Antibacterial effects of fresh onion (Allium cepa L.) bulbs against clinical bacteria

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Abstract
The purpose of study was to determine the effects of fresh onion bulbs extracts on pathogenic bacteria in an attempt to seek active components present in the onion bulbs. Standard procedures were used for plant extraction and antibacterial susceptibility tests. The extracts were active against Escherichia coli, Staphylococcus aureus, Streptococcus Spp and Enterococcus Spp. while Pseudomonas aeruginosa and Bacillus subtilis were resistant to plant extracts and standard antibiotics. Minimum inhibitory concentration were 25mg/ml and 50mg/ml respectively for ethanol and methanol extracts for E. coli and Streptococcus Spp. The phytochemical results indicated the presence of saponins phenols, tannins, steroid, alkaloids, cardiac glycosides and flavonoid. Anthraquinone and terpenoid were absent. Conclusively, water extracts of fresh onion bulbs was not potent against clinical isolates however ethanol and methanol extracts were active. Fresh onions bulb extracts contained low concentrations of phytochemical compounds.

Keywords: Fresh onion (Allium cepa L.) bulbs, antibacterial effect, phenols.

1. Introduction
Antibiotic resistance is currently a global concern and significant research is taking place to tackle this issue (WHO, 2014; Choski et al., 2019). Acquired resistance is the major contributor to baseline resistance and is a major threat to the continued success of antibiotics (Lior and Bjerrum, 2014). Due to the alarming rate of increased acquired antibiotic resistance in pathogens it is imperative that researchers explore other safe and sustainable alternatives to antibiotics. (Antunes et al., 2016; Umarau et al., 2017; Davis et al., 2018). Numerous medicinal plants have been utilized as traditional medicines globally for human therapeutic use to treat diseases of pathogenic origin (Bensky et al., 2014). Plant extracts consist of numerous bioactive compounds including polyphenols, terpenes, and phytosterols (Lilehoj et al., 2018) and exhibit multiple modes of action to inhibit or kill bacteria (Chitemerere and Mukanganyama, 2014). An increasing number of research has demonstrated that many plants used in traditional medicine have antimicrobial activity. It is necessary to establish scientific evidence for the bactericidal activity of plant extracts as they may provide a source for the development of novel antibacterial agents. Attempt have been made previously to seek antibacterial agents to solve many problems associated with microbial illnesses. Antibacterial activities of some plants and their parts have been carried out previously (Adomi, 2006, Adomi, 2008; Adomi and Umukoro, 2010, Adomi and Oseh-Jovy, 2020a; Adomi, 2020b; Adomi, 2021a, Adomi, 2021b) in an attempt to seek active component of plants and also to verify scientifically the basis of use in traditional medicine. Efforts are also being made by scientists and most especially clinical microbiologists for more medicine to curb ever expanding ailments plaquing mankind.

Onion belongs to the family Amaryllidaceae; the plant is either biannual or perennial (depending on the cultivar), and
smells when crushed (WHO, 1999). Onion (Allium cepa L.) has been valued as a food and a medicinal plant since ancient times. It is cultivated widely as a vegetable bulb plant known and consumed worldwide (FAO, 2012). It is a short duration horticulture crop which grows at low latitudes. It is known as the “Queen of the kitchen,” due to its aroma, flavour, taste, and the medicinal features of its compounds (Griffiths et al., 2002). Onions have been used by man since his existence, and they are still being used till date globally. Over this long period, some people use onions in large quantities while others rejected and hardly use them (Koch and Lawson, 1996). Onions have been planted around the world, in at least 175 countries, for about 5000 years. In Egypt, the spherical bulb is regarded as a symbol of the universe. The word onions are probably taken from the latin unus meaning “one,” the Romans introduced the plant to Britain, and Britain, to the Americans (Burnie et al., 1999). In distant past, onions was utilized by Egyptians and worshipped believing the concentric rings represented eternal life and usually buried their dead with it. The ancient Greeks consumed lots of it, thinking it had the ability to improve athlete prowess and cleanse blood. The ancient American used onion to treat cold, coughs, breathing problems and asthma. Hippocrates suggested the use of onion as a laxative, diuretic and emmenagogue also, for the management of pneumonia, and healing putrid injuries (Koch and Lawson, 1996). It is used for the treatment of gastrointestinal disorders, asthma, and bronchitis. The recommended mean oral dose of fresh onion is 50g or 20g of dried onion daily. Fresh onion bulb or extracts are also used topically for treatment of insect stings and warts.

