

Research Paper



Detoxification of bacterial toxins in soured soups using the extract of aidan tree (*tetrapleura tetraptera* L.) fruit

Saturday Okpabi^{1*}, Uche Joy Nzube², Robert Chioma Lilia³, Prof. K. H. Ogbonda⁴

^{1*,2,3,4}Biology Department, Faculty of Natural and Applied Sciences, Ignatius Ajuru University of Education, Rumuolumeni Port Harcourt, Rivers State, Nigeria.

Article Info

Article History:

Received: 03 December 2023

Revised: 12 February 2024

Accepted: 20 February 2024

Published: 08 April 2024

Keywords:

Detoxification

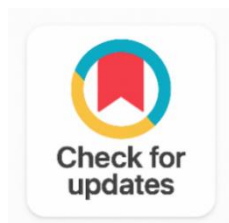
Bacterial Toxin

Soured Soups

Extract

Aidan Tree

Native Soup



ABSTRACT

The study evaluated the use of *Tetrapleura tetraptera* fruit extract to detoxify bacterial toxins in soured soups. *Tetrapleura tetraptera* fruit extracts were prepared and analyzed for phytochemicals. Three different types of soured soups (egusi, ogbono, and Abua native soup) were used. The soups were prepared and left for 24 hours at room temperature to sour. The pour-plate method and streaking techniques were used to isolate and identify the bacteria. The lateral flow assay device, Biothreat Alert test strip, and enzyme immunoassay kits were used to detect the bacterial toxins, while the extracts were used to test the detoxifying effectiveness of the bacterial toxins. The results showed that *Lactobacillus* sp, *Bifidobacterium* sp, *Streptococcus* sp, *Pediococcus* sp, *Leuconostoc* sp *Bacillus cereus*, *Clostridium perfringens*, *Campylobacter jejuni*, *Vibrio parahaemolyticus*, *Yersinia enterocolitica*, *Salmonella* sp, *Shigella* sp, *Escherichia coli*, *Staphylococcus aureus*, *Listeria monocytogenes* were associated with the soured soups; alkaloids, cardiac glycosides, flavonoids, phenols, saponins, steroids, tannins, and terpenoids were present. The quantity of saponin (5.670 ± 0.15), steroid (4.102 ± 0.13), terpenoid (3.194 ± 0.03), and alkaloid (0.596 ± 0.01) were very high in concentrations; phenol (0.373 ± 0.02) and tannin (0.366 ± 0.02) were present in moderately high concentrations, while cardiac glycoside (0.007 ± 0.00) and flavonoid (0.073 ± 0.00) were present in low concentrations. The toxins detected in the soured soups included *Bacillus cereus* and *Staphylococcus aureus* exotoxins. Therefore, it is concluded that the extract of *Tetrapleura tetraptera* has no detoxifying effect on the stability of the bacterial toxins and should, therefore, be discouraged.

Corresponding Author:

Saturday Okpabi

Biology Department, Faculty of Natural and Applied Sciences, Ignatius Ajuru University of Education, Rumuolumeni Port Harcourt, Rivers State, Nigeria.

Email: okpabis@gmail.com

Copyright © 2024 The Author(s). This is an open access article distributed under the Creative Commons Attribution License, (<http://creativecommons.org/licenses/by/4.0/>) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

1. INTRODUCTION

The preservation of soups is a critical issue faced by rural communities in Nigeria, particularly due to the lack of appropriate preservation methods and limited access to electricity. As a result, soups often get fermented or spoiled, leading to significant food waste [1]. In rural areas with sparse electricity supply, the absence of refrigeration and other modern preservation techniques poses challenges for keeping soups fresh and safe for consumption over extended periods. When soups are not properly preserved, they may become contaminated with harmful pathogenic bacteria, which often secrete poisons (toxins) into the soups, making them unfit for consumption [2]. Consumption of such contaminated soups can lead to food infections, food poisoning, and other illnesses, posing significant health risks to individuals [3], [4]. The fermentation process in soups is primarily driven by the metabolic activities of bacteria, which produce poisonous substances that cause the soups to sour. This souring process further exacerbates the problem, making the soups unsuitable for consumption and contributing to the economic burden [5]. To address this issue and make the soups safe for consumption, the paper aimed to focus on identifying and removing the bacterial toxins responsible for the souring of soups. By understanding the types of bacteria and toxins involved and developing techniques to neutralize or eliminate these harmful substances, we can make the soups safe to eat even after they have soured [6]. The lack of available preservation techniques in the context of limited resources emphasizes the urgency and importance of this research. The successful development of a method of neutralizing the poison (microbial) in the soups could significantly improve food safety, reduce food waste, and alleviate the economic burden on the less fortunate population in rural communities [7]. It would also contribute to addressing food security issues, as fewer soups would be wasted and communities could better utilize available resources to sustain themselves. Many tribes and cultures in Nigeria make use of red palm oil, onions, and even aidan tree fruit to restore and recover their sour soups [8]. Charcoal was used as a preservative in soup, removing bad taste and smell. It was heated to restore its original taste, with onions, red palm oil, crayfish, and seasoning cubes added. This practice is still prevalent in some localities today [9].

