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Bacterial population of environmental surfaces in child-care centres in Omoku and Port Harcourt, Nigeria

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Abstract
The aim of this study was to enumerate the bacterial population isolated from environmental surfaces in child-care centres in Omoku and Port Harcourt, Nigeria. Four child-care centres in Omoku and Four from Port Harcourt, Nigeria were selected for the study and samples from different environmental surfaces (Table, play mat, toilet seat, feeding area, diaper changing area, toys, window and door handle surfaces) using sterile swab sticks moistened with sterile peptone water. Aliquots of 0.1 ml from the 10^3 dilutions were inoculated on agar plates of different culture media using the spread-plate method. The following culture media were used for the inoculation; Mannitol salt agar, Eosine methylene blue agar, MacConkey agar, *Salmonella-Shigella* agar, and Nutrient agar. The inoculated media were incubated at 35 °C for about 24 hours. An indirect cell count was performed in which the number of colonies produced on the various culture media plates were counted and recorded, and the information was used to calculate the amount of microorganisms in the original sample. The total heterotrophic bacterial counts of diapers changing surfaces in four schools located in Omoku ranged from 3.6×10^4 to 7.7×10^4 CFU/cm^2 while those obtained from schools in Port Harcourt ranged from 2.2×10^4 to 6.1×10^4 CFU/cm^2. The total coliform, faecal coliform, *Salmonella-Shigella* and staphylococcal counts of diapers changing surfaces in Omoku ranged 3.1×10^4 to 3.7×10^4, 2.8×10^2 to 3.9×10^2, 0.3×10^3 to 1.4×10^3, and 0.4×10^3 to 3.5×10^3 CFU/cm^2 respectively. While that of Port Harcourt ranged from 0.4×10^4 to 3.3×10^4, 0.8×10^2 to 5.0×10^2, 0.8×10^3 to 4.7×10^3 and 3.8×10^3 to 18.5×10^3 CFU/cm^2 respectively. The mean faecal coliform counts of door handle from schools in Omoku ranged from 0.9×10^3 to 4.8×10^3 CFU/cm^2, while those from schools in Port Harcourt ranged from 0.2×10^3 to 0.32×10^3 CFU/cm^2. The total heterotrophic bacterial load of the feeding areas of schools in Omoku ranged from from 0.7×10^4 to 1.6×10^4 CFU/cm^2 while the mean range of 8×10^4 to 1.3×10^4CFU/cm^2 was recorded for schools in Port Harcourt. The total heterotrophic bacterial count of play mats of schools located in Omoku ranged from 0.1×10^4 to 4.3×10^4 CFU/cm^2, while that of Port Harcourt ranged 0.3×10^4 to 0.40×10^4 CFU/cm^2. There were significant differences ($P < 0.05$) in the counts of the total heterotrophic bacteria for tables, *Salmonella-Shigella* counts for toilet seats and total staphylococcal counts for both toilet seats and toys. The faecal coliform bacteria of playmat and window surfaces showed no variance from both schools in Omoku and Port Harcourt. These results revealed that the high bacterial prevalence of these bacterial isolates in environmental surfaces of child-care facilities especially in faecal coliform and staphylococci is of public health importance especially as they could pose danger. Good hygiene including washing of hands and mopping of floors with disinfectants and clean water is highly recommended.

Keywords: Population, environmental surfaces, bacterial counts, total heterotrophic bacteria, total coliform, faecal coliform, *Salmonella-Shigella*

1. Introduction
There is a growing reliance of working parents on day-care facilities for childcare (Lee *et al*., 2007) [8]. Day-care setting is constantly exposed to different microorganisms from the kids, teachers, and parents. Kids are therefore at risk of contracting infectious diseases from day-care facilities. This raises concerns about the health and safety of toddlers attending day-care institutions. Families have become dependent on the child-care Industry for their growing children and most children spend most of their day in these day-care or school environments. A day-care centre is a facility where infants and children are kept while their parents are at work. Day-care workers look after the children's needs in terms of feeding, nurse care,

excretion, and general comforts (Sule & Awe, 2019) [12]. Child health care is the primary barometer that can be used to assess a country's social and economic well-being. Furthermore, the nutritional status of U5 children is regarded as a major indicator of a household's living standard, as well as a determinant of child survival (Olaniyan & Sunkanmi, 2012) [2].

