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Potentials of Using Allium Cepa (Onion) Leaves Extracts as Antibacterial Agent: A Strategy of Converting Agricultural Waste to Wealth

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Abstract- This research investigated the phytochemical constituents and antibacterial activity of Allium cepa (onion) leaves extracts as a potential source of natural antimicrobial agents. Several analyses were conducted, beginning with qualitative phytochemical screening, followed by antibacterial evaluation using some selected bacterial strains. Standard antibiotic (ceftriaxone) served as a control. The phytochemical analysis revealed the presence of alkaloids, flavonoids, tannins, saponins, steroids, and cardiac glycosides with some amount of carbohydrates and proteins, while anthraquinones were absent. Antibacterial assays showed concentration-dependent inhibition zones, with maximum activity at 100 mg/mL: Staphylococcus aureus (3.04 mm), Escherichia coli (2.68 mm), and Bacillus subtilis (2.35 mm), while Shigella showed no inhibition. By contrast, ceftriaxone (50 mg/mL) produced higher zones ranging from 3.28 – 4.85 mm across all organisms. The MIC and MBC values confirmed inhibitory and bactericidal effects at 75 mg/mL for Staphylococcus aureus and Escherichia coli, 50 mg/mL for Bacillus subtilis, and 25 mg/mL for Shigella.

Index Terms- Phytochemicals, Antibacteria, Onion, Allium cepa, Shigella, Bacteria

I. INTRODUCTION

Allium cepa Leaves (Onion) is one of the most widely consumed vegetables worldwide, valued not only as a dietary staple but also as a medicinal plant. It contains a broad spectrum of phytochemicals such as flavonoids, phenolic acids, organosulfur compounds, and saponins, which contribute to its antioxidant, anti-inflammatory, and antimicrobial properties [1-6]. These bioactive compounds have positioned onion as an important candidate for studies focused on natural alternatives to synthetic drugs. Phytochemical investigations of onion leaves specifically have revealed alkaloids, tannins, flavonoids, saponins, terpenoids, and cardiac glycosides. These compounds are responsible for various biological activities, including antibacterial and antifungal effects [7 - 9]. The leaves are often underutilized compared to the bulbs, yet they hold significant potential as sources of therapeutic compounds. Antibacterial screening of onion leaf extracts has demonstrated significant inhibition against pathogenic microorganisms such as Staphylococcus aureus and Escherichia coli. These activities are strongly associated with the presence of thiosulfinates, flavonoids, and phenolic constituents [10-12]. Such findings suggest that onion leaves could be exploited as inexpensive and accessible antimicrobial agents. Comparative studies among different Allium species highlight that while garlic is often reported to have stronger antimicrobial activity, onion leaves also possess meaningful inhibitory effects. The synergy between organosulfur compounds and polyphenols enhances their antibacterial properties, particularly in hydroalcoholic extracts [13-15]. This makes onion a valuable plant for further comparative phytochemical and biological studies.

Advanced analytical techniques such as HPLC and GC–MS have facilitated the identification of major compounds in onion leaf extracts, including quercetin, rutin, and kaempferol. These flavonoids, along with sulfur derivatives, have been shown to disrupt bacterial cell walls and inhibit enzymatic processes essential for microbial survival [16-17]. Their abundance in onion leaves strengthens the case for their antibacterial application. With the increasing global challenge of antibiotic resistance, the search for new antibacterial agents from plant sources is critical. A comprehensive study of the phytochemical constituents and antibacterial activity of onion leaves extracts will offer insight into their potential as natural alternatives to conventional antibiotics. This research seeks to contribute to the growing body of evidence supporting onion as a promising medicinal plant [18].

Plate 1: Allium cepa Leaves

II. MATERIALS AND METHODS

All chemicals and solvents were obtained from LobaChemie and used as received. Mular Hilton nutrient agar and broth were used for the antibacterial analysis.The test organisms were obtained from the Department of Biological Science, Sokoto State University. The microorganisms are standard laboratory strains of *Staphylococcus aureus* (gram +ve), *Escherichia coli* (gram –ve), *Bacillus subtilis* (gram +ve), and *Shigella* (gram –ve).

Sample Collection and Treatment

The sample *Allium cepa* (Onion) leaves were collected in July, 2025 from Vegetable section, Sokoto Central Market, Sokoto State, Nigeria. The leaves were washed with clean water to remove the dirt and dried under shade for three days. The dried leaves of *Allium cepa* were ground using mortar and pestle then sieved to obtain fine powder. The powdered samples were stored in an air tight water free polyethene bag and used for the analysis.

