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Weaning Food Preparations Consumed in Umuahia, Nigeria: Evaluation of The Bacteriological Quality

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Abstract: Bacteriological quality of weaning foods given to children = 2 years was evaluated by estimating bacterial cell count. Bacterial count (geometric mean) ranged from 3.92±0.39 to 6.14±0.23 $\log_{10}$ cfu per ml or g. All food samples examined (pap (*akamu*), rice, moi- moi, agidi, beans, yam and stew, cereals, garri, beverages) were contaminated with some bacterial species. Moi-moi and beans had the highest bacterial counts of 6.14×0^5 and 6.0×10^5 cfu mL^{-1} or g, respectively. The least contaminated foods were cereals and beverages with counts of 3.92×10^4 cfu mL^{-1} or g and 4.49×10^4, respectively. Contamination of foods increased with storage time and type of food. Reheated food had higher bacterial count than freshly cooked food. Foods prepared by maids were found to be more contaminated (poor hygiene standard) than that prepared by mothers, nannies or siblings. Analysis of variance (ANOVA) showed no significant difference ($p>0.05$) between food given to children at home and those served in day-care centers although the later gave a higher bacterial count. Contaminated foods had high levels of *Escherichia coli, Staphylococcus aureus* while *Streptococcus* sp. and *Bacillus* sp. were also identified as common food borne pathogens. Growth profile of isolated organisms in food revealed a logarithmic phase of growth despite the low pH during the first 6 h. Counts of *E. coli* and *S aureus* increased from 10^4 to above 10^8 after 24 h at 37°C. Thus, in combating acute bacterial food borne diseases, the control of time factor during cooking and storage of food should receive special attention in education on health and food safety as well as improving general hygiene.

Key words: Diarrhoea, weaning, bacterial contamination, child care

INTRODUCTION

Contaminated weaning foods have been implicated in quite a large number of diarrhoeal diseases among infants and young children in developing countries. There is evidence of increased diarrhoeal morbidity at the transition from exclusive breast-feeding to a mixed diet as a result of food contamination. Up to 70% of diarrhoeal episodes could be due to pathogens transmitted through food, transmission being mainly by faecal-oral route[1].

The precise extent of food-borne diarrhoeal diseases in young children is not documented although it is estimated that there are approximately 1.3 thousand million episodes and almost 5 million deaths in children younger than 5 years each year[2]. In 1990, over 3 million children were reported died[3]. Knowledge of the specific pathogens that cause diarrhoeal diseases and their epidemiology is critical for the implementation of specific intervention strategies[4]. In a review[1], Esrey and Feachem concluded that there are three main practices in food handling which increase the risk of food borne diseases. First is preparing food several hours before consumption and storing it at ambient temperature that favors microbial growth and formation of toxins. Second I s insufficient cooking or reheating of preserved food. Third, is preparing and serving food in contaminated utensils. Hands, water, utensils, feeding bowls, raw ingredients and the surrounding environment are potential sources of pathogens in infant foods. In areas where there is no organized sewage disposal, contamination of food invariably makes the weaning period most hazardous particularly with respect to diarrhoeal disease. Bryan[5] found out that 44% of dishes prepared in rural Kenya were unsafe hygienically due to contaminated cooking and feeding utensils. The incidence of food contamination also increased among families where mothers leave children in the care of child/minders or siblings[6].

Since storage time invariably increases bacterial load, it therefore becomes necessary to know the growth pattern of pathogens during storage of weaning food since it is possible that these microorganisms may gain access to these foods before, during or after preparation. Result of such investigation can be used to access the risk of such pathogens occurring in infant food and further educateaged<2years using microbial load as or basis and hence mothers or child handlers on adequate

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hygiene procedures.This work was therefore carried out to evaluate the bacteriological quality of weaning food given to children evaluate the hygiene standards of those preparing and feeding the children their foods. It will also serve as a means of education to child handlers and may go a long way in reducing childhood morbidity.

MATERIALS AND METHODS

Collection of food samples: The study was carried out in Umuahia metropolis, Abia State, Nigeria. A study population of one hundred and fifty households was randomly pooled for the survey. Out of these, thirty households refused to have their infants' food given out to a stranger. The remaining one hundred and twenty accepted skeptically. Three day care centres were also approached with the proprietress of one declining. Altogether one hundred and fifty three different weaning foods were sampled. The households were visited without prior information to avoid deliberate improvement of hygiene.

All food samples were collected in sterile disposable containers (Sterilin). Each sample was properly identified with a number code, subject name, type of food, condition of food (fresh or reheated) and time lag between cooking/heating and feeding (if known). Samples were sent to the laboratory in a cold box containing ice-blocks within 2 hrs. Table 1 shows the different types of food collected.

