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Assessment of Some Bacteriological Quality of Streams of Upper Awash River, Central Ethiopia

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Abstract
Supply of safe and adequate drinking water is very essential for the health of human beings. But water safety and sanitations in the developing countries has been a serious problem threatening the community of these nations. The major objective of this article is to communicate the microbial quality of water of streams of upper Awash River. The indicator microbial quality of the water samples from streams of upper Awash River was analyzed using membrane filtration techniques, while heterotrophic bacteria, Bacillus spp and Staphylococcus aureus count were done by using specific isolation techniques for the particular organisms. From the five different sampling sites site 1 and 4 showed highest total coliform count ranging from 17300 to 22000 cfu/100ml. Escherichia coli count ranged from 200to 11300 cfu/100ml; Entrococcus count ranged from 178to 2300 cfu/100ml; Clostridium perfringes ranged from 66- to 201 cfu/100ml. Hetrotrophic plate count ranged from 350-2380 cfu/100ml. Bacillus spp count ranged from320-990 cfu/100ml. Staphylococcus aureus count ranged 300-1950 cfu/100ml.The result of this investigation indicated that all the indicator microorganisms and other bacterial species were found to be above the minimum number of the specific group of the organisms showing that the water of streams of upper Awash river is highly exposed to certain pollution and it need some means of purification before it used for the intended purposes.
Keywords/ phrases: bacteriological; indicator microorganisms, Streams; water quality
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Introduction

Next to oxygen water is the most important substances for human existence (White,1969). It is an essential resource which sustains agriculture, allow aquatic life, supports industry, produce hydroelectric power, permit aquatic transport, ensures personal hygiene, maintain clean environment, and it used for sport as well as for recreations (Ministry of Water Resources, 1997, Falkenmark, 1982). In addition, water is very essential for washing, cooking, bathing, food processing, brewing, beverage bott lingetc. (Oesterholt *et al*., 2007). This therefore necessitates a constant supply of potable water to all human communities but most of the area slacks good improved water supply especially in developing countries . However, there is a lot of time,effort, and money that is required or invested so as to find a suitable source of water supply (Oester holt *et al*., 2007).Unfortunately, such water sources are scarce and even when they are available; they are seldom safe for consumption. There is therefore a great need for water to be treated effectively in order to make it potable and safe for humans. Water quality can have a major impact on both individuals and communities health. According to the World Health Organization, about 1.4 million children die from diarrhea due unsafe drinking water (WHO; 2011).Therefore, for good human health, it is important to ensure that drinking water is well treated before consumption (Massoud *et al*., 2010).

Consumption of contaminated drinking water was associated with 80 percent of disease and one third of death in developing countries (Mellor *et al*., 2013;Echoru*et al*., 2015). Therefore, an essential basic requirement for health protection is to provide the public with safe and adequate drinking water (WHO 2011).

In all urban areas in the developed countries, supply of adequately treated water is done by municipal authorities (Howard and Bartram 2005). For the case of developing countries, there is little or no access to such treated water and therefore it is often very difficult or even impossible to get portable water. This is because of inadequate funds available to the municipal for appropriate treatment and supply of water and this being so, a large percentage of people in developing countries have to rely on their own personal efforts in order to get safe drinking water (Howard and Bartram 2005).

Water for consumption is supposed to be free from microbial contamination but it's not obvious that all microorganisms present in water are harmful. The most dangerous microorganisms are the coliforms which are part of the normal flora of the gut of warm blooded animals. The presence of contamination with any of these organisms and others can be tested using various indicators. However the development of such indicators has never been such an easy job and up to date, there still remains considerable arguments about the best indicators for testing microbial contamination (Jeong*et al*., 2011).

Besides, sustainable social and economic development is largely dependent on water resources. However, securing water (quality and quantity) to satisfy the needs of the humans and ecosystems is one of the primary issues challenging the 21stcentury(Amangabara and Ejenma, 2012). Compounding the problem is the fact that water quality is one of the most sensitive issues worldwide, potentially influenced by many natural and

anthropogenic factors. Theses includes: sources of water, the degree of its evaporation , types of rocks and minerals it has encounter (i.e. geology and mineralogy of the water shade) , geological processes within the aquifer, velocity and direction of water movement, and the time it has been in contact with reactive minerals (Freeze and Cherry, 1979). It is also affected by external pollution agencies such as effluents from agricultural return flows, industrial and domestic activities (Srinvasa moothy *et al*., 2012).