2. RELATED WORK

[10], Examined *Clostridium botulinum* toxin inactivation in selected soured soups in South-South region of Nigeria and revealed that *Clostridium botulinum* neurotoxin was denatured at a temperature of 100°C. Also, the serological activity of the toxins treated with palm oil was unaffected, whereas, the joint treatment with heat and palm oil remained denatured.

[9], Evaluated detoxification of bacteriotoxins in sour soups using red oil, heat, and onion and indicated that bacterial toxins detected in the sour soups included *Bacillus cereus* toxins, *Clostridium botulinum* toxins, and *Staphylococcus aureus* toxins and that the test materials were not able to detoxify the sour soups.

[11] Investigated “the correlation between dominant bacteria and primary metabolites during the fermentation of sour soup. They found that fermentation led to a decrease in the presence of native bacterial strains, with lactic acid bacteria (LAB) becoming the dominant microbes and this shift in microbial composition resulted in changes in the relative abundance of bacteria and variations in the types and quantities of metabolites.

In a study conducted by [12], the objective was to examine and identify bacterial toxins and thermal stability of enterotoxins in food present in food sellers and some vegetables found in market and

revealed that all the soup samples that were tested had different levels of bacterial toxins and growth, ranging from 1.0×10^5 to 3.0×10^6 cfu/ml.

[13] Assessed the bacteria in recently spoiled ofe-okwu soup, isolated several strains of Bacillus, and hypothesized that these microbes could generate toxins in the soup.

[14] Found that Bacillus cereus spores exhibited resilience to the sterilization process, as they remained intact and maintained stability for a minimum of three years at temperatures as high as 00°C.

2.1. Aim and Objectives of the Study

The aim of the study was to use Tetrapleura tetraptera fruit extract to detoxify (render ineffective) bacterial toxins in soured soups. The objectives were to:

1. determine the phytochemical composition of Tetrapleura tetraptera fruit extract (qualitative and quantitative);
2. isolate and characterize the bacteria in the soured soups;
3. investigate (test) some soured soups for contamination with Bacillus cereus toxins, Clostridium botulinum toxins, and Staphylococcus aureus toxins;
4. ascertain the detoxifying effect of Tetrapleura tetraptera fruit extract on Bacillus cereus toxin, Clostridium botulinum toxin, and Staphylococcus aureus toxin in soured soups;

3. METHODOLOGY

3.1. Phytochemical Analysis of Aidan Fruit Extract

Collection of Aidan Plant Fruit: The Aidan Plant fruits (Tetrapleura tetraptera) were purchased from a fruit vendor at Abua main market and subsequently transferred to the laboratory for processing and analysis.

Production of Fruit Extract: The fruits were dehydrated in a shaded environment for a duration of three weeks and subsequently crushed into a fine powder using a sterile electric blender. Three duplicates of about four grammes (4g) of powdered fruit were immersed in thirty millilitres (30 ml) of hot water. Every conical flask was sealed with cotton wool that was wrapped in aluminium foil and placed in a rotary shaker for a duration of two days. The extracts were purified using a sterile filter paper (Whatman No 1).

3.2. Phytochemical Analysis (Qualitative)

Flavonoid (Alkaline Reagent Test): A precisely measured volume of 0.2 ml of the extract was subjected to the addition of six drops of a 2% sodium hydroxide solution. The appearance of a vibrant yellow hue, which subsequently turned into a transparent solution upon the introduction of a weak acid, serves as evidence for the existence of flavonoids in the extract [15].

Alkaloids (Mayer's Test): The extract was diluted in 5 ml of 1% dilute hydrochloric acid and filtered. The amount of extract used was 0.5 ml, which is equivalent to one half gramme. The filtrate underwent treatment with Mayer's reagent, which is composed of Potassium mercuric iodide. A favourable indication for alkaloids in the extract is observed when a yellow-colored precipitate forms [15].

