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Colonization of School Children (Age less than 5 years) Lunch Box by bacterial Pathogens Associated with Infant Diarrhea in Shomolu Local Government Area, Lagos, Nigeria

Abstract

Introduction: The incidence of Infant diarrheal disease is a global problem which is more prevalent in developing countries. The survey was conducted to evaluate infant diarrhea, among school children and implicated bacterial pathogens in Shomolu Local Government Area, Lagos State, Nigeria.

Materials and Methods: Ethical permissions were obtained, research questionnaires were administered to mothers with children below five years old and forty five samples i.e., lunch boxes/baskets obtained in groups of nine at five different schools within the study area were used for the analysis. Research questions were generated, various hypotheses were tested, and analysis of questionnaires (using SPSS Data analysis), case records and laboratory results were appropriately analyzed using conventional methods of isolating both enterobacteriaceae and other Gram positive organisms as basis for generating inferences in the study.

Result: All the examined lunch boxes/baskets were contaminated with at least two bacterial species and five bacteria species were recovered from the samples namely; *Staphylococcus aureus, Escherichia coli, Salmonella typhi, Shigella dysenteriae* and *Pseudomonas aeruginosa. Staphylococcus aureus* (29.4%) was the most frequent isolates followed by *Shigella dysenteriae* (26.5%), *Salmonella typhi* (24.5%) and *Pseudomonas aeruginosa* (11.8%) while *Escherichia coli* (7.8%) were the least frequent isolates.

Conclusion: Unhygienic practices, inadequate education and knowledge about basic food safety issues were responsible frequent diarrheal episodes. Food safety education of the target population and enforcement of existing laws on environmental sanitation and hygiene as a basic preventive strategy is hereby recommended.

Keywords: Infant diarrhea; Lunch box; pathogens; contaminant; hygiene

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Introduction

Colonization of children's lunch box (under 5 years old) by pathogens of bacterial origin like *Staphylococcus aureus, Escherichia coli* and other members of the genera *Enterobacteriaceae* which are causative agents of diarrhea, dysentery, food poisoning and intoxication could be a problem of public health and epidemiological concern which could pose a serious threat to the health of the young ones, since diarrhea is majorly transmitted through the fecal-oral route, which includes ingestion of food or water contaminated by feces, contact of person-to-person and direct contact with feces that had been

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infected [1]. The incidence of Infant diarrheal disease is a global problem which is more prevalent in developing countries due to poverty, limited education, poor environmental sanitation and inadequate water supplies and it is grossly under-reported/ under-estimated because of poverty and/or ignorance among the affected group of people who constitute a large percentage of a developing country [2].

The period of rapid growth and increased demand for calories and proteins is called infancy [3]. Infant mortality is a major public health concerns as debilitating picture of poverty, malnutrition and diseases still constitutes a huge decimal in Nigeria and demographic data on Infant Mortality Rate (IMR) are still hugely inadequate because many deaths occur at home and are not recorded in official statistics [4].

Reports from various parts of the world have implicated various pathogens such as Parasites (e.g., *Giardia lamblia, Entamoeba histolytica*), Bacteria (e.g., *Escherichia coli, Salmonella species, Klebsiella species, Enterobacter species e.t.c.,*) and Viruses (e.g., *Rotavirus*) with the outbreak of infant diarrheal disease [3-6].

There is variation in the risks factors with child's age, the local environment and the pathogens involved [7]. The risk factors include the following: sanitation factors, malnutrition, eating habits, breastfeeding, socio-economic factors, hygienic practices, immunodeficiency, consumption of food sold by vendors, waterrelated factors, seasonal distribution and demographic factors.

The association of the following with risk of diarrhea morbidity in children had been revealed by some studies i.e., unsafe food storage, eating cold left overs, eating food with long and dirty finger nails, children not washing hands after defecation and before meals, presence of flies inside the house or kitchen, mothers not washing hands before preparing food or feeding children, dirty utensils or feeding bottles, presence of animals inside the house, unsafe food storage and unhygienic domestic places like yard, living room or parlor and kitchen [8-11].

Eating of contaminated foods (e.g. vegetables, ice cubes, fruits, water, locally made juices or drinks, seafood and raw meat) i.e., by the local population or tourists visiting foreign countries with poor sanitation and warm climates, is also a significant factor [12]. One of the most important causes of severe diarrhea is Rotavirus and other viruses which may be important causes of diarrheal disease in humans are enteric Adenoviruses, Astroviruses, Caliciviruses, Norwalk-like viruses, and Norwalk viruses.

