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Research article

Isolation, Identification and Antimicrobial Susceptibility Profile of *E.Coli* (O157:H7) From Fish in Lake Hawassa, Southern Ethiopia -

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Abstract

Escherichia coli belongs to the family Enterobacteriaceae, is a facultative anaerobic, non-spore forming, and gram-negative bacilli bacteria. The isolation and identification of *E. coli* O157:H7, and its antimicrobial susceptibility have been done in fish from skin and muscle samples taken from Lake Hawassa in Southern Ethiopia. A total of 343 healthy fish was randomly sampled (212 skin swab and 131 muscle swab) which comprise three species Nile tilapia, African catfish and Barbus (golden) fish. From a total of 343 fish samples, 80 (23.3%) *E. coli* was isolated, while 8 (2.3%) strain O157:H7 was isolated. In case of part of fish where swab samples were taken from a total of 212 skin swabs and 131 muscle swabs 69(32.54%) and 11(8.4%) *E. coli* was isolated, respectively. Similarly, from 212 skin swabs and 131 muscle swabs, 7(3.3%) and 1(0.76%) pathogenic strain (O157:H7) of *E. coli* was isolated, respectively. The occurrence of *E. coli* from fish skin and muscle is statically significant with p-value (0.000). Therefore, *E. coli* was more prevalent in fish skin than muscle. However the occurrence of *E. coli* and its strain O157:H7 in Nile tilapia, African catfish and Barbus fish was not showing a Significance difference, where that p-value is greater than 0.05. *E. coli* O157:H7 isolates were tested with nine available antimicrobial drugs. All eight isolates were 100% sensitive to ciprofloxacin, gentamicin, trimethoprim, and Sulfamethoxazole. Whereas all of the isolates were 100% resistant to some antibiotics like ampicillin and cefoxitin. Isolates for tetracycline were 1(12.5%), 4(50%) and 3(37.5%) were resistant, intermediate and susceptible, respectively. And 3(37.5%) resistant, 1(12.5%) intermediate and 4(50%) isolates were susceptible for streptomycin. For doxycycline 5(62.5%), 2(25%) and 1(12.5%) were susceptible, intermediate and resistant, respectively. *E. coli* and the strain O157:H7 are among the common microbial threats to the fishery practice. Indiscriminate uses of antibiotics to treat bacterial infection promote the development of drug-resistant bacteria. Further study should be conducted in the fish environment for assessment of water quality, isolation and identification of pathogenic microorganism that has great zoonosis importance like *E. coli* O157:H7.

Keywords: *E. coli*; Fish; Lake Hawassa; Muscle; O157:H7; Skin

ABBREVIATIONS

µg: microgram; m: micrometer; CLSI: Clinical and Laboratory Standard Institute; Conc.: Concentration; FAO: Food and Agricultural Organization of the United Nations; Hrs: hours; IFT: Institute of Food Technologists; Km: kilometer; LFDP: Lake Fisheries Development Project Working Paper; NMA: National Metrology Agency; OIE: Office International des Epizooties; SMA: Sorbitol MacConkey Agar; WHO: World Health Organization; X²: Pearson's chi-square value

INTRODUCTION

Aquaculture and fisheries are important sources of food and income, and sustaining the livelihood for many people across the world [1,48]. Fish and sea-food constitute an important and cheap food source of protein for a many group of the world population [2,49]. Fish contains good quality protein and other necessary nutrients that make it a valuable food. Fish consumption become a popular part of food in many areas of the world, while in some Asian countries fishery supplied the major source of animal protein [3,50].

Ethiopia is a country without sea outlet, where fish supply is entirely depending on inland lakes and rivers, but fishery is still a cheap source of protein. The water bodies of Ethiopia are assumed to accommodate about 7,400 km². There are also rivers with travel a lot of kilometers to the different directions of the country. The country's total annual fish producing potential was estimated to be 51,481 metric tons per year on a maximum sustainable yield basis [4]. Like other countries, in Ethiopia different threats such as urbanization and population growth, expansions of, industrialization, and other water related development activities have resulted in an impact in the fish production [5].

