International Journal of Chemical Studies

P-ISSN: 2349–8528
E-ISSN: 2321–4902
IJCS 2019; 7(1): 33-36

Received: 22-11-2018
Accepted: 24-12-2018

Saroj Yadav
Assistant Professor, Department of Textile and Apparel Designing I.C. College of Home Sciences, CCS Haryana Agricultural University, Hisar, Haryana, India

Correspondence
Saroj Yadav
Assistant Professor, Department of Textile and Apparel Designing I.C. College of Home Sciences, CCS Haryana Agricultural University, Hisar, Haryana, India

Assessment of antimicrobial activity of selected plant extracts for application on textiles

Saroj Yadav

Abstract
The increasing awareness of public about various ill effects of microorganisms on human health, there is an increasing demand for natural antibacterial materials for development of eco-friendly textiles for health care and hygiene application. Plants have been a valuable source of natural products and are known to be chemically balanced, effective and least injurious with none or reduced side effects as compared to synthetic compounds. The present study was undertaken to investigate the preliminary antimicrobial properties of ten plant sources against *S. aureus (gram +)* and *E. coli (gram -)* bacteria. Aqueous and ethanol extracts of each plant were tested for their antibacterial performance using agar well diffusion method. The results indicated that all the ten plants exhibited antibacterial activity against selected bacteria. The yield of extraction in both the solvents varied from 40ml to 46 ml/50ml of extract. *Jamun* leaves exhibited maximum yield of extract (46ml/50 ml). Maximum phenolic content was found in ethanol extract of *neem* leaves i.e. 482.01 mg GAE/g and lowest phenolic content was found in *bael* leaves i.e. 11.14 mg GAE/g. *Safeda* and *bael* leaves extracts in 70% ethanol exhibited higher zone of inhibition against *S. aureus* (23mm, 20mm, respectively) and *E. coli.* (20mm, each).

Keywords: Plant extracts, ethanol and aqueous extracts, microorganisms, antimicrobial activity

Introduction
For a long period of time, plants have been a valuable source of natural products for maintaining human health and their use as medicines could be traced as far back as the beginning of human civilization. The earliest mention of medicinal use of plants in Hindu culture is found in "Rigveda", which is said to have been written between 4500 - 1600 B.C. and is supposed to be the oldest repository of human knowledge (Rastogi and Mehrotra, 2002) [5]. The plants produce numerous natural chemical compounds with different biological activities, which serve as plant defense mechanisms against predation by micro-organisms, insects or herbivores.

Microbes are the tiniest creatures which cannot be seen with the naked eye and can be found almost everywhere in the environment. Micro-organisms include a variety of organisms like bacteria, fungi, algae and viruses. Bacteria are unicellular organisms which can further be subdivided as either Gram positive (e.g. *Staphylococcus aureus*) and Gram negative (e.g. *E. coli*), or spore forming and non-spore forming types. Antimicrobials are the agents that either kill micro-organisms or inhibit their growth by interfering with the necessary mechanism of the microbe's cell. These agents work either by the slow release of the active ingredient or by surface contact with the microbes (Gupta and Bhaumik, 2007; Sood, 2014) [3].

Antimicrobials of plant origin have enormous therapeutic potential as they are effective in the treatment of infectious diseases while mitigating the side effects of the synthetic antimicrobials. The natural plant products are known to be chemically balanced, effective, less toxic and least injurious with none or reduced side effects as compared to synthetic medicines. Plants have long been employed in treating and preventing various ailments. Crude plant extracts or plant derived components are a reliable source to discover new antimicrobial agents (Hemaiswarya *et al*., 2008) [4].

Though, there is a vast resource of antimicrobial agents derived from plants, which can be used for imparting useful antimicrobial property to the textiles substrates, yet the utilization of plant based herbal products depends upon their bulk availability, extractability, antimicrobial efficacy, durability, non-allergenic reaction to skin, shelf life and cost. Thus the aim of the present work is to assess the antimicrobial activity of different plant extracts for application on textiles.

Materials and methods

Collection and Selection of Plant Material: On basis of available review, an exhaustive list of 50 plants having antimicrobial property was prepared. Out of the prepared list, ten plants having good bacterial resistance, locally available and in abundance were selected for the study. Only renewable parts of the plants were used. Matured fresh leaves of selected plant sources were collected from CCS Haryana Agricultural University, Hisar campus.

a. **Drying of the samples:** The fresh leaves were collected, washed to remove debris and dried at 40 ± 1^0 C in hot air oven.
b. **Sample preparation:** After being completely dried, the material was crushed into small pieces, pulverized into coarse powder and stored in air tight containers free from environmental climatic changes, till usage.

