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Comparative antibacterial potential of *Tetrapleura tetraptera* extracts on some common food pathogens

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ABSTRACT

The harmful effects of chemical preservatives on food safety have spiralled interest in natural antimicrobial agents. This study investigates the antibacterial potential of *Tetrapleura tetraptera* (Aridan pods) extracts against common foodborne pathogens.To evaluate the antimicrobial efficacy of Dimethyl Sulphate, Methanolic, and n-hexane extracts from *Tetrapleura tetraptera* pods and assess their potential as natural alternatives to synthetic preservatives.The sensitivity was tested using the agar well diffusion method against isolated organisms from food, including *Escherichia coli, Salmonella typhi, Serratia marcescens, Staphylococcus aureus*, and *Staphylococcus epidermidis*. Zones of inhibition were measured to assess efficacy.The zone of inhibition measurements against *Escherichia coli*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Salmonella*, and *Serratia marcescens* were as follows: 20.00 mm, 18.00 mm, 19.6 mm, 16.5 mm, and 17.00 mm, respectively, for the dimethyl sulfate extracts; 15.30 mm, 14.00 mm, 11.07 mm, 16.0 mm, and 17.0 mm for the methanolic extracts; and 13.00 mm, 11.24 mm, 13.00 mm, 12.00 mm, and 15.00 mm for the n-hexane extracts. significant difference ($p > 0.05$) as revealed by analysis of variance was noticed on the isolates for the dimethyl sulfate extracts but not for the aqueous or methanolic extracts of *Tetrapleura tetraptera* pod.*Tetrapleura tetraptera* extracts demonstrate potent antibacterial properties, suggesting their potential as natural alternatives to synthetic antibiotics in food preservation.

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1. Introduction

Foodborne diseases are a growing global issue, with more frequent outbreaks linked to the consumption of contaminated food (1).

The rise of antibiotic-resistant microorganisms emancipating from food consumption further complicates the challenge, highlighting the need for new antimicrobial agents that can ensure food safety without affecting food quality (2).

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Numerous studies have explored viable alternatives to synthetic antibiotics, with plants emerging as promising and sustainable options (3).

Traditional methods of food preservation often rely on chemical additives and synthetic antibiotics. While these methods have been successful, there are growing concerns about their negative effects on human health and the environment (4). This has led to an increasing interest in exploring natural alternatives, such as plant-derived antimicrobial compounds, which offer both food safety and environmental sustainability (5).

Tetrapleura tetraptera which is commonly known as Aridan tree in Southern Nigeria belongs to the Mimosaceae family (6, 7, 8). The fruit of *Tetrapleura tetraptera* possess a fragrant, characteristically, pungent aromatic odour. This may account for the insecticidal potential of the plant (9). The extract of the plant has been reported to be toxic to tadpoles (10) and larvae of *Anopheles gambiae* (11).

The *Tetrapleura tetraptera*, commonly referred to as the Aridan tree in Southern Nigeria, belongs to the Mimosaceae family (6, 7, 8). Its fruit is known for its distinctively pungent and aromatic fragrance, which may contribute to its insecticidal properties. Studies have shown that extracts from the plant (9) are toxic to tadpoles (10) and the larvae of *Anopheles gambiae* (11).

Beyond its pesticidal potential, *Tetrapleura tetraptera* is widely used as a culinary spice in traditional dishes across rural areas in countries like Ghana and Nigeria. It is rich in phytochemicals and bioactive compounds (6, 8) which are believed to be responsible for its therapeutic properties (12).

Ethnobotanical studies and scientific validations have highlighted the plant's diverse medicinal uses, including its cytoprotective, piscicidal, anti-gonadotropic, antimutagenic, and antimalarial properties (13). It has been employed in the management of conditions such as inflammation, arthritis, hypertension, diabetes, schistosomiasis, and epilepsy (8, 9). The plant also exhibits antioxidant, analgesic (11), aphrodisiac, anticonvulsant, anti-ulcerative, neuromuscular, hypotensive, cardiovascular, and hypoglycemic activities. Furthermore, it is used to control intestinal parasites (14) and is traditionally consumed by breastfeeding mothers to prevent postpartum contractions.

