removed by prior streaking on selective plates, resulting in a sample containing little or no target DNA for molecular analysis.

In summary, the results of this study indicate that direct DNA release through bacterial lysis, combined with the addition of ≥2 μg BSA, represents an effective system for the RT-PCR detection of MRSA from screening swabs, and potentially could be applied to similar pathogen transport systems. In addition, the authors suggest that equipment used for specimen collection, transport and processing should be accessed and optimised for RT-PCR assays. □

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Prevalence of *Salmonella typhi* among food handlers from bukkas in Nigeria

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Salmonella enterica serovar *typhi* is the aetiological agent of typhoid fever and causes an estimated 16.6 million cases and 600,000 deaths worldwide each year. A syndrome similar to typhoid fever is caused by paratyphoid serotypes of *Salmonella* species.[1]

Salmonella typhi is transmitted through food or water contaminated with faeces from infected persons, persistent excretors or from chronic asymptomatic carriers who handle food. Humans are the only host for *S. typhi* and there are no known environmental reservoirs.[1,2] Typhoid fever remains a major public health problem in many developing countries.[2] It is a sporadic disease in developed countries, occurring mainly in travellers returning from overseas. It can also produce the occasional point-source epidemic.[3]

In endemic areas, identified risk factors for the disease include eating food prepared outside the home (e.g., ice cream, flavoured iced drinks) by street vendors,[4] drinking contaminated water,[5] close contact with an infected person,[6] poor housing with inadequate facilities for personal hygiene,[7] and the recent use of antimicrobial drugs.[4]

Typhoid fever is among the major widespread diseases affecting the population in Nigeria and has been rated eighth among these common infections.[8] Nigeria, like many other tropical and developing countries, has been described as an endemic zone for typhoid fever.[9,10]

In Nigeria, transmission of typhoid fever occurs all year round but rates are slightly higher in April and July, coinciding with the height of the hot, dry season and the onset of the rainy season, respectively.[11] The highest number of cases of typhoid fever are recorded during the rainy season in south-east Nigeria.[10] Typhoid fever has been reported in all age groups and classes in Nigeria.[12] Owing to the irregular nature of bacterial shedding, several samples

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Table 1. Antibiotic sensitivity pattern of *Salmonella typhi* and other isolates obtained from stool and water samples.

	AM (10 μg)	COT (25 μg)	CHL (30 μg)	NAL (30 μg)	OFL (30 μg)	CIP (5 μg)	TET (30 μg)
S. typhi (*n*=3)	R	3 (100%)	1 (33.3%)	3 (100%)	3 (100%)	3(100%)	1 (33.3%)
S. cholerasuis (*n*=1)	R	R	R	1 (100%)	1 (100%)	1(100%)	1 (100%)
S. enteritidis (*n*=3)	R	2 (66.7%)	R	3 (100%)	3 (100%)	3(100%)	2 (66.7%)

AM: ampicillin, COT: cotrimoxazole, CHL: chloramphenicol, NAL: nalidixic acid, OFL: ofloxacin, CIP: ciprofloxacin, TET: tetracycline.

should be examined in order to identify carriers.[13]

Up to 10% of convalescing patients with untreated typhoid excrete *S. typhi* in faeces for up to three months, while 1–4% become long-term carriers, excreting the organism for more than a year. Furthermore, up to 25% of long-term carriers have no history of typhoid. Chronic carriage is more common among women and the elderly and in patients with cholelithiasis.[14]

Excretion of *Salmonella* species by humans may continue long after clinical cure. Approximately 5% of patients cured of typhoid fever remain carriers for months or even years. Antibiotics are usually ineffective in eradicating carriage (even if the organism is susceptible) because the site of carriage may not allow penetration by the antibiotic agent.[15,16]

The main aim of this pilot study is to establish the prevalence and plasmid profile of *S. typhi* in healthy food handlers in bukkas (local fast-food restaurants) in Lagos. This is the first report on typhoid carriage among this group in Nigeria.

A total of 53 stool samples and 36 water samples (used in the preparation of foods and for drinking) were collected from different apparently healthy food handlers in June and July 2006. The samples were collected in leak-proof containers and were transported to the molecular biology and biotechnology laboratory of the Nigerian Institute of Medical Research within an hour of collection for processing. The food handlers were not on antibiotic therapy.

The samples comprised 13 stools and 10 water samples from the Oshodi area, three stools and two water samples from Ojuelegba, nine stools and four water samples from Yaba, 11 stools and six water samples from Ketu, and 17 stools and 14 water samples from the Mafoluku area. All samples were collected in duplicate.