Bacteriological analysis: Approximately I g. of each the food sample was weighed out into a sterile Bijou bottle on a Mettler balance. The sample was crushed with sterile glass rod, blended in 2 mL of sterile saline and serially diluted. Triplicate plates of nutrient agar and MacConkey agar were inoculated each with 0.1 mL of each dilution and spread plated over the agar surface with a sterile (bent) glass rod. The plates were examined after 24 hrs of incubation at 37°C. Colonies were counted and identity was confirmed by standard bacteriological methods[7].

Growth study of *Escherichia coli* and *Staphylococcus aureus*: Thefateof twotestisolates, *E. coli and S. aureus* in two common weaning foods namely pap and rice was examined. Colonies of overnight cultures of isolates were suspended in sterile normal saline and the turbidity adjusted to match that of Mcfarland standard (10^8cfu mL^{-1}) and serially diluted to a concentration of 10^4cfu mL^{-1}.

The food samples (pap and rice) were purchased from Umuahia market and prepared aseptically. The rice sample was crushed into tiny bits using sterile glass mortar while the Pap was allowed to cool to about 35°C. Approximately 1g of the food samples was added to different test tubes and diluted with 9 mL of sterile water, respectively. Each food sample was subsequently inoculated with 1 mL of test isolates to obtain 10^4 cfu mL^{-1}[8]. A 0.1 mL of samples was immediately withdrawn and incubated for 24 hrs at 37°C on MacConkey agar and blood agar for E. *coli* and *Staph aureus* respectively. Subsequently the samples were withdrawn at 6 h interval up to 24 h and incubated to determine number of viable pathogens. Also the pH of the samples were taken at the various intervals.

Statistical analysis: Analysis of variance (ANOVA) using Statistical Package for Social Sciences (SPSS), Fishers least significant Difference (F-LSD) and student's statistics[9] for comparing the geometric means of becterical counts.

Table 1: Bacterial contamination (geometric mean count, GMC) in relation to type of food

Type of food	Frequency (%)	GMC±SD (log_{10} cfu mL^{-1} or g)
Pap (Akamu)	31 (27.3)	5.64±0.63
Garri/soup	16 (17.5)	5.59±0.79
Rice	26 (17.0)	5.77±0.49
Beans	11 (7.2)	6.00±0.49
Agidi	11 (7.2)	5.88±0.14
Yam/stew	7 (4.6)	5.56±0.54
Beverages	10 (6.5)	4.49±0.25
Moi-moi	13 (6.5)	6.14±0.23
Dry cereal	11 (7.2)	3.92±0.39
*Others	17 (11.1)	5.81±0.51

* Spaghetti and indomie

Table 2: Bacterial contamination (GMC) distributed according to condition of food

Food condition	Frequency (%)	GMC±SD (log_{10}cfu mL^{-1} or g)
Freshly cooked	111 (72.5)	5.35±0.78
Reheated	22 (14.4)	5.35±0.78
Overnight reheated	20 (13.1)	6.13±0.24

Table 3: Bacterial contamination (GMC) in relation to duration of storage

Time interval between cooking and serving (in h)	Frequency (%)	GMC±SD (log_{10} cfu mL^{-1} or g)
<1h	48(38.7)	4.85±0.76
1-3h	23(18.5)	5.41±0.65
>3h	53(42.7)	6.00±0.39

Table 4: Bacterial contamination (GMC) in relation to who prepared and served the food

Prepared/served by	Frequency (%)	GMC ± SD (log_{10} cfu mL^{-1}or g)
Mother/mother	53 (34.6)	5.11±0.71
Mother/maid	6 (3.9)	5.77±0.44
Mother/nanny	25 (16.3)	5.95±0.53
Mother/sibling	8 (5.2)	5.87±0.62
Maid/maid	39 (25.5)	5.95±0.60
Maid/nanny	4 (2.6)	5.72±0.62
Sibling/sibling	12 (7.8)	5.57±0.58
Maid/mother	2 (1.3)	6.02±0.30
Nanny/nanny	4 (2.6)	3.90±0.13

Table 5: Comparison of bacterial counts (GMC) on food of children taken to daycare centre and those left at home

Location	Frequency (%)	GMC±SD (log_{10}cfu mL^{-1} or g)
Home	120 (78.4)	5.53±0.74
Daycare Centre	33 (21.6)	5.68±0.84

*Nanny served children taken to day care centres

Table 6: Types of bacterial isolates

Isolates	Frequency (%)
Staphylococcus aureus	20 (50)
Escherichia coli	12 (30)
Bacillus cereus	2 (5)
Streptococcus sp.	6 (15)

RESULTS

The types of weaning foods investigated are shown in Table 1. The different food samples categorized into 10 groups were obtained from 120 households and two day-care centres. Altogether bacterial counts ranged from 3.92±0.39 to 6.14±0.23 $\log_{10}$ cfu per mL^{-1} or g of food sample. Statistical showed significant difference in counts between types of foods (F_{cal} = 21.19, $p<0.05$).