In the food industry, ensuring food safety is critical due to the risk that contaminated food can harbor harmful microorganisms, which poses significant public health risks. Food-associated pathogens such as *Escherichia coli*, *Salmonella* species, *Staphylococcus* species, *Listeria monocytogenes* are commonly associated with foodborne illnesses and have demonstrated resistance to antibiotics, highlighting the urgent need for effective control measures against these harmful organisms (15).

Previous studies on the antimicrobial properties of *Tetrapleura tetraptera* have primarily investigated the use of petroleum ether, water-based, and alcohol-based extracts (16), cold water and alcohol extracts (17), as well as acetone, water, and ethanol extracts (18) as extraction solvents. Consequently, this research focuses on evaluating the effects of dimethyl sulfate, ethanol, and water extracts of *Tetrapleura tetraptera* pod on certain bacterial microorganisms associated with food spoilage.

2. Materials and Methods

2.1. Sample collection

Dry and wet *Tetrapleura tetraptera* pods were obtained from Lafenwa market, Ogun State, Nigeria. The pods were washed and air-dried. The samples were then blended to obtain fine powder.

2.2. Source and isolation of food pathogens

Five different food samples with three replicates were used. Food samples such as kunu, zobo, suya, rice, and salad were collected from different locations to obtain difference in environmental pathogens. Food samples were collected in sterile universal containers and zip lock bags and transported to the microbiology laboratory for the isolation process.

Serial dilution and culturing techniques were used to isolate foodborne pathogens. Samples were diluted in sterile water, and different dilutions were plated on selective media such as Mannitol Salt Agar (MSA) and Xylose Lysine Deoxycholate (XLD) agar. Plates were incubated at 37°C for 24 h, and colonies were sub-cultured to obtain pure isolates.

2.3. Identification of isolates

Bacterial isolates were identified using both macroscopic and microscopic techniques. Gram staining was performed to determine Gram reaction, followed by biochemical tests such as catalase, indole, citrate, and motility tests to confirm the identity of the pathogens. Isolates identified include *Escherichia coli, Staphylococcus aureus, Staphylococcus epidermidis, Salmonella typhi, and Serratia marcescens.*

2.4. Method of extraction

The extraction process employed the soaking technique as outlined by Kigigha et al. (12, 19), with minor adjustments. A total of 10 g of the pulverized samples were separately extracted using 20 mL of dimethyl sulfate and methanol. The samples were left to soak for 72 h, after which the mixture was first strained through muslin cloth and then filtered using Whatman filter paper. They were allowed to evaporate before being concentrated at 40°C and stored at 4°C.

n-Hexane Extraction: 40g of powdered *Tetrapleura tetraptera* pods were subjected to Soxhlet extraction using 100 mL of n-hexane. The extracted oil was concentrated using a rotary evaporator and stored at 4°C.

2.5. Microbial inhibition analysis

The inhibition zones produced by the different isolates were assessed using the agar well diffusion technique as previously outlined by (20, 21) with slight modification by Agu et al. (22), Kigigha et al. (19) and Izah et al. (23).

Approximately 0.4 mL of the test microbes suspended in peptone water and incubated for 24 h were evenly spread onto Mueller-Hinton agar plates. Three wells, each 6 mm in diameter, were created using a sterile cork borer. About 2 mL of each extract was individually introduced into the wells. A positive control was also set up (1% Ciprofloxacin). All the plates were incubated for 24 h. The zones of inhibition were measured using a metre rule in millimetres.

2.6. Statistical analysis

The statistical analysis was performed using the Statistical Package for Social Sciences (SPSS) software. Data were presented as mean ± standard error. Significant differences were analyzed using one-way analysis of variance, and the Tukey Honestly Significant Difference (HSD) test was employed to identify the source of variations observed, with a significance level set at $p=0.05$.

