Usuda et al., J Clin Case Rep 2017, 7:9
DOI: 10.4172/2165-7920.10001021

Journal of Clinical Case Reports

Case Report Open Access

Gastroenteritis in Japanese Females Caused by *Shigella flexneri* without Definite Infection Route: A Case Report and Mini-Review of Literature

Daisuke Usuda[1,2]*, Shinya Yamada[3], Kento Takeshima[2], Ryusho Sangen[2], Masaharu Iguchi[3], Yasuhiro Kawai[1], Yuji Kasamaki[2], Yoshitsugu Iinuma[1] and Tsugiyasu Kanda[2]

[1]*Department of Infectious Diseases, Kanazawa Medical University, Japan*
[2]*Department of Community Medicine, Kanazawa Medical University Himi Municipal Hospital, Japan*
[3]*Department of Respiratory Medicine, Kanazawa Medical University Himi Municipal Hospital, Japan*

Abstract

In Japan, the incidence of Shigellosis is low at approximately 150 cases per year, with the majority of these being imported, mainly from Asia. Sporadic domestic occurrences are also reported, the majority of which absent epidemiological association. We report a case of gastroenteritis caused by *Shigella flexneri* in a 60-year-old Japanese female who recovered fully following treatment with levofloxacin. While we were unable to identify the route of infection, we suspected imported frozen marine products that may have been handled or processed in an unsafe manner. Shigellosis should be included in the differential diagnosis when presented with a patient exhibiting symptoms of gastrointestinal infection, even in the absence of a history of travel to an endemic area.

Keywords: *Shigella flexneri*; Japan; Infection route

Introduction

Shigellosis is a significant problem throughout the world with the highest prevalence in tropical and subtropical climates, particularly in areas where living standards may be low and access to sufficient safe drinking water and proper excreta disposal systems are limited [1]. Bacterial dysentery due to *Shigella* species is a major cause of morbidity and mortality with 165 million cases occurring annually worldwide and 1 million associated deaths. *Shigella* is the third leading cause of diarrhea mortality in children under five years of age [2].

In Japan, the number of reported cases had been decreasing annually, and the rate has remained generally flat since 2013 at approximately 150 cases per year [3]. Most of these are imported cases, especially from Asia, including India, Indonesia, the Philippines, and Cambodia, in decreasing order of occurrence [3]. However, domestic cases are also reported [3]. According to bacterial strain, reported cases of *Shigella sonnei* are most frequent followed by *S. flexneri* [3,4]. Domestic outbreak due to *S. sonnei* occurred at a kindergarten in 2014, and an elementary school in 2015. Domestic outbreak due to *S. flexneri* has not been reported [3]. With the exception of outbreak, the majority of domestic cases have occurred sporadically with unknown epidemiological association [3]. As far as we know, there have been no reported cases of domestic sporadic gastroenteritis caused by *Shigella,* including *S. flexneri*, with identified infectious route in Japan. We report a case with a brief review of literature.

Case Report

In June, a 60-year-old Japanese female presented to our hospital complaining of sudden-onset physical fatigue, fever, epigastralgia, vomiting and watery diarrhea. She was well nourished and reported her occupation as housewife. She visited our hospital regularly for hypertension, diabetes mellitus, and hyperlipidemia, and was under treatment with aliskiren, azelnidipine, sitagliptin, and rosuvastatin. She was negative for immunodeficiency syndrome, and denied the use of tobacco or alcohol. She had no history of travel abroad for the past ten years, and denied unspecified sex partners. She had received a medical check-up four month prior to her visit and no abnormality had been detected. She had no history of eating raw or undercooked food for the past 7 days. She was married and lived with her husband of the same age, who was retired and had no relevant medical history. Her husband and neighbors were negative for similar symptoms.

Her vital signs revealed tachycardia and high-grade fever. Physical examination revealed tenderness to the epigastric fossa. Routine blood tests showed leukocytosis, elevation of C-reactive protein and decline of sodium and potassium, while the results of urinalysis were within normal limits. Plain chest-abdominal computed tomography showed thickening mucosa of the ascending colon and small intestine (Figure 1). A tentative diagnosis of gastroenteritis without definite suspected infection route was made and she was admitted under contact infection precautions.