Determination of Moisture Content

The fruit were taken in a pre-weighed empty dish (W0) and their resulting weight was denoted by (W1). The dish containing the fruit sample was transferred into an oven at 100 0C for 24 hours then removed, and the weight of the dish containing the dried sample was determined and denoted by (W2). The percentage moisture content of the fruit was calculated using equation below.

$$\%\ \text{moisture} = \frac{W_1 - W_2}{W_1 - W_0} \times 100 \qquad (1)$$

Where W_0 = weight of the empty dish
W_1 = weight of empty dish + sample
W_2 = weight of to the empty dish + dry sample.

Extraction of Allium cepa Leaves

The leaves powder (98 g) was extracted by maceration in a 1000 cm^3 glass stoppered conical flask. Ethanol (200 cm^3) was used as solvent. The leaves powder was left in contact with the solvent for 24 hours with intermittent stirring to ensure maximum extraction. After 24 hours, the extracts were filtered using Whatman filter No.1 and the filtrates were transferred to evaporation dishes and concentrated using air circulated oven at temperature 80 ^{0}C. The percentage yield of the extracts was calculated as follows:

$$\%\ \text{extract yield} = \frac{\text{Weight of extract}}{\text{Initial weight of sample}}\ x\ 100\text{----}(2)$$

Qualitative Phytochemical Screening of Extract.

The tests for flavonoids, tannins, saponnins, steroids/triterpenoids, alkaloids, cardiac glycosides and anthraquinones were carried according to the methods described by [10-12].

Antibacterial Tests

The antibacterial test was conducted using the petri dish plate method described by [19]. The Nutrient Agar plates prepared according to manufacturer's instruction were allowed to solidify for 15 minutes at room temperature and incubated without inoculum for 24 hours at 37 ^{0}C to ensure the sterility of the medium. The Nutrient Agar plates were flooded with 1 ml of the inoculum and the excess was removed using Pasteur pipette. 4-6 wells (cups) of about 6 mm in diameter were cut on each Nutrient Agar plate using a sterile cork borer and the agar plugs were removed using a sterile ampoule file. The extract solution (0.1 mL) was placed in each of the wells and were allowed to settle for two hours at room temperature and then incubated for 24 hours at 37 ^{0}C.

The inhibition zone was observed and then recorded in millimeters using a transparent ruler. Standard antibiotics were used.

Minimum Inhibitory Concentration (MIC)

This was carried out as described by [20]. Minimum Inhibitory Concentration (MIC) was defined as the lowest concentration where no visible turbidity would be observed in the test tubes. The MIC was determined for the micro-organisms were prepared using the broth dilution technique the stock solution (20 mg1ml) of the fruit extract was prepared by dissolving 0.2g of the fruit extract in 10 ml DMSO three concentration were prepared (0.25mlmL, 0.50mglmL, 0.75mglmL) from the stock solution using serial dilution technique and later inoculated with 0.2ml, suspension of the test organism, After 24 hours incubation at 37 °C, the tubes were observed for turbidity. The lowest concentration where no turbidity was observed was determine and noted as the Minimum Inhibitory Concentration (MIC).

Minimum Bactericidal Concentration (MBC)

The minimal bactericidal concentration was determined from broth dilution test resulting from the MIC tubes as described previously (2.4.1) by inoculating the content of each test tube on a nutrient agar plate. The plates were then incubated at 37 °C for 24 hours. The lowest concentration of the extract that showed no growth was noted and recorded as the minimum bactericidal concentration [20].

III. RESULTS AND DISCUSSION

Percentage Moisture Content of Allium cepa Leaves

The details of the percentage moisture content obtained from *Allium cepa* leaves were given in Table 1. The leaves had a percentage moisture content of 75.0 %.

Table 1: Percentage moisture content of Allium cepa leaves (%)

Sample	W_0	W_1	W_2	Moisture content (%)
Allium cepa leaves	5.0	103	29.5	75.0 %

Key: - W_0 = Weight of empty dish, W_1 = Weight of empty dish + sample, W_2= Weight of empty dish + dry sample

Extraction of Allium cepa Leaves

The mass of the extract and percentage yield obtained from Allium cepa leaves were given in Table 2. The extract has a mass of 24.5 g with percentage yield of 25.0 % as shown in the table below.