It is well known that the quality and safety of the drinking water continues to be an important publichealth issue, because its contamination has been frequently described as responsible for the transmission of infectious diseases that have caused serious illness and associated mortality worldwide (Hrudey and Hrudey 2007, Marshall, *et al*. 2006, Jones *etal.,* 2007, Peace and Mazunder, 2006).

The World Health Organization estimated that up to 80% of all sicknesses and diseasesof world is caused by inadequate sanitation, polluted water or unavailability of water. InEthiopia over 60% of the communicable diseases are due to poor environmental health conditions arising from unsafe and inadequate water supply and poor hygienic and sanitation practices (WHO, 2004). Several studies have confirmed that water related diseases not only remain a leading cause of morbidity and mortality worldwide but that the spectrum of diseases is expanding and the incidence of many water-related microbial disease is increasing (WHO, 2003). Diarrhea remains a major killer in children and it is estimated that 80% of all illness in developing countries is related to water and sanitation; and that 15% of all child deaths under the age of 5 years in developing countries results from diarrheal diseases (WHO, 2003).

In rural areas and villages of Ethiopia, water for human consumption, drinking, washing (bathing,laundry), for preparation of food etc, is obtained from rivers, streams, shallow wells, spring lakes, ponds, and rainfall. Unless water is made safe or treated for human consumption, it may behazardous to health and transmit diseases. The main contaminants of these water sources arehuman excreta, animal waste and effluent because of open field defecation practices. The aim of this article is to communicate the microbial quality of water of upper Awash river, Central Ethiopia

Materials and methods

Description of Study area

This investigation was carried out on streams of upper Awash River in Chilimo forest around Ginchi town which is located in West Shoa Zone, Oromia Regional State. Ginchi is located seventy five kilometers west of Addis Ababa on the Addis Ababa-Nekamte road. The Woreda has a total area of 109,729 ha with altitudinal range from 2000-3200 meter above sea level. Based on the (CSA,2007) population census; the total population of Dandi District is estimated at 256,896. Its favorable climatic condition for both crops and livestock production has been attributed for more population in the Woreda. This Woreda is well known for its rich water resources among which Awash River basin and Lake Dandi are the most important natural resources in the distinct. Lake Dandi is one of the highland lakes found in the district, and has a high tourist potential. The Awash River basin is well known and it covers a catchment area of 110,000 km2 and serves as home for 10.5 million inhabitants. The river originates from a high plateau near Ginchi town, and flows along the rift valley into the Afar triangle and ends in Saline Lake Abbe.

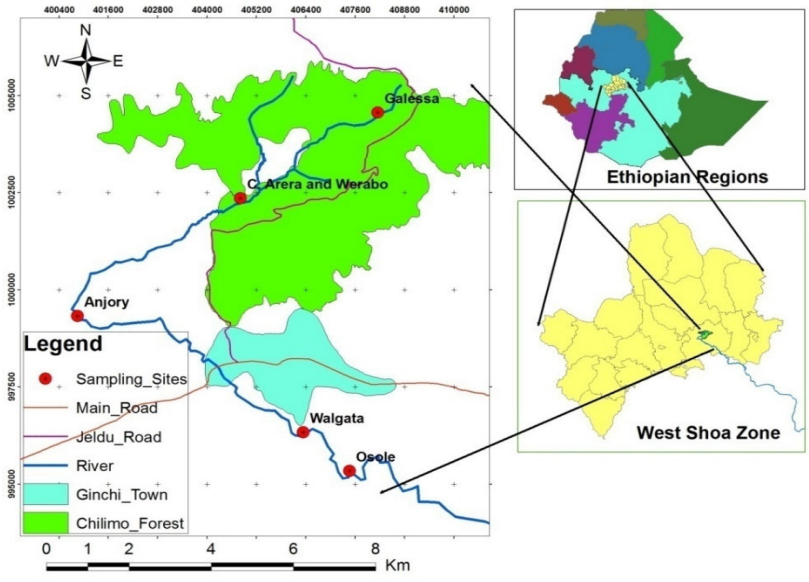

Figure1: Site map of the study area

Sampling sites and sample collection

Representative sampling sites were selected by purposive sampling method and each sample was taken directly from the streams of upper Awash River in two weeks once, and total of six times from the month of December 2017to March 2018.