Sulbactam/ampicillin 9 g/day was started as empiric therapy; however, fever and abdominal symptoms persisted. 2/2 sets of blood culture test taken at admission were negative. Cystic forms of *Entamoeba histolytica* were not observed on microscopic examination of stool, and serological fluorescent antibody technique were negative for amebic antibody. On day 3 of hospitalization, colonies grown on bromothymol

Figure 1: Chest-abdominal computed tomography in admission.

***Corresponding author:** Daisuke Usuda, Department of Infectious Diseases, Kanazawa Medical University, Ishikawa-ken, Japan, Tel: +81-76-218-8452; Fax: +81-76-218-8453; E-mail: united19771108@yahoo.co.jp

Received August 07, 2017; **Accepted** September 20, 2017; **Published** September 25, 2017

Citation: Usuda D, Yamada S, Takeshima K, Sangen R, Iguchi M, et al. (2017) Gastroenteritis in Japanese Females Caused by *Shigella flexneri* without Definite Infection Route: A Case Report and Mini-Review of Literature. J Clin Case Rep 7: 1021. doi: 10.4172/2165-7920.10001021

Copyright: © 2017 Usuda D, et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

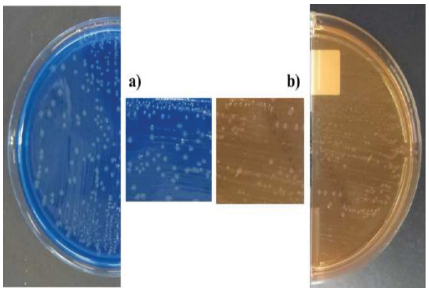

Figure 2: Colonies grown on bromothymol blue lactate agar (a) and Salmonella-Shigella agar (b) using patient's stool.

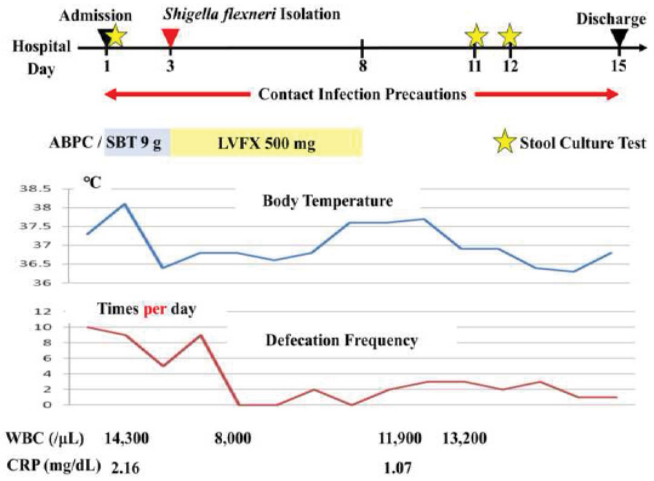

Figure 3: Clinical course of the patient.

blue lactate agar and Salmonella-Shigella agar using stool resulted in a diagnosis of gastroenteritis caused by *S. flexneri* (Figure 2). It was resistant to sulbactam/ampicillin by drug susceptibility testing, and we changed antibiotics to 500 mg/day intravenous administration of levofloxacin. On the same day, her fever resolved and her abdominal symptoms lessened. She was started on oral ingestion on day 4 hospitalization. Later, levofloxacin was terminated on day 7 of hospitalization. She complained of epigastric distress and fever on day 8 of hospitalization. Her temperature rapidly decreased and abdominal symptoms resolved without antibiotics or antipyretic drugs. On day 9 of hospitalization, esophagogastroduodenoscopy revealed normal findings. Stool cultures taken on day 11 and 12 of hospitalization were negative. The patient recovered fully and was discharged on day 15 of hospitalization without the need for contact infection precautions (Figure 3). We confirmed her continuing health on outpatient follow up two weeks after discharge. The infection route remained unknown.

Discussion

This case was neither imported nor the result of outbreak at a restaurant or institution, which are the most common cause of infection in Japan. While the infection route was not identified, there was a report of a mass gastroenteritis outbreak caused by *Shigella* spp in which imported frozen marine products including attached ice was suspected as the source of infection. In addition, unsafe food handling and processing can serve as a vehicle for transmission [4-6]. From this point of view, we suspected that contaminated imported frozen marine products might have been the route of infection. On the other hand, shigellosis has been reported in men with a history of homosexual activity in developed countries, including Japan; therefore, attention to sexual behavior as a potential route of transmission is recommended [7,8].