Terpenoids (Salkowski's Test): 0.1 ml of the extract was combined with 0.5 ml of chloroform, and then 1 ml of concentrated sulphuric acid was added. The occurrence of a reddish-brown solid that forms when two substances react is a sign that terpenoids are present in the extract [15].

Tannins (Ferric Chloride Test): A precisely measured amount of the extract (0.2ml) was combined with an equal proportion of distilled water in a test tube. Subsequently, three drops of diluted ferric chloride were introduced. The appearance of a brownish blue or dark hue indicated the presence of tannins in the extract [15].

Steroids (Liebermann-Burchard's Test): The 0.5 ml sample was combined with chloroform and sulphuric acid, resulting in the observation of a red colour in the chloroform layer below. This colour change confirms the presence of steroids.

Saponins (Foam Test): A precisely measured amount of 0.2 ml of the extracts was combined with 6 ml of distilled water in a graduated cylinder. The mixture was rapidly agitated for a duration of 15 minutes.

Observing the occurrence of bubbles or long-lasting foam for a duration of 10 minutes can serve as an indication of the existence of saponins in the extract [15].

Phenols (Ferric Chloride Test): A precisely measured quantity of 0.2 millilitres of the extracts will be combined with 2 millilitres of a 5% aqueous ferric chloride solution. The presence of phenols in the extract is indicated by the development of a bluish hue, as reported by [15].

Cardiac Glycoside Test: Exactly two (2.0 ml) milliliters of the extract was dissolved in two (2) ml of chloroform; two (2) ml of sulphuric acid was added carefully and shaken gently. A reddish-brown colouration is an indication of the presence of a steroidal ring [15].

3.3. Quantitative Phytochemical Analysis

Determination of Total Flavonoid Content: The flavonoid content of *T. tetraptera* extract was measured using the aluminium chloride colorimetric method, with quercetin as the standard. Concentrations of 10, 25, 50, 75, and 100 µg/ml were produced in methanol. The mixture was then mixed with distilled water, 5% sodium nitrate, aluminium chloride, and sodium hydroxide. The absorbance of the reaction mixture was measured at 510nm using a single-beam UV-VIS spectrophotometer. The total flavonoids were quantified using a linear equation derived from a standard curve with quercetin. The results were reported as milligrammes of quercetin equivalent (QE) per gramme of dry extract. The extract was then combined with chloroform and concentrated sulphuric acid. A positive indication for steroids in the extract was observed when a red color appeared in the lower chloroform layer.

Determination of Total Phenol Content: The quantification of the phenolic compounds in the *T. tetraptera* extract was carried out by employing “the Folin-Ciocalteu reagent. Gallic acid will serve as the reference standard for constructing the calibration curve. A 0.5 ml portion of gallic acid solutions with concentrations of 10, 20, 40, 80, and 100 µg/ml were combined with 2ml of Folin-Ciocalteu reagent, which had been diluted 1:10 with de-ionized water. The resulting mixture was then neutralised with 4ml of a 7.5% sodium carbonate solution. The reaction mixture was left at room temperature for 30 minutes with periodic agitation to facilitate the development of colour. The intensity of the produced blue colour was quantified at a wavelength of 765 nm using a single-beam UV-VIS spectrophotometer (UV mini-1240). The total phenols content was quantified using a linear equation derived from a standard curve constructed with gallic acid. The results were represented as milligrammes per gramme of gallic acid equivalent (GAE) of dry extract, following the method described by [15].

Determination of Total Saponin Content: A solution of diosgenin, vanillin reagent, and 72% sulphuric acid was prepared by adding 5000 µL of water to 100 µL of diosgenin, 500 µL of vanillin reagent, and 5 ml of sulphuric acid. The solution was then heated at 60°C for 10 minutes, cooled, and absorbance recorded at 544nm. The total saponin content was determined using a standard curve and expressed as mg/g diosgenin equivalent.

Determination of Total Alkaloid Content: A crude extract of *T. tetraptera* was mixed with 10% hydrochloric acid in ethanol and filtered. The filtrate was concentrated using a rotary evaporator and then precipitated with 15 drops of concentrated ammonium hydroxide. Following a sedimentation period of 3 hours, the liquid portion was discarded, and the solid particles were rinsed with a solution of 0.1M ammonium hydroxide and then separated by filtration. Subsequently, the remaining substance was subjected to drying and weighing, and the proportion of alkaloid was determined by dividing the weight of the residue by the weight of the sample multiplied by 100.

