

An epidemiological surveillance of asymptomatic typhoid carriers associated in respect to socioeconomic status in India

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Introduction

The term ‘enteric fever’ was coined to embrace both typhoid and paratyphoid. It was known as a generalized infection of the reticuloendothelial system and intestinal lymphoid tissue accompanied by sustained fever and bacteraemia. Typhoid has remained a major public health issue in many tropical countries and the highest burden of the disease is in Asia (Pang et al. 1998). The global burden of typhoid was estimated as 16,000,000 cases and 600,000 deaths per year (Pang and Puthuchery 1983; Pang and Bhutta 1995). Currently global pandemics are being caused by the less common serovar of *S. typhi* (Miko et al. 2002).

The asymptomatic carrier state in humans has had serious consequences on public health and the exclusive source for typhoid bacillus was asymptomatic typhoid carriers. An individual could asymptotically carry the typhoid bacilli for days to years without any symptoms of typhoid fever (Bigelow and Anderson 1933; White and Parry 1996; Senthilkumar and Prabakaran 2005). They usually excrete as many as 10^{11} virulent typhoid bacilli per gram of faeces. These organisms reside in the scarred foci of the biliary tract, migrate through the bile ducts and over the vast surface area of intestinal epithelium; however, they did not cause typhoid fever in the carrier (Hornick et al. 1970).

S. typhi usually possesses a polysaccharide Vi (virulence) surface antigen that was associated with enhanced capacity to invade the host. Vi antigen of *S. typhi* and its relation with high serum titres of *S. typhi* carrier state, reports about the usefulness of Vi serology in the detection of the carrier state (Felix and Pitt 1934; Felix 1938; Lanata et al. 1983) and establishes a chronic asymptomatic infection in the gall bladder (Kauffmann 1954; Prouty et al. 2002).

Asymptomatic typhoid carriers were deficient in IgM and anatomical alterations in the biliary tract lead to prolong bacterial persistence (Cheronokhvosstova et al. 1969). The carrier state occurred disproportionately often following infection not only in women but also in older patients, with biliary carriage rather than the urinary carriage (Hornick 1985).

Identification of asymptomatic typhoid carriers as the source of an outbreak usually required multiple cultures for several weeks (Engleberg et al. 1983). Positive asymptomatic carriers from food handlers significantly contributed to the spread of *S. typhi*, *S. paratyphi* A and *S. paratyphi* B. Therefore, a periodic assay of *Salmonella* spp in foodstuffs and food handlers could help in the control of food borne diseases (Sasikumar et al. 2005).

Materials and methods

Collection of the sample

Based on historical background, a total of 1,000 suspected individuals who previously suffered from typhoid have been chosen for this study among the food handlers working in restaurants, first class and second class hotels, roadside hotels, roadside vendors, fruit juice stalls, milk men, slaughter houses, chicken stalls and fried fish stalls. Early morning faecal samples from 100 individuals from each group were collected and transported to the laboratory by using screw-

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capped tubes with Cary-Blair medium (Himedia, India); three samplings were done at an interval of 15 days.

Identification of the *Salmonella* spp

The preliminary morphological and biochemical tests were performed for the identification of *S. typhi*, *S. paratyphi* A and *S. paratyphi* B viz. Gram's staining, motility test, catalase test, oxidase test, sugar fermentation, indole test, methyl red test, Voges–Proskauer test, citrate utilization test, triple sugar iron test and urease test. They were also identified on the basis of growth pattern observed on enrichment medium (Selenite-F broth), differential medium (Mac Conkey Agar), and selective medium (Bismuth Sulphite Agar, Xylose, Lysine and Deoxycholate Agar; Himedia, India).

Serological test for the confirmation of *Salmonella* spp

The bacterial agglutination test (high titre serum agglutination test) was adopted to identify *S. typhi* (Lim and Fok 1987), *S. paratyphi* A and *S. paratyphi* B. This procedure was carried out on an ordinary glass slide. A drop of saline was placed in the circle and a small amount of culture of *S. typhi*, *S. paratyphi* A and *S. paratyphi* B from the solid medium was emulsified using an inoculation loop. A drop of antiserum of *S. typhi*, *S. paratyphi* A and *S. paratyphi* B were added respectively. Then, both were mixed and observed for agglutination.

Results

Screening of asymptomatic typhoid carriers

A total of 1,000 food handlers in Tamil Nadu were subjected to screening for asymptomatic typhoid carriers. Of them, 88 individuals have shown positive result for asymptomatic typhoid carriers (Fig. 1). In the 88 positive cases, 67 were identified as carrying *S. typhi*, 16 *S. paratyphi* A and 5 *S. paratyphi* B. According to the gender, out of 88 (8.8 %) cases 70 (79.54 %) cases were males and 18 (20.45 %) were females (Table 1). Based on their history profile, both men and women were considered as being 74 (84 %) acute and 14 (16 %) chronic asymptomatic carriers without any symptoms of typhoid fever.