Extract Preparation: Extraction refers to separating the desired extract by physical and chemical means with aid of solvent. Solvents play a vital role in the extraction process. The extraction efficiency and quality of phenolics is dependent on the type of solvent, its concentration that varies with the plant sources and its physical state. The extraction of plant materials was done using following methods:

a. **Aqueous extraction:** Aqueous extract was prepared by soaking 2g of plant powder in 50 ml distilled water in a stainless steel vessel overnight to loosen the cell structure. The mixture was centrifuged and filtered to separate the extract and remove plant remnants.
b. **Ethanol extraction:** 2g of plant powder was soaked in 50 ml of 70 % ethanol in a stainless steel vessel overnight to loosen the cell structure. The mixture was centrifuged and filtered to separate the extract and remove plant remnants.

Yield count: The mixture was centrifuged and filtered by using Whatmann No.1 filter paper to separate the extract and remove plant remnants**.** After separating plant, the amount of extract was measured in ml.

Testing of extracts

a. Determination of Total Phenolic Content (TPC): The amount of total phenolics in extracts was determined according to the Folin-Ciocalteu procedure. Samples (2ml, triplicates) were introduced into test tubes; 1.0 ml of Folin-Ciocalteu's reagent and 0.8 ml of sodium carbonate (7.5%) were added. The tubes were shaken well to mix the contents and allowed to stand for 30 min. Absorption at 765 nm was measured using Systronics UV-vis spectrophotometer. The total phenolic content was expressed as gallic acid equivalents (GAE) in milligrams per gram dry material.

b. Bio-Assay: The effect of various plant extracts on the two bacterial strains i.e. *Escherichia coli* and *Staphylococcus aureus* was assayed by Agar well diffusion method. The minimum concentrations of the plant extracts to inhibit the microorganisms were also determined by dilution method using plant fractions serially diluted in sterile nutrient broth.

c. Antimicrobial testing of extracts: The antimicrobial activity of samples was evaluated using agar disc diffusion assay. Briefly, a 24 and 48 hours old culture of selected bacteria was mixed with sterile physiological saline (0.9%) and the turbidity was adjusted to the standard inoculums of Mac Farland scale 0.5 (10^6 colony forming unit (CFU) per ml). Petri plates containing 20 ml of Nutrient Agar was used for antibacterial activity. The inoculums was prepared on the surface of the solidified media and Whatman No.1 filter paper discs (5 mm in diameter) impregnated with the sample (20µl/ disc) were placed on the plates. Streptomycin was used as positive control for bacteria. Plates inoculated with the bacteria were incubated for 24 hour at 37^0C. The diameters of zone of inhibition were measured in millimeters (zoi mm).

Results and Discussion

Plants have always been important sources of medicines since ancient times and majority of the population still relies on one or other forms of plant based traditional remedial measures for different diseases. Plant based natural constituents can be derived from any part of the plant like bark, leaves, flowers, roots, fruits, seeds, etc. The leaves of ten plants sources were explored for antibacterial activities. The sources selected for the present study are recorded in Table-1.

Table 1: Plant materials selected for the bacterial resistance properties

S. No.	Local name	Scientific name	English name	Part used
1.	*Aonla*	*Phyllanthus emblica*	Indian gooseberry	Leaves
2.	*Bael*	*Aegle marmelos*	Golden apple	Leaves
3.	*Curry patta*	*Murraya koenigii*	Curry leaves	Leaves
4.	*Safeda*	*Eucalyptus globulus labill*	Eucalyptus	Leaves
5.	*Mehandi*	*Lawsonia inermis*	Henna	Leaves
6.	*Jamun*	*Syzygium cumini*	*Java Plum*	Leaves
7.	*Neem*	*Azadirachta indica*	Indian lilac	Leaves
8.	*Nimboo*	*Citrus limon*	Lemon	Leaves
9.	*Peepal*	*Ficus riliigosa*	Sacred ficus	Leaves
10.	*Sagun/ Sagwan*	*Tectona grandis*	Teak	Leaves

Yield of plant materials in aqueous and ethanol extraction mediums

The yield of the extracts obtained per 50 ml of solvent is presented in Table 2. It is noticed from the table that the extract yields varied from 41.50 to 46 ml/50 ml in aqueous extracts. *Jamun* leaves exhibited maximum yield of 46ml/50 ml and Nimboo and Mehandi leaves exhibited lowest yield i.e. 41.50ml/50 ml.