A preliminary survey was conducted in the November 2017 together with some general information about the topography, physical characteristics of the area and human activities along the course of streams of upper Awash River. Sampling sites were selected based on their exposure to organic wastes and various human activities that pollute the water.The topographic features and habitat structure were also taken in to consideration while selecting the sampling sites. Accordingly, five sampling sites were selected along the longitudinal zonation of the upper Awash River and designated as; sampling site one at the middle of chilimo forest called:Galesa site (Aw1), the second site is Arera (Aw2), which is two Km far from site Aw1 in the forest,Anjori site (Aw3), after it leaves the chilimo forest 11km far from Aw2,Wamurasako site or welgeta (Aw4),which is 10km far from Aw3 and the fifth site is Osole site (Aw5)which is two km far from Aw4.

Water sample collection

The water samples from the study area were collected inthe brown bottle which is 500ml capacity. The bottles were pre sterilized by autoclaving. Double composite samples were collected from each study site. The collected water samples were immediately put in the ice box in order to avoid the loss of the microorganisms though injury and transported to Biology Laboratory department of Biology College Natural and Computational sciences, Ambo University and processed with 8 hrs for the indicator microbial load.

Water samples processing and incubation

The water samples brought to the laboratory were filtered through filter membrane. sample was filtered using 0.45 mm pore size, 47 mm diameter filter membrane as described by NOM-127-SSA1-1994and APHA, Whatman cellulose nitrate membrane filters (Sartorius, Vienna, Austria) and were put in triplicate in the selective isolation and enumeration purposes as described below.

Total coliform and E.coli

Escherichia coli detection with chromocult coliform agar (www.himedialabs.com).Recently, application of defined substrate medium technology with particular selective growth conditions and the simultaneous detection of ß-D-galactosidase and ß-D-glucuronidase activity have become widespread tools for the detection of *E. coli* in water and wastewater (Brenner, 1993, 1996; clark.et *al*, 1991; rice et al, 1990; Venkateswaran.*et al*, 1996).For simultaneous enumeration of *Total coliform and E. coli*, filters were placed onto CCA plates and incubated at 37°C for 24hrs,the*total coliforms* and *E. Coli* isolates on CCA were enumerated. Blue, blue black or violet colonies on plates were classified as *E.coli*, red colonies on plate were *total coliform* (Sartorius, Vienna, Austria).

Enterococcus

M-enterococcus agar base was used for *enterococcus* (www.himedialabs.com).mediawas amended to select against possible background bacteria by the addition of cefsulodin (5 mg/liter; Sigma, Vienna, Austria), while those for *enterococcus* were placed on m-enterococcus agar base plates, and incubatedat 37°C for 48 hrs.Red colonies on M-enterococcus agar base were classified as *enterococcus* (Sartorius, Vienna, Austria).

Clostridium perfringes

Fluorocult-tryptose sulfite cycloserine (F-TSC)(Merck, Darmstadt, Germany) was used to isolate *Clostridium* perfringes (www.himedialabs.com) supplement (0.4 g/liter; Merck)was added on the media to against possible background bacteria, To select for spores of CP, samples were first preheated at 75°C for 15 min in a water bath before filtration. After filtration, the membrane filters were placed on F-TSC agar plates, put in an anaerobic jar containing Anaerocult Anaerobic system (Merck), and incubated at 44°C for24 h in a dry incubator), while black colonies growing on F-TSC plateswere *Clostridium perfringes* (Sartorius, Vienna, Austria).