Bacterial communities in day-care and schools are largely shaped by factors that influence bacterial dispersal, recruitment, and persistence. These include architectural design (e.g. ventilation), building condition, outdoor environmental factors such as climate, vegetation, and light, as well as various bacteria sources (e.g. humans) (Beasley *et al*., 2022) [1]. Children and teachers' behaviours can have an impact on these bacterial communities, both across and within classrooms. For example, eating areas and bathrooms have distinct bacterial communities due to how occupants contribute to the dispersal of different bacterial taxa in these spaces (i.e. through food transfer and waste processes) (Beasley *et al*., 2022) [1].

Children have also had less exposure to microorganisms, making them more likely to catch and transmit pathogens or opportunistic pathogens, as well as to suffer adverse effects from contact in densely populated facilities. Recent molecular studies of microbial diversity in indoor environments have revealed significant bacterial contamination and highlighted how little we know about such contamination (Lee *et al*., 2007) [8].

Due to the high concentration of young children and their continual interaction, day-care centers in Nigeria are the perfect setting for infectious diseases to proliferate. Young children may spread viruses that cause diarrhoea by sharing toys, using play mats or filthy surfaces, and putting pillows, books, and other objects in their mouths (Ledwaba *et al*., 2019) [7].

Contaminated environmental surfaces can be sources of indirect transmission but cleaning and disinfection are common interventions aimed at reducing contamination levels.

School children that live in regions where access to soap and water is difficult are particularly at risk of exposure to these microorganisms (Singh, 2010) [11]. Cases of children with these diseases have been infected with bacteria like *Escherichia coli*, *Salmonella*, *Shigella*, and *Campylobacter species* being the most prevalent types (Webb & Starr, 2005) [13].

2. Materials and Methodology

2.1 Samples collection

Four child-care centres in Omoku and Port Harcourt, Nigeria were selected for the study. Samples were collected between 8 a.m. and 10 a.m., just after hygiene procedures in the centres had been observed, and with approximately 80% of the children present. A measured portion of the surfaces (10 cm x 10 cm table surfaces, 10 cm x 10 cm play mats, 10 cm x 10 cm feeding area surfaces, 2cm x 2 cm toilet seat surfaces, 5 cm x 5cm diaper changing surfaces, 2 cm x 2 cm toys, 2 cm x 2 cm windows, and 2 cm x 2 cm door handles) was swabbed with a sterile swab sticks on each occasion. The swab sticks containing the samples were placed in a box containing ice cubes and promptly transported to the microbiology laboratory at Rivers State University for bacteriological analysis.

2.2 Isolation of bacteria

Swab sticks from each sampling site were separately placed and agitated in 10ml sterile peptone water (Sule & Awe, 2019) [12]. Inoculated peptone water were subjected to ten-fold serial dilution to obtain 10^{-3} dilutions. Aliquots of 0.1 ml from the 10^{-3} dilutions were inoculated on agar plates of different culture media using the spread-plate method. The following culture media were used for the inoculation; Mannitol salt agar, Eosine methylene blue agar, MacConkey agar, *Salmonella-Shigella* agar, and Nutrient agar. The inoculated media were incubated at 35 °C for about 24 hours. All the media were prepared according to the manufacturers' instruction and sterilised in an autoclave at 121ºC for 15 minutes at 15 PSI.

2.3 Microbial Count

After incubation, the ensuing colonies produced on the different culture media plates were counted. Counts on the different culture media were used to calculate the population of the corresponding bacterial groups as follows: counts on Mannitol salt agar for *Staphylococcus aureus*, counts on Eosine methylene blue agar for *Escherichia coli*, counts on MacConkey agar for coliforms, counts on *Salmonella-Shigella* agar for salmonella and shigella, counts on Nutrient agar for total heterotrophic bacteria.

2.4 Statistical Analysis

Numerical data were analyzed using the statistical package for social science (SPSS version 12). Descriptive data were summarized using frequencies and cross tabulations. Statistical significance of the data was determined using Analysis of Variance (ANOVA). Significance and mean separation of the data were determined using the T-test.

3. Results

The population of different bacterial groups in the diaper changing surfaces, door handles, feeding area surfaces, play mat surfaces, tables, toilet seat surfaces, toys and windows are presented in Tables 1 to 8, respectively.

