

Comparative Analysis of Antibacterial Property of Bamboo Fabric Treated with *Calotropis gigantea* and *Psidium guajava* leaf extract

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Abstract:

In this study two environmentally favourable herbs, *Calotropis gigantea* (**C. gigantea** or commonly known as Aakada) and *Psidium guajava* (**P. guajava** or commonly known as Guava), were extracted in methanol and used as antibacterial agents. Bamboo fabric was pretreated with harad (**Terminalia chebula**) as pre-moderating agent. The extract of *P. guajava* and *C. gigantea* were applied to the pretreated bamboo fabric in different concentrations of bioactive compound (100 µg/mg of fabric (10.0 % own weight of fabric (owf)), 50 µg/mg of fabric (5.0%owf), 25 µg/mg of fabric (2.5%), 12.5 µg/mg of fabric (1.25%)) by Spray-dry-cure process at 50±2 °C.

Bamboo fabric treated with *Calotropis gigantea* at 12.5 µg/mg of fabric weight showed 99.5 % and 87.6% antibacterial activity against *Staphylococcus aureus* (*S. aureus*) and *Escherichia coli* (*E. coli*) respectively. While *Psidium guajava* leaves extract shows 99.16 % antibacterial effect for *S. aureus* & 67.66% for *E. coli* for 50 µg/mg of fabric of fabric.

Keywords: antibacterial activity, Bamboo fabric, *Calotropis gigantea*, *Psidium guajava*, *Terminalia chebula*

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1. Introduction

Psidium guajava L. (guava), a fruit plant belonging to the family Myrtaceae, is found all over the world. Guava leaves, roots, and fruits have been used for the prevention and treatment of diarrhea [1, 2]. In several studies, guava showed significant antibacterial activity against common food borne diarrhea-causing bacteria such as *Staphylococcus spp.*, *Shigella spp.*, *Salmonella spp.*, *Bacillus spp.*, *E. coli*, *Clostridium spp.*, and food spoilage bacteria such as *Pseudomonas spp.* [2-5].

C. gigantea is a widely growing plant native to India, Indonesia, Malaysia, Philippines, Thailand, Sri Lanka and China, commonly known as milk weed or crown flower weed. *C. gigantea* is latex bearing plant and release the latex after a tissue injury. Plant latex is a mixture of alkaloids, tannins, gum, sugars, starch, resins and protein [20].

The focus of this investigation was to determine antimicrobial activity of *C. gigantea* and *Psidium guajava* L. against pathogenic microorganisms for bamboo treated sample in vitro testing.

A variety of textile items finished with antimicrobial finish created a greater awareness of the clean lifestyle practiced by the current generation. The inherent properties of textile fibres and the structure of the substrate favor the growth of microorganisms. When the fabric is worn close to the skin, cross-infection by pathogens results in the development of odor.

Regenerated bamboo fabric, also known as bamboo viscose or bamboo rayon, is a textile material produced from bamboo cellulose. Bamboo fabric itself has inherent antimicrobial properties due to the presence of a substance called "Bamboo Kun". Bamboo Kun is believed to provide resistance to bacteria and fungi, inhibiting their growth on the fabric's surface [6].

Antimicrobial agents can be found in extracts from a variety of medicinal plants and herbs, including leaves, stems, flowers, fruits, roots, seeds, and bark. These antimicrobial substances, which are mostly derived from plants include terpenoids, essential oils, alkaloids, lectins, polypeptides, and polyacetylenes [7]. These compounds display antioxidant as well as antibacterial effects. It would be advantageous for both medical industry personnel and the public; if recyclable, eco-friendly, long-lasting antimicrobial fabrics that are effective against germs are developed.

The antibacterial property of *P. guajava* and *C. gigantea* leaves extract on regenerated bamboo fabric has not been studied in detail or reported in scientific literature. Therefore, limited information is available on the potential effects of *Psidium guajava* and *Calotropis gigantea* leaves extract on regenerated bamboo fabric.

However, it is known that *P. guajava* and *C. gigantea* leaf possess antimicrobial properties due to the presence of bioactive compounds such as flavonoids, phenolics, and tannins. These compounds have been reported to exhibit antibacterial activity against various bacteria, including *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*, and *Salmonella* species.