Shigella, a gram-negative bacterium, is responsible for Shigellosis/bacillary dysentery; and four serotypes are seen in Asia: *S. flexneri*, *S. sonnei*, *S. dysenteriae*, and *S. boydii*. While these are a global concern, they predominate in developing countries [9]. *S. flexneri* is the most common species of *Shigella* causing diarrhea and dysentery in developing countries in Asia [10,11]. In Japan, *S. flexneri* is estimated to account for 18% of bacterial dysentery [12]. We have included a table of published articles related to *S. flexneri* in Asia from January 2014, searched in the PubMed database using the keywords *S. flexneri*, Asia, and human (Table 1). The search yielded 35 articles including a total 42,942 cases [9,10,13-38]. According to the table, the greatest number of articles came from China with 13 articles and 35,794 cases, followed by India with 6 articles and 972 cases.

Nation	Period	Population	Number of cases	Reference
China	From 2005 to 2010	All ages	27,023	[13]
China	From 2005 to 2014	All ages	4,009	[14]
China	From 2003 to 2013	Not specified	1,610	[15]
China	From 2006 to 2012	All ages	801	[16]
China	From January 2001 to December 2014	All ages	660	[9]
China	From January 2001 to December 2011	Not specified	624	[17,18]
China	From January 2004 to December 2011	All ages	569	[19]
China	From 2006 to 2011	All ages	132	[20]
China	From January 2008 to December 2012	Not specified	112	[21]
China	From January 2008 to December 2013	Children aged from birth –14 years	99	[22]
China	From 1997 to 2006	Not specified	59	[10]
China	From 1981 to 2010	Not specified	56	[23]
China	From 2005 to 2013	Not specified	40	[24]
India	From September to October 2007	All ages	461	[25]
India	From 2000 to 2004	Children aged <5 years	218	[26]
India	From January 2001 to December 2014	All ages	98	[9]
India	From 2009 to 2012	All ages	80	[27]
India	Over 3 years (Not specified)	Children aged from birth–59 months	47	[28]
India	From 1945 to 2012	All ages	8	[29]
Bangladesh	Over 3 years (Not specified)	Children aged from birth–59 months	415	[28]
Bangladesh	From 2006 to 2011	Not specified	200	[30]
Bangladesh	From July 2012 to June 2015	Children aged 2–59 months	88	[31]
Bangladesh and Vietnam	From 1995 to 2010	Not specified	33	[32]
Vietnam	From 2006 to 2009	Children aged <15 years	33	[33]
Vietnam	From January 2001 to December 2014	All ages	7	[9]
Iran	From January 2001 to December 2014	All ages	454	[9]
Iran	From June 2013 to August 2014	Not specified	31	[34]
Iran	From 2008 to 2010	Pediatric patients	15	[35]

Pakistan	From January 2001 to December 2014	All ages	242	[9]
Pakistan	Over 3 years (Not specified)	Children aged from birth–59 months	84	[28]
Pakistan	From December 2011 to November 2013	All ages	25	[36]
Israel	From 1998 to 2012	All ages	3,428	[37]
Israel	From 2005 to 2009	All ages	1,058	[38]
Nepal	From January 2001 to December 2014	All ages	99	[9]
Thailand	From January 2001 to December 2014	All ages	24	[9]
Total			42,942	

Table 1: Published articles of *Shigella flexneri* in Asia from January 2014.

Shigella organisms can survive transit through the stomach due to their lower susceptibility to acid than another bacteria exhibit. For this reason, as few as 10 to 100 organisms can cause disease [39]. There are several virulence factors contributing to the colonization and invasion of epithelial cells and the eventual death of host cells [34]. *Shigella* also has the ability to carry multiple plasmids in relation to virulence [40]. The ial gene was the most frequent virulence gene among isolated bacterial strains followed by ipaH, set1B, and set1A; and *S. flexneri* possessed all (ial 66%, ipaH 59%, set1A 12%, and set1B 22%) [35].