3.1 Bacterial Population of Diaper Changing Surfaces

Results of the bacterial load of diaper changing surfaces in Omoku and Port Harcourt schools are presented in Table 1. The mean value of total heterotrophic bacterial counts of diaper changing surface samples from four schools in Omoku ranged from 3.6×10^4 to 7.7×10^4 CFU/cm^2, whereas those from schools in Port Harcourt ranged from to 2.2×10^4 to 6.1×10^4 CFU/cm^2. The total coliform, faecal coliform, *Salmonella-Shigella* and staphylococcal counts of diaper changing surfaces from Omoku ranged from 3.1×10^4 to 3.7×10^4, 2.8×10^2 to 3.9×10^2, 0.3×10^3 to 1.4×10^3, and 0.4×10^3 to 3.5×10^3 CFU/cm^2 respectively, while the ranges of total coliform, faecal coliform, *Salmonella-Shigella* and staphylococcal counts of diaper changing surfaces from four schools located in Port Harcourt ranged from 0.4×10^4 to 3.3×10^4, 0.8×10^2 to 5.0×10^2, 0.8×10^3 to 4.7×10^3 and 3.8×10^3 to 18.5×10^3 CFU/cm^2 respectively.

3.2 Bacterial Population of Door Handles

Results of the mean bacterial load of the door handle surfaces from the various schools of the different locations are presented in Table 2. The mean range of total heterotrophic bacterial load of door handles of schools located in Omoku ranged from 0.2×10^5 to 1.9×10^5

CFU/cm^{2,} while those from schools in Port Harcourt ranged from 1.9×10^5 to 2.2×10^5 CFU/cm^2. The total coliform, faecal coliform, *Salmonella-Shigella* and total *Staphylococcal* counts of door handles of schools in Omoku ranged from 0.2×10^5 to 1.5×10^5, 0.9×10^3 to 4.8×10^3, 0.2×10^4 to 1.2×10^4, and 3.6×10^4 to 5.0×10^4 CFU/cm^2 respectively, while the ranges of total coliform, faecal coliform, *Salmonella-Shigella* and total *Staphylococcal* counts of door handles of the four schools located in Port Harcourt ranged from 0.2×10^5 to 0.79×10^5, 0.2×10^3 to 0.32×10^3, 0.1×10^4 to 0.86×10^4 and 0.1×10^4 to 0.45×10^4 CFU/cm^2 respectively.

3.3 Bacterial Population of Feeding Area Surfaces

Results of the mean bacterial load of the feeding area surfaces of the four different schools in Omoku and Port Harcourt are presented in Table 3. The mean range of total heterotrophic bacterial load of the feeding area surfaces of schools in Omoku ranged from 0.7×10^4 to 1.6×10^4 CFU/cm^2 while the mean range of the total heterotrophic bacterial counts of pupils feeding area surfaces in schools located in Port Harcourt ranged from 0. The total coliform, faecal coliform, *Salmonella-Shigella* and total *Staphylococcal* counts of pupils feeding area surfaces in schools located in Omoku ranged from 0.6×10^3 to 8.3×10^3, 0.02×10^2 to 0.13×10^2, 0.8×10^2 to 6.3×10^2, and 3.9×10^2 to 4.1×10^2 CFU/cm^2 respectively, while the ranges of total coliform, faecal coliform, *Salmonella-Shigella* and total *Staphylococcal* counts of pupils feeding area surface in the four schools located in Port Harcourt ranged from 0.1×10^3 to 11.0×10^3, 0.2×10^2 to 37.8×10^2, 0.8×10^2 to 9.2×10^2 and 0.5×10^2 to 4.9×10^2 CFU/cm^2 respectively.

3.4 Bacterial Population of Play Mat Surfaces

The mean bacterial load of the play mat surfaces of the four different schools in Omoku and Port Harcourt is presented in Table 4. Results showed that the total heterotrophic bacterial count of play mat surface of schools located in Omoku ranged from 0.1×10^4 to 4.3×10^4 CFU/cm^2, while the total heterotrophic bacterial counts of schools in Port Harcourt ranged from 0.3×10^4 to 0.40×10^4 CFU/cm^2. The total coliform and total *Staphylococcal* counts of play mat surfaces of schools in Omoku ranged from 0.4×10^2 to 1.8×10^2 and 0.2×10^4 to 1.4×10^4 CFU/cm^2 respectively, while the ranges of total coliform and total *Staphylococcal* counts of play mat surface of the four schools located in Port Harcourt ranged from 0.1×10^2 to 0.50×10^2, 0 to 0 and 0.2×10^4 to 1.5×10^4 CFU/cm^2 respectively. Result recorded no faecal coliform counts and no *Salmonella-Shigella* counts for both locations.

