JMID/
Journal of Microbiology and Infectious Diseases

2017; 7 (1): 29-35
doi: 10.5799/ahinjs.02.2017.01.0249

RESEARCH ARTICLE

Antimicrobial Susceptibility among circulating *Salmonella typhi* serotypes in Children in Jakarta, Indonesia

Mirari Prasada Judio[1], Mulya Karyanti[1], Lia Waslia[2], Decy Subekti[2], Bambang Supriyatno[1], Kevin Baird[2]

ABSTRACT

Objectives: Indonesia is known for high incidence of typhoid fever especially in children. This study aimed to observe antibiotic susceptibility in circulating *Salmonella typhi* serotypes in children with typhoid.

Methods: A cross sectional study design was conducted. A total of 142 blood samples from children between 1-18 years old clinically diagnosed with suspected typhoid fever were recruited between January 2012 and July 2013 from six health centers in Jakarta. Confirmed cases were retrieved based on *S. typhi* isolate finding in blood culture. Antimicrobial susceptibility was investigated and PCR was used to detect *S. typhi* serotypes using *fliB*, *fliC* and *aroC* genes.

Results: The prevalence of confirmed typhoid case based on isolate finding was 22 (15.5%). Twenty of *S. typhi* isolates expressed *fliC* gene carrying H:d allele, the other two expressed j allele, while only two samples expressed *fliB*, all showed no difference in pathogenicity and antimicrobial resistance.

Conclusions: Circulating serotypes found in typhoid children in Jakarta, Indonesia are still susceptible even to the firstline antimicrobials. Thus, chloramphenicol, ampicillin and co-trimoxazole are still recommended. *J Microbiol Infect Dis 2017; 7(1): 29-35*

Keywords: *Salmonella typhi*, child, serogroup, microbial sensitivity test

INTRODUCTION

It has been reported that incidence of typhoid fever was estimated to be approximately 21 million in year 2000 with highest incidence among children in South Central and South East Asia and the impact is difficult to measure because often clinical features is confused with many other febrile infections [1,2].

In Indonesia, the incidence of typhoid fever is 900,000 cases per year, with yearly morbidity of 20,000. Confirmed case typhoid fever was 1000 case per 100,000 populations per year [3] with high prevalence of 64% found in school age children between 3-19 years old [4].

The burden of typhoid fever however remained largely unknown in many regions where diagnostic methods and surveillance systems are not yet implemented [5]. The same is true in Indonesia where the magnitude of disease is difficult to measure since bacteriology laboratories are lacking in numbers.

Meanwhile, emerging issues on increasing trend of *S. typhi* with antimicrobial resistance generates concern in endemic countries since almost two decades ago [6-8]. A study of typhoid fever in all ages in Indonesia showed increased of microbial resistance towards three first line drugs: ampicillin, chloramphenicol and co-trimoxazole as well as ciprofloxacin [9].

Meanwhile bacterial pathogenicity is known related to *S. typhi* expression of two flagellar antigens encoded by phase 1 and phase to flagellin genes *fli*C and *fliB*.

It has been hypothesized that some *S. typhi* isolates found in Indonesia express high genetic

[1]Department of Child Health, University of Indonesia
[2]Clinical Microbiology, Eijkman-Oxford Clinical Research Unit
Running title: Antimicrobial Susceptibility among circulating S. typhi serotypes in Children
Correspondence: Dr. Mirari Prasadajudio, Department of Child Health, Sub-division Infectious and Tropical Disease, CiptoMangunkusumo Hospital, Medical Faculty, University of Indonesia, Jalan Salemba Raya 6, Jakarta Pusat 10430, Indonesia, Email: mirari23@yahoo.com
Received: 09 March 2016, Accepted: 21 December 2016

heterogeneity if compared to other endemic regions [10,11].

A flagellar-antigen study observed that there was not only *fli*C gene carrying H:d allele (*fliC*-d) found circulating in Indonesia , but there was also another different flagellar gene *fli*C carrying H:j allele (*fliC*-d) found. Expression of the later allele correlated with milder clinical manifestation of typhoid fever [12].

