

Prevalence of Escherichia Coli O157:H7 and Associated Factors in Under-five Children in Eastern Ethiopia

Dawit Kassaye Getaneh

Haramaya University

Lemessa Oljira Hordofa

Haramaya University College of Health and Medical Sciences

Desalegn Admassu Ayana

Haramaya University College of Health and Medical Sciences

Tesfaye Sisay Tessema

Addis Ababa University

Lemma Demissie Regassa (✉ esraeldemiss@gmail.com)

Haramaya University, Dire Dawa, Ethiopia <https://orcid.org/0000-0002-5461-5348>

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Abstract

Background: *Escherichia coli* O157:H7 is one of the most potent zoonotic pathogens that causes mild diarrhea and leads to hemolytic uremic syndrome or death. This study was aimed to assess the prevalence and determinants of *Escherichia coli* O157:H7 related to diarrhea among under-five children with acute diarrhea.

Methods: A cross-sectional study design was carried out in 2018 on 378 under-five-year children recruited randomly from Hospitals in Eastern Ethiopia. Stool specimens were collected and processed using enrichment, differential and selective medium. Among isolates, *Escherichia coli* O157:H7 was confirmed using latex test (Oxoid, Basingstoke, Hants, England). Factors associated with *E. coli* O157:H7 infection were identified using binary and multivariable logistic regression. Associations were reported by odds ratio with 95 % confidence interval.

Results: Magnitude of *E. coli* O157:H7 related diarrhea was 15.3% (95%CI: 11.8-19.5). The *E. coli* infection was positively associated with rural residence (AOR;3.75, 95%CI:1.26-11.20), consumption of undercooked meat (AOR;3.95, 95%CI: 1.23-12.67), raw vegetables and/or fruit juice (AOR;3.37, 95%CI:1.32-8.62), presence of bloody diarrhea (AOR;4.42, 95% CI:1.78-10.94), number of under-five children in a household (AOR;7.16, 95%CI: 2.90-17.70), presence of person with diarrhea in a household (AOR;4.22, 95% CI: 1.84-12.69), owning domestic animal (AOR;3.87, 95% CI: 1.48-10.12) and uneducated mother (AOR;3.14, 95%CI: 1.05-9.42).

Conclusion: The magnitude of *E. coli* O157:H7 related diarrhea among under-five children is relatively high in Eastern Ethiopia. The *E. coli* infection was associated with sanitation and hygiene in a household. Thus, education focused on food cooking and handling, child care and household sanitation associated with animal manure in rural resident children are helpful it.

Introduction

Diarrheal disease is the second leading cause of mortality and morbidity among under-five (U5) children of the developing world. It is responsible for the death of more than 1,400 children every day, and 88% of deaths are concentrated in South East Asia and sub-Saharan Africa [1]. In the past five years, community-based prevalence studies in different regions of Ethiopia have indicated the magnitude of diarrhea to be within the range from 8–36% [2–4].

Though *Escherichia coli* (*E. coli*) is a ubiquitous intestinal bacterial flora of animals and humans, there are several pathogenic stains associated with diarrhea, collectively called *diarrheagenic E. coli* namely; *enterohaemorrhagic E. coli* (EHEC), *enterotoxigenic E. coli*, *enteropathogenic E. coli*, *enteroaggregative E. coli*, *enteroinvasive E. coli*, and *diffusely adherent E. coli* [5]. The EHEC serotype *E. coli* O157:H7 produces a powerful toxin responsible for the severe cause of hemolytic uremic syndrome, end-stage renal failure, and even death in humans [6]. *E. coli* O157:H7 is responsible for 20% of foodborne outbreaks globally [7]. It is predominantly associated with childhood diarrhea with an isolate rate of 39.3% in Khuzestan

province of Iran [8], 33.6% in Norway [9], and up to more than 20% in Nigeria [10, 11] from a community-based prevalence studies.

