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## Prevalence and antimicrobial susceptibility pattern of Salmonella and Shigella among food handlers in catering establishments at Debre Markos University, Northwest Ethiopia



Abeba Mengist\*, Getachew Mengistu, Alemayehu Reta

Department of Medical Laboratory Science, College of Medicine and Health Sciences, Debre Markos University, Debre Markos, Ethiopia

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#### ABSTRACT

Background: Food-borne diseases are a major health problem in developing countries including Ethiopia. This study determined the prevalence and antibiotic susceptibility patterns of Shigella and Salmonella among food handlers working in student and staff food service establishments at Debre Markos University.

Methods: A laboratory-based cross-sectional study was conducted among 220 food handlers from January 2015 to June 2016. Stool and fingernail samples from the subjects were cultured on bacteriological culture medium, and Shigella and Salmonella were isolated and identified following standard procedures. Antimicrobial susceptibility testing was performed for all isolates using the Kirby–Bauer disk diffusion method.

Results: The overall prevalence of Shigella and Salmonella in this study was 5.9%, with 3.6% of stool specimens testing positive for Salmonella species and 2.3% testing positive for Shigella species. None of the food handlers had positive cultures for Shigella or Salmonella in respect of their fingernail specimens. The isolation of either Shigella or Salmonella had a significant inverse relationship with the number of service years (p = 0.017). All isolates of Shigella and Salmonella were 100% susceptible to ciprofloxacin, norfloxacin, and gentamicin. However, all isolated pathogens were resistant to ampicillin (100%). Conclusions: The findings of this study highlight the importance of food handlers in the transmission of

Conclusions: The findings of this study highlight the importance of food handlers in the transmission of pathogens to the customers (students and the general population). Screening of food handlers, training for food handling and hand hygiene practices, and regular monitoring of the food handling practices should be done in order to avoid diseases that can be acquired through improper food handling, like bacterial infections.

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#### Introduction

Food-borne diseases remain a major public health problem across the globe. The problem is severe in developing countries due to difficulties in securing optimal hygienic food handling practices. In developing countries, up to 70% of cases of diarrheal disease are associated with the consumption of contaminated food (Zeru and Kumie, 2007). The health status of the food handlers, their personal hygiene, and their knowledge and practice of food hygiene play an important role in food contamination (Mudey et al., 2010).

E-mail address: abymaa@gmail.com (A. Mengist).

Food can be contaminated by physical, chemical, and microbiological agents. The microbial agents responsible for food-borne diseases are bacteria, viruses, parasites, and fungi. Poisoning represents another food-borne disease and is caused by harmful toxins or chemicals, such as poisonous mushrooms and enterotoxins secreted by some bacteria (CDC, 2005). Therefore, food handlers who harbor and excrete microbial agents may contaminate foods by transmission from their faces via their fingers into the food processing chain, and finally to healthy individuals (Kaferstein and Abdussalam, 1999). Compared to other parts of the hand, the area beneath the fingernails harbors the most microorganisms and is most difficult to clean (Jones and Angulo, 2006).

The bacteria important to transmission by food handlers include Salmonella, Campylobacter, Listeria, pathogenic *Escherichia coli*, Yersinia, Shigella, Enterobacter, and Citrobacter (CDC, 2005).

<sup>\*</sup> Corresponding author at: Department of Medical Laboratory Science, College of Medicine and Health Sciences, Debre Markos University, P.O. Box 269, Debre Markos, Ethiopia.

Food-borne diseases are a serious threat to people in Africa, with an intolerable public health burden and causing massive economic losses. According to the most recent World Health Organization (WHO) estimates, 700 000 deaths per year in Africa are due to food and water-borne related diseases. These outbreaks only represent the tip of the iceberg, as many more cases that are sporadic have gone unreported (Mohammedaman and Getaneh, 2016).

A sudden outbreak of food poisoning due to Salmonella Newport occurred at the University of Gondar, Ethiopia (Asefa et al., 1994). Accordingly, 23% of students manifested symptoms of the disease, and Salmonella Newport was isolated from the stool of six students and three food handlers in the study area (University of Gondar) (Asefa et al., 1994). In Ethiopia, a country subject to more aggravated situations and challenges, food safety issues are not well understood and have received little attention. The aim at this study was, therefore, to assess the prevalence of Salmonella and Shigella and to evaluate drug resistance patterns of the isolates among food handlers in catering establishments at Debre Markos University, in the northwest of Ethiopia.

#### Materials and methods

Study design, area, period, and participants

This laboratory-based cross-sectional survey included 220 food handlers working in student cafeterias and staff lounges at Debre Markos University during the period from January 2015 to June 2016. The university is found in the northwestern part of Ethiopia in the town of Debre Markos. This town is located 300 km northwest of Addis Ababa.

