ANTI-BACTERIAL ACTIVITY OF BAMBOO (*Bambusa vulgaris*) SHOOT ETHANOLIC SKIN EXTRACT AGAINST *Staphylococcus aureus* and *Escherichia coli*

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ABSTRACT

The Bamboo (Bambusa vulgaris) Shoot is proven to have an antibacterial activity. However, the antibacterial activity of the Bamboo Shoot Skin has not yet been studied. It is for this reason that the researchers investigated its antibacterial property. The ethanolic extract from the Bamboo Shoot Skin was obtained and subjected to phytochemical screening to identify what components are present. The results revealed that saponins, flavonoids, and phenols were present. The antibacterial property is linked to their presence. The researchers formulated different concentrations of the extract and tested these through disc diffusion assay to measure the zone/s of inhibition (ZOI) against *Staphylococcus aureus* and *Escherichia coli*. Experimental test design was used in the study. The mean ZOI of the Experimental groups (25% w/v, 50% w/v, 75% w/v) against *Staphylococcus aureus* were 9 mm, 14 mm, 15 mm, respectively, and the mean of the ZOI of the Experimental group (25% w/v, 50% w/v, 75% w/v) against *Escherichia coli* were 8 mm, 11 mm, 11 mm, respectively. To test the significant difference, the researchers used One-way ANOVA. POST-HOC Tukey test was utilized to determine significant difference of variable to variable thereafter. Analysis revealed that there is significant difference on the antibacterial property of the ethanolic shoot skin extract of Bamboo (B. vulgaris) and the Positive control (Amoxicillin) against both *Staphylococcus aureus* and *Escherichia coli*; meaning, the Amoxicillin has better antibacterial activity. In terms of (ZOI) exhibited, there is no significant difference between the Experimental groups. However, based on the ZOI reference chart, Bamboo Shoot Skin ethanolic extract has an active and partially active antibacterial activities against S. aureus and E. coli, respectively. It is recommended to test the extract against other bacterial species, produce an antibacterial product from it, and test other parts or species of bamboo.

Key words: b*amboo shoot, Bamboo shoot skin, Escherichia coli, Staphylococcus aureus, zone of Inhibition, ethanolic extract*

INTRODUCTION

Staphylococcus aureus, gram-positive spherical cocci, “may be present among the indigenous flora of the skin, eye, upper respiratory tract, gastrointestinal tract, urethra, and, infrequently, vagina” (Hall & Woods, 2017). Infections arise from various causes like decrease or defects in the immunity, damage in the natural immunity defenses like skin and mucosa, and presence of artificial implants in the

body (Ibid.) It is known to cause skin infections, endocarditis, osteomyelitis, and toxic shock syndrome (Aryee & Edgeworth, 2017; Hall & Woods, 2017). In the recent times, there have been increasing rates of antibiotic resistance in S. aureus. Notably, in the Philippines in 2011, Mendes et al (2011) reported that methicillin-resistant S. aureus (MRSA) constituted 59% of S. aureus isolated compared to the report of Song et al (2011) with 38.1% (2004-2006).

Escherichia coli, gram-negative bacilli that are usually part of normal flora of the intestines, can incidentally cause diseases like urinary tract infection (90% of cases), diarrheal diseases (i.e., enterohemorrhagic [Shiga toxin producing], enterotoxigenic, enteroinvasive, enteropathogenic, enteroadherent), sepsis, and meningitis (leading cause in children) (Brooks, Caroll, Butel, Morse, & Mietzner, 2013; Hall & Woods, 2017). Also like *S. aureus*, *E. coli* also has an increasing case of antibiotic resistance (Research Institute for Tropical Medicine, 2016).

