



DETECTION OF *SALMONELLA* SPECIES AMONG PATIENTS WITH SUSPECTED CASES OF TYPHOID FEVER IN WUDIL GENERAL HOSPITAL

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ABSTRACT

The prevalence of *Salmonella typhi* infection among patients with reported cases of typhoid fever in Wudil general hospital was investigated by serological examination of blood samples and by culture method of stool collected from patients attending the Hospital. A total of 140 each of blood and stool samples were analyzed using the method of Widal slide agglutination test and selective cultivation on *Salmonella-Shigella* (SS) agar respectively. Out of this, 112 (80%) of the samples were found to be positive, while 28 (20%) of the samples were found to be negative for both Widal agglutination test and culture on SS agar. In the study, males were more infected by *S. typhi* than females. The presence of these organisms among patients with symptoms of typhoid fever is a threat to public health especially if immunocompromised patients are involved. Based on the research findings it can be seen that substantial numbers of reported patients were exposed to predisposing factors for typhoid fever infection. Hence it is recommended that action should be expedited in the provision of safe drinking water to the public; discourage defecation in an open area and the improvement of personal hygiene.

Keywords: *Salmonella typhi*, Typhoid fever, Detection, Patients,

INTRODUCTION

Salmonella belongs to the family of the bacteria Enterobacteriaceae. The family is a large heterogeneous group of gram-negative, non-spore forming rods, facultative anaerobic, catalase positive and oxidase negative, whose natural habitat is the intestinal tract of human and animals. Other members include *Escherichia*, *Shigella* and *Enterobacter* (Adam *et al.*, 2007) *Salmonellae* are generally motile with peritrichous flagellation that cause infection even at low dose (Tan *et al.*, 2019). Typhoid and paratyphoid fevers are bacterial infections transmitted by the ingestion of food or water contaminated with faeces of an infected person containing the bacterium *S. typhi* or *S. paratyphi* (Wain *et al.*, 2014; Shu'aibu *et al.*, 2018). The organism enters through the gastrointestinal tract and spreads through the circulatory system (bacteremia), inflaming the lining of the small and large intestine (intestinal mucosa). The initial manifestation of this disease include sudden onset of a high fever, a headache and nausea, loss of appetite, diarrhea and enlargement of the spleen are also observed (Chin, 2016). Severe cases can lead to delirium or coma, and may be life threatening (Humphrey, 2004).

Another strain, *S. paratyphi* A remains uncommon in Africa (Marks *et al.*, 2017). If treatment is started early, the injury produced by typhoid fever is reversible and restricted. This results in a lower mortality rate of less than 1% among treated persons who possess an antibiotic-susceptible strain of *S. typhi*, making the outcome and prognosis for patients a positive one

(Agwu *et al.*, 2009).

Typhoid fever infections are one of the diseases that require public concern, most especially in the developing countries. It remains a major cause of enteric disease and a significant public health problem (Crump and Mintz, 2010) and is now very prevalent in developing world of which Nigeria belongs (Parry, 2015). It is therefore very imperative to study the disease in order to put it under check and thus curtailing its devastating effects. This study was aimed at finding out the incidence of typhoid fever in Wudil General Hospital, Kano State, Nigeria. The outcome of the work would increase awareness about typhoid fever, treatment and preventive measures to limit the occurrence of the disease in the community and the general public.

MATERIALS AND METHODS

Study Area

The area of this study Wudil town is located in the south east of Kano, along Kano–Maiduguri Road, on a Latitude 110°N and 480°N and Longitude 80°E 510°E. Wudil Local Government Area of Kano State covers about 458km² and has approximately 262,400, inhabitants according estimated population in 2016 by National Population Census (NPC/ National Bureau of Statistics, 2006).

Ethical Consideration

Ethical approval to carry out this study was obtained from the Kano State, Health Management Board after which a written

consent was obtained from each patient before sample collection.

Sample Collection

The blood and stool samples (n=140 for each) used for this research were collected from Hematology Department of Wudil General Hospital on a daily basis in a sterilized container and taken to the laboratory for processing and analysis. Two millimeters (2ml) of blood samples were collected intravenously using syringe and put inside test-tube for centrifugation at 5,000 rpm for 5 minute to separate the serum from the red blood cells. The stool samples were collected in a sterile sampling bottle for analysis.