Although there is a paucity of epidemiological information among children in Ethiopian, a prevalence of 14% was reported on a Hospital-based cross-sectional study at Bahir-Dar [12]. In low resource countries like Ethiopia, where childhood diarrheal disease is a common, significant unsanitary condition with livestock product consumption habits, close relationship with reservoir animals, poor health infrastructure, there is no comprehensive information about the Epidemiology of *E. coli* O157: H7. Hence, this study aims to determine the prevalence of *E. coli* O157: H7 and its associated factors among under-five children in Eastern Ethiopia.

Materials And Methods

Study Area and period

The study was carried out in Hospitals found in Eastern Ethiopia, namely Haramaya District Hospital (HH) and Hiwot-Fana Specialized University Hospital (HFSUH) from January to March 2018. HH is primary hospital located at approximately 509 km East of Addis Ababa, the capital City of Ethiopia and 19 Km ahead to reach Harar. Geographically, this area lies between 9⁰26'N latitudes and 42⁰ 01'E longitudes, at an altitude of 2047 meters above sea level. HFSUH, the largest government Hospital in Eastern Ethiopia, is found in Harar town, the capital City of Harari region. The region has a total population of 183,344, of which 92,258 were males and 91,086 females, with 99,321 (54.17%) of the population are urban inhabitants [13]. Geographically the region is situated between Latitude: 9°18'49" N and Longitude of 42°07'05" E at an elevation of 1917 m above sea level. The area has a mean temperature ranging from 10 to 18 °C with relative humidity of 65%. It receives an average annual rainfall of 800 mm with a bimodal distribution of the seasonal pattern peaking in mid-April and mid-August of the year; however, there is a variation from year to year.

Study Design

A Hospital-based cross-sectional study design was used to determine the prevalence of *E. coli* O157:H7 and its associated factors among children with acute diarrhea.

Population

All under-five children with acute diarrhea who visited HH and HFSUH were considered the source populations. From the source populations, those who selected to participate in the study were considered the study population.

Inclusion and Exclusion Criteria

All under-five children with acute diarrhea who visited HH and HFSUH during the study period were included. Those on antibiotics at the preceding week and the study period were excluded not to hide the etiology of diarrhea.

Sampling method and procedure

The sample size was determined based on the previous study findings [14] following single and double proportion formula for the magnitude of *E. coli* O157: H7 and the associated factors, respectively. As a result, 378 under-five-year children were planned to include. Two Hospitals (HFSUH and HH) from Eastern Ethiopia were selected randomly.

The sample size was proportionally allotted based on their patient case flow. A three-month (September, October, and November) mean diarrhea cases were estimated from registration books of under-five children to guess the probable case flow to each Hospital a day. Then the study subjects were recruited consecutively until the desired sample was reached (Fig. 1).

Data collection and Stool Sample Collection

After written informed voluntary assent was obtained, a structured pretested questionnaire was used and completed by two trained laboratory technicians in a face-to-face interview basis. Participants were also asked and guided by the laboratory technicians to give the stool sample. On-spot gross examination of stool samples was performed to note the type of diarrhea at the collection time. Swabs with visible stool staining were prepared and collected in a sterile hi-culture test tube with Buffered Peptone Water (BPW) (Oxoid Ltd., Hampshire, England) and labelled. Lastly, the sample was transported in an icebox within 24 hours of collection to the Haramaya University laboratory and stored at refrigeration temperature (+ 4 °C). However, all the samples were processed by well-experienced laboratory experts within two days of sample collection.

Laboratory Processing: Isolation and Identification of *E. coli* O157:H7 (S1)

