

**Research Paper** 

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# « ANTIBACTERIAL ACTIVITY OF Ricinodendronheudelotii SEED EXTRACT ON SELECTED BACTERIA »

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Abstract: This study investigated the invitroantibacterial activities of ethanol seed extract of Ricinodendronheudelotii against Staphylococcusaureus, Escherichiacoli and Bacilluscereus. The extracts were tested against the bacteria using the disc diffusion method at various concentrations of400mg/kg, 800mg/kg, 1200mg/kg, 1600mg/kg and the undiluted. Data obtained showed resistance of Staphylococcus aureus and Bacillus cereus to all concentrations of the seed extract except at undiluted concentration with inhibition zones at 10mm and 14mm respectively. Escherichia coli was inhibited by the seed extract at 16mm each for 400mg/kg, 800mg/kg and the undiluted concentration at 13mm. Staphylococcusaureus, Escherichiacoli and Bacilluscereus showed no significant inhibition at lower concentration of the extract while the undiluted extract showed inhibition against the three organisms used at 10mm, 13mm and 14mm respectively. It can be concluded that the ethanol seed extract of Ricinodendronheudelotican be used as a potential antimicrobial therapy at undiluted concentration against Staphylococcusaureus, Escherichiacoli and Bacilluscereus.

Keywords: Ricinodendronheudelotii, Antibacterial, Staphylococcusaureus, Bacilluscereus, Escherichiacoli.

#### Introduction:

Bacteria exist as unicellular, or cell clusters. They are widespread in nature and are the major causes of disease and infections such as skin infection, food poisoning and food borne illnesses, as well as causing serious harm to the human body e.g. *Staphylococcusaureus*. However, Bacteria are important to human in many ways, as they aid in fermentation of foods, treat sewage, produce fuel, enzymes and other bioactive compounds etc<sup>11</sup>. They are also essential tools in biology as model organisms, make up the human micro biota including the essential gut flora. They are the pathogens responsible for many infectious diseases, and as such are the target of hygiene measures. Bacteria rapidly reproduce and are also able to freely exchange genes through conjugation, transformation and transduction, even between widely divergent species<sup>1</sup>.

*Staphylococcusaureus* is a gram-positive, round shaped bacterium that is a member of normal flora of the body, frequently found in the nose, respiratory track and on the skin. It is the most common cause of skin infection

including abscesses, respiratory infections and food poisoning.

*Escherichiacoli* also known as *E.coli* is a gram-negative facultative anaerobic rod –shaped which is commonly found in the lower intestine of warm-blooded organism (endotherms). Its strains are harmless but some serotypes can cause serious food poisoning in their host and occasionally responsible for product recalls due to food contamination.

*Bacilluscereus* is a Gram-positive, rod-shaped, aerobic, motile, beta hemolytic bacterium commonly found in soil and food; some strains are harmful to human and cause food borne illness.

*Ricinodendronheudelotii*is a plant with food, medicinal and industrial values<sup>9</sup>. The phytochemical screening of the seed *of Ricinodendronheudelotii*as reportedrevealed the phytochemical constituent of the plant, suggesting its curative potency for various diseases, its potential use therapeutically and in pharmaceutical industries<sup>7</sup>.

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### Materials and methods:

### **Collection of Test Organisms**

Cultured and identified bacteria (*Staphylococcusaureus*, *Escherichiacoli* and *Bacilluscereus*) were obtained from Braithwaite Memorial Specialist hospital in Port Harcourt, Rivers State and were sub- cultured to get stock culture of the organisms.

# **Antibacterial Susceptibility Testing**

Three already identified isolates were used for the Bacterial susceptibility test: *Escherchia coli, Staphylococcus aureus* and *Bacillus cereus*.

Selective media were prepared for the growth of the various Bacteria namely; Maintol salt agar, Macconkey agar and Nutrient agar. The media was used to sub-culture *S.aureus, E coli* and *B. cereus* respectively.

#### **Media Preparation**

MacConkey Agar, Manitol Salt Agar and Nutrient Agar were prepared according to the manufacturers recommended procedure<sup>3</sup>.

The solidified media were dried in an oven at  $65 - 70^{\circ}$ C to eliminate moisture on the surface of the media before streaking for sub-culture.

**Sub-culturing of Isolates:** The identified isolates were subcultured and stored in glycerol. The 24 hour culture was used as test organism for the anti-bacterial susceptibility testing.

# **Antibacterial Susceptibility Testing**

Kirby-Bauer disc diffusion test method of 1996 was adopted for the susceptibility testing. The procedure involved the suspension of a spoonful of the confluent growth of the pure culture. Macfar land standard was compared with the test tube and the density of the test suspension was adjusted to that of the standard by adding more bacteria or more sterile saline. The plate was inoculated by dipping a sterile swab into the inoculums. Excess inoculums were removed by pressing and rotating the swab against the side of the tube above the level of the liquid. The swab was streaked all over the surface of the medium three times, rotating the plate through an angle of 60 degree after each application. The inoculums were left to dry for 5 minutes at room temperature with the lid closed. The appropriate antimicrobial impregnated discs were placed on the surface of the agar using a flamed sterilized forceps. Each disc was gently pressed down to ensure complete contact with the agar surface and inoculated at 35 degree for 24 hours. The diameter of each zone was measured using a meter rule and recorded in mm.