Fish is an important source of better quality protein consumed by human. However, fish are susceptible to a many varieties of disease-causing organisms like bacterial pathogens, most of which are zoonotic, causing disease in human [6]. The occurrence of diseases in fish is the result of the association between pathogen, fish, and water bodies. To study pathogenic microorganisms on fish is by understanding the surrounding factors influencing them; it allows the development of advanced and adequate measures for the prevention and controls of many diseases that limit influence production [7].

The intensification of production and increase in stocking density has made fish farming more vulnerable to disease [8].

Human diseases caused by pathogens transmitted from fish and the aquatic environment is quite common depending on the degree of contact with fish, related environment, dietary habits and the immune status of the exposed individual. Bacterial species are often facultative pathogenic for fish and maybe isolated from fish without apparent symptoms of the disease on it. Human infection caused by different pathogens from fish is either from food or direct contact during hobby [9].

Escherichia coli are a rod-shaped bacillus having about 2µm long and 0.5µm in diameter with a cell volume of 0.6 to 0.7µm³ [10]. It is a normal inhabitant in the intestines of animals and humans. Its recovery from food has public health concern due to the possible presence of pathogenic strains which lead to severe gastrointestinal disturbance [11]. It is widely used as an indicator of the bacteriological contamination of food and environments most commonly from the fecal origin [12].

E. coli is one of the common indicators of hazardous conditions during collection and processing of fish and its products [13]. Most strains of *E. coli* are normally present in intestinal tract of both humans and animals as commensals, while there is some pathogenic strain that causes strong diarrhea to animals and humans. Pathogenic *E. coli* distinguished from normal flora by their possession of virulence factors. The specific virulence factors can be used, to separate these organisms into path types [14].

E. coli O157:H7 is one of the most known strains to contain path types that can cause foodborne infection in humans and animals. *E. coli* O157:H7 path types have been found in the intestines of healthy animals [9,15]. It produces a toxin that causes disease in animals as well as humans. *E. coli* that causes diarrheal disease may belong to many serotypes, but most severe in human and animal infections are caused by *E. coli* O157:H7 serotypes [16]. Outbreak of diseases like diarrhea due to consumption of fish contaminated with enterotoxigenic *E. coli* O157:H7 has been reported in Japan [17].

Drugs particularly antibiotics possess an ultimate importance in treatment, control and prevention of illness and death caused by infectious diseases in both animals and humans [18]. As a result of

antibiotics that are released into water bodies via the excretion of humans and animals different metabolites of parent compounds present in the aquatic environment [19]. In another case the poor hygienic and stressful conditions present in a water environment, the potential risk of aquaculture fish for bacterial infections is high. Due to this, large amounts of different grades of antimicrobials are used in fish feed for control and treatment purposes in aquaculture facilities worldwide [20]. These huge uses of antibiotics in fish have resulted in increased strains resistant to these drugs. Resistant strains therefore, put a potential impact on therapy of fish diseases, human diseases and as well as an animal disease [21].

Indiscriminate uses of antibiotics to treat many bacterial infections in both human and animal promote the development of drug-resistant bacteria. It is difficult to treat drug-resistant bacteria with common antibiotics. In case of fish different bacteria like *E.coli* and antimicrobials are released into water bodies via the excretion of human and animals. It causes infection of aquatic animals with different bacteria and the development of drug-resistant bacteria.

Here are the objectives of this study

- ❖ To isolate *E.coli* from fresh fish obtained from Lake Hawassa and assessing how water contamination impacts on fish quality
- ❖ To identify *E.coli* O157:H7 from fresh fish muscle and skin
- ❖ To assess the susceptibility of *E.coli* O157:H7 against selected antibiotic that usually used to treat the bacterial infection

METHODS AND MATERIALS

Study area

The study was conducted from November 2017 to June 2018 in fresh healthy fish found in Lake Hawassa. Lake Hawassa is one of the most beautiful natural gifts of Ethiopia. Hawassa city is located at 275 km south of Addis Ababa, and is the capital city of the Southern Nations, Nationalities and Peoples' Regional State. Hawassa is settled at an elevation of 1700 meter above sea level, and a latitude of 7° 04'N and a longitude 38° 31'E on the cliff of the Great Rift Valley (Figure1). The mean annual range of precipitation and temperature are 950-1200 mm and 27°C, respectively [22]. The Lake stretches 16 km from the north to south direction and extends 8 km from west to east direction having an approximate water volume 1.3 billion meter cube. The maximum depth of the Lake is 21.6 m with mean depth 11 m [23]. It has an area of 97 km² [24]. The common landing site and fish market of Lake Hawassa fishery are Amorage del but illegal fishermen also land their catches at other shores of the lake.