Patients with *Shigella* gastroenteritis typically present with high fever, abdominal cramps, and bloody, mucoid diarrhea: fever (30% to 40%), abdominal pain (70% to 93%), mucoid diarrhea (70% to 85%), bloody diarrhea (35% to 55%), watery diarrhea (30% to 40%), and vomiting (35%) [41]. Both severe intestinal disease and extra-intestinal manifestations of shigellosis occur with infection by any of the four species of *Shigella*; however, they are most common with *S. dysenteriae* type 1 [41]. Among several severe extra intestinal manifestations, encephalopathy is one of the most common complications; and it has been associated with a higher rate of death [31]. The common manifestations of *Shigella* encephalopathy are seizure, altered consciousness and coma [31]. Encephalopathy is usually reversible; however, it can sometimes fulminate and be fatal [31].

The magnitude of shigellosis in developing countries is largely unknown because an affordable detection method is not available [42]. Current laboratory diagnosis of *Shigella* spp. is laborious, time-consuming and has low sensitivity [42]. Hence, a molecular-based diagnostic assay which simultaneously amplifies four specific genes to identify *Shigella* genus was developed in a single reaction to detect and differentiate *Shigella* spp [42]. Validation with 120 *Shigella* strains and 37 non-*Shigella* strains yielded 100% specificity [42]. In addition, the sensitivity of pentaplex polymerase chain reaction (PCR) assay was further improved following preincubation of stool samples in gram-negative broth [42]. The developed pentaplex PCR assay is robust and is capable of providing information about target genes essential for the identification of the *Shigella* genus and the three *Shigella* species responsible for the majority of shigellosis cases [42]. Pulsed-field gel electrophoresis (PFGE) is convenient for evaluation of the clonal relatedness of the strains; however, in such geographical areas where the same clone is in circulation, use of antibiotic resistance typing and/or plasmid profile analysis together with PFGE would be useful for precise discrimination of *Shigella* strains [43,44].

Antibiotic treatment of shigellosis is required to prevent mortality [45]. Patients are often treated with antimicrobial agents to reduce the duration of illness, and the treatment is known to reduce the period of *Shigella* excretion [46]. Antibiotic susceptibility testing should be performed at the time of stool and/or blood culture, and at the start of outbreak; and antibiotic use should be restricted to cases of *Shigella* classified as severe based on resistance pattern variations observed in different regions [24]. Although antimicrobial susceptibility was considerably high to most commonly used drugs, genetic determinants were variable between countries over time; therefore, the data stress the need for a more careful selection of antibiotics in the treatment of shigellosis [47]. The majority of *S. flexneri* is sensitive to 3rd generation cephalosporins, and quinolones, especially ciprofloxacin, are commonly employed in the treatment of shigellosis [36,46]. However, in recent years, some *Shigella* strains have rapidly developed resistance to antimicrobial agents [9,23,46,48]. For example, more than 80.0% of isolates of *S. flexneri* have proven resistant to ampicillin, nalidixic acid, and tetracycline; and in China from 2004 to 2014, *Shigella* isolates were highly resistant to nalidixic acid (89%), ampicillin (89%), tetracycline (88%), and sulfamethoxazole (83%). During the study period, isolates resistant to ciprofloxacin and cefotaxime increased, respectively, from 9 and 8% in 2005 to 4 and 30% in 2014 [14,17,18]. Multi-drug-resistant *Shigella,* including extended spectrum β-lactamases producing *Shigella,* is also emerging; and has the potential to become a serious threat to public health [11]. Here, several mechanisms are responsible for the development of antimicrobial resistance in *Shigella* strains [46]. One prominent mechanism is the mutation of the genes related to antimicrobial resistance [46]. For example, about quinolone resistance, the target regions where mutations occur in the organism have been identified as topoisomerase IV (parC and parE) and DNA gyrase (gyrA and gyrB) [18,46]. Another mechanism depends on plasmid-mediated antimicrobial resistance [40,46]. For example, *S. flexneri* is divided into at least 19 serotypes, the majority of which being modifications of the same basic O-antigen by glucosylation and/or O-acetylation of its sugar residues by phage encoded serotype-converting genes [49]. Plasmid-encoded gene modification is an important serotype conversion mechanism in *S. flexneri*, in addition to glucosylation and O-acetylation [49]. Furthermore, quinolone resistance (PMQR) determinants are qnr, aac(6')-Ib-cr, and qepA [40,46]. Considering the above, monitoring of the drug resistance of Shigella strains should be strengthened, and rational use of antibiotics is required [17,50]. In addition, guidelines for therapy should be monitored and modified based on regional reports of resistance to antimicrobial agents [11].