#### Data collection

This study was approved by the Ethics Review Board of Debre Markos University. Data were collected by the data collectors after obtaining written informed consent using a well-structured questionnaire designed to obtain socio-demographic data and other relevant data related to the food handlers' number of years of service, medical screening status, status of certification, status of diarrhea, education, and hand-hygiene practices. Participants with a history of using antibiotic(s) 2 weeks prior to the study were excluded. Those food handlers positive for Shigella or Salmonella infection were informed to take appropriate treatment based on standard guidelines.

#### Sample collection and transport

Sterile cotton swabs (Puritan Low-Lint Cylindrical Tip) were prepared and dipped into saline-containing sterile test tubes to collect samples from the fingernails. Using these moistened cotton swabs, fingernail contents were collected from both hands of each subject by rubbing all over the surface under the nails. These samples were kept with normal saline in a test tube until they were inoculated onto the respective culture media; the delay between sample collection and inoculation was not more than 5 min. A stool specimen was collected from each food handler in a clean stool cup. Instructions were given to the participants on how to collect a proper stool sample (Esparar et al., 2004).

Stool sample culture and identification of Salmonella and Shigella species

All of the samples were enriched in Selenite F broth for 18 h prior to inoculating onto Salmonella–Shigella agar plates (Oxoid, Hampshire, UK) (World Health Organization, 2018). After 24 h of

incubation at  $37\,^{\circ}$ C, isolates were identified following standard procedures using biochemical tests such as indole, motility, lysine decarboxylase, triple sugar iron agar, citrate, and urea (Vandepitte and Verhaegen, 2003).

#### Antimicrobial susceptibility testing

Antimicrobial susceptibility tests were performed on Müller–Hinton agar (Oxoid, Hampshire, UK) by disk diffusion method. The following antimicrobial agents were used: ampicillin ( $10 \mu g$ ), tetracycline ( $30 \mu g$ ), chloramphenicol ( $30 \mu g$ ), gentamicin ( $10 \mu g$ ), norfloxacin ( $10 \mu g$ ), co-trimoxazole ( $25 \mu g$ ), and ciprofloxacin ( $10 \mu g$ ). Resistance and sensitivity was interpreted according to the National Committee for Clinical Laboratory Standards, 2011).

#### Data processing and analysis

All statistical calculations were done using IBM SPSS Statistics for Windows version 20 (IBM Corp., Armonk, NY, USA). Descriptive statistics were computed to determine the rate of Shigella and Salmonella and other variables. The relationships between the presence of infection and various risk factors were tested using the Chi-square test. A p-value of  $\leq$ 0.05 was considered indicative of a statistically significant association.

#### Results

Socio-demographic data

A total of 220 individuals participated in this study, with a response rate of 95.83%. Among them, 69.1% were female and 30.9% were male. The age of the study participants ranged from 18 to 43 years (mean age  $25.1\pm4.1$  years). The majority (85.9%) of the participants were between the ages of 21 and 30 years. None of the study participants were illiterate. The majority of food handlers (45.9%) had 1–2 years of work experience. Out of the total participants, 15.5% were certified for training in food handling and preparation and 62.7% had previously undergone a medical checkup including stool examination (Table 1).

With regard to hand-washing practices, 97.3% of food handlers reported that they had a habit of hand-washing after using the toilet. However, few of the food handlers (10%) reported that they had a habit of hand-washing after touching different parts of their body (hair, nose, and ears) between handling food items (Table 2).

 $Pathogen\ prevalence\ and\ associated\ risk\ factors$ 

As shown in Table 3, 13 (5.9%) of the stool specimens tested positive for either *Shigella* or *Salmonella* species. *Salmonella* species (n = 8; 3.6%) had a higher frequency compared to *Shigella* species (n = 5; 2.3%), while no pathogen was isolated from the stool of 207 (94.1%) participants. None of the food handlers had positive cultures for Shigella or Salmonella in respect of their fingernail specimens.

Different factors were assessed for a possible association with pathogen (Shigella and Salmonella) infections among the study participants (Tables 2 and 4). The highest proportion of infection (10%) was seen among the age group of  $\leq$ 20 years and no pathogens were isolated from the age groups of 31–40 and 41–50 years; however, the difference was not statistically significant (p = 0.73), as shown in Table 4. All of the isolated pathogens were from those who were not certified in food preparation and handling.

As illustrated in Table 4, the isolation rate of potential foodborne bacteria in stool samples was relatively higher among food

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