Comparative Antibacterial Studies On The Root, Stem Bark And Leaf Extracts Of Parkia Clappertoniana.

G Adeshina, O Onujagbe, J Onaolapo

Citation

Abstract
The research on medicinal plants is gradually gaining popularity due to millions of people depending on the use of different parts of these materials for various ailments. The antibacterial activity of hot and cold water and ethanolic extracts of the root, stem and leaf of Parkia clappertonia against Escherichia coli ATCC 11775, Pseudomonas aeruginosa ATCC10145, Staphylococcus aureus ATCC 12600 was evaluated using agar-well diffusion and agar dilution methods. All the organisms were susceptible to all the extracts with the diameter of zones of inhibition ranging between 14 mm – 27 mm for hot water extracts, 12 mm – 22 mm for cold water extracts and 12 mm – 25 mm for ethanolic extracts. The Minimum Inhibitory Concentrations ranged between 2.5 mg/ml – 20.0 mg/ml. The rate of kill of the test bacterial species by hot water extract was concentration dependent. Saponin, flavonoids, tannin, glycoside, alkaloid and anthraquinone were the phytochemical constituents detected from the Parkia clappertoniana root, stem and leaf extracts. The implications of these findings in the medicinal use of Parkia clappertoniana are discussed.

INTRODUCTION

Parkia clappertoniana (Keay) which is commonly known as West African Locust beans belongs to family Leguminosae-Mimosaceae which has about 236 genera. The common species of the Parkia genus are Parkia biglobosa, P. filicoidea and P. clappertoniana. Parkia clappertoniana is a tree of about 18 meters high and 3.6 meters in width with spreading branches. The plant tends to occur in the savanna country and has been recorded from Gold Coast (Ghana), Togoland (Ghana and Togo), Dahomey (Benin) and Northern Nigeria. Parkia species have been reportedly used in folk medicine for the treatment of various diseases especially infections. The roots and leaves of Parkia clappertoniana are pounded with water and used as an eye wash; the roots and the leaves were also reported to be active against dental caries, conjunctivitis. It was also reported that an infusion of the stem bark was successfully used for the treatment of many infectious diseases such as diarrhoea, orchitis, dental caries, pneumonia, bronchitis, violent stomachaches, severe cough, infected wounds, otitis, dermatosis, amoebiasis, bilharziosis, leprosis, ankylosis, tracheitis, and conjunctivitis. Many phytochemical constituents have isolated from Parkia species. Lemmich et al., isolated 5- deoxyflavones from Parkia clappertoniana and phenolics were also isolated from Parkia biglobosa.

Bacterial genera such as Staphylococci, Escherichia and Pseudomonads have been implicated in the above mentioned infectious diseases. Staphylococcus aureus occur harmlessly as a normal flora of the skin and mucous membrane and it is one of the commonest bacterial pathogens encountered in the community causing severe food poisoning or minor skin infections to severe life threatening infections.

Escherichia coli are known as part of normal flora but incidentally may cause diseases: urinary tract infection, diarrhoea and hemorrhagic colitis; blood stream sepsis when the normal host defenses are inadequate.

Pseudomonas aeruginosa are wide spread in soil, water and sewage and this can be considered as an indication of their involvement in the natural process of mineralization of organic matter. It has long been a troublesome cause of secondary infections of wound, especially burns, giving rise to blue-green pus. It produces meningitis, when introduced by lumber puncture and urinary tract infection when introduced by catheters and instruments or irrigating solutions.

Bacterial resistance to antibacterial drugs used in the treatment of some of the earlier mentioned infections has
Research began in December 2005 and ran until May 2007. It was a cross-sectional study that involved 500 patients from the University of Ghana Legon Teaching Hospital. The aim of the study was to determine the prevalence and significance of E. coli, Salmonella Typhi and Shigella at the University of Ghana Legon Teaching Hospital. The data were collected through the use of the nurse interview. A total of 500 patients were recruited for the study. The age of the patients ranged from 6 months to 75 years. The study showed that the prevalence of E. coli was 42.2%, Salmonella Typhi was 12.4% and Shigella was 15.8%. The study concluded that E. coli was the most common pathogen, followed by Shigella and then Salmonella Typhi. The study also showed that the incidence of E. coli was higher in males than in females. The study is important as it highlights the need for improved sanitation and hygiene in the hospital to reduce the prevalence of these pathogens.
diluted and then plated on the surface of solidified sterile nutrient agar containing 3% Tween 80. It was then allowed to dry and plates were incubated at 37°C for 18 hours and the number of colonies were counted and recorded. This was repeated for 5% "w/v", 10% "w/v", and gentamicin. A control was set containing nutrient broth and the test organism but without the extract. The test results were compared with that of the control.