3.5 Bacterial Population of Table Surfaces

The mean bacterial load of Table surfaces in the four different schools in Omoku and Port Harcourt is presented in Table 5. Results showed that the total heterotrophic bacterial count of the tables of schools located in Omoku ranged from 0.7×10^3 to 5.9×10^3 CFU/cm^2, while the total heterotrophic bacterial counts of schools in Port Harcourt ranged from 0.8×10^3 to 13.4×10^3 CFU/cm^2. The total heterotrophic bacterial counts of Table surfaces of schools in Omoku and Port Harcourt location showed a significant difference (*p<0.05*). Results also showed that The total coliform, faecal coliform, *Salmonella-Shigella* and total *Staphylococcal* counts of the table surfaces of schools in Omoku ranged from 0.2×10^3 to 1.5×10^3, 0.3×10 to 2.0×10, 0.2×10^2 to 0.85×10^2, and 0.7×10 to 4.0×10 CFU/cm^2 respectively, while the ranges of total coliform, faecal coliform, *Salmonella-Shigella* and total *Staphylococcal* counts of the table surfaces of the four schools located in Port Harcourt ranged from 0.2×10^3 to 2.2×10^3, 0.4×10 to 4.0×10, 0.3×10^2 to 27.0×10^2 and 0.9×10 to 3.9×10 CFU/cm^2 respectively.

3.6 Bacterial Population of Toilet Seat Surfaces

Results of the bacterial load of the Toilet seats surfaces investigated in the four different schools in Omoku and Port Harcourt is presented in Table 6. Results showed that the total heterotrophic bacterial counts of the four different toilet seats of schools in Omoku ranged from 2.1×10^5 to 4.2×10^5 CFU/cm^2, while the total heterotrophic bacterial counts of the four different toilet seats of schools in Port Harcourt ranged from 1.8×10^5 to 3.5×10^5 CFU/cm^2. Results also showed that there was no significant difference ($P\geq0.05$) in the total heterotrophic counts of the toilet seats in all the schools despite the locations. The total coliform and faecal coliform counts of the four different toilet seats of schools in Omoku ranged from 2.1×10^4 to 2.5×10^4 and 0.2×10^2 to 8.9×10^2 CFU/cm^2, while the total coliform and faecal coliform counts of the four different toilet seats of schools in Port Harcourt ranged from 0.4×10^4 to 3.8×10^4 and 0.05×10^2 to 0.12×10^2 CFU/cm^2. Statistically, there was no significant difference ($P \geq 0.05$) in the total coliform and faecal coliform counts of the toilets in all the schools despite the locations. The *Salmonella-Shigella* counts of the four different toilet seats of schools in Omoku ranged from 1.2×10^3 to 1.4×10^3 CFU/mL while the *Salmonella-Shigella* counts of the four different toilet seat surfaces of schools in Port Harcourt ranged from 0.05×10^3 to 0.23×10^3 CFU/cm^2. The total *Staphylococcal* counts of the four different toilet seats of schools in Omoku ranged from 0.7×10^4 to 1.5×10^4 CFU/cm^2, while the total *Staphylococcal* counts of the four different toilet seats of schools in Port Harcourt ranged from 1.9×10^4 to 3.3×10^4 CFU/cm^2. The *Salmonella-Shigella* counts and total *Staphylococcal* counts of Toilet seat surfaces of schools in Omoku and Port Harcourt location showed a significant difference (*P<0.05*).

3.7 Bacterial Population of Toys

Results of the bacterial load of the toys used by pupils in the various schools across Omoku and Port Harcourt are presented in Table 7. Results showed that the total heterotrophic bacterial counts of Toys used by pupils in some schools located in Omoku ranged from 2.3×10^5 to 2.5×10^5 CFU/cm^2, while the total heterotrophic bacterial counts of Toys used by pupils in some schools located in Port Harcourt ranged from 1.5×10^5 to 2.1×10^5 CFU/cm^2. There was no recorded significant difference ($P\geq0.05$) in the total heterotrophic bacterial counts of the toys used by pupils in the different schools both in their location and across the locations. The total coliform counts of Toys used by pupils in some schools located in Omoku ranged from 1.0×10^4 to 4.7×10^4 CFU/cm^2, while the total coliform counts of Toys used by pupils in some schools located in Port Harcourt ranged from 0.2×10^4 to 14.6×10^4 CFU/cm^2. The faecal coliform counts of Toys used by pupils in some schools located in Omoku ranged from 0.7×10^2 to 31.0×10^2 CFU/cm^2, while the faecal coliform counts of Toys used by pupils in some schools located in Port Harcourt ranged from