Table 2 shows the bacterial counts obtained from freshly prepared foods, reheated foods and food left overnight. Overnight reheated foods had significantly higher counts than the others (F_{cal} = 19.89; $p<0.05$).

The time interval between preparation of food and administration of food (Table 3) to the infants ranged from <1 h to>3 h. It was noted that bacterial load increased significantly with increase in storage time. The mean values ranged from 4.85±0.76 to 6.0±0.39 $\log_{10}$ cfu per mL^{-1} or g (F_{cal} = 45.13; $p<0.05$).

The level of hygiene was evaluated by assessing the extent of contamination from persons who prepared and administered the food. The highest mean (6.02±0.30$\log_{10}$ cfu per g or mL) count was with food prepared by maid and served by mother (Table 4; F_{cal} = 10.76; p<0 .05).

Bacterial counts between food of children taken to daycare centre were compared to those who stayed at home (Table 5). Although the home given food showed lower mean count as compared to that of day care centers, there was no significant difference (F_{cal} = 0.96, $p>0.05$). Out of the 40 randomly selected isolates, 20 (50%) were identified to be *Staphylococcus aureus*, 12 (30%) were *Escherichia coli*, 2 (5%) were Bacillus *cereus* and 6 (15%) were identified as *Streptococcus* sp. (Table 6).

Figure 1 and 2 show the increase in numbers of surviving enteropathogens and the pH level in the prepared food samples of pap and rice, during 24 hrs. at 37°C. Both *E. coli* and *S. aureus s.aureus* survived in the food respectively while that of rice dropped from 6.82-5.21 and 5.9-5.2, respectively for the *E.coli* and *S. aureus* (Fig. 2). A count of *S. aureus* was slightly higher than *E. coli* in both food samples. *E. coli* and *S. aureus* were not inoculated in the control

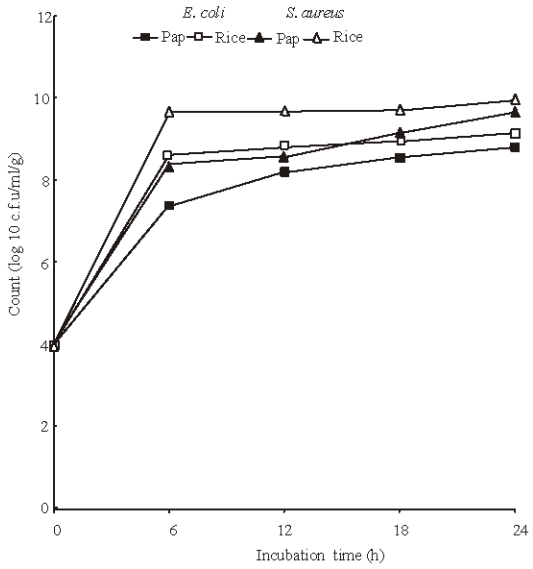

Fig.1: Growth of *E. Coli* and *S.aureus* in pap and rice

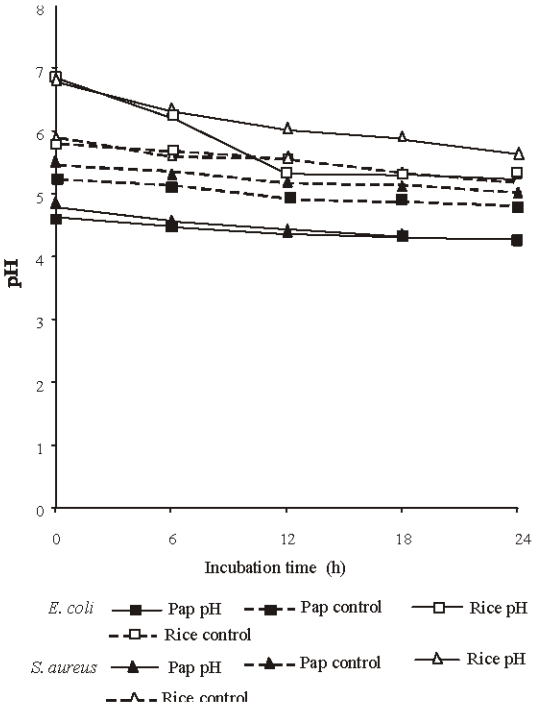

Fig. 2: pH of the food samples innoculated with the test bacteria

DISCUSSION

Weaning period is the most critical stage in child development because it is the time that the child is gradually introduced to solid diet. At this time the immunity is still developing and most children exposed to unhygienic standards frequently suffer from infantile diarrhoea[10]. The main factors, which determine food hygiene, include handling, preparation techniques and

