



Research Article

www.ijrap.net



PREPARATION OF ANTIBACTERIAL DISCS FROM AYURVEDIC DRUGS

Mallika KJ^{1*}, Shashirekha KS²

¹Professor, Department of Samhita and Siddhanta, Shri Dharmasthala Manjunatheshwara College of Ayurveda and Hospital Hassan, Karnataka, India

²Microbiologist, Department of Roganidana, Shri Dharmasthala Manjunatheshwara College of Ayurveda and Hospital Hassan, Karnataka, India

Received on: 27/04/18 Accepted on: 24/07/18

*Corresponding author

E-mail: drmallikakj@gmail.com

DOI: 10.7897/2277-4343.094102

ABSTRACT

In clinical practice of Ayurveda various compound preparations are in use to treat infectious diseases, out of which few single herb powders and compound preparations were tested against human pathogenic bacteria like *Escherichia coli*, *Proteus vulgaris*, *Enterococcus faecalis*, *Staphylococcus aureus*, *Streptococcus pyogenes*, *Pneumococcal pneumonia* isolated from human body fluids. Antimicrobial susceptibility of these bacteria against Ampicillin (10mcg) antibiotics was conducted to determine antibiotic pattern in all the isolates. It was observed that the Nimbadiguggulu, Kaishoraguggulu, Arogyavardhini, Chandraprabhavati, Bilvadigulika, Kadhiradivati, Triphalaguggulu, Lakshadiguggulu; and churna of Chandana, Maricha, Harithaki, Jathiphala, Pippali, Shunti, Raktachandana, Madhuyasti, Guduchi, Vacha, Vidanga, Chitraka shown significant susceptibility against the standard drug Ampicillin (10mcg). These observations support the use of these drugs in preventing as well as in controlling the infections in clinical practices of Ayurveda.

Keywords: Churna, Vati, Antibiotic, Susceptibility, Herbal Disc

INTRODUCTION

Ayurveda explains microorganisms as external agents for the causation of diseases. However, the explanations are rudimentary and scattered throughout the Ayurveda literature. However, there is elaborate description of treatment protocol for various infectious diseases like vidradi (abscess), vrana (wound) etc in the texts, which highlights that Ayurveda was well equipped to face such diseases. With this background in this research antimicrobial discs were prepared with well-known and frequently used medicines in Ayurveda practice¹. Treatment of microbes were emphasised for strengthening the internal environment there by to prevent from getting disease and to fight against the external causes. The benefits of using plant-derived medicine are relatively safer than synthetic drugs. As these medicines have diverse range of bioactive molecules and play an important role in the maintenance of health.² Adding these formulation as co medication may increase the antimicrobial spectrum and action potency. Recently, it has been demonstrated that many human pathogenic bacteria have developed resistance against several synthetic drugs. So, it is time to find alternatives with Ayurveda formulations or with single drugs. Microbiologists and other researchers had many successes in learning how microbes cause certain infectious diseases and how to combat those microbes. Unfortunately, microbes are much better at adapting to new environments than people. Having existed on Earth for billions of years, microbes are constantly challenging human newcomers with ingenious new survival tactics. Many microbes are developing new properties to resist drug treatments that once effectively destroyed them. Drug resistance has become a serious problem worldwide. Changes in the environment have put certain human populations in contact with newly identified microbes that cause diseases have never seen before, or that previously occurred only in isolated populations. Time has come to find effective remedy to the present scenario. Hence an attempt

was made to screen the antibacterial potential of herbal preparations in the control and prevention of enteric bacterial infections with the objective of preparing the antimicrobial disc from Ayurveda drugs and determining the antimicrobial susceptibility of Ayurveda Drugs (Vati and Churna) against various pathogenic Bacterial species.

MATERIALS AND METHODS

Collection of sample: *Escherichia coli*, *Proteus vulgaris*, *Enterococcus faecalis*, *Staphylococcus aureus*, *Streptococcus pyogenes*, *Pneumococcal pneumonia*, isolated from pus and urine samples were collected from IPD and OPD patients of Sri Dharmasthala Manjunatheshwara college of Ayurveda—and hospital B M Road, Hassan, India.

Collection drug: It was procured from Sri Dharmasthala Manjunatheshwara Ayurveda pharmacy (GMP-ISO 9001:2008 Certified) Lakshminarayana Nagar, Udupi. Karnataka, India.

Ethical clearance was obtained for carrying out this research No: SDMCAH/IEC/104/12-13 and study was carried out as per the good ethical practice guidelines

Materials used: Mac Conkey agar, Mueller Hinton Agar, Sterile swabs, what's man Number 1 filter paper, Distilled water, weighing balance, pestle and Mortar, Punching machine Standard antibiotic disc (Ampicillin-10µg), ethanol, forceps, 0.5 McFarland test standard McFarland reference card, Sterilized vials Hot air oven.

Preparation of Ayurvedic disc cartridges

Ayurveda discs were prepared from 10gms of drugs. The compound formulations used for the purpose were Nimbadi

guggulu, Kaishora guggulu, Arogyavardhini, Chandraprabha vati, Bilvadi gulika, Kadhiraadi vati, Triphala guggulu, and Lakshadi vati and the powders used were Chandana, Maricha, Harithaki, Jathiphala, Pippali, Shunti, Raktachandana, Madhuyasti, Guduchi, Vacha, Vidanga, and Chitraka.