The Bamboo is abundant in the Philippines. In fact, in 2009, Philippines is the sixth largest exporter of bamboo worldwide (Mayuga, 2016). Bamboo has various uses. For example, its roots and leaves have been used as medical products. Its medicinal properties like anti-oxidant, anti-cancer, and anti-inflammatory have been supported by studies (Lee, Baek, & Han, 2001; Zhang, Jiao, Liu, Wu, & Zhang, 2008). The antibacterial property of leaves and shoot skins of another bamboo species, *Phyllostachys pubescens,* were shown in a number of studies (Tanaka, Shimizu, & Kondo, 2013; Afrin, Tsusuki, Kanwar, & Wang, 2012). Also, in the study of Owokomoto (2011), the leaves of B. vulgaris L. were found to have an antibacterial property. However, studies on the anti-bacterial activity of bamboo (*B. vulgaris*) shoot skin have not been reported. Moreover, bamboo shoot skins are usually discarded when it is cooked.

Hence, this study aimed to examine the anti-bacterial activity of Bamboo (*Bambusa vulgaris*) shoot skin extracts against *Staphylococcus aureus* and *Escherichia coli*.

Research Questions

This study aimed to examine the anti-bacterial activity of Bamboo (Bambusa vulgaris) shoot ethanolic skin extracts against food spoilage bacteria:

Specifically, it aimed to answer the following:

1. What are the phytochemical components present in the local variety of Bamboo (*Bambusa vulgaris*)?
2. Using various concentrations of the Bamboo (*Bambusa vulgaris*) shoot ethanolic skin extract, what is the zone of inhibition against the selected bacteria?
    a. 25% w/v

a.1. against *Staphylococcus aureus*
a.2. against *Escherichia coli*

b. 50% w/v
b.1. against *Staphylococcus aureus*
b.2. against *Escherichia coli*

c. 75% w/v
c.1. against *Staphylococcus aureus*
c.2. against *Escherichia coli*

3. Which concentration of Bamboo (*Bambusa vulgaris*) shoot ethanolic skin extract has the widest zone of inhibition?
    a. 25% w/v
    b. 50% w/v
    c. 75% w/v
4. Is there a significant difference on the antibacterial activity of Bamboo (*Bambusa vulgaris*) shoot ethanolic skin extract against *Staphylococcus aureus* between:
    a. Positive Control vs. Experimental Group (25% w/v)
    b. Positive Control vs. Experimental Group (50% w/v)
    c. Positive Control vs. Experimental Group (75% w/v)
    d. 25% w/v Experimental Group vs. 50% w/v Experimental Group
    e. 25% w/v Experimental Group vs. 75% w/v Experimental Group
    f. 50% w/v Experimental Group vs. 75% w/v Experimental Group
5. Is there a significant difference on the antibacterial activity of Bamboo (*Bambusa vulgaris*) shoot ethanolic skin extract against *Echerichia coli* between:
    a. Positive Control vs. Experimental Group (25% w/v)
    b. Positive Control vs. Experimental Group (50% w/v)
    c. Positive Control vs. Experimental Group (75% w/v)
    d. 25% w/v Experimental Group vs. 50% w/v Experimental Group
    e. 25% w/v Experimental Group vs. 75% w/v Experimental Group
    f. 50% w/v Experimental Group vs. 75% w/v Experimental Group

Hypotheses

1. There is no significant difference in the antibacterial activity between the Positive Control (Amoxicillin) and the Experimental Group at 25% w/v against *Staphylococcus aureus.*

2. There is no significant difference in the antibacterial activity between the Positive Control (Amoxicillin) and the Experimental Group 50% w/v against *Staphylococcus aureus*

3. There is no significant difference in the antibacterial activity between the Positive Control (Amoxicillin) and the Experimental Group 75% w/v against *Staphylococcus aureus*