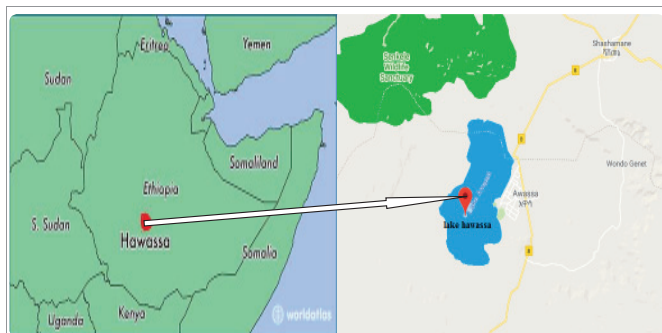


Figure 1: Map of Hawassa city (study area) and the location of Lake Hawassa in Southern Ethiopia.

Study animals

The study was conducted on fishes from Lake Hawassa. In Lake Hawassa Commercially important species are Nile tilapia (*Oreochromis niloticus*), African catfish (*Clarias gariepinus*) and Barbus (golden) fish (*Lepidothamnus intermedius*) [24]. Nile tilapia constitutes about 90% of the total production, while African catfish and Barbus (golden) fish contribute only about 7-8% and 2-3%, respectively. However, the contribution of catfish rises to 20% of the total landing during the fasting periods of the Ethiopia Orthodox Tewahido Church followers [23].

Sample size determination

The sample size required for the study was determined using an expected prevalence of 50% as there is no previous research work on isolation of *E.coli* from fish in Lake Hawassa. The required sample number was obtained using the field formula [25] with a 95% confidence interval and 5% absolute precision.

$$n = \frac{1.96^2 \cdot P_{exp} (1 - P_{exp})}{d^2}$$

Where, P_{exp} = expected prevalence

d= absolute precision;

n = sample size.

Based on this, the required sample size was 384 animals, but due to a financial problem related to the cost of fish, laboratory reagents 343 fish were sampled and considered for the study.

Study design, sample collection

A cross-sectional investigation was conducted for isolation and antimicrobial susceptibility profile of *E.coli* with main focus on strain O157:H7 from 343 randomly selected fresh fish sample in Lake Hawassa particularly at Amorage del. In Lake Hawassa most species of fish in the market was Nile tilapia. The percentage of African catfish and Barbus (golden) fish was much lower than that of Nile tilapia. Due to this reason sampling was 146 samples Nile tilapia, 121 samples African catfish and 76 samples Barbus (golden) fish.

The fish was transported with sterile plastic and icebox to Hawassa university microbiology laboratory for sampling. The sterilize cotton swab was wiped randomly against from muscle and skin of fish. A swab stick of sterile cotton swabs was immersed into buffered peptone water; the shaft of the stick was pressed against the tube wall to break and through it. The remaining part of the stick swab was sealed in the tube. The swab was marked, numbered and incubated at 37°C for one day.

Isolation and identification of *E.coli* and *E.coli* O157:H7

The swab was incubated firstly on buffered peptone water enriched media at 37°C for 24 hrs. And then the sample incubated in buffered peptone water cultured on MacConkey agar at 37°C for 24 hrs. *E.coli* isolates which revealed characteristics colony morphology such as smooth, circular, pink colony in MacConkey agar sub-cultured into Eosin methylene blue agar for 24 hours at 37°C. Bacterial colonies which show the typical characteristic and color of *E.coli* were taken and sub-cultured on Eosin Methylene Blue agar. The bacterial colony appeared green metallic sheen was regarded as *E.coli*.