Discs of 10mm Diameter were prepared by punching what's man No 1 filter paper and preserved in the sterilized vials. Under the sterilized condition 2 gm of drug was weighed with the help of electronic weighing balance and made into fine powder with help of pestle and mortar. By adding 8 ml of distilled water the fine powder of drugs was made into a thick paste so that it could firmly stick on what's man no 1 filter paper discs. In a hot air oven, the discs coated with drugs were placed for drying at 40-45°C for 2 hours; the prepared discs were stored in sterilized vials with label of medicine.

Antimicrobial susceptibility of Ayurveda Drugs

The Bacterial species taken for the study were from the subjects suffering from various infectious diseases like UTI, Fever, Wound infection, Gastro Intestinal Tract infection. Urine and pus sample were collected and culture was done after 24 hours of incubation. The colonies obtained were used for the study.

A loop full of colony was transferred into tube of sterile saline. Turbidity thus obtained was diluted equivalent to the 0.5 McFarland test standard. The diluted tube and the 0.5 McFarland test standards were held to-gather against the black-lined McFarland reference card to accurately rate the turbidity.

Within 15 minutes of diluting, a sterile swab was dipped into the properly adjusted inoculums, so that the suspension was slightly lifted out and swab was firmly rotated several times against wall of the tube to express excess fluid. To obtain an even inoculation entire surface of glass plate with the swab was turned with stroking in 60 degree. Then lid was Closed and waited for 3-5 minutes before applying the drug impregnated discs. Using aseptic technique by means of a dispenser discs were applied on inoculation plates such that ayurvedic drug discs and standard Antibiotic disc were side by side, then discs were pressed firmly by sterile swab to make contact with the surface. Inoculated plate was incubated in incubator at 37°C for 24 hours in an inverted position. The plates were examined after 16-24 hours incubation. Zone showing complete inhibition by gross visual inspection were measured in millimetre, by holding the ruler behind the inverted plate over a black non-reflective surface. Results obtained were tabulated (Table 1,2).

RESULTS

The Vati and churna that were studied shown zone of inhibition against both gram positive and gram negative microorganisms (Figure 1). The test organisms were isolated from the patients suffering from various infectious diseases like UTI, Fever, Wound infection, Gastro Intestinal Tract infection. Isolated organisms include *Escherichia coli*, *Proteus vulgaris*, *Enterococcus faecalis*, *Staphylococcus aureus*, and *Streptococcus pyogenes* and *Pneumococcal pneumonia*. The standard drug used as control was Ampicillin (10mcg).

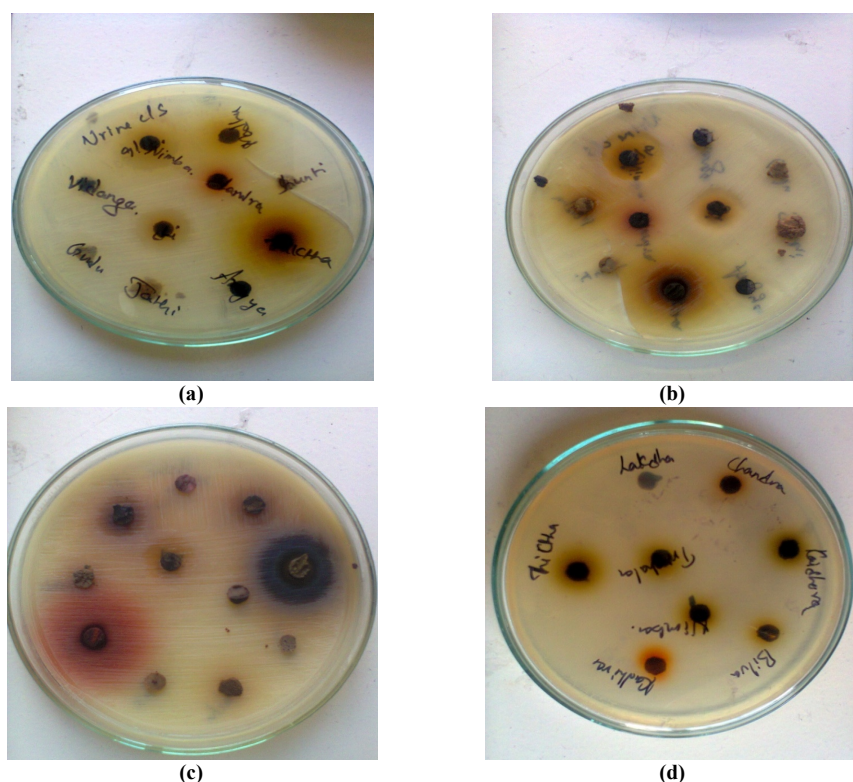


Figure 1: (a-d) Nutrient Agar plates of antimicrobial disc showing zone of inhibition

Table 1: Zone of inhibition of different vati and churna against bacterial species