The sample was enriched by Modified tryptone soy broth supplemented with Novobiocin (mTSB + N) at the ratio of 1:9 BPW to mTSB + N and incubated overnight at $41.5 \pm 0.5^{\circ}\text{C}$ (24 hrs.). All media required for the isolation and biochemical characterization of *E. coli* were prepared as per the manufacturer's instruction. The enriched sample was cultured aerobically at 37°C for 24 hours onto MacConkey Agar (IVD, UK) (for easy identification of lactose fermenting organisms) and Eosin Methylene Blue Agar (Oxoid LTD, Basingstoke, England), for easy identification of the green metallic sheen appearance characteristic of *E. coli* colonies. Colony morphology and reaction on agar media were noticed and registered. Biochemical tests were performed on presumptive *E. coli* colonies using the standard bacteriological procedures described by Quinn *et al.* (2011) [15]. Isolates with biochemical test results for indole, methyl red, Voges-Proskauer, and citrate utilization (IMViC) (+ + - -), respectively, were presumed as *E. coli* isolates (S1). Furthermore, chromogenic Rainbow Agar O157 (Biolog, Hayward, HUSA) and Sorbitol MacConkey containing cefixime and tellurite (CT-SMAC) (Oxoid, England) media were used to notice the typical *E. coli* O157: H7 colonies after overnight culture (24hrs) at 37°C . *E. coli* isolates that appeared pink on Rainbow O157 and non-sorbitol fermenting on CT-SMAC were presumed as presumptively *E. coli* O157: H7

colonies [16, 17]. Then confirmation of *E. coli* O157:H7 was performed using *E. coli* O157:H7 latex test (Oxoid, Basingstoke, Hants, England). A drop of test latex was dispensed near the edge of the circle on the reaction card. Using a loop, up to 10 separate presumptive colonies were emulsified in a drop of 0.85% saline solution to increase the probability of detecting *E. coli* O157:H7. Latex quality control was performed before samples were tested using control latex. Agglutination of the test latex within a minute was registered as a positive result.

Study Variables

Dependent/Outcome Variables: *E. coli* O157:H7 related diarrhea

Independent /Explanatory Variables

The explanatory variables included were the following: -

- Socio-demographic, behavioral, and environment-related factors concerning the study participant (under-five-year children) include; age, residence, sex, sharing a home with animals, animal manure contact, breastfeeding status, type of diarrhea, and food consumption habits.
- Socio-demographic, behavioral, and environmental factors related to the households of the child includes; the age of the mother, marital status of the mother, total family size, diarrhea in the household other than the sampled child, number of under-five children, hand washing habit, child food preparing condition, drinking water treatment method and per capita water consumption.

Data Quality Control

The questionnaire was pretested on 5% sample for one-week period. Two-day training was given to laboratory technicians to ensure their understanding of the data collection technique. Supervision was made daily and the filled questioner is collected back by checking its completeness on a daily base. During laboratory analysis, calibrated equipment and standard operating procedures were used. Culture media were prepared and sterilized based on the manufacturer's instruction. All the dispensed culture Media were checked for sterility before culturing the samples after 37°C overnight incubation. Appropriate standard strain, in this case, *E. coli* O157: H7 (ATCC 43894) was used as a positive control, and Biosafety cabinet level II was used during each stool culture and culture transfer. Double data entry was conducted and identified errors were corrected after revising the original questionnaire and recording format.

Data Analysis

The data were coded, entered, and cleaned using EpiData version 4.2. Then the data were exported into STATA version 14 for processing and analysis. Descriptive statistics: mean, standard deviation, and proportion were used to present the findings. Binary logistic regression was conducted and variables with $p \leq 0.25$ were selected as a candidate to be included in the multivariable logistic regression analysis in an attempt to control for potential confounding variables, and the adjusted odds ratio was determined. Collinearity between variables was also checked by the standard error, and Hosmer and Lemshow test

assured the model fitness. Throughout the data presentation, a p-value of less than 0.05 was considered statistically significant.

Results

Sociodemographic Characteristics

Data was collected from 365 (96.6% response rate), 206(56.4%) from HH, and 159 (43.6) were from HFSUH. Of the total, 191 (52.33%) were males. The mean age of children was 18.7 months (SD \pm 14.5) and 28.94 (SD \pm 5.68) for mothers. Most of the children's mothers were married (81.94%) and 70.7% rural residents. More than halve (200/365) had no formal education, and 79.45% (290/365) were housewives. A hundred sixty-seven (55.67%) Children's' fathers had no formal education, and 199 (66.33%) farmers (Table 1).

