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Prevalence of *Salmonella Enterica* Subspecies *Enterica* Serovar Typhi in Hospital Landfill: An Environmental Cross Sectional Study

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ABSTRACT

Background: Research indicates that *Salmonella enterica* subspecies *enterica* serovar Typhi (ST), the causative agent of enteric fever, is evolving with the emergence of new strains that require distinct treatment approaches and exhibit varying clinical manifestations. This study focuses on investigating the prevalence of *Salmonella enterica* subspecies *enterica* serovar Typhi from hospital dumping sites.

Methods: A total of 207 samples were collected from different locations (A, B and C) of hospital landfill in Awka, Anambra State, and screened for ST using standard microbiological techniques. The prevalence of ST was determined through a cross-sectional study design.

Results: The study revealed that 61.84% of the 207 samples analyzed were positive for ST. Notably, samples from location A had the highest occurrence of ST, accounting for 79.81% of the positive samples, which was statistically significant ($p < 0.05$). Six ST strains were identified: STCMCST, STCT18, STWGS1146, ST311189, STERL12960, and ST2018K. Among these, STCMCST was the most prevalent, occurring significantly more frequently ($p < 0.05$) across all sample types, from the three locations. The frequency distribution of the ST strains was as follows: STCMCST (65.62%), STCT18 (23.44%), STWGS1146 (5.47%), ST311189 (3.13%), STERL12960 (1.56%), and ST2018K (0.78%).

Conclusion: This study has confirmed the presence of various ST strains in the samples, with STCMCST being the most prevalent. To mitigate the risk of ST infections, proper hygiene practices and safe disposal of hospital waste are essential preventive measures.

Keywords: Prevalence, Strains, Enteric, Manifestation, Typhi



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INTRODUCTION

Salmonella is a genus of rod-shaped, gram-negative bacteria that are facultative anaerobes belonging to the family Enterobacteriaceae.¹ The genus comprises two main species: *Salmonella enterica* and *Salmonella bongori*. With over 2600 serovars described globally, *Salmonella enterica* is a diverse group of bacteria that can cause various illnesses in humans and animals.² One of the most significant serovars is *S. enterica* serovar Typhi, which is commonly found in contaminated water, food products, soil samples, and waste deposits.³ As a zoonotic bacterium, *S. enterica* serovar Typhi has spread globally, exhibiting varying host specificity and infectivity.⁴ The serovar is a primary cause of gastrointestinal disorders, food-borne illnesses, and severe systemic illnesses, including typhoid and paratyphoid fever.⁵

Although *S. enterica* serovar Typhi is also present in fresh and marine surface waters, excessive contamination of these environments can originate from a variety of sources, including discharges from wastewater treatment plants, urban and agricultural runoff pollution, and overworked septic systems.⁶ There is a significant chance of catching infections brought on by *S. enterica* serovar Typhi because of the organism's high retention in water bodies. Occasionally, organisms develop outside of their hosts, leading to inter-host survival.⁷

According to research, hospital landfills include a wide variety of *Salmonella* serovars.^{7,8} Infected hospital patients may release *S. enterica* serovar Typhi into the environment via their faeces and other bodily fluids. Typhi is often the cause of the majority of infectious infections linked to drinking tainted water.⁹ Additionally, there is a connection between hospital disposal locations and the prevalence of *S. enterica* serovar Typhi in water. Refuse tainted by sick patients is carried to various water bodies during water runoff, where the organism continues to grow.⁷

Although a number of researchers, like,⁷ have studied the incidence of *Salmonella* serovars in water samples, but there was no documented study on the prevalence of various strains of *S. enterica* subspecies *enterica* serovar Typhi in hospital landfills majorly in Awka, Anambra State. Therefore, the purpose of this study is to assess the frequency of various *S. enterica* subspecies *enterica* serovar Typhi strains in hospital landfills at different locations in Awka. The findings of this study will aid in the prevention and management of illnesses brought on by various *Salmonella* strains.