Ayurvedic drug	Organisms											
	<i>Escherichia coli</i>				<i>Proteus vulgaris</i>				<i>Enterococcus faecalis</i>			
	S	Ms	R	Std	S	Ms	R	Std	S	Ms	R	Std
Nimbadi guggulu	22	-	-	22	-	18	-	17	-	16	-	17
Kaishora guggulu	26	-	-	22	24	-	-	17	20	-	-	17
Arogyavardhini vati	-	18	-	22	-	-	14	17	-	18	-	17
Chandraprabha vati	-	18	-	22	-	16	-	17	-	18	-	17
Bilvadi gulika	-	16	-	22	-	18	-	17	20	-	-	17
Kadhiradi vati	20	-	-	22	20	-	-	17	-	18	-	17
Triphala guggulu	-	18	-	22	-	18	-	17	22	-	-	17
Lakshadi guggulu	-	-	12	22	-	-	10	17	-	-	12	17
Chandana churna	-	-	10	22	-	18	-	17	-	16	-	17
Maricha,	-	18	-	22	-	-	10	17	-	-	08	17
Harithaki	-	18	-	22	-	16	-	17	-	18	-	17
Jathiphala	-	-	10	22	-	16	-	17	-	16	-	17
Pippali	-	16	6	22	-	18	-	17	-	-	10	17
Shunti	-	-	08	22	-	-	08	17	-	14	-	17
Raktachandana	-	14	-	22	-	18	-	17	-	-	08	17
Guduchi	-	16	-	22	-	16	-	17	-	-	08	17
Vacha	-	-	12	22	-	-	10	17	-	-	10	17
Vidanga	20	-	-	22	-	18	-	17	-	18	-	17
Chitraka churna	20	-	-	22	-	18	-	17	20	-	-	17

(S- Sensitive, MS-Moderately sensitive, R-Resistance, Diameter of Zone of Inhibition in mm)

Table 2: Zone of inhibition of different vati and churna against bacterial species

Ayurvedic drug	Organisms											
	<i>Staphylococcus aureus</i>				<i>Streptococcus pyogens</i>				<i>Pneumococcal pneumonia</i>			
	S	Ms	R	Std	S	Ms	R	Std	S	Ms	R	Std
Nimbadi guggulu	22	-	-	28	22	-	-	-	20	-	-	26
Kaishora guggulu	-	-	08	28	-	16	-	-	20	-	-	26
Arogyavardhini vati	-	-	08	28	-	-	08	-	-	16	-	26
Chandraprabha vati	-	-	08	28	-	-	10	-	-	-	08	26
Bilvadi gulika	22	-	-	28	-	18	-	-	-	16	-	26
Kadhiradi vati	22	-	-	28	24	-	-	-	-	18	-	26
Triphala guggulu	24	-	-	28	22	-	-	-	22	-	-	26
Lakshadi vati	-	-	08	28	-	-	08	-	-	10	-	26
Chandana churna	-	-	10	28	-	-	08	-	-	-	08	26
Maricha,	-	16	-	28	20	-	-	-	-	18	-	26
Harithaki	-	-	08	28	-	18	-	-	-	18	-	26
Jathiphala	-	-	08	28	-	-	10	-	-	18	-	26
Pippali	-	-	08	28	-	16	-	-	-	18	-	26
Shunti	20	-	-	28	-	16	-	-	-	16	-	26
Raktachandana	-	18	-	28	-	-	10	-	-	-	10	26
Guduchi	-	-	08	28	-	16	-	-	-	16	-	26
Vacha	-	-	10	28	-	16	-	-	-	18	-	26
Vidanga	24	-	-	28	22	-	-	-	22	-	-	26
Chitrakachurna	24	-	-	28	20	-	-	-	20	-	-	26

(S- Sensitive, MS-Moderately sensitive, R-Resistance. Diameter of Zone of Inhibition in mm)

DISCUSSION

In the present study Nimbadi guggulu had shown 22mm of zone of inhibition for *Escherichia coli* (Std-22), 18mm for *Proteus vulgaris* (Std-17), 16mm for *Enterococcus faecalis* (Std-17), 20mm for *Staphylococcus aureus* (Std-28), 22mm for *Streptococcus pyogens*, and 20mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). Previous works on Leaf and bark of Nimba (*Azadirachta indica*) showed inhibition zone against *Vibrio cholerae* and *Bacillus subtilis*, while *Escherichia coli* and *S.typhi* are less susceptible to neem extract^{7,8}. Nimbadi guggulu as a combination had shown sensitivity towards *Escherichia coli* this may be due to the combined action of drugs in the compound than independent action of the Nimba (*Azadirachta indica*).

Kaishora guggulu⁹ used in various skin manifestation had shown better Zone of inhibitions for *Escherichia coli* (Std-22), 20mm for

Proteus vulgaris (Std-17), 18mm for *Enterococcus faecalis* (Std-17), 22mm for *Staphylococcus aureus* (Std-28), 24mm for *Streptococcus pyogens*, and 18mm for *Pneumococcal pneumonia* (Std-26). kaishore guggulu has Triphala and guduchi (*Tinospora cordifolia*) as the main ingredient. Yogesh S. Biradar et al found that Triphala inhibits the strains *Escherichia coli*, *Staphylococcus aureus*, and other bacteria. According to their published report, the aqueous and ethanol extracts of Triphala and Triphala Mashī exhibited a broad-spectrum antimicrobial activity against all the microorganisms from human secretions and from pathology lab with prior diagnosis.¹⁰ Guduchi (*Tinospora cordifolia*) found protective in rats against mixed bacterial abdominal sepsis¹¹ and in mice against *Escherichia coli* induced peritonitis¹².