Table 1
 Characteristics of the sample (children) and their parents, Eastern
 Ethiopia, 2018.

Variables	Category	Magnitude (%)
Child Age (months)	≤ 12months	197 (53.97%)
	> 12months	168 (46.03)
Maternal age (years)	15-24yrs	108 (30.17)
	25-34yrs	161 (44.97)
	35 - 4	89 (24.86)
Child sex	Male	191 (52.33)
	Female	174 (47.67)
Maternal educational level	No formal	257 (70.41)
	Formal	108 (29.59)
Father's educational level	No formal	221(73.67)
	Formal	79 (26.33)
Mother occupation	House wife	290 (79.45)
	Merchant	48 (13.15)
	Farmer	14 (3.84)
	Government	13 (3.56)
Family size	≤ 4	193 (52.88)
	> 4	172 (47.12)
Number of U5 children in HH	0-1	224 (62.22)
	2-3	136 (37.78)
Residence	Rural	258 (70.68)
	Urban	107 (29.32)
Exclusive breast feeding	Yes	52 (14.40)
	No	309 (85.60)
Person with diarrhea in the house	No	239 (65.48)
	Yes	126 (34.52)

Notes: U5, under five years old, HH; household.

Regarding the household and environmental characteristics, 40% (146/365) of households have only one room, 81 (36.49) sharing room with animals. More than half households (253/365) had traditional latrine and 11.78% use open field as a latrine. The majority of the households (62.47%) get water from taps while 9% use surface water. Overall, 22.74% eat under-cooked meat, 23.20% uncooked vegetables, 12.60% drink raw milk, and 22.47% fruit juice. Household, environmental and behavioral characteristics are described in Table 2.

Table 2
Household, environmental and behavioral characteristics of study participants, Eastern Ethiopia, 2018.

Variables	Category	Magnitude (%)
Number of rooms in HH	One	146 (40.00)
	Two	150 (41.10)
	More than two	69 (18.90)
Animal ownership	No	143 (39.18)
	Yes	222 (60.82)
Sharing home with live stokes (n = 222)	No	141 (63.51)
	Yes	81 (36.49)
Type of latrine	Traditional	253 (69.32)
	Open field	43 (11.78)
	Public	36 (9.86)
	Ventilated improved pit	33 (9.04)
Source of water	Tap	228 (62.47)
	Well	104 (28.49)
	Surface	33 (9.04)
Handwashing means	Water only	206 (57.87)
	Water and ash	103 (28.93)
	Water and soap	47 (13.20)
Household water treatment	No	233 (64.36)
	Aqua tab	70 (19.34)
	Boiling	34 (9.39)
	Filtering	25 (6.91)
Eating under cooked meat	No	282 (77.26)
	Yes	83 (22.74)
Drinking raw milk	No	319 (87.40)
	Yes	46 (12.60)

Notes: HH; household

Variables	Category	Magnitude (%)
Eat raw vegetable	No	278 (76.80)
	Yes	84 (23.20)
Drink fruit juice	No	283 (77.53)
	Yes	82 (22.47)
Notes: HH; household		

Prevalence and associated factors of *E. coli* O157:H7 related diarrhea

Upon laboratory investigation, *E. coli* O157:H7 prototype was found in 15.3% (95% CI: 11.8–19.4) children. Almost half of the sample (181/365) was presented with watery diarrhea, while 13.7% with mucus and 36.71% dysentery.

The prevalence of *E. coli* O157:H7 related diarrhea was significantly associated with rural residence, under-cooked meat consumption, raw vegetable consumption, dysentery, type of diarrhea, number of under-five children contained in the household, educational status of mothers, livestock ownership, and households with a history of diarrhea (Table 3).

Table 3

Multivariate analysis results of associated factors of *E. coli* O157:H7 among under-five children with diarrhea, Eastern Ethiopia, 2018.

