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## Bacterial Profile of Livestock Farms in South-East Nigeria

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### ABSTRACT

The unregulated practice of livestock production has endangered the public health sector through the multiplication and spread of bacterial pathogens. This study investigated the bacterial profiles of livestock farms in Aba, Umuahia, Okigwe and Mbaise in the Southeastern part of Nigeria. Air was sampled with passive sedimentation technique; water samples were collected randomly from the farm water sources while hand swabs from the farmers and feeds were collected with sterile swab sticks and containers respectively. Total heterotrophic bacterial count (THBC) was analyzed by pour plate method; total coliform count (TCC) was determined by membrane filter technique while total potential pathogenic bacterial count (TPPBC) was examined by growing the samples in some selective agar media. Of the four cities studied, Aba had the highest THBC ( $28.43 \pm 0.3 \times 10^5$ ,  $26.70 \pm 0.7 \times 10^5$ ,  $26.26 \pm 0.5 \times 10^5$  CFU/ml), TPPBC ( $17.47 \pm 0.5 \times 10^5$  CFU/ml and  $20.02 \pm 0.5 \times 10^5$  CFU/ml) and TCC ( $24.06 \pm 0.4 \times 10^5$ ,  $17.93 \pm 0.6 \times 10^5$  and  $22.36 \pm 0.4 \times 10^5$  CFU/ml) for pig, cow and poultry farms respectively while Mbaise had the least value. A total of thirteen (13) bacterial species were isolated in the study but, only *Escherichia coli*, *Staphylococcus aureus*, *Klebsiella sp.*, *Salmonella sp.*, *Proteus mirabilis* and *Bacillus subtilis* were commonly distributed in the four cities. *Bacillus subtilis*, *Salmonella sp.* and *Staphylococcus aureus* were isolated more in Okigwe, Aba and Umuahia respectively than Mbaise. *Salmonella sp.* (60.00%) had the highest occurrence followed by *Staphylococcus aureus* (55.33%) while *Proteus mirabilis* (4.50%) had the lowest occurrence. High bacterial loads were obtained in the study especially in Aba. Livestock farmers should consider proper hygienic measures in order to limit the spread of pathogenic bacteria among surrounding communities.

### INTRODUCTION

Livestock farming is currently one of the leading agricultural practices in developing countries that is economically viable and performed both in urban and rural areas (Thornton, 2010). Urban livestock is a farming activity that is practiced within the urban centers. It is an important aspect that develops the urban areas. Apart from the economic impact on cities, urban livestock also produces negative effects for instance, increased health failure, environmental contamination and spread of diseases (Asadu *et al.*, 2021). Livestock farming contributes to climate change which consequently affects the distribution of bacteria and other unhealthy chemical substances (Grossi *et al.*, 2015). Livestock farms are agents of bacterial transmission and animal-related pathogens, especially the antibiotic-resistant strains. Livestock diseases are very significant worldwide based on their effects on the environment and inhabitants. These diseases produce direct effects on human and animal health and negatively affect the economy and food supply (Thornton, 2010; Gebreyes *et al.*, 2020).

Nigeria is geopolitically grouped into six zones including the South-East which is made up of Enugu, Anambra, Imo, Abia, and Ebonyi States. The occupation of the southeasterners is mainly trading, crop production and livestock farming (Nwanta *et al.*, 2011). The zone has so many urban towns with growing populations such as Aba, Umuahia, Okigwe and Mbaise. A lot of urban

agricultural activities take place in these towns especially rearing of sheep, goats and pig (Asadu *et al.*, 2021). They are managed in both intensive and semi-intensive systems. Livestock farming generate animal protein for consumers and revenue to the sellers (Nwanta *et al.*, 2011). Most livestock farmers largely venture into pig farming while 65% included poultry, and 31% indulge in goat and sheep production (Nwanta *et al.*, 2011). These animals are sources of direct and indirect disease spread. Microbial pathogens are transmitted via excreta, urine and flesh of animal hosts. They can also be isolated from feeds, drinking water, rain splashes, feeding troughs and hands of farmers (Alegbeleye *et al.*, 2018).

Microorganisms are microscopic living organisms that survive either in their natural environment or in the body of both animals and humans. They are an important part of atmospheric particulate matter, water bodies, soil and are closely associated with human health. The growth and spread of these microbes are dependent on the available nutrients, hosts and locations (Köhl *et al.*, 2019). Bacteria are the most highly bountiful microbes in livestock farms. Their ubiquity and survival mechanisms within and outside of the hosts make them most successful in disease transmission among other pathogens. They are distributed through mediums such as urine, faeces and hides of livestock and from their aerosols (Klous *et al.*, 2016). Several strains of pathogenic bacteria such as *Escherichia coli*, *Vibrio sp.*, *Shigella sp.* and *Salmonella sp.* and

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some non-pathogenic bacteria have been isolated from air, water and soil (Ugbogu *et al.*, 2016). Through human and animal activities, these bacteria are spread thereby resulting in infectious diseases (Ali *et al.*, 2021). This study is aimed at determining the bacterial species that are predominant in the livestock farms in four cities of the southeast.