MATERIALS AND METHODS

Study Area: Awka is the capital city of Anambra State, located in the southeastern region of Nigeria. The city is situated approximately 25 kilometers east of Onitsha, a major commercial hub. Awka is bounded by neighboring towns and cities, including Nibo, Nise, and Amansea (plate 1). The city's geography is characterized by a tropical rainforest climate, with two distinct seasons: the rainy season (April to October) and the dry season (November to March). The temperature is typically high throughout the year, with average minimum and maximum temperatures ranging from 25°C to 32°C. Awka's residents are primarily engaged in farming, trading, and other commercial activities. The city's coordinates are approximately 6.217°N 7.067°E. With a rich cultural heritage and growing economy, Awka serves as an important urban center in Anambra State.

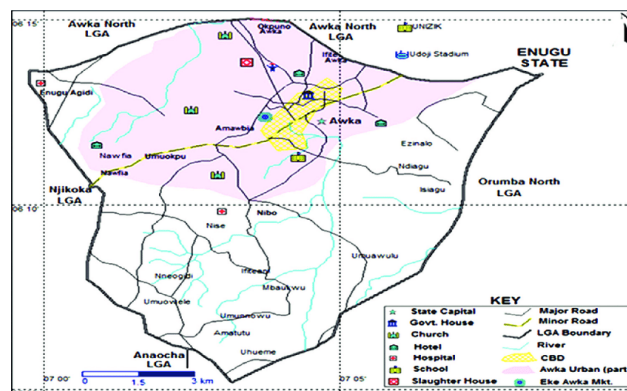


Plate 1: Map of Awka metropolis indicating the collection regions (hospitals)

Sample Size: The sample size was calculated using the formula below

$$N = \frac{Z^2 \cdot P \cdot (1-P)}{e^2}$$

P = The population size (16 %)

E = The margin of error (0.05)

Z = The Z-value extracted from the Z-Table (1.96)

$$n = \frac{1.96 \cdot 1.96 \cdot 0.16 \cdot 0.84}{0.05 \cdot 0.05} = 207$$

Sample collection, Handling and transportation: A total of 207 soil samples from hospital landfill were randomly collected from different sites in Awka, Anambra State. The litter from the soil surfaces was carefully scrapped out using sterile stainless spoon. The soil auger was derived to a plough depth of 15 cm in the farm land, and soil sample was drawn up to 10 samples from each sampling unit into a sterile tray. The samples

were thorough mixed and foreign materials such as roots, stones, pebbles and gravels were carefully removed. The soil sample was then reduced to half by quartering the sample. Quartering was carried out by dividing the soil sample into four equal parts and the two opposite quarters were discarded and the remaining two quarters were mixed. The process was repeated for the rest of soil samples used for this study. The samples were carefully labeled and then kept in a disinfected cooler, to maintain its temperature and stability of the number of the isolates. The samples were transported to the laboratory for analysis.¹⁰

Isolation of the Organism: This was carried out using the method of ¹¹. Five grams (5.0 g) of the soil sample was weighed using electronic weighing balance (DC-300), and the sample was aseptically dissolved in 15.0 ml of sterile normal saline in a 50 ml beaker (Pyrex), then make up the volume to 50.0 ml prior to serial dilution. One milliliter aliquot was aseptically transferred into a sterile test tube (PYREX) containing 9.0 ml of the diluent (sterile normal saline) and from this; ten-fold serial dilutions were made up to 10^{-3} , then 1.0 mL of the sample from last dilution was plated on Petri dishes (60 mm OD × 55 mm ID × 13mm high). This was followed by the addition of Deoxycholate citrate agar medium (DCA/Biotech) prepared according to the manufacturer's directions. All the plates in triplicates were incubated inverted at $37 \pm 2^\circ\text{C}$ for 24-48 h.¹¹

Characterization and Identification of the Isolates: The isolates were sub cultured on nutrient agar (Biotech), incubated in inverted position at 37°C for 24 h. The isolates were characterized and identified using their colonial and morphological descriptions ¹¹, biochemical reactions ^{9, 11} and molecular characterization ^{12, 13, 14}. The colonial description was carried out to determine the colours of the isolates on agar media plates, their sizes, edges, consistencies and optical properties of the isolates. The biochemical tests carried out were catalase, oxidase, citrate, indole, methyl red, voges Prokauer's, urease, hydrogen sulphide production, sugar fermentation, gelatine hydrolysis and lysine decarboxylase tests

Motility test: The motility test was performed using a semi-solid medium consisting of 5g of bacteriological agar and 2g of nutrient broth in 1L of distilled water. After sterilization by autoclaving, 10 ml portions were dispensed into test tubes and allowed to set. The test organisms were inoculated by stabbing the medium to half its depth with a sterile needle. The tubes were

incubated at 37°C for 24 hours in a vertical position, as described.¹¹ Motility was indicated by growth radiating from the stab line.