Variables	Categories	COR (95%, CI)	AOR (95%, CI)
Childs age	> 18	2.44 (1.37–4.35)	2.16 (0.78–5.97)
	≤ 18	1.00	1.00
Residence	Rural	2.83(1.29–6.21)	3.75(1.26–11.20)***
	Urban	1.00	1.00
Consume under cocked meat	Yes	5.07(2.78–9.25)	3.95(1.23–12.67)***
	No	1.00	1.00
Raw milk consumption	Yes	1.00	1.00
	No	0.39(0.18–0.89)	0.63(0.21–1.86)
Consume Raw Vegetable	Yes	2.99 (1.66–5.36)	3.37(1.32–8.62)**
	No	1.00	1.00
Type of diarrhea	Watery	1.00	1.00
	Bloody	3.94(2.04–7.62)	4.42(1.78–10.94) **
Number of under-five children	1–2 child	1.00	1.00
	3–4 children	4.44(2.41–8.17)	7.16(2.90–17.70)***
Mothers Education level	No formal	1.87 (0.9–4.18)	3.14(1.05–9.42)**
	Formal	1.00	1.00
Animal ownership	Yes	2.70(1.37–5.31)	3.87(1.48–10.12)**
	No	1.00	1.00
Diarrhea in Household	Yes	3.64(2.02–6.57)	4.22(1.84–12.69)***
	No	1.00	1.00
Drinking water-treatment method	Boiling	1.00	1.00
	Aqua tab	1.78 (0.35–9.06)	1.03(0.14–7.16)
	Straining	6.22(1.17–33.2)	6.8 (0.78–17.5)
	Nothing	3.32(0.76–14.4)	3.61(0.64–20.55)
Hand washing ways usually practiced	Water only	1.00	1.00
Notes COR; crude odds ratio, AOR; adjusted odds ratio, *, p-value < 0.05, **, p-value < 0.01, ***, p-value < 0.001			

Variables	Categories	COR (95%, CI)	AOR (95%, CI)
	Ash & water	0.36 (0.17–0.78)	1.32 (0.48–3.63)
	Soap & water	0.26(0.08–0.87)	0.29 (0.07–1.27)
Notes COR; crude odds ratio, AOR; adjusted odds ratio, *; p-value < 0.05, **; p-value < 0.01, ***; p-value < 0.001			

Multivariable analysis showed the odds of *E. coli* O157:H7 related diarrhea among children from rural residence were 3.75 times (AOR:3.75; 95%CI:1.26, 11.20) higher than children from urban residential. The likelihood of *E. coli* O157:H7 related diarrhea is increased by about four-fold (AOR:3.95; 95%CI:1.23, 12.67) in children who consumed under-cooked meat compared to those who did not eat undercooked meat. Children with the history of consuming raw vegetables were 3.37 (AOR:3.37; 95%CI:1.32, 8.62) times more likely to have *E. coli* O157:H7 related diarrhea than those without a history of consuming raw vegetables. Children with dysentery (bloody diarrhea) were 4.42 (AOR: 4.42; 95%CI:1.78, 10.94) times more likely to have *E. coli* O157:H7 related diarrhea than those with watery diarrhea. The likelihood of *E. coli* O157:H7 related diarrhea was seven times (AOR:7.16; 95%CI:2.90, 17.70) higher among children from a household with more than two children compared to household with less than two children. Children from families who owned livestock were 3.87 (AOR: 3.87; 95%CI:1.48, 10.12) times more likely to have *E. coli* O157:H7 related diarrhoea compared to children from families who did not have livestock. The odds of *E. coli* O157:H7 related diarrhoea were fourfold (AOR; 4.22; 95%CI:1.84, 12.69) greater among children from families with a recent history of diarrhoea compared to children from household without a history of diarrhoea.

Discussion

This Hospital-based study provides results on the magnitude of *E. coli* O157:H7 and associated factors in under-five children with diarrhea in Eastern Ethiopia. The magnitude of *E. coli* O157:H7 among the children was 15.3% (95% CI: 11.8–19.4) and associated with being rural residents, under cooked and raw vegetable consumers, history of dysentery type of diarrhea, number of children, mothers uneducated, owned livestock, and history of diarrhea in the household.