Another study observing 143 serovars of *S. typhi* in Indonesia for 30 years identified rare single nucleotide polymorphisms (SNPs) also from flagellar genes, suggested that there are approximately 8 haplotypes circulating in Jakarta namely H46, H45, H59, H85, H84, H1, H51, H50 and H8, but majority of haplotype found were H59 (73%) dan H8 (24%). Haplotype H59: consists of 4 subgroups based on its gene expression: *fliC:* d, *fliC:* j and *fliB* (z66)+*fliC:*d and *fliB* (Z66)+ *fliC:*j [13].

Some of the variants were observed to associate with microbial resistance. Haplotype H58 for example are frequently isolated in some South East Asia regions, and related to resistance towards nalidixic acid [14]. This is a preliminary study to observe circulating *S. typhi* isolates in children in Indonesia, and to find possible antibiotics resistance related to their expressions.

METHODS

Study setting and participants

We performed a multicenter cross sectional study between January 2012 to July 2013 in outpatient and inpatient wards of three public hospitals: Cipto Mangunkusumo, Budi Asih and Soelianti Saroso hospitals and three primary health care centers in North and East Jakarta: Puskesmas Pademangan, Ancol and Jatinegara. The centers were selected based on distance to main research center. The study was approved by Medical Faculty University of Indonesia ethical committee, and affirmed by Health Department. The children were included after parents' approval if they were between 1 and 18 years old with history of fever three days or more, clinically diagnosed as typhoid fever by physicians. Children who had been clearly diagnosed with infectious disease other than typhoid fever were excluded from the study.

Data and blood collections

In total 5 ml blood were withdrawn for further examination, 2 ml were separated for complete blood count analysis and semi-quantitative colorimetric test Tubex®TF (IDL Biotech AB, Sweden), while the other 3 ml was inserted into pediatrics culture medium (BD Bactec PedsTM Plus Medium, USA) for blood culture assessment. Tubex®TF test of patients from both Ciptomangunkusumo and Budi Asih hospitals were analyzed within hospital laboratory, whereas the rest were analyzed in an appointed private laboratory, which located within less than 10 kilometers distance to each center involved in study. Tubex®TF test were conducted according to manufacture procedure. Positive result was defined with a reading of $\geq$4. Meanwhile blood collected for biomolecular analysis, were transported to Clinical Microbiology Eijkman-Oxford Clinical Research Unit (EOCRU), Jakarta.

Blood analysis

Blood were then cultured in BD BactecTM 9050 blood culture system at 37 ^{o}C (Beckton, Dickinson and Company, USA), blood with both positive or negative results were then reconfirmed by 24 hours incubation at temperature 37 ^{o}C on Salmonella-Shigella (SS) and Mac Conkey agar plate. Samples were excluded for biomolecular analysis when no growth found for *S. typhi*. Whereas samples with positive growth on both agar plates were incubated for another 24 hours at temperature 37 ^{o}C. Any isolate growth were identified as *S. typhi* after verifications with gram staining, Kliger ion Agar isolation, motility test with Motility Indole Ornithine and Difco ®Agglutination test (Beckton Dickinson Company, USA) using all the procedures as instructed. *S. typhi* isolates were then incubated for 24 hours at 37 ^{o}C in Mueller-Hinton Agar medium (Beckton, Dickinson and Company, USA) for antibiotic susceptibility testing with 9 antibiotics discs: ceftriaxone, amikacin, chlorampenicol, kanamycin, ciprofloxacin, nalicidic Acid, gentamycin, amoxiclav, sulfamethoxazole and imipenem. Antibiotics susceptibility was evaluated as described elsewhere.