Heterotrophic plate Count

One ml of water sample was transferred aseptically to 9ml of distilled sterilized water and homogenized by Vortex machine (Biocote) and a serial dilutions of (10^{-1}, 10^{-2}, 10^{-3} ,$10^{-4,}$ 10^{-5}) was made by taking 1 ml from homogenized sample and adding to sterile test tube containing 9ml sterile distilled water and mixed properly by vortex machine. 0.1 ml of each dilution was spread plated on a Plate Count Agar (PCA) medium and incubated at 28°Cfor 48 hours. All colonies that are 0.5 mm or larger in diameter were counted using digital colony counter. The number (N) of CFU/ ml of water sample was calculated (Robert*et al*, 2003).

Staphylococcus aurous

One ml of sample water was transferred aseptically to 9ml of distilled sterilized water and homogenized by Vortex machine (Biocote) and a serial of dilutions (10^{-1}, 10^{-2}, 10^{-3} ,$10^{-4,}$ 10^{-5}) was made by taking 1 ml from homogenized sample and adding to sterile test tube containing 9ml sterile distilled water and mixed properly by vortex machine then 0.1 ml of the dilutions 1:10, was spread plated on Manitol Salt Agar (MSA) and incubated at 37^0C for 24-48 hour . Golden yellow and Orange colonies were counted using digital colony counter and the colony was confirmed by staining and recorded as a result.

Bacillus

In order to count the *Bacillus* spps (aerobic spore formers) in the water samples taken from the streams of upper awash river the serial diluted water samples were first preheated at 75°C for 15 min in a water bath before inoculation in order to kill all the vegetative cells and 0.1ml of the heated and cooled water samples was spread plated on the nutrient agar and incubated at 32^0C for 18-24 hours. After incubation the colony formed on the nutrient agar was confirmed by gram staining and counted as *Bacillus* spps (aerobic endospore formers).

Gram staining

Bacillus

The slide was placed on the staining rack then a one colony of bacteria was placed on the slide and flood with malachite green then the bottom of the slide was heated with flame then allowed for 5 minute and washed with water finally the slide was fully covered with safranim and rinsed with tap water and air dry then the bacteria was examined under oil immersion

Statistical analysis

All the statistical analyses were done with the Statistical Package for the Social Sciences, version 24.The significance of differences in bacterial counts and physicochemical parameters of water samples from the stream of upper Awash River between sampling sites were determined by one-way analysis of variance (ANOVA). P value of <0.05 was considered significant.

Results

Total coliforms

In this investigations, the count of *total coliform* between the sampling rounds shows significant variation (P<0.05). The count of *total coliform* for three month from five sampling sites indicate that sampling sites Aw1 and Aw4 showed higher total coliform ranged from 1780CFU/100ml to 17300 CFU/100ml and 2050 CFU/100ml to 22000 CFU/100ml respectively. The minimum total coliform mean value was observed from the water sample taken from site Aw3 which was ranged between12200 CFU/100ml to1440 CFU/100ml (Table 1).

Table 1;*Total coliform count* CFU/100ml

Organism	Site	CFU/100ml Round 1	CFU/100 ml Round 2	CFU/100ml Round 3	CFU/100ml Round 4	CFU/100ml Round 5	CFU/100ml Round 6
TC	Aw1	1780^a	1600^c	2670^c	17300^d	15900^b	11000^a
	Aw2	1980^c	1280^b	2590^c	10300^c	16800^d	13600^c
	Aw3	2210^d	2290^e	1440^a	3200^a	6600^a	12200^b
	Aw4	2490^e	2050^d	2650^c	20900^e	19500^e	22000^e
	Aw5	1900^b	1010^a	2350^b	8700^b	16400^c	18300^d

Figures followed with the same superscript letter are not significantly different from each other (p ≥0.05)

Escherichia coli

The result obtained from laboratory analysis of *E.coli* of water samples from streams of upper Awash River during the six round of sampling showed mean values ranged from 200 CFU/ml to 11300 CFU ml/100ml.In the six sampling round, round one has highest load and round two has the lowest load. *E.coli* count was highest in the water sample taken from Aw3:11300 CFU/100ml and lowest in the water sample taken from site AW5: 200 CFU/100ml.There was no significant difference between impacted sites (p > 0.05) from the count of of*E.coli*in as compared to water sample taken from five different sampling sites