3. Results

Isolation and identification of food pathogens

Five bacterial isolates were obtained from food samples and identified using biochemical tests. Table 1 presents the results of these tests for each isolate, confirming the organisms based on the Catalase, Citrate, H2S, Indole, and Motility tests.

Table 1. Biochemical identification of isolated bacteria from food samples.

Biochemical tests	*Escherichia coli*	*Staphylococcus aureus*	*Staphylococcus epidemidis*	*Salmonella typhi*	*Serratia marcescens*
Catalase	+	+	+	+	+
Citrate	-	+	-	-	+
H_2S	-	-	+	+	-
Indole	+	-	-	-	-
Motility	+	-	-	+	+
Gram staining	-	+	+	-	-
Shape	Rod	Cocci	Cocci	Rod	Rod

3.1. Phytochemical composition

Table 2 presents the results of the phytochemical analysis, indicating elevated levels of alkaloids (21.53 ± 0.01%), saponins (13.17 ± 0.07%), tannins (7.36 ± 0.03%), and flavonoids (7.93 ± 0.08%) in the pods of *Tetrapleura tetraptera*. Fig. 1 illustrates the statistical comparison of the phytochemical quantities

Table 2. Result of phytochemical analysis of *Tetrapleura tetraptera* pod.

S/N	Alkaloids	Saponins	Tannins	Flavonoids	Total Phenolic compound
1	21.53 ± 0.01	13.17 ± 0.07	7.36 ± 0.03	7.93 ± 0.08	3.07 ± 0.02
2	20.98 ± 0.02	12.98 ± 0.02	5.63 ± 0.04	7.76 ± 0.04	2.69 ± 0.00
3	20.95 ± 0.15	13.03 ± 0.01	5.73 ± 0.02	7.76 ± 0.06	2.77 ± 0.02
4	21.13 ± 0.01	13.31 ± 0.01	7.31 ± 0.03	8.25 ± 0.05	3.02 ± 0.01

Values are expressed as Mean ± SD, n=3 for each replicate

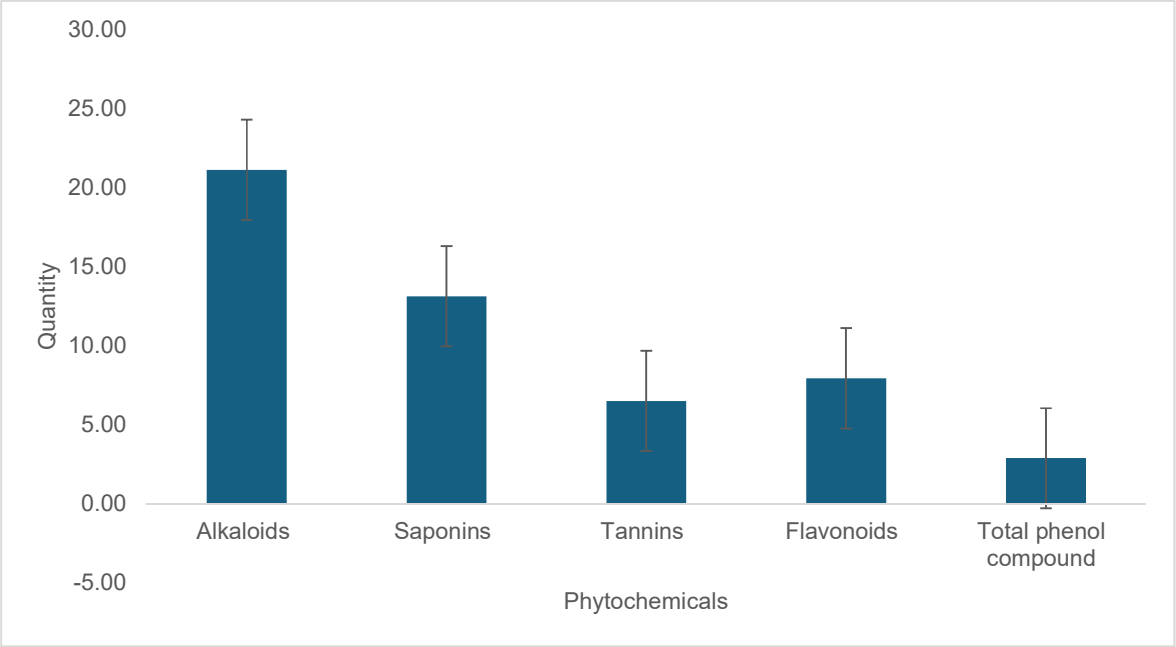

Figure 1. Phytochemical content of *Tetrapleura tetraptera*.