Arogyavardhini Vati had shown 18mm of zone of inhibition for *Escherichia coli* (Std-22), 14 mm for *Proteus vulgaris* (Std-17), 18mm for *Enterococcus faecalis* (Std-17), 08mm for *Staphylococcus aureus* (Std-28), 08mm for *Streptococcus*

pyogens, and 16mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). Physico-chemical analysis and evaluation of antibacterial and antifungal activity of 5ml of 100% Arogyavardhini vati solution conducted by Savanur et.al showed 30mm of Zone of inhibition for *Staphylococcus aureus* and 28mm for *Escherichia coli* against the standard control Benzathine penicillin. other study it was observed that Arogyavardhini vati was having antibacterial activity against both gram positive and gram negative bacterial species¹³. *Picrorhiza kurroa* is the main ingredient of arogyavardhini proved for its antimicrobial activities.¹⁴

Chandraprabha vati which was in use for diseases of urinary tract, had shown 18mm of zone of inhibition for *Escherichia coli* (Std-22), 16mm for *Proteus vulgaris* (Std-17), 18mm for *Enterococcus faecalis* (Std-17), 08mm for *Staphylococcus aureus* (Std-28), 10mm for *Streptococcus pyogens*, and 08mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). Shilajit is one of the ingredients of this compound; the anti-microbial study of shilajit showed that among gram positive organism; *Streptococcus pneumonia* and *B.subtilis* showed greater zone of inhibition (20mm). While *Staphylococcus aureus* showed a zone of (17mm) and *S.saprophyticus* showed a zone of (15mm). Among gram negative organism, *Shigella dysenteriae* and *Salmonella paratyphi A* showed greater zone of (17mm) inhibition. *Escherichia coli* showed a zone of (16mm), *P.florescense* and *Klebsiella oxytoca* showed a zone of (16mm), *Salmonella typhi* showed a zone of (14mm) and *Citrobacter* showed a zone of (13mm)¹⁵.

Bilvadhigulika had shown 16mm of zone of inhibition for *Escherichia coli* (Std-22), 18mm for *Proteus vulgaris* (Std-17), 20mm for *Enterococcus faecalis* (Std-17), 22mm for *Staphylococcus aureus* (Std-28), 18mm for *Streptococcus pyogens*, and 16mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). Bilvadhigulika which is commonly used in the treatment of poison and skin manifestations;¹⁶the crude ethanolic extract of Bilva (*Aegle marmelos*) had proved antimicrobial activity against gram positive and negative at different concentrations. It was observed that at 2.5 mg/ml concentration, it exhibits about 25.7mm of zone inhibition for *Escherichia coli*, 19.9mm for *Pseudomonas aeruginosa*; gram positive *Staphylococcus aureus* 29.0 mm; and *Bacillus subtilis*, a maximum zone of inhibition about 28.1 mm was recorded but *Staphylococcus aureus* doesn't exhibit zone of inhibition¹⁷. Even fixed oil from seeds of *Aegle marmelos* and its unsaponifiable portion were also found to be active against various strains of gram positive and gram negative bacteria¹⁸.

Khadiradvati which is having khadira (*Acacia catechu*) as ingredient had shown 20mm sensitivity for *Escherichia coli* (Std-22), 20mm for *Proteus vulgaris* (Std-17), 18mm for *Enterococcus faecalis* (Std-17), 22mm for *Staphylococcus aureus* (Std-28), 24mm for *Streptococcus pyogens*, and 18mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). In a study of the ethanolic bark extract of *Acacia catechu* Wild it was found more effective against *Staphylococcus aureus*, *Shigella dysenteriae* and *Escherichia coli* with a zone of inhibition of 22mm, 20mm and 19 mm diameter (at conc 300 µg.) respectively¹⁹.

In the present study Triphala guggulu had shown 18mm of zone of inhibition for *Escherichia coli* (Std-22), 18mm for *Proteus vulgaris* (Std-17), 22mm for *Enterococcus faecalis* (Std-17), 24mm for *Staphylococcus aureus* (Std-28), 22mm for *Streptococcus pyogens*, and 22mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). This compound formulation is a combination of triphala and guggulu as its ingredient²⁰. Rani

and Khullar et al screened for plants to treat enteric diseases and reported antibacterial activity of triphala against multidrug resistant enteric *Salmonellae typhi*; Tambekar and Saratkar et al showed that *Terminalia bellirica* and *Terminalia chebula* were strong antimicrobial agents against enteric pathogens whereas *Emblica officinalis* was moderate²¹; Panthi and Chaudhary recorded potent antibacterial activity against *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Escherichia coli* by *Terminalia chebula* (Haritaki)²²; Jagtap and Karkera reported that the extract of *Terminalia chebula* inhibited the salivary bacteria and potential as an anti-caries agent²³.