2. Materials and Method

2.1 Plant Collection and Extraction

Red onions were bought on 20th of July 2022 from Ekpan market, Uvwie local Government area of Delta State. Part of the onions were taken to the herbarium section of Botany Department, Delta State University, Abraka for identification and voucher specimen with number DelsuH133 was assigned and kept in the laboratory. The other part was taken for extraction. The onions were washed under tap water and cut into bits after the outer dry cover was peeled. The raw onions (100g) were soaked in the 300ml of methanol, ethanol and water seperately for 48hrs, filtered and evaporated to dryness (Oyebode and Fajilade, 2021; Ola-Madather and Madiagwu (2014). The various extracts were stored under 4°C before use.

Bacterial cultures used in this experiment were clinical isolates and included Escherichia coli, Staphylococcus aureus and Streptococcus sp Pseudomonas aeruginosa, Acinetobacter baumanii. The cultures were standardized according to Clinical Laboratory Standard Institute (CLSI,2019). Broth culture grown overnight was diluted with sterile normal saline and compared with Mcfarland standard (0.5 x10^6). The susceptivity test was investigated by streaking the standardized culture in solidified. Mueller hinton agar. Holes were bored in strategic points on the agar and 200µl of each extract was introduced into hole and then incubated for 24-48hours after diffusion for two hours (Eltaweel, 2013). Normal saline was used as the negative control, and antibiotics as positive control. The diameters of the zones of inhibition were measured with a ruler. The different extracts which were active on bacteria was investigated for the minimum inhibition concentration by broth dilution method (Omotola et al., 2018). The extracts were diluted from 50% to 6.25% in sterile Mueller hinton broth, 2ml of each extract dilution was added 2mls of broth. The
inoculum (0.1ml) was then introduced and incubated at 37°C for 24hrs. Broth without inoculum, diluted extract without inoculum, inoculated broth without extract were the controls. The lowest concentration of extract with no visible growth at the end of incubation was taken as the minimum inhibitory concentration (Tagoe and Gbadago, 2010).

3. Results and discussion

3.1 Results

The results of the investigation of the antibacterial effects of fresh onion bulbs against some pathogenic bacteria including *Staphylococcus aureus*, *Escherichia coli*, *Streptococcus Spp.* *Pseudomonas aeruginosa*, *Enterococcus Spp.* and *Acinetobacter baumanii* are presented in Table 1.

Table 1: Antibacterial effects of fresh onion (*Allium cepa* L.) bulbs against clinical bacteria at 50mg/ml

<table>
<thead>
<tr>
<th>Allium cepa L.</th>
<th>Diameter of Inhibition (mm)</th>
<th><em>S. aureus</em></th>
<th><em>E. coli</em></th>
<th><em>P. aeruginosa</em></th>
<th><em>B. subtilis</em></th>
<th><em>A. baumanii</em></th>
<th>Enterococcus Spp.</th>
<th>Streptococcus Spp.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Ethanol</td>
<td>0</td>
<td>20</td>
<td>0</td>
<td>18</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Methanol</td>
<td>19</td>
<td>15</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>14</td>
<td>15</td>
<td>0</td>
</tr>
<tr>
<td>Rifampin (30mcg)</td>
<td>15</td>
<td>15</td>
<td>0</td>
<td>0</td>
<td>14</td>
<td>15</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Table 2: MIC of Fresh onions (*Allium cepa* L.) bulbs Extract.

