

Research Article

## Comparative Analysis of Antibacterial and Antifungal Properties of Traditional Medicinal Plants of Shimla and Solan, Himachal Pradesh.

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### ABSTRACT

Human race is in great danger due to the development of drug resistance in pathogens. Medicinal plants produce an array of secondary metabolites to protect themselves against pathogens and can be exploited as phytomedicines for humans. In view of the benefits of medicinal plants, 41 medicinal plants of ethnomedicinal importance, belonging to 33 plant families were collected from Solan and Shimla regions of Himachal Pradesh, India. The ethanolic extracts of different parts (flowers, leaves and barks) of these plants were tested for antimicrobial activity against *Staphylococcus aureus*, prokaryotic bacterial pathogen and *Candida albicans*, an eukaryotic fungal pathogen using agar well diffusion assays. Collectively, the medicinal plants exhibited 2 fold more antimicrobial activity against prokaryotic pathogen (*S. aureus*) as compared to the eukaryotic pathogen (*C. albicans*). For *S. aureus*, 40 medicinal plants and their different parts exhibited antimicrobial activity whereas for *C. albicans* only 21 medicinal plants showed antifungal activity. Amongst all medicinal plants, *Hypericum perforatum* (flowers) extract showed highest antimicrobial activity against *S. aureus* with the zone of inhibition of 19 mm and found bactericidal. On the other hand, *Cymbopogon citrate* (leaf) extract showed highest antifungal activity of 10 mm and fungicidal effect. The extracts of *H. perforatum*, *C. citrate* and *T. arjuna* were strong antimicrobial against both pathogens. It is hypothesized that phytochemicals present in these three plants might be following the same mechanism of action causing death of *S. aureus* and *C. albicans*. These findings have an implication for the development of phytomedicines/ drugs with pathogen-specific effect.

**Keywords:** Medicinal plants, antibacterial, antifungal and Himachal Pradesh.

### INTRODUCTION

Human infectious diseases particularly those involving bacteria, fungi and viruses are the leading cause of death in tropical countries [Frean and Blumberg, 2008]. Development of antimicrobial resistance to antibiotics is one of the major challenges to combat infectious diseases caused by bacterial and fungal pathogens. Antibiotic resistance is very common in *Staphylococcus aureus* for methicillin [Mulligan *et al.*, 1993], *Escherichia coli* for ampicillin, cephalothin and trimethoprim-sulfamethoxazole [Sahm *et al.*, 2001] and *Salmonella typhi* for ampicillin, chloramphenicol, streptomycin, sulfonamides and tetracycline [Briggs and Fratamico, 1999]. Several isolates of *C. albicans* have developed resistance against fluconazole [Jack and Sobel, 2006], azole drug [White and Goetz, 1994] and amphotericin B [Kelly *et al.*, 1997]. Over expression of efflux pumps related to *MDR1* gene (multiple drug resistance gene 1) in *S. aureus* [Ramage *et al.*, 2002] and *CDR1* gene (multidrug resistance gene 1) in *Candida* [White *et al.*, 2002] have been proposed to be the underlying mechanisms of drug resistance in *S. aureus* and *C. albicans*, respectively. Multidrug resistance in clinical bacteria like *Staphylococcus aureus* is responsible for nosocomial infections [Mulligan *et al.*, 1993] and skin diseases [Lina *et*

*al.*, 1999]. With time, isolates of *S. aureus* have developed resistance to many additional antibiotics like vancomycin, clindamycin and - lactam antibiotics (Lewis and Jorgensen, 2005). Various mechanisms like activation of efflux pumps [Piddock, 2006], alterations in drug efflux transporters and drug targets [Loscher, 2005] have been studied for drug resistance. Due to the development of antimicrobial resistance, higher dosages for longer time and even the combination of different drugs is recommended, leading to an increase in the side effects and drug toxicity in the patients [Greenblatt *et al.*, 1977]. As a result, there is a pressing demand to search for new drugs, which are more potent at low dosage and are patient friendly.