4. There is no significant difference in the antibacterial activity between the Experimental Group 25% w/v) and the Experimental Group 50% w/v against *Staphylococcus aureus*
5. There is no significant difference in the antibacterial activity between the Experimental Group 25% w/v and the Experimental Group 75% w/v against *Staphylococcus aureus*
6. There is no significant difference in the antibacterial activity between the Experimental Group 50% w/v and the Experimental Group 75% w/v against *Staphylococcus aureus*
7. There is no significant difference in the antibacterial activity between the Positive Control (Amoxicillin) and the Experimental Group at 25% w/v against *Escherichia coli*
8. There is no significant difference in the antibacterial activity between the Positive Control (Amoxicillin) and the Experimental Group 50% w/v against *Escherichia coli*
9. There is no significant difference in the antibacterial activity between the Positive Control (Amoxicillin) and the Experimental Group 75% w/v against *Escherichia coli*
10. There is no significant difference in the antibacterial activity between the Experimental Group (25% w/v) and the Experimental Group 50% w/v against *Escherichia coli*
11. There is no significant difference in the antibacterial activity between the Experimental Group 25% w/v and the Experimental Group 75% w/v against *Escherichia coli*
12. There is no significant difference in the antibacterial activity between the Experimental Group 50% w/v and the Experimental Group 75% w/v against *Escherichia coli*

Significance of the Study

The researchers advocate the use of common local herbs or grass such as the Common Bamboo (*Bambusa vulgaris*) for the development of new organic or natural anti-bacterial agents having known the negative side effects of commercially known antibiotics. The Bamboo is well known for its usefulness. For example, its roots and leaves have been used as medical products. Also, studies on the medicinal properties of bamboo trees have shown the anti-oxidant, anti-cancer, and anti-inflammatory properties.

Literature Review

Traditional and Alternative Medicine Act of 1997

Using herbal medicine was a practice long time ago. The community utilizes their available resources in order to prevent and manage their diseases such as infections. Herbal medications are not only considered for their effective

and cost-effective way of preventing or managing infections, but also, due to their constant availability in the community. Republic Act 8423 (RA 8423) also known as the Traditional and Alternative Medicine Act of 1997, focuses on developing different traditional health-related management in the country. Drugs for prevention, cure, lessening signs and symptoms, diagnosis and maintaining a healthy lifestyle with lower price are needed to be explored and developed. The alternative medications undergo methods of proper compounding. (Jose N. Nolledo, 2015) This law encourages the indigenous people to share their traditional medicines and for people to study more about the safety and effectiveness of these alternative medicines. The health care professionals should become aware of these alternative medications and promote them to their patients. By this, our countrymen would have more alternative medicines than that of the existing drugs which are costly. The cheaper the medicines get, the more patients will comply with medication. (Jose N. Nolledo, 2015)

Common Bamboo (*Bambusa vulgaris*)

Bambusa vulgaris, commonly named as Bamboo or Kauayan-kiling, belongs to the family Poaceae. The plant is the most commonly encountered bamboo in cultivation in southeast asia and is grown panthropically,being the only Asian specie that is common in the new world. Bamboo is naked at the base, without spiny branches and grows up to 17 meters high, 15 centimeters in diameter. Bamboo grows almost everywhere in tropical countries like the Philippines, particularly in places close to water such as on riverbanks and by streams. Because it is easily found and so easily replaced, it is treated with an almost casual disregard and valued only lightly. It is indeed a relatively cheap raw material. The plant can easily be grown and harvested. It can be found in varied climates, from the cold mountainous regions to the hot tropical areas. (Stuart, 2016)

Traditional use of Bamboo (*Bambusa vulgaris*)

Bamboo plants play a significant role in traditional Asian medicine, especially in China and Japan. Biomedical investigations on the health-benefiting effects as well as toxicity of different parts and species of bamboo have been carried out worldwide since the 1960s. Also, the Common Bamboo (*Bambusa vulgaris*) is a folk remedy for kidney problems, phthisis, abortifacient, used to treat coughs and mucous and help alleviate fever and various inflammatory conditions. (Panee, 2015)