## METHODOLOGY

### Study Area

The study was conducted in the four most populated cities in Nigeria's South-East region, namely Aba, Umuahia (Abia State) and Okigwe, Mbaise (Imo State). The selection of the cities was based on random sampling where livestock activities are predominant. The samples were collected from urban areas where livestock farming is vigorously practiced. These four cities stretch from latitude 4°50' to 7°20' N and longitude 6°51' to 8°20' E. It has common boundary with Benue State in the North, in the East it is bounded by Cross River and Akwa Ibom States, in the West by Delta State and River Niger (Kalu & Zakiora, 2019).

The zone has diverse ecological variations and land mass of 22,525 km<sup>2</sup> (Madu, 2006). Its annual rainfall is between March and October while the dry season starts from November and ends in February (Kalu & Zakiora, 2019). The study was carried out from December 2019 to April, 2022. A total of 600 samples (air, water, soil, feeds) were collected from pig, poultry and cow farms including hand swabs of their keepers. Feeds were obtained only from poultry and pig farms in Aba, Umuahia, Okigwe and Mbaise while air, water and soil samples were collected from pig, cow and poultry farms in the four cities of the two states using scientific standards. Hand swabs of the livestock workers were also collected with sterile swab sticks in all the farms.

### Air Quality Sampling

Passive air sampling was performed using settle plates. Freshly prepared nutrient agar (NA), blood agar (BA), Salmonella-Shigella agar (SSA), MacConkey (MCA) and Thiosulphate citrate bile salt sucrose (TCBS) plates were allowed to solidify and dry. The plates were exposed at the height of 1.5 m above the ground for 60 min at various locations in the poultry farm, cow ranches and pig farms. The samples were sealed, labeled appropriately, put inside sterile polythene bags, transported to the Laboratory, and incubated at 30°C for 24 h for bacterial growth. The experiment was repeated in triplicate and expressed as CFU/plate/hour.

### Water quality sampling

Water samples were collected in 2.5 litre plastic containers and transported to the laboratory for analysis (Ugbogu *et al.*, 2016). Ten-fold serial dilution of the water samples collected from Aba, Umuahia, Okigwe and Mbaise were performed according to method described by Harley and Prescott (2002).

### Soil Quality Sampling

Briefly, 2 kg of soil samples were collected in sterile polythene bags using soil auger at 30 cm depth (Bhat *et al.*, 2011). The soil samples were collected from different locations in the pig farm, poultry farm and cow farm. Soil samples were placed on ice in a cooler box immediately after collection and transported to the laboratory for analysis.

### Hand Swab Sampling

Hand swab samples were collected with sterile swab sticks from the hands of the livestock farmers and properly labeled. In the laboratory, 5 ml of normal saline was transferred into the swab sticks and allowed to stand for 10 min. Thereafter, ten-fold serial dilution ( $10^{-1}$ - $10^{-4}$ ) was performed with the solution, and appropriate dilution inoculated on agar plates and incubated (Sampson *et al.*, 2019).

### Heterotrophic Bacteria

After ten-fold serial dilution, 1 ml from  $10^{-4}$  was pipetted onto NA plates in triplicates. The discrete colonies in each NA plate were counted and recorded in CFU/ml for water, soil, hand swabs and feed samples and CFU/plate/hour for air sample. Plates with colonies between the ranges of 30-300 were counted. The heterotrophic bacterial count was recorded as total heterotrophic bacterial count (THBC).

### Total Coliform Bacterial Count

#### Water

TCC was performed using membrane filter technique according to method described by Harley and Prescott (2002) with slight modification. Briefly, after serial dilution, 100 ml of the water sample from  $10^{-4}$  dilution was transferred onto a membrane filter with pore size of 0.45 µm. After filtration, the absorbent paper was laid carefully on the MacConkey agar plate with sterile tweezers. The plates were incubated at 30°C for 24 h. After incubation, total coliform bacterial colonies were enumerated with the help of a magnifying glass.

#### Soil

Ten-fold serial dilution ( $10^{-1}$ - $10^{-7}$ ) was performed according to the method described by Adhikari *et al.* (2007) with slight modifications. Briefly, 10 g of soil was suspended into 90 ml of distilled water and mixture shaken properly. After serial dilution, 50 ml filtrate from  $10^{-4}$  was transferred onto a membrane filter (0.45 µm). The absorbent paper after filtration was transferred onto MacConkey agar plate and incubated for 24 h at 30°C. For confirmation, inoculums from the MacConkey agar plate were inoculated in tubes containing 10 ml of lactose bile broth. The mixture was incubated for 24 h at 30°C for fermentation to occur.