According to reports, it is estimated by World health organization that 80% of population of developing countries is dependent on the traditional medicinal plants as a source of drug [Farnsworth, 1988]. Herbal medicines have been used traditionally to treat infections since 3000 BC [Philip, 2011]. Some examples of plant based drugs are aspirin from willow bark (*Salix spp.*) as analgesic/ antipyretic [Setty and Sigal 2005] and anti-inflammatory drug [Vane and Botting, 1998]. Other examples of plant based drugs are vincristine, vinblastine, colchicine, ellipticine and lepachol along with flavopiridol, which are

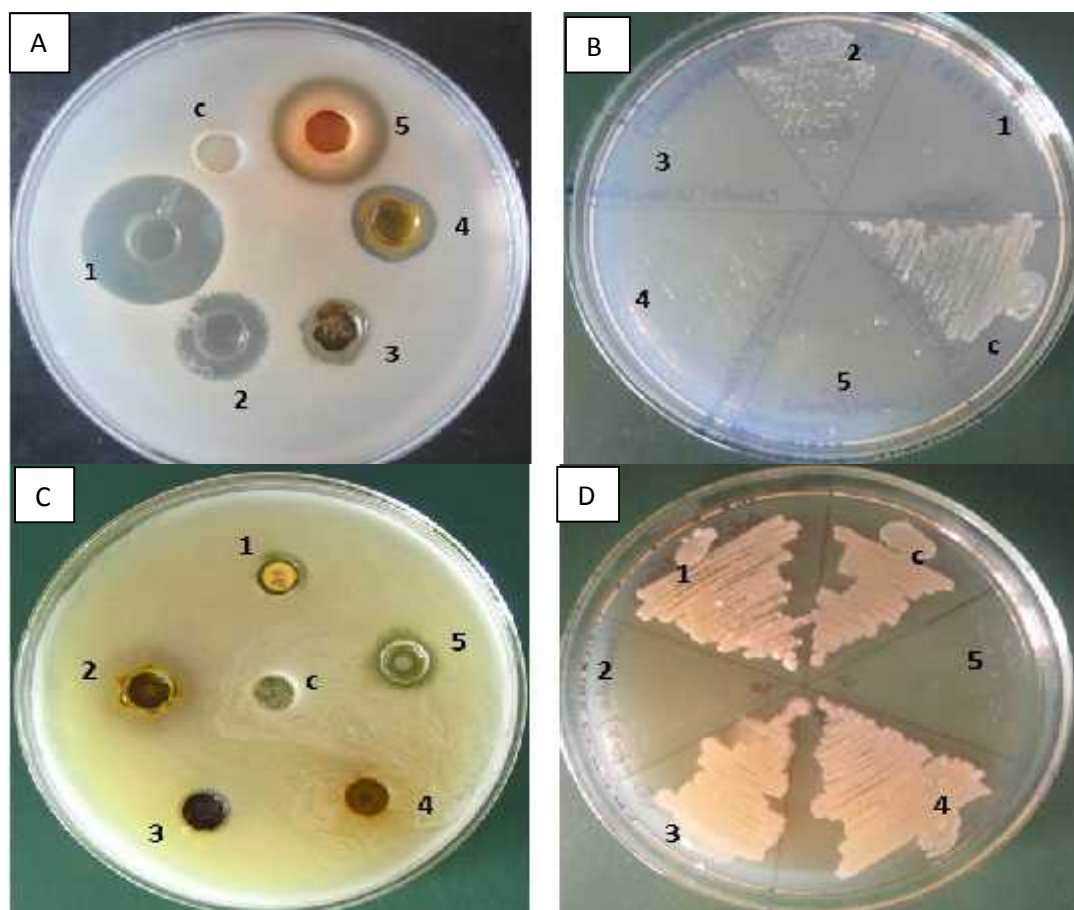


Figure 1: Evaluation of antimicrobial activity of extracts of medicinal plants by agar well diffusion assay. A & C: Antimicrobial assay of medicinal plant extracts against *S. aureus* and *C. albicans*, respectively. B & D: Bacteriostatic/Bactericidal and Fungistatic/fungicidal assay. The wells were loaded with ethanolic extracts (40 $\mu$ g) of the following plants: (A, B): c- Solvent control (ethanol), 1- *Cymbopogon citrates* (leaves), 2- *erythromycin* (50  $\mu$ g), 3- *Colebrookea oppositifolia* (axial), 4- *Berberis aristata* (leaves), 5- *Embllica officinalis* (leaves). (C, D): c- Solvent control (ethanol), 1- amphotericin B (100  $\mu$ g), 2- *Pinus roxburghii* (bark), 3- *Cannabis sativa* (leaves), 4- *Berberis aristata* (fruits), 5- *Ferula asafoetida* (latex). Fig 1 Sharma et al

used to cure cancer [Mukherjee et al, 2001]. Medicinal plants like *Embllica officinalis*, *Terminalia chebula*, *Terminalia bellerica*, *Plumbago zeylanica* and *Holarrhena antidysenterica* have been reported to possess antibacterial properties [Ahmad et al., 1998]. *Ginkgo biloba* was reported to be an effective source for treating tinnitus [Drew and Davies, 2001] and *Gongronema latifolium* was reported as a source of antimalarial drug [Adebajo et al., 2013]. Phytocompounds such as cinnamaldehyde, eugenol and geraniol were found to be active alone or in combination with fluconazole and amphotericin B against pre-formed biofilms of *Candida albicans* [Khan and Ahmad, 2012].

