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Original Research Article

Antibacterial Effect of *Piper guineense* and *Aframomum melegueta* Extracts on *Escherichia coli* and *Staphylococcus aureus* among Patients Attending Medical Care at Imo State Specialist Hospital Umuguma, Owerri, Imo State, Nigeria

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Abstract

This study was conducted using one hundred samples collected from female patients attending medical care at Imo State Specialist Hospital Umuguma Owerri, Imo State between the age group of 18-60 years. They were screened of *Staphylococcus aureus* and *Escherichia coli*, 50 positive subjects each were picked for the study, to know the efficiency of *Piper guineense* and *Aframomum melegueta* on these bacterial infections. Both hospitalized and out patients of the hospital were used for this study. Out of this 35 subjects had significant growth for the both *S. aureus* and *E. coli* infections, 19 subjects had heavy growth *S.aureus* on chocolate agar and 21 subjects had significant growth of *E.coli* on MacConkey agar. Twenty five subjects were negative out of the 50 used for sensitivity test on *S.aurues* and they have the zone of inhibition of 2^{++} at 500mg/10ml concentration of *P. guineense* and its inhibitory zone for 30 subjects of 250mg/10ml *P. guineense*, 125mg *P. guineense* showed resistance to *S.aurues*. *A. melegueta* showed an inhibitory zone of 1^{+} at 500mg/10ml concentration and resistant at 250mg/10ml and 125mg/10ml concentration for *S.aurues*. It is better to use of both *P. guineense* and *A. melegueta* because it effect has been tested on the sensitivity of bacterial infections.

Keywords

Aframomum melegueta
Antibacterial effect
Piper guineense
Plant extracts

Introduction

An antibacterial is a compound or substance that kills or slows down the growth of bacteria the term is often used synonymously with the term *antibiotic(s).* However, with increased knowledge of the causative agents of various infectious diseases, *antibiotic(s)* has come to denote a broader range of antimicrobial compounds including anti-fungal and other compounds. The term *antibiotic* was first used in 1942 by Selman Waksman and his collaborators in journal articles to describe any substance produced by a microorganism that is antagonistic to the growth of other microorganisms in high dilution (Waksman, 1947).This definition excluded substances that kill bacteria, but are not produced by microorganisms (such as gastric juices and hydrogen peroxide). It also excluded synthetic antibacterial compounds such as the sulfonamides. Many antibacterial compounds are relatively small molecules with a molecular weight of less than 2000 atomic mass units. *Staphylococcus aureus* is a bacterial species also known as "golden staph" and Oro staphira, is a facultative anaerobic Gram-positive coccal bacterium.

It is frequently found as part of the normal skin flora on the skin and nasal passages. It is estimated that 20% of the human population are long-term earners of *S. aureus* (Kluytmans et al., 1997). *S, aureus* is the most common species of *Staphylococcus* to cause *Staph* infections. *S. aureus* is a successful pathogen due to a combination of bacterial immuno-evasive strategies. One of These strategies is the production of carotenoid pigment staphyloxanthin, which responsible for the characteristic golden colour of *S. aureus* colonies. This pigment acts as a virulence factor, primarily by being a bacterial antioxidant which helps the microbe evade the reactive oxygen species which the host immune system uses to kill pathogens (Kluytmans et al., 1997; Clauditz et al., 2006).

S. aureus can cause a range of illnesses, from minor skin infections, such as pimples, impetigo, boils (furuncles), cellulitis folliculitis, carbuncles, scalded skin syndrome, and abscesses, to life-threatening diseases such as pneumonia, meningitis, osteomyelitis, endocarditis, toxic shock syndrome (TSS) bacteremia, and sepsis. Its incidence ranges from skin, soft tissue, respiratory,one, joint, endovascular to wound infections. It is still one of the five most I common causes of nosocomial infections and is often the cause of postsurgical wound infections. Each year, some 500,000 patients in American hospitals contract a staphylococcal infection (Liu et al., 2005).

Escherichia coli is one of the well known and significant species of bacteria living as gut fauna in the lower intestines of mammals. The number of individual *E. coli* bacteria in the faeces that a human excretes in one day averages between 100 billion and 10 trillion. For this reason, *E. coli* has been used in water analysis as the indicator of faecal contamination. However, the bacteria are not confined to this environment, and specimens have also been located, for example, on the edge of hot springs.

Objectives

1. To evaluate the antibacterial effect of *Piper guineense* on *E. coli* and *S. aureus.*
2. To evaluate the effect of *Aframomum melegueta* on *E. coli* and *S.aureus.*
3. To determine synergistic effect of *P. guineense* and *A. melegueta* on *E.coli* and *S.aureus*.

Materials and methods

Study area

This study was conducted in Imo State Specialist Hospital, Umuguma Owerri, Imo State.

Research design

This work was carried out on patients assessing medical care in Imo State Specialist Hospital, Umuguma.

Study population

One hundred (100) samples were randomly collected from female patients between ages IS and above who access medical care at Imo State Specialist Hospital, Umuguma, Owerri, Imo State. Fifty samples that yielded growth of *S. aureus* and *E.coli* were used for the sensitivity of this study**.**

Selection criteria inclusive

Patients whose consent was obtained were selected for the study.

Exclusive

1. Those who refused to consent.

2. Those that are not on any antibiotic therapy.

Specimen collection

Urine/HVS samples were collected from the test subjects using dry sterile universal transparent containers and swab sticks. The subjects were properly educated on how to aseptically collect the urine sample into the sterile container to prevent contamination of the sample while the HVS was collected by the nurses using sterile speculum. The samples were immediately inoculated using Blood agar, Chocolate agar and MacConkey agar. Nutrient agar was used for sensitivity.