Lakshadhi vati had shown 12mm of zone of inhibition for *Escherichia coli* (Std-22), 10mm for *Proteus vulgaris* (Std-17), 12mm for *Enterococcus faecalis* (Std-17), 08mm for *Staphylococcus aureus* (Std-28), 08mm for *Streptococcus pyogens*, and 10mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). Laksha (*Laccifer lacca*), Asthisamhrut (*Cissus quadrangularis*), kakubha (*Terminalia arjuna*), Ashwagandha (*Withania somnifera*), Nagabala (*Grewia populifolia*), Guggulu (Purified *Commiphora mukul*) are the ingredient of lakshadi guggulu²⁴. Acetone extract of *Laccifer lacca* showed antimicrobial activity against five microbes i.e. *Escherichia coli*, *Staphylococcus aureus*, *Bacillus subtilis*, *Xanthomon ascitri* and *Erwinia carotovora* the highest being against *Escherichia coli*²⁵. The ethyl acetate extract and methanol extract of both fresh and dry stem of *Cissus quadrangularis* exhibited antimicrobial activity against gram-positive bacteria including *Bacillus subtilis*, *Bacillus cereus*, *Staphylococcus aureus* and *Streptococcus* species²⁶. This preparation is used in fracture healing; anti-microbial activity supports the usage of drug even to protect the wound by infection²⁷.

All the compound formulation studied indicated those formulations can be a choice in treating the diseases with microbial manifestations.

Single Herbs (churna)

Maricha (*Piper nigrum* Linn) had shown 18mm of zone of inhibition for *Escherichia coli* (Std-22), 10mm for *Proteus vulgaris* (Std-17), 08mm for *Enterococcus faecalis* (Std-17), 16mm for *Staphylococcus aureus* (Std-28), 20mm for *Streptococcus pyogens*, and 18mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). The acetone extract of *Piper nigrum* Linn (Maricha) was strong antibacterial against *S. epidermidis* and *Staphylococcus aureus*, while moderate against *Proteus vulgaris*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Escherichia coli*, *Klebsiella pneumoniae*, *S. typhimurium* and *Enterobacter aerogenes*.

Shunti (*Zingiber officinale*) had shown 08mm of zone of inhibition for *Escherichia coli* (Std-22), 08mm for *Proteus vulgaris* (Std-17), 14mm for *Enterococcus faecalis* (Std-17), 20mm for *Staphylococcus aureus* (Std-28), 18mm for *Streptococcus pyogens*, and 16mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). *Zingiber officinale* has been showed to have antimicrobial activity *Pseudomonas aeruginosa*, *Escherichia coli*, *Klebsiella pneumoniae*, *S. typhimurium*, whereas aqueous and ethanolic extract was mild antibacterial against *S. epidermidis*, *Proteus vulgaris*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Escherichia coli*, *Klebsiella pneumoniae*, *Enterobacter aerogenes*.

Pippali churna (*Piper longum*) had shown 16mm of zone of inhibition for *Escherichia coli* (Std-22), 18mm for *P.vulgaries* (Std-17), 10mm for *Enterococcus faecalis* (Std-17), 08mm for *Staphylococcus aureus* (Std-28), 16mm for *Streptococcus*

pyogens, and 18mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). Methanol extract of *Piper longum* (pippali) was strong antibacterial against *S. epidermidis* while acetone extract was strong antibacterial against *S. epidermidis*, *Staphylococcus aureus*, *Escherichia coli* and *Enterobacter aerogenes*.²⁸

Ethanol extract of Trikatu churna showed moderate antibacterial against all the bacterial pathogens while methanol extract was strong antibacterial against *S. epidermidis* and *Staphylococcus aureus*. It is the common ingredient in most of the Ayurveda formulation as it increases the bioavailability of drug. However along with its anti-microbial action enhances its utility in various diseases.

Chitraka churna (*Plumbago zeylanica*) had shown 20mm of zone of inhibition for *Escherichia coli* (Std-22), 18mm for *Proteus vulgaris* (Std-17), 20mm for *Enterococcus faecalis* (Std-17), 24mm for *Staphylococcus aureus* (Std-28), 20mm for *Streptococcus pyogens*, and 20mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). A study revealed that root extracts of chitraka possess potential antibacterial activity against *Bacillus subtilis*, *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Proteus vulgaris* and highest against *Helicobacter pylori*²⁹. Chitraka known for its tikshna and ushna guna is commonly used in gastrointestinal tract manifestations.

Chandana (*Santalum album*) had shown 10mm of zone of inhibition for *Escherichia coli* (Std-22), 18mm for *Proteus vulgaris* (Std-17), 16mm for *Enterococcus faecalis* (Std-17), 10mm for *Staphylococcus aureus* (Std-28), 08mm for *Streptococcus pyogens*, and 08mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). The study revealed that *Santala album* has antibacterial activity in leaf, bark and seed methanol extract against *Bacillus subtilis*, *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Escherichia coli*³⁰.

Rakthachandana (*Pterocarpus santalinus*) had shown 14mm of zone of inhibition for *Escherichia coli* (Std-22), 18mm for *Proteus vulgaris* (Std-17), 08mm for *Enterococcus faecalis* (Std-17), 18mm for *Staphylococcus aureus* (Std-28), 10mm for *Streptococcus pyogens*, and 10mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). Other study showed Raktachandana (*Pterocarpus santalinus*) Stem bark extract having maximum activity against *Enterobacter aerogenes* (21.5 mm), *Alcaligenes faecalis* (20.0 mm), *Escherichia coli* (18.5 mm), *Pseudomonas aeruginosa* (18.0 mm), *Proteus vulgaris* (18.0 mm), *Bacillus cereus* (17.5 mm), *Bacillus subtilis* (17.0 mm) and *Staphylococcus aureus* (16.5 mm), whereas the leaf extract showed maximum activity against *Escherichia coli* (19.5 mm), *Alcaligenes faecalis* (18.0 mm), *Enterobacter aerogenes* (17.5 mm) and *Pseudomonas aeruginosa* (16.5 mm)³¹.