Himachal Pradesh is inhabited by a diverse range of plants with ethnomedicinal features and thus a valuable source for developing novel antimicrobials based on phytocompounds. In the present study, we aimed to identify and compare the antimicrobial activities of some medicinal plants from the rich biodiversity of Himachal Pradesh against an eukaryotic (*C. albicans*) and a prokaryotic (*S. aureus*) human pathogen.

## MATERIAL AND METHODS

Microbial strains: Erythromycin resistant strain of *Staphylococcus aureus* (ATCC-43300) was obtained from Post Graduate Institute of Medical Education and Research, Chandigarh and was stored on nutrient agar plates and slants at 4 $^{\circ}$  C. *Candida albicans* (ATCC- 90028) was procured from IMTECH, Chandigarh and was stored on slants of Yeast peptone dextrose (YPD) agar medium at 4 $^{\circ}$  C.

Culture media and growth conditions: *S. aureus* was cultured in nutrient broth and Muller Hinton agar (MH) was used to perform agar well diffusion assay [Olila and Opuda-Asibo, 2001]. YPD broth was used to culture *C. albicans* and YPD agar was used for antifungal assay for [Kim, et al., 2009]. The media used in this study were purchased from Himedia Labs, India. Erythromycin and Amphotericin B discs were purchased from MP Biomedicals, Inc. USA and Himedia Labs, India, respectively. Incubation temperature was 37 $^{\circ}$  C for *S. aureus* and 30 $^{\circ}$  C for *C. albicans*.

Collection of plant material: The plant material used in this study was collected during the month of September-December from Solan (altitude 1350m, temperature 20-30  $^{\circ}$ C, humidity 55-68%) and Shimla (altitude 2202m,

temperature 12-25 °C, humidity 62-80%) regions of Himachal Pradesh. These plants were verified from herbarium of Dr Y.S Parmar University of Horticulture and Forestry, Nauni, Himachal Pradesh.

Preparation of plant extracts: The collected plant material was thoroughly washed with running tap water, followed

by surface sterilization with 1% H<sub>2</sub>O<sub>2</sub> and then washed with autoclaved distilled water. The surface sterilized plant material were dried in oven at 40 °C for 5 days or until they were dried completely. Dried plant material was crushed in mixer grinder to make fine powder. The fine powder (5 g) was extracted in soxhlet apparatus

Table 1. Antimicrobial activity of medicinal plants extracts against *S. aureus* and *C. albicans*.