The antibacterial action of *Psidium guajava* and *Calotropis gigantea* leaf extracts on bamboo fabric was highlighted in the current investigation [8].

2. Materials and Methods

2.1. Fabrics and Study area

This study was carried out at Shri Vaishnav Vidyapeeth Vishwavidyalaya Indore, India. For the application of active compounds two varieties of fabrics regenerated bamboo fabrics in the bleached state were used. The test fabrics were sourced online, and the construction particulars are calculated.

2.2. Extraction of active compound

Fresh leaves of *Psidium guajava* leaves were collected from Burhanpur, M.P, India & *Calotropis gigantea* leaves were collected from Sendhwa. The leaves were washed under running tap water and dust was removed from the leaves. The leaves were dried at room temperature for 15 days, after that the dried leaves are grounded into fine powder in a mixer grinder (Philips mixer grinder and HL7701) [9] and the active compound was extracted by the maceration method in methanol (8). The active compound in powder form was taken from beaker and the extracts were collected in plastic bottle and stored at 4 °C temperature and was utilized for the finishing of fabrics.

2.3. Test Organisms

In this investigation, two types of pathogens were used as test organisms. Table 1 had a list of the organism's name, culture type, and collection source. Working cultures were kept at 4°C and kept on Nutrient Agar slants. The cultures of the Nutrient Agar slants were occasionally transplanted onto new Nutrient Agar slants.

Table 1 - Test Organisms

Organisms	No. of type culture	Source
<i>Staphylococcus aureus</i>	2079	National Chemical Laboratory, Pune
<i>Escherichia coli</i>	2065	NCL, Pune

2.4. Preparation of Petri dish

Media The antibacterial activity of *P. guajava* and *C. gigantea* on bamboo cloth was evaluated using two different types of media.

2.4.1. Nutrient Agar

(NA) used as the foundational medium. Susceptibility testing using nutrient agar is generally reliable. It was made with dehydrated Nutrient agar.

2.4.2. Nutrient Broth

(NB) For the inocula production, nutrient broth was made. All the parts of the Nutrient Agar were present in the nutritional broth, but not the agar.

2.4.3. Preparation of Nutrient Agar

In accordance with the manufacturer's instructions, nutrient agar was made from a commercially available dehydrated foundation. It was allowed to cool at room temperature right after autoclaving. In order to achieve a uniform depth of around 4 mm, the freshly prepared and cooled medium was poured into a sterile glass flat-bottomed Petri plate on a level, horizontal surface. After allowing the agar medium to settle to room temperature, the plate was either utilised right away or kept in an incubator (37°C). A sterile cotton swab was dipped into the suspension after each plate had been infected with the test bacterium, which had previously been adjusted to the 0.5 McFarland standard solution [10].

2.4.4. Preparation of Inocula

From a cryogenic vial, one loopful of the inoculum of each test organism was transferred into 9 ml of sterile Nutrient Broth (NB), where it was cultured for 24 hours at 37°C. The Nutrient Agar plate was then streaked with one loop of the NB culture, which was then cultured there for 24 hours at 37°C. The test organisms' inocula was created by transferring 3 to 4 colonies from nutrient agar cultures into 9 ml of sterile NB, which was then incubated at 37°C for 12 to 18 hours to form colonies. After an overnight incubation, colonies were chosen and transferred to a glass tube containing sterile physiological saline and carefully vortexed. The turbidity of each bacterial suspension is then contrasted with that of the 0.5 McFarland standard solution (which has approximately 1.5 10⁸ CFU/mL)[10].

2.5. Preparation of Standard Fabric Sample

After washing of bamboo fabric, pre-mordanting has been done by using harad(5% own weight of fabric) at 50°C temperature, M: L (material to liqueur) =1:10 for 1 hour in laboratory water bath. Harad treated sample is used as standard sample [11].

2.6. Application of bioactive agent on test fabrics

Natural extracts of *P. guajava* and *C. gigantea* were applied on the bamboo fabric samples; by using different concentrations (100µg, 50µg, 25µg, 12.5µg /mg of fabric weight), which has been pretreated with Harad as a cross-linking agent, using Spray-dry-cure process. Fabric samples were prepared in the laboratory water bath at temperature 50± 2°C.