#### Hand Swab

A ten-fold serial dilution ( $10^{-1}$ -  $10^{-4}$ ) was conducted,

thereafter 1 ml from the  $10^{-4}$  dilution was inoculated onto MAC plate using spread plate technique for 24 h at  $37^{\circ}\text{C}$ .

### Total Potential Pathogenic Bacteria

TPPB was enumerated with selective media selected for potential pathogenic bacteria. SSA for *Salmonella* sp. and *Shigella* sp.; TCBS for *Vibrio cholera* and *Vibrio parahaemolyticus*; EMB for *Escherichia coli* and *Enterobacter aerogenes*; MSA for *Staphylococcus aureus*; Blood agar for *Streptococcus pyogenes* and MCA for *Pseudomonas aeruginosa*. For air samples, each plate was exposed to air for 60 min; aliquots from water, soil and feeds were seeded onto the media plates while hand swabs were inoculated onto the plates. After incubation for 24 h at  $30^{\circ}\text{C}$ , the bacterial species were identified based on their colony appearances (Lama *et al.*, 2013). The colonies on each plate were counted with magnifying glass.

### Feed

Total heterotrophic bacterial count, total potential pathogenic bacterial count and total coliform counts were performed according to the method described by Onyeagba (2015) and Adhikari *et al.* (2007) with slight modifications.

### Characterization and Identification of Bacterial Isolates

The bacterial isolates were characterized based on their colonial/cultural characteristics, macroscopic and microscopic appearances including elevation, margin, colour, size and surface texture and afterwards, biochemical reactions. The isolates were further confirmed by culturing them in selective media. These approaches were done according to Onyeagba (2015). The results were compared with standard reference of Bergey's Manual of Determinative Bacteriology (Buchanan and Gibbons, 1974).

### Isolation and Maintenance of Pure Culture

The representative bacterial colonies were sub-cultured on freshly prepared nutrient agar plates by streaking. The pure cultures were sub-cultured onto nutrient agar slants in bijoux bottles and incubated at  $30^{\circ}\text{C}$ . After 24h, the slants were kept in the refrigerator at  $-4^{\circ}\text{C}$  for storage until further processing.

### Statistical Analysis

The results were expressed as mean  $\pm$  SD using graph pad prism graphical statistical package version 5. The student t-test at  $p < 0.05$  was applied to assess the difference between the mean for variables in triplicates and two-way analysis of variance (ANOVA) for more than two variables followed by Bonferreni post hoc test.

## RESULTS AND DISCUSSION:

### Bacteria Profile of the Pig Farms in the Four Southeastern Cities

The total heterotrophic bacteria count (THBC) from

the pig farms varied between  $6.13 \pm 0.6 \times 10^5$  and  $28.43 \pm 0.3 \times 10^5$  CFU/ml while total potential pathogenic bacteria count (TPPBC) ranged from  $9.83 \pm 1.0 \times 10^5$  to  $26.23 \pm 0.4 \times 10^5$  CFU/ml. The value obtained for total coliform bacteria count (TCC) was between  $12.73 \pm 0.5 \times 10^5$  and  $24.06 \pm 0.4 \times 10^5$  CFU/ml. Of the four cities, Aba ( $28.43 \pm 0.3 \times 10^5$ ,  $24.06 \pm 0.4 \times 10^5$ ,  $26.23 \pm 0.4 \times 10^5$  CFU/ml) had the highest values. THBC (TCC) were higher in soil samples while hand swabs of workers had higher TPPBC than other samples. Air samples had the least counts of bacteria. The THBC, TPPBC and TCBC of samples obtained from air, water, soil, feeds and hand swabs of farmers in pig farm are shown in Table 1.

### Bacteria Profile of the Cow farms in the Four Southeastern Cities

THBC of air, soil and hand swabs from the cow ranged from  $10.50 \pm 0.6 \times 10^5$  to  $26.70 \pm 0.7 \times 10^5$  CFU/ml; TPPBC had values between  $9.26 \pm 0.5 \times 10^5$  and  $17.47 \pm 0.5 \times 10^5$  CFU/ml. The values of TCC were between  $9.03 \pm 0.6 \times 10^5$  and  $18.33 \pm 0.5 \times 10^5$  CFU/ml. In Aba and Mbaise, the highest value for THBC and TPPBC ( $26.70 \pm 0.7 \times 10^5$  and  $17.47 \pm 0.5 \times 10^5$  CFU/ml) and TCC ( $18.33 \pm 0.5 \times 10^5$  CFU/ml) respectively were obtained. Soil samples had the highest bacterial count (Table 2).