S. No.	Family	Plant	Parts used	<i>S. aureus</i> Zone of inhibition (mm)*	Bacteriostatic /Bactericidal	<i>C. albicans</i> Zone of inhibition (mm)*	Fungicidal/ fungistatic
1.	Combretaceae	<i>Terminalia arjuna</i>	Bark	11.5±0.5	Bactericidal	5±0.5	Fungicidal
2.	Myrtaceae	<i>Eugenia Jambolana</i>	Leaves	9.5±0.5	Bacteriostatic	4±0.5	Fungistatic
3.	Anacardiaceae	<i>Rhus cotinus</i>	Leaves	5.5±0.5	Bactericidal	2±0.5	Fungicidal
4.	Liliaceae	<i>Allium cepa</i>	Bulb	6.0±0	Bacteriostatic	Not detected	Not detected
5.	Berberidaceae	<i>Berberis aristata</i>	Leaves Fruits	6.0±0 6.0±1.0	Bactericidal Bactericidal	4.0±0 Not detected	Fungicidal Not detected
6.	Juglandaceae	<i>Juglans regia</i>	Leaves	5.5±0.5	Bactericidal	3.0±0	Fungicidal
7.	Pinaceae	<i>Pinus roxburghii</i>	Bark Leaves	7.0±1.0 9.5±0.5	Bactericidal Bactericidal	2±0.5 2±0.5	Fungicidal Fungicidal
8.	Ericaceae	<i>Rhododendron viscosum</i>	Flower	7.5±0.5	Bacteriostatic	Not detected	Not detected
9.	Piperaceae	<i>Piper nigrum</i>	Fruit	4.5±0.5	Bacteriostatic	Not detected	Not detected
10.	Solanaceae	<i>Withania somnifera</i> <i>Datura metel</i> <i>Capsicum annum</i>	Fruit Leaves Fruit Fruit	4.5±0.5 11.0±1.0 1.5±0.5 5.0±0	Bactericidal Bactericidal Bacteriostatic Bacteriostatic	Not detected Not detected 2±1.0 Not detected	Not detected Not detected Fungistatic Not detected
11.	Punicaceae	<i>Punica granatum</i>	Flowers Leaves	9.5±0.5 4.5±0.5	Bacteriostatic Bacteriostatic	Not detected Not detected	Not detected Not detected
12.	Lythraceae	<i>Lawsonia alba</i>	Leaves	11.0±1.0	Bactericidal	Not detected	Not detected
13.	Zingiberaceae	<i>Zingiber officinale</i> <i>Curcuma longa</i>	Tuber Tuber	3.5±0.5 4.0±1.0	Bacteriostatic Bactericidal	Not detected Not detected	Not detected Not detected
14.	Euphorbiaceae	<i>Emblica officinalis</i>	Leaves	7.5±0.3	Bactericidal	7±0.5	Fungistatic
15.	Ranunculaceae	<i>Nigella sativa</i>	Seed	7.5±0.5	Bactericidal	Not detected	Not detected
16.	Rutaceae	<i>Zanthoxylum armatum</i> <i>Ruta graveolens</i> <i>Aegle marmelos</i>	Leaves Fruit Fruit	4.5±0.5 4.5±0.5 6.5±0.5	Bacteriostatic Bacteriostatic Bacteriostatic	Not detected Not detected 5±0.5	Not detected Not detected Fungistatic
17.	Cannabidaceae	<i>Cannabis sativa</i>	Leaves	5.5±0.5	Bactericidal	2±0.5	Fungistatic
18.	Myristicaceae	<i>Myristica fragrans</i>	Fruit Flower	7.5±0.5 5.5±0.9	Bactericidal Bactericidal	Not detected Not detected	Not detected Not detected
19.	Salicaceae	<i>Populus nigra</i>	Leaves	6.5±0.5	Bactericidal	3±0.5	Fungistatic

Table 1. Antimicrobial activity of medicinal plants extracts against *S. aureus* and *C. albicans*.

