Evaluation of a Qualitative Antibody Detection Method of Typhoid Fever

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Abstract
Whole South East Asia including Pakistan is the hyper-endemic areas for typhoid fever. Clinical diagnosis is not definitive. Blood or bone marrow culture which is still considered as the gold standard in cases of typhoid fever. However bacterial culture facilities are often unavailable and time consuming. there is real need of rapid serological diagnostic test. The present study was conducted to evaluate the % age of IgG and IgM antibodies against the Salmonella typhi by Typhidot test. A total of 100 cases of typhoid fever were collected from 1st January 2015 to 31st December 2015 from Pathology Laboratory in Karachi, Pakistan, on which typhidot procedure was followed. The results of typhidot were IgG positive in 11(11%) cases, and IgM positive in 6(6%) cases and both were collected from 1

INTRODUCTION

Typhoid fever still remains a greater health problem worldwide. Salmonella typhi is the etiological agent of typhoid fever. This disease is considered as endemic in the South- East Asia, Indian sub-continent, Africa, the Middle-East, South and Central America, due to insufficient supply of pure water and uncontrolled pollution of sewage water (Gillespie, 2003). Salmon first gave the name Salmonella. He first isolated Salmonella choleraesuis, from porcine intestine (Pegues et al., 2005). Wide range of diseases in humans and animals are caused by Salmonella organism and it is active commensals.

In 1843, Austin Flint did classic study about the Epidemiology of typhoid fever. Different clinical symptoms of typhoid fever are characterized by sustained fever, headache, malaise, anorexia, relative bradycardia, constipation or diarrhea, and nonproductive cough (Crump et al., 2004). S. typhi cause typhoid fever and S. Paratyphi cause Paratyphoid fever. In early 19th century, the disease appeared as a significant infectious disease (Jenkins et al., 2009). In Pakistan typhoid fever is a prevalent disease (Yoshikawa et al., 1980; Hanon, 1991). The global annual burden was estimated at approximately 12 million cases for 2010 (Mogasale et al., 2014).

Salmonella typhi affects various organs of the body and it resides the lymphatic tissues of the small intestine, liver, spleen and bloodstream of infected persons. The disease is pathogenic for both humans and mammals and they are associated with inflammatory reactions (Thong et al., 2000). Contaminated food or drinking water is a main source of infection due to which bacteria can enter the human body where they multiply and disperse in the body via blood circulation (World Health Organization, 2008).

For S. typhi the infectious dose is about 108 organisms by ingestion and it varies with gastric acidity (Hornick et al., 1967). In summer and wet season, infection is most frequent in young children and older people (Jerrold and Turner, 2010). It is estimated that 1%-5% of patients become chronic carriers, which depends on their age and S. typhi persist in the gall bladder of infected persons (World Health Organization, 2003). The disease primarily influences developing areas of the world where sanitary measures and pure water are unavailable (World Health Organization, 2003).

Laboratory tests are performed for the diagnosis of disease. Isolation of the pathogen is done through blood and bone marrow. The disease confirmation from these samples is the most suitable method (Abdoel et al., 2007). Typhoid fever is a significant cause of morbidity and mortality and it is due to emergence of antibiotic resistance
among Salmonella species and as well as delay in diagnosis and appropriate therapy (Bhutta, 1996). In 1940’s several antibiotics were used successfully for the treatment of typhoid fever and such antibiotics were chloramphenicol, ampicillin and trimethoprim-sulphamethoxazole (Ivanoff et al., 1997). Salmonella typhi strains showed resistance to multidrugs and it was first observed in 1987 in Pakistan (Yoshikawa et al., 1980; Hanon, 1991). Due to which other curative methods are recommended to treat typhoid fever (Yanagi et al., 2009; Aggarwal et al., 2011). Antibiotic resistant bacteria are causing great damage to public health (Iqbal et al., 2015). The present study was conducted to evaluate the percentage of IgG and IgM antibodies against the Salmonella typhi by Typhidot test.

MATERIALS AND METHODS

The data of typhoid fever was collected in a Pathology Laboratory in Karachi, Pakistan, from 1st January 2013 to 31st December 2013. The total 100 cases which were clinically diagnosed as typhoid fever by various clinicians and sent to laboratory for the typhidot test were collected during above mentioned period. Typhidot procedure was followed in these cases. The study was approved by Ethical Review Board of Jinnah University for Women, Karachi, Pakistan.

Collection of blood samples

Five ml blood in disposable syringe was taken from antecubital vein aseptically and transferred to sterile centrifuge tube slowly just to avoid hemolysis. It was allowed to clot and then centrifuged at the rate of 3000 rpm for 15 minutes. Separated serum was transferred with the help of disposable Pasteur pipette to sterile capped tubes and stored at -20 °C until processed for analysis. Repeated thawing and freezing were avoided.

Sample preparation

Serum and plasma were separated from whole blood according to standard procedure.

Typhidot (3 Hours)

Typhidot (Dot-EIA) test for specific detection of IgG & IgM to Salmonella typhi (Mehmood et al., 2015) was performed using kits manufactured by Reszon Diagnostics International Sdn. Bhd. No. 12A, Jalan TP5 Taman Perindustrian UEP 47600 Subang Jaya Selangor DE, Malaysia. The data obtained was analyzed statistically using SPSS v16.0.

RESULTS

The results of typhidot were IgG positive in 11(11%) cases, and IgM positive in 6(6%) cases and both (IgG & IgM) positive in 9 (9%). The results of typhidot were negative in 74(74%) (Figure 1).

DISCUSSION

Typhoid fever still remains a greater health problem worldwide. Approximately >100/100,000 cases/year are the highest incidence reported in South-East and South-central Asian countries (Gillespie, 2003). In Pakistan, approximately an annual incidence of 413/100,000 person/year was reported (Crump et al., 2010). We collected total 100 cases of typhoid fever and Typhidot procedure was followed in these cases. Sensitivity and specificity of this test is more than 90% and it is reported in different studies (Choo et al., 1994; Jackson et al., 1995; Lu-Fong et al., 1996; Jesudason et al., 2006). The collected data of 100 cases showed Positive IgG% was 11% which indicates presence of IgG antibodies as it possibly as a result of current infection, previous infection, relapse or re-infection. Positive IgM% was 6% which indicates presence of IgM antibodies and it showed acute typhoid fever, infection at the initial stage. Positive both (IgG & IgM) % was 9% which indicates presence of IgG and IgM antibodies against S. typhi and it showed acute typhoid fever but infection at the middle stage. The percentage of negative cases was 74% which indicates absence of IgG, IgM and both (IgG & IgM) antibodies, no typhoid fever was suspected. Similarly Mehmood et al. (2015) reported 47 (32.4%) patients had only IgM positive on Typhidot, 7(4.8%) had both IgM and IgG positive and 91(62.8%) had both IgM and IgG negative. According to a recent study 10 cases were positive and only 01 case was negative by Typhidot test among 11 blood culture positive cases (Garg, 2017).

CONCLUSION

Typhidot is a Dot-EIA test to diagnose typhoid fever. It is relatively rapid serological test, simple, reliable, less expensive test which fulfills needs of clinicians especially in areas where laboratory services are limited. This test is
helpful in those areas where blood culture facilities are unavailable. Stages of typhoid infection can easily indicate and results can also be easily interpreted in this test. It is less time consuming procedure and it has high sensitivity and specificity as compared to Widal test.

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CONFLICT OF INTEREST
There is no conflict of interest.

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