# Experimental Design for antibacterial activity of ethanol seed extract.

The experimental set up comprised of the test organism and the seed extract which was done at varying concentrations. The concentration and organism to be tested were arranged in six (6) Groups.

**GROUP 1:** Application of a spectrum antibacterial drug (CIPROFLOXACIN) to the test organisms in sets of A, B C: *Staphylococcus aureus, Escherichia coli and Bacillus cereus* respectively. Ciprofloxacin was used as a positive control and sterile distilled water was used as a negative control.

**GROUP 2:** Four percent concentration of the extract was applied to the test organisms in varying sets (A, B, C) of *S. aureus, E. coli, and B. cereus* respectively and incubated at  $35^{\circ}$ C for 24 hours.

**GROUP 3:** Eight percent concentration of the extract was applied to the test organisms *S.aureus, E. coli, and B. cereus* in set of A, B, C respectively and incubated at  $35^{\circ}$ C for 24 hours.

**GROUP 4:** Twelve percent concentration of the extract was applied to the test organisms in sets of A, B, C respectively and incubated at  $35^{\circ}$ C for 24 hours.

**GROUP 5:** Sixteen percent concentration of the extract was applied to the test organisms in sets of A, B, C and incubated at  $35^{\circ}$ C for 24 hours.

**GROUP 6:** 100% undiluted concentration of the extract was applied to the test organisms in sets of A, B, C and incubated at  $35^{\circ}$ C for 24 hours.

After 24 hours of incubation, the set up was observed for zones of inhibition and recorded.

## RESULTS

Table 1. Susceptibility and resistance of the test organisms to the seed extract.			
	Set A. <i>S.aureus</i> (mm)	Set B <i>, E.coli</i> (mm)	Set C. <i>B.cereus</i> (mm)
GROUP 1	40	27	33
GROUP 2	_	16	_
GROUP 3	_	16	_
GROUP 4	_	_	_
GROUP 5	_	_	_
GROUP 6	10	13	14

Table 1: Susceptibility and Resistance of the test organisms to the seed extract.



Fig 1: Effect of ethanol seed extract of R. heudelotii on Bacterial activity of S.aureus, E. coli and B. cereus.

## Discussion

The effect of the ethanol seed extract of *Ricinodendronheudelotii* on the microbial activity of *Staphylococcusaureus, Escherichiacoli* and *Bacilluscereus*was carried out. The selected bacteria for the research work are known for their high antimicrobial resistant patterns.

From the results in Table 1, all the test bacteria in Group 1 which served as the positive control, were susceptible to the antibiotics (Ciprofloxacin), showing a clear zone of inhibition. *Staphylococcusaureus* was shown to be more susceptible with 40mm zone of inhibition, followed by *Bacilluscereus* 33mm, and *Escherichiacoli* 27mm.

Results in Group 2 indicatesthat*Escherichiacoli* was susceptible (16mm zone of inhibition) while *Bacilluscereus* and *Staphylococcusaureus* were resistant, Group 3 representing the extract at 8% concentration revealed the susceptibility of *Escherichiacoli* with 16mm zone of inhibition, while *Staphylococcusaureus* and *Escherichiacoli* were resistant to the treatment. Group 4 and 5 represents the extract at 12% and 16% concentrations respectively, the test result showed resistance of the organisms to the extract at both concentrations.

Group 6 represents the seed extract at undiluted concentration. The test organisms were susceptible showing zone of inhibition of *Staphylococcus aureus*, 10mm, *Escherichiacoli*, 13mm and *Bacilluscereus* 14mm.

Staphylococcusaureus, Pseudomonasaeruginosas, Escherichiacoli, Bacilluscereus when tested with oil extract of Ricinodendronheudelotiias reported by Olasehindeet al.<sup>8</sup>, were resistant at all concentrations of the extract <sup>8</sup>whereas this study have revealed the susceptibility of *Escherichiacoli* to the seed extract of Ricinodendronheudelotii plant while Staphylococcusaureus and Bacilluscereus were resistant at low concentration. However, all three organisms showed susceptibility at undiluted concentration of the plant seed extract, although the zone of inhibition was smaller compared to the control of Ciprofloxacin. This could be as a result of altered purity of the isolated organisms.

Therefore this research suggests the potency of the undiluted extract to inhibit microorganism. This could be be due to the presence of some phytochemicals in the seed extract of *Ricinodendronheudelotii*, which has part of its potencies as anti-inflammatory, antimicrobial and anti-oxidant<sup>7</sup>.

### CONCLUSION

This study has revealed the potency of ethanol seed extract of *Ricinodendronheudelotii* against Bacterial activities at undiluted concentration.

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