The prevalence of *E. coli* infection was in agreement with the prevalence report of study (14%) in Bahir-Dar town, Ethiopia [12] and it is comparable with study (20%) in Benin City, Nigeria [10]. However, it is higher than the report of a study conducted in central Ethiopia in 2017 [18] and southern Ethiopia [19]. It is also much higher than studies conducted in other African countries [20, 21]. The discrepancy might be explained by the difference in sample size, source of study population difference, method of sample collection and specificity of polymerase chain reaction.

Moreover, compared to hospitals based studies of the same age group with diarrhea, the current prevalence was higher than studies conducted in some Asian countries, as the reported prevalence was 1.14% in Iran [22], and 4.61% in India [23, 24]. Compared to the South American study, Argentina, the finding is also higher than the 10.1% report of Rivero *et al.* (2010) [25]. Generally, the relatively high

prevalence of the organism obtained in the current finding might be because of the cosmopolitan and high level of animal-to human interaction in the study subjects than the above-mentioned countries, indicating *E. coli* O157:H7 to be an important diarrhea causing pathogen in the study population.

It was found that children from rural residents were about four times more likely to had *E. coli* O157:H7 compared to urban resident children. The finding is consistent with other studies conducted elsewhere [26, 27]. The positive association in the existence of high-density reservoir animals as a predictor of infection [28, 29] and the higher probability of rural residents to have animal contact there by the predictor of rural residence for childhood diarrhea in this age group [30, 31] might support the finding.

Consuming undercooked meat and raw vegetables is significantly associated with the prevalence of *E. coli* O157:H7. This finding is in line with the report of previous studies [32–34]. This could be evidenced by the high level of animal manure contamination of vegetables through either irrigation by untreated surface water or animal manure usage as a fertilizer [35]. As indicated in this study, animal manure usage as a fertilizer was significantly associated with infection to the organism. The organism was isolated from vegetable samples [36], can survive outside the host reservoirs [37]. There were lettuce associated large outbreaks raised in Sweden in the year 2005 [38]. However, there are some reports, which claim that, eating raw vegetables as a protective factor for the occurrence of *E. coli* O157:H7 infection [39].

The likelihood of *E. coli* O157:H7 was more than four times higher among children with bloody diarrhea relative to those who experience watery diarrhea. Previous study reported was more likely to be isolated from visibly bloody stool than without visible blood and was the pathogen most commonly isolated from visibly bloody stool that yielded a bacterial enteric pathogen [33, 40, 41].

Meanwhile the association of raw milk consumption and *E. coli* O157:H7 was not found statistically significant, although some authors described as consumption of raw milk is a risk factor for the occurrence of *E. coli* O157:H7 infection [42, 43]. However, the interpretation must be cautious because of the limitations posed by the small sample size (12.6%) of the children had consumed raw milk.

Age differences were not found independently associated with occurrence of *E. coli* O157:H7. However, Kargar and Homayoon found that children (< 2) years) of age were at highest risk of infection with *E. coli* O157:H7 [44]. Since the data related to exposure status of the child were not mentioned, it is difficult to justify. The contrary reported as the odds of detection proportion of *E. coli* O157:H7 were high in those children above the mean age in more than 11%. The discrepancy seen with the current finding might be attributed to the older child (1–17 years) age group in the later study, which will influence the social and cultural behavior [45].

Children from households with livestock were four times more likely to contract *E. coli* O157:H7 than households without livestock, which is consistent with the report of study on cattle owners [46] and study conducted in Harare, Zimbabwe [47]. Since ruminants had been identified as the major reservoir of *E. coli* O157:H7, there could be a cross-infection to the children through either by direct live animal contact or with animal manure. A matched case-control targeted particularly in children under three years of age in

Germany also indicates nine times the risk of developing infection in those who touched ruminants including goats and sheep [14]. In the meantime, studies reported that contact with farm animal manure or cattle and living in or visiting a place with farm animals be well-established risk factors for the occurrence of *E. coli* O157:H7 infection [32, 48].