2.7 Evaluation of antimicrobial activity by the quantitative method

By using callipers to measure the zones of inhibition in millimetres close to the agar surface, antibacterial activity was measured, and the results were reported. The moment at which growth was completely stopped was considered to be the end point. In this experiment, the assay was carried out three times on each sample. Each sample performed testing for fabric weights of 100g, 50g, 25g, and 12.5g/mg. The untreated and the treated fabric samples were used for testing, and antimicrobial efficiency was determined in terms as percentage inhibition of antibacterial zone (PIAG)[12,13].

$$\text{PIAG(\%)} = (\text{Area of sample} - \text{Area of control}) / \text{Area of control} \times 100$$

2.8 Statistical analysis

The inhibition zones were calculated as means ± S. D. (n=3). The significance among different data was evaluated by analysis of variance (ANOVA) using Microsoft Excel program. Significant differences in the data were established by least significant difference at the 5% level of significance.

3. Result and Discussion

The disc (treated fabric sample) diffusion method was used in the current investigation to test *P. guajava* and *C. gigantea* for antibacterial activity against *S. aureus* and *E. Coli* bacteria [14]. By evaluating the existence or absence of the inhibitory zone at various concentrations, the results were evaluated.

This conclusion is logical given that the antibacterial activity of leaf extract is influenced by a number of variables, including: a) various species, b) composition and concentration, c) microbial species and its occurrence level, d) processing conditions and storage [15-16]. It should be mentioned that the concentration of the extracts from *P. guajava* and *C. gigantea* affected their antibacterial activity [17].

3.1. Fabric specification

Quality particulars of fabric are given in table 2.

Table 2 - Fabric Specification

Bamboo Fabric	Plain weave
EPI	108
PPI	90
Warp Count and weft Count	40's Ne and 40's Ne

3.2. Percentage Extraction of leaves in Methanol

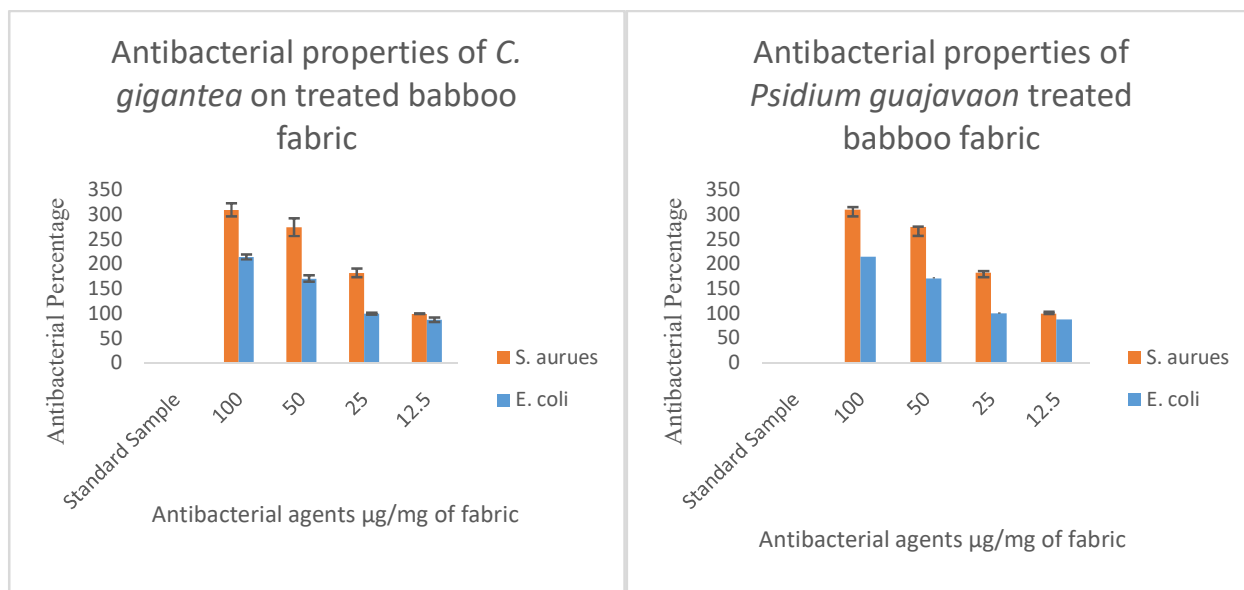
Crude or bioactive agents of *C. gigantea* and *P. guajava* leaves in methanol gives maximum extract for *P. guajava* leaves as compared to *C.gigantea*. It shows that maximum phytochemicals were extracted for *P. guajava* leaves. (Table.3)