Table 3. Zones of Inhibition (mm) of *Tetrapleura tetraptera* pods against some bacterial isolates.

Isolates	Extracts		
	Dimethyl sulfate	Methanol	n- hexane
Escherichia coli	20.00 ±1.53$_a$	15.30± 0.83$_a$	13.00± 0.04$_a$
Staphylococcus aureus	18.00± 1.02$_a$	14.00± 0.80$_a$	11.00± 0.02$_a$
Staphylococcus epidermidis	19.60± 1.00$_a$	11.70± 0.75$_a$	13.00± 0.04$_a$
Salmonella	16.50± 0.07$_b$	16.00± 0.88$_a$	15.00± 0.06$_a$
Serratia marcescens	17.00± 0.09$_b$	17.00± 0.98$_a$	15.00± 0.06$_a$

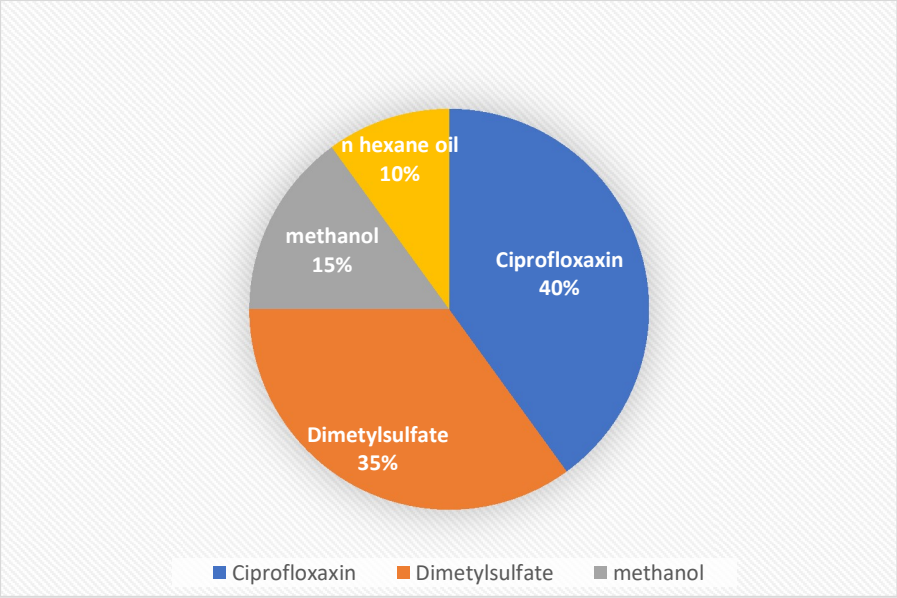

Figure 2. Percentage zones of inhibition exhibited by extracts against bacterial isolates.

3.2. Antimicrobial activity

Table 3 shows the zones of inhibition exhibited by *Escherichia coli, Staphylococcus aureus, Staphylococcus epidermidis, Salmonella and Serratia marcescens,* when exposed to Dimethyl sulfate, methanolic and n-hexane extracts of *Tetrapleura tetraptera* and positive control (1% ciprofloxacin). **Fig. 2** shows the frequency of the zones of inhibition exhibited by the microorganisms using the extracts and antibiotic.

4.Discussion

Five distinct bacterial strains were isolated from food samples, including *Escherichia coli*, *Salmonella typhi*, *Serratia marcescens*, *Staphylococcus aureus*, and *Staphylococcus epidermidis*, which aligns with the findings of Liu et al. (24). The isolates were identified based on their cultural, biochemical, and morphological characteristics, as described in the work of Topić Popović et al. (25).