Determination of Tannin: The ground sample was solubilized in distilled water and agitated for 1 hour. The liquid was then poured into a 50-ml flask. A 0.1 gram tannic acid solution was dissolved in 100 ml of distilled water and transferred to a 50-ml flask. A blank specimen was prepared using 5 ml of distilled water. A Folin-Dainas reagent and sodium carbonate solution were added. The absorbance of the dark color was measured in a spectrophotometer after incubation at 25°C for 90 minutes.

Determination of Cyanogenic Glycosides: A colorimetric method was employed to analyze the absorbance of *T. tetraptera* in a solution. The mixture was mixed with distilled water and cyanide solution, and an alkaline picrate paper was suspended above the mixture. The setup was left undisturbed for 18

hours, then rinsed with distilled water the next day. The absorbance was measured using a spectrophotometer at a wavelength of 540 nm, with a reagent blank serving as a reference.

3.4. Isolation and Characterization of Bacteria in Soured Soups

Preparation of Soups: The study used three soup varieties, egusi, ogbono, and Abua local soup, made from materials from Abua main market in Rivers State, Nigeria. The soups included fish, clams, shrimps, periwinkles, pepper, palm oil, crayfish, onion bulb, seasoning cubes, salt, cocoyam, pumpkin leaves, fish, and beef. The ingredients were meticulously cleansed, fermented for 48 hours, and then filtered using a membrane filter to remove solid particles. The soups were prepared according to local techniques.

Isolation of Bacteria in Soured Soups: Each soured soup sample was homogenised with five millilitres (5 ml) and one millilitre (1 ml) from the resulting mixture was utilised for serial dilutions. The pour-plate method and streaking procedures were employed to isolate the bacteria that were present. The isolates were examined using oil immersion lens (x 100) to analyse their colonial morphology. The plates were incubated in sets of three under aerobic and anaerobic conditions at a temperature of 37° for a duration of 24 hours. Subsequently, individual colonies were transferred to new growth medium to cultivate pure isolates, which were then preserved on nutrient Agar slants for conducting biochemical tests such as Gram's stain, oxidase, coagulase, catalase, indole, citrate, sugar fermentation, MR-VP, and motility. This experiment employed three various types of soup, including egusi, ogbono, and Abua local soup. The ingredients for the soups were obtained from Abua main market. The ingredients used in the different types of soup were thoroughly cleaned, and the soups were then cooked using specific local techniques and allowed to ferment for 48 hours. Afterward, the fermented soups were subjected to filtration using a membrane filter to remove any undesirable particles.

3.5. Testing some Soured Soups for Contamination with *Bacillus cereus*, *Clostridium botulinum*, and *Staphylococcus aureus* Toxins

Procurement of Test Kits: The lateral flow assay device (Duopath cereus Enterotoxin immunoassay) and Biothreat Alert test- strip were purchased from Merck, White House Station, New Jersey, USA, while Enzyme Immunoassay (EIA) Kit (Staphylococcal Enterotoxin A-E), were obtained from R-Biopharm GmbH, Darmstadt, Germany.

Detection of *Bacillus Cereus* Toxin in Soured Soups: The lateral flow assay device (Duopalh® cereus Enterotoxin Immunoassay) was used for the detection of *Bacillus cereus* toxin. Ten milliliters (10 ml) of sample buffer (provided) were mixed with the soured soup samples and homogenized and centrifuged at 7000 rpm for 30 minutes. The supernatant was passed through 3.45µm membrane filter. Each filtered sample was supplemented with 150 microliters (150µl) and introduced into the immunoassay port, adhering to the guidelines provided by the manufacturer. A positive result was shown by the presence of a red line after a 20-minute incubation period at room temperature. Tests were considered valid only when control lines were visible. Two red lines indicate positive while one red line indicates negative results.

Detection of *Clostridium Botulinum* Toxin: Biothreat Alert test strip for the detection of BONT/A/B was used. Fifty milliliters (50 ml) of soured soup sample was mixed with 10 ml of sample buffer homogenized with a bench top stomacher (Seward, Cincinnati, OH) to make a homogenous suspension. The food-buffer mixture was centrifuged at 7,000 x g for 30 mm. at 4°C to remove solid particles, and then filtered through a membrane filter. Five hundred microliters (500µl) of soured soup sample supernatant were thoroughly mixed with 500µl of sample buffer in a glass test tube and used for the experiment. Each test device was removed from a protective pouch and placed on a flat surface. 150µl of the sample was placed into the round sample sort according to the manufacturer's instructions and results were recorded visually after 15 minutes. The appearance of red lines indicates positive (one of the red lines is the control line) while only one red line indicates negative results.