Phytochemical properties of Bamboo

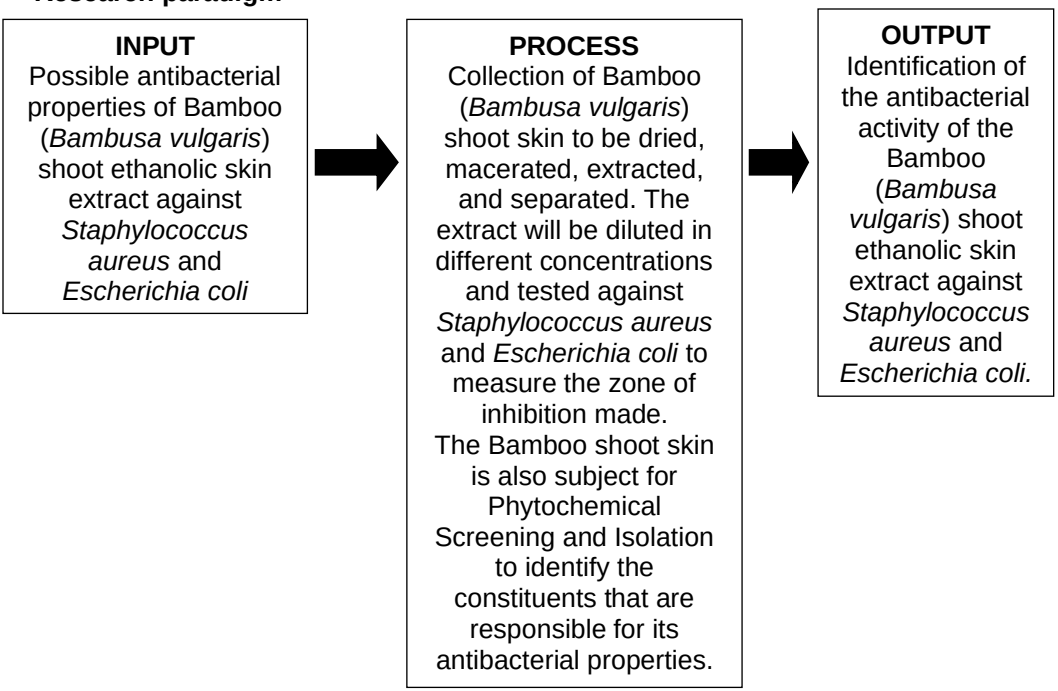
Preliminary phytochemical screening of solvent extracts viz. methanol and ethyl acetate of fermented *Bambusa balcooa* were carried out. Study reveals the presence of tannins, steroids, phenols, glycosides, flavanoids, carbohydrates and

proteins. FT-IR spectrum in the mid infrared region (4000-400cm-1) shows the presence of N-H, C=O, C-H, C=C and C-O, C-C, C-O bonds responsible for alkyl groups, methyl groups, alcohols, ethers, esters, ketones, carboxylic acid, anhydrides and deoxyribose confirming the qualitative results. (Singh, 2012)

Pharmacological uses of Bamboo

Bamboo leaves have antioxidant properties. Antioxidants help to keep the balance of free radicals and ward off any excess amount that can cause a large variety of problems like heart disease and even cancer. It is also great for healthy skin. In Nigeria, a drink of macerated leaves is taken against venereal diseases, while in DR Congo, the leaves form a part of preparations used for treatment of measles. Other ethno-medicinal usages include treatment of malaria fever, inflammations, ulcers, and wound healing. (Chongtham, 2011)

Research paradigm



METHODS

Research Design

The experiment was conducted at the Medical Technology Laboratory of University of Saint Louis, Tuguegarao City and Department of Science and Technology Provincial laboratory from October to November 2018.

Sample Technique

The botanical verification and authentication of the plant material was done by the Bureau of Plant Industry, Department of Agriculture Region 2 Office

1. The researchers minced one hundred grams of fresh bamboo shoot skin sample and soaked in a ninety five percent ethyl alcohol.
2. Muslin cloth was used to collect the filtrate.
3. The separation of the extract from the ethyl alcohol was done using rotary evaporator.

Data Gathering Procedure

The following procedures were conducted during the laboratory experimentation:

1. Collection, Preparation, and Processing of Bamboo shoot skin

1.1. Collection. The plant sample of shoot skin of Bamboo (*Bambusa vulgaris*) was collected in Balzain, Tuguegarao, Cagayan in the months of October to November.