Table 2: Mass of Extract (g) and percentage yield of Allium cepa leaves (%)

Extract	Original mass (g)	Mass (g) of extract	Percentage yield (%)
Ethanol	98.0	24.5	25.0

Qualitative Phytochemical Screening of Allium cepaLeaves Extract

The results of the qualitative phytochemical screening of the extract obtained were shown in Table 3. Saponins, flavonoid, tannins, alkaloid, steroids, carbohydrates and protein, were detected, while anthraquinones were not detected.

Table 3: Qualitative Phytochemical Screening of Allium cepa leaves extracts

Phytochemicals	Ethanol
Carbohydrates test	
(a) Molich's test	+
(b) Fehling test	+
Protein test	
(a) Xanthoprotic test	+
Saponins	
(a) Froth's test	+
Flavonoids	
(a) Alkaline test	+
(b) Ferric chloride test	+
(c) Shinoda's Test	+
Tanins	
(a) Ferric chloride test	+
(b) Lead acetate test	+
Alkaloid	
(a) Mayer's test	+
(b)Wagner's reagent	+
(c) Hager's test	+

Steroids and Inter-terpenoids test	
(a) Salkowski's test	+
(b) Libermann-Richard's test	+
Anthraquinone	
(a) Borntrager's test	-
Cardis glycoside's test	+

Keys: - (+) = detected (-) = not detected

Antibacterial Test

The results obtained from the antibacterial test were shown in table 4a – c.

Table 4a: Antibacterial activity of *Allium cepa* (Onion) leaves extracts

Concentration (mg/mL)	Zone of Inhibition (mm)			
	S. aureus	*E. coli*	*B. subtilis*	*Shigella*
25	0.00	0.00	0.75	0.00
50	0.00	1.25	1.38	0.00
75	2.70	2.30	1.93	0.00
100	3.04	2.68	2.35	0.00

Table 4b: Antibacterial activity of standard antibiotic (Ceftriaxone)

Antibiotic Conc. (mg/mL)	Zone of Inhibition (mm)			
	S. aureus	*E. coli*	*B. subtilis*	*Shigella*
Ceftriaxone (50 mg/mL)	4.05	4.85	4.71	3.28

Table 4c: MIC and MBC of *Allium cepa*leaves extract

Test Organisms	MIC (Mg/mL)	MBC (Mg/mL)
S. aureus	75.00	75.00
E. coli	75.00	75.00
B. subtilis	50.00	50.00
Shigella	25.00	25.00

Key: - MIC = Minimum Inhibitory Concentration, MBC = Minimum Bactericidal Concentration

IV. DISCUSSION

The results obtained from the moisture content and solvent extraction of *Allium cepa* leaves revealed percentage moisture content of 75.0 %, indicating that the leaves have high water content (Table 1). The mass of the extract obtained was 24.5 g, with a percentage yield of 25.0 % (Table 2). This indicates that ethanol as a solvent is effective for extracting the leaves. It also shows that the leaves contain appreciable amount of polar compounds as ethanol is a polar solvent. The phytochemical screening of the extract (Table 3) revealed the presence of saponins, flavonoids, tannins, alkaloids, steroids, and cardiac glycosides with some amount of carbohydrates and proteins, whereas anthraquinones were absent. The presence of flavonoids, tannins, and alkaloids is particularly significant since these classes of compounds are well documented for their antimicrobial, antioxidant, and anti-inflammatory properties [21]. Flavonoids such as quercetin and rutin, which are abundant in onion leaves, are known to disrupt microbial membranes and inhibit nucleic acid synthesis [22]. The detection of saponins and steroids further supports the potential pharmacological value of onion leaves. Saponins possess surface-active properties that can cause leakage of cellular contents in bacteria, while steroids and terpenoids are associated with membrane stabilization and immunomodulation [23]. The absence of anthraquinones suggests that onion leaves may not rely on this class of compounds for antimicrobial activity, which is consistent with prior studies where anthraquinones were more common in root-based extracts of other medicinal plants than in leafy tissues [24].