Detection of *Staphylococcus Aureus* Toxin: Enzyme Immunoassay (EIA) Kit which utilizes five monovalent capture antibodies against SEA of SEE (Staphylococcal Enterotoxin A-E), was used. The soured soup sample was diluted with phosphate-buffered saline and adjusted to pH 7.4. The buffered sample was transferred to reaction tubes and sealed with Silicon stoppers to avoid enclosure of air. The enterotoxin

assay will be performed by the methods recommended by the manufacturer of the kit and results will be recorded visually after 15 minutes. One hundred and fifty microliters (150µl) of sample were added into each of the immunoassay wells and allowed to stand for 15 minutes after which results is read as positive or negative. A positive result appears yellow which is indicative of *S. aureus* toxin while a negative result appears light blue colour.

3.6. Detoxifying Effect of Tetrapleura Tetraptera Fruit Extract on Bacillus Cereus Toxin in Soured Soups

Five milliliters (5 ml) of Tetrapleura tetraptera fruit extracts were added into each soured soup (10 ml) sample and the mixture was transferred into a pot placed on fire and heated to a temperature of 100° C. After cooling, 150µl of the sample was placed into the round sample sort (lateral flow assay device) according to the manufacturer's instructions and results were recorded visually after 15 minutes. The appearance of red lines indicates positive (one of the red lines is the control line) while only one red line indicates negative results.

3.7. Detoxifying Effect of Tetrapleura Tetraptera Fruit Extract on Clostridium Botulinum Toxin in Soured Soups

Five milliliters (5 ml) of Tetrapleura tetraptera fruit extracts were added into each soured soup (10 ml) sample and the mixture was transferred into a pot placed on fire and heated to a temperature of 100° C. After cooling, 150µl of the sample was placed into the round sample sort (Biothreat Alert test strip) according to the manufacturer's instructions and results were recorded visually after 15 minutes. The appearance of red lines indicates positive (one of the red lines is the control line) while only one red line indicates negative results.

3.8. Detoxifying Effect of Tetrapleura Tetraptera Fruit Extract on Staphylococcus Aureus Toxin in Soured Soups

Five milliliters (5 ml) of Tetrapleura tetraptera fruit extracts were added into each soured soup (10 ml) sample and the mixture was transferred into a pot placed on fire and heated to a temperature of 100° C. After cooling, 150µl of the sample was placed into the round sample sort (Enzyme Immunoassay (EIA) Kit) according to the manufacturer's instructions and results were recorded visually after 15 minutes. The appearance of red lines indicates positive (one of the red lines is the control line) while only one red line indicates negative results.

4. RESULTS AND DISCUSSION

Table 1. Phytochemical Composition of Tetrapleura Tetraptera Fruit Extract

Phytochemical Constituent	Method of Analysis	Relative Presence	G/Kg
Alkaloid	Alkaline Reagent Test	+++	0.596± 0.01
Cardiac glycoside	Picrate Colorimetric Test	+	0.007± 0.00
Flavonoid	Mayer's Test	+	0.073±0.00
Phenol	Ferric Chloride Test	++	0.373±0.02
Saponin	Foam Test	+++	5.670±0.15
Steroid	Liebermann Burchard's Test	+++	4.102±0.13
Tannin	Ferric Chloride Test	++	0.366±0.02
Terpenoid	Salkowski's Test	+++	3.194±0.03

+: present in low concentration, ++: present in moderately high concentration, and +++: present in very high concentration.

Table 2. Isolation of Bacteria in Soured Soups

Soup Samples	Associated Bacteria
Egusi soup	Lactobacillus sp, Bifidobacterium sp, Streptococcus sp, Pediococcus, sp Leuconostoc sp Bacillus cereus, Clostridium perfringens, Campylobacter jejuni, Vibrio parahaemolyticus
Ogbono soup	Lactobacillus sp, Bifidobacterium sp, Streptococcus sp, Pediococcus, sp Leuconostoc sp, Yersinia enterocolitica, Salmonella sp, Shigella sp.
Native soup	Lactobacillus sp, Bifidobacterium sp, Streptococcus sp, Pediococcus, sp Leuconostoc sp, Escherichia coli, Vibrio parahaemolyticus, Staphylococcus aureus, Campylobacter jejuni, Listeria monocytogenes.