DNA were extracted from these isolates using QIAmp DNA mini kit® (Qiagen, Hilden,

Germany) with procedure as described by manufacture. Using the Polymerase chain reaction (PCR) we analyzed genomic DNA *S. typhi* in blood specimens. Primers were developed from flagellin genes: *fliC*, *fliB*(z66) and *aroC*. The amplification of *fliC* primers were performed with *fliC*_F: 5'-TTA-ACG-CAG-TAA-AGA-GAG-3' (F= Forward), *fliC*_R: 5'-ATG-GCA-CAA-GTC-ATT-AAT-AC-3' (R= reverse), and produced 1521 base pair for d-allele, while 1273 bp for j-allele. fli-B (z66) z66 flag_F: 5'-ATG-GCA-CAA-GTC-ATC-AAT-AC-3' and z66 flag_R:5'-TTA-ACG-CAG-CAG-AGA-CAG-TAC-3' produced 1479 base pair amplicons. Aro-C were used as control: aro-C flag_F: 5'-CCT-GGC-ACC-TCG-CGC-TAT-AC-3', *aroC*_R: 5'-CCA-CAC-ACG-GAT-CGT-GGC-G-3'. [13]

As control samples for DNA amplification were DNA *S. typhi* of patients from Oxford Clinical Research Unit Vietnam with positive expression on above genes were applied. PCR were performed with PCR toptaq DNA polymerase (Qiagen, Hilde, Germany) with concentration of DNA 20 ng/ml, cycled for 35 times on a Thermocycler PCR System 9700 (Applied Biosystem, California, USA) and result products were analyzed on 1% agarosa gel with 2 µl loading dye (Promega, Madison, USA), for each 5 µl samples. The size was measured by comparing the migration available with 100 bp DNA ladder (Promega, Madison, USA).

Data measures

All demographic and clinical data were analyzed using Statistical package for social studies version 20.0 (IBM, SPSS, New York, USA).

RESULTS

This study observed 69 cases of typhoid fever (48.6%) which were seen at hospital both in and out patients, while the other 73 cases (51.4%) at primary health centers. Demographic and clinical characteristics for typhoid fever patients in this study are presented in table 1. From 142 patients clinically diagnosed and treated as typhoid fever, only 22 patients (15.5%) were confirmed case of typhoid fever with identified *S. typhi* bacteria in their blood culture. The youngest age of confirmed typhoid case was 3 year-old while and the oldest age was 15 year-old with fever between 6-8 days prior to diagnosis.

Evaluation on treatment prior to sampling showed that 139 out of 142 (97.8%) patients had received medications from either primary health care or private clinics or hospitals during period of disease (Table 2). It is also observed that none who received chloramphenicol showed positive *S. typhi* in blood culture, except for one patient with combine treatment chloramphenicol and ampicillin, however the medications were given less than 24 hours before blood culture were retrieved.

PCR results showed that 20 out of 22 *S. typhi* samples were expressing fliC with d-flagellin allele, which generated DNA fragment of 1521 bp, while 2 other sampels were expressing j-allele at 1273 bp, by deletion or shortening of base pairs. (Figure 1)

The *fliB* (z-66) gene was confirmed by amplification of 1479-bp products in 2 samples (Figure 2). Both samples which expressed z-66 antigen also harbored a *fli*C j allele in chromosome. All *S. typhi* isolates found showed no difference in affecting disease severity and showed susceptibility towards all the antibiotics tested.

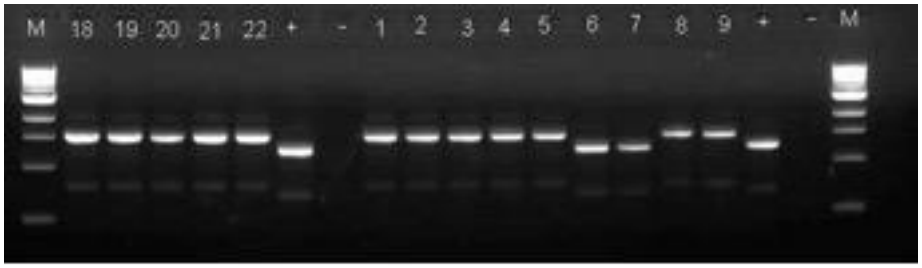

Figure 1. FliC expression, all were positive for d-allele except sample no 6 and 7 were positive for j-allele.