Hydrogen sulphide production test: Hydrogen sulphide production was tested using triple sugar iron (TSI) agar, following the method described by ¹¹. The TSI agar was prepared according to the manufacturer's instructions, sterilized by autoclaving, and cooled to 45°C . The isolate was then aseptically inoculated into the medium by vertical stabbing and streaked on the surface. After incubation at 37°C for 24-48 hours, the presence of hydrogen sulphide production was indicated by a darkened coloration of the medium. This color change is a positive indicator of H_2S production, allowing for the identification of isolates capable of producing this compound.

Urease test: The urease test was performed as described by ¹¹. Urease broth was prepared according to the manufacturer's instructions and sterilized. Isolates were aseptically inoculated into the medium and incubated at 37°C for 48 hours. A positive result was indicated by a pink or red coloration, signifying the presence of urease enzyme. This color change occurs when the organism's urease enzyme hydrolyzes urea, resulting in an alkaline pH shift.

Methyl red: The methyl red test was performed as described by ¹¹. Isolates were inoculated into glucose phosphate broth and incubated at 37°C for 48 hours. After adding 0.4% methyl red solution, results were read immediately. A bright red color indicated a positive result, while a yellow color indicated a negative result. This test detects stable acid production from glucose fermentation.

Voges-Proskauer test: The Voges-Proskauer test was performed as described by ¹¹. Isolates were inoculated into glucose phosphate broth and incubated at 37°C for 48 hours. After incubation, KOH and α -naphthol solutions were added. A positive reaction was indicated by the development of a pink color within five minutes, detecting acetoin production. This test identifies bacteria that ferment glucose and produce acetoin.

Citrate utilization: The citrate utilization test was performed as described by ¹¹. Isolates were stabbed into Simmon's Citrate Agar and incubated at 37°C for 48 hours. A positive result was indicated by growth and a blue color change, while a negative result showed no growth and the original green color was retained. This test detects the ability of bacteria to utilize citrate as a sole carbon source.

Catalase test: The catalase test was performed as described by ¹¹. A smear of the isolate was made on a slide, and a drop of 3% hydrogen peroxide (H₂O₂) was added. Prompt effervescence (bubbling) indicated catalase production, confirming the isolate's ability to break down H₂O₂ into water and oxygen. This test differentiates catalase-positive from catalase-negative bacteria.

Oxidase test: The oxidase test was performed as described by ¹¹. Two drops of freshly prepared oxidase reagent were placed on filter paper, and a smear of the isolate was applied. A blue-black coloration within 15 seconds indicated a positive result, detecting the presence of cytochrome c oxidase enzyme. This test identifies bacteria that produce this enzyme.

Sugar fermentation test: The sugar fermentation test was conducted as described by ¹¹. This test assessed the ability of isolates to metabolize various sugars (glucose, mannitol, mannose, maltose, sorbitol, inositol, and lactose), resulting in acid and/or gas production. Peptone water with bromocresol purple indicator and inverted Durham tubes were prepared and sterilized. Sugar solutions were added aseptically, and the medium was inoculated with isolates. After incubation at 37°C for 48 hours, cultures were examined for acid production (indicated by a color change from purple to yellow) and gas formation (presence of bubbles in Durham tubes). This test determines the metabolic capabilities of bacteria in utilizing specific sugars.

Indole test: The indole test was performed as described by ¹¹. Isolates were cultured in peptone water and incubated at 37°C for 48 hours. KOVAC's reagent was added to the broth, and a red layer forming on top indicated a positive result, signifying the presence of indole produced by bacterial hydrolysis of tryptophan. This detects the enzyme tryptophanase in bacteria. The test helps identify bacteria based on their ability to produce indole.