1.2. Preparation. The Bamboo shoot skin was washed, and the extraneous dirt were removed with distilled water.

1.3. Processing/Extraction

1.3.1. The researchers minced and soaked about five hundred (500) grams of fresh bamboo shoot skin sample with a ninety five percent (95%) ethyl alcohol and covered with tightly closed container.

1.3.2. The sample was macerated for 24 hours and the filtrate was placed in a tightly sealed container.

1.3.3. The filtrate was put in a water bath until sticky gum-like viscous extract remained. This is the crude ethanolic extract.

1.3.4. The ethanolic crude extract was concentrated on a Rotary Vacuum Evaporator until viscous.

1.3.5. The extract was evaporated to dryness in a water bath at a controlled temperature below 60°C. Flame test

was done to confirm that ethanol has completely evaporated.

2. Phytochemical Screening

The Bamboo shoot skin extract was submitted for testing for flavonoids, tannins and saponins.

3. Semi-purification and Confirmation of the presence of flavonoid

3.1. Part of the residue was dissolved about 10 mL of 2M HCl and then an equal amount of ethyl acetate was added.

3.2. The aqueous acid layer was separated from the ethyl acetate layer using a separatory funnel.

3.3. The process was repeated until the aqueous acid layer is almost colorless.

3.4. Ethyl acetate extracts were combined and placed in an evaporating dish. The mixture was heated over a water bath until incipient dryness. Flame test was done to confirm that ethyl acetate has completely evaporated.

3.5. About 0.5 g of the residue was dissolved in ten (10)mL of 80% Ethyl alcohol to confirm the presence of flavonoids in the extract.

3.6. It was then filtered to eliminate any insoluble residue and the solution was divided in three test tubes labelled as A, B, and C reserving test tube A as the control.

3.7. The three labelled test tubes were subjected to the confirmatory chemical tests for flavonoids, namely Bate-smith and Metcalf method.

4. Preparation of Extract Concentrations

Dissolve the residue in distilled water based on the table below:

Concentration (%)	Extract (mg)	Distilled Water (uL)
25	500	1500
50	1000	1000
75	1500	500

5. Microbial Testing

The Bamboo (*Bambusa vulgaris*) shoot ethanolic skin extract was submitted to Department of Science and Technology and was tested for antibacterial activity using *Staphylococcus aureus* and *Escherichia coli.*

5.1. Preparation for the culture media:

5.1.1. About a desired amount of dehydrated culture media (Mueller's Hinton agar) was dissolved in appropriate volume of distilled water in an Erlenmeyer flask.
5.1.2. It was then heated warmly until the solution was cleared.
5.1.3. The mouth of the Erlenmeyer flask was plugged and was ready for sterilization.

5.2. Sterilization Method

5.2.1. The prepared culture media and clean dry petri dish test tube and flask (properly wrapped) that were used in the assay method was placed in an autoclave machine.
5.2.2. Operation of the autoclave: the autoclave was covered, and the air outlet was open. The autoclave was heated, and the air outlet is closed as soon as steam came out. The pressure was allowed to rise until the desired pressure or temperature was reached (121 degree Celsius or 15 LBS).
5.2.3. The temperature or pressure was maintained from 15-20 minutes.
5.2.4. After 15-20 minutes, the pressure was dropped to zero and the escape valve was opened before opening the autoclave.

5.3. Disc Diffusion Method

5.3.1. Antimicrobial susceptibility was determined following the Kirby-Bauer method adapted from the method compiled by (Guevarra, 2005).
5.3.2. A 100 µL of the filtrate was loaded into a disc, placed on Mueller-Hinton agar inoculated with E. coli and S. aureus and incubated at 37 °C for 24 hours for bacterial species while 24 – 72 hours at room temperature for bacteria.
5.3.3. The positive control used is Amoxicillin suspension and distilled water is used for negative control.
5.3.4. Measurement of the Zone of Inhibition
5.3.5. After the prescribed incubation time, the metal cylinders were removed, and the zones of inhibition were measured
5.3.6. The Zone of Inhibition was measured with a ruler or calliper.