Identification of *S. typhi* and *S. paratyphi* isolates

The production of red colour deposits in the Selenite F broth, jet-black colour colonies on BSA, red smooth colonies on XLD and colourless colonies on Mac Conkey Agar by the isolates indicate the isolates belong to *Salmonella* spp. Based on the cultural characteristics, preliminary tests

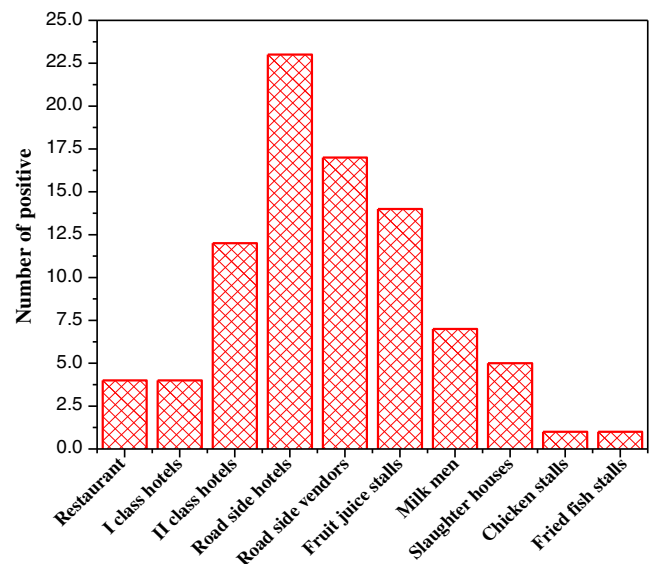


Fig. 1 Incidence of asymptomatic typhoid carrier with the respect to different food handlers and their socioeconomic status

and biochemical tests, the isolates were identified as *S. typhi*, *S. paratyphi* A and *S. paratyphi* B (Table 2).

Bacterial agglutination test for confirmation of *Salmonella* spp

A clear agglutination of emulsified colony of *S. typhi*, *S. paratyphi* A and *S. paratyphi* B with respective antiserum on the glass slide (bacterial agglutination test) confirms that the isolates were *S. typhi*, *S. paratyphi* A and *S. paratyphi* B respectively.

Discussion

Typhoid fever continues to be an important infection in endemic countries and among travelers to these areas. Typhoid fever has been declining in many middle-income countries, whereas the bulk of cases occur in the Indian subcontinent and in south-east Asia (Meltzer and Schwartz 2010). Poor personal hygiene and inadequate food handling can easily potentiate the transmission of *S. typhi*. Several food products kept at room temperature were found to favour the growth of *Salmonella* spp. The food handlers prominently play a role in disseminating typhoid bacilli through different food products and water (Lin et al. 1988; Senthilkumar and Prabakaran 2005). Similarly, the present work is also carried out to screen the typhoid asymptomatic carriers with respect to different socio economic status among the food handlers in Tamil Nadu, India.

Of the typhoid patients, about 3–5 % of individuals become asymptomatic carriers after an acute typhoid episode. There is

Table 1 Incidence of asymptomatic typhoid carrier state among the various food handlers

Groups of food handlers	Time after typhoid episode (months)/No. of samples collected			No. of asymptomatic typhoid carriers					
				Age group (years)		Gender		Status of carrier	
	0–3	3–6	6–12	15–30	30–60	Male	Female	Acute	Chronic
Restaurant	30	50	20	1	3	3	1	4	–
1st class hotels	26	40	44	–	4	3	1	4	–
2nd class hotels	37	60	3	4	8	6	6	10	2
Roadside hotels	40	16	44	2	21	19	4	18	5
Roadside vendors	53	36	11	1	16	14	3	14	3
Fruit juice stalls	36	42	22	3	11	12	2	13	1
Milk men	42	37	31	1	6	7	–	5	2
Slaughter houses	47	40	13	1	4	5	–	4	1
Chicken stalls	25	42	33	–	1	1	–	1	–
Fried fish stalls	30	46	24	–	1	–	1	1	–

an established infection in their gall bladder of the infected individuals after the symptoms disappeared, serving as a reservoir for the spreading of the typhoid fever (Kauffmann 1954; Prouty et al. 2002; Gonzalez-Escobedo et al. 2011). Transmission of typhoid fever is chiefly through the asymptomatic carriers—those who work as the food handlers in hotels and restaurants (Sasikumar et al. 2005). This present study also revealed that 8.8 % of the asymptomatic typhoid carriers were found among the food handlers.