Table 2; *Escherichia coli count CFU*/100ml

Organism	Site	CFU/100ml Round 1	CFU/100ml Round 2	CFU/100ml Round 3	CFU/100ml Round 4	CFU/100ml Round 5	CFU/100ml Round 6
E.coli	Aw1	290^a	^{260b}	230^a	380^b	670^b	1980^b
	Aw2	360^b	200^a	610^c	690^c	790^c	3690^e
	Aw3	11300^d	310^c	210^a	210^a	220^a	2890^c
	Aw4	990^c	810^d	850^d	730^c	870^d	3320^d
	Aw5	300^a	200^a	450^b	350^b	700^b	510^a

Figures followed with the same superscript letter are not significantly different from each other (p ≥ 0.05)

Enterococcus

In this study, the highest *enterococcus* load was observed in round six and the lowest *enterococcus* load was observed in round one. Highest *Enterococcus* count was observed in the water samples taken from the site 4(Aw4); followed by the water sample taken from site 1 (Aw1), and the mean values were between 178 CFU/100ml to 2300CFU/100ml and 218CFU/100ml to 2270CFU/100ml respectively. The lowest *Enterococcus* count was observed in the water samples taken from site 5 (Aw5) and the mean value was ranged from 96CFU/100ml to 710

CFU/100ml. Over all there were significant differences between impacted sites ($p < 0.05$) and the less impacted site from the point of view of *Enterococcus* count (Table 3).

Table3 :*Enterococcus count* CFU/100ml

Organism	Site	CFU/100ml Round 1	CFU/100ml Round 2	CFU/100ml Round 3	CFU/100ml Round 4	CFU/100ML Round 5	CFU/100ml Round 6
Enterococcus	Aw1	218^{c}	248^{c}	332^{c}	440^{c}	1070^{b}	2270^{d}
	Aw2	144^{b}	112^{a}	266^{b}	358^{b}	1180^{c}	2030^{c}
	Aw3	346^{d}	330^{d}	126^{a}	158^{a}	134^{a}	1960^{b}
	Aw4	178^{bc}	406^{e}	480^{d}	402^{bc}	1280^{d}	2300^{d}
	Aw5	96^{a}	186^{b}	254^{b}	120^{a}	100^{a}	710^{a}

Clostridium perfringes

The highest *Clostridium perfringes* count was observed in round six and the lowest *Clostridium perfringes* was observed in round two. The highest *Clostridium perfringes* was detected from the water sample taken from site 2(Aw2) which was ranged between 201 CFU/100ml to 66 CFU/100ml and the lowest *Clostridium perfringes* counted was observed in water samples taken from site(Aw3) which was ranged 23 CFU/100ml to 95 CFU/100ml. There was no significant difference between the reference site and impacted sites ($p \geq 0.05$) for the number of *Clostridium perfringes* was count.

Table: 4. *Clostridium perfringes count* CFU/100ml

Organism	Site	CFU/100ml Round 1	CFU/100ml Round 2	CFU/100ml Round 3	CFU/100ml Round 4	CFU/100ml Round 5	CFU/100ml Round 6
Clostridium	Aw1	140^{b}	48^{a}	115^{b}	58^{a}	84^{b}	198^{b}
	Aw2	109^{b}	66^{ab}	56^{a}	180^{c}	201^{c}	156^{b}
	Aw3	61^{a}	61^{ab}	95^{ab}	48^{a}	23^{a}	56^{a}
	Aw4	150^{b}	88^{ab}	109^{b}	107^{b}	53^{ab}	168^{b}
	Aw5	119^{b}	104^{b}	51^{a}	45^{a}	31^{a}	87^{a}

Heterotrophic plate count (HPC) bacteria

Water samples taken from different streams of upper Awash River showed highly significant variation ($p \geq 0.05$) for *Heterotrophic* plate count. The highest HPC was observed in round four while the lowest HPC was observed in round one. Highest *Heterotrophic plate* count was observed from the water sample taken from site4(Aw4), followed by the water sample taken from site 1 (Aw1) and the lowest CFU was observed in Aw3.The *heterotrophic bacteria* isolated during the study consisted of pigmented (yellow, orange, pink and red) and non-pigmented (white) colonies. The high levels of HPC bacteria in the drinking water at Aw4may be due to different anthropogenic activities and wastes discharge in to the water channel in this site.