storage practices. These are generally evaluated from the level of bacterial contamination[1]. In this study bacterial count was used as a measure of hygienic standard of food given to children<2years. This is because the impact of diarrhoea and associated malnutrition is greatest among samples within 24 hrs. *E. coli* in pap increased from 1.0×10^4 to8.8×10^8 cfu mL^{-1} or g while *S. aureus* increased from 1.0×10^4 to 9.7×10^8 cfu mL^{-1} or g (Fig. 1).

Altogether there was a decrease in the pH of the food samples during incubation, with pap dropping from 4.62-4.26 and 4.76-4.25 for *E. coli* and *S. aureus* children in this age bracket[1]. From the result, food given to children was highly contaminated to levels of 10^3 to 10^7 for the various food samples. The level of contamination varied with the type of food tested. Moi-moi and beans had higher bacterial counts (6.14±0.23 and 6.0±0.33 $\log_{10}$ cfu per g or mL) than the other foods. These foods are highly proteinaceous and due to their high nutritional content may serve to enhance the proliferation of these organisms. Dry commercial baby foods such as custard, cerelac etc. had a reduced microbial load probably because dry ingredients are subject to fewer risk of contamination and do not facilitate bacterial growth as readily as wet preparation. Also these cereals were given almost immediately after preparation thereby ensuring safety against microbial proliferation. Regardless of the fermentative nature of the food, pap was highly contaminated. It might be possible that the food may have been contaminated from the cooking or feeding utensils.

This is because the viscous nature of pap prevents it from being cooked for a long period. Due to the short period of cooking some contaminating pathogens may not be killed.

As would be expected, bacterial counts were higher in reheated and left overnight foods than in freshly prepared foods. However, some freshly prepared foods were highly contaminated as a result of long time lapse between cooking and serving. (or sample collection).

Bacterial counts were high in foods stored>4 hrs. Due to the economy of the country and limited fuel supply, most mothers who work to supplement the family needs prepare food in bulk and leave in care of childminders or siblings. Sometimes, these foods (some of which are meant to be eaten immediately after preparation like pap) stay for long hours before being served to the infants without reheating. These foods may get contaminated during or after cooking. Storing for long period at ambient temperature therefore becomes health threatening as a single bacterium can multiply to 500 million bacteria in 10 hrs[11].

Foods prepared and served by mother gave the lowest mean count with the exception of one prepared and served by nanny. It is important to note that food given by nanny in the day-care centers were mostly 'dry' foods which were reconstituted anytime the child was to be fed thus minimizing the risk of contamination. Food prepared and served by maid was highly contaminated. It is possible that simple hygiene procedures like handwashing, cleaning of feeding bottles is sometimes absent among these childminders and it is possible that they contaminate the food during handling and preparation Although foods given to children left at home showed lower bacterial count than those taken to day-care centers did, statistically there was no significant difference ($p>0.05$). The type of food most children taken to day care centers are given may explain this. For convenience purposes most proprietors insist on foods like Cerelac, Nutrend which they prepare and serve to infants on demand.

The ability of enteropathogenic *E. coli* and *S. aureus* to survive in different environmental conditions has been well documented. The ability of pathogenic microorganism to survive in fermented food is controversial as diverse authors have different views. Studies carried out[12] show that lactic acid fermented foods with pH of <4 suppress the growth of foodborne pathogens. However, in a similar study[13] enteropathogenic *E. coli*, *S. aureus*, *Salmonella* sp. resisted or adapted to the growth inhibitory conditions in lactic fermented products with a pH<4.0.

In this study, *E. coli* and *S. aureus* both survived and multiplied in both pap and rice (most common weaning foods) despite the low pH of pap. Bacteria counts of both organisms increased from 10^4 to above 10^8 after incubation. The growth was exponential within the first 6 h after which it reduced, this might be explained using the work of Foster and Hall which reported that some microorganisms produce an acid-tolerance response system that protects them against severe acid stress for longer period[14]. This phenomenon and the fact that enteropathogenic *E. coli* have a double cell membrane making them less sensitive to growth inhibiting factors offers an explanation for the relatively long survival of the organism in pap. From this study therefore we can infer that acidity and pH cannot be relied upon to control the growth of *E. coli* and *S. aureus* in pap.