A loop of stool sample was inoculated aseptically in a McCartney bottle containing 9 mL selenite F broth. One water sample was measured aseptically into a McCartney bottle containing 5 mL selenite F broth. These were incubated at 37°C for 24 h and then streaked on Salmonella–Shigella agar and incubated for 24 h at 37°C. The isolates were identified and characterised using the method of Cowan.[16]

The NCCLS disc-diffusion method[1] was used for antimicrobial susceptibility testing. Antimicrobial discs tested included ampicillin (10 μg), cotrimoxazole (25 μg), chloramphenicol (30 μg), gentamicin (10 μg), nalidixic acid (30 μg), ofloxacin (30 μg), co-amoxyclav (30 μg) tetracycline (30 μg) and ciprofloxacin (5 μg).

Plasmid DNA isolation was performed using the alkaline lysis method of Birnboim and Doly.[17] Results were analysed statistically using the χ^2 test.

Salmonella spp. (three *S. typhi* [5.7%], three *S. enteritidis* [5.7%] and one *S. choleraesuis* (1.9%]) were recovered from seven (13. 2%) of the 53 stool samples processed. None of the water samples yielded a *Salmonella* sp. Table 1 shows the antibiotic sensitivity pattern of the isolates. Table 2 shows the different molecular weights of plasmids carried by the *Salmonella* species isolated.

In this study, the prevalence of *S. typhi* among healthy food handlers in Lagos was 5.7%. It has been reported that food vendors are carriers of a variety of bacterial enteropathogens, including *S. typhi*.[18] This study confirms this finding.

Inadequate personal hygiene can facilitate the transmission of these pathogens via food to humans. The serving stage is a critical point in the street food industry[19] because enteropathogens can survive on the hands for over three hours.[20] Therefore, carriers of *S. typhi* and other *Salmonella* species who handle food in bukkas may pose a risk to their customers.

The major sources of water for the food handlers were public tap water and bore hole water. According to Nickerson and Sinskey,[21] *Salmonella* spp. do not multiply significantly in the natural environment, but they can survive for weeks in water and for several years in soil if

Table 2. Molecular weight of plasmids harboured by *Salmonella typhi* and other *Salmonella* species.

C/N	Sample number	Isolate	Number of plasmids	Molecular weight (kb)
2	S03	*S. typhi*	2	14.125, 35.481
3	S10	*S. enteritidis*	0	None
4	S22	*S. typhi*	1	14.125
5	S37	*S. typhi*	2	14.125, 35.481
6	S43	*S. cholerasuis*	3	14.125,56.234, 35.481
7	S45	*S. enteritidis*	3	14.125,56.234, 35.481
8	S51	*S. enteritidis*	2	14.125, 35.481

temperature, humidity and pH are favourable. It can be concluded that the carriers of *S. typhi* and other *Salmonella* species probably did not acquire the microorganisms from the water used for drinking and in food preparation.

Ampicillin, chloramphenicol and tetracycline are are used as first-line treatment in typhoid fever. The high resistance to these agents corroborates the findings of Olukoya *et al.*[22] and Smith *et al.*,[23] who showed that easy access to drugs has led to increasingly resistant strains.

The findings of this study agree with those of Chukwuani *et al.*,[24] who showed that the quinolones fleroxacin and ciprofloxacin possess greater potential and benefits as first-line therapy for the management of typhoid fever in this environment.

In this study, the three isolates of *S. typhi* that were resistant to ampicillin harboured a plasmid of low molecular weight (14.125 kb), while the two isolates of *S. typhi* resistant to tetracycline harboured plasmids of high molecular weight (35.4 kb).

Plasmids in salmonellas may code for antibiotic resistance; however, this study did not ascertain whether or not the antibiotic resistance was plasmid-mediated.

The rate of resistance or susceptibility to a particular antibiotic depends not only on the plasmid harboured but also on the rate of exposure to these antibiotics. Therefore, the high rate of multidrug resistance demonstrated by these isolates would seem to make a case for the appropriate use of drugs in this environment.

It should be noted that the number of isolates studied here was low; however, these preliminary findings indicate the need to implement environmental laws covering food preparation and sanitation.

The high level of multidrug-resistant *Salmonella* species indicates a need for appropriate measures in managing typhoid fever and its complications. Furthermore, the antibiotics used in health institutions to control enteric infections should be reviewed periodically.

In view of the high incidence of typhoid fever and antibiotic resistance in Nigeria, typhoid fever vaccination programmes[25] should be implemented, as the new-generation vaccines have been found to be very effective and relatively inexpensive. However, the fluoroquinolone antibiotics remain the drug of choice for the management of typhoid fever in this environment. □

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