S. No.	Family	Plant	Parts used	<i>S. aureus</i> Zone of inhibition (mm)*	Bacteriostatic /Bactericidal	<i>C. albicans</i> Zone of inhibition (mm)*	Fungicidal/ fungistatic
20	Laureceae	<i>Cinnamomum tamala</i>	Bark	5.5±0.5	Bacteriostatic	4±0.5	Fungicidal
21.	Meliaceae	<i>Azardica indica</i>	Leaves	6.5±0.5	Bacteriostatic	Not detected	Not detected
22.	Umbelliferae	<i>Ferula asafoetida</i>	Latex	5.5±0.5	Bactericidal	3±1.0	Fungicidal
23.	Liliaceae	<i>Allium sativum</i>	Bulb	6.5±0.5	Bacteriostatic	Not detected	Not detected
24.	Myrtaceae	<i>Callistemon citrinus</i>	leaves	5.5±0.3	Bactericidal	Not detected	Not detected
		<i>Syzygium aromaticum</i>	Flower bud	6.5±0.5	Bactericidal	3±0.5	Fungistatic
25.	Lamiaceae	<i>Ajuga reptans</i>	Flower	5.5±0.5	Bactericidal	Not detected	Not detected
		<i>Colebrookea oppositifolia</i>	Leaves	9.5±0.5	Bactericidal	1±0.5	Fungicidal
			Axial	2.5±0.5	Bactericidal	2±0.3	Fungicidal
			Bark	5.5±0.5	Bactericidal	1±0.5	Fungicidal
		<i>Thymus vulgaris</i>	Whole plant	5.5±0.5	Bacteriostatic	Not detected	Not detected
26.	Euphorbiaceae	<i>Euphorbia hirta</i>	Fruit	5.5±0.5	Bacteriostatic	3±0.5	Fungistatic
27.	Compositae	<i>Taraxacum officinale</i>	Leaves	7.5±0.5	Bactericidal	3.0±0	Fungistatic
28.	Hypericaceae	<i>Hypericum perforatum</i>	Flowers	19.0±0	Bactericidal	6±0.5	Fungicidal
29.	Fabaceae	<i>Abrus precatorius</i>	Seeds	7.5±0.5	Bacteriostatic	1±0.5	Fungicidal
30.	Poaceae	<i>Cymbopogon citrate</i>	Leaves	14.5±0.5	Bactericidal	10±1.0	Fungicidal
31.	Malvaceae	<i>Bombax ceiba</i>	Flowers	2.5±0.5	Bacteriostatic	1±0.2	Fungicidal
32.	Convolvulaceae	<i>Cuscuta reflexa</i>	Stem	Not detected	Not detected	Not detected	Not detected
33.	Menispermaceae	<i>Tinospora cordifolia</i>	Stem	3.5±0.5	Bactericidal	Not detected	Not detected

\*All the experiments were repeated two times to calculate the standard deviation of the zone of inhibition. SD denotes the standard deviation. The data represents the average of the two experiments ± SD.

[Hawthorne et al., 2000] using ethanol. The extracts were evaporated in rota-evaporator at 40° C. The dried extract was stored at -20° C until further use. Antimicrobial assay was performed by dissolving the dried extract in ethanol to the concentration of 1 mg/ml prior to use.

Antimicrobial assay: Antimicrobial activity was measured by using agar well diffusion method [Perez et al., 1990]. Erythromycin (50 µg/well) and amphotericin B discs (100 µg/disc) were taken as the positive antibiotic control for *S. aureus* and *C. albicans*, respectively and solvent (ethanol) used for diluting the extracts was used as negative control. The amount of extract used for the antimicrobial assay was 40µg. The zone of clearance (mm) for two independent experiments of each plant extract and the antibiotic was measured and the standard deviation (SD) of values from two independent experiments was calculated. To assay the bactericidal or bacteriostatic activity and

fungicidal/ fungistatic activity of extracts/ antibiotics, cells were carefully scraped from the zone of clearance around the well and streaked on nutrient agar ( for *S. aureus*) or YPD agar (for *C. albicans*) plates, and observed for the growth after 24 h of incubation at the respective temperatures.