Table 1. Demographic and clinical characteristics of typhoid infected patients based on blood culture.

Variables	Blood culture (+) (n=22), n (%)	Blood culture (-) (n=120), n(%)
Gender (M)	15 (68.2)	64 (53.3)
Age		
1- 4 years old	2 (9.1)	22 (18.3)
5-9 years old	13 (59.1)	54 (45)
10-14 years old	6 (27.3)	37 (30.8)
15-18 years old	1 (4.5)	7 (5.8)
Residential area		
North Jakarta	1 (4.5)	22(18.3)
East Jakarta	16 (72.7)	64 (53.3)
Central Jakarta	2 (2)	10 (8.3)
South Jakarta	3 (13.6)	12 (10)
West Jakarta	0 (0)	2 (1.7)
Greater Jakarta	0 (0)	10 (8.3)
Place of diagnosis		
Hospital	16 (72.7)	53 (44.2)
Primary health care	6 (27.3)	67 (55.8)
Length of fever		
3-5 days	4(18.2)	56 (46.7)
6-8 days	10(45.5)	37 (30.8)
9-11 days	5(22.7)	17 (14.2)
12-14 days	2(9.1)	8 (6.7)
>15 days	1(4.5)	2 (1.7)
Clinical manifestation		
Abdominal pain	1 (4.5)	7 (5.8)
Nausea	3 (13.6)	30 (25)
Diarrhea	1 (4.5)	3 (2.5)
Constipation	0 (0)	6 (5)
Abdominal pain & nausea	6 (27.3)	24 (20)
Constipation &nausea	2 (9.1)	8 (6.7)
Diarrhea &nausea	9 (40.9)	25 (20.8)
Non specific	0 (0)	17 (14.2)

Table 2. History of antibiotics treatment prior to microbiological test.

	Blood culture (+), n=22 (%)	Blood culture (-), n=120 (%)	p value
Prior antibiotics			
Yes	8 (36.4)	45 (37.5)	0.820
No	0 (0)	2 (1.7)	
Unknown	14 (63.6)	73 (60.8)	
Length of antibiotics			
<3 days	5 (22.7)	20 (16.7)	0.832
3-7 days	1 (4.5)	10 (8.3)	
>7 days	0 (0)	2 (1.7)	
No antibiotics	0 (0)	2 (1.7)	
Unknown	16 (72.7)	86 (71.7)	
Antibiotics			
CLPI	0 (0)	9 (7.5)	0.713
PEN + AMP/AMP	1 (4.5)	5 (4.2)	
Cephalosporin	1 (4.5)	11 (9.2)	
AMP & CRF	1 (4.5)	5 (4.2)	
No Antibiotics	0 (0)	2 (1.7)	

AMP=Ampicillin, CLP=Chloramphenicol, PEN=penicillin, AMX=amoxicillin

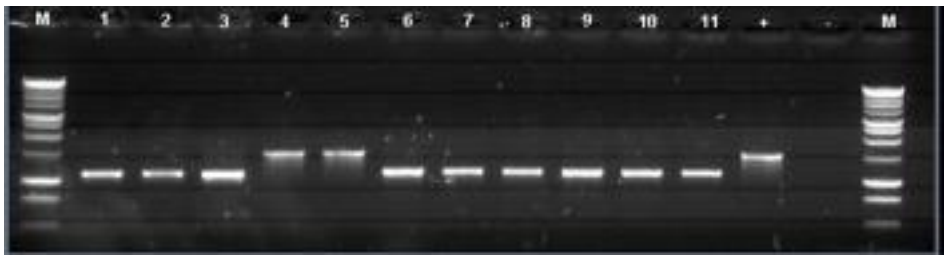

Figure 2. *FliB* (z66) expression could be seen in sample no 4 and 5.