5.3.7. If the perimeter of the Zone of Inhibition is clearly defined, measure its diameter; otherwise, measure its radius.

5.3.8. The ruler or caliper was held on the underside of the Petri dish and made a direct reading in millimeters. The size of the antimicrobial sample was included in the measurement.

5.3.9. The readings of multiple zones were taken to obtain an average size.

6. Waste Disposal

6.1. All the materials used during the experimentation in the University Laboratory were properly sterilized and biochemical were properly disposed of and coordinated with the research Adviser and the personnel in charge of the laboratory.

6.1.1. Waste dry products were placed inside color coded bags collected by Authorized personnel in charge of disposal.

6.1.2. Waste chemicals were diluted using running water and disposed through the sink.

6.2. Disposal of the materials and Biological wastes used during the Microbial experimentation in the DOST were taken charge by the authorities in the facility.

Ethical Considerations

This Research paper underwent evaluation by the University Research Ethics Board. The Researchers made sure that no living creatures were harmed during the experiment and that microbial organisms were carefully handled and disposed to avoid unwanted infections.

Data Analysis

The results gathered was tabulated and subjected to statistical treatment. To test the significant difference, the researchers used One-way ANOVA and an additional test or POST-HOC test was used to determine variable to variable significance testing.

RESULTS

Table 1. *Result of the Organoleptic Test of the Bamboo (Bambusa vulgaris) Shoot Ethanolic Skin Extract.*

Extract	Observation
Appearance	Clear Golden-brown Solution
Color	Golden-brown color

Smell	Strong, earthy odor

Table 1 shows that the appearance of the *Bamboo (Bambusa vulgaris) Shoot Ethanolic Skin Extract* has a clear golden brown color and has a strong earthy odor.

Table 2. *Phytochemical Screening.*

Chemical Constituents	Results
Saponins	+
Tannins	-
Flavonoids	+
Phenols	+
Alkaloids	+

Legend: Presence (+), Absence (-)

Based on the Phytochemical Test done by the researchers Cuzzamu, et al. who conducted a study on the anti-inflammatory activity of Bamboo (Bambusa vulgaris) leaf extract, table 2 shows that saponins, flavonoids, and phenols are present in the extract while tannin is absent.

Table 3. *Antibacterial activity of Bamboo (Bambusa vulgaris) Shoot Skin Ethanolic Extract and Amoxicillin against Staphylococcus aureus and Escherichia coli.*

ANTIBACTERIAL ANALYSIS									
Sample Number	**Sample Description**	**ZONE OF INHIBITION (mm)**							
		Escherichia coli				**Staphylococcus aureus**			
		R1	R2	R3	**MEAN**	R1	R2	R3	**MEAN**
MIC-1606	**25 % Common Bamboo Ethanolic Extract**	7	8	9	**8**	11	8	8	**9**
MIC-1607	**50% Common Bamboo Ethanolic Extract**	9	10	14	**11**	16	13	12	**14**
MIC-1608	**75% Common Bamboo Ethanolic Extract**	10	12	11	**11**	15	15	15	**15**
MIC-1609	**Positive Control**	42	36	46	**41**	64	57	55	**59**
Negative Control: Distilled Water		6	6	6	**6**	6	6	6	**6**

DOST interpretation:
< 10 mm = inactive
10-13 mm = partially active

14-19 mm = active
>19 mm = very active

Table 3 shows the susceptibility pattern that the positive control, Amoxicillin, has the highest mean which indicates very active against *Staphylococcus aureus* .The zone of inhibition of all the experimental groups of Bamboo (*Bambusa vulgaris*) shoot ethanolic skin extract exhibits a partial activity against *Escherichia coli* while it exhibits an activity against *Staphylococcus aureus.*