Phytochemical analysis showed that *Tetrapleura tetraptera* pods contain considerable amounts of tannins, alkaloids, saponins, flavonoids, and phenols, all of which are known for their antimicrobial properties. The presence of these compounds highlights the potential of *Tetrapleura tetraptera* extracts as effective antimicrobial agents, as reported by Topić Popović et al. (25). Various studies have suggested that the medicinal properties of plants, including *Tetrapleura tetraptera*, are attributed to their bioactive components (3, 19).

For instance, Achi (17) found tannins and glycosides in ethanol and cold water extracts of *Tetrapleura tetraptera* whole pods, but no alkaloids, saponins, flavonoids, or anthraquinones. Ebana et al. (16) reported the presence of reducing compounds (such as polyphenols, phlorotannins, anthraquinones, and hydroxymethyl anthraquinones), alkaloids, flavonoids, glycosides, and the absence of tannins and saponins in aqueous and ethanolic extracts of *Tetrapleura tetraptera*. Lin et al. (18) explored the mechanism of *Tetrapleura tetraptera* root extract against *E. coli* and *Staphylococcus aureus*, concluding that the plant could disrupt respiratory metabolism by inhibiting the organisms through the Embden–Meyerhof–Parnas and hexose monophosphate pathways.

The extracts of *Tetrapleura tetraptera* pods exhibited antibacterial activity, consistent with previous research. Ebana et al. (16) reported that petroleum ether, aqueous, and ethanolic extracts of *Tetrapleura tetraptera* are effective against *E. coli*, *Staphylococcus aureus*, and *Pseudomonas aeruginosa*. Similarly, Achi (17) found that cold water and ethanol extracts of *Tetrapleura tetraptera* are potent against *Staphylococcus aureus*, *E. coli*, and *Pseudomonas aeruginosa*. A study by Adusei et al. (26) also showed that water, acetone, and ethanolic extracts of *Tetrapleura tetraptera* are effective against *Staphylococcus aureus*, *E. coli*, *Pseudomonas aeruginosa*, and *Proteus* species. However, the zones of inhibition observed in this study differed from those previously reported, which may be attributed to differences in the sources of the isolates, extract concentrations, solvents used, the physical condition of the plant material (such as age), and the strains of microbial isolates, among other factors (27).

The zone of inhibition by extracts of *Tetrapleura tetraptera* pods followed this order: Ciprofloxacin > Dimethyl sulfate > Methanol > n-hexane for all isolates. The variation between extracts may be due to differences in the solvent properties used in the study. It has been reported that solvent polarity affects the zone of inhibition in plants with antibacterial properties (16).

The variation in the mean values of the different isolates may be due to differences in their metabolism, physiology, nutrition, and biochemistry (28). Environmental factors such as temperature and pH of the extracts might also contribute to significant variations among the isolates (29).

Tetrapleura tetraptera has shown potency against *Escherichia coli*, *Salmonella*, *Serratia marcescens*, *Staphylococcus aureus*, and *Staphylococcus epidermidis*, suggesting it may serve as a broad-spectrum antibiotic. Moreover, based on the study's findings, the known antibiotics exhibited higher activity than the various extracts of *Tetrapleura tetraptera*. This variation may be due to the specificity of the known antibiotics and their concentrations. Additionally, many of the known antibiotics tested have previously been shown to be effective against both gram-positive and gram-negative organisms (9, 28).

5.Conclusion

The isolation and identification of five bacterial strains from food samples, including *Escherichia coli* and *Staphylococcus aureus*, underscore the relevance of these pathogens in food safety. The phytochemical analysis of *Tetrapleura tetraptera* pods demonstrates their significant antimicrobial properties, attributed to the presence of various bioactive compounds such as tannins, alkaloids, and flavonoids. These findings align with existing literature that supports the medicinal potential of *Tetrapleura tetraptera* extracts. While the extracts showed broad-spectrum antibacterial activity against the isolated pathogens, variations in the zones of inhibition compared to previous studies can be attributed to several factors, including the source of isolates, extract concentration, and solvent properties. The study reinforces the promise of *Tetrapleura tetraptera* as a natural antimicrobial agent, suggesting its potential use in developing alternative treatments for bacterial infections. Further research is warranted to optimize extraction methods and explore the mechanisms of action, which may contribute to the development of effective, plant-based antimicrobial agents.