Table 5: Heterotrophic plate count CFU/ml

organism	site	CFU/ml Round 1	CFU/ml Round 2	CFU/ml Round 3	CFU/ml Round 4	CFU/ml Round 5	CFU/ml Round 6
HPC	Aw1	900^{d}	980^{d}	990^{d}	1100^{d}	1900^{d}	1790^{d}
	Aw2	720^{c}	730^{b}	890^{c}	980^{c}	1340^{c}	1290^{c}
	Aw3	350^{a}	520^{a}	410^{a}	310^{a}	730^{a}	530^{a}
	Aw4	960^{e}	1100^{e}	1110^{e}	990^{c}	2320^{e}	2380^{e}
	Aw5	430^{b}	860^{c}	470^{b}	610^{b}	980^{b}	880^{b}

Bacillus

Water samples taken from different site of streams of upper Awash River showed significant differences for the *Bacillus spp* count. The highest *Bacillus* spp count was observed in round five and the lowest bacillus spp was observed in round one. The highest *Bacillus* spp count was recorded from the water sample taken from the site 4(Aw4) followed by the water sample taken from site1 (Aw1). The lowest *Bacillus* spp count was observed in the water samples taken from site 3 (Aw3).

Table6: Bacillusspp count CFU/ml

Organism	Site	CFU/ml Round 1	CFU/ml Round 2	CFU/ml Round 3	CFU/ml Round 4	CFU/ml Round 5	CFU/ml Round 6
Bacillus	Aw1	560^{c}	730^{d}	540^{b}	670^{d}	990^{d}	790^{c}
	Aw2	470^{b}	610^{c}	500^{b}	620^{c}	710^{b}	550^{b}
	Aw3	320^{a}	320^{a}	420^{a}	390^{a}	490^{a}	470^{a}
	Aw4	590^{c}	700^{d}	660^{c}	940^{e}	840^{c}	910^{d}
	Aw5	330^{a}	460^{b}	430^{a}	510^{b}	530^{a}	580^{b}

Staphylococcus aureous

The water samples taken from different sites of the upper Awash River showed significant differences for the count of *Staphylococcus*. The highest *staphylococcus aureous* was observed in round six and the lowest *Staphylococcus aureous* was observed in round one. The highest *Staphylococcus* count was observed from the water sample taken from site 4(Aw4) and the mean value range from 580 CFU/ml to 1950 CFU/ml. the lowest *Staphylococcus* count was observed from the water sample taken from the site 3 (Aw3) and the mean value range from 300 CFU/ml to 800 CFU/ml. Figure 2 indicates the picture of the different bacterial isolated colony and light microscope photo of the bacillus and *Staphylococus spp*

Table 13: StaphylococcusCFU/ml

Organism	Site	CFU/ml Round 1	CFU/ml Round 2	CFU/100ml Round 3	CFU/100ml Round 4	CFU/100ml Round 5	CFU/100ml Round 6
Staphylococcus	Aw1	730^{e}	910^{d}	560^{b}	1020^{e}	690^{c}	2640^{c}
	Aw2	520^{c}	760^{b}	670^{c}	790^{c}	1990^{e}	1930^{b}
	Aw3	300^{a}	530^{a}	380^{a}	460^{b}	320^{a}	800^{a}
	Aw4	580^{d}	840^{c}	860^{d}	940^{d}	1690^{d}	1950^{b}
	Aw5	370^{b}	490^{a}	360^{a}	400^{a}	610^{b}	770^{a}

value followed with the same superscript letter are not significantly different from each other ($p \geq 0.05$)