### Bacteria Profile of the Poultry Farm in the Four Southeastern Cities

From Table 3, the THBC varied between  $9.53 \pm 0.8 \times 10^5$  and  $26.26 \pm 0.5 \times 10^5$  CFU/ml; TPPBC was within the range of  $9.86 \pm 0.4 \times 10^5$  to  $20.20 \pm 0 \times 10^5$  CFU/ml while the values  $9.97 \pm 0.8 \times 10^5$  to  $22.36 \pm 0.4 \times 10^5$  CFU/ml were for TCC. The highest counts of THBC ( $26.26 \pm 0.5 \times 10^5$  CFU/ml) were obtained in Aba and Mbaise. TPPBC and TCC were more only in Aba ( $20.20 \pm 0.5 \times 10^5$  and  $22.36 \pm 0.4 \times 10^5$  CFU/ml) respectively. The hand swabs of workers produced the highest count for TPPBC while highest amount of THBC and TCC appeared more in soil samples. Result for the enumeration of bacteria in poultry farm is shown in Table 3.

### Distribution of Bacteria in Aba, Umuahia, Okigwe and Mbaise

Of the thirteen different bacterial strains isolated from the four cities, only six, namely *Escherichia coli*, *Staphylococcus aureus*, *Klebsiella* sp., *Salmonella* sp., *Proteus mirabilis* and *Bacillus subtilis* were commonly distributed among the four cities (Table 4)

### Percentage Bacterial Occurrence in Aba, Umuahia, Okigwe and Mbaise

*Salmonella* spp (60.00%) obtained from Aba were significantly higher ( $p < 0.01$ ) than those obtained from Umuahia (25.00%), Okigwe (18.33%) and Mbaise (32.33%). *Staphylococcus aureus* (55.33%) in Umuahia was appreciably higher ( $p < 0.05$ ) than others (50.00%, 15.67% and 8.33%) obtained. In Okigwe, *Bacillus subtilis* (38.33%)

obtained was significantly ( $p < 0.01$ ) higher than the other three cities 15.00% (Aba), 24.00% (Umuahia) and 19.00% (Mbaize). From the result, the livestock in Mbaize had the lowest percentage bacterial occurrence while Aba had the highest. *Proteus mirabilis* was the least isolated bacteria in all the cities while *Salmonella sp.*, *Klebsiella sp.* and *Escherichia*

*coli* were predominantly isolated. Result for the percentage bacterial occurrence of bacteria commonly distributed in Aba, Umuahia, Okigwe and Mbaize is presented in Figure 1.

Values with different numbers as superscripts within a row for the same parameter are significantly different

**Table 1:** THBC, TPPBC and TCC of air, water soil, hand swabs and feed samples of pig farm in Aba, Umuahia, Okigwe and Mbaize.

	Aba			Umuahia			Okigwe			Mbaize		
	THBC $\times 10^5$	TPPBC $\times 10^5$	TCC $\times 10^5$	THBC $\times 10^5$	TPPBC $\times 10^5$	TCC $\times 10^5$	THBC $\times 10^5$	TPPBC $\times 10^5$	TCC $\times 10^5$	THBC $\times 10^5$	TPPBC $\times 10^5$	TCC $\times 10^5$
Air (CFU/ plate/ hour)	18.96 $\pm$ 0.4 a <sup>1</sup>	21.33 $\pm$ 0.4 b <sup>1</sup>	ND	11.36 $\pm$ 0.4 a <sup>2</sup>	20.23 $\pm$ 0.4 b <sup>2</sup>	ND	12.20 $\pm$ 0.3 a <sup>3</sup>	17.50 $\pm$ 0.5 b <sup>3</sup>	ND	13.20 $\pm$ 0.3 a <sup>4</sup>	19.00 $\pm$ 0.3 b <sup>4</sup>	ND
Water (CFU/ ml)	20.33 $\pm$ 0.6 a <sup>1</sup>	12.73 $\pm$ 0.7 b <sup>1</sup>	22.16 $\pm$ 0.7 c <sup>1</sup>	12.30 $\pm$ 0.4 a <sup>2</sup>	11.66 $\pm$ 0.2 b <sup>2</sup>	16.86 $\pm$ 0.3 c <sup>2</sup>	13.23 $\pm$ 0.3 a <sup>3</sup>	20.06 $\pm$ 0.3 b <sup>3</sup>	17.80 $\pm$ 0.6 c <sup>3</sup>	13.96 $\pm$ 0.2 a <sup>4</sup>	12.30 $\pm$ 0.5 b <sup>4</sup>	21.00 $\pm$ 0.4 c <sup>4</sup>
Soil (CFU/ ml)	28.43 $\pm$ 0.3 a <sup>1</sup>	23.10 $\pm$ 0.3 b <sup>1</sup>	23.26 $\pm$ 0.4 c <sup>1</sup>	27.50 $\pm$ 0.5 a <sup>2</sup>	22.06 $\pm$ 0.2 b <sup>2</sup>	21.16 $\pm$ 0.4 c <sup>2</sup>	23.33 $\pm$ 0.3 a <sup>3</sup>	20.33 $\pm$ 0.5 b <sup>3</sup>	19.20 $\pm$ 0.4 c <sup>3</sup>	28.16 $\pm$ 0.5 a <sup>4</sup>	15.56 $\pm$ 0.6 b <sup>4</sup>	20.46 $\pm$ 0.4 c <sup>4</sup>
Hand swabs (CFU/ ml)	19.43 $\pm$ 0.5 a <sup>1</sup>	15.46 $\pm$ 0.6 b <sup>1</sup>	20.50 $\pm$ 0.7 c <sup>1</sup>	11.43 $\pm$ 0.4 a <sup>2</sup>	12.83 $\pm$ 0.7 b <sup>2</sup>	14.76 $\pm$ 0.6 c <sup>2</sup>	6.13 $\pm$ 0.6 a <sup>3</sup>	9.83 $\pm$ 1.0 b <sup>3</sup>	12.73 $\pm$ 0.5 c <sup>3</sup>	11.46 $\pm$ 0.5 a <sup>4</sup>	26.23 $\pm$ 0.4 b <sup>4</sup>	20.36 $\pm$ 0.6 c <sup>4</sup>
Feeds (CFU/ ml)	21.16 $\pm$ 0.4 a <sup>1</sup>	20.36 $\pm$ 1.3 b <sup>1</sup>	24.06 $\pm$ 0.4 c <sup>1</sup>	14.93 $\pm$ 0.3 a <sup>2</sup>	19.60 $\pm$ 0.7 b <sup>2</sup>	21.53 $\pm$ 0.5 c <sup>2</sup>	12.90 $\pm$ 0.3 a <sup>3</sup>	17.56 $\pm$ 0.5 b <sup>3</sup>	22.33 $\pm$ 0.5 c <sup>3</sup>	14.60 $\pm$ 0.5 a <sup>4</sup>	19.23 $\pm$ 0.3 b <sup>4</sup>	19.23 $\pm$ 0.5 c <sup>4</sup>