Overall fatality rate was 10% and did not differ significantly by species; and the case-fatality-rate was significantly higher among the children with *Shigella* encephalopathy accompanied by symptoms including of documented seizure and unconsciousness, malnutrition, hyponatremia, and lower stool frequency [31,41]. Therefore, early identification and aggressive management may help to reduce morbidity and death, especially in resource-limited settings [31].

Conclusion

In summary, we experienced a case of gastroenteritis caused by *S. flexneri* whose infection route we were unable to identify; however, we suspected imported frozen marine products. The incidence is low in Japan although sporadic cases or outbreaks are sometimes confirmed. Therefore, we concluded that it should be included in the differential diagnosis when encountering patients with symptoms of gastrointestinal infection, even in the absence of a history of travel to an endemic area. In addition, constant epidemiological surveillance, improvement of personal hygiene and environmental sanitation are recommended for control of pathogen infection, especially for food handlers.

References

1. Abera G (2004) Shigellosis in Ethiopia: Review of studies conducted since 1974. Ethiop J Biol Sci 3: 191-235.

2. Kotloff KL, Winickoff JP, Ivanoff B, Clemens JD, Swerdlow DL, et al. (1999) Global burden of Shigella infections: Implications for vaccine development and implementation of control strategies. Bull World Health Organ 77: 651.

3. https://www.niid.go.jp/niid/ja/route/intestinal/1469-idsc/iasr-news/6818-441p03.html

4. https://www.niid.go.jp/niid/ja/iasr/510-surveillance/iasr/graphs/1524-iasrgb.html

5. Nishiyama M, Yano K, Tanimatsu T, Takahashi S, Miyawaki Y (2001) An Epidemic of dysentery due to *Shigella flexneri* Ib, S. flexneri IIIa, IVa. Igakukensa 50: 933-936.

6. Mama M, Alemu G (2016) Prevalence, antimicrobial susceptibility patterns and associated risk factors of Shigella and Salmonella among food handlers in Arba Minch University, South Ethiopia. BMC Infect Dis 21: 686.

7. Okame M, Adachi E, Sato H, Shimizu S, Kikuchi T, et al. (2012) *Shigella sonnei* outbreak among men who have sex with men in Tokyo. Jpn J Infect Dis 65: 277-278.

8. Borg ML, Modi A, Tostmann A, Gobin M, Cartwright J, et al. (2012) Ongoing outbreak of *Shigella flexneri* serotype 3a in men who have sex with men in England and Wales, data from 2009-2011. Euro Surveill 29: 17.

9. Kahsay AG, Muthupandian S (2016) A review on Sero diversity and antimicrobial resistance patterns of Shigella species in Africa, Asia and South America, 2001-2014. BMC Res Notes 30: 422.

10. Zhang N, Lan R, Sun Q, Wang J, Wang Y, et al. (2014) Genomic portrait of the evolution and epidemic spread of a recently emerged multidrug-resistant *Shigella flexneri* clone in China. J Clin Microbiol 52: 1119-1126.

11. Nath R, Saikia L, Choudhury G, Sharma D (2013) Drug resistant *Shigella flexneri* in and around Dibrugarh, north-east India. Indian J Med Res 137: 183-186.

12. http://www.nih.go.jp/niid/en/

13. Zhang H, Si Y, Wang X, Gong P (2016) Patterns of bacillary dysentery in China, 2005-2010. Int J Environ Res Public Health 27: 164.

14. Chang Z, Zhang J, Ran L, Sun J, Liu F, et al. (2016) The changing epidemiology of bacillary dysentery and characteristics of antimicrobial resistance of Shigella isolated in China from 2004-2014. BMC Infect Dis 18: 685.

15. Qiu S, Xu X, Yang C, Wang J, Liang B, et al. (2013) Shift in serotype distribution of Shigella species in China, 2003-2013. Clin Microbiol Infect 21: 252.e5-8.