Table 3. Bacterial Toxins Detected in Soured Soups

Soured Soup	Bacteria Toxin Detected
Egusi Soup	Bacillus cereus toxin
Ogbono Soup	None
Native Soup	Staphylococcus aureus enterotoxin

Absence of toxin; -

Presence of toxin; +

Table 4. Detoxification of Bacteria Toxins Treated with Tetrapleura Tetraptera Extract

Soured Soup	Bacteria Toxin Treated with T. Tetraptera Extract	Observation
Egusi Soup	Bacillus cereus toxin	Bacillus cereus toxin detected
Native Soup	Staphylococcus aureus enterotoxin	Staphylococcus aureus enterotoxin detected
Ogbono Soup		None of the toxins were detected including Clostridium botulinum toxin

Table 1 indicated that alkaloids, cardiac glycoside, flavonoid, phenol, saponin, steroid, tannin, and terpenoid were present; the quantity of alkaloids, saponin, steroid, and terpenoid was present in very high concentrations; phenol and tannin were present in moderately high concentrations; and cardiac glycoside and flavonoid were present in trace concentrations. The qualitative phytochemical examination of *T. tetrapleura* aligns with the findings of, who investigated flavonoids, tannins, and alkaloids and observed significant quantities of these chemical components in the examined plants. Also, it agrees with that of [16], who revealed “the presence of flavonoids, phenols, tannins, saponins, terpenoids, phlebotannin, D-fructose, piperazine, octodrine, glycidol, glyceraldehydes, 6-octadecenoic acid, and 9,12-octadecenoic acid.

Table 2 showed that *Lactobacillus* sp, *Bifidobacterium* sp, *Streptococcus* sp, *Pediococcus* sp, *Leuconostoc* sp *Bacillus cereus*, *Clostridium perfringens*, *Campylobacter jejuni*, *Vibrio parahaemolyticus*, *Yersinia enterocolitica*, *Salmonella* sp, *Shigella* sp, *Escherichia coli*, *Staphylococcus aureus*, and *Listeria monocytogenes* were associated with the soured soups. This agrees with the work of [7] 2009, who reported that “these organisms are major food spoilage microorganisms. The study also agrees with the findings of [12], [13] whose works isolated these organisms from various cooked food items”.

Table 3 shows that *Bacillus cereus* toxin was detected in the soured egusi soup; *Staphylococcus aureus* enterotoxin was also observed in the native soup; and none was detected in ogbono soup. This is in line with the findings of [3], [10], [9], [11], which noted “the presence of *B. cereus* toxins in rice dishes. This means that *B. cereus* toxin is mostly found in rice, rice products, grains, oriental dishes, spice meats, vegetable dishes, soups, and sauces. Moreover, detecting *B. cereus* toxins in soured egusi soup may be due to the presence of its heat-resistant spores from the ingredients (melon seed or vegetable) used in the preparation of the soup, which, when the soup cooled down, germinated, multiplied, and produced toxin. Inadequate temperature management occasionally plays a part in *B. cereus* food poisoning; the bacteria

multiply in food that has been precooked and then not heated up enough or else not adequately cooled down beforehand. Also, *B. cereus* generates spores that can survive high heat, and are still capable of generating viable cells at low temperatures. These spores then often secrete toxins that are heat-stable.

Table 4 indicates the ineffectiveness of the *Tetrapleura tetraptera* extract in detoxifying the bacterial toxins present in the soured soup samples. This is supported by the findings of Pat-Harkins (2015) who noted that “heating soured soup does not denature the toxins in it. Also, it agrees with the findings of Stewart (2003) who noted that Staphylococcal Enterotoxins (SEs) are heat-stable and resist high temperatures and environmental conditions of drying and freezing, and that usual cooking procedures, pasteurization, and drying do not inactivate these toxins. It also ties in with the findings of Odike and Ogbonda (2019) [6] who indicated that red oil and onion extract could not detoxify the presence of bacteria and toxins in the soured soups. However, the findings of [10] showed that diarrhoeal toxin of *B. cereus* can be inactivated by heating with other treatments for 5 minutes at 56°C”.