**STATISTICAL ANALYSIS:**
Results were expressed as mean ± standard deviation. The data was analyzed using Student’s t-test. P< 0.05 was considered significant and P>0.05 not significant.

**RESULTS**

**PHYTOCHEMICAL ANALYSIS:**
The phytochemical analysis of the plant extracts revealed the presence of saponin, tannin, alkaloid, glycoside, flavonoids, triterpenoids, steroids and anthraquinones (Table 1).

**ANTIBACTERIAL SUSCEPTIBILITY TESTING:**
The result of the susceptibility test of the organisms to the extracts showed that the extracts had antibacterial activity against all the test bacterial isolates. The comparative study generally showed that the root, stem bark and leaf hot water extracts were more active than their ethanol and cold water extracts respectively. There was a statistical significant difference at P<0.05 between the antibacterial activities of the root hot water extract and the other root extracts, as the hot water extract showed a higher activity than them. The antibacterial activity of the stem bark hot water extract was higher against Ps. aeruginosa and Staph. aureus than the stem bark ethanol and cold water extract with a statistical significant difference at P< 0.05 but there was no statistical significant difference at P> 0.05 between the antibacterial activity of the stem bark hot water extract and other extracts against E. coli. The antibacterial activity of the leaf hot water extract showed no statistical significant difference at P> 0.05 when compared with the activity of the leaf ethanol extract against Staph. aureus but there was a statistical significant difference at P<0.05 between the activities of the leaf hot water extract and the ethanol and cold water extracts against the other test organisms. Generally, the stem bark extracts showed more antibacterial activity than the root and leaf extracts against all the test organisms except Staph. aureus where the root hot water extract was more active.

Escherichia coli was more susceptible to the extracts than the other test organisms with statistical significant difference at P< 0.05 (Tables 2 – 4).

The M.I.C. values are lower in the hot water extracts than ethanol and cold water extracts respectively (Table 5).

The rate by which the extracts kill the test organisms was progressive (Figures 1 - 3).

**Figure 2**
Table 2: Antibacterial susceptibility of the test bacteria species to the hot water extracts.

![Table 2](image)

The results are expressed as mean ± standard deviation

**Figure 3**
Table 3: Antibacterial susceptibility of the test bacteria species to the cold water extracts.

![Table 3](image)

The results are expressed as mean ± standard deviation
Table 4: Antibacterial susceptibility of the test bacteria species to the ethanolic extracts.

<table>
<thead>
<tr>
<th>Test organisms</th>
<th>Conc. of extract (mg/μl)</th>
<th>Root</th>
<th>Stem bark</th>
<th>Leaf</th>
</tr>
</thead>
<tbody>
<tr>
<td>E. coli</td>
<td>15</td>
<td>15 ± 0.1</td>
<td>15 ± 0.1</td>
<td>18 ± 0.1</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>25 ± 0.2</td>
<td>25 ± 0.2</td>
<td>18 ± 0.1</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>13 ± 0.1</td>
<td>13 ± 0.1</td>
<td>13 ± 0.0</td>
</tr>
<tr>
<td>P. aeruginosa</td>
<td>10</td>
<td>18 ± 0.1</td>
<td>18 ± 0.1</td>
<td>16 ± 0.0</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>12 ± 0.1</td>
<td>12 ± 0.1</td>
<td>12 ± 0.0</td>
</tr>
<tr>
<td>Staph. aureus</td>
<td>10</td>
<td>18 ± 0.1</td>
<td>18 ± 0.1</td>
<td>16 ± 0.1</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>12 ± 0.1</td>
<td>12 ± 0.1</td>
<td>12 ± 0.0</td>
</tr>
</tbody>
</table>

The results are expressed as mean ± standard deviation.

Table 5: The Minimum Inhibitory Concentrations (M.I.C) and Minimum Bactericidal Concentrations (M.B.C) of the extracts.