The positive result from the above media was inoculated into nutrient agar and tested by different biochemical test:

- Catalase test: to observe slightly bubble production



- Simon citrate test: no color change from green to blue
- Methyl red test: red color indicative of acid production
- Triple sugar iron agar slant culture by stab method and yellow slant yellow butt presence of gas bubbles and absence of black precipitate in the indicative of *E. coli*
- **Indole test:** red ring at the top of culture and Vogues Proskauer tests no color change was observed.

Isolates that were identified to be *E. coli* were sub-cultured onto Sorbitol MacConkey medium from nutrient agar medium. Sorbitol MacConkey agar and plates were just incubated at 37°C for 20-24 hrs. The interesting situation here is that *E. coli* O157:H7 cannot undergo fermentation on sorbitol medium, and therefore, it results in colorless colonies. However, other strains of *E. coli* can ferment this medium and form pink/red colonies [26]. Then after non fermenting colonies from the SMA were transferred to agglutination assessment with the *E. coli* O157:H7 test kit. *E. coli* O157 or H7 antigens on the test organisms coated with antibodies on latex blobs were forming a visible antigen-antibody precipitate [27]. Bacterial colonies showing an agglutination reaction were considered as *E. coli* O157:H7 positive.

Antimicrobial susceptibility

Antimicrobial sensitivity was done with the disk diffusion method using Mueller-Hinton agar. First of all, a suspension of a sample in saline solution was prepared by adjusting with the 0.5 McFarland turbidity standards. The susceptibility of the *Escherichia coli* strains has been checked in relation to nine groups of antimicrobials, including streptomycin (10 µg), ampicillin (25 µg), gentamicin (10 µg), Sulfamethoxazole (100 µg), cefoxitin (30 µg), tetracycline (30 µg), ciprofloxacin (5 µg), trimethoprim (5 µg) and doxycycline (30 µg) was added into Mueller-Hinton agar plate using sterile forceps. The plate was incubated for 24 hrs at 37°C under aerobic condition. Then zone of Inhibition was measured with calipers in millimeter and interpreted as susceptible, intermediate and resistant as expressed in table below (Table 1), based on Clinical Laboratory Standards (CLSI). The antimicrobial susceptibility has been done eight times for each antibiotic drug.

Table 1: Zone diameter interpretive standards chart for the determination of antibiotic sensitivity and resistance status by the Disk Diffusion method for *Enterobacteriaceae* [51].

Name of antibiotics	Dose/disk	Inhibitory zone diameter to nearest millimeter (mm)		
		Sensitive (S)	Moderately sensitive (MS)	Resistant (R)
Streptomycin	10 µg	≥ 15	12 - 14	≤ 11
Sulfamethoxazole	100 µg	≥ 16	11 - 15	≤ 10
Cefoxitin	30 µg	≥ 18	15 - 17	≤ 14
Tetracycline	30 µg	≥ 15	12 - 14	≤ 11
Doxycycline	30 µg	≥ 14	11 - 13	≤ 10
Ampicillin	25 µg	≥ 17	14 - 16	≤ 13
Ciprofloxacin	5 µg	≥ 21	16 - 20	≤ 15
Trimethoprim	5 µg	≥ 16	11 - 15	≤ 10
Gentamicine	10µg	≥ 15	13 - 14	≤ 12

Data analysis

The generated data was entered into Microsoft Excel spreadsheet. The total isolated *E. coli* and specifically *E. coli* O157:H7 from fish muscle and skin was determined using standard formula. Stata 13 statistical software, Pearson's chi-square, and Fisher's exact test were used to associate fish species and site of swab sampling from the fish organ as a risk factor with the prevalence of *E. coli* and specifically pathogenic strain *E. coli* O157:H7. The difference was considered to be significant when *p*-value less than 0.05, while insignificant if greater than/equals to 0.05.