5. CONCLUSION

The study indicated that alkaloids, cardiac glycoside, flavonoid, phenol, saponin, steroid, tannin, and terpenoid were present; the quantity of alkaloid, saponin, steroid, and terpenoid was present in very high concentrations; phenol and tannin were present in moderately high concentrations; and cardiac glycoside and flavonoid were present in trace concentrations. *Tetrapleura tetraptera* extracts did not denature the toxins produced by *S. aureus* and *B. cereus*, implying that *Tetrapleura tetraptera* extracts did not have any effect on the stability of the toxins produced by the bacteria in the soured soups. It is therefore unsafe to consume soured soups, even when treated with the extracts of *Tetrapleura tetraptera*. The use of the extracts of *Tetrapleura tetraptera* in the treatment of soured soups as a way of repairing the soup for consumption is a myth.

5.1. Recommendations

Based on the findings of this study, the researcher hereby submits the following recommendations:

1. Spoiled soups of any kind should not be consumed.
2. Local communities should discontinue the use of the extract of aidan fruit to treat soured soups as a means of restoration for consumption since the bacterial toxins could not be denatured by the extract.

Acknowledgments

The authors have no specific acknowledgments to make for this research.

Funding Information

This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

Author Contributions Statement

Name of Author	C	M	So	Va	Fo	I	R	D	O	E	Vi	Su	P	Fu
Saturday Okpabi	✓	✓	✓	✓		✓		✓	✓	✓	✓			
Uche Joy Nzube		✓	✓		✓	✓			✓	✓			✓	✓
Robert Chioma Lilia	✓	✓		✓			✓	✓			✓	✓		
Prof. K. H. Ogbonda		✓	✓		✓	✓			✓	✓		✓		✓

C : Conceptualization

M : Methodology

So : Software

Va : Validation

I : Investigation

R : Resources

D : Data Curation

O : Writing - Original Draft

Vi : Visualization

Su : Supervision

P : Project

administration

Fo : **F**ormal analysis

E : **W**riting - Review & **E**dit

Fu : **F**unding acquisition

Conflict of Interest Statement

The authors declare that there are no conflicts of interest regarding the publication of this paper.

Informed Consent

All participants were informed about the purpose of the study, and their voluntary consent was obtained prior to data collection.

Ethical Approval

The study was conducted in compliance with the ethical principles outlined in the Declaration of Helsinki and approved by the relevant institutional authorities.

Data Availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

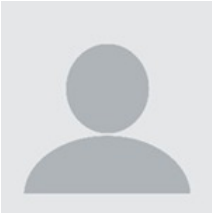
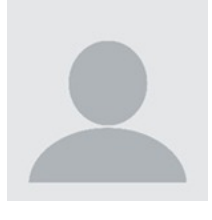
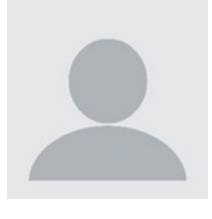

REFERENCES

- [1] Anderson, C. D., & Williams, E. F. (2021). Bacterial toxins and their mechanisms of action. Springer, 87-110.
- [2] L. M. Brown and R. S. Davis, 'Role of bacterial toxins in host immune response', in Proceedings of the International Conference on Microbiology, 2023, pp. 42-47.
- [3] S. J. Oomes, A. C. Van, J. O. Hehenkamp, H. Witsenboe, J. M. Van, and S. Brul, 'The characterization of Bacillus spores occurring in the manufacturing of (low acid) canned products', International Journal of Food Microbiology, vol. 120, pp. 85-94, 2007. doi.org/10.1016/j.ijfoodmicro.2007.06.013
- [4] A. C. Vanzuiglan and S. J. Oomes, 'Detecting bacterial spores in soup manufacturing', International Journal of Food Microbiology, vol. 8, no. 7, pp. 37-39, 2009.
- [5] E. A. Szabo and A. M. Gibson, 'Foodborne microorganisms of public health significance. Australian Institute of Food Science and Technology', pp. 917-922, 2003.
- [6] N. Agata, M. Ohta, and K. Yokoma, 'Production of Bacillus cereus emetic toxin (cereulide) in various foods', International Journal of Food Microbiology, vol. 73, pp. 23-27, 2002. [doi.org/10.1016/S0168-1605\(01\)00692-4](https://doi.org/10.1016/S0168-1605(01)00692-4)
- [7] A. M. Richard, C. Pamelo, and O. Champe, 'Lippincott Illustrated Reviews', pp. 98-102, 2007.
- [8] S. Luebke, R. W. & Koepsell, S. A. (2000). Detection of bacterial toxins by cell culture assay
- [9] O. Odike and K. H. Ogbonda, 'Bacteria toxins in sour soup', Reiko International Journal of Science and Technology Research, vol. 9, no. 2, pp. 69-79, 2019.
- [10] O. Odike, 'Clostridium botulinum Toxin Inactivation in Selected Soured Soups in South-South Region of Nigeria', International Journal of Microbiology and Applied Sciences, vol. 2, pp. 76-82, 2023.
- [11] Salminen, S., Wright, A. & Ouwehand, A. C. (2014). Lactic acid bacteria: microbiological and functional aspects. CRC press, 116-213.
- [12] S. R. Tatini, 'Thermal stability of enterotoxins in food', Journal of Milk Food Technology, vol. 39, no. 6, pp. 432-438, 2011. doi.org/10.4315/0022-2747-39.6.432
- [13] O. C. Eruteya, F. S. Ire, and C. C. Aneke, 'Inhibitory potential of Ocimum gratissimum L. on bacterial implicated in "Ofe Akwu" soup spoilage', International Journal of Microbiology Research, vol. 19, no. 3, pp. 1-8, 2017. doi.org/10.9734/MRJI/2017/32554
- [14] J. D. Smith and A. B. Johnson, 'Molecular mechanisms of bacterial toxin production', Journal of Microbiology, vol. 45, no. 3, pp. 167-182, 2022.

