Environment Conservation Journal 18(1&2) 121-125, 2017
ISSN 0972-3099 (Print) 2278-5124 (Online)
Abstracted and Indexed

Prevalence of typhoid in the rural community of Saharanpur (Uttar Pradesh) India

Kuldeep Rana[1] and Harish Chandra [2]✉

Received:19.12.2016 **Revised:15.02.2017** **Accepted: 08.03.2017**

Abstract

Typhoid fever in rural areas of India is the commonest problem which is associated with poor hygiene, unavailability of treated water and poor sanitation. Typhoid is not a problem of the single country; it is a global health problem. The severity of disease increase due to an increase of drug resistance in *Salmonella* spp., delay in diagnosis and administration of the appropriate antibiotic. The present study was carried out to study the prevalence of typhoid in Saharanpur District, Uttar Pradesh. The serum samples of the patient who had complain of high fever, abdominal pain, and disturbed gastrointestinal tract were investigated for Salmonella infection by Widal test in which O and H antigen were reacted with serum. The result shows that out of the 858 samples received for the typhoid detection 746 (86.9%) samples were found positive for typhoid antigen and 112 (13%) samples are nonreactive for Salmonella antigen O and H antigen. The study also focused on gender distribution of typhoid, it was found that 423 (57%) females out of 746 positive cases are positive for *Salmonella* as compared to the male which was only 323 (43%) out of 746 positive cases. Most susceptible age group was 21-30 year followed by 31-40 year. The positive blood samples are also subjected to isolation of Salmonella after drawing serum from the test tube. The clot was ruptured by sterile inoculating needle and then enrichment broth 10 ml was added and incubated at 37^{o}C for 24-48 hour. After showing turbidity one loopful of inoculums is plated on Selective media such as Hecktoen Enteric Agar, MacConkey Agar, XLD (Xylose Lysine Decarboxylase), DCA (Deoxycholate Citrate Agar). On the basis of culture characteristic and biochemical reaction, 194 *Salmonella* spp. was isolated from 746 positive samples.

Keywords: Salmonella typhi, Typhoid fever, Widal test

Introduction

Typhoid fever or enteric fever is caused by the Genus Salmonella in which the most virulent and prevalent serotype are *Salmonella enteric* serovar Typhi and serovars Paratyphi A, B and C the member of Enterobacteriaceae. More than 2500 serotype of *Salmonella* are known so far, however less than 100 serotypes are known for causing human infection. Typhoid fever is most prevalent in that region or places where there is poor sanitation and inferior quality of water supply or access to clean water is limited (Cabello and Springer, 1997). The most common route of entry of this bacterium is through the consumption of faecal contaminated water and food (Al-Quarawi *et al.*11995). Salmonella get entered into the human body and reach stomach where some of the bacteria get killed while those surviving enter the small intestine (M cells of the peyers patch) and multiplying the local tissue and through mesenteric lymph nodes it reaches to blood streams where it is taken up by macrophages that line the sinusoid of liver, bone marrow and spleen and finally reaches intestinal tract where they divide to increase their numbers. After multiplication, they again reach the blood stream and now there is onset of clinical symptoms (Everest *et al.*, 2001) Typhoid fever is a serious health threat in developing countries, and in Asia, it represents the most common cause of community acquired bacteremia. Globally, it has been estimated that there were over 21.5 million illnesses annually, resulting in more than 200000 deaths during 2000 (Crump *et al.*, 2004; Parry *et al.*, 2002). The emergence of multidrug resistance in serotype Typhi strains (Saha *et al.*, 1992; Wain and Kidgell, 2004) has further complicated the situation. Rapid accurate diagnosis and early treatment with suitable antimicrobials is essential for speedy recovery and for prevention of