The loss of water from the body, which is always associated with the disturbance of salt metabolism is termed diarrhea and the symptoms include extreme thirst, fever and weight loss, special risk is attributed with infants with diarrhea and are vomiting [13].

The signs of dehydration are not evident until there is an acute loss of fluid about four to five percent of the body weight and the signs and symptoms include the following; dry mouth and throat, loss of the elasticity of the skin, fast and weak pulse, reduction in the amount of urine and sunken fontanelle which lead to death to shock and ultimately death of children under five years old [14].

Thirst and dryness of the mouth may be experienced by patients with mild dehydration while orthostatic hypotension with syncope i.e., fainting upon standing caused by a reduction in the volume of blood, which causes a drop in the blood pressure upon standing, severe weakness, kidney failure, shock, a diminished urine output, confusion, acidosis i.e., too much acid in the blood and coma [15].

Frequent watery or loose stools, risks of dehydration, loss of appetite with or without vomiting and damage to the intestine (especially, when bloody diarrhea is involved) and the signs and symptoms of diarrhea can be identified by severity of manifestation, which include severe diarrhea, moderate diarrhea and mild diarrhea. In severe diarrhea, there is signs of rapid weight loss, weak pulse and cold limbs (shock or coma), by the child. Signs of rapid deep breathing, loss of skin elasticity, sunken funtanelle and eyes are characteristics of moderate diarrhea. Children affected by mild diarrhea show signs of being thirsty, dry mouth, weight loss and passage of less urine [16].

Also, there is the passage of frequent loose and watery stools, fever (particularly if there is an infectious cause), abdominal pain or cramps and bleeding [17].

Lunch boxes/baskets of school children below five years old were chosen because it contains food items which are one of the basic necessities of life apart from clothing and shelter. It also transcends beyond all geographical locations, races and cultures globally and any hazard due to biological agents could be life threatening or fatal.

The study is designed to isolate and identify possible bacteria contaminants responsible for infant diarrhea associated with lunch boxes/baskets of children below five years old in Shomolu Local Government Area, Lagos, Nigeria.

Materials and Methods

Study Site/Area

The targeted community was Shomolu Local Government situated in Lagos State, Nigeria. It has a population of 403, 569 people according to the census conducted in 2006 and the geographical coordinates of 6° 32′ 27″ North, 3° 23′ 14″ East. The location decimated with poor structures and household overcrowding where majority in the population are traders and few people in the study sites involved in white collar jobs like teaching, accounting and the rest (**Figure 1**).

Ethical Considerations

Ethical permissions were obtained from the Local Government Chairman and Chief Medical Officer of Health Centers in the project community at Shomolu Local Government Area, Lagos State with a detailed letter obtained from the project supervisor. Oral permission was given and a copy of the letter bearing the seal of approval i.e., official stamp issued from the local government office was shown to the head masters/mistresses of schools where samples were collected and to the officer in charge of medical records in the health center where diarrhea case records were obtained.

Questionnaires Administration and Analysis

One hundred and ten questionnaires were administered to mothers having children below five years old in the study area, screened for detailed information and ten questionnaires were discarded due to lack of completeness. The selected questionnaires were analyzed with SPSS data analysis, research questions were generated, hypotheses were tested and appropriate inferences were made.

Sample Collection

Forty-five samples were taken from the lunch boxes or baskets of children below five years old at five different schools located in Shomolu local government, Lagos state in groups of nine namely:

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- 1. Felitorah Children School, Adetayo Osho Street (code: FL).
- 2. Kabma-F International School, Femi Adebule Street (code: KF).
- 3. Holly-moore Nursery and Primary school, Awolaja Street, off Oju Square (code: HM).
- 4. Fehope Nursery and Primary School, Akoka (code: FH).
- 5. Trix Villa Schools, Fola-agoro (code: TV).

Procedure of Sample Collection

Swab sticks moisturized with sterile water aseptically using hypodermic syringe, were used in swabbing the lunch boxes/ baskets of children (below 5 years old) and later sorted into a zipped plastic bag aseptically for analysis.

Inoculation of Samples

Forty-five samples was inoculated into the nutrient broth in groups of nine, in accordance with code from the source and labeled appropriately.