- [15] K. R. Khandelwal, 'Preliminary photochemical screening in Practical Pharmacognosy Techniques and Experiments', Nirali Publication, vol. 3, pp. 149-156, 2001.
- [16] D. A. Akinpelu and T. M. Onakoya, 'Antimicrobial Activities of Medicinal Plants used in Folklore Remedies in South-Western', African Journal, vol. 5, pp. 1078-1081, 2006.

How to Cite Saturday Okpabi, Uche Joy Nzube, Robert Chioma Lilia, Prof. K. H. Ogbonda. (2024). Detoxification of bacterial toxins in soured soups using the extract of aidan tree (*tetrapleura tetraptera* l.) fruit. International Journal of Agriculture and Animal Production (IJAAP), 4(1), 88-97. <https://doi.org/10.55529/ijaap.42.35.45>

BIOGRAPHIES OF AUTHORS

	<p>Saturday Okpabi, is an academic affiliated with the Biology Department, Faculty of Natural and Applied Sciences at Ignatius Ajuru University of Education, Rumuolumeni, Port Harcourt, Rivers State, Nigeria. He is engaged in biological research and science education within one of Nigeria's prominent universities of education. His work contributes to the advancement of biological sciences in the Niger Delta region, supporting the development of qualified science educators and researchers in Rivers State and the broader Nigerian academic community. Email: okpabis@gmail.com</p>
	<p>Uche Joy Nzube, is a researcher and academic in the Biology Department, Faculty of Natural and Applied Sciences at Ignatius Ajuru University of Education, Rumuolumeni, Port Harcourt, Rivers State, Nigeria. She is dedicated to advancing biological sciences research and education in the Niger Delta region. Her academic contributions support the training and development of science educators in Nigeria. Based in Rivers State, she plays an active role in promoting quality science education and research within the Nigerian university system. Email: spectabilisjoy70@gmail.com</p>
	<p>Robert Chioma Lilia, is a dedicated academic and researcher in the Biology Department, Faculty of Natural and Applied Sciences at Ignatius Ajuru University of Education, Rumuolumeni, Port Harcourt, Rivers State, Nigeria. With a passion for biological sciences, she contributes meaningfully to research and science education in the Niger Delta region. Her scholarly pursuits focus on fostering academic excellence and nurturing the next generation of science educators in Rivers State, making a lasting impact on the growth of natural sciences within the Nigerian educational landscape. Email: liliachioma@gmail.com</p>
	<p>Prof. K. H. Ogbonda, is a distinguished Professor in the Biology Department, Faculty of Natural and Applied Sciences at Ignatius Ajuru University of Education, Rumuolumeni, Port Harcourt, Rivers State, Nigeria. A highly respected academic and seasoned researcher, he has made significant contributions to the advancement of biological sciences in Nigeria. With decades of scholarly experience, Prof. Ogbonda has mentored numerous researchers and science educators, shaping the academic landscape of the Niger Delta region. His expertise and leadership continue to inspire excellence in biological research and science education across Rivers State and beyond. Email: Kemkahumphery.ogbonda@iaue.edu.ng</p>