( $p < 0.05$ ;  $p < 0.01$ ;  $p < 0.001$ ;  $p < 0.0001$ ). Values with similar numbers as superscripts within a row for the same parameter are not significantly different ( $p > 0.05$ ). Each alphabet represents similar parameter within a column. Key: THBC-Total heterotrophic Bacteria Count; TPPBC-

Total Potential Pathogenic Bacteria Count; TCC-Total Coliform Count; ND-Not determined.

Values with different numbers as superscripts within a row for the same parameter are significantly different ( $p < 0.05$ ;  $p < 0.01$ ;  $p < 0.001$ ;  $p < 0.0001$ ). Values with

**Table 2:** THBC, TPPBC and TCC of air, water soil, hand swabs and feed samples of cow farm in Aba, Umuahia, Okigwe and Mbaize

	Aba			Umuahia			Okigwe			Mbaize		
	THBC $\times 10^5$	TPPBC $\times 10^5$	TCC $\times 10^5$	THBC $\times 10^5$	TPPBC $\times 10^5$	TCC $\times 10^5$	THBC $\times 10^5$	TPPBC $\times 10^5$	TCC $\times 10^5$	THBC $\times 10^5$	TPPBC $\times 10^5$	TCC $\times 10^5$
Air (CFU/ plate/ hour)	18.56 $\pm$ 0.3 a <sup>1</sup>	11.70 $\pm$ 0.4 b <sup>1</sup>	ND	10.50 $\pm$ 0.6 a <sup>2</sup>	10.33 $\pm$ 0.5 b <sup>2</sup>	ND	10.66 $\pm$ 0.5 a <sup>3</sup>	10.86 $\pm$ 0.3 b <sup>3</sup>	ND	11.46 $\pm$ 0.5 a <sup>4</sup>	10.66 $\pm$ 0.3 b <sup>4</sup>	ND
Water (CFU/ ml)	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Soil (CFU/ ml)	26.70 $\pm$ 0.7 a <sup>1</sup>	11.70 $\pm$ 0.4 b <sup>1</sup>	17.93 $\pm$ 0.6 c <sup>1</sup>	26.16 $\pm$ 0.5 a <sup>2</sup>	16.46 $\pm$ 0.4 b <sup>2</sup>	16.16 $\pm$ 0.3 c <sup>2</sup>	26.33 $\pm$ 0.2 a <sup>3</sup>	15.60 $\pm$ 0.3 b <sup>3</sup>	16.33 $\pm$ 0.5 c <sup>3</sup>	25.40 $\pm$ 0.4 a <sup>4</sup>	16.20 $\pm$ 0.3 b <sup>4</sup>	15.36 $\pm$ 0.4 c <sup>4</sup>
Hand swabs (CFU/ ml)	20.16 $\pm$ 0.5 a <sup>1</sup>	10.80 $\pm$ 0.3 b <sup>1</sup>	10.96 $\pm$ 0.2 c <sup>1</sup>	18.20 $\pm$ 0.4 a <sup>2</sup>	9.26 $\pm$ 0.5 b <sup>2</sup>	9.03 $\pm$ 0.6 c <sup>2</sup>	17.80 $\pm$ 0.9 a <sup>3</sup>	9.33 $\pm$ 0.5 b <sup>3</sup>	9.33 $\pm$ 0.3 c <sup>3</sup>	17.16 $\pm$ 0.3 a <sup>4</sup>	9.33 $\pm$ 0.4 b <sup>4</sup>	9.50 $\pm$ 0.4 c <sup>4</sup>
Feeds (CFU/ ml)	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND



