The antimicrobial activities of Ethanolic extracts of *Basella alba* on selected microorganisms

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**ABSTRACT**

The antimicrobial effects of ethanolic extract of *Basella alba* against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, and *Candida albican* was determined using the agar cup plate method. The phytochemical components of the ethanolic extracts of the leaf and stem of *B. alba* showed the presence of tannin, terpene, steroid, saponin, anthraquinone, and with carbohydrate only in the stem extracts. The result of this study showed that all the organisms except *Candida albican* were susceptible to 60mg/ml and 100mg/ml of extract. The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) were also determined. The result obtained showed that the MIC and MBC for the ethanolic extract of the leaf and stem of *P. aeruginosa*, *E. coli* was 50mg/ml, while the MIC and MBC of *S. aureus* was 100mg/ml for the ethanolic extract of the leaf and stem of *B. alba*. The result of this study suggests that the ethanolic extracts of *B. alba* could be suitable for the treatment of diseases caused by *S. aureus*, *P. aeruginosa* and *E. coli*.

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1. Introduction

Of the world population, about three quarter relies on plants and their extracts for their healthcare (Jachak, 2007). Plants provide the possibility of an alternative strategy in exploration for new drugs (Abalaka et al., 2011).
Infectious diseases, which account for the significant proportion of the health problems, are most often catered for by this system of medicine (Yasmin et al., 2009). Herbal drug analyses the part or parts of a plant used for the preparation of herbal and traditional medicines (for examples: leaves, flowers, seeds, roots, barks, stems, etc.) (Kayode and Kayode, 2011). The progressing failure of chemotherapeutics and resistance to antibiotics exhibited by pathogenic microbial infectious agents has lead to the screening of several medicinal plants for their potential antimicrobial activity (Elizabeth, 2005; Yogisha and Raveesha, 2009). Plants constitute many biologically active compounds that possess ability and criteria for development as medicinal agents (Abalaka and Oyewole, 2011).

_Basella alba_ is one of such belonging to the family Basellaceae, a fast growing vegetable, native to tropical Asia, probably originating from India or Indonesia and extremely heat tolerant. It is commonly known as Malabar, Ceylon, East-Indian, Surinam and Chinese spinach (Bamidele, et al., 2010; Sushila et al., 2010). The Yoruba natives calls it “amunututu” (Olajire and Azeez, 2011), and the Akwa-Ibom natives of Nigeria calls it “Atameme” (Mepba, et al., 2007). Of more than twenty leafy vegetables consumed in South-Western Nigeria, there are several reports on routine cultivation on only eight. Fewer than six are actually grown for commercial purpose, while some others like _Basella_ grow wild and are under-explored (Odueso, 2011). The paste of root of red _B. alba_ along with rice washed water is taken in the morning in empty stomach for one month to cure irregular periods by the rural people of Orissa, India. Leaves of _B. alba_ is used for the treatment of hypertension by Nigerians in Lagos, and malaria in cameroonian folk medicine (Anandarajagopal, et al., 2011). It is high in vitamin A, vitamin C, vitamin B9 (folic acid), calcium, magnesium and several vital anti-oxidants. It is low in calories by volume and high in protein per calorie. In addition, the cooked roots and leaves have been reported to be used in the treatment of diarrhoea and as laxative, respectively. The flowers are used as an antidote for poisons (Bamidele, et al., 2010). It is administered in gonorrea and balanitis. The mucilaginous liquid obtained from the leaves and tender stalks of this plant is a popular remedy for habitual headaches. The flowers are used as an antidote to poisons and also as diuretic and febrifuge. A paste of the root is applied to swellings and is also used as a rubefaciency, leaf juice is used in Nepal to treat catarrh and is applied externally to treat boils (Sushila et al., 2010). It is also a safe aperient for pregnant women and its decoction has been used to alleviate labour. Moreover, it is locally reported to be used in the treatment of anaemia (Bamidele, et al., 2010). A red dye is obtained from the juice of the fruits. It has been used as rouge and also as a dye for official seals (Rodda, et al., 2012).

The aim of this study is to determine the phytochemical components of ethanolic extracts of _Basella alba_, determine the antimicrobial spectrum of the ethanolic extracts of the leaf and stem of _B. alba_ on _Staphylococcus aureus_, _Pseudomonas aeruginosa_, _Escherichia coli_, and _Candida_ sp., and also to determine the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of the extracts on the test organisms.

2. Materials and methods

2.1. Collection and preparation of samples

Fresh plants of _Basella alba_ were collected from the environs of Ilorin town, Kwara State, Nigeria. The leaves were separated from their stems and were air-dried for five weeks in microbiology laboratory of Federal University of Technology, Minna. The dried materials were blended using sterilized electric blender and well packaged for subsequent analysis.

2.2. Collection of specimen

Pure cultures of _Pseudomonas aeruginosa_, _Staphylococcus aureus_, _Escherichia coli_, and _Candida albicans_ were obtained from microbiology laboratory of Federal University of Technology, Minna. Niger State and were subcultured in agar slants.

2.3. Phytochemical screening of the extracts

Then the screening of the plant extract was carried out according to the method described by Odebiyi and Sofowora (1978) and, Trease and Evans (1989) for the purpose of detecting active components like glycosides, tannin, alkaloid, terpene, steroids, phenolics, saponins, anthraquinone, carbohydrate, and flavonoids.
2.4. Extraction of materials

Ethanol was used as solvents for the extraction of the plant materials using reflux extraction method by suspending 50g of blended sample in 400ml of 75% ethanol for 3 hours. The extracts were filtered and the solvent was evaporated using a steam bath at 60°C.