Sub Culturing into Various Selective, Differential and General Media (i.e., Nutrient Agar)

The isolates were sub cultured from nutrient agar into mannitol salt agar to screen for bacteria belonging to *Staphylococci* and into mackonkey agar, eosine methylene blue and *Salmonella Shigella* agar respectively to screen for bacteria belonging to *Enterobacteriaceae* in that order respectively. Then later sub cultured into nutrient Agar, so as to obtain fresh culture for biochemical tests. Conventional methods of isolation of various microorganisms were adopted using standard biochemical analysis including catalase, coagulase, DNAse, motility and indole tests.

Results Discussion and Conclusion

(Tables 1 and 2, Figure 2)

 Table 1. Relative Occurrence of Infant Diarrhea and Method Used in Cleaning of Lunch Boxes/Baskets.

		Method used in cleaning of lunch boxes/baskets				
		Washing with soap and water	Wiping with napkins and foam	Use of baby wipes	Others	
Relative occurrence of infant diarrhoea	Yes	50 (69.4%)	9 (75%)	1 (100%)	3 (100%)	
Total		72 (100%)	12 (100%)	1 (100%)	3	

 Table 2. Relative Occurrence of Infant Diarrhea and Frequency of Cleaning Lunch

 Boxes/Baskets.

		Relative occurrence of infant diarrhea		Total
		Yes	No	
Frequency of cleaning lunch boxes/baskets	Daily	37 (66.1%)	19 (33.9%)	56 (68.3%)
	Weekly	18 (85.7%)	3 (14.3%)	21 (25.6%)
	Twice weekly	4 (80.0%)	1 (20.0%)	5 (6.1%)
Total		59 (72.0%)	23 (28.0)	82(100%)

Percentage Occurrence of Bacterial Isolates Recovered per Samples

Among the 45 samples i.e., lunch boxes/food baskets collected (**Table 3**) all were positive to bacterial pathogens causing diarrhea, in which *Staphylococcus aureus* was isolated from 30 samples (29.4%); *Shigella dysenteriae* was detected in 27 samples (26.5%); *Salmonella typhi* in 25 samples (24.5%); *Pseudomonas aeruginosa* in 12 samples (11.8%) and *Escherichia coli* in 8 samples (7.8%).

Percentage Occurrence of Bacterial Isolates Recovered per School

The percentage occurrence (**Table 3**) of bacteria pathogens recovered from samples (i.e., lunch boxes/food baskets) obtained

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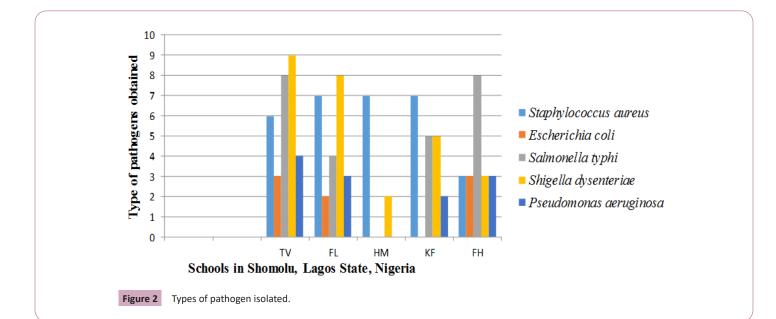


Table 3. Percentage Contribution of Bacterial Isolates Recovered from Samples Obtained in the Study Area.

School Code	Chambula a saus auraus	Escherichia coli	Salmonella typhi	Chigollo dugostavioo	Pseudomonas	Total	
School Code	Staphylococcus aureus	Escherichia coli	Saimonena typni	Shigella dysenteriae	aeruginosa		(%)
TV	6	3	8	9	4	30	29.4
FL	7	2	4	8	3	24	23.5
HM	7	0	0	2	0	9	8.8
KF	7	0	5	5	2	19	18.6
FH	3	3	8	3	3	20	19.6
Total	30	8	25	27	12	102	
Percentage	29.40%	7.80%	24.50%	26.50%	11.80%		100

Table 4. Relative Occurrence of Infant Diarrhea and Level of Formal Education.