16. Zhang J, Wang F, Jin H, Hu J, Yuan Z, et al. (2015) Laboratory monitoring of bacterial gastroenteric pathogens Salmonella and Shigella in Shanghai, China 2006-2012. Epidemiol Infect 143: 478-485.

17. Xu Y, Zhuang L, Kang H, Ma P, Xu T, et al. (2016) Prevalence, resistance patterns, and characterization of integrons of *Shigella flexneri* isolated from Jiangsu Province in China, 2001-2011. Eur J Clin Microbiol Infect Dis 35: 1347-1353.

18. Qin T, Bi R, Fan W, Kang H, Ma P, et al. (2016) Novel mutations in quinolone resistance-determining regions of gyrA, gyrB, parC and parE in *Shigella flexneri* clinical isolates from eastern Chinese populations between 2001 and 2011. Eur J Clin Microbiol Infect Dis 35: 2037-2045.

19. Zhang J, Jin H, Hu J, Yuan Z, Shi W, et al. (2014) Antimicrobial resistance of *Shigella* spp. from humans in Shanghai, China, 2004-2011. Diagn Microbiol Infect Dis 78: 282-286.

20. Zhang Y, Zhao Y, Ding K, Wang X, Chen X, et al. (2014) Analysis of bacterial pathogens causing acute diarrhea on the basis of sentinel surveillance in Shanghai, China, 2006-2011. Jpn J Infect Dis 67: 264-268.

21. Zhang CL, Liu QZ, Wang J, Chu X, Shen LM, et al. (2014) Epidemic and virulence characteristic of *Shigella* spp. with extended-spectrum cephalosporin resistance in Xiaoshan District, Hangzhou, China. BMC Infect Dis 15: 260.

22. Zhang H, Pan F, Zhao X, Wang G, Tu Y, et al. (2015) Distribution and antimicrobial resistance of enteric pathogens in Chinese paediatric diarrhoea: a multicentre retrospective study, 2008-2013. Epidemiol Infect 143: 2512-2519.

23. Wang J, Liang F, Wu XM, Qi W. (2014) Transfer patterns of integron-associated and antibiotic resistance genes in S. flexneri during different time intervals in Tianjin, China. Indian J Med Microbiol 32: 256-260.

24. Cui X, Yang C, Wang J, Liang B, Yi S, et al. (2015) Antimicrobial Resistance of *Shigella flexneri* serotype 1b isolates in China. PLoS One 3: e0129009.

25. Taneja N, Mewara A (2016) Shigellosis: Epidemiology in India. Indian J Med Res. 143: 565-576.

26. Dutta S, Jain P, Nandy S, Matsushita S, Yoshida S (2014) Molecular characterization of serologically atypical provisional serovars of Shigella isolates from Kolkata, India. J Med Microbiol 63: 1696-1703.

27. Aggarwal P, Uppal B, Ghosh R, Krishna Prakash S, Chakravarti A, et al. (2016) Multi drug resistance and extended spectrum beta lactamases in clinical isolates of Shigella: A study from New Delhi, India. Travel Med Infect Dis 14: 407-413.

28. Livio S, Strockbine NA, Panchalingam S, Tennant SM, Barry EM, et al. (2014) Shigella isolates from the global enteric multicenter study inform vaccine development. Clin Infect Dis 59: 933-941.

29. Appannanavar SB, Goyal K, Garg R, Ray P, Rathi M, et al. (2014) Shigellemia in a post renal transplant patient: A case report and literature review. J Infect Dev Ctries 13: 237-239.

30. Iqbal MS, Rahman M, Islam R, Banik A, Amin MB, et al. (2014) Plasmid-mediated sulfamethoxazole resistance encoded by the sul2 gene in the multidrug-resistant *Shigella flexneri* 2a isolated from patients with acute diarrhea in Dhaka, Bangladesh. PLoS One 9: e85338.

31. Afroze F, Ahmed T, Sarmin M, Smsb Shahid A, Shahunja KM, et al. (2017) Risk factors and outcome of Shigella encephalopathy in Bangladeshi children. PLoS Negl Trop Dis 28: e0005561.

32. Baker KS, Dallman TJ, Ashton PM, Day M, Hughes G, et al. (2015) Intercontinental dissemination of azithromycin-resistant shigellosis through sexual transmission: a cross-sectional study. Lancet Infect Dis 15: 913-921.