Table 2: Yield of selected plant materials in different extraction mediums

S. No.	Plant Names	Yield of extracts (ml/ 50 ml)	
		Aqueous medium	Ethanol medium
1.	*Aonla*	45.00	45.50
2.	*Bael*	44.00	44.00
3.	*Curry patta*	45.00	45.00
4.	*Safeda*	45.00	44.00
5.	*Mehandi*	41.50	43.00
6.	*Jamun*	46.00	44.00
7.	*Neem*	40.00	44.00
8.	*Nimboo*	45.50	45.00
9.	*Peepal*	45.00	45.00
10.	*Sagun/ Sagwan*	41.50	44.00

In case of ethanol medium of extraction yield of extract ranged from 43 to 45.50 ml/50 ml. The maximum yield was for *aonla* leaves i.e. 45.50 ml/50 ml and the lowest yield was exhibited by Mehandi leaves (43 ml/50 ml).Whereas the yield variation in extracts was dependent upon the extraction solvent and plant material used.

Table 3: Total phenolic content of selected plant extracts

S. No.	Plants extracts	TPC in extracts (mg GAE/g)*	
		Aqueous extract	Ethanol extract
1.	*Aonla*	31.58	86.42
2.	*Bael*	04.21	11.14
3.	*Curry patta*	06.44	22.48
4.	*Safeda*	117.51	286.04
5.	*Mehandi*	01.59	11.17
6.	*Jamun*	10.74	36.18
7.	*Neem*	167.24	482.01
8.	*Nimboo*	23.71	42.06
9.	*Peepal*	02.65	15.66
10.	*Sagun/ Sagwan*	13.17	55.15

Total phenolic content of plant extracts in different mediums of extraction

Phenolics are effective antioxidants and antimicrobial agents. According to Cowan, 1999, plants are rich in a wide variety of secondary metabolites such as tannins, terpenoids, alkaloids and flavonoids which have been *in vitro* to have antimicrobial properties.

Table 3 highlight presence of total phenolic content of the selected plant extracts. It is observed that, in ethanol extract maximum phenolic content was found in *neem* leaves (482.01 mg GAE/g) followed by *safeda* leaves (286.04 mg GAE/g**)**, *aonla* leaves (86.42 mg GAE/g), Sagun leaves (55.15 mg GAE/g), Nimboo leaves (42.06 mg GAE/g), *jamun* leaves (36.18 mg GAE/g), curry patta leaves (22.48 mg GAE/g), *peepal* leaves (15.66 mg GAE/g), Mehandi leaves (11.17 mg GAE/g) and *bael* leaves (11.14 mg GAE/g).

In aqueous extract also maximum phenolic content were exhibited in *neem* leaves (167.24 mg GAE/g) followed by *safeda* leaves (117.51 mg GAE/g), *aonla* leaves (31.58 mg GAE/g), Nimboo leaves (23.71 mg GAE/g), Sagun leaves (13.17 mg GAE/g), *jamun* leaves (10.74 mg GAE/g), curry leaves (6.44 mg GAE/g), *bael* leaves (4.21 mg GAE/g), *peepal* leaves (2.65 mg GAE/g) and Mehandi leaves (1.59 mg GAE/g). In both the extraction medium *neem* leaves showed the highest total phenolic content. The result of the present study indicated that the ethanol extraction medium exhibited the highest amount of phenolic compounds as compared to aqueous extract (Table 3).

Bio assay

Natural extracts were screened for their antimicrobial activity against *Escherichia coli* and *Staphylococcus aureus* bacteria using agar well diffusion method and the zone of inhibition was measured.

Table 4: Antimicrobial activity of selected plant sources

S. No.	Plant extracts	Antimicrobial activity Zone of inhibition (mm)			
		Aqueous extract		Ethanol extract	
		S. aureus (gram +)	*E. coli (gram -)*	*S. aureus (gram +)*	*E. coli (gram -)*
1.	*Aonla*	12	11	18	16
2.	*Bael*	16	12	20	20
3.	*Curry patta*	12	07	20	18
4.	*Safeda*	16	09	23	20
5.	*Mehandi*	06	10	17	17
6.	*Jamun*	03	06	16	15
7.	*Neem*	04	06	18	16
8.	*Nimboo*	06	07	12	12
9.	*Peepal*	05	03	14	12
10.	*Sagun/ Sagwan*	08	04	13	11

Anti-bacterial activity <6mm: Week; 7-12 mm: Moderate; >12mm: Strong

The results of screening test of plant sources against *S. aureus (gram +)* and *E. coli* (gram -) bacteria are reported in Table 4. In aqueous medium of extraction among the different plant extracts the *bael* leaves showed the higher zone of inhibition i.e. 16 and 12 mm against *S. aureus (gram +)* and *E. coli* (gram -) bacteria, respectively followed by *aonla* leaves (12 and 11mm), *safeda* leaves (16 and 09 mm), curry patta (12 and 07 mm).