0.2×10^2 to 1.5×10^2 CFU/cm^2. Statistical analysis showed that there was no significant difference ($p\geq0.05$) in the total coliform counts and faecal coliform counts of toys used by pupils in the four different schools in Omoku and Port Harcourt. The *Salmonella-Shigella* counts of Toys used by pupils in some schools located in Omoku ranged from 0.2×10^4 to 0.84×10^4 CFU/cm^2, while the *Salmonella-Shigella* counts of Toys used by pupils in some schools located in Port Harcourt ranged from 0.3×10^4 to 15.2×10^4 CFU/cm^2. The total *Staphylococcal* counts of Toys used by pupils in some schools located in Omoku ranged from 0.5×10^3 to 1.9×10^3 CFU/cm^2 while the total *Staphylococcal* counts of Toys used by pupils in some schools located in Port Harcourt ranged from 0.8×10^3 to 8.4×10^3 CFU/cm^2. There was no recorded significant difference ($p\geq0.05$) in the total heterotrophic bacterial counts, total coliform counts, faecal coliform counts and *Salmonella-Shigella* counts of the toys in the different schools in Omoku and Port Harcourt. The total *Staphylococcal* counts of Toys used by pupils of some schools in Omoku and Port Harcourt location showed a significant difference ($p<0.05$).

3.8 Bacterial Population of Window Surfaces

Results of the bacterial load of the window surfaces in the various schools across Omoku and Port Harcourt are presented in Table 8. Results showed that the total heterotrophic bacterial counts of windows in some schools located in Omoku ranged from 0.3×10^5 to 2.5×10^5 CFU/cm^2, while the total heterotrophic bacterial counts of windows in some schools located in Port Harcourt ranged from 0.3×10^5 to 1.3×10^5 CFU/cm^2. The total coliform counts of windows in some schools located in Omoku ranged from 0.1×10^4 to 1.3×10^4 CFU/cm^2, while the total coliform counts of window surfaces in some schools located in Port Harcourt ranged from 0.2×10^4 to 1.0×10^4 CFU/cm^2. There was no count of the faecal coliform counts in the window surfaces of the various schools located in Omoku and Port Harcourt. The *Salmonella-Shigella* counts of windows in some schools located in Omoku ranged from 0.2×10^3 to 1.3×10^3 CFU/cm^2, while the *Salmonella-Shigella* counts of windows in some schools located in Port Harcourt ranged from 0.1×10^3 to 0.73×10^3 CFU/cm^2. The *Staphylococcal* counts of window surfaces in some schools located in Omoku ranged from 1.0×10^3 to 4.7×10^3 CFU/cm^2, while the *Staphylococcal* counts of windows in some schools located in Port Harcourt ranged from 0.8×10^3 to $.4\times10^3$ CFU/cm^2. There was no recorded significant difference ($p\geq0.05$) in the total heterotrophic bacterial counts, total coliform counts, *Salmonella-Shigella* counts and total *Staphylococcal* counts of the windows in the different schools in Omoku and Port Harcourt.

Table 1: Bacterial Load (CFU/cm^2) of Diaper Changing Surfaces of Schools in Omoku and Port Harcourt

Sample Location	THB ($\times10^4$)	TCC ($\times10^4$)	FC ($\times10$)	SSC ($\times10^3$)	TSC ($\times10^3$)
Omoku	7.7±3.6^a	3.7±3.1^a	3.9±2.8^a	1.4±0.3^a	3.5±0.4^a
PH	6.1±2.2^a	3.3±0.4^a	5.0±0.8^a	4.7±0.8^a	18.5±3.8^a
P-value	0.18	0.77	0.69	0.18	0.19

*Means with similar superscript (alphabet) along the columns have no substantial difference ($p\geq 0.05$)

Keys: THB = total heterotrophic bacteria, TCC = total coliform counts, FC = Faecal coliform, SSC = *Salmonella-Shigella* count, TSC = total staphylococcal counts,

Table 2: Bacterial Load (CFU/cm^2) of Door Handle Surfaces of Schools in Omoku and Port Harcourt

Sample location	THB ($\times10^5$)	TCC ($\times10^5$)	FC ($\times10^3$)	SSC ($\times10^4$)	TSC ($\times10^4$)
Omoku	1.9±0.2^a	1.5±0.2^a	4.8±0.9^a	1.2±0.2^a	5.0±3.6^a
PH	2.2±1.9^a	0.79±0.2^a	0.32±0.2^a	0.86±0.1^a	0.45±0.1^a
P-value	0.73	0.26	0.09	0.55	0.34

*Means with similar superscript (alphabet) along the columns have no substantial difference ($p\geq 0.05$)

Keys: THB = total heterotrophic bacteria, TCC = total coliform counts, FC = Faecal coliform, SSC = *Salmonella-Shigella* count, TSC = total staphylococcal counts,

Table 3: Bacterial Load (CFU/cm^2) of the Feeding Area Surface of Schools in Omoku and Port Harcourt