33. Thompson CN, Thieu NT, Vinh PV, Duc AN, Wolbers M, et al. (2016) Clinical implications of reduced susceptibility to fluoroquinolones in paediatric Shigella sonnei and *Shigella flexneri* infections. J Antimicrob Chemother 71: 807-815.

34. Hosseini Nave H, Mansouri S, Emaneini M, Moradi M (2016) Distribution of genes encoding virulence factors and molecular analysis of Shigella spp. isolated from patients with diarrhea in Kerman, Iran. Microb Pathog 92: 68-71.

35. Ranjbar R, Bolandian M, Behzadi P (2017) Virulotyping of Shigella spp. isolated from pediatric patients in Tehran, Iran. Acta Microbiol Immunol Hung 1: 71-80.

36. Raza A, Mahboob A, Nizammudin S, Nazeer SH, Sultan F (2016) Shigella infections: A two year experience in cancer patients. J Pak Med Assoc 66: 37-39.

37. Cohen D, Bassal R, Goren S, Rouach T, Taran D, et al. (2014) Recent trends in the epidemiology of shigellosis in Israel. Epidemiol Infect 142: 2583-2594.

38. Peleg I, Givon-Lavi N, Leibovitz E, Broides A (2014) Epidemiological trends and patterns of antimicrobial resistance of Shigella spp. isolated from stool cultures in two different populations in Southern Israel. Diagn Microbiol Infect Dis 78: 287-291.

39. DuPont HL, Levine MM, Hornick RB, Formal SB (1989) Inoculum size in shigellosis and implications for expected mode of transmission. J Infect Dis 159: 1126.

40. Mandal J, Sangeetha V, Nivedithadivya, Das A, Parija SC. (2013) Characterization of extended-spectrum beta-lactamase-producing clinical isolates of *Shigella flexneri*. J Health Popul Nutr 31: 405-408.

41. Khan WA, Griffiths JK, Bennish ML (2013) Gastrointestinal and extra-intestinal manifestations of childhood shigellosis in a region where all four species of Shigella are endemic. PLoS One 17: e64097.

42. Ojha SC, Yean Yean C, Ismail A, Singh KK (2013) A pentaplex PCR assay for the detection and differentiation of Shigella species. Biomed Res Int 2013: 412370.

43. Eftekhari N, Bakhshi B, Pourshafie MR, Zarbakhsh B, Rahbar M, et al. (2013) Genetic diversity of Shigella spp. and their integron content. Foodborne Pathog Dis 10: 237-242.

44. Saran B, Erdem B, Tekeli FA, Sahin F, Aysev AD. (2013) Characterization of Shigella strains isolated in ankara, Turkey by antimicrobial resistance models, plasmid profile analysis and pulsed-field gel electrophoresis. Mikrobiyol Bul 47: 35-48.

45. Mamatha B, Rituparna C (2012) Decreased susceptibility to antimicrobials among *Shigella flexneri* isolates in Manipal, South India-A 5-year hospital based study. Southeast Asian J Trop Med Public Health 43: 1447-1451.

46. Jeon YL, Nam YS, Lim G, Cho SY, Kim YT, et al. (2012) Quinolone-resistant

Shigella flexneri isolated in a patient who travelled to India. Ann Lab Med 32: 366-369.

47. Ahmed SF, Klena J, Husain T, Monestersky J, Naguib A, et al. (2013) Genetic characterization of antimicrobial resistance of *Shigella flexneri* 1c isolates from patients in Egypt and Pakistan. Ann Clin Microbiol Antimicrob 2: 9.

48. Thompson CN, Anders KL, Nhi Le TQ, Tuyen HT, Van Minh P, et al. (2014) A cohort study to define the age-specific incidence and risk factors of Shigella diarrhoeal infections in Vietnamese children: A study protocol. BMC Public Health 17: 1289.

49. Sun Q, Lan R, Wang J, Xia S, Wang Y, et al. (2013) Identification and characterization of a novel *Shigella flexneri* serotype Yv in China. PLoS One 30: e70238.

50. Gu B, Zhou M, Ke X, Pan S, Cao Y, et al. (2015) Comparison of resistance to third-generation cephalosporins in Shigella between Europe-America and Asia-Africa from 1998 to 2012. Epidemiol Infect 143: 2687-2699.