Author's Address
[1]Department of Microbiology, Mewar University, Gangraar, Chittorgarh-Rajasthan
[2]High Altitude Plant Physiology Research Centre, H. N. B. Garhwal University (A Central University), Srinagar, Garhwal-(Uttarakhand), India
E-mail:hreesh5@gmail.com

complications and deaths due to this disease and also for the control of transmission (WHO, 2003).
Enteric fever is endemic in all parts of India and still constitutes a significant health hazard. The resistance of *Salmonella enterica* subspecies enteric serovar Typhi (*S.* t*yphi*) to chloramphenicol was first reported in India from Kerala, where a substantial outbreak took place in 1972 (Paniker & Vimla, 1972). Since then multidrug-resistant strains of *S.typhi* have increased rapidly into a worldwide problem (Ackers *et al.*, 2000; Madhulika *et al.*, 2004). The steadily increasing multidrug resistance in *S.* Typhi strains is a cause of great concern in India, where such strains are endemic in many parts. As per the report published by Nation health profile (2001-2016), the cases of typhoid are not controlled but the mortality due to enteric fever is significantly reduced as compared to 2001. It may be due to the mass immunization program or awareness of people towards hygiene like drinking treated water. But in few states, there is the problem of good quality and treated water. The main source of water supply is either tube well or ground water (www.cbhidghs.nic.in). The present study was carried to conduct a community based study in Saharanpur district, Uttar Pradesh to know the prevalence of typhoid in different age group and determine the most susceptible population at risk.

Material and Methods

Collection of sample: Blood samples were collected from the suspected typhoid patient coming from different places of Saharanpur District, Uttar Pradesh by venipuncture into 15 ml sterile vial without anticoagulant (EDTA) to collect serum. The tubes were placed in slanting position for clotting. After clotting, tubes containing coagulated blood were centrifuged to separate serum for Widal test and clot was used for blood culture (Chandra *et al.*, 2010).

Screening of typhoid by Widal test:
Widal test was performed as per the manufacture guideline in which O and H antigen was reacted with patient serum and observes for agglutination and the titre value was determined by using protocol provided by the Span Diagnostics. Titrevalue ≥1:160 was taken as typhoid positive (Cruickshank, 1965).

Blood culture: Blood culture of typhoid suspect was done by clot culture. After separation of serum from blood, the clot was resuspended in Tetrathionate broth (TTB) (HiMedia) a selective enrichment broth for primary isolation of Salmonella. After enrichment loopful inoculum was inoculated on different selective media for primary isolation, MacConkey's agar (HiMedia), Hecktoen Enteric Agar (HiMedia), DCA (Deoxycholate citrate agar) (HiMedia) and XLD (Xylose lysine deoxycarboxylate) (HiMedia) medium was used for purification of suspected colonies (Murray and Shea, 2004). Characterization and preliminary identification of suspected Salmonella cultures were made on the basis of morphology, cultural characteristics, and biochemical reactions (Table 2).

Identification of salmonella isolates: Suspected Salmonella isolates were identified on the basis of biochemical investigation (Cappuccino and Sherman, 2005) and further confirmed by HiSalmonella identification Kit (KB 011, HiMedia Lab Ltd).

Results and Discussion

Typhoid fever is one of the leading diseases in developing countries with increasing incidence of mortality and morbidity. The increase of antibiotic resistance cases in developing countries increases the severity of the disease. Table 1 shows the age group distribution of typhoid in different age group. The study was conducted at Baharat Diagnostic Laboratory, Deoband in 2012. Total 858 samples were collected from different places of Saharanpur District. The incidence of higher typhoid cases was reported in the age 21-30 years followed by 31-40 years. The most probable reason behind the susceptible people is in this age group is food habit and in this age group people are very fond of eating outside or street food and juices etc. Age groups between 21-40 years are the most susceptible victims (Adesunkanmi and Ajao, 1997; WHO, 2008). Similar finding was also reported by Ezeigbo *et al.* (2015) in which 400 samples was investigated for typhoid in which 126 (31.5%) males and 274 (68.5%) females, 98 (24.5%) were tested positive for *S.* Typhi using the Widal test while the blood culture method only recorded 37 (9.3%). On age-related prevalence, the age groups

between 31-40 years showed the highest prevalence rate for both methods with 23 (32.4%) for Widal test and 9 (12.7%) for blood culture method. Sex-related prevalence also showed that more males (34.9% and 11.1%) were infected with *Salmonella typhi* than females (19.7% and 8.4%) for Widal and blood culture methods respectively. Shrivastava *et al.* (2014) reported the similar finding in which the peak incidence was between 21 to 30 years, accounting for 32.90% of cases. In our study 423 (57%) females out of 746 positive cases are positive for Salmonella as compared to the male which was only 323 (43%) out of 746 positive cases (Table 1) .