Vidanga (*Embelia ribes*) had shown 20mm of zone of inhibition for *Escherichia coli* (Std-22), 18mm for *Proteus vulgaris* (Std-17), 18mm for *Enterococcus faecalis* (Std-17), 24mm for *Staphylococcus aureus* (Std-28), 22mm for *Streptococcus pyogens*, and 22mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). Embelin, the principal constituent of the fruit of *Embelia ribes* was proved to have antibacterial property against both gram positive and gram-negative bacteria. Even at very high doses (100 mg/disc) embelin did not exhibit any antibacterial property on *Escherichia coli* and *Klebsiella pneumoniae*. At 25 mg/disc dose the zone of inhibition is not observed when experimented, at high doses (50 mg/disc and 100 mg/disc) mild antibacterial property was observed on *Streptococcus faecalis*, *Salmonella typhi* and *Vibrio cholerae* organisms³², other study revealed aqueous and ethanol extracts of

Embelia ribes exhibited significant antibacterial activity against gram-positive and gram-negative strains with minimum inhibitory concentration (MIC) ranging from 1.5 to 100 mg/ml. The most susceptible organism to the ethanolic extract was *Bacillus subtilis* and *Pseudomonas aeruginosa*³³.

Jathiphala (*Myristica fragrans*) had shown 10mm of zone of inhibition for *Escherichia coli* (Std-22), 16mm for *Proteus vulgaris* (Std-17), 16mm for *Enterococcus faecalis* (Std-17), 08mm for *Staphylococcus aureus* (Std-28), 10mm for *Streptococcus pyogens*, and 18mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). Various extracts and the essential oil of nutmeg seeds have presented strong antimicrobial activity against gram-positive and gram-negative bacteria, as well as a variety of fungi. Some important antimicrobial compounds reported in nutmeg seeds are a-pinene, b-pinene, p-cymene, carvacrol, and b-caryophyllene³⁴.

Vacha (*Acorus calamus*) had shown 12mm of zone of inhibition for *Escherichia coli* (Std-22), 10mm for *Proteus vulgaris* (Std-17), 10mm for *Enterococcus faecalis* (Std-17), 10mm for *Staphylococcus aureus* (Std-28), 18mm for *Streptococcus pyogens*, and 18mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). Alcohol extract of *Acorus calamus* showed antibacterial activity against *Staphylococcus aureus*, *Escherichia coli*, *Bacillus subtilis*, *Staphylococcus citreus*, *Bacillus megaterium*, *Salmonella paratyphi*. A and B, *Salmonella marcescens*, *Proteus vulgaris* and *Shigella dysomei*³⁵.

Guduchi (*Tinospora cordifolia*) had shown 16mm of zone of inhibition for *Escherichia coli* (Std-22), 16mm for *Proteus vulgaris* (Std-17), 08mm for *Enterococcus faecalis* (Std-17), 08mm for *Staphylococcus aureus* (Std-28), 16mm for *Streptococcus pyogens*, and 16mm of zone of inhibition for *Pneumococcal pneumonia* (Std-26). Maximum antibacterial activity was observed against *S. sanguinis* (23 mm) and lowest antimicrobial activity against *S. salivarius* (17 mm) by agar well diffusion³⁶.

CONCLUSION

The present study revealed that single herb powder and compound formulations were showing sensitivity against pathogenic bacteria which have been isolated from human samples. In compound formulation Nimbadi guggulu and in single herb powder chitraka (*Plumbago zeylanica*) had shown the sensitivity towards all microbes viz *Escherichia coli*, *Proteus vulgaris*, *Enterococcus faecalis*, *Staphylococcus aureus*, *Streptococcus pyogens*, *Pneumococcal pneumonia*. All compound formulations exert antimicrobial effect by phytochemical attack on the bacterial cell wall e.g. carvacrol - a phenol in organ and thyme breaches defensive cell membrane enabling the bacteria to be destroyed. Hence there is need of further in vitro researches to understand the mechanism of antimicrobial effect of Ayurveda herbs. As new microbes emerge, and old microbes re-emerge there is a need for finding safe, cost effective drug, which may be an ideal replacement in certain bacterial infections.

ACKNOWLEDGEMENT

We are thankful for the Rajiv Gandhi University of Health Sciences Karnataka, Bengaluru for funding the research and also, we acknowledge support of Sri Dharmasthala Manjunatheshwara College of Ayurveda & Hospital, Hassan, India.