RESULT

A total of 343 healthy fish were sampled (212 skin swab and 131 muscle swab) from a total of 146, 121 and 76 Nile tilapia, African catfish and Barbus (golden) fish, respectively. All swab specimens have been tested for the isolation of *E. coli* as well as *E. coli* O157:H7. From a total of 343 samples, 80(23.32%) *E. coli* were isolated, and 8(2.35%) *E. coli* strain O157:H7 were identified. In part of fish where swab samples were taken 212 skin swabs and 131 muscle swabs, 69(32.54%) and 11(8.4%) total *E. coli* were isolated, respectively. Similarly, from 212 skin swabs and 131 muscle swabs 7(3.3%) and 1(0.76%), pathogenic strain *E. coli* O157:H7 were isolated, respectively.

During sampling 146 Nile tilapia (83 skin and 63 muscle swabs), 121 African catfish (73 skin and 48 muscle swabs) and in Barbus (56 skin and 20 muscle swabs) species of fish were sampled. Nile tilapia 30(36.14%) skin swab and 5(7.93%) from muscle swab *E. coli* were isolated whereas 4(4.8%) from skin and 0(0%) from muscle swab *E. coli* O157:H7 was isolated. African catfish 23(31.5%) from the skin and 4(8.33%) *E. coli* was isolated and from these 2(2.74%) from the skin and 1(2.08%) from muscle swabs *E. coli* O157:H7 strain was isolated. Barbus (golden fish) 16(28.57%) from the skin and 2(10%) from muscle swab *E. coli* was isolated from these 1(1.78%) from skin swab and 0(0%) from muscle swabs *E. coli* O157:H7 was isolated. The study did not examine for variations with maturity and size of fish.

The occurrence of *E. coli* from muscle and skin has a significant association, where *p*-value less than 0.05, that means *E. coli* is present more in fish skin than fish muscle. The other species of fish with *E. coli* prevalence and *E. coli* O157:H7 with a site of sampling have no significant difference that *p*-value greater than 0.05.

The table below (Table 2) showed that the occurrence of *E. coli* has no significant difference between Nile Tilapia, Catfish and Barbus species of the fish *p*-value (0.947). Whereas *E. coli* isolates in skin and muscle have a significant difference with *p*-value 0.000.

Table 2: Total occurrence of *E. coli* in three species (Nile tilapia, African catfish and Barbus (golden) fish) and site of swab with its statistical analysis.

Species of fish	The total sample is taken	Total <i>E. coli</i> isolates	X ²	<i>p</i> - value
Nile tilapia	146	35(24%)	0.1089	0.947
African catfish	121	27(22.31%)		
Barbus (golden fish)	76	18(23.68%)		
Total	343	80(23.32%)		
Site swab sample			26.4057	0.000
Muscle	131	11(8.4%)		
skin	212	69(32.55%)		
total	343	80(23.32%)	26.4057	0.000



The isolation of *E.coli* O157:H7 in three species of fish and site of swab sample taken has no significant difference with *p*-value 0.905 and 0.161, respectively (Table 3). Statistical analysis was calculated with Stata 13 statistical software, Fisher's exact test.

Table 3: Percentage of *E.coli* O157: H7 in 3 different species and site of swab sample with statistical analysis.

Species of fish	The total sample is taken	<i>E.coli</i> O157: H7 isolates	<i>p</i> - value
Nile tilapia	146	4(2.74%)	0.905
African catfish	121	3(2.48%)	
Barbus (golden fish)	76	1(1.31%)	
Total	343	8(2.35%)	
Site swab sample			
Muscle	131	1(0.76%)	0.161
skin	212	7(3.3%)	
total	343	8(2.33%)	

Anti-microbial susceptibility profile of *E.coli* O157:H7

Isolated *E.coli* O157:H7 have been tested with nine available antibiotics with disc diffusion method (Table 4). From this isolate, all were susceptible to drug ciprofloxacin, trimethoprim, gentamicin, and sulfamethoxazole and resistant for ampicillin and cefoxitin. Four isolates were susceptible, three isolates were resistant and one isolate was intermediate for streptomycin. For tetracycline one isolates were resistant, three isolates were susceptible and four isolates were intermediate. In the case of doxycycline, five isolates were susceptible and two isolates were intermediate and one isolate resistant. *E.coli* O157:H7 isolated from fish muscle was resistant for three drugs (streptomycin, ampicillin, and ciprofloxacin) and intermediate for doxycycline and tetracycline whereas susceptible for streptomycin, ciprofloxacin, trimethoprim, and gentamicin.

