential sources novel antimicrobial activity compounds especially against bacterial pathogens. In vitro studies in this work showed that the plant extracts inhibited bacterial growth but their effectiveness varied.The medicinal values of the secondary metabolites are due to the presence of chemical substances that produce a definite physiological action on the human .Solmonella sp which infects a human and animal species **(Furowicz and Terzolo**) against found to effective, These plant extract also inhibited the fungal growth, Its cure the skin diseases,inflammation, white patches, it has anti-malarial activity and Mosquito repellent.***Vitex negundo***-By disc diffusion method and Streptococcus mutants express a very clear indication of inhibitory activity and they show wide spectrum of inhibitory activity and they show wide spectrum inhibition to all the extracts. Ecoli and streptococcus mutants were resistant to all the extracts. *Klebsiella pneumonia,Vibrio cholerae, streptococcus mutants, Eschericia coli* have inhibitory activity next only to the *Salmonella paratyphi*. These organisms very sequentially given using their sensitivity pattern to the the extracts.***Ocimum basilicum***The secondary metabolites exert antimicrobial activity through different mechanisms. Tannins have been found to form irreversible complexes with proline rich proteins **(Shimada,2006)** resulting in the inhibition of cell protein to provide to typical tanning effect which is important for the treatment of inflamed or ulcerated tissue).

CONCLUSION

Our current studies, the phytochemical screening of extracts of *Azardica indica, Vitex negundo, Oscimum basilicum.* These plant powder residents excellently inhibit parasites. The report is the First work done using repellent cakes as 3 herbal plant Parasite repellents using natural binders. Antimicrobial Origins from plants & detect its ability to treat illnesses caused by unaffected micro-organisms is wanted. Our consequences of the study of these plants presented its aptitude to be a new source of

ISSN: 2278-4632
Vol-10 Issue-5 No. 1 May 2020

natural products brings to the culture with new thoughts escape the Parasites.