similar numbers as superscripts within a row for the same parameter are not significantly different ( $p>0.05$ ). Each alphabet represents similar parameter within a column.

Key: THBC-Total heterotrophic Bacteria Count; TPPBC-Total Potential Pathogenic Bacteria Count; TCC-Total

Coliform Count; ND-Not determined.

Values with different numbers as superscripts within a row for the same parameter are significantly different ( $p<0.05$ ;  $p<0.01$ ;  $p<0.001$ ;  $p<0.0001$ ). Values with similar numbers as superscripts within a row for the same

**Table 3:** THBC, TPPBC and TCC of air, water soil, hand swabs and feed samples of poultry farm in Aba, Umuahia, Okigwe and Mbaise

		Aba			Umuahia			Okigwe			Mbaise		
		TPPBC ×10 <sup>5</sup>	TCC ×10 <sup>5</sup>	THBC ×10 <sup>5</sup>	TPPBC ×10 <sup>5</sup>	TCC ×10 <sup>5</sup>	THBC ×10 <sup>5</sup>	TPPBC ×10 <sup>5</sup>	TCC ×10 <sup>5</sup>	THBC ×10 <sup>5</sup>	TPPBC ×10 <sup>5</sup>	TCC ×10 <sup>5</sup>	
Air (CFU/ plate/ hour)	15.20± 0.4 a <sup>1</sup>	20.13± 0.5 b <sup>1</sup>	ND	9.53± 0.8 a <sup>2</sup>	18.23± 0.5 b <sup>2</sup>	ND	10.40± 0.6 a <sup>3</sup>	19.50± 0.5 b <sup>3</sup>	ND	10.26± 0.5 a <sup>3</sup>	16.30± 0.5 b <sup>4</sup>	ND	
Water (CFU/ ml)	20.70± 0.6 a <sup>1</sup>	10.33± 0.5 b <sup>1</sup>	21.26± 0.4 c <sup>1</sup>	11.53± 0.3 a <sup>2</sup>	11.90± 0.2 b <sup>2</sup>	17.00± 0.5 c <sup>2</sup>	12.36± 0.4 a <sup>3</sup>	9.86± 0.4 b <sup>1</sup>	16.80± 0.4 c <sup>3</sup>	13.56± 0.6 a <sup>4</sup>	12.03± 0.4 b <sup>2</sup>	19.03± 0.4 c <sup>4</sup>	
Soil (CFU/ ml)	26.26± 0.4 a <sup>1</sup>	13.23± 0.5 b <sup>1</sup>	22.36± 0.4 c <sup>1</sup>	22.36± 0.5a <sup>2</sup>	12.33± 0.4 b <sup>2</sup>	20.93± 0.5 c <sup>2</sup>	26.10± 0.4 a <sup>1</sup>	12.30± 0.4 b <sup>2</sup>	18.73± 0.4 c <sup>3</sup>	26.26± 0.5 a <sup>1</sup>	12.46± 0.5 b <sup>2</sup>	19.26± 0.4 c <sup>3</sup>	
Hand swabs (CFU/ ml)	19.06± 0.4 a <sup>1</sup>	20.20± 0.5 b <sup>1</sup>	15.00± 0.8 c <sup>1</sup>	11.76± 0.5 a <sup>2</sup>	13.13± 0.7 b <sup>2</sup>	13.26± 0.4 c <sup>2</sup>	12.83± 0.2 a <sup>3</sup>	14.26± 0.3 b <sup>3</sup>	9.97± 0.8 c <sup>3</sup>	12.66± 0.4 a <sup>3</sup>	14.26± 0.3 b <sup>3</sup>	15.36± 0.5 c <sup>1</sup>	
Feeds (CFU/ ml)	20.16± 0.6 a <sup>1</sup>	16.53± 0.3b <sup>1</sup>	12.43± 0.4 c <sup>1</sup>	14.33± 0.5a <sup>2</sup>	13.23± 0.4b <sup>2</sup>	11.50± 0.5 c <sup>1</sup>	12.76± 0.6 a <sup>3</sup>	12.40± 0.5 b <sup>3</sup>	10.23± 0.6 c <sup>2</sup>	13.36± 0.5 a <sup>4</sup>	13.83± 0.3 b <sup>2</sup>	11.20± 0.4 c <sup>1</sup>	

parameter are not significantly different ( $p>0.05$ ). Each alphabet represents similar parameter within a column.