Table 4: *E.coli* O157: H7 isolates and their susceptibility to antimicrobial drugs.

Drugs	Conc.	Frequency of <i>E.coli</i> isolate susceptibility per drugs		
		resistant	Intermediate	susceptible
Streptomycin	10 µg	3(37.5%)	1(12.5%)	4(50%)
Sulfamethoxazole	100 µg	0(0%)	0(0%)	8(100%)
Cefoxitin	30 µg	8(100%)	0(0%)	0(0%)
Tetracycline	30 µg	1(12.5%)	4(50%)	3(37.5%)
Doxycycline	30 µg	1(12.5%)	2(25%)	5(62.5%)
Ampicillin	25 µg	8(100%)	0(0%)	0(0%)
Ciprofloxacin	5 µg	0(0%)	0(0%)	8(100%)
Trimethoprim	5 µg	0(0%)	0(0%)	8(100%)
Gentamicine	10µg	0(0%)	0(0%)	8(100%)

DISCUSSION

E.coli presence in aquaculture accredited to animal waste pollution of the water bodies [28]. Its isolation from fish samples results from animal dung contamination of the water. The contamination of food and environment with a bacteriological condition like *E.coli* is almost originated from human and animal feces [12].

Isolation of *E.coli* was done by taking swab samples from the skin and muscle of fresh fish. Isolation of *E.coli* from fish muscle and skin has a statically significant difference with *p*-value 0.000 or 69(32.54%)

and 11(8.4%) from skin and muscle, respectively. *E.coli* is more found in fish skin than muscle. However, the overall isolation percentage of *E.coli* and *E.coli* O157:H7 in fish might not be representative of the *E.coli* and *E.coli* O157:H7 distribution in fish in the study area. Because this study used the sample size which is below the actual sample requirement for the study due to financial related problems.

Skin has a chance to contact with water and other environment contaminated with feces that have many pathogenic bacteria [29]. This study is similar to Rafael [30] who isolated more *E.coli* from fish skin than muscle and they conclude that fish muscle can contaminate with different bacteria during harvesting but the skin has direct contact to the environment it can easily contact with many pathogenic or non-pathogenic *E.coli* [30].

The same to [30] and a pun *et al.* (1991) in this study skin is a more vulnerable organ of fish for *E.coli* than muscle. *E.coli* occurrence from muscle was much lower than skin [30,31]. Pao and his collaborators described that the detection of *E.coli* on fish muscle aggravated by intestinal waste contamination at the time of harvesting. The usual occurrence of *E.coli* in fish muscle has a major relation with pollution of water with manure and other wastes that contaminate fish when at harvesting or processing [32].

The samples taken from different fish species namely Nile tilapia, African catfish and Barbus (golden fish) for identification of the most critical species vulnerable to infection with *E.coli* O157:H7 is not statically significant *p*-value (0.905). The result of current study indicates that there was no a significant association among the fish species considered as to the occurrence of *E.coli* infection. This is not in agreement with previous reports higher infection in Plankton feeders, Nile tilapia species for *E.coli* than Catfish [33]. This potential disagreement may arise from the difference in sample size used, ecosystem of study area, or sampling methods.

Fish will infected with *E.coli* O157:H7 while at harvesting or during further production stages. During the current study pathogenic *E.coli* O157:H7 have been dealt from fish muscle and skin 1(0.76%) and 7(3.32%) respectively. Romero Ayulo *et al.* (1994) isolate pathogenic *E.coli* O157:H7 from fish muscle the result is almost similar to this study. They isolate one *E.coli* O157:H7 from 30 fish meat samples 1(3.33%) [34]. As Wang and Doyle (1998) report that *E.coli* O157:H7 can survive several days in water. It can contaminate skin due to direct contact with water and muscle due to stress during harvesting [35]. Water environment pollution with wastes may be the main source of sources of *E.coli* O157:H7.