## RESULTS

The rationale of the present study was to compare the antimicrobial activity of the medicinal plant extracts against representative eukaryotic (*C. albicans*) and prokaryotic (*S. aureus*) pathogens. The hill state of Himachal Pradesh is rich in tribal and traditional cultural population, predominated by the use of traditional medicinal plants for curing most ailments [Uniyal et al., 2006]. In the present study, information from folklore medicine was used to collect various ethnomedicinally

important plants of Himachal Pradesh for the comparative analysis of their antibacterial and antifungal properties. Ethanolic extracts of different plants parts, i.e. flowers, leaves and barks were used for antimicrobial analysis against *S. aureus* and *C. albicans* by well diffusion method. While *S. aureus* is a gram-positive bacterial pathogen infecting humans, *C. albicans* is an opportunistic fungal pathogen. The clearance of bacterial growth around the well (referred to as zone of inhibition) containing the respective plant extract/ antibiotic was evaluated to assess the antimicrobial activity. A representative result of antimicrobial assays [Fig 1A, 1C] and the corresponding bactericidal/ fungicidal assays (Fig1 B and D) are shown in Figure 1.

Out of 48 extracts (ethanolic) of 41 medicinal plants (Table 1), 47 showed antibacterial activity against *S. aureus* and 24 extracts were found to be antifungal against *C. albicans* (Table 1). Inhibition zones of erythromycin and amphotericin B were observed to be 5 mm and 2 mm respectively, and both the drugs exhibited static effects on the respective pathogen growth (Table 1).

The maximum zone of inhibition was shown by extracts of *H. perforatum* (flowers), *C. citrate* (leaves), *L. alba* (leaves), *W. somnifera* (leaves) and *P. roxburghii* (leaves) against *S. aureus*. The antimicrobial activity of these plants extracts was bactericidal. Amongst the five extracts, the maximum zone of inhibition (19 mm) of *S. aureus* growth was observed with the extract of *H. perforatum* (flowers), whereas the remaining 4 extracts produced zone of inhibition in the range of 10- 15 mm (Table 1).

On the other hand, maximum antifungal activity against *C. albicans* was shown by extract of *C. citrate* (leaves) with zone of inhibition of 10 mm and observed to be fungicidal (Table 1). The extracts of *E. officinalis* (leaves), *H. perforatum* (flower), *A. marmelos* (fruit), *E. jambolana* (leaves), *B. aristata* (leaves), *C. tamala* (bark) and *T. arjuna* (bark) exhibited weak antifungal activity (zone of inhibition < 4mm), while the remaining 15 plants extracts showed moderate inhibition of *C. albicans* growth (zone of inhibition >4 mm). The antifungal activity of extracts of *C. citrate*, *E. officinalis* (leaves), *B. aristata* leaves), *C. tamala* (bark) and *H. perforatum* (flower) were fungicidal but those of *A. marmelos* (fruit) and *E. jambolana* (leaves) were fungistatic (Table 1).

Comparison of antibacterial versus antifungal activities revealed that the extracts of certain medicinal plants namely *T. arjuna*, *E. jambolana*, *B. aristata* (leaves), *A. marmelos* (fruit), *C. tamala* (bark), *H. perforatum* (flower) and *C. citrates* (leaves) exhibit strong antimicrobial activity against both the pathogens. On the other hand, extract of *Cuscuta reflexa* (stem) has not shown any antimicrobial activity against either of the tested pathogens. Analysis of the mode of antimicrobial action revealed that out of 48 extracts of 41 medicinal plants, ~ 50% extracts were bactericidal against *S. aureus*, whereas for *C. albicans*, only 30% plant extracts were fungicidal.

## DISCUSSION

In the recent years, medicinal plants have attracted a lot of researchers globally. These plants provide promising

potential to cure human diseases. Himalayan plants are very rich in the medicinally potent compounds. It has been reported that about 35 plant species are commonly used by local people of tribal communities of chhota Bhangal, Western Himalayas for curing various diseases [Uniyal et al., 2006]. Many reports are available on antibacterial and antifungal properties of medicinal plants and some of these observations have helped in identification of potent medicinal plants of Himalayas and other region that have been used to cure bacterial and fungal infections [Muanza et al., 1994; Ahmad et al., 1998; Srinivasan et al., 2001; Rios and Recio 2005; Kumar et al., 2006].