Sample location	THB ($\times10^4$)	TCC ($\times10^3$)	FC ($\times10^2$)	SSC ($\times10^2$)	TSC ($\times10^2$)
Omoku	1.6±0.7^a	8.3±0.6^a	0.13±0.02^a	6.3±0.8^a	4.1±3.9^a
PH	1.3±0.8^a	11.0±0.1^a	37.8±0.2^a	9.2±0.8^a	4.9±0.5^a
P-Value	0.26	0.47	0.33	0.42	0.67

*Means with similar superscript (alphabet) along the columns have no substantial difference ($p\geq 0.05$)

Keys: THB = total heterotrophic bacteria, TCC = total coliform counts, FC = Faecal coliform, SSC = *Salmonella-Shigella* count, TSC = total staphylococcal counts,

Table 4: Bacterial Load (CFU/cm^2) of Play Mat Surface of Schools in Omoku and Port Harcourt

Sample location	THB ($\times10^4$)	TCC ($\times10^2$)	FC ($\times10^2$)	SSC ($\times10$)	TSC ($\times10^4$)
Omoku	4.3±0.1^a	1.8±0.4^a	0±0^a	0±0^a	1.4±0.2^a
PH	0.40±0.3^a	0.50±0.1^a	0±0^a	0±0^a	1.5±0.2^a
P-value	0.31	0.27	No Variance	0.11	0.84

*Means with similar superscript (alphabet) along the columns have no substantial difference ($p\geq 0.05$)

Keys: THB = total heterotrophic bacteria, TCC = total coliform counts, FC = Faecal coliform, SSC = *Salmonella-Shigella* count, TSC = total staphylococcal counts,

Table 5: Bacterial Load (CFU/cm^2) of the Table Surfaces of Schools in Omoku and Port Harcourt

Sample Location	THB ($\times10^3$)	TCC ($\times10^3$)	FC ($\times10$)	SSC ($\times10^2$)	TSC ($\times10$)
Omoku	5.9±0.7^a	1.5±0.2^a	2.0±0.3^a	0.85±0.2^a	4.0±0.7^a
PH	13.4±0.8^b	2.2±0.2^a	4.0±0.4^a	27.0±0.3^b	3.9±0.9^a
P-value	0.03	0.45	0.28	0.05	0.97

*Means with similar superscript (alphabet) along the columns have no substantial difference ($p\geq 0.05$).
Significant difference noticed for means with superscript (alphabet) b ($p<0.05$).

Keys: THB = total heterotrophic bacteria, TCC = total coliform counts, FC = Faecal coliform, SSC = *Salmonella-Shigella* count, TSC = total staphylococcal counts

Table 6: Bacterial Load (CFU/cm^2) of the Toilet Seat Surfaces of Schools in Omoku and Port Harcourt

Sample Location	THB (×10^5)	TCC (×10^4)	FC (×10^2)	SSC (×10^3)	TSC (×10^4)
Omoku	4.2±2.1^a	2.5±2.1^a	8.9±0.2^a	1.4±1.2^b	1.5±0.7^a
PH	3.5±1.8^a	3.8±0.4^a	0.12±0.05^a	0.23±0.05^a	3.3±1.9^b
P-value	0.41	0.74	0.17	0.006	0.02

*Means with similar superscript (alphabet) along the columns have no substantial difference ($p \geq 0.05$)
Significant difference noticed for means with superscript (alphabet) b ($p<0.05$).

Keys: THB = total heterotrophic bacteria, TCC = total coliform counts, FC = Faecal coliform, SSC = *Salmonella-Shigella* count, TSC = total staphylococcal counts

Table 7: Bacterial Load (CFU/cm^2) of the Toys of Schools in Omoku and Port Harcourt

Sample Location	THB (×10^5)	TCC (×10^4)	FC (×10^2)	SSC (×10^4)	TSC (×10^3)
Omoku	2.5±2.3^a	4.7±1.0^a	31.0±0.7^a	0.84±0.2^a	1.9±0.5^a
PH	2.1±1.5^a	14.6±0.2^a	1.5±0.2^a	15.2±0.3^a	8.4±0.8^b
P-value	0.55	0.14	0.19	0.51	0.03

*Means with similar superscript (alphabet) along the columns have no substantial difference ($p \geq 0.05$)

Keys: THB = total heterotrophic bacteria, TCC = total coliform counts, FC = Faecal coliform, SSC = *Salmonella-Shigella* count, TSC = total staphylococcal counts

Table 8: Bacterial Load (CFU/cm^2) of the Window Surface of Schools in Omoku and Port Harcourt