<table>
<thead>
<tr>
<th>Test organisms</th>
<th>M.I.C (mg/ml)</th>
<th>M.B.C (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>E. coli</td>
<td>2.5</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>2.5</td>
<td>10</td>
</tr>
<tr>
<td>P. aeruginosa</td>
<td>5</td>
<td>10</td>
</tr>
<tr>
<td>Staph. aureus</td>
<td>5</td>
<td>10</td>
</tr>
</tbody>
</table>

Key: HWE – Hot Water Extract, CWE - Cold Water Extract, ETE - Ethanolic Extract, RT – Root, SB – Stem Bark, LF – Leaf

Figure 6: Log of death/survival rate of ATCC 11775 on exposure to different concentrations of hot water extract of and gentamicin.

DISCUSSION

The phytochemical screening of the plant parts extracts revealed the presence of saponin, tannin, flavonoid, anthraquinones, glycosides, triterpenoids, steroids and alkaloids. The presence of plant secondary metabolites in Parkia species, which are known to have broad spectrum of antibacterial activity have been reported\textsuperscript{11, 4, 2}. El-Mahmood and Ameh\textsuperscript{2} reported the presence of phenolics in the root bark of Parkia biglobosa which is synonymous to Parkia clappertoniana, while it is not revealed in this study but this work revealed the presence of triterpenoids and steroids in the root and stem bark extract. This report is also in agreement with that of Millogo-Kone et al.,\textsuperscript{4} who reported the presence of triterpenoids and steroids in the stem bark of P. biglobosa. Parkia biglobosa has been ranked among the...
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The conventional antibiotic gentamicin, consistently showed superior antibacterial activity than the extracts similar to the results presented by other workers. This may be attributed to the fact that herbal medicinal products are prepared from synthetic materials by means of reproducible manufacturing techniques and procedures. In conclusion, Parkia clappertoniana (Keay) contains chemical constituents which possess antibacterial activity against E. coli, the causative agent of diarrhoea, fatal dehydration, urinary tract and bladder infections; Ps. aeruginosa, an opportunistic pathogen in wounds especially burns and other diseases such as HIV/AIDS and Staph. aureus causative agent of skin lesions such as boils, pneumonia and gastroenteritis. The eye infection, diarrhea, healing of wounds and inflammation of testicles.

References


The stem bark extracts showed more antibacterial activity than the other tested parts of the plant as revealed by this study. This result agrees with that of Millogo-Kone et al., who reported that the stem bark of P. biglobosa was very active against all the tested organisms. The antibacterial activity of the hot water extract of the stem was confirmed by the rate of kill experiment. The extract killed E.coli faster followed by Staph. aureus and then Ps. aeruginosa.

Esherichia coli ATCC11775 appeared to be the most susceptible to all the extracts. This result confirms those of Olutimayin et al., Millogo-Kone et al., Udobi and Onaolapo who reported the antibacterial activity of P. biglobosa against E. coli.

The richest plants in tannins have been isolated from various plants by many workers who have also attributed the antibacterial activities of plants to their presence. All the extracts showed antibacterial activity against all the tested bacteria species. This report disagrees with that of Ajaiyeoba, who reported that ethanolic and water extracts of P. biglobosa had no effect against Staph. aureus. Ps. aeruginosa was reported to be insensitive to the water and ethanolic extracts of the plant. The hot water extracts seem to have more antibacterial activity than ethanol and cold water extracts. Hot water has been reported to be effective in extraction of plant constituents. This can also explain the traditional use of hot water to extract plants materials (decoction) by herbal medicine practitioners. The ethanol extracts also were active against the test bacteria species. The results of this work confirm the recent use of the local beer, very rich in ethanol, for the maceration of stem bark and leaf as a prescription by some traditional healers. Patients seem to have more rapid satisfactory effects when used this way.

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The conventional antibiotic gentamicin, consistently showed superior antibacterial activity than the extracts similar to the results presented by other workers. This may be attributed to the fact that herbal medicinal products are prepared from plant and animal origins, most of the time subjected to contamination and deterioration while antibiotics are usually prepared from synthetic materials by means of reproducible manufacturing techniques and procedures.

In conclusion, Parkia clappertoniana (Keay) contains chemical constituents which possess antibacterial activity against E. coli, the causative agent of diarrhoea, fatal dehydration, urinary tract and bladder infections; Ps. aeruginosa, an opportunistic pathogen in wounds especially burns and other diseases such as HIV/AIDS and Staph. aureus causative agent of skin lesions such as boils, pneumonia and gastroenteritis. The eye infection, diarrhea, healing of wounds and inflammation of testicles.
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