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Authorship contribution:

Badmos Amina: Conceptualization, Supervision, Writing – review and editing, Methodology, Project administration. **Samsudeen Onikede and Abigeal Oluremi**: Data curation, Formal analysis, Investigation. Writing – original draft.

Declaration of Competing interest

The authors declare that there is no competing interest.

Data Availability

Data are made available in the body of the article.

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References

1. Farias AdS, Akutsu RdCCdA, Botelho RBA, Coelho Araújo WM, Silva IC, Sávio KE, et al. Food safety conditions in home-kitchens: A cross-sectional study in the federal district/Brazil. Int J Environ Res Public Health. 2020;17(13):4897.
2. Wu-Wu JWF, Guadamuz-Mayorga C, Oviedo-Cerdas D, Zamora WJ. Antibiotic resistance and food safety: perspectives on new technologies and molecules for microbial control in the food industry. Antibiotics. 2023;12(3):550.
3. Epidi J, Izah S, Ohimain E. Antibacterial and synergistic efficacy of extracts of *Alstonia boonei* tissues. Br J Appl Sci. 2016;1(1):21-6.
4. Izah S, Etim N, Ilerhunmwuwa I, Silas G. Evaluation of crude and ethanolic extracts of *Capsicum frutescens* var. minima fruit against some common bacterial pathogens. Int J Complement Altern Med. 2019;12(3):105-8.
5. Izah SC, Zige DV, Alagoa KJ, Uhunmwangho E, Iyamu A. Antibacterial efficacy of aqueous extract of *Myristica fragrans* (Common Nutmeg). EC Pharmacol Toxicol. 2018;6(4):291-5.
6. Koma OS, Olawumi OO, Godwin E-U, Theophilus OA. Phytochemical screening, in-vitro antimicrobial activity and antioxidant characteristics of *Tetrapleura tetraptera* extracts. Eur J Med Plants. 2016;17(2):1-10.
7. Enema O, Adesina S, Umoh U, Eseyin O. Gas chromatography-mass spectroscopy (GC-MS) studies of fixed oil of leaf of *Tetrapleura tetraptera* Taub. (Mimosaceae). J Pharmacogn Phytochem. 2019;8(6):1237-41.
8. Enema O, Umoh U, Thomas P, Adesina S, Eseyin O. Phytochemical and antioxidant studies of leaf of *Tetrapleura tetraptera* (schum and thon) Taubert (Mimosaceae). Br J Pharm Med Res. 2019;4:1865-75.
9. Ojewole JA, Adewunmi CO. Anti-inflammatory and hypoglycaemic effects of *Tetrapleura tetraptera* (Taub) [Fabaceae] fruit aqueous extract in rats. J Ethnopharmacol. 2004;95(2-3):177-82.
10. Oniya M, Idowu O. Evaluation of the toxicity of the oil extract of *Tetrapleura tetraptera* on tadpoles [Amphibia: Anura]. FUTA J Res Sci. 2013;1:172-6.
11. Seiyaboh EI, Odubo TC, Izah SC. Larvicidal activity of *Tetrapleura tetraptera* (Schum and Thonn) Taubert (Mimosaceae) extracts against *Anopheles gambiae*. Int J Adv Res Microbiol Immunol. 2020;2(1):20-5.
12. Kigigha L, Izah S, Ehizibue M. Activities of *Aframomum melegueta* Seed Against *Escherichia coli*, *Staphylococcus aureus* and *Bacillus* species. Point J Bot Microbiol Res. 2015;1(2):23-9.
13. Adesina S, Iwalewa E, Johnny I. *Tetrapleura tetraptera* Taub-ethnopharmacology, chemistry, medicinal and nutritional values-a review. Br J Pharm Res. 2016;12(3):1-22.
14. Aladesanmi AJ. *Tetrapleura tetraptera*: Molluscicidal activity and chemical constituents. Afr J Tradit Complement Altern Med. 2007;4(1):23-36.
15. Ullah F, Ayaz M, Sadiq A, Ullah F, Hussain I, Shahid M, et al. Potential role of plant extracts and phytochemicals against foodborne pathogens. Appl Sci. 2020;10(13):4597.
16. Ebana R, Edet U, Ekanemesang U, Ikon G, Etok C, Edet A. Antimicrobial activity, phytochemical screening and nutrient analysis of *Tetrapleura tetraptera* and *Piper guineense*. Asian J Med Health. 2016;1(3):1-8.

