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Research Article

THE ANTIMICROBIAL ACTIVITIES OF ETHANOLIC EXTRACTS OF *BASELLA ALBA* ON SELECTED MICROORGANISMS

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ABSTRACT

Agar cup plate method was used to determine the antimicrobial effects of *Basella alba* against *Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli* and *Candida albican*. Ethanolic extracts of the leaf and stem of *B. alba* revealed the presence of tannin, terpene, steroid, saponin, anthraquinone, and with carbohydrate present only in the stem extracts. The result of this study showed that *S. aureus, P. aeruginosa* and *E. coli* were susceptible to 60mg/ml and 100mg/ml of the extract while *Candida albican* was resistant. The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) for the ethanolic extract of the leaf and stem were also determined. The MIC and the MBC for the leaf and stem extract of *P. aeruginosa* and *E. coli* was 50mg/ml while the MIC and the MBC for the leaf and stem extract of *S. aureus* was 100mg/ml. The result of this study suggests that the ethanolic extracts of *B. alba* was not suitable for the treatment of disease caused by *Candida albican* but could be suitable for the treatment of diseases caused by *S. aureus, P. aeruginosa* and *E. coli.*

Keywords: Phytochemical, cup plate, ethanolic extract, antimicrobial, MIC, MBC

INTRODUCTION

A medicinal plant is any plant which in one or more of its organ contains substances that can be used for the synthesis of useful drugs[1]. Right from the ancient times to our modern days, plants have formed the basis of traditional medicine[1-5]. Today, millions of people around the world consume plant derived medicines as part of traditional medicine for a range of medical conditions[4]. Medicinal plants have been proven to have several curative abilities[5]. Plants provide the possibility of an alternative strategy in exploration for new drugs[6-10]. Bioactive constituents are produced by plants secondary metabolites. Some of these compounds such as phenolic acids, flavonoids, tannins, terpenes, steroids, alkaloids, saponins and anthraquinone are present in varying proportion in different medicinal plant[2,4]. The leaves, flowers, seeds, roots, barks and stems have been used for the preparation of herbal and traditional medicine due to resistance that have been reported from antibiotics and chemotherapeutics[8]. Infectious diseases, which account for the significant proportion of the health problems, are most often catered for by this system of medicine[9] . With the hope of getting safer, effective and newer drugs, several medicinal plants are being screened[10]. However, the identification of these plants and the investigation of the quality and toxicity as well as method for preserving them are very important[4].

Basella alba is a fast growing vegetable. It belongs to the family *Basellaceae,* 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[11,12]. 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[13]. *Basella alba* is high in calcium, magnesium, anti-oxidants, vitamins A, C and B9 (folic acid). 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, and also as diuretic and febrifuge[11]. It is administered in gonorrhoea and balanitis. The mucilaginous liquid obtained from the leaves and tender stalks of this plant is a popular remedy for habitual headaches. A paste of the root is applied to swellings and is also used as rubefacients, leaf juice is used in Nepal to treat catarrh and is applied externally to treat boils[12]. 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[11].

The aims and objectives of this study were 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 albicans*, and also to determine the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of the extracts on the test organisms.

MATERIALS AND METHODS

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, Niger State, Nigeria. The dried materials were blended using sterilized electric blender and well packaged for subsequent analysis.

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.

Phytochemical screening of the extracts

The phytochemical screening of the plant extract was carried out according to the method described by Odebiyi and Sofowora[14] and Trease and Evans[15] for the purpose of

detecting active components like glycosides, tannin, alkaloid, terpene, steroids, phenolics, saponins, anthraquinone, carbohydrate, and flavonoids.

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 3hours. The extracts were filtered and the solvent was evaporated using a steam bath at 60^{o}C.

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.*[16]. Nutrient agar was prepared according to the standard concentration and autoclave at 121^{o}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^{o}C for 24 hours.

Determination of Minimum Inhibitory Concentration (MIC)

The minimum inhibitory concentration (MIC) of the test organisms was determined using the tube dilution technique. Nine millilitre (9ml) of the nutrient broth was pippeted 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^{o}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)[17].

Determination of Minimum Bactericidal Concentration (MBC)

The plant extract was serially diluted from 10 to 10^{10} dilution. One millilitre (1ml) of each of the dilutions representing a known .concentration of the extract was introduced into 9ml of sterile nutrient broth in test tubes. The mixture was then inoculated with 0.1ml culture of the test organisms previously standardized to 10^{6}cfu/ml. It was then incubated at 37^{o}C for 24hours. The least concentration of plant extract in the test tube with no turbidity was taken as the MIC. Subsequently, tubes that indicated no turbidity was plated out on nutrient agar plates and absence of growth after incubation for 24hours confirms the MBC as described by Hugo and Russell[18].

RESULTS

Phytochemical screening of the extracts

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

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 of 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* in all the concentrations examined.

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 both the leaf and stem while *S. aureus* showed MIC and MBC values of 100mg/ml for the leaf and stem.

Table 1: Phytochemical components of the ethanolic extracts of *Basella alba*

Phytochemical component	Leaf extracts	Stem extracts
Glycosides	-	-
Alkaloid	-	-
Terpene	+	+
Tannin	+	+
Steroids	+	+
Phenolics	-	-
Saponins	+	+
anthraquinone	+	+
Carbohydrate	-	+
Flavonoids	-	-

+ = present
- = absent

Table 2: Antimicrobial activity of Ethanol extracts of *Basella alba*

Test organisms	40mg/ml		60mg/ml		100mg/ml		20mg/ml
	Leaf	Stem	Leaf	Stem	Leaf	stem	Control
P. aeruginosa	0	0	4	5	11	17	19
E. coli	2	0	5	7	14	13	22
S. aureus	0	0	0	0	6	7	24
C. albican	0	0	0	0	0	0	14

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

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

Test organisms	Leaf		Stem	
	MIC (mg/ml)	MBC (mg/ml)	MIC (mg/ml)	MBC (mg/ml)
P. aeruginosa	50	50	50	50
E. coli	50	50	50	50
S. aureus	100	100	100	100

DISCUSSION

The phytochemical component of the crude extracts of *Basella alba* leaf and stem revealed the presence of tannin, terpene, steroid, saponin, and anthraquinone, but carbohydrate was only present in the stem extracts. The result of this study is similar to previous report with an exception of the presence of flavonoids and phenolic compounds[18-19] and the absence of saponin and anthraquinone[20]. 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 activity of *B. alba* extracts[20]. The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) for *P. aeruginosa* and *E. coli* were 50mg/ml, while MIC and MBC for *S. aureus* was 100mg/ml of the leaf and stem extract. The higher MIC and MBC values *S. aureus* may be due to the gram positive clustered nature of *S. aureus*. However, *C. albican* was resistant to the ethanolic extracts of *B. alba* in all the concentrations examined. The result of this study revealed that *P. aeruginosa, E. coli* and *S. aureus* were susceptible to the ethanolic extract of leaf and stem of *Basella alba* and could be suitable for the treatment of diseases caused by these organisms while *Candida albican* was resistant.

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