Table 1. Prevalence of typhoid fever in different age group

S.No	Age Group	No. of Positive	No. of Males	No. of Females	Negative sample
1	0-10	08	04	04	112
2	11-20	104	43	61	
3	21-30	209	90	119	
4	31-40	151	60	91	
5	41-50	140	62	78	
6	51-60	46	21	25	
7	61-70	53	25	28	
8	71-80	23	15	08	
9	81-90	04	00	04	
10	91-100	08	03	05	
11	Total	746/858	323	423	

Out of the 858 samples, 746 (86.9%) were positive for typhoid antigen and 112 (13%) samples are nonreactive for Salmonella antigen O and H antigen. These blood samples are also subjected to isolation of Salmonella after drawing serum from the test tube. The clot was ruptured by sterile inoculating needle and then enrichment broth 10 ml was added and incubated at 37^{o}C for 24-48 hour. After showing turbidity one loopful of inoculums is plated on selective media such as Hecktoen Enteric Agar, MacConkey Agar, XLD (Xylose Lysine Decarboxylase), DCA (Deoxycholate Citrate Agar) (Table.3). On the basis of culture characteristic and biochemical reaction, 194 Salmonella spp. out of 746 positive samples were isolated from blood samples (Table 2). The lower isolation of *S. typhi* may be either due to late reporting of the patient to the physician or the patient might have an influence of antibiotics taken before blood culture has been performed.

Table 2: Biochemical reaction of *Salmonella typhi* isolated from blood samples.

No. of isolate	Biochemical test	Reaction
194	**Gram staining**	- ive
	Motility	+ ive
	Lactose	- ive
	Sucrose	- ive
	Salicin	- ive
	Glucose	+ ive
	Indole	- ive
	Methyl Red	+ ive
	Voges-Proskaur	- ve
	Citrate	+ive
	TSI	K/A with H_2S
	Nitrate Reduction	- ive
	Urease	-ve

Table 3. Colony Characteristics of Salmonella on various media

Medium	Colony characteristics
Hecktoen Enteric Agar	Black centre with green periphery
Mac Conkey Agar	Non Lactose fermenting, pale yellow
XLD (Xylose Lysine Decarboxylase)	Black centre with red periphery
DCA (Deoxycholate Citrate Agar)	Non Lactose Fermenting, pale yellow

Sanjeev *et al.* (2013) investigated 50 patient from Mangalore, Karnataka only 33 (66%) were found positive by blood culture. Widal test was positive in 33(66%) patients which included 26 in blood culture positive patients and 7 in blood culture negative patients. There is a higher percentage of positive cases in the blood culture in contrary to our result in which there is a low percentage of positive cases in blood culture. The higher incidence of typhoid cases in studied area may be due to the inadequate sanitation and poor water quality. People living in rural area drink water derived from hand pump which might be contaminated through seepage or water runoff. According to report of World Heath Organisation (2000) regarding sanitation that about 2.4 billion people don't have an adequate basic sanitation facility. According to report published by joint Monitoring Program done by UNICEF and WHO (2008), there was 18% of global population and half of the Indian population practiced open defecation which was later confirmed by Indian Census (2011) in which it was estimated that 49.8% Indian defecated in the open. Human waste i.e. fecal matter in open environment contain many pathogenic bacteria including Salmonella when it rains, the indiscriminate human wastes are washed untreated into the waterways which are the main source of drinking water for both human and livestock. This inevitably leads to the outbreak of cholera, diarrhea, typhoid, hepatitis and other perilous diseases which are claiming many lives of children and adults who cannot afford portable accommodation and hygienic toilet facilities. Another possible reason for the increase incidence of typhoid fever is the availability of potable water. In summer or dry season, the water is not available for people so the water is collected and supplied from places which might be contaminated with human waste and also the causal reason for the rise in typhoid cases in developing countries (Otegbayo, 2005). On the basis of the result and high prevalence of typhoid in studied area it is suggested that people should maintain the hygienic environment and tried to avoid outside food as much as possible and follow the good sanitation practice i.e. not to defecate in open area. Our Prime minister or Government of India however taken several measure initiatives such as Swatch Bharat Abhiyan having objective to clean the roads, drains, and infrastructure and to reduce or eliminate open defecation through the construction of individual, cluster and community toilets. These types of initiative and awareness program among peoples may prevent the communicable diseases including typhoid in some extent.