		Relative of	Total (%)	
		Yes	Νο	Total (%)
	M.Sc	1 (100.0%)	0 (100.0%)	1
	B.Sc. / B.Pharm/ B.A.	11 (91.7%)	1 (8.3%)	12
	HND	7 (87.5%)	1 (12.5%)	8
Level of formal education	OND	12 (63.2%)	7 (36.8%)	19
	SSCE	21 (53.8%)	18 (46.2%)	39
	SS 3	9 (90.0%)	1 (10.0%)	10
	JS 3	1 (100.0%)	0 (0.0%)	1
	Primary School	5 (100.0%)	0 (0.0%)	5
	No Formal Education	3 (75.0%)	1 (25.0%)	4
	NCE	1 (100.0%)	0 (0.0%)	1
Total (%)		71	29	100

from the five schools in increasing order is as follows: Holly-moore Nursery and Primary School (8.8%), Kabma-F International School (18.6%), Fehope Nursery and Primary School (19.6%), Felithorah Children School (23.5%) and Tris Villa School (29.4%).

Percentage Composition of Bacterial Isolates Recovered per School

The most frequent isolated bacteria recovered from the lunch boxes obtained from Tris Villa School are *Shigella dysenteriae* (30.0%) and others in decreasing order are as follows: *Salmonella typhi* (26.7%), *Staphylococcus aureus* (20.0%), *Pseudomonas*

aeruginosa (13.3%) and Escherichia coli (10.0%).

The most frequent isolated bacteria recovered from the lunch boxes obtained from Felithorah Children School are *Shigella dysenteriae* (33.3%) and others are as follows: *Staphylococcus aureus* (29.2%), *Salmonella typhi* (16.7%), *Pseudomonas aeruginosa* (12.5%) and *Escherichia coli* (8.3%).

The most frequent isolated bacteria recovered from the lunch boxes/food baskets obtained from Holly-moore Nursery and Primary School are *Staphylococcus aureus* (77.8%) and few *Shigella dysenteriae* (22.2%) but *Escherichia coli* (0.0%), *Salmonella typhi*

(0.0%) and Pseudomonas aeruginosa (0.0%) were absent.

The most frequent isolated bacteria recovered from the lunch boxes obtained from Kabma-F International School are *Staphylococcus aureus* (36.8%) and others are as follows: *Salmonella typhi* (26.3%), *Shigella dysenteriae* (26.3%) and *Pseudomonas aeruginosa* (10.5%) but *Escherichia coli* (0.0%) was absent.

The most frequent isolated bacteria recovered from the lunch boxes/food baskets obtained from Fehope Nursery and Primary School are *Salmonella typhi* (40.0%) and others are as follows: *Staphylococcus aureus* (15.0%), *Escherichia coli* (15.0%), *Shigella dysenteriae* (15.0%) and *Pseudomonas aeruginosa* (15.0%).

The colonization of lunch boxes/baskets with pathogenic bacteria to could be from frequently used surfaces like door handles, school desks, kitchen surfaces and toilet flushes. Also, long and dirty finger nails which can easily retain small amounts of fecal matter, packing and eating with unwashed hands and the use of napkins especially if it is used for other purposes like cleaning of kitchen, tables, dusting of furniture etc.

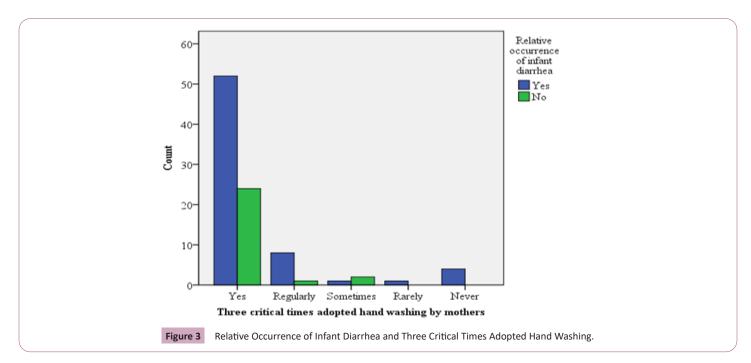
Infant diarrheal infection is most common among children whose lunch boxes or food baskets are cleaned with baby wipes (100.0%) and other methods of cleaning (100.0%), as shown in (**Table 5**). In this study, occurrence of infant diarrhea was also high in children whose lunch boxes or food baskets are wiped with napkins and foam (75.0%) and lowest (i.e., in comparison to others) in those washed daily with soap and water (69.4%) which constitute the most adopted method of cleaning (81.8%). Therefore, the use of appropriate cleaning methods of lunch boxes or food baskets is important in the reduction of gastro-intestinal infections which includes infant diarrhea, since food storage for later consumption could increase the risk of infant diarrhea disease, which is in agreement with the works of Cain cross [18]. That lack of hygiene leads to increase in infant diarrhea infection.