Pathogenic strain of *E.coli* has been identified in water bodies in Brazil [36], while in Scotland the pollution of aquatic environments by *E.coli* O157:H7 was detected [37]. Many evidences support the contamination of fish may arise from animal and human wastes, probably by flood from the environment in the water bodies. In contrast to current study result, *E.coli* O157:H7 patho type has been isolated from the gastrointestinal tract but not detected from fish muscle [38].

Current study revived that all eight *E.coli* O157:H7 isolates were sensitive to ciprofloxacin, gentamicin, trimethoprim, and sulfamethoxazole. In another case, they show tolerance to ampicillin and cefoxitin at a resistance level. Likewise, Bekele [39], while this result was opposite to Magwira *et al.* (2005) from Botswana showed that *E.coli* O157:H7 tolerance to ampicillin has not been detected [40]. For tetracycline 12.5%, 50% and 37.5% were resistant,



intermediate and susceptible respectively. Many researches indicated *E.coli* O157:H7 is not responding to tetracycline treatment [41,39,42], which fits results of current study. However, Mohammed [43] showed that *E.coli* O157:H7 was susceptible to tetracycline. The isolates were 37.5% resistant 12.5%, intermediate and 50% susceptible for streptomycin this result almost similar to Beyi [44]. For doxycycline 62.5%, 25% and 12.5% were susceptible, intermediate and resistant respectively.

Development of drug resistance/tolerance by *E.coli* can be achieved via mutation. For example, adaptation to fluoroquinolone has often been due to acquisition of mobile genetic elements, and has also been the significant factor for broad-spectrum penicillin's (e.g. ampicillin or amoxicillin) resistance. Bacterial tolerance to third-generation cephalosporin is developed through the modification of the enzymes called extended-spectrum beta-lactamase; which alters the activity of beta-lactam antibacterial drugs. Extended-spectrum beta-lactamase is shared among bacteria and /or bacterial species. As *E.coli* strains with this enzyme are adapted to several antibacterial agents, Carbapenems usually act as the major treatment option for severe infections [45].

There is evidence supporting the sharing of resistant bacteria between among livestock, aquatics animals and human via food production, and pose critical effect to threat public health [46]. World organization for animal health (OIA) suggested that aquatic animal health should rely on constant monitoring and diseases surveillance of anti-microbial resistant microbes those have interaction with aquatic animal [47]. Generally, fish infection with strain *E.coli* O157:H7 probably results from the environment at harvesting and production process of fish. Hygienic fishery, handling and fish processing techniques are helpful in reducing indiscriminate use of antibiotics for controlling antimicrobial transmissions.

CONCLUSION AND RECOMMENDATION

Among the critically hazardous foodborne diseases increased attention with public health is *E.coli* and its pathogenic strain O157:H7. *E.coli* O157:H7 in three species of fish namely Nile tilapia, Catfish, and Golden fish from muscle and skin were isolated. Eight *E.coli* O157:H7 were identified from the skin and muscle of three species of fish. The presence of *E.coli* O157:H7 in fish is an indication of the contamination of water bodies with animal and human feces. Fish muscle infection with potentially hazardous *E.coli* O157:H7 probably at harvesting and production process of fish. Without selection uses of antibiotics to treat bacterial infection in both human and animal promote the development of drug-resistant bacteria. It is difficult to treat drug-resistant bacteria with common antibiotics. There is a research gap in isolation and identification of *E.coli* O157:H7 from fish both in Ethiopia and in the world.

Therefore, based on aforementioned conclusive remarks, the following recommendations are forwarded

- ❖ The fisherman around Lake Hawassa needs to be educated about the risk of food born disease sanitary and hygienic method of fish handling.
- ❖ The hygienic condition of the lake should be improved by controlling the flood that entre into the lake during a rainy time and controlling the waste disposal of restaurant and hotels around the lake.
- ❖ There should be regular antibiotics sensitivity testing to

E.coli O157:H7 to select effective antibiotics and also help to reduce the problem of drug resistance development towards commonly used antibiotics.

- ❖ Further study should be conducted to establish the stage at which contamination occurs so that corrective measure can be measure.

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