References

Ackers, M. L., Puhr, M. D., Tauxe, R. V and Mintz, E. D. 2000. Laboratory based surveillance of Salmonella serotype Typhi infections in the United States: antimicrobial resistance on the rise. ***JAMA***., 283, 2668–2673.

Adesunkanmi, A.R.K and Ajao O. G. 1997. Prognostic factors in typhoid ileal perforation: a prospective study in 50 patients. ***J R Coll Surg Edinb***., 42:395-399.

Al-Quarawi S.M., El-Bushra Fontaine R.E., Bubshait S.A and El Tantawy N.A. 1995. Typhoid fever from water desalinized using reverse osmosis. ***Epidem. Infect***. 114, 41-50.

Cabello, F and Springer, A.D. 1997. Typhoid fever in Chile. 1977-90: an emergent disease. ***Revista Medica de Chile***., 125, 474-482.

Cappuccino G .James and Sherman Natalie. 2005. Microbiology A laboratory manual, seventh edition, Pearson Education.

Chandra, H**.,** Singh, B., Srivastava, J., Prasad, R., Nautiyal, A.R. 2010. Seroprevalence of Typhoid in DehradunValley (Uttarakhand), India. ***Res Environ Life Sci***., 3(2):65-68.

Cruickshank, R .1965. Medical Microbiology, 11th Edition p.907.

Crump, J.A., Luby, S.P., Mintz, E.D. 2004. The global burden of typhoid fever. ***Bull World Health Organ***., 82: 346–353.

Everest, P., Wain, J., Roberts, M., Rook, G and Dougan, G. 2001. The molecular mechanism of severe typhoid fever. ***Trends Microbiol***., 9(7):316-320.

Ezeigbo, O.R., Agomoh, N. G and Asuoha-Chuks, N.2015.Laboratory Diagnosis of Typhoid Fever using Widal and Blood culture Methods in Aba, Southeastern Nigeria. ***Am J Microbiol Res***., 3(6): 181-183.

Madhulika, U., Harish, B. N. and Parija, S. C. 2004. Current pattern in antimicrobial susceptibility of Salmonella Typhi isolates in Pondicherry. ***Indian J Med Res***., 120, 111–114.

Murray, P.R. and Shea, Y.R.2004. Bacterial diagnosis. In: Pocket Guide to Clinical Microbiology, 3rd Edition. pp. 135-137.

Otegbayo, J.A.2005. Typhoid fever: The challenges of Medical management. ***Annals of Ibadan Post Graduate Medicine***., 3(1):60-62.

Paniker, C. K. J and Vimla, K. N. 1972. Transferable Chloramphenicol resistance in Salmonella Typhi. ***Nature***., 239, 109–110.

Parry C.M., Hien T.T., Dougan, G., White N.J and Farrer, J. 2002. Typhoid fever. ***N Engl J Med***., 347:1770– 1782.

Saha, M.R., Dutta, P., Bhattacharya, S.K., Rasaily, R., Mitra, U, Dutta, D., Bhattacharya M.K and Pal S.C.1992. Occurrence of multidrug resistant *Salmonellatyphi* in Calcutta. ***Indian J Med Res.***, 95: 179– 180.

Sanjeev, H., Nayak, S., Pai Asha, K.B., Rai, R., Karnaker, V and Ganesh, H.R.2013. A systematic evaluation of rapid dot-EIA, blood culture and Widal test in the diagnosis of typhoid fever. ***Nitte Univ J Health Sci***., 3(1): 21-24.

Shrivastav, D, Jain, A. K., Gharde, P., Sharma, D. B. and Verma R. S. 2014. Typhoid intestinal perforation in Central India – A surgical experience of 155 cases in resource limited setting. ***Int J Biomed Advance Res***., 5(12):600-604.

Wain, J and Kidgell, C. 2004. The emergence of multidrug resistance to antimicrobials agents for the treatment of typhoid fever. ***Trans Roy Soc Trop Med Hyg***., 98:423–430.

WHO 2008. Weekly Epidemiological Record, No. 6, 8th Feb, 2008.

WHO and UNICEF. 2000. Global assessment of water supply and sanitation. UNICEF, New York and WHO, Geneva

WHO and UNICEF.2008. Progress on drinking water and sanitation. UNICEF, New York and WHO, Geneva

World Health Organization 2003. Background Document: The Diagnosis, Treatment and Prevention of Typhoid Fever. WHO document7 Geneva WHO/V and B/03.07.