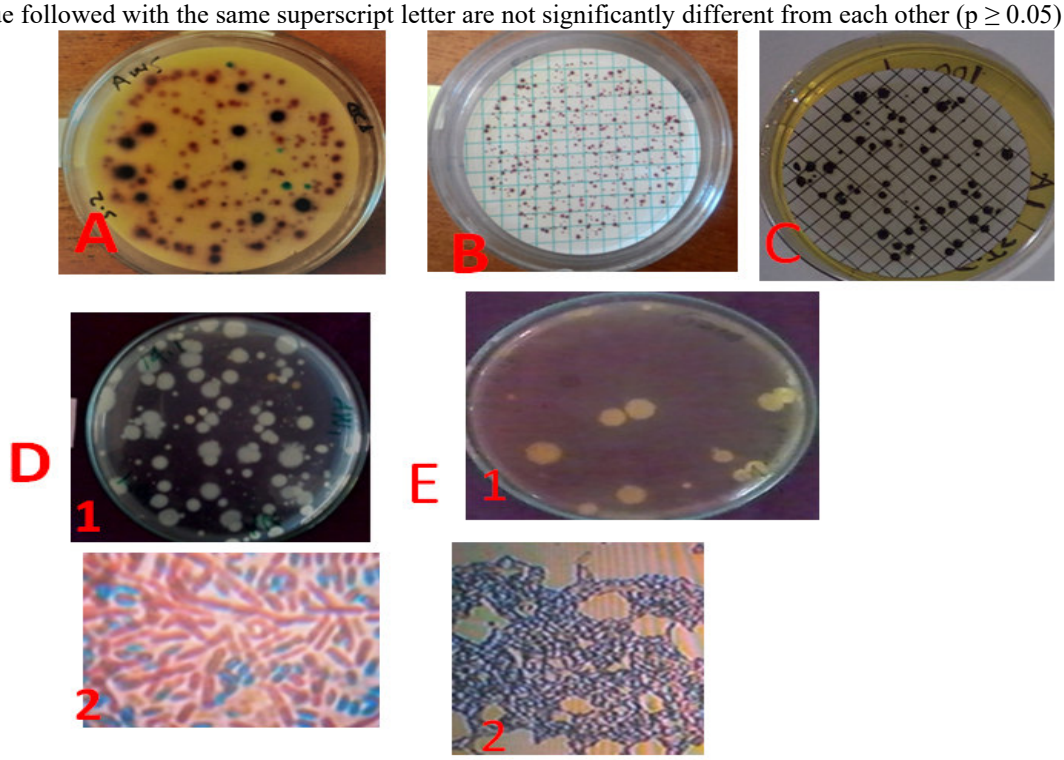

Fig2: The different indicator microorganisms isolated in this investigations A) coloforms and E.coli.; B) Entrococcus, C) Clostridium prefringins, 1D) hetrotrophic plate count, 2) Endospore of Bacillus spp, E)1, staphylococcus isolate ; 2, Gram reaction of Staphylococcus

Discussion

The results of this study indicated that, the count of *total coliform* for three month from five sampling sites indicate that sampling sites Aw1 and Aw4 showed higher total coliform ranged from 1780CFU/100ml to 17300 CFU/100ml and 2050 CFU/100ml to 22000 CFU/100ml respectively. This is in line with the reports of Mohammed Yasin *et al*., (2015), which indicated that all water samples collected from unprotected water sources in Jima zone were positive for total coliforms and fecal coliforms (FC). And FC was detected in 80 % of the total samples with counts ranging between 0.67 and 266.67 CFU/100 ml although 66.67 % of tap water samples were negative for FC.Reportes from the study of Rural Communities of Dire Dawa Administrative Council indicated that, about 83.34% of the water sample was positive for indicator bacteria (*Desalegn Amenu etal.2012*)**.**The similarities this indicator microorganisms count may be due to the fact that all the water sources were unprotected as well as untreated, from the all sampling round, round one has highest load and round two has the lowest load,*for E.coli* count in the water sample taken from Aw3:11300 CFU/100ml and lowest in the water sample taken from site AW5: 200 CFU/100ml.This result is in agreement with the report ofAbebe Berhanu and Dejene Hailu (2015)