The present study revealed that the ethanolic extracts of *H. perforatum* (flowers and leaves), *C. citrate* (leaves), *T. arjuna* (bark), *W. somnifera* (leaves) and *L. alba* (leaves) have strong antibacterial potential against *S. aureus*. Consistent with our study, the extracts of different species of *Hypericum* and *L. alba* have been shown to exhibit strong activity against bacterial pathogens [Sakar and Tamer 1990; Ghosh et al., 2008; Khan et al., 2009; Sharma et al. 2011]. The flowering and aerial parts of *Hypericum* spp. known for the bioactive compounds like hypericins, hyperforins and flavanoids have been abundantly used in the pharmaceutical drug formulations as well as in cosmetics products [Guedes et al., 2012]. It would thus be interesting to examine the role of these bioactive compounds of *Hypericum* spp. used in this study in antibacterial action against *S. aureus*.

In our study, the growth of *C. albicans* was also found to be inhibited by *H. perforatum* extracts. Similarly, xanthenes isolated from the roots of *Hypericum perforatum* were reported to be antifungal against *Candida albicans*, non-*albicans* *Candida* species, *Cryptococcus neoformans* and other dermatophytes [Noemi et al., 2013]. The chloroform, methanol and aqueous extracts of *Lawsonia* spp. were observed to be antifungal for various dermatophytic spp. [Sharma et al., 2011]. However, in the present study, ethanolic extract of *Lawsonia alba* failed to inhibit growth of *C. albicans*. This could be due to the difference in the type of solvent used for the extraction in the two studies.

The leaf extract of *C. citrate* was reported to be more potent against gram positive as compared to gram negative bacteria, and also observed to be antifungal against various species of *Candida* [Ferdous et al., 1992; Silva et al., 2008]. Consistent with these findings, *C. citrate* extracts showed strong activity against *S. aureus* and *C. albicans* in our study.

*W. somnifera* is well known for its antimicrobial features [Singariya et al., 2012]. A monomeric glycoprotein isolated from *W. somnifera* root tubers and leaves was found to be potent microbial agent against phytopathogenic fungi and bacteria [Ghosh 2009; Girish et al., 2006]. In our study, *W. somnifera* extract was antibacterial against *S. aureus* but was not active against *C. albicans*.

Many researchers have documented the antibacterial activity of *T. arjuna* against different bacteria [Fakruddin et al. 2011; Bhuiyan et al., 2011; Jothiprakasham et al., 2013; Ghosh et al., 2008]. In our study, we observed that

the extracts of *T. arjuna* (bark) have a very strong antimicrobial activity against both bacterial and fungal pathogens. Thus, our study forms the first report of antifungal activity of *T. arjuna*.

According to Kumar et al [2006], extracts of *P. roxburghii* (stem and cone) were observed to be antimicrobial against various pathogens and in this study, we observed that the bark and leaf extracts of *P. roxburghii* have antibacterial activity against *S. aureus*, but in the case of *C. albicans*, their inhibitory effect was minimal.

In this study, the extracts of *C. oppositifolia* (leaves, axial and bark) were observed to be bactericidal and fungicidal. The flavonoid isolated from leaves of *C. oppositifolia* was reported antimicrobial against bacteria [Mahapatra 2013], and an acteoside isolated was found to be synergistic in combination with amphotericin B against fungal pathogens [Ali et al., 2011].

In the present study, the extracts of *P. granatum* (flower, leaves) were strong antibacterial agents, but no antifungal activity was observed against *C. albicans*. The peel extract of *P. granatum* was found to be antibacterial and antifungal for certain species [Negi and Jayaprakasha 2003; Dutta et al., 1998].