Table 4. *Descriptive Statistics of the Disc Diffusion Assay Results of Bamboo (Bambusa vulgaris) Shoot Ethanolic Skin Extract against Staphylococcus aureus.*

(I) Treatment	(J) Treatment	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
25% Extract	50% Extract	-4.333	2.236	.286	-11.49	2.83
	75% Extract	-6.667	2.236	.068	-13.83	.49
	Positive Control	-49.333*	2.236	.000	-56.49	-42.17
50% Extract	25% Extract	4.333	2.236	.286	-2.83	11.49
	75% Extract	-2.333	2.236	.730	-9.49	4.83
	Positive Control	-45.000*	2.236	.000	-52.16	-37.84
75% Extract	25% Extract	6.667	2.236	.068	-.49	13.83
	50% Extract	2.333	2.236	.730	-4.83	9.49
	Positive Control	-42.667*	2.236	.000	-49.83	-35.51
Positive Control	25% Extract	49.333*	2.236	.000	42.17	56.49
	50% Extract	45.000*	2.236	.000	37.84	52.16
	75% Extract	42.667*	2.236	.000	35.51	49.83

Table 4 shows that there is a significant difference on the disc diffusion assay of the positive control versus all other extracts against staphylococcus aureus.

Table 5. *Analysis of Variance of the Treatment Groups against Staphylococcus aureus*

Treatment		N	Mean	F-Ratio	p - value	Decision at 0.05 level
Zone of Inhibition	25% Extract	3	9.33	211.596	.000	**Reject Ho**
	50% Extract	3	13.67			
	75% Extract	3	16.00			
	Positive Control	3	58.67			

Table 5 indicates that at 0.05 level of significance, there is a significant difference among the extracts and positive control on their zones of inhibition against Staphylococcus aureus. Specifically, the positive control was most effective as compared with the plant extracts.

Table 6. *POST-HOC Test of the Treatment Groups against Staphylococcus aureus*

Treatment		Mean Difference	p-value	Decision
25% Extract	50% Extract	-4.333	0.286	Accept Ho
	75% Extract	-6.667	0.068	Accept Ho
	Positive Control	-49.333*	0.000	Reject Ho
50% Extract	75% Extract	-2.333	0.730	Accept Ho
	Positive Control	-45.000*	0.000	Reject Ho
75% Extract	Positive Control	-42.667*	0.000	Reject Ho

Table 6 indicates that there is a significant difference between 25% extract and positive control with a mean difference of -49.333, This means that the zone of inhibition for 25% extract is less than that of the positive control. There is also a significant difference between 50% extract and Positive Control with a mean difference of -45.000 which means that the zone of inhibition for 50% extract is less than that of the positive control. And lastly, there is a significant difference between 75% extract and positive control, with a mean difference of -42.667 and means that the zone of inhibition for 75% extract is less than that of the positive control.

Table 7. *Descriptive Statistics of the Disc Diffusion Assay Results of Bamboo (Bambusa vulgaris) Shoot Skin Extract against Escherichia coli.*

(I) Treatment	(J) Treatment	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
25% Extract	50% Extract	-3.000	2.392	.613	-10.66	4.66
	75%	-3.000	2.392	.613	-10.66	4.66

	Extract					
	Positive Control	-33.333*	2.392	.000	-40.99	-25.67
50% Extract	25% Extract	3.000	2.392	.613	-4.66	10.66
	75% Extract	.000	2.392	1.000	-7.66	7.66
	Positive Control	-30.333*	2.392	.000	-37.99	-22.67
75% Extract	25% Extract	3.000	2.392	.613	-4.66	10.66
	50% Extract	.000	2.392	1.000	-7.66	7.66
	Positive Control	-30.333*	2.392	.000	-37.99	-22.67
Positive Control	25% Extract	33.333*	2.392	.000	25.67	40.99
	50% Extract	30.333*	2.392	.000	22.67	37.99
	75% Extract	30.333*	2.392	.000	22.67	37.99

Table 7 shows that there is a significant difference on the disc diffusion assay of the positive control versus all other extracts against Escherichia coli.