Sample location	THB (×10^5)	TCC (×10^4)	FC (×10^2)	SSC (×10^3)	TSC (×10^3)
Omoku	2.5±0.3^a	1.3±0.1^a	0.0±0.0^a	1.3±0.2^a	4.7±1.0^a
PH	1.3±0.3^a	1.0±0.2^a	0.0±0.0^a	0.73±0.1^a	6.4±0.8^a
P-value	0.19	0.56	No variance	0.31	0.67

*Means with similar superscript (alphabet) along the columns have no substantial difference ($p \geq 0.05$)

Keys: THB = total heterotrophic bacteria, TCC = total coliform counts, FC = Faecal coliform, SSC = *Salmonella-Shigella* count, TSC = total staphylococcal counts

4. Discussion

The use of microbiological methods in monitoring environmental surfaces has become one of the best tools for evaluating the hygiene of an environment since it determines the types of bacteria, fungi or viruses that are present or predominant in such environments. This conforms to the study by Cosby (2005) [2] that to measure bacterial contamination on contact surfaces and facilities, environmental microbiological surveys have been frequently used. This study investigated the bacteriology of environmental surfaces in Day-care and Nursery school centres in Omoku and Port Harcourt locations of Rivers State. The bacteriological counts especially in the total faecal coliforms, staphylococcal counts and total heterotrophic bacteria in the door handles, diaper changing, feeding area, play mat, toilet seat and window surfaces were generally high and showed high fluctuations in the schools within and across the geographical locations.

The highest total heterotrophic bacterial counts were recorded from diaper changing surfaces in schools from Port Harcourt location and were substantially higher ($p \geq 0.05$) than counts recorded in schools from Omoku. Although there was disparity in the bacteriological counts of diapers changing surfaces from schools in Omoku. Despite this variation in counts, there was no substantial difference ($p \geq 0.05$) in the total heterotrophic bacterial counts of diapers obtained from pupils in Omoku schools even though diapers from some schools in this region had similar and the highest total heterotrophic bacterial counts in this location. Comparative analysis of both locations (Omoku and Port Harcourt) also showed no substantial differences ($p \geq 0.05$) in the counts of the total heterotrophic bacteria and the highest count was recorded in diaper changing surfaces from schools located in Port Harcourt. The total heterotrophic bacterial load of diaper changing surfaces from pupils in the eight different schools across the two locations in the present study were higher than the total heterotrophic load in the range of 1.0 x 10^3 to 2.0 x 10^3 CFU/g reported by Eteyen *et al*. (2020) [3].

The total coliform counts was higher in the diaper changing surfaces of schools located in Omoku followed by the toilet seat surfaces of schools in this same location while the feeding area surfaces of schools in Port Harcourt region recorded higher total coliform counts than those in the Omoku region. The lowest total coliform count was recorded from play mat surfaces in both locations. The counts of the total coliform bacteria of door handles, diapers changing area, feeding area, play mat, tables, toilet seat, toys and window surfaces from the schools in Omoku and Port Harcourt showed no substantial differences ($p \geq 0.05$) despite the disparity in the counts. The counts of the total faecal coliform bacteria of diapers changing areas, door handles, feeding areas, tables, toilets and toys shows that they were no counts in the faecal coliform bacteria in the play mat surfaces and window surfaces of the various schools in Omoku and Port Harcourt. Although, the faecal coliform counts of the table surfaces was higher in schools from Port Harcourt, schools in Omoku recorded higher counts in faecal coliforms than those recorded in schools from Port Harcourt.

More so, results also showed detectable counts in the *Salmonella-Shigella* and *staphylococcal* in all schools from both locations with Omoku having higher counts than those from Port Harcourt for *Salmonella-Shigella* and *staphylococcal* counts. The presence of micro-organisms in the raw materials during manufacturing, low amounts of antimicrobial agents engrained in the diaper, managing processes during diaper exchange, transportation, and other environmental and intrinsic factors could all be attributed to the high total heterotrophic bacterial, total coliforms, faecal coliform, and staphylococcal counts in the diapers (Eteyen *et al*., 2020) [3]. Some of the intrinsic factors could be associated with the amount of faecal matter in the diaper changing surfaces as well as the various activities carried out by the toddlers. Furthermore, the variation in bacteriological counts between schools in different locations could be linked to facility design and construction, which may affect the availability of hand washing facilities and the separation of distinct age groups of children inside the centers (Cosby, 2005) [2] as well as the forms of hygiene

being practiced in the various schools.