Sample analysis

For the blood sample, Widal Slide Agglutination titres Test Method were employed: ANTEC febrile antigen kit (United Kingdom) was used for the Widal test. The standard Widal slide agglutination test method was used to screen blood sample for *Salmonella* infection. Two different stained antigens (somatic 'O' antigens and flagella 'H' antigens) were used in rapid agglutination test method. Using a pipette, a drop of undiluted serum was dispensed into a row of eight (8) places on the slide. Then the reagent bottles were rigorously shaken and a drop of the undiluted antigen suspensions were added to each serum drop that marked as *Salmonella typhi* 'O', *S. Paratyphi* 'OA', *S. Paratyphi* 'OB', *S. Paratyphi*, 'OC', and *Salmonella typhi* 'H', *S. Paratyphi* 'HA', *S. Paratyphi* 'HB', *S. Paratyphi*, 'CH', These were thoroughly mixed with the aid of stirring stick on the slide and gently rotated, then the reaction mixtures were observed after a minute as reported by Kabiru *et al.* (2018). Meanwhile, the stool samples were inoculated into Salmonella and Shigella agar. The inoculated SS agar was then incubated for overnight. The non-lactose fermenting colonies observed with dark centers indicating hydrogen sulfide production

depicting the prevalence of *S. typhi*.

RESULTS AND DISCUSSION

Table 1 shows the prevalence of *Salmonella typhi* infections in relation to gender, from 140 subjects tested for *Salmonella typhi* from the Wudil General Hospital, 78 (55.7%) females and 62 (44.3 %) males. The result showed that 112 (80%) of the samples analyzed for *Salmonella* agglutinations were sera positive, while 28 (20%) were sera negative. It also showed that 57 (40.7%) of the sera from males were positive and 55 (39.3%) of the sera from females were also positive. In this present study, 112 (80%) of the 140 (100%) blood samples analyzed were positive for Widal reaction. Meanwhile 80% (112/140) of the stool samples showed transparent colonies with dark centers on SS agar, a positive biochemical reaction for *S. typhi*. This indicates a high prevalence of *Salmonella* infection among the patients presenting with symptoms at Wudil general Hospital. This study agrees with the observations of Outi *et al.* (2009) and Adeleke *et al.* (2006) in a similar study on Widal reaction. Also, 112 patients out of 140 were found positives for *Salmonella typhi*, given a prevalence level of 80%, an indication of high prevalence of the disease in the study area, the value is a little bit low compared to 81.2% obtained in a study at College of Health Sciences Bingham University Teaching Hospital in Karu, Nassarawa State, Nigeria as reported by Ramil (2012). Also, Okonko, (2010) observed high prevalence of 80.1% in University of Ilorin Teaching Hospital, Ilorin, Kwara State. Furthermore, the high prevalence of *Salmonella* is common throughout the globe as it is the most widespread foodborne pathogen with higher prevalence than *Vibrio spp.* and *Staphylococcus aureus* (Zhang *et al.*, 1998; Shu'aibu *et al.*, 2019). This may be as a result of improper food handling practices and/or poor personal hygiene.

Table 1: Incidence of *Salmonella typhi* infections in relation to gender

Sex	No. of Sera Tested (%)	No. of Sera Positive (%)	No. of Sera Negative (%)	Stool sample Positive (%)
Female	78 (55.7)	55 (39.3)	23 (16.4)	55 (39.3)
Male	62 (44.3)	57 (40.7)	5 (3.6)	57 (40.7)
Total	140 (100)	112 (80)	28 (20)	112 (80)

Table 2 indicates the distribution of *Salmonella* agglutination titres in 62 male subjects and 78 female patients in Wudil General Hospital, Kano Nigeria. This is also in agreement with the findings of Adeleke *et al.* (2006) in which 62 male patients, the *Salmonella* 'O' titres were higher than those of the 'H' whereas in 78 female patients, *Salmonella typhi* 'H' titres were higher than those of 'O'. This differs from what was reported in a similar study by Ibekwe *et al.* (2008) where 82 apparently normal males had higher titre of *Salmonella typhi* 'H' titres and 118 apparently normal female had higher *Salmonella typhi* 'O' titres. Agglutinins to *S. typhi* were the most prevalent among the

sera tested at various dilutions in both males and females. In the study, 57 (91.9%) had higher titres for *S. typhi* 'O' titres and 54 (87.1 %) for *S. typhi* 'H' titres than in the female with 54 (69.2%) for the *S. typhi* 'O' titres and 72 (92.3%) for the *S. typhi* 'H' titres. Agglutinin level for the typhoid and paratyphoid group tested in this study were evidently very frequently found in the sera of the subjects. The levels of agglutinin of *salmonella paratyphi* C-H 17 (27.6%) and *salmonella typhi* C-O 18(29.0%) in the males were however, low. Titres above 1:80 occurred in more than 91.9% and 92.3% of the male and female samples respectively and as such could be used in the presumptive