DNA Extraction and Purification: Bacterial and fungal strains were cultured on Nutrient Agar and Sabouraud Dextrose Agar, respectively. Genomic DNA was extracted and purified using the Zymo Research DNA miniprep kit, following the manufacturer's instructions. The quality of extracted DNA was assessed using a Nanodrop mass spectrophotometer.^{12, 13, 14}

DNA Amplification and Gel Electrophoresis: PCR amplification was performed using a Master cycler Nexus Gradient, with a reaction mixture containing primer, template DNA, water, and master mix. The PCR

program consisted of initial incubation at 94°C for 5 minutes, followed by 35 cycles of denaturation, annealing, and elongation, with a final extension period at 72°C for 10 minutes. Amplified products were electrophoresed in 1.0% agarose gel and documented using a gel documentation apparatus.^{12, 13, 14}

DNA Sequencing and Computational Analysis: The 16S rRNA amplified PCR products were sequenced using an ABI DNA sequencer. Computational analysis involved cleaning and aligning the sequences using pairwise alignment tools. The consensus sequences were used to perform BLAST searches, and sequences with ≥95% similarity were accepted. The maximum scores, total scores, and accession numbers of the isolates were also assessed.^{12, 13, 14}

Determination of Prevalence of the Isolates in the Studied Samples: The occurrences of different strains of the bacterial isolates associated with the water samples drawn from the rivers, boreholes and soil samples collected from different hospital landfills were counted and recorded according to the method described in the study published by ¹⁵. The number of the occurrences of the predominate pathogenic bacteria were counted, and their percentages of occurrences were appropriately calculated and recorded.

Statistical Analysis: The data generated were expressed in percentage and Tables. The significance of the prevalence was determined using Chi square) at 95% confidence level. Pairwise comparison was carried out in an excel sheet using student "t" test from IBM SSPS version 30.0 ¹⁵

RESULTS

A total of 207 samples were drawn from different locations of hospital landfills in Awka, and the study revealed that a total of 128 samples were positive to test bacterial isolates as shown in Table 1. The study also revealed that the samples drawn from location A recorded the highest (79.81%) occurrences of the test isolates whereas the samples from location B recorded the lowest (47.83%) occurrences of the test isolates. The study also showed that an association exist between the sample sources and the occurrences of the test isolates as the data obtained in the study were statistically significant ($p < 0.05$), as the p-value was 0.0017.

The test bacterial isolates (code 15, 16, 17, 18, 19 and 20) exhibited similar appearances on Deoxycholate citrate agar (DCA) and Nutrient Agar (NA) plates as they showed colourless and dark centered and greyish white

colours respectively (Table 2). They exhibited circular and entire colonies on DCA and NA with smooth surfaces. They varied in colony sizes and elevations. Isolates 15 and 19 showed low convex elevation whereas isolates 16, 17, 18 and 20 showed convex elevation. The isolates were Gram negative rods, arranged singly/pairs with peritrichous flagella. They were non-spore formers and non-capsulated.

The isolates showed positive reactions to catalase, hydrogen sulphide, methyl red, lysine decarboxylase, and nitrate reduction. They tested negative for oxidase, citrate, indole, urease, and Voges-Proskauer. The isolates were also unable to hydrolyze gelatin. The sugar utilization patterns of the isolates varied. Glucose, xylose, and maltose were utilized by the isolates. However, arabinose, lactose, and sucrose were not utilized. The isolates exhibited varying abilities to utilize trehalose, mannitol, inositol, dulcitol, sorbitol, salicin, and mucate. These results are presented in Tables 3 and 4. The isolates demonstrated distinct biochemical characteristics. The variation in sugar utilization patterns suggests diversity among the isolates.