Door handles are known to be in constant touch and the presence of microorganisms in door handles may not be limited by environmental factor alone but by exchange of microbial flora in hands of users. Thus, the high counts of total heterotrophic bacteria, total coliforms, and staphylococci in the current investigation could be due to the handles' continuing use by different individuals. After repeated contact with different persons, inanimate environmental surfaces such as door handles can get directly contaminated with bacteria (Hassan *et al*., 2022) [4]. In this study, door handle recorded the highest total heterotrophic bacteria counts from schools in Omoku and showed no substantial difference ($p \geq 0.05$) from counts recorded from the two locations. The total heterotrophic bacterial counts of door handle from the four different schools in Port Harcourt showed no significant differences despite the disparity in the counts recorded from schools in that location. The faecal coliform counts of door handle from schools located in Omoku were lower than those reported in schools from Port Harcourt. Results also showed no substantial differences in the faecal coliform counts of door handles among schools located in Omoku and Port Harcourt. Results also showed that while there were detectable counts for coliform, and staphylococci in door handles from schools in Omoku location and schools in Port Harcourt location. This disparity in counts associated with the door handles are also to a large extent associated with the level of hygiene especially hand washing hygiene that is practiced. The use of soap and clean water to wash hands especially after using the toilet or touching inanimate object could reduce bacteriological loads in door handles. In a previous study, poor hand hygiene practices after making use of toilet as well as lack of adequate cleaning and sanitation of facilities has been suggested as major factors that contributes to microbial contamination of door handles (Maryam *et al.*, 2014) [5].

Bacteriological contamination of environment such as play mat surfaces, feeding area surfaces and table surfaces of child day-care could be an important route in the spread of infectious diseases like diarrhoea amongst children and caregivers (Moritz, 2010) [6]. The total heterotrophic bacterial, faecal coliform and staphylococcal counts in the present study on children play mat surfaces, feeding area surfaces and table surfaces were generally high. The total heterotrophic bacterial counts of feeding area surfaces had higher counts from schools in Port Harcourt than from schools in Omoku location. Despite these variations recorded between schools, there was no significant difference ($p \geq 0.05$) in the total heterotrophic bacterial counts of student feeding area in schools located in Omoku and Port Harcourt. More so, in comparing the total heterotrophic bacterial counts of the feeding area surfaces in both locations, the feeding area in Port Harcourt schools were higher than values recorded for feeding areas of schools in Omoku. For the total faecal coliform counts, there were no substantial differences in the counts recorded in the feeding area within schools in the respective locations.

More so, the total heterotrophic bacterial counts of play mat surfaces in schools in the various locations despite the disparity in counts do not show any substantial difference ($p \geq 0.05$). There were no counts recorded for the faecal coliform in the various play mat surfaces within and across the two locations. Furthermore, there was a substantial difference ($p<0.05$) in the total heterotrophic bacterial counts of Tables of schools in Omoku and Port Harcourt. There was no significant difference ($p \geq 0.05$) in the faecal coliform counts of Tables in schools in the various locations. There was also a substantial difference in the *Salmonella-Shigella* and total *Staphylococcal* count in toilet seat surfaces of schools in Omoku and Port Harcourt locations. Some of the factors that may have influenced the bacterial contamination in these environments could be overcrowding and general use of play mat surfaces and table surfaces. In the present study, all the children used same play mats, tables and were kept together in the feeding area in their different schools. Thus, contamination of these surfaces could have resulted from exchange of normal flora of children as well as their caregivers. It has been documented that the concentration of faecal coliforms in surface, air, and hand samples was related to the age of children because faecal coliforms are often associated with diaper changing and this component decreases as children become potty trained (Cosby, 2005) [2].

Other factors that could also contribute to the increased bacteriological load in these areas as well as in the windows could be attributed to human activities like talking, sweeping, indoor and outdoor air as well as wearing shoes on these environments (Sule and Awe, 2019) [12]. The toys used by the toddlers in the day care centres showed presence of bacteria and these could be attributed to the normal flora on body surfaces of the toddlers as well as the microorganisms present in the environment to which they are found. It is well documented that microorganisms are ubiquitous and this makes them readily available in all environments (Prescott *et al.*, 2011) [10].

5. Conclusion

Based on the data obtained for the bacterial population from these environmental surfaces, it was concluded that the bacterial load in the various samples: diapers changing areas, feeding areas and toys from the various schools were generally very high. Although there are no standards in the bacterial load acceptable in these samples but the high bacterial counts especially in faecal coliform and staphylococci is of public health importance.

6. Conflict of Interest

Not available

7. Financial Support

Not available

8. References

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