*E. jambolana* (leaves) extracts showed antibacterial and antifungal activity, with static effects on both the microbes, but does not cause complete killing of microbes (bacteriostatic/ fungistatic). The antibacterial, cytotoxic and antifungal activity of this plant has also been reported by other research groups [Coutinho et al., 2010 and Santos et al., 2012]. In another study, *P. granatum* (fruit) and *E. jambolana* (leaves) have been found to inhibit *Salmonella* serovars by 90- 100% [Voss- Rech et al., 2011].

While the ethanolic extracts of *N. sativa* (seeds), *M. fragrans* (fruit) and *E. officinalis* (leaves) have shown similar extent of antibacterial activity and bactericidal effect, *M. fragrans* (flower) extract was least active against *S. aureus*. Ferdous et al. [1992] also reported antibacterial activity of the volatile oil of *Nigella sativa* seeds against 37 isolates of *Shigella*. Amongst these three plants extracts, only *E. officinalis* (leaves) extract exhibited antifungal activity but not *N. sativa*. On the contrary, Khan et al. [2003] reported that *N. sativa* (seeds) are inhibitory against candidiasis in mice. The antifungal and antibacterial activity was also reported in *M. fragrans* plant extracts prepared in different solvents [Asgarpanah and Kazemivash, 2012; Dipankar et al., 2012; Dorman and Deans, 2000].

Extracts of *B. aristata* fruit and leaves showed bactericidal activity against *S. aureus* but in the case of *C. albicans*, antifungal activity was observed only in leaves of this plant. The antimicrobial activity of *B. aristata* was also reported by different researchers [Srinivasan et al., 2001; Freile, et al., 2003; Mathur, et al., 2011].

The plant extracts observed to be bactericidal for *S. aureus* were *T. officinale* (leaves), *P. nigra* (leaves), *S. aromaticum* (flower bud), *R. cotinus* (leaves), *J. regia* (fruit), *T. cordifolia* (stem), *C. longa* (bulb), *C. sativa* (leaves), *C. citrinus* (leaves), *A. reptans* (flower) and *F. asafetida* (latex). All of these plants were reported to be antibacterial and antifungal against various pathogens

[Izzo, et al., 1995; Digrak, et al., 2001; Svetaz, et al., 2010; Sitara et al., 2008; Naz et al., 2010; Cock 2008; Srinivasan et al., 2001; Hendriani et al., 2009; Popescu and Kopp 2013; Rani and Khullar 2004; Abdou 1972; Ahmad and Beg 2001 and Khlifi et al., 2013]. On the other hand, extracts of *A. precatorius* (seed), *R. viscosum* (flower), *A. marmelos* (fruit), *A. sativa* (bulb), *Z. armatum* (leaves), *R. gravelons* (fruit), *P. nigrum* (seeds), *C. tamala* (bark), *C. annum* (fruit), *D. metel* (fruit), *A. indica* (leaves) were observed to be bacteriostatic. All of these plants were reported to be medicinally important and used to cure various diseases [Dorman and Deans 2000, Pandey 2012, Erturk 2006, Sai Ram et al., 2000, Kagale 2004].

Amongst all the 41 plants tested, extracts of *Cuscuta reflexa* (stem) did not show any antimicrobial and antifungal activity for the both the tested pathogens, whereas methanol extract of *Cuscuta reflexa* (stem) was reported to be antibacterial [Pal, et al., 2006]. This could be due to the differences in geographical location of the plant or the solvent of extraction.

In the present study, we found remarkable differences in the activity of medicinal plants of Shimla and Solan against prokaryotic (*S. aureus*) and eukaryotic (*C. albicans*) microbes. Surprisingly, *H. perforatum* (flower and leaf extract), *C. citrate* (leaf) and *T. arjuna* (bark) were strong antimicrobials against both *C. albicans* and *S. aureus*, reflecting similarities in their mechanism of action against prokaryotic and eukaryotic pathogens. The possible hypothesis could be that the bioactive components of extracts target the basic cellular processes e.g nucleic acid synthesis.

Thus, our results reveal mechanistic differences as well as similarities in the antimicrobial properties of the tested medicinal plants against a prokaryotic pathogen (*S. aureus*) versus a eukaryotic pathogen (*C. albicans*).

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