The nucleic acid extracted from the bacterial isolates revealed that the nucleic acids were all DNA (1.80 - 1.90) as shown in table 5 and plate 2. The amplicons generated from the extracted DNA were electrophoresed to confirm the purity of the amplicons as shown in plate 1. *Salmonella enterica* subspecies *enterica* serovar Typhi strain CMST (STCMST), ST serovar Typhi strain WG-S1146 (STWG-S1146), ST serovar Typhi strain ERL12960 (STERL12960), ST serovar Typhi strain 311189 (ST311189), ST serovar Typhi strain CT18 (STCT18), and ST serovar strain 2018K (ST2018K)

The investigation displayed the isolates' prevalences on samples taken from hospital landfills at different locations in Awka, Anambra State (Table 7). In the examined samples, the isolates' occurrences were statistically non-significant ($p \geq 0.05$) as the p-value was 0.201. The analysis also revealed that the isolates were more prevalent in the samples from location A, followed by location B, and location C was the least. Additionally, the study revealed that STCMST was most prevalent among the isolates encountered from the studied samples, and statistically significance ($p < 0.05$) when compared to the occurrences of other isolates, as their p-values were 0.012, 0.003, 0.005, 0.033 and 0.002 respectively. ST2018K recorded the least occurrence in the samples. There were no STERL12960, ST311189, or

ST2018K detected in locations B and C. STWGS1146 was also not detected in location C.

Table 1: Sample sources and occurrences of the test isolates

Source	Number	Positive (%)	Negative (%)
Location A	69	53 (79.81)	16 (20.19)
Location B	69	33 (47.83)	36 (53.17)
Location C	69	42 (60.87)	27 (39.13)
Total	207	128 (61.84)	79 (38.16)

DF= (3-1)(2-1) = 2; $\chi^2 = 12.76$; p-value = 0.0017



Table 2: Morphological Characteristics of the Isolates

Parameter	Isolate 15	Isolate 16	Isolate 17	Isolate 18	Isolate 19	Isolate 20
Appearance on DCA	Colourless and dark centered	Colourless and dark centered	Colourless and dark centered	Colourless and dark centered	Colourless and dark centered	Colourless and dark centered
Appearance on NA	Greyish White	Greyish White	Greyish White	Greyish White	Greyish White	Greyish White
Size (mm)	2.40	2.60	2.20	2.80	2.30	2.60
Surface	Smooth	Smooth	Smooth	Smooth	Smooth	Smooth
Colony Shape	Circular	Circular	Circular	Circular	Circular	Circular
Elevation	Low Convex	Convex	Convex	Convex	Low Convex	Convex
Edge	Entire	Entire	Entire	Entire	Entire	Entire
Gram reaction	—	—	—	—	—	—
Shape of Cell	Rod	Rod	Rod	Rod	Rod	Rod
Cell Arrangement	Singly/Pairs	Singly/Pairs	Singly/Pairs	Singly/Pairs	Singly/Pairs	Singly/Pair s
Motility	+	+	+	+	+	+
Nature of Flagella	Peritrichous	Peritrichous	Peritrichous	Peritrichous	Peritrichous	Peritrichous
Spore Test	—	—	—	—	—	—
Capsule Test	—	—	—	—	—	—

Table 3: Biochemical Characteristics of the Isolates

Parameter	Isolate 15	Isolate 16	Isolate 17	Isolate 18	Isolate 19	Isolate 20
Catalase	+	+	+	+	+	+
Oxidase	—	—	—	—	—	—
Citrate	—	—	—	—	—	—
Indole	—	—	—	—	—	—
Urease	—	—	—	—	—	—
H₂S	+	+	+	+	+	+
MR	+	+	+	+	+	+
VP	—	—	—	—	—	—
Nitrate Reduction	+/--	+	+	+	+	+
Gelatin Hydrolysis	—	—	—	—	—	—
Lysine décor bioxylase test	+	+	+	+	+	+

Table 4: Sugar Utilizing Potential of the Isolates

Sugar	Isolate 15	Isolate 16	Isolate 17	Isolate 18	Isolate 19	Isolate 20
Glucose	+	+	+	+	+	+
Maltose	+	+	+	+	+	+
Arabinose	—	—	—	—	—	—
Lactose	—	—	—	—	—	—
Sucrose	—	—	—	—	—	—
Xylose	+	+	+	+	+	+
Trehalose	+/--	+	+/--	+	+	+
Mannitol	+/--	+	+	+/--	+	+