<table>
<thead>
<tr>
<th>Allium cepa L.</th>
<th>Diameter of Inhibition (mm)</th>
<th><em>S. aureus</em></th>
<th><em>E. coli</em></th>
<th><em>Streptococcus Spp.</em></th>
<th><em>A. Baumanii</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Ethanol</td>
<td>25mg/ml</td>
<td>-</td>
<td>25mg/ml</td>
<td>-</td>
<td>50mg/ml</td>
</tr>
<tr>
<td>Methanol</td>
<td>25mg/ml</td>
<td>25mg/ml</td>
<td>50mg/ml</td>
<td>50mg/ml</td>
<td></td>
</tr>
</tbody>
</table>

Table 3: Phytochemical test of *Allium cepa* L

<table>
<thead>
<tr>
<th>Constituents</th>
<th>Saponins</th>
<th>Phenols</th>
<th>Tannins</th>
<th>Steroids</th>
<th>Alkaloids</th>
<th>Cardiac glycosides</th>
<th>Flavonoids</th>
<th>Terpenoids</th>
<th>Anthraquins</th>
</tr>
</thead>
<tbody>
<tr>
<td>Qualitative</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Quantitative (mg/100g)</td>
<td>4.43</td>
<td>16.00</td>
<td>5.27</td>
<td>3.56</td>
<td>0.07</td>
<td>0.84</td>
<td>2.51</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

Key: + low concentration, ++ moderate concentration, +++ high concentration, - negative
The water extract was not potent against any bacteria in this study. Ethanol extract was active against *Escherichia coli* and *Acinetobacter baumannii*. Methanol extract was the most active in this study being potent against *E. coli*, *S. aureus* and *Streptococcus* Spp. The standard antibiotic was active against *E. coli*, *S. aureus*, *Streptococcus* Spp and *Enterococcus* Spp. However, *Pseudomonas aeruginosa* and *Bacillus subtilis* were resistant to both extract and standard antibiotics. *Streptococcus* Spp. was equally resistant to standard antibiotics, water and ethanol extracts of *Allium cepa* Linn. Table 2 shows MIC of *Allium cepa* extracts was 25mg/ml for ethanol and methanol extracts, while 50mg/ml for *E. coli* and *Streptococcus* Spp. The phytochemical results indicated the presence of saponins (4.43mg/100g), phenol (1600 mg/100g), tannins (5.27 mg/100g steroid (3.56 mg/100g), alkaloids (0.07 mg/100g) Cardiac glycosides (084 mg/100g), flavonoid (2.51 mg/100g however, anthraquinones and terpenoid were absent.

### 3.2 Discussion

Three crude extracts were obtained from onion (*Allium cepa* L.) bulbs. Water extract was not active against any bacteria. The reason could be that the active component present was very minimal in the fresh onion bulbs used. The phytochemical result buttresses this fact where the phytochemical compounds concentration present in onions bulb was low (table 3). The method of plant extraction affects the activity of plant extractives. Cold maceration method was used, ruling out the possibility of heat being a factor in affecting the activity of the extract. However, the result was consistent with the findings of Omotala et al. (2018) in that aqueous extract was not potent against *E. coli*. Methanol and ethanol extracts were active against *E. coli*, and *Streptococcus* Spp. and *Staphylococcus aureus* (15mm, 19mm, 19mm), and ethanol was active against *E. coli* and *A. baumannii*. The activity of these extracts against the pathogens showed that these extracts (ethanol and methanol) contained active compounds; also solvent extracted the active components which could be a valuable feature for medicine and could be used to treat sickness caused by the pathogens. Jesmirhene, et al. (2021) found ethanol extracts more active than water extracts against pathogens studied, which aligned with this research. Method of obtaining plant extracts could play a role in susceptibility of microorganisms to plant extracts. Kobrah et al. (2016) obtained their extracts by blending fresh onions bulb and the filtrate was used for their experiment. Their results showed activity against the bacteria studied. Aqueous extracts of onions in another study was active against the organisms. The onion bulbs were air dried at room temperature and milled. The powdered sample was soaked in water, filtered, and evaporated to dryness at 20°C. In this study however, fresh onions bulbs were cut into bits and extracted through cold maceration using water, ethanol, and methanol as solvents. Dry onions bulbs could have higher antibacterial potential. It is recommended that extracts of dry onions bulb should be investigated to determine if more phytochemical compounds correlate with higher activity.
Conclusion
Fresh water extract of onion bulb was not active against clinical bacteria. However, ethanol and methanol extract showed activity. Qualitative and quantitative Phytochemical tests of fresh onion bulb showed low content. Extracts from dry onion bulb could have more activity.

References


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