Table 8. *Analysis of Variance of the Treatment Groups against Escherichia coli*

	Treatment	N	Mean	F-Ratio	p -value	Decision
Zone of Inhibition	25% Extract	3	8.00	86.485	.000	Reject Ho
	50% Extract	3	11.00			
	75% Extract	3	11.00			
	Positive Control	3	41.33			

Table 8 indicates that at 0.05 level of significance, there is a significant difference among the extracts and positive control on their zones of inhibition against Escherichia coli. Specifically, the positive control was most effective as compared with the plant extracts.

Table 9. *POST-HOC Test of the Treatment Groups against Escherichia coli*

Treatment	Treatment	Mean Difference	p-value	Decision
25% Extract	50% Extract	-3.000	0.613	Accept Ho
	75% Extract	-3.000	0.613	Accept Ho

	Positive Control	-33.333*	0.000	Reject Ho
50% Extract	75% Extract	0.000	1.000	Accept Ho
	Positive Control	-30.333*	0.000	Reject Ho
75% Extract	Positive Control	-30.333*	0.000	Reject Ho

Table 9 indicates that there is a significant difference between 25% extract and positive control with a mean difference of -33.333. This means that the zone of inhibition for 25% extract is less than that of the positive control. There also a significant difference between 50% extract and positive control with a mean difference of -30.333 which means that the zone of inhibition for 50% extract is less than that of the positive control. And lastly, there is a significant difference between 75% extract and positive control, with a mean difference of -30.333. This means that the zone of inhibition for 75% extract is less than that of the positive control.

DISCUSSION

The research study intended to determine the antibacterial activity of the ethanolic shoot skin extract of Bamboo (Bambusa vulgaris). To attain the objective of the study, the researchers did phytochemical screening and microbial testing (Disc Diffusion Method). The phytochemical screening revealed that saponins, flavonoids, and phenols are present in the extract while tannins are absent. Flavonoids are well known as antibacterial agents against a wide range of pathogenic microorganisms. With increasing prevalence of untreatable infections induced by antibiotic resistance bacteria, flavonoids have attracted much interest because of the potential to be substitute for antibiotics (Xie, Y. 2014).

Amoxicillin was used as a reference standard for comparing the antibacterial potential of the ethanolic shoot skin extract of Bamboo (*Bambusa vulgaris*). Results showed that the extract has zones of inhibition that shows microorganisms have less susceptibility than the positive control. Through paper disc diffusion assay, the average mean of the zone of inhibition of the different concentrations (25% w/v, 50% w/v, 75% w/v) against *Staphylococcus aureus* was 9 mm, 14 mm, 15 mm, respectively, was less effective than the positive control and the average mean of the zone of inhibition of the different concentrations (25% w/v, 50% w/v, 75% w/v) against *Escherichia coli* was 8 mm, 11 mm, 11 mm respectively, which was also less effective to that of the positive control. Zone of inhibition studies showed significant ($p < 0.05$) inhibition capacity of different concentrations of plant extract against *Staphylococcus aureus* and *Escherichia coli*. Analysis of the obtained results shows significant antibacterial activity of the ethanolic shoot skin extract of Bamboo (*Bambusa vulgaris*) to *Staphylococcus aureus* and *Escherichia coli*.

CONCLUSION

Bamboo shoot skin ethanolic extract has weaker antibacterial activity copmpared with the commercially prepared amoxicillin. Nonetheless, based on the zone of inhibition reference chart, bamboo shoot skin ethanolic extract has active and partially active antibacterial activities against s. aureus and e. coli, respectively.

RECOMMENDATIONS

Based on the foregoing findings of the study, the following are recommended:

1. To produce/formulate an anti-bacterial product using the Bamboo (*Bambusa vulgaris*) shoot skin extract.
2. To further test the extract on bacterial species other than what was used in this study.
3. To compare the antibacterial activity of bamboo shoot skin with other commercially prepared antibiotics.
4. To utilize other bamboo species other than *Bambusa vulgaris*.

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