The susceptibility to carry the typhoid germs also varies with gender and age. Women exceed men as carriers by a ratio of 3:1. Carrier state occurs more commonly in elders (Hornick et al. 1970). In contrast, the present investigation revealed that among the food handlers, men can serve as more potential carriers of typhoid germs than women.

A series of over 9,000 cases reported in London during 1871–1894 represented clinical enteric fever in urban communities of all ages and the data shows that maximum susceptibility is found in people between the ages of 15–25 years (Goodall and Washbourn 1896). In this the present investigation, a typhoid asymptomatic carrier state was found among the people in the age group 15–60 years. Of the people who were suffering from typhoid fever during their lifetime, some were cured, while a few continued to

harvest and excrete typhoid bacilli in their faeces. Based on the life history of such persons, they were found to be asymptomatic typhoid carriers.

Outbreaks of typhoid fever from restaurants through contaminated food by infected asymptomatic food handlers have also been reported from the United States (Taylor et al. 1984). In the US, out of 1,013 cases of typhoid fever from 1975 to 1985, 28 % were related to an outbreak. The source of exposure was unknown in most cases, but 21 % were associated with newly discovered carriers, 9 % with previously identified carriers and 3 % with exposure to laboratories (Royan et al. 1989).

A total of 138 enteric fever cases were found in the United Kingdom during 2005–2010. Of these, 65 % and 35 % cases were caused by *S. typhi* and *S. paratyphi* respectively. These individuals, in the age group 16–40 years and under 16 years, acquired the infection abroad. Indian, Pakisthani, Bangladesh patients have been the majority (87 %) of the typhoid cases. New entrant and foreign visitors accounted for 10 % of the cases, 92 % cases had a history of recent foreign travel; 57 % patients had travelled to visit friends and relatives and 23 % patients were new entrants/foreign visitors from the Indian subcontinent (Reddy et al. 2011).

Table 2 Identification of the isolates based on preliminary and biochemical characteristics

Isolates	Gram	Mot	Catalase	Oxidase	Glu	Lac	Suc	Xylu	d-tar	Ind	MR	VP	Cit	TSI	U
<i>S. typhi</i>	-Ve	+	+	-Ve	A	–	–	A	A	–	+	–	–	Ak/A H ₂ S	–
<i>S. paratyphi</i> A	-Ve	+	+	-Ve	AG	–	–	–	–	–	+	–	+	Ak/AG	–
<i>S. paratyphi</i> B	-Ve	+	+	-Ve	AG	–	–	AG	–	–	+	–	+	Ak/AGH ₂ S	–

A acid; AG acid and gas; Ak alkaline; H₂S hydrogen sulphide

G gram staining; M motility; C catalase; O oxidase; G glucose; L lactose; MAL maltose; MAN mannitol; S sucrose; I indole; C citrate utilization; TSI triple sugar iron; U urease; Ak alkaline; A acid H₂S; + positive; – negative

The present study revealed the workers in roadside (26.13 % carriers) and second class hotels (13.63 % carriers) live in relatively very poor hygienic condition as compared to the first class hotel workers. Therefore, there is a high probability of spreading the typhoid bacilli to susceptible persons via food handlers in roadside hotels and second class hotels. They pose a public health problem to the human race as reservoirs of *S. typhi*, *S. paratyphi* A and *S. paratyphi* B.

Salmonella can stay alive in an asymptomatic carrier in human gall bladder for years. An antibiotic treatment of the carriers is often ineffectual against *S. typhi* due to the emergence of drug resistant strains. Thus, the multi-drug resistant *S. typhi* has recently turned out to be a center of attention in our country (Senthilkumar and Prabakaran 2005).

The World Health Organization has been recommended for the programmatic use of new-generation typhoid vaccines in high-risk areas of countries where typhoid fever is still endemic. Past and current typhoid vaccination programs that have taken place in Thailand (using the old whole-cell vaccine) and in China, Vietnam and India (using the new-generation injectable Vi polysaccharide vaccine; DeRoeck et al. 2008).

Conclusion

In hotels, the cooks and waiters could easily contaminate water and foodstuffs while handling them. Often their nails remain wet and dirty and their poor sanitation after defaecation lends a favourable environment to the growth and spread of the bacteria. Their palms too carry many enteric pathogens. Washing their hands with only pure water is inadequate to keep them sterile. In addition, foodstuffs would favour the growth of *S. typhi*, *S. paratyphi* A and *S. paratyphi* B. The improper disposal of stools after defecation could also lead to severe water pollution. Particularly at the time of flooding, it would lead to a large-scale outbreak of typhoid, creating public health problems of a severe magnitude. Therefore, we conclude that following hygienic practices, health education programmes, treating the carrier with a suitable chemotherapeutic agent and vaccination among the public would help us to get rid of the problem of typhoid in the developing countries.

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Conflict of interest The authors declare that they have no conflict of interest.

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