DISCUSSION

Interpretation of results in this study reflects of what occurs daily in pediatrics practice in Indonesia or developing countries, where lack of diagnostic resources would restrict the ability of physician to perform adequate evaluations of diagnostic test [15]. Decision on treatment often is established solely based on clinical judgment from symptoms appeared, therefore impose over-diagnose of typhoid fever which showed that most patients in this study have previously been given medications by other physicians. Prompt management must therefore be established to help reduce the rate of antimicrobial resistance.

We observed typhoid cases diagnosed in primary health care are lower if compared to hospital where these diagnostic tools are readily available. The highest frequency of age group in this study for typhoid fever were between 5 and 9 years-old, which is similar to the range observed in Crump's study [1]. Our study inclusion age were between 1 and 18 year-old to reduce the bias of fever and gastrointestinal issues due to other disease, which is common in infants such as Rotavirus. Children age less than two years were prompt to higher risk of rotavirus-positive diarrhea with incidence of 1185 of 1345 (88%) patients during study period [16]. Moreover we also believe that infants under one year-old in our country are still being breastfed and receive home-prepared meal, whereas there is a higher possibility that infants above 1 year-old would receive meals prepared from outside home with higher dietary diversity, which impose children to higher risk for typhoid fever infection.

It is also suggested in this study that male subjects has higher tendency to suffer from typhoid fever if compared to female. The phenomenon is similar to what was observed in Bangladesh large number study between 2005 and 2009 with number of male cases surpassed female cases. The explanation was gender predilection might be the reflection of healthcare seeking culture related to patriarchy where young sons would be more likely to be valued than female, thus more likely to be taken to hospital [17]. This hypothesis, as well as possibility of difference in hand hygiene practice, toilet etiquette, or outdoors exposure between boys and girls are yet to be proven.

We applied Tubex®TF a semi-quantitative test as part of our screening. However its result is based on inhibition reaction between patient's IgM antibodies and monoclonal antibodies to bind to *S. typhi* O9 lipopolisaccharide, which usually emerge between day 6 and 8 onset of disease [2], thus patients may no longer have antigen circulating in the blood but the antibodies can still be detected. Duration of fever observed in confirmed case patients were between 6-8 days prior to diagnosis, which is concurrent to the previous finding. Blood which were collected during late infection would show antibodies abundantly, while antigen would have been cleared by the body. This suggests that test should be applied instead in the early typhoid fever infection [18]. This shows that Tubex may be applied in area where blood culture is not feasible however the use should be considered within a week from onset of disease. Yet, careful interpretation must be made in areas of endemicity like Indonesia where there is often a low background level of antibodies circulating in normal population and variety between areas and times [19].

Although gold standard for typhoid fever is finding of *S. typhi* isolate in bone marrow aspirate culture [20], but detecting *S. typhi* isolate in blood culture can also confirm the diagnosis of typhoid fever [2]. The small number of positive culture found in this study may be

related to possible factors of limitations on laboratory media, the presence of antibiotics volume of specimen cultured and time of collection, which should be adequately taken between 7-10 days of disease onset [2]. This study had minimized any possible bias from other isolate, which may grow in non-ox bile laboratory media. Any isolates found in positive culture were verified and identified using several microbiological tests and conducted in one centralized laboratory, specimens were also delivered within 24 hours since blood retrieval. However the optimal volume of blood withdrawn would also affect the ability to detect infection in the blood. It is suggested that in infants and children the blood volume collected should be 0.5 ml for bodyweight less than 4 kilograms, while up to 5 ml for body weight of 4 kilograms [21]. Meanwhile WHO suggested volume of 5 ml for blood culture containing 45 ml broth, although it did not distinctively specify the age [2]. Our blood samples were withdrawn between 1-3 ml in 40 ml pediatrics culture medium. To obtain standardized 5 ml blood in all children were not as simple in practice and documentation on blood volume retrieved would help in analysis for future reference.