In this study it was indicated that the relative occurrence of infant diarrhea increases with decrease in the frequency of cleaning of lunch boxes or food baskets i.e., daily (66.1%), weekly (80.0%) and weekly (85.7%) This means that the lower the frequency of cleaning of lunch boxes or baskets the higher the risk or chances of children being infected. This is however anticipated and not surprising because the accumulation of dirt and food remains due to infrequent cleaning of lunch boxes/baskets enhances the proliferation of pathogens which can cause infant diarrhea when digested. Also, the highest frequency of cleaning of lunch boxes or baskets enhances the removal of teaming microbial populations that may have accumulated in the lunch boxes or baskets during or after school hours which will reduce the risk of children being infected.

The results in (**Figure 3**) depicted that all the children of mothers who rarely (100.0%) and never (100.0%) adopted the three critical times hand washing had diarrhea, but there was a reduction among children whose mothers adopted the three critical times

 Table 5. Frequency and Methods Used in Cleaning Lunch Boxes/Baskets.

Frequency of cleaning lunch box and basket					
	Washing with soap and water	Wiping with napkins and foam	Use of baby wipes	Others	Total
Daily	49 (81.7%)	7 (11.7%)	1 (1.7%)	3 (5.0%)	60 (68.2%)
Weekly	19 (82.6%)	4 (17.4%)	0 (0.0%)	0 (0.0%)	23 (26.1%)
Twice weekly	4 (80.0%)	1 (20.0%)	0 (0.0%)	0 (0.0%)	5 (5.7%)
Total	72 (81.8%)	12 (13.6%)	1 (1.1%)	3 (3.4%)	88 (100.0%)



hand washing as follows: regularly (88.7%), yes i.e., only affirmed without stating the frequency (67.9%), and sometimes (50.0%). It is however, surprising that mothers who practice regular hand washing at three critical times had high occurrence of infant diarrhea (88.7%) but generally infant diarrhea can be reduced with washing of hands at three critical times. This agrees with the work of Tumwine [19], which reported that reduction in the occurrence of infant diarrhea can be achieved through proper hand washing. Also, most mothers wash their hands at three critical times (81.0%) in the study area and this may be due to the existence of various enlightenment programs which uses different avenues to enhance better accessibility by mothers e.g., the media, free health care services, and various community campaigns like the stop diarrhea initiative and save the children project which are organized by the government and international organizations in the study area.

The report in (**Table 4**) indicated that the occurrence of infant diarrhea is very high among the following groups with respect to level of education i.e., M.Sc. (100.0%), NCE (100.0%), JS 3 (100.0%), Primary school (100.0%), SS 3 (90.0%) and HND (87.5%); High among mothers with no formal education (75.0%) and OND (63.2%) but lowest among mothers with SSCE (53.8%). Also, the level of education most common to mothers in the study area is SSCE (39.0%).

The colonization of lunch boxes/baskets with pathogenic bacteria to could be from frequently used surfaces like door handles, school desks, kitchen surfaces and toilet flushes. Also, long and dirty finger nails which can easily retain small amounts of fecal matter, packing and eating with unwashed hands and the use of napkins especially if it is used for other purposes like cleaning of kitchen, tables, dusting of furnitures etc.

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Conclusion

Unhygienic practices (as observed in structural questionnaires analysis and sampling process) like neglect or lack of proper cleaning of lunch boxes, unhygienic handling, etc., could aid the transmission or spread of pathogens. Innocent explorative/playful attitudes/behaviors of children like keeping of dirty socks in their lunch boxes after school hours, keeping of fallen/used utensils in the lunch boxes, eating of foods e.g. meats/opened biscuits or sweets which had accidentally poured or dropped inside the lunch boxes could be important risk factors in the transmission of infectious agents.

Recommendation

Washing of hands with soap and water before packing and eating from the lunch boxes/baskets should be maintained so as to prevent the transference of bacteria from the skin (i.e., Staphylococcus aureus which is an agent of food poisoning), from nails (long and dirty finger nails could harbor bacteria from the genus Enterobacteriaceae e.g. Escherichia coli which could cause diarrhea, since it is mostly transmitted through the fecal-oral route).

Lunch boxes should be clean every day with soap and water. Infrequent cleaning enhances the proliferation of pathogen organisms which could be harmful to children's health.

The use of baby wipes and napkins in cleaning of lunch boxes/ baskets should be avoided, since these materials cannot remove bacteria but only aid in moving them around in apparently cleaned lunch boxes/baskets because they are not designed for removing pathogens.

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