JIMMA UNIVERSITY COLLEGE OF NATURAL SCIENCES SCHOOL OF GRADUATE STUDIES DEPARTMENT OF BIOLOGY



MICROBIOLOGICAL AND PHYSICHO CHEMICAL QUALITIES OF DRINKING WATER SOURCES IN SEKA CHEKORSA TOWN AND ITS SURROUNDINGS, JIMMA ZONE, SOUTH WEST ETHIOPIA

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ABBREVIATIONS AND ACRONYMS

MLS	Membrane Lauryl Sulfate		
NTUs	Nephelo-metric Turbidity Units		
TC	Total coli forms		
TTC	Thermo tolerant coliforms		
TCR	Total coliform rule		
FC	Faecal coliform		
WHO	World Health Organization		
APHA	American Public Health Association		
UNCF	United Nations Children's Fund		
CFU	Colony Forming Unit		
BOD	Biological oxygen demand		
TSS	Total suspended substances		
TDS	Total Dissolved Solid		
VFSS	Volatile fraction of suspended solid		
PCA	Plate count agar		
MPN	Most probable number		
EMB	Eosine Methylene Blue		
FAAS	Flame Atomic Absorption Spectrometer		
LB	Lactose broth		
TSI	Triple Sugar Iron		
SIM	Sulfur Indole motility		
WWAP	World water assessment program		
USEPA	United states environmental protection agency		
ANOVA	Analysis of variance		
CSA	Central statistical authority		
EPA	Environmental protection agency		
SPSS	Statistical package for the social sciences		
ADWG	Australian Drinking Water Guidelines		

OPERATIONAL DEFINITION OF TERMS

Double strength broth: Refers to broth made using twice the normal amount of broth powder.

Single strength broth: is the broth that contains the normal amount of broth powder as instructed by the manufacturer.

Aerobic Bacteria: Bacteria which can live and reproduce only in the presence of "free" or dissolved oxygen.

Aseptic Conditions: Free of contamination by living microorganisms, i.e. bacteria.

Coliform: A group of bacteria which can be used as an indicator of pollution. Major portion of this group of organism lives in the intestinal tracts of warm blooded animals, including human being.

Colony: A group of bacteria growing on a supporting surface. The colony is considered to be the result of the growth and reproduction of a single cell.

Disinfection: To destroy most (but not necessarily all) of the harmful or objectionable microorganisms by means of chemicals, heat, ultraviolet light, etc.

Fermentation: The process by which bacteria convert organic matter into carbon dioxide and water.

Fermentation Tube: A container designed to allow easy identification of gas production.

Fecal Coliform: A subclass of the coliform bacteria which originate almost exclusively in the intestinal tract of warm blooded animals.

MPN: The most probable number (MPN) of coliform or fecal coliform bacteria per unit volume of a sample. It is expressed as the number of organisms which are most likely to have produced the laboratory results noted in a particular test.

Sterilization: Destruction or removal of all viable or living organisms.

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Abstract

Water is basic necessity for human health and biological activity within living things. The quality of drinking water has always been a major health concern, especially in developing countries. This study focused on the assessment of microbial quality and some physicochemical properties of drinking water samples of different sources in seka chekorsa town, Jimma zone, and its surroundings. The standard plate count method was used for bacteriological analysis for enumeration of total and fecal coliforms; Most Probable Number (MPN) method was used. Physico-chemical analysis was done following standard procedures of American Public Health Association (2011). Results of the current study showed that only 37% of population in the study area had access to potable drinking water. About 56.6% of the study subjects dispose waste material in open field. The entire water sample from wells and springs were positive for total coliform and fecal coliform. Fecal coliform were recorded in 60% of the samples with a mean value ranging from 0.5 to 5.26 CFU/100ml, but 64% of tap water samples were found negative for fecal coliform, and E. coli were not detected in all tap water samples. Entrobacteriaceae, Bacillus and Pseudomonas were among the dominant bacterial groups frequently isolated from the water samples. Salmonella was detected in two water samples (one each from well and spring) while Shigella species were encountered in none of the water sample. The mean temperature of all the three categories of water samples ranged between 20.2°C to 24.4°C and that of pH was between 5.85 and 8.56. The lowest and highest mean values of total dissolved solid recorded from the water samples were 136 and 331mg/l, respectively. The mean concentration of total suspended solid and turbidity values ranged between 11.2 and 47.4mg/l and 1.53 to 55.08 NTU, respectively. Likewise, electro-conductivity and Nitrate concentrations ranged from 43.1 to 407µs/cm and 0.535 to 14.764mg/l, respectively. In general, the results of the present study have shown that some of the Physico chemical and bacteriological parameters had values beyond the maximum recommended limits set earlier. Thus, it is recommended that the government and the other responsible authorities have to take appropriate corrective measures to curb the existing health problems through improvement of access to potable water and regular monitoring of the existing condition besides awareness development on hygienic practices.