From Tubex®TF isolates retrieved in confirmed case typhoid, we did not observe any difference in disease severity between patients infected by *S. typhi* which antigen expressed *fliC*-j allele or H:j variant and others who were infected by *S. typhi* with H:d variants. A previous study which study flagellar serotypes of *S. typhi* in Indonesia hypothesized that *fliC*- j allele experience deletion in some area of the gene, thus there would be a change in antigen dominant epitope which is associated with decreased severity of illness. [12] However our observation were made based on very small number of H:j variants, thus it is not possible to claim otherwise. Furthermore, our study is in concordance with previous study in adult patients, which claimed that strains harboring z66 (*fliB*) occupy and silence the *fliC*-j chromosomal allele [13]. Gen *fliB* is known to be dominant in its expressions, and able to suppress the protein which is located in plasma pBSSB1 within *fli*C so it would not be expressed [22]. From twenty-two isolates in this study, two samples showed expression of *fli*C variants j were also the same sample, which expressed *fliB* and experienced 251 bp deletions. Strains harboring z66 antigen were observed to be associated with Indonesia, and yet there is no evidence of the bacterial spreading into other area where typhoid is endemic [13]. Further studies in Indonesia, which evaluate evolution of typhoid serotypes and virulence of circulating serotypes, are required.

Although we observed that cephalosporins were often prescribed for clinically diagnosed typhoid fever especially in primary health care where laboratory test is not feasible, this study is in concordance with other typhoid fever study in adult patients in Jakarta that most isolates found are still susceptible to 1st line antibiotics [23].

To our knowledge this is the first study of *S. typhi* serotype in children in Indonesia. This initial study could be used as pilot study for larger scale research.

The limitation of the study is related to the relative small numbers of confirmed typhoid fever cases, which contribute merely 15.4% of all clinically diagnosed typhoid fever. Thus, perhaps explain why this small number of isolates failed to show difference in clinical manifestation of typhoid fever in correlation to expression of j or d allele in the genes found in the isolate.

Conclusion

Confirmed case of typhoid fever based on *S. typhi* isolate finding in patients were only 22 out of 142 (15%) from all typhoid case diagnosed clinically by physicians. *S. typhi* serotypes currently circulating in children in Jakarta are fliC (H:j, and H:d) and fli-B, which are still sensitive towards chloramphenicol, ampicillin and co-trimoxazole. Therefore, these first-line antimicrobial should be prescribed at any time in children with typhoid fever where laboratory confirmation is limited.

Acknowledgements: We thank Dr. Stephen Baker from Oxford Unit Clinical Research Unit (OUCRU), Vietnam for supplying control strains and information that have assisted with this study. This study was funded by The Li-kai-Shing foundation, and received small equipment grants from The German Academic Exchange Service (DAAD).

Declaration of conflicting interests: The authors declared no conflicts of interest with respect to the authorship and/or authorship of this article.

REFERENCES

1. Crump JA, Luby SP, Mintz ED. The global burden of typhoid fever. Bull World Health Organ 2004;82:346-253.

2. World Health Organization. Background document: The diagnosis, treatment and prevention of typhoid fever. Geneva: The Organization; 2003. http://www.who.int/rpc/TFGuideWHO.pdf

3. Ochiai RL, Acosta CJ, Danovaro-Holliday MC, et al. A study of typhoid fever in five Asian countries: disease burden and implications for controls. Bull World Health Organ 2008; 86:260-268.

4. Centers for Disease Control and Prevention Indonesia. Ministry of Health Republic of Indonesia (2007) Typhoid fever prevalence.

5. Buckle GC, Walker CL, Black RE. Typhoid fever and paratyphoid fever: Systematic review to estimate global morbidity and mortality for 2010. J Glob Health 2012; 2:010401.

6. Parry CM, Threlfall EJ. Antimicrobial resistance in typhoidal and nontyphoidal salmonellae. Curr Opin Infect Dis. 2008; 21:531-538.