Key: THBC-Total heterotrophic Bacteria Count; TPPBC-Total Potential Pathogenic Bacteria Count; TCC-Total Coliform Count; ND-Not determined.

## DISCUSSION

Livestock farming is a major contributor to small and medium scale enterprises and has been advocated for at both the state and federal levels. The spread of bacteria in livestock farms has been a major challenge in public

**Table 4:** Bacterial distribution in all the four cities

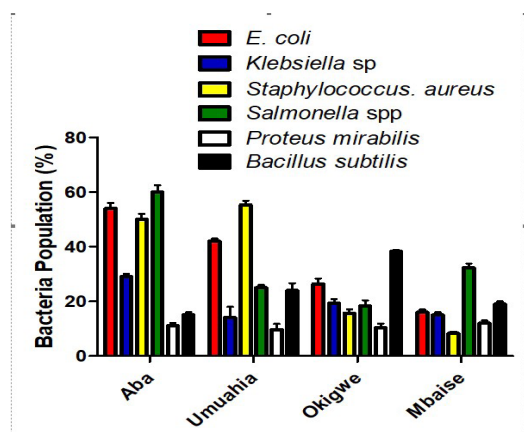
Isolates	Aba	Umuahia	Okigwe	Mbaise
<i>Escherichia coli</i>	+	+	+	+
<i>Klebsiella sp.</i>	+	+	+	+
<i>Staphylococcus aureus</i>	+	+	+	+
<i>Enterobacter sp.</i>	+	-	+	+
<i>Salmonella sp.</i>	+	+	+	+
<i>Proteus mirabilis</i>	+	+	+	+
<i>Pseudomonas sp.</i>	+	+	-	+
<i>Shigella sp</i>	+	-	+	-
<i>Vibrio cholera</i>	+	+	-	+
<i>Vibrio parahaemolyticus</i>	+	-	-	+
<i>Bacillus subtilis</i>	+	+	+	+
<i>Streptococcus pyogenes</i>	+	-	-	+
<i>Chromobacterium violaceum</i>	+	-	+	+

Key: + = present; - = absent

health sector particularly in the South-East region.

Total heterotrophic bacteria are the most multifaceted group of microorganisms with different nutritional and survival requirements. Some are primary colonizers while others are secondary invaders directly deriving their nutrients from the primary colonizers. The compositions of these microbes differ based on the location and nutrient available (Serves *et al.*, 1996). According to World Health Organization (WHO, 2018), the standard bacterial

count stipulated for drinking water is approximately 100 CFU/ml. Infectious diseases result when that quantity is exceeded. From this study, THBC was very significant in pig farm (Table 1) cow farm (Table 2) and poultry farm (Table 3) ( $28.43 \pm 0.5 \times 10^5$ ;  $26.70 \pm 0.7 \times 10^5$  and  $26.26 \pm 0.5 \times 10^5$  CFU/ml) respectively. The high values obtained could be attributed to water and soil pollution and improper disposal of waste generated from homes and industries (Ali *et al.*, 2021). These wastes serve as



**Figure 1:** Percentage occurrence of bacteria in Aba, Umuahia, Okigwe and Mbaise.

breeding sites for bacteria (Agodo *et al.*, 2016). During rainfall, splashes of water from these wastes percolate through underground water and affect the streams while the surrounding air become polluted.

THB, TC as well as the hand swabs of livestock keepers were assessed from Air, water, soil and livestock. THBC and TCC were appreciably higher in soil followed by water than air, feeds and hands of keepers. Soil and water support the multiplication of these microbes due to availability of nutrients. High values of THBC and TCC in soil samples is as a result of their ability to tap the available nutrients in soil sediments more than water (Adhikari *et al.*, 2007). According to Adhikari *et al.* (2007), the availability of nutrient and water retaining capacity of soil bacteria tend to increase their survival rate. Air particles do not have enough resources to sustain microbes; the chemical preservatives used in producing feeds inhibit the survival of bacteria (Soriano, 2020). The TPPBC were seen to increase among hand swabs ( $26.23 \pm 0.4 \times 10^5$  and  $20.20 \pm 0.5 \times 10^5$  CFU/ml) of pig and poultry keepers respectively while soil sample ( $17.47 \pm 0.5 \times 10^5$  CFU/ml) had the highest TPPBC for cow farm. These increased values could result from improper handling of the excreta, feeds and drinking water of the animals (McAllister and Toppt, 2012) and due to frequent close contact of livestock farmers with their livestock in our area (Klous *et al.*, 2016). Most pathogenic bacteria which colonize the skins and excreta of host animals can be transmitted as zoonotic pathogens to humans (Klous *et al.*, 2016).