Table 3 - Percentage Extraction of leaves in Methanol

S. No.	Antibacterial Agents (5gm)	Methanol(100ml) as extraction medium
1	<i>Calotropis gigantea</i>	6.5%
2	<i>Psidium guajava</i>	8.9%

3.3. Evaluation of antimicrobial activity by disc(fabric) diffusion test (quantitative) [18,19] by Antibacterial assay and ANOVA

The antimicrobial activity of the test fabrics treated with plant extracts was studied by the disc diffusion method. The level of antibacterial activity of fabrics treated with *Calotropis gigantea* and *Psidium guajava* plant leaves extracts was assessed by examining the extent of bacterial growth in the contact zone between the agar and the specimen and the width of the inhibition zone around the specimen. All the treated fabric samples showed significant inhibitory activity. It is observed that the zone of inhibition for the samples treated with *C. gigantea* leaf extract showed optimum antibacterial effect at 12.5 µg/mg of fabric sample while *P. guajava* leaf extract showed good results at 50 µg/mg of fabric sample as shown in graph 1 and 2. Table number 3 shows - Statistical Analysis of the effect of concentration of *C. gigantea* and *P. guajava* on antibacterial property (Linear Regression and ANOVA). This shows that the effect of concentration of *C.gigantea* on *S. aureus* and *E. coli* is 95% significant because *p*-value is lower than 0.05 while for *P.guajava*; null hypothesis was rejected as *p* value is less than 0.05 for *S. aureus* but there is no significant difference of concentration on antibacterial effect against *E.coli*.



Graph 1 - Antibacterial properties of *C.gigantea* on untreated Bamboo Fabric **Graph 2 - Antibacterial properties of *P.guajava* on treated Bamboo Fabric**

Table 3-Effect of concentration of *Calotropis gigantea* and *P.guajava* on antibacterial property

(Linear Regression and ANOVA)

	SS	df	F- Value	p-value	R Square	F crit
Effect of concentration of <i>Calotropis gigantea</i> on <i>S. aureus</i>	57686.72	1	10.98	0.016122	0.8106	5.98
Effect of concentration of <i>Calotropis gigantea</i> on <i>E. coli</i>	18608.42	1	7.28	0.0356054	0.933	5.98
Effect of concentration of <i>Psidium guajava</i> on <i>S. aureus</i>	4950.12	1	2.749	0.0148	0.995	5.98
Effect of concentration of <i>Psidium guajava</i> on <i>E. coli</i>	4950.125	1	2.74	0.148351	0.883	5.987378

4. Conclusion

Even though gram-negative bacteria were less sensitive to the antimicrobial property of guava leaves, it can be inferred from this experiment that Aakada leaves had stronger antimicrobial (antibacterial and antifungal) properties than Guava leaves extract. Considering their antimicrobial properties, polyphenolic chemicals may be the most likely source of the antifungal and antibacterial effects of guava and Aakada leaf extract. According to the research, both leaf extracts offer good anti-microbial properties for bamboo fabric. Therefore, using an herbal extract as an antibacterial treatment for textiles is a novel approach with a wide range of applications and commercial potential. The extract exhibited inhibition of bacterial growth for gram positive microbes *S. aureus* and gram-negative microbes *E. coli*. Bamboo fabric treated with *Calotropis gigantea* at 12.5 µg/mg of fabric weight showed 99.5 % antibacterial effect against *S. aureus* & for *E. coli* is 87.6% while *Psidium guajava* leaves extract showed 99.16 % antibacterial effect for *S. aureus* & 67.66% for *E. coli* for 50 µg/mg of fabric. These findings suggest that *P. guajava* and *C. gigantea* leaves extract has the potential to enhance antimicrobial properties of bamboo fabric, making it a promising outcome for antimicrobial textile applications. Hence, these leaves extract may be successfully employed in making antibacterial bandage fabric.

According to current research, the leaves of *P. guajava* and *C. gigantea* have the potential to be an excellent choice in the stalk for a natural antibacterial agent against topical wounds caused by *E. coli* and *S. aureus*.

5. Conflict of interest

Author declares there is no conflict of interest in publishing the article.

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