7. Rowe B, Ward LR, Threlfall EJ. Multidrug-resistant *Salmonella typhi*: a worldwide epidemic. Clin Infect Dis 1997; 24 (Suppl 1):S106-9.

8. Mirza SH, Beeching NJ, Hart CA. Multi-drug resistant typhoid: a global problem. J Med Microbiol 1996; 44:317-319.

9. Hatta M, Ratnawati N. Enteric fever in endemic areas of Indonesia: an increasing problem of resistance. J Infect Dev Ctries 2008;2:279-282.

10. Hatta M, Sultan AR, Pastoor R, Smits HL. New flagellin gene for *Salmonella enterica serovar Typhi* from the East Indonesian archipelago. Am J Trop Med Hyg 2011; 84:429-434.

11. Moshitch S, Doll L, Rubinfeld BZ, et al. Mono- and bi-phasic Salmonella typhi: genetic homogeneity and distinguishing characteristics. Mol Microbiol 1992; 6:2589-2597.

12. Grossman DA, Witham ND, Burr DH, et al. Flagellar serotypes of *Salmonella typhi* in Indonesia: relationships among motility, invasiveness, and clinical illness. J Infect Dis 1995; 171:212-216.

13. Baker S, Holt K, van de Vosse E, Roumagnac P, et al. High-throughput genotyping of *Salmonella enterica serovar Typhi* allowing geographical assignment of haplotypes and pathotypes within an urban District of Jakarta, Indonesia. J Clin Microbiol 2008; 46:1741-1746.

14. Le TA, Fabre L, Roumagnac P, Grimont PA, Scavizzi MR, Weill FX. Clonal expansion and microevolution of quinolone-resistant *Salmonella enterica serotype typhi* in Vietnam from 1996 to 2004. J Clin Microbiol 2007;45:3485-3492.

15. Banoo S, Bell D, Bossuyt P, et al. Evaluation of diagnostic tests for infectious diseases: general principles. Natur Rev Microbiol 2006;4 (Suppl 9):S21-S31.

16. Soenarto Y, Aman AT, Bakri A, et al. Burden of severe rotavirus diarrhea in indonesia. J Infect Dis. 2009;200 (Suppl 1):S188-S194.

17. Dewan AM, Corner R, Hashizume M, Ongee ET. Typhoid Fever and its association with environmental factors in the Dhaka Metropolitan Area of Bangladesh: a spatial and time-series approach. PLoS Negl Trop Dis 2013; 7(1):e1998.

18. Tam FC, Ling TK, Wong KT, Leung DT, Chan RC, Lim PL. The TUBEX test detects not only typhoid-specific antibodies but also soluble antigens and whole bacteria. J Med Microbiol 2008; 57:316-323.

19. Clegg A, Passey M, Omena M, Karigifa K, Suve N. Re-evaluation of the Widal agglutination test in response to the changing pattern of typhoid fever in the highlands of Papua New Guinea. Acta Tropica 1994; 57:255-263.

20. Gasem MH, Dolmans WM, Isbandrio BB, Wahyono H, Keuter M, Djokomoeljanto R. Culture of *Salmonella typhi* and *Salmonella paratyphi* from blood and bone marrow in suspected typhoid fever. Trop Geograph Med 1995; 47:164-167.

21.Gonsalves WI, Cornish N, Moore M, Chen A, Varman M. Effects of volume and site of blood draw on blood culture results. J Clin Microbiol 2009; 47:3482-3485.

22. Baker S, Hardy J, Sanderson KE, et al. A novel linear plasmid mediates flagellar variation in *Salmonella typhi*. PloS Pathogen 2007; 3:605-610.

23. Moehario LH, Tjoa E, Kalay V, Abidin A. Antibiotic susceptibility patterns of *Salmonella typhi* in Jakarta and surounding areas. In: Kumar Y (Edt) Salmonella a diversified superbug. Rijeka: InTech 2012. p: 91-98.