The ethanol extract of *Allium cepa* leaves displayed a concentration-dependent antibacterial effect (Table 4a) with zone of inhibition ranging between 0.75 mm to 3.04 mm. At 25 mg/mL, there was no observable inhibition against *Staphylococcus aureus*, *Escherichia coli*, or *Shigella*, though a mild inhibition of 0.75 mm was recorded against *Bacillus subtilis*. As the concentration increased to 50 mg/mL, activity against *Escherichia coli* (1.25 mm) and *Bacillus subtilis* (1.38 mm) became evident, indicating that higher doses enhance the extract's efficacy. The maximum inhibition was observed at 100 mg/mL, producing zones of 3.04 mm (*Staphylococcus aureus*), 2.68 mm (*Escherichia coli*), and 2.35 mm (*Bacillus subtilis*), while *Shigella* remained resistant across all concentrations. This trend supports the notion that the antibacterial potency of onion leaves is dose-dependent, with more pronounced inhibition at higher concentrations [25]. The strong activity against *Staphylococcus aureus* and *Escherichia coli* highlights the sensitivity of both Gram-positive and Gram-negative organisms to onion phytochemicals such as quercetin, rutin, and thiosulfinates [26]. The resistance of *Shigella* is likely due to its robust outer membrane, which restricts the penetration of phytochemicals, a pattern previously reported for Gram-negative enteric pathogens [27].

The standard antibiotic ceftriaxone exhibited significantly higher antibacterial activity compared to the onion leaves extract (Table 4c) with zone of inhibition ranging between 3.28 mm to 4.85 mm. At 50 mg/mL, inhibition zones were 4.05 mm (*Staphylococcus aureus*), 4.85 mm (*Escherichia coli*), 4.71 mm (*Bacillus subtilis*), and 3.28 mm (*Shigella*). These values were appreciable when compared with the values obtained for the extract in controlling bacterial growth. It is important to note that while ceftriaxone is highly effective, the global rise of antibiotic resistance is reducing its long-term reliability. Plant extracts such as onion leaves, may provide alternative or complementary strategies. Their complex phytochemical mixtures can act on multiple microbial targets simultaneously, potentially reducing the likelihood of resistance development [28]. Thus, onion extracts could serve as supportive agents, especially in settings where access to standard antibiotics is limited or resistance is widespread.

The MIC and MBC results (Table 4c) revealed further insights into the antibacterial potential of the onion leaves extracts. For *Staphylococcus aureus* and *Escherichia coli*, both MIC and MBC were 75 mg/mL, indicating that growth inhibition and bacterial killing occurred at the same concentration. While for *Bacillus subtilis*, the MIC and MBC were lower (50 mg/mL), suggesting a stronger bactericidal effect of the extract on this organism. Interestingly, *Shigella* displayed the lowest MIC and MBC values (25 mg/mL), despite showing no visible inhibition zones in Table 4a. These findings highlight the complexity of antibacterial assessments, as disc diffusion methods (zone of inhibition) may not always align perfectly with broth dilution techniques (MIC/MBC). The apparent discrepancy in *Shigella*'s response could be due to differences in extract diffusion rates in agar compared to liquid media [29]. Nonetheless, the relatively high MIC and MBC values across organisms suggest that onion leaf extracts, while active, require substantial concentrations to achieve bactericidal effects. The comparable MIC and MBC values observed for most organisms suggest that the extract exerts both inhibitory and lethal effects at the same concentration. This dual action is advantageous, as it implies that once inhibitory levels are reached, bacterial eradication is also possible [30].

V. CONCLUSION

This study focused on investigation of the phytochemical constituents and antibacterial activity of *Allium cepa* (onion) leaves extracts. From the background of this research, the importance of exploring plant-derived antimicrobials was emphasized in response to the rising threat of antibiotic resistance. The literature review further highlighted the rich phytochemical profile of onion leaves and their potential as an accessible natural remedy. The results obtained from this research confirmed the presence of key secondary metabolites including alkaloids, flavonoids, tannins, saponins, steroids, and cardiac glycosides with some carbohydrates and proteins, while anthraquinones were absent. These findings provide strong evidence of the biochemical richness of onion leaves and their possible role in bioactivity. Antibacterial assays revealed that the ethanol extracts of *Allium cepa* leaves exhibited selective inhibitory effects. The maximum inhibition zone recorded was 3.04 mm against *Staphylococcus aureus* at 75 mg/mL, followed by 2.68 mm against *Escherichia coli* and 2.35 mm against *Bacillus subtilis*. In contrast, *Shigella* that demonstrated no inhibition at all tested concentrations. The Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) confirmed bactericidal activity at 75 mg/mL for *Staphylococcus aureus* and *Escherichia coli*. Compared with the standard antibiotic ceftriaxone, which produced inhibition zones between 3.28 – 4.85 mm, the extract showed appreciable activity, suggesting it as an alternative or complementary antimicrobial source. This study concludes that onion leaves, often regarded as agricultural waste, contain significant phytochemicals with demonstrable antibacterial potential. The presence of bioactive compounds supports their use in ethnomedicine and justifies their potentials in developing new antibiotics.

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