Sugar	Isolate 15	Isolate 16	Isolate 17	Isolate 18	Isolate 19	Isolate 20
Inositol	+	+	+	+	+/--	+
Dulcitol	—	—	—	+/--	—	—
Arabitol	—	—	—	—	—	—
Surbitol	+	+	+	+	+	+/--
Salicin	+/--	—	—	—	+/--	—
Mucate	—	—	+/--	—	—	—
Malonate	—	—	—	—	—	—

Table 5: Purity of Nucleic Acids of the Bacterial Isolates

Isolate	Conc. (ng/μl)	ABS ₂₆₀	ABS ₂₈₀	ABS ₂₆₀ ----- ABS ₂₈₀
15	101.50	3.0142	1.6382	1.84
16	110.50	3.2146	1.7283	1.86
17	104.20	3.0640	1.6743	1.83
18	117.30	3.4010	1.8187	1.87
19	109.10	3.2018	1.7307	1.85
20	107.50	3.1008	1.6852	1.84

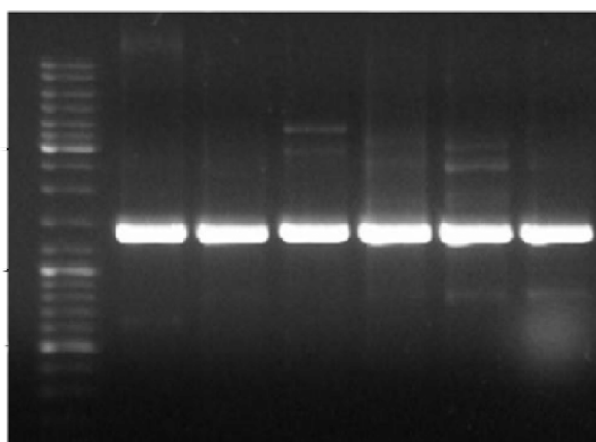


Plate 2: Gel Representation of the Amplicons

Table 6: Characteristics and Identities of the Isolates

Isolate Code	Max Score	Total Score	Query Cover (%)	E-value	Percent Identity	Accession Number	Description of the Isolates
15	3620	3620	100	0.0	100	CP053702.1	<i>Salmonella enterica</i> subspecies <i>enterica</i> serovar Typhi strain CMST (STCMST)
16	3620	3620	100	0.0	100	CP040575.1	<i>Salmonella enterica</i> subspecies <i>enterica</i> serovar Typhi strain WG-S1146 (STWG-S1146)



Isolate Code	Max Score	Total Score	Query Cover (%)	E-value	Percent Identity	Accession Number	Description of the Isolates
17	3620	3620	100	0.0	100	LT904894.1	<i>Salmonella enterica</i> subspecies <i>enterica</i> serovar Typhi strain ERL12960 (STERL12960)
18	6874	7660	100	0.0	100	CP029645.1	<i>Salmonella enterica</i> subspecies <i>enterica</i> serovar Typhi strain 311189 (ST311189)
19	6096	11283	100	0.0	100	AL513383.1	<i>Salmonella enterica</i> subspecies <i>enterica</i> serovar Typhi strain CT18 (STCT18)
20	4257	4257	100	0.0	100	CP044007.1	<i>Salmonella enterica</i> subspecies <i>enterica</i> serovar Typhi strain 2018K (ST2018K)

Table 7: Prevalence of the Isolates in the Sample

Isolate	Location A (%)	Location B (%)	Location C (%)	Total (%)
STCMCST	28 (21.88)	22(17.19)	34 (26.56)	84 (65.62)
STWGS1146	6 (4.69)	1 (0.78)	0 (0.00)	7 (5.47)
STERL12960	2 (1.56)	0 (0.00)	0 (0.00)	2 (1.56)
ST311189	4 (3.13)	0 (0.00)	0 (0.00)	4 (3.13)
STCT18	12 (9.38)	10 (7.81)	8 (6.25)	30 (23.44)
ST2018K	1 (0.78)	0 (0.00)	0 (0.00)	1 (0.78)
Total	53(41.41)	33 (25.78)	42 (32.81)	128 (100)