The fact that growth was exponential within the first six should serve as warning to those who handle children since most weaning foods are prepared in bulk and stored for>4 h. This food if contaminated becomes a rich medium for microorganisms, encouraging their growth and eventual production of toxin. Although toxin production was not evaluated in this study, storage at ambient temperature for up to four hours is known to encourage their production.

Since the nutrition of infants and young children depend closely on the education of their mothers on food safety, a programme to educate mothers on food safety

principles should therefore be considered to be an integral part of every primary health care system. This should be incorporated into national infant feeding or food and nutrition programmes.

CONCLUSION

The hazards of inadequate water supply and sewage disposal are nationwide and the effects on health are serious, such as increased prevalence of severe gastroenteritis[15]. Therefore, Hazard Analysis Critical Control Point which is a system or procedure by which preventive or control measure which can eliminate, prevent or minimize hazards associated with food production should be studied. This will help to identify the risk points encountered during food production and administration and thereby alert food handlers.

REFERENCES

1. Esrey, S.A. and R.G. Feacham, 1989. Interventions for control of diarrhoeal diseases among young children: Promotion of food hygiene. Geneva: Control of Diarrhoeal Disease Programmed. World Health Organization, 1-22. (WHO/CDD/89, Research Report No. 30).
2. Claeson, M. and M.H. Merson, 1990. Global progress in the control of diarrheal diseases. Pediatric Infectious Disease J., 9: 345-55.
3. World Health Organization, 1993. Food Safety Unit. Contaminated food: A major source of diarrhoea and associated malnutrition among infants and young children. Geneva: WHO Working Group on Infant Feeding 4 (Facts about infant feeding issue No. 3). pp: 4.
4. Tawfeek,H.I.,N.H. Najim and S. Al-Mashikhi 2002. Studies on diarrhoeal illness among hospitalized children under 5 years of age in Baghdad during 1990-97. Eastern Mediterranean Health J., 8: 127-135.
5. Bryan, F., 1988. Hazard analysis of food prepared by migrants living in a new settlement at the outskirts of Lima, Peru. J. Food Protect., 51: 314-328.
6. Iroegbu, C.U., H.N. Ene-Obong, A.C. Uwaegbute and U.V. Amazigo, 2000. Bacteriological quality of weaning food and drinking water given to children of market women in Nigeria: Implications for Control of Diarrhoea. J. Health Popul. Nutr., 18: 157-162.
7. Collins, C.H. and P.M. Lyne, 1970. Microbiological Methods Butterworths, London, pp: 403-408.
8. Feresu, S. and H. Nyati, 1990. Fate of pathogenic and non-pathogenic *Escherichia coli* stains in two fermented milk productions. J. Applied Bacteriol., 69: 814-821
9. Steele, R.G.D. and J.H. Torrie, 1960. Principles and procedures of statistics with reference to biological sciences Mc Graw Hill Book Co. Inc. New York.
10. Black, R.E., K.H.G. Lopez de Romana, Brown, N. Bravo, O.G. Bazalar and H.C. Kanashiro, 1989. The incidence and aetiology of infantile diarrhoea and major routes of transmission in Huascar, Peru. Am. J. Epidemiol., 189: 785-799.
11. Motarjemi, P., F. Kaferstein, G. Moy and F. Quevedo, 1993. Contaminated weaning food: a major risk factor for diarrhoea and associated malnutrition Bulletin of the WHO., 71: 79-92.
12. Nout, M.J.R., F.M. Roumbouts and A. Havellar, 1989. Effect of accelerated natural lactic fermentation of infant foods ingredient on some pathogenic microorganisms. Intl. J. Food Microbiol., 8: 351-361.
13. Mensah, P., A.M. Tomkim, B.S. Drusar and T.J. Harrison, 1991. Antimicrobial effect of fermented Ghanaian maize dough. J. Applied Bacteriol., 70: 203-210.
14. Foster, J.W. and H.K Hall, 1991. Induced pH homeostasis and the acid tolerance response of *Salmonella typhimurium*. J. Bacteriol., 173: 5129-5135.
15. Food and Agriculture Organization of the United Nations, 1995. Evaluation of food and nutrition situation in Iraq. Executive summary. Ecology of Food and Nutrition, 35: 105-110.