who investigated, Bacteriological and Physicochemical Quality of Drinking Water Sources and Household Water Handling Practice Among Rural Communities of Bona District, Sidama Zone-Zouthern, Ethiopia. According to these authors**,** majority (86 %) of the protected springs and wells in the study area did not fulfill the WHO's criteria for drinking water quality standards. Water schemes with high sanitary risk scores had high number of E. coli/100 ml of sample water. Highest *Enterococcus* count was observed in the water samples taken from the site 4(Aw4) and followed by the water sample taken from site 1 (Aw1), and the mean values were between 178 CFU/100ml to 2300CFU/100ml and 218CFU/100ml to 2270CFU/100ml respectively. The lowest *Enterococcus* count was observed in the water samples taken from site 5 (Aw5). Enterococci are frequently isolated in soil, plants, vegetables (Facklam, and Teixeira, . 1997) , in a variety of small-scale cheeses produced from cow, goat and buffalo milk (Andrighetto, *et al*., 2001)and in various foods (Franz, et al., 19990). Because of their high concentration in feces and their long survival in the environment, enterococci have been proposed as water fecal contamination indicators (US EPA 1986; Chenoweth, and Schaberg,1990). Epidemiological studies have indeed demonstrated a strong correlation between the presence of enterococci in water and disease risk (Kay*et al*. 1994). The highest *Clostridium perfringes* was detected from the water sample taken from site 2(Aw2) which was ranged between 201 CFU/100ml to 66 CFU/100ml and the lowest *Clostridium perfringes* counted was observed in water samples taken from site(Aw3) which was ranged 23 CFU/100ml to 95 CFU/100ml. *Clostridium perfringes* (CP) is an anaerobic, Gram-positive, rod-shaped bacillus which produces spores. Obligate anaerobic bacteria cannot tolerate oxygen and die in the presence of oxygen (Willey *et al*., 2008).*Clostridium perfringes* can have serious health impacts on humans including gangrene and gastrointestinal disease. Highest *Heterotrophic plate* count was observed from the water sample taken from site4 (Aw4), followed by the water sample taken from site 1 (Aw1) and the lowest CFU was observed in Aw3.The *heterotrophic bacteria* isolated during the study consisted of pigmented (yellow, orange, pink and red) and non-pigmented (white) colonies, (Carter *et al.,* 2000) isolated pigmented and non-pigmented colonies (especially yellow and orange colonies) in drinking water are an indication of a change in the water quality. Divekulu Siyum and DelelegnWoyessa (2013) reported , the mean cfu/ml of aerobic mesophilic bacteria (AMB) was 815.4; while the mean cfu/ml was 264 and 306.8 for coliforms and enterobacteriaceae, respectively. The isolated colonies also displayed diverse morphological and biochemical features.The highest *Bacillus* spp count was observed in round five and the lowest *Bacillus* spp was observed in round one. The highest *Bacillus* spp count was recorded from the water sample taken from the site 4(Aw4) followed by the water sample taken from site1 (Aw1). The lowest *Bacillus* spp count was observed in the water samples taken from site 3 (Aw3). The highest *Staphylococcus aureous* was observed in round six and the lowest *Staphylococcus aureous* was observed in round one. The highest *Staphylococcus* count was observed from the water sample taken from site 4(Aw4)and the mean value range from 580 CFU/ml to 1950 CFU/ml. the lowest *Staphylococcus* count was observed from the water sample taken from the site 3 (Aw3) and the mean value range from 300 CFU/ml to 800 CFU/ml. Adegboyega et al. 2015 reported that t*he bacteriological parameters analysed were total viable count which had values ranging from 2.02 x 102 to 6.08 x103cfu/ml. Bacteria isolates were identified as Bacillus sp, Escherichia coli, Pseudomonas sp, Salmonela spp, Aeromonas spp and Vibrio cholera.* The fact that, about 83.34% of the water sample was positive for indicator bacteria shown that the three selected Peasant Association had risk of contamination. High concentration of microbiological indicators in all water sources of this study area have demonstrated the presence of pathogenic organisms which constitute a threat to anyone consuming or in contact with these waters. (Desalegn Amenu *et al*., 2012**).** The majority of the drinking water sources is either of unacceptable quality or grossly polluted.

Conclusions

The bacteriological quality of most water samples analyzed in the current study did not meet the standards set for drinking water. The total colifrom bacteria, E.coli, *Entrococcus*, *Clostriuduim prifringens*,heteroptrophic count and *Staphylococcus aurous* count where by far higher than the WHO criteria for the different water uses. Thus, with the current high dependence on alternative water sources other than tap water, it calls for awareness development on hygienic handling of spring and streams besides designing protections and regular purification strategies by the concerned bodies.

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