Children whose mothers had no formal education were three times more likely to have *E. coli* O157:H7 than mothers with formal education. It is possible to suggest that mother's education level might have a positive influence in preventive measures of diarrhea such as household hygiene and sanitation. Similarly, poor level of household sanitation had been reported as predictor of *E. coli* O157:H7 infection [39]. Evidences strength of the presence of low awareness on hygiene and sanitation among low-income mothers and this could expose a child to several cross-contamination from one source to the other, indicative of poor personal hygiene [49].

Children from a household member with diarrhea prior to his/her diarrhea manifestation were four times more likely to contract *E. coli* O157:H7. The same finding was obtained by Rivas in their case-control study [39]. In addition, a child who was from a household having three to four under-five children had seven times higher odds of *E. coli* O157:H7 than a household with a single to two children. Household under-five year number is a well-studied determinant factor associated with childhood diarrhea through its effect on overall household sanitation and hygiene level [50–53]. Although the levels of contamination of the water sources are not well identified, drinking water treatment methods were not found associated with the occurrence of the organism. However, other researchers reported drinking contaminated water sources as a predictor for the occurrence of *E. coli* O157:H7 infection [36, 54–57].

However, the result obtained in this research work might be difficult to infer about the general source population because of the design. Besides, as study crosses sectional type, the absence and the existence of the organism may not declare the absence of other enteric pathogens as a cause of diarrhea.

Conclusions

The magnitude of *E. coli* O157:H7 among under-five children is found high. The likelihood of *E. coli* O157:H7 prevalence was increased among rural residents, under cooked and raw vegetable consumers, who had dysentery, type of diarrhea, family with more than two children, uneducated mother, availability of livestock, and presence of family members with a history of diarrhea.

In line with the above conclusion, creating awareness on childcare and household sanitation is essential. Local health authorities should work in collaboration with the local agriculture and livestock development bureau to take preventive hygienic measures associated with animal manure and the role of livestock's and livestock products in the transmission pathway of *E. coli* O157:H7 to humans in all the interfaces in rural residents. Finally, we recommend future research efforts must target at a community level transmission chain of *E. coli* O157:H7 and its health impact on under five years.

List Of Abbreviations

BPW Buffered Peptone Water

CT-SMAC Cefixime Tellurite Supplemented Sorbitol Macconkey Agar

E. coli *Escherichia coli*

EHEC Enterohaemorrhagic *Escherichia coli*

HFSUH Hiwot-Fana Specialized University Hospital

HH Haramaya Hospital

IHRERC Institutional Health Research Ethics Review Committee

IMVIC Indole, Methyl red, Voges-Proskauer and citrate

mTSB+N Modified tryptone soy broth supplemented with Novobiocin

U5 Under five

Declarations

Ethical Consideration

Initially, ethical clearance was obtained from Institutional Health Research Ethics Review Committee (IHRERC) of Haramaya University. Formal letter written to hospitals was obtained in order to get permission from the Harari regional health bureau and the Haramaya district health bureau. An informed, voluntary, written and signed consent was obtained from the head of the respective Hospitals. Further an assent was obtained from parents/guardians of the children after the aim of the study, and nature of the study is fully explained to them. Parents/guardians of the child were also informed of the confidentiality of the information obtained and about their full right to refuse or discontinue participating in the research. There could be some psychological discomfort to the parents/guardians pertaining to stool sample collection and delay in the laboratory analysis report, thus they were convinced by consciously informing on the benefit of this study in this vulnerable age group. Besides, they were explained and assured if in case they are unable to afford the payment associated with confirmed cases after laboratory analysis, as it will be to be covered by the researcher.