REFERENCES

- Pandey MM, Rastogi S, Rawat AK. Indian herbal drug for general healthcare: An overview. *Int J Alter Med*. 2008 Jan;6:1–5.
- D. H. Tambekar and S. B. Dahikar. Antibacterial activity of some Indian Ayurvedic preparations against enteric bacterial pathogens Single and polyherbal preparations. *J Adv Pharm Technol Res*. 2011 Jan-Mar; 2(1): 24–29.
- Kenneth. J Ryan. *Sherries medical microbiology an introduction to infectious diseases*. McGraw hill publication; 4thed; 2004: 215
- J.G. Collee. A.G. Fraser. B.P.Marmion.Mackie and McCartney *Practical medical microbiology*. Elsevier publication; 14thed; 2007; 356
- AnejaK.R.,Pranayjain and Raman aneja. *A text book of basic and applied microbiology*. New age international publishers Chakraborty P. 1sted; 2008: 726-737.
- Chakraborty P- *A text book of Microbiology*. New central book agency(P) Ltd. 2nded; 2009: 93-104, 238-257,312-326, 343-351Ranjit R. Raut, Ajit R. Sawant,Bhagyashree B. Jamge *Antimicrobial activity of Azadirachta indica (Neem) against Pathogenic Microorganisms Journal of Academia and Industrial Research (JAIR)Dec 2014Vol 3, Issue 7 327-29*
- Lakshmi. T, Geetha R.V AnithaRoy,“In Vitro Anti Bacterial Activity Of Ethanolic Bark Extract Of Acacia Catechu Willd Against Enteric Pathogens”, *Int. J. DrugDev. & Res.*, July-Sep 2011, 3(3): 328-33
- Sharangadhara.Vidhyadhara editor Sharangadhara Samhita. 2nd section Vatalkalpana. Choukhamba Publications. Varanasi: 1984 P.186 2. Shloka no. 70-81.
- Yogesh S. Biradar, SheetalJagatap, K. R. Khandelwal, and Smita S. Singhanian. *Exploring of Antimicrobial Activity of Triphala Mashi An Ayurvedic Formulation. Evidence-Based Complementary and Alternative Medicine*, vol. 5, no. 1, pp. 107-113, 2008.
- Dhanukar SA, Thatte UM, Pai NR, More PB, Karandikar SM. Immunotherapeutic modification by *Tinospora cordifolia* of abdominal sepsis induced by coecal ligation in rats. *Ind J Gastroenterol* 1988; 7:21-23.
- Thatte UM, Chhabria S, Karandikar SM, Dahanukar SK. Immunotherapeutic modification of *Escherichia coli* abdominal sepsis & mortality in mice by Indian Medicinal Plant. *Ind Drugs* 1987; 25:95-97
- Wijenayake *et al.* Antimicrobial potential of two traditional herbo metallic drugs against certain pathogenic microbial species *BMC Complementary and Alternative Medicine BMC series open, inclusive and trusted* 201616:365 <https://bmccomplementaltermmed.biomedcentral.com/articles/10.1186/s12906-016-1336-1>
- Deepti Ratheea, Permender Ratheeb, Sushila Ratheeb, Dharmender Rat. *Phytochemical screening and antimicrobial activity of Picrorrhiza kurroa*, an Indian traditional plant used to treat chronic diarrhoea. *Vol9, 2, Nov 2016, Pages S1307–S1313*
- Aliya Hayat *et al.* Antimicrobial Activity of Shilajit. *Vol 4 (2), July 2013; 10-12* Available from file:///C:/Users/Dept.%20of%20Samhitha/Downloads/11-1-28-1-10-20170414.pdf
- Christa, Suneeva S *et al.* Modulatory effect of Chandraprabha Vati on antimicrobial peptides and inflammatory markers in kidneys of mice with urinary tract infection. *Iranian journal of kidney diseases* 7 5 (2013): 390-8.
- Brahmanada tripathy. *Ashtanga Hridaya, Uttara Sthana, Chapter 36, Sarpa Visha Pratishedha Adhyaya Verse: 84–85* Chaukhambha Surbharti Publication, Varanasi; 2015
- E N katesan, D. Karrunakarn, C. M. Kumar, S. Selva, Palani Swamy. *Identification of Phytochemical Constituents of Aegle marmelos Responsible for Antimicrobial Activity against Selected Pathogenic Organisms. Ethnobotanical Leaflets: Vol. 2009: Iss. 11 , Article 4.* Available at: <http://opensiuc.lib.siu.edu/ebl/vol2009/iss11/4>
- Lakshmi.T, GeethaR.V, AnithaRoy, *In Vitro Anti Bacterial Activity Of Ethanolic Bark Extract Of Acacia CatechuWilld Against Enteric Pathogens*, *Int. J. DrugDev. & Res.*, July-Sep 2011, 3(3): 328-334
- Lakshmi.T, Geetha R.V, Anitha Roy. *In Vitro AntiBacterial Activity of Ethanolic Bark Extract Of Acacia Catechu Wild Against Enteric Pathogens*. *Int. J. Drug Dev. & Res.*, July-Sep 2011, 3(3): 328-334
- Brahmanand Tripathi, Sharangdhar Samhita, Chaukhambha Surbharti Publication, Varanasi; 2013 *Madhyam Khanda 7/82-83, p. 137.*
- Tambekar, d.h. *et al.* antibacterial properties of contents of triphala: a traditional Indian Herbal preparation. *Continental J. Microbiology*. 2007, 1 (3): 8 - 12,
- Panthi, M.P. and R.P. Chaudhary, 2006. Antibacterial activity of some selected folklore medicinal plants from West Nepal. *Scient. World*, 4: 16-21 referred in Aparna Upadhyay, Pooja Agrahari and D.K. Singh, 2014. *A Review on the Pharmacological Aspects of Terminalia chebula*. *International Journal of Pharmacology*, 10: 289-298. <https://scialert.net/abstract/?doi=ijp.2014.289.298> (accessed on July 21, 2018)
- Jagtap, A.G. and S.G. Karkera, 1999. Potential of the aqueous extract of *Terminalia chebula* as an anticaries agent. *J. Ethnopharmacol.* 68: 299-306 [https://doi.org/10.1016/S0378-8741\(99\)00058-6](https://doi.org/10.1016/S0378-8741(99)00058-6) (accessed on June 10, 2018)
- Ambikadatta Shashtri, Bhaishajya Ratnavali.(*Kanjiv lochan Eng Translation*) Chap.49/14 reprint ed. Chaukhambha Sanskrita Sansthan, Varanasi, 2009, P. 775
- Dinesh D. Wanule *Lac as a promising antibacterial: “Economic Booster” for Lac Cultivation*. *AJBPR Issue 1(Vol. 6) 2016 ISSN: 2231-2560* Available from : <http://www.saepub.com/get-paper1.php?table=AJBPR&id=124>
- Garima Mishra *et al* *Int.J. PharmTech Res*.2010,2(2)pp 1298-1310 http://cissus.org/wp-content/uploads/CissusQuadrangularis_Case Study 01.pdf
- Kaundal *et al.* Analysis of the effect of laksha guggulu (an ayurvedic formulation) on fracture healing In a rat model. *J Res Educ Indian Med*, Jan.-March 2014; Vol. XX (1): 1-8
- Evaluation of antibacterial potential of Trikatu churna and its ingredients: An in vitro study. https://www.researchgate.net/publication/280216728_Evaluation_of_antibacterial_potential_of_Trikatu_churna_and_its_ingredients_An_in_vitro_study [accessed Jun 12, 2017].
- G.Renuga *et al* Evaluation on anti microbial potential of root extracts *Plumbago zeylanica* L against human intestinal microflora. *International Journal of Pharmaceutical and Biological Research (IJPBR)*. Aug-Sep 2013 Vol 4 (4) 146-58
- R. Sreedevi, T. Damodharam. *Phytochemical and Antibacterial Activities of Santalum album*. *Int. J. Pharm. Sci. Rev. Res.*, 33(1), July – August 2015; Article No. 52, Pages: 280-283
- K Manjunatha. *Antibacterial Activity of Pterocarpus santalinus* [Internet]. [cited 2018Jun10]. Available from: fulltext/57ee7aa108ae280dd0ad5c5426447177_Antibacterial_activity_of_Pterocarpus_santalinus.pdf&p=DevEx.LB.1,5067.1
- Lal B, Mishra N. *Importance of Embeliaribes: An Update* [Internet]. *Intl. journal of pharmaceutical sciences and research*. [cited 2018Jun10]. Available from: <http://ijpsr.com/bft-article/importance-of-embelia-ribes-an-update/?view=fulltext>.