diagnosis of enteric fevers in the study area. The Widal test reaction involves the use of bacterial suspensions of *S. typhi* and *S. paratyphi* 'A' and 'B', treated to retain only the 'O' and 'H' antigens. These antigens are employed to detect corresponding antibodies in the serum of a patient Suspected of having typhoid fever. The IgM somatic O antibody appears first and represents the initial serologic response in acute *salmonella* infection,

while the IgG flagella H antibody usually develops more slowly but persists for longer time (Olopoenia *et al.*, 2000; Hoffman *et al.*, 2006; Washington and Henry, 2012). Also in this study, male patients were more Widal test positive than the female patients. This is probably a reflection of different eating habits and level of personal hygiene.

Table 2: Distribution of *Salmonella* agglutination titres among male and female from the study

<i>Salmonella</i> Antigen	No of sera tested	No of Widal Positive (%)	No of Widal Negative (%)
<i>S. paratyphi</i> AO	62 male 78 female	23 (37.1%) 25 (32.1%)	39 (62.9%) 53 (67.9%)
<i>S. paratyphi</i> BO	62 male 78 female	19 (30.6%) 28 (35.9%)	43 (69.4%) 50 (64.1%)
<i>S. paratyphi</i> CO	62 male 78 female	18 (29.0%) 24 (30.8%)	44 (71.0%) 54 (69.2%)
<i>S. typhi</i> O	62 male 78 female	51 (82.3%) 54 (69.2%)	11 (17.7%) 24 (30.8%)
<i>S. paratyphi</i> AH	62 male 78 female	20 (32.3%) 24 (30.8%)	42 (67.7%) 54 (69.2%)
<i>S. paratyphi</i> BH	62 male 78 female	21 (33.9%) 29 (37.2%)	41 (66.1%) 49 (62.8%)
<i>S. paratyphi</i> CH	62 male 78 female	17 (27.6%) 22 (28.2%)	45 (72.6%) 56 (71.8%)
<i>S. typhi</i> H	62 male 78 female	54 (87.1%) 72 (92.3%)	8 (12.9%) 6 (7.7%)

Table 3 reveals the prevalence of *Salmonella typhi* infections in relation to age, it shows the result obtained from 140 (100%) subjects tested for *Salmonella typhi* in relation to age in which 47 (33.6%) were children and 93 (66.4%) adult. The result shows the prevalence of positive sera (*Salmonella typhi* agglutinin titres) in relation to age of the patients tested. It showed that 76 (54.3%) of the samples analyzed for *Salmonella typhi* agglutinin titres were sera positive, while 64 (45.7%) were sera negative, it also showed that 50 (35.7%) of the sera from adult were sera positive and 26 (18.6%) of the sera from children were also sera positive Table 3. This incidence rate is similar to the reports of Onabowale and Ogunbiya, (1996) and Kabiru *et al.*, (2012) which reported that the peak age group for typhoid infection was 21 – 30 years. Similarly, Ajayi *et al.* (2015) found high prevalence among males of age group 10-25 years. This incidence could be attributed to the life pattern of the age groups, which involves their active nature, engaging frequently in physical activities, consuming more foods and drinks that are hawked and probably contaminated by young people.

Table 3: Prevalence of *Salmonella typhi* in relation to age

Age	Total No Tested (%)	No of Sera Positive (%)	No of Sera Negative (%)
Children	47 (33.6%)	26 (18.6%)	21 (15%)
Adult	93 (66.4%)	50 (35.7%)	43 (30.7%)
Total	140 (100%)	76 (54.3%)	64 (45.7%)

CONCLUSION

The findings of this study shows that adults were more affected than children also; the males were affected higher than the females. The Widal agglutination test was found to be useful in

the diagnosis of *Salmonella spp.* infection. But still the *Salmonella* infection could occur irrespective of age or gender. The prevalence of the *Salmonella typhi* infection in the study was found to be 80% for both blood and stool which needs

serious measures to address the problem.

RECOMMENDATIONS

An urgent control measure should be put in place in order to avert the disease epidemic. Proper awareness about Typhoid fever should be conducted to reduce the incidence of this disease among the people living the community. Proper sanitary measures should be taken in the preparation of food and drinking water should be made safe by boiling before drinking. The environment should also be kept clean at all times especially from human waste products. Routine screening of the food handlers in the area need to be carried and infected individuals should be treated adequately after proper medical investigations to avoid indiscriminate abuse of antibiotics, which can lead to drug resistance.

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