17. Achi O. Composition and antibacterial activities of *Tetrapleura tetraptera* Taub. pod extracts. 2010.
18. Lin L, Agyemang K, Abdel-Samie MAS, Cui H. Antibacterial mechanism of *Tetrapleura tetraptera* extract against *Escherichia coli* and *Staphylococcus aureus* and its application in pork. J Food Saf. 2019;39(6):e12693.
19. Kigigha LT, Selekere RE, Izah SC. Antibacterial and synergistic efficacy of acetone extracts of *Garcinia kola* (Bitter kola) and *Buchholzia coriacea* (Wonderful kola). J Basic Pharmacol Toxicol. 2018;2(1):13-7.
20. Lino A, Deogracious O. The *in vitro* antibacterial activity of *Annona senegalensis*, *Securidacca longipendiculata* and *Steganotaenia araliacea*-Ugandan medicinal plants. Afr Health Sci. 2006;6(1):31-5.
21. Doherty V, Olaniran O, Kanife U. Antimicrobial activities of *Aframomum melegueta* (Alligator pepper). Int J Biol. 2010;2(2):126-31.
22. Agu G, Thomas B. Antibacterial activities of ethanol and aqueous extracts of five Nigerian medicinal plants on some wound pathogens. Nat Sci. 2012;10(2):78-84.
23. Izah SC, Aseibai ER. Antibacterial and synergistic activities of methanolic leaves extract of lemon grass (*Cymbopogon citratus*) and rhizomes of ginger (*Zingiber officinale*) against *Escherichia coli*, *Staphylococcus aureus* and *Bacillus subtilis*. Acta Sci Microbiol. 2018;1(6):26-30.
24. Liu Y, Wei C, Wan H, Liang X, Jiang T, Dong Y, et al. Preliminary Study on Rapid and Simultaneous Detection of Viable *Escherichia coli* O157:H7, *Staphylococcus aureus*, and *Salmonella* by PMA-mPCR in Food. Molecules. 2023 Aug 3;28(15):5835.
25. Topić Popović N, Kepec S, Kazazić SP, Strunjak-Perović I, Bojanić K, Čož-Rakovac R. Identification of environmental aquatic bacteria by mass spectrometry supported by biochemical differentiation. PLoS One. 2022 Jun 3;17(6):e0269423.
26. Adusei S, Otchere JK, Oteng P, Mensah RQ, Tei-Mensah E. Phytochemical analysis, antioxidant and metal chelating capacity of *Tetrapleura tetraptera*. Heliyon. 2019 Nov 1;5(11):e02762.
27. Aderibigbe AO, Iwalewa EO, Adesina SK, Agboola OI. Studies of behavioural and neural mechanism of Aridanin isolated from *Tetrapleura tetraptera* in mice. Int J Pharmacol. 2010;6(4):480-486.
28. Kigigha LT, Biye SE, Izah SC. Phytochemical and antibacterial activities of *Musanga cecropioides* tissues against *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Proteus* and *Bacillus* species. Int J Appl Res Technol. 2016;5(1):100-7.
29. Guglielmi P, Pontecorvi V, Rotondi G. Natural compounds and extracts as novel antimicrobial agents. Expert Opin Ther Pat. 2020 Dec 1;30(12):949-62.