33. Bishnu P. Marasini, Pankaj Baral, Pratibha Aryal, et al., "Evaluation of Antibacterial Activity of Some Traditionally Used Medicinal Plants against Human Pathogenic Bacteria," BioMed Research International, vol. 2015, Article ID 265425, 6 pages, 2015. <https://doi.org/10.1155/2015/265425>. Accessed on June 10 2018
34. Antioxidant and Antimicrobial Activity of Nutmeg (*Myristica fragrans*). Available from: https://www.researchgate.net/publication/264972570_Antioxidant_and_Antimicrobial_Activity_of_Nutmeg_Myristica_fragrans [accessed Jun 12, 2017].
35. Asha Devi S, Deepak Ganjewala. Antimicrobial activity of *Acorus calamus* (L.) rhizome and leaf extract [Internet]. <http://www2.sci.u-szeged.hu/ABS>. 2009 [cited 2018Jun10]. Available from: file:///C:/Users/e3t/Downloads/2667-Article Text-2771-1-10-20180605.pdf
36. Screening of Antibacterial Activity of *Tinospora cordifolia* Miers. Extracts Against Dental Pathogens [Internet]. An Economic Study of the Demand for Red Meat in the Kingdom of Saudi Arabia using Almost Ideal Demand System – Sci Alert Responsive Version. [cited 2018 June15]. Available from: <https://scialert.net/abstract/?doi=jpt.2013.28.34>

Cite this article as:

Mallika KJ and Shashirekha KS. Preparation of antibacterial discs from Ayurvedic drugs. Int. J. Res. Ayurveda Pharm. 2018;9(4):21-27 <http://dx.doi.org/10.7897/2277-4343.094102>

Source of support: Rajiv Gandhi University of Health Sciences Karnataka, Bengaluru, India, Conflict of interest: None Declared

Disclaimer: IJRAP is solely owned by Moksha Publishing House - A non-profit publishing house, dedicated to publish quality research, while every effort has been taken to verify the accuracy of the content published in our Journal. IJRAP cannot accept any responsibility or liability for the site content and articles published. The views expressed in articles by our contributing authors are not necessarily those of IJRAP editor or editorial board members.