2.5. Antimicrobial susceptibility test

The Susceptibility test of the test organisms to ethanolic extracts of *Basella alba* at concentrations of 100mg/ml, 60mg/ml, and 40mg/ml was carried out using agar cup plate technique as described by Silver *et al.* (1997). Nutrient agar was prepared according to the standard concentration and autoclave at 121°C for 15 minutes. It was then poured on to plates and allowed to solidify after which wells were made on the agar media using a sterile cup borer. Standardized inoculum of each test organisms was spread on to agar plates so as to achieve a confluent growth. Different concentration of the extract was introduced into the wells equidistant from one another. The plates were then incubated at 37°C for 24 hours.

2.6. Determination of Minimum Inhibitory Concentration (MIC)

The minimum inhibitory concentration (MIC) of the test organisms was determined using the tube dilution technique. 9ml of the nutrient broth was pipetted into various test tubes containing concentrations of 100mg/ml, and 50mg/ml of the extract. The overnight culture of the test organisms diluted at 10^6 cfu/ml was added to the test tubes and then incubated at 37°C for 24 hours. The least concentration of the extract that did not indicate any visible growth of the incubated organisms in broth culture was taken as the minimum inhibitory concentration (MIC) (Hugo and Russel, 1983; Babayi *et al.*, 2004).

3. Results

3.1. Phytochemical screening of the extracts

Table 1 shows the phytochemical screening of the ethanolic extracts of *Basella alba*. The result indicates the presence of tannin, terpene, steroids, saponins, anthraquinone, in the leaf and stem extracts of the plant, while carbohydrate is present only in the stem.

<table>
<thead>
<tr>
<th>Phytochemical component</th>
<th>Leaf extracts</th>
<th>Stem extracts</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glycosides</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Tannin</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Alkaloid</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Terpene</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Steroids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Phenolics</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Saponins</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>anthraquinone</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Carbohydrate</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

+ = present; - = absent

3.2. Antimicrobial activities of the ethanolic extracts of *Basella alba*

Table 2 shows the zones of inhibitions (mm) of ethanolic extracts of *B. alba* on *P. aeruginosa*, *E.coli*, *S. aureus* and *C. albicans* at concentrations 40mg/ml, 60mg/ml and 100mg/ml and control (20mg/ml). The control had a higher antimicrobial effect on the tested organisms than the ethanolic extract of the *B. alba*. 
Table 2
Antimicrobial activity of Ethanol extracts of Basella alba.

<table>
<thead>
<tr>
<th>Test organisms</th>
<th>40mg/ml</th>
<th>60mg/ml</th>
<th>100mg/ml</th>
<th>20mg/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Leaf</td>
<td>Stem</td>
<td>Leaf</td>
<td>Stem</td>
</tr>
<tr>
<td>P. aeruginosa</td>
<td>0</td>
<td>0</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>E. coli</td>
<td>2</td>
<td>0</td>
<td>5</td>
<td>7</td>
</tr>
<tr>
<td>S. aureus</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>C. albicans</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Control; ciprofloxacin (bacteria) and fusin (yeast).

3.3. Minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of the ethanolic extract of B. alba on the test organisms

Table 3 shows the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of the test organisms on the ethanolic extract of Basella alba. The MIC and MBC were 50mg/ml for P. aeruginosa, and E. coli of the leaf and stem of B. alba while S. aureus showed MIC and MBC values of 100mg/ml for the leaf and stem.

Table 3
The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of the ethanolic extracts Basella alba.

<table>
<thead>
<tr>
<th>Test organisms</th>
<th>Leaf MIC (mg/ml)</th>
<th>Leaf MBC (mg/ml)</th>
<th>Stem MIC (mg/ml)</th>
<th>Stem MBC (mg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>P. aeruginosa</td>
<td>50</td>
<td>50</td>
<td>50</td>
<td>50</td>
</tr>
<tr>
<td>E. coli</td>
<td>50</td>
<td>50</td>
<td>50</td>
<td>50</td>
</tr>
<tr>
<td>S. aureus</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
</tbody>
</table>

4. Discussion

The phytochemical component of the crude extracts of Basella alba leaf and stem reveals the presence of tannin, terpene, steroid, saponin, and anthraquinone, but stem extracts only contains carbohydrates. The result of this study supports previous work with an exception of the presence of flavonoids and phenolic compounds (Olajire and Azeez, 2011; Nehar et al., 2011) and the absence of saponin and anthraquinone (Phadungkit, et al., 2012). The antimicrobial activity showed P. aeruginosa E. coli and S. aureus were susceptible to 60mg/ml and 100mg/ml concentration of the extract except C. albicans. The presence of these phytocompounds may be responsible for the antibacterial potency of B. alba extracts (Phadungkit, et al., 2012). The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of P. aeruginosa, and E. coli were 50mg/ml, while for S. aureus is 100mg/ml of the leaf and stem. The result of this study showed that P. aeruginosa E. coli and S. aureus were susceptible to the ethanolic extract of leaf and stem of Basella alba which is in support of the study of Yasmin et al., (2009) and Sushila, et al., (2010).

5. Conclusion

The result of this study suggests that the ethanolic extracts of B. alba could be suitable for the treatment of diseases caused by Staphylococcus aureus, Pseudomonas aeruginosa, and Escherichia coli except Candida albicans.

References