Since the result of the laboratory takes about weeks, the address of the participants, mobile phone numbers of the family and in those who did not have a mobile phone their relative address of the family was documented. Until the laboratory report, the physician on duty treated all the children accordingly. Soon the laboratory work ends, for those children positive for presumptive *E. coli* O157:H7 their parents were communicated about their child. To date from the total 56 cases identified, only 34 of them were

reached. Up on phone communication report of their respective family made after a week 9 of them has not been recovered well although the diarrhea was not as previously. However, even if they are well informed to bring the child to Hospital for better advice and management of the case, until this document is submitted only 3 of the cases were returned to the Hospital and got the service through the physician on duty. It will be tried to convince the family who refused to bring their child and in the upcoming days, we keep reaching all the rest family of the child through the health extension workers of their respective Kebeles taking into account the consequence of the case if not managed accordingly.

Consent for publication

Not applicable

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Competing interests

The authors declare that they have no competing interests

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Authors' contributions

DK: conceptualize the study, write the proposal, analyzed, interpreted the patient data and was a major contributor in writing the manuscript. LO, DA and TS: edit and revise the manuscript, and supervise the study, LDR: perform statistical analysis, interprets results and write the manuscript and all authors read and approved the final manuscript.

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Figures

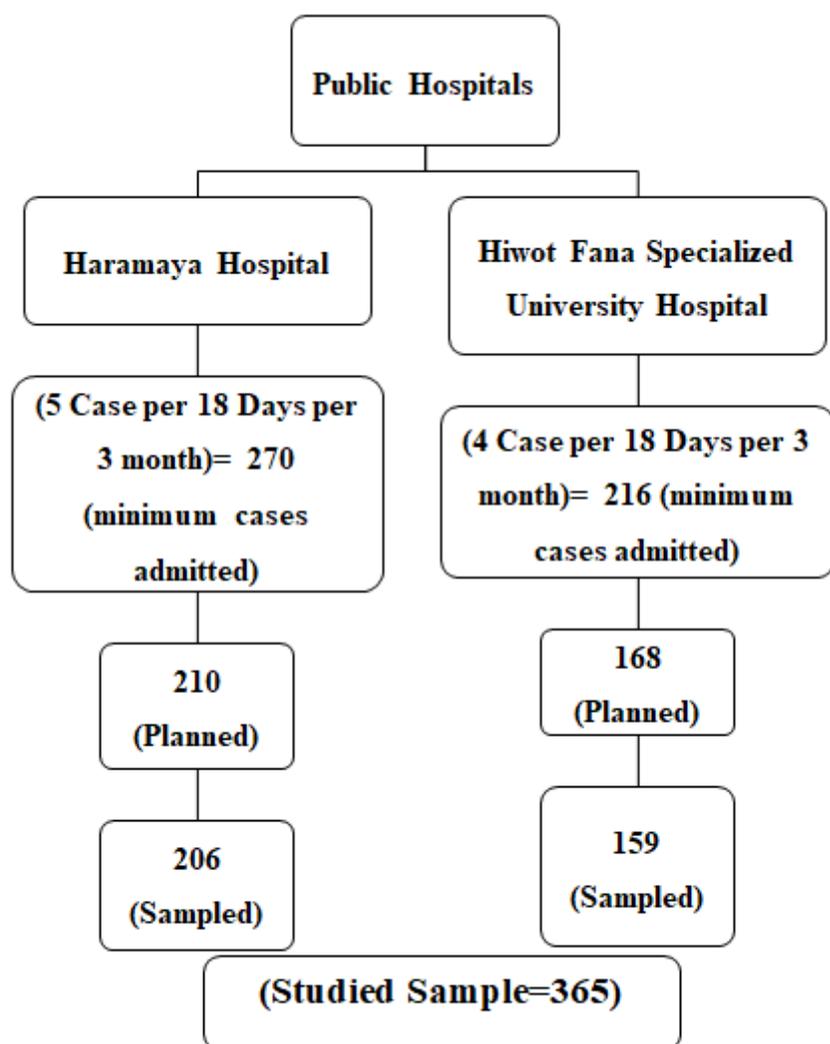


Figure 1

Schematic representation of sampling procedure for selecting study population, Eastern Ethiopia, 2018.

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