In ethanol medium of extraction, the *safeda* leaves extract showed the higher zone of inhibition i.e. 23 and 20 mm against *S. aureus (gram +)* and *E. coli* (gram -) bacteria, respectively followed by *bael* leaves (20 mm each), curry *patta* leaves (20 and 18 mm), *aonla* leaves and *neem* leaves exhibited same zone of inhibition i.e. 18 and 16 mm, respectively. The lowest zone of inhibition was shown by

Nimboo leaves extract which was 12 mm for both gram positive and gram negative bacteria.

Ahmmad *et al*, 1998 [1] stated that out of 82 plants, 56 exhibited antibacterial activity against one or more test pathogens and extracts of five plants showed strong and broad spectrum activity as compared to rest of 51 plant extracts which demonstrated moderate activity. On the whole the alcoholic extracts showed greater activity than their corresponding aqueous and hexane extracts.

Selvamohan *et al*, 2012 [7] reported that preliminary screening of different medicinal plants for antimicrobial activity depicted that the methanolic extract of *Phyllanthus niruri* exhibited maximum inhibitory zone (30 mm) against *Staphylococcus* sp. While the ethanol and aqueous extracts of *Murrya koenigii, Cynodon dachylon, Lawsonia inermis* and *Adha-thoda vasica* showed least inhibitory activity. The antimicrobial assay by agar-well diffusion method revealed that methanol extract of medicinal plants exhibited broad spectrum activity against tested isolates as compared to ethanol and aqueous extracts.

The results are also supported by the findings of Sood, 2014 which stated that the aqueous extracts of *Eucalyptus citriodora, Pinus roxburghii* and *Woodfordia fruticosa* exhibited highest zone of inhibition against *Staphylococcus aureus* and *Pseudomonas* spp. bacteria.

Conclusions

Plants are used by large proportion of the Indian population for curing one or the other health problems. The reasons for this may be true improvement of disease conditions after herbal treatment, no harmful side effects and cost effectiveness as compared to other forms of treatment. In the present study the results were encouraging as all the selected ten plants appeared to contain substances that had antimicrobial properties. It is concluded that aqueous extract of *jamun* leaves exhibited maximum yield of extraction (46 ml/50 ml) and in case of ethanol extracts, *aonla* leaves showed highest yield (45.50 ml/50 ml) in comparison to the other nine plant extracts tested and repeated maceration resulted in higher yield of plant extracts. Presence of total phenolic content was found to be maximum in *neem* leaves extract in both aqueous (167.24 mg GAE/g) and ethanol (482.01 mg GAE/g) mediums of extraction. In ethanol medium of extraction, all the selected plant extracts showed strong zone of inhibition against gram +ve and gram –ve bacteria.

References

1. Ahmad I, Mehmood Z, Mohmmad F. Screening of some Indian medicinal plants for their antimicrobial properties. Journal of Ethnopharmacology. 1998; 62(2):183-193.
2. Cowan MM. Plant products as antimicrobial agents. Clinical microbiology reviews. 1999; 12(4):564-582.
3. Gupta D, Bhaumik, S. Antimicrobial treatments for textiles. Indian Journal of Fibre and Textile Research. 2007; 32(6):254-263.
4. Hemaiswarya S, Kruthiventi AK, Doble M. Synergism between natural products and antibiotics against infectious diseases. Phytomedicine. 2008; 15(8):639-652.
5. Rastogi RP, Mehrotra BN. Glossary of Indian Medicinal Plants. National institute of science communication, New Delhi, 2002.
6. Rios JL, Recio MC. Medicinal plants and antimicrobial activity. Journal of Ethnopharmacol. 2005; 100: 80-84.
7. Selvamohan T, Ramadas V, Kishore SSS. Antimicrobial activity of selected medicinal plants against some selected human pathogenic bacteria. Advances in Applied Science Research. 2012; 3(5):3374-3381.
8. Sood A. Application of herbal extracts on covering fabric of sanitary napkins for bacterial resistance. Ph. D. Thesis, CCS Haryana Agricultural University, Hisar, 2005.