Aba, from the results had the highest isolated THB, TC and TPPB. The increase in THBC, TCC and TPPBC in Aba can be attributed to the location of the pig and cow farms and the activities being performed. Aba River, where the research was conducted is a tributary of Imo River and all abattoir activities take place there. The river runs through two local governments in the State, and serve as sites for washing slaughtered animals and burning of hides (Ngozi and Humphrey, 2019). Of the sixteen bacteria isolated from the study (Table 4), only *Escherichia coli*, *Staphylococcus aureus*, *Klebsiella sp.* *Salmonella sp.*, *Proteus mirabilis* and *Bacillus subtilis* were distributed in

the four cities. The bacterial species isolated from the farms in the four cities were in agreement with Adogo *et al.* (2016).

Percentage occurrence of *Escherichia coli*, *Salmonella sp.*, *Klebsiella sp.*, *Staphylococcus aureus*, *Bacillus subtilis* and *Proteus mirabilis* is shown in Figure 1. *Salmonella sp.* have the highest occurrence at 60.00% while the least occurring bacteria was *Proteus mirabilis* (4.50%). *Bacillus subtilis* appeared more in Okigwe than in all the other cities. *Salmonella sp.* are Gram negative rods in the family of Enterobacteriaceae and more than 2,500 serovars have been identified. They are ubiquitous bacteria that survive in dry environments and water bodies for a long time (WHO, 2018). The strains that inhabit the colons of pig and cow according to WHO, are invasive and life threatening to humans and become major concerns to public health. From the study, *Salmonella species* have the highest percentage occurrence especially in Aba. Due to lack of potable drinking water and good roads to assess it, the dwellers depend on the Aba River. They drink, bathe, cook and wash clothes with the water without proper treatment. These activities lead to increase in spread of *Salmonella sp.* and *Escherichia coli* which subsequently cause waterborne diseases (Ali *et al.*, 2021). The ability to survive in low moisture environments contributes to their widespread. They have high osmo-tolerant membranes, filamentous cells and strong metabolic process (Finn *et al.*, 2013). In contrast, *Proteus mirabilis* recorded the lowest percentage occurrence. *Proteus mirabilis* is a motile bacterium that survives better in alkaline and urea-rich environments. It causes urinary tract infections, wound infections and kidney stones in humans (Zafar *et al.*, 2019). Their low percentage values in all the four cities can be attributed to the fact that *P. mirabilis* is a human pathogen that is rarely transmitted and isolated in livestock except when a person comes in contact with poultry flesh and droppings (Nahar *et al.*, 2014). The microbe has been reported to be transmitted through person-to-person contacts and food (Zafar *et al.*, 2019).

*Bacillus subtilis* is a spore forming bacterium that is air-borne. The formation of spores is a survival mechanism utilized by *B. subtilis* to thrive even in a challenging environment (Ravine, 2019). Only few literatures attributed *B. subtilis* to causing human diseases. They are reported to be beneficial in medicine as they can be used in producing probiotics, vaccines and enzymes (Piewngam and Otto, 2019; Sun *et al.*, 2018). From the study, Okigwe had the highest percentage of *B. subtilis*. This could result from the economic activities carried out in the city. Livestock farming and crop production are major occupation of Fulani occupants; deliberate disposal of animal dungs, farm waste and pesticides could increase the spread of spores of *B. subtilis* (Jorgensen *et al.*, 2015).

Comparing the percentage of occurrence of bacteria in the four cities, it can be seen that Aba had the highest bacterial loads compared with Mbaise that had the least percentage. This could be attributed to the type of economic activity predominant in these two areas

(Okoro and Ibe, 2017). Aba is a metropolitan city with large population of residents having diverse culture and business inclinations. It is the center for commercial activity in the South-east. As a center for Small and Medium Scale Enterprise (SME) in Nigeria (Agu *et al.*, 2019), small businesses that involve waste generation and degradation of ecosystem usually take place which lead to increase in the spread of bacteria and infectious diseases (Wizor, 2019).

## CONCLUSION

Livestock farming has remained one of the veritable means of improving the livelihood of the South-easterners. The challenge often experienced is the spread of pathogenic bacteria directly from the farm animals or the agents surrounding them such as air, water and soil. Of all the cities studied, Aba had the highest loads of bacteria. This calls for concerted effort by all stakeholders residing in the city and by extension the entire South-east as the disease resulting from the spread of these microbes from Aba could reach to other South-east States. Holistic hygienic practices should be promoted by livestock keepers as health workers should routinely conduct inspection on those farms and markets where these animals are reared and consumed respectively.

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