Laboratory procedure

All media were commercially purchased and the manufacturers' preparations and standard operational procedures (SOPs) were strictly observed. *P. guineense* and *A. melegueta* were used to make sensitivity disc. The seeds of *P. guineense* and *A. melegueta* were crushed into powdered form and serial dilution with distilled water was made in ratio 1:10 w/v that is 1g of the powder to 10ml of distilled water (500mg). Then 1ml of 500mg concentration was added to 10ml of distilled water to make 250mg v/v; 125mg was also prepared by taking 1ml of 250mg to 10ml of distilled water v/v; Filter paper was used to make sensitivity disc by placing a drop of each preparation on the disc, this was dried on a hot air oven.

Culture procedure

A loopful of urine sample was inoculated on the Blood agar, CLED agar using a wire loop of 3mm diameter, HVS sample was inoculated on Chocolate agar and MacConkey agar, Blood agar and Deoxycholate agar. The inoculums were then aseptically streaked over the culture media to produce discrete colonies. The Chocolate agar was placed in an anaerobic jar. Others were incubated at 37°C in an incubator for 24 h and discrete colonies of *S. aureus* and *E. coli* were isolated. The pathogens were identified using both appearance and biochemical tests.

Sensitivity procedure

A sterile wire-loop was used to pick a loopful of the colony which was isolated from the culture media, and was inoculated into a Nutrient agar. The inoculums were then aseptically streaked over the culture media and the sensitivity disc of *P. guineense* and *A. melegueta* were placed on the inoculum. This was incubated at 37°C in an incubator for 24h and the degree of sensitivity was ascertained.

Results and discussion

One hundred (100) subjects from Imo State Specialist Hospital, Umuguma were screened of *S.aureus* and *E.coli*, 50 positive subjects each were picked. These subjects were both out patients and hospitalized patients and they met the research criteria. Out of these 35 subjects had significant growth for both *S. aureus* and *E.coli* infections, 19 subjects had heavy growth of *S.aureus* on chocolate agar and 21 subjects had significant growth of *E.coli* on MacConkey agar;25 subjects were negative for *S.aureus* and *E.coli* (Table 1).

Out of the 50 subjects for the *S.aureus*, 41 of them had an inhibitory zone of 2++ at 500mg/10ml concentration of *P. guineense* and 1+ inhibitory zone for 30 subjects of 250mg/10ml *P. guineense*, 125mg *P. guineense* showed resistance to *S.aureus*. *A. melegueta* showed an inhibitory zone of 1+ at 500mg/10ml concentration and resistant atr 250mg/10ml and 125mg/10ml concentrations for *S.aureus*. Thirty two (32) subjects had 2++ at 500mg/10ml concentration of *Piper guineense* on *E.coli*, 6 0 subjects showed 1+ at 250mg/10ml concentration and 12 subjects were resistant at 125mg/10ml concentration for *E.coli*. *Afromomium melegueta* showed an inhibitory zone of + at 500mg/10ml concentration on 15 subjects and resistant at 250mg/10ml and 125mg/10ml concentrations for *E.coli* for all subjects. The studied population was made up of subjects between ages 18-60 years and of 18 45 years old (Tables 2 and 3).

Table 1. Distribution of the study population.

Categories	%
No. of subjects screened	100
Positive subjects for both *S.aurues* and *E.coli*	35
No. of positive *S.aureus* subjects used	19
No. of positive *E.coli* subjects used	21
No. of negative subjects	25

Table 2. Inhibition to *Piper guineense* on *S.aureus* and *E. coli*.

No. of subjects	500mg/10ml	250mg/10ml	125mg/10ml
S. aureus			
41	++	nil	nil
30	nil	+	nil
E. coli			
32	++	nil	nil
6	nil	+	nil
12	nil	nil	nil

Table 3. Inhibition to *Afromonium melengueta* on *S. aureus* and *E. coli*.

No. of subjects	500mg/10ml	250mg/10ml	125mg/10ml
S. aureus			
23	+	nil	nil
27	nil	nil	nil
E. coli			
15	+	nil	nil
35	nil	nil	nil

The antibacterial treatment may select for bacterial strains with physiologically or genetically enhanced capacity to survive high doses of antibacterials. Under certain conditions, it may result in preferential growth of resistant bacteria, while growth of susceptible bacteria is inhibited by the drug (Liu et al., 2005). This may be the case in this study which showed that *P. guineense* may have a higher susceptibility than *A. melengueta* since 41 subjects showed inhibitory zone of 2+ and 32 subjects out of 50 showed also inhibitory zone of 2+ at 500mg/10 concentration on *S.aureus* and *E.coli* respectively. This was not very evident in *A. melengueta* where 23 subjects out of 50 showed an inhibitory zone of just 1+ on *S.aureus* and *E.coli* at 500mg 10ml concentration.

There was a high resistance of *A. melengueta* on both *S.aureus* and *E.coli*. This may mean that this plant does not have bacterial effect or may show some sensitivity if a study is carried out on other microorganisms. *P. guineense* can be used as antibacterial sensitivity since there is a significant susceptibility to *S.aureus* and *E.coli.*

The study showed that *P. guineense* has antibacterial effects on *S.aureus* and *E.coli* because of the high sensitivity shown when it was used as sensitivity drug. The zone of inhibition was marked (2++). Based on the above reason, *P. guineense* may be developed pharmacologically to be use as an antibiotic drug. There is little or no sensitivity of *A. melengueta* hence this may not be useful as antibiotic drug or further work may be carried out on it for other bacteria to ascertain its effect.

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