DF =10, $X^2 = 13.45$; p-value = 0.201. P-values of STCMCST vs STWGS1146, STERL12960, ST311189, STCT18 and ST2018K were 0.012, 0.003, 0.005, 0.033 and 0.002 respectively

DISCUSSION

The presence of *Salmonella enterica* serovar Typhi in the studied landfill samples is consistent with previous research findings^{2, 16, 17}. The cultural, morphological, biochemical and molecular characteristics of the *Salmonella* isolates align with existing literature^{16, 18, 19, 20, 21}. The occurrence of these strains in hospital landfills may be attributed to poor disposal practices and the ability of the organisms to survive in such environments. The study revealed that the isolates were most predominant in samples collected from location A, which could be due to the high population density and presence of surface water bodies that harbor anthropogenic activities. This finding corroborates previous studies^{8, 20, 22}. For instance, a study in Anambra State, Nigeria, reported the predominance of *Salmonella enterica* serovar Typhi in surface water samples²⁰. Another study suggested that *Salmonella enterica* Typhi can survive in aquatic environments by entering a viable but non-culturable state or residing within protozoa, potentially leading to a water-hospital-landfill

transmission chain.⁸ These findings highlight the importance of proper waste management and sanitation practices in preventing the spread of *Salmonella* infections. The presence of *Salmonella enterica* serovar Typhi in landfill samples underscores the need for further research into the environmental and public health implications of poor waste disposal practices.

The detection of various *Salmonella enterica* serovar Typhi strains, including STCMST, STWGS1146, STERL12960, ST311189, and STCT18, in landfill samples agrees with previous research findings^{8, 15, 20, 21} but varies in strains. The presence of these pathogens in landfill samples highlights the potential health risks to individuals exposed to these environments. As noted by previous studies, the presence of *Salmonella* species in these samples underscores the need for monitoring and control measures to minimize the risk of infection.¹⁷ These findings emphasize the importance of proper waste management and sanitation practices to prevent the spread of *Salmonella* infections.

The highest occurrences of STCMST in the landfill samples observed in the present study could be

attributed to the diversity of this strain within the study area. Studies have shown that genetic variation; genomic heterogeneity, antigenic variation, horizontal gene transfer and presence of dispensable (strain-specific) genes which confer fitness advantages to a particular strain, are associated with diverse epidemiological settings among different strains of microorganisms^{23, 24}. Also, the occurrence of STCMST mostly in the samples collected from location A, could be attributed to poor sanitation, indiscriminate sewage disposal, dumping of refuse, uncontrolled flooding and other anthropological activities within the study area. Similar conclusion was drawn^{8, 17, 21}

Many researchers^{25, 26, 27} have also reported that the survival of organisms, such as *Salmonella enterica* serovar Typhi, in hospital landfills can be attributed to several factors, including: Poor waste disposal practices, ability to survive in environment, presence of nutrients, moisture and humidity and lack of proper sanitation and hygiene. These factors can contribute to the persistence and transmission of pathogens, including *Salmonella enterica* serovar Typhi, in hospital landfills.

Contribution to Knowledge

This study identifies the predominant *Salmonella enterica* serovar Typhi strains in hospital landfills in Uli, Anambra State, Nigeria. The findings provide valuable insights into the epidemiology of typhoid fever in the region, serving as a guide for controlling and preventing diseases associated with these isolates. The study's results can inform public health strategies and interventions to mitigate the spread of typhoid fever in the community.

CONCLUSION

This study confirms the presence of diverse *Salmonella enterica* serovar Typhi strains in hospital landfills, including STCMCST, STWGS1146, STERL12960, STCT18, and ST2018K. Notably, STCMCST was the most predominant strain, suggesting its potential significance in the epidemiology of typhoid fever in the region. The detection of these strains in hospital landfills highlights the risk of transmission and spread of typhoid fever. The predominance of STCMCST warrants further investigation into its role in the region's typhoid epidemiology. Understanding the distribution and prevalence of these strains can inform public health strategies to control and prevent typhoid fever.

Conflict of Interest Declaration: Nil

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