Keyword: *Water, Physico - chemical, bacteriological, coliform, most probable number, fecal coliform, total coliform, Disinfection, Fermentation Tube, Aseptic Conditions.*

1. Introduction

1.1. Background

Water is one of the most important and abundant compounds of the ecosystem. All living Organisms on the earth need water for their survival and growth. As of now only earth is the Planet having about 70 % of water. But due to increased human population, industrialization, Use of fertilizers in the agriculture and man-made activity is highly polluted with different harmful contaminants. Therefore it is necessary that the quality of drinking water should be checked at regular time interval, because due to use of contaminated drinking water, human Population suffers from varied of water borne diseases. It is well known that access to safe water and sanitation are important in reducing disease transmission (Newton, 2003). The world health organization estimated that approximately 80% of all sickness and disease in the world are caused by inadequate sanitation, polluted water or unavailability of water (WHO, 2004). Most infectious diseases are caused by pathogenic microorganisms such as bacteria, virus, protozoa and other parasites that cause life threatening conditions in relation to drinking water. Insufficient treatment of water and contamination of the water by various pathogenic microorganisms can be potential to cause water borne diseases of diverse symptoms. The symptoms for these illnesses range from fever and malaise to gastro-intestinal symptoms such as diarrhea and stomach aches. Waterborne illnesses usually occur when sanitation and living conditions of human beings are generally poor in addition to lack of safe water sources (Verheyen et al., 2009). For instance, Study from some East African countries revealed that determinants of diarrhea morbidity are attributed to poor hygiene (unsafe disposal of faces and wastes), education level of household, obtaining water from wells or surface sources and per capita water used for cleaning (Tumwine et al., 2002).

The risk of diseases like diarrhea becomes worse when people continuously drink or use water obtained from wells, live in poor housing conditions, and have low family income and low educational attainment. Water borne diseases can enter in to lake, river, stream, ground water and other body of water through any anthropogenic and natural processes worldwide. Any time such contaminated water may be used for drinking, cooking, swimming or other purposes and hence, there could be a risk that such organism will enter the body and cause disease and death (WHO, 2008). Certainly the majority of these illnesses are due to waterborne diseases, there is a need for improvements in water supply and

sanitation services. Moreover, improvement must be accompanied by activities to promote change in health related waterborne diseases (WHO, 2008). According to (Farris and Kaba, 2009), assessment of the microbiological quality of drinking water primarily aims at protecting people who consume water, which may contain pathogens that lead to water borne diseases and illnesses. Therefore, this study will be designed to assess the microbiological and physicochemical quality of drinking water from different sources in seka chekorsa town and its surroundings.

1.2. Statement of the Problem

Water quality deterioration is a big issue in many countries water supply system, which may be a result of many interconnected physical, chemical, and biological factors. It may or may not be at the source only rather it may happen after leaving the source, on the path from source to consumers tap. Moreover, it is also very difficult to identify strictly the cause as well as the place of pollution. Because of this by assessing water quality based on indicator parameters such as turbidity, Biological oxygen demand, PH, and faecal microorganisms different reports showed that significant level of water pollution both at sources and distributions systems. This indirectly determines the risk of ingesting pathogens and chemicals with polluted water APHA (2008). The reports also showed that water sources and distribution systems of towns and rural communities alike have serious water quality problems. Assessment of bacteriological and Physico-chemical qualities of urban source water and tap water distribution systems in sub-city of Addis Ababa (Mengstayehu, 2007) showed contaminations of water by indicator bacteria such as total coli forms, fecal coli forms and/or faecal streptococci. Health Office reports indicate that among the top ten diseases registered in the area waterborne disease take the higher ranks and among all typhoid, cholera, helminthiasis and diarrhea were the most rapidly occurring waterborne diseases (Addis sub-city, 2003). A study conducted in Jimma town showed that very high nitrate concentration in protected springs indicating the presence of organic pollution (Sofonias and Tsegaye, 2006). These results provide convincing evidence that water quality problems are both in urban and rural drinking water sources. This condition would create high health risks to users unless there is timely intervention. Therefore, evaluation of microbial and physicochemical water quality status of urban and rural drinking water source is very important. To this effect, the present study was designed to evaluate the safety status of different water sources being used for drinking in seka chekorsa town and its surroundings.

1.3. Significance of the study

In Ethiopia, wells and springs are the dominant sources of drinking water used to supply major urban and rural communities. According to most research reports the quality of drinking water sources are classified as grossly polluted. This showed that the drinking water source contamination problems in the country. Thus, this study is expected to generate useful data providing accurate information on the quality of drinking water that the consumers are using for drinking purpose. The finding of this research addresses the microbiological and physicochemical quality of water sources in Seka town and its surroundings. It also enriches the available literature on water quality analysis in Ethiopia besides serving as baseline data for further study in other areas

Hypothesis

1. The water sources could be vulnerable to bacterial and Physico chemical contamination.

2. There could be difference in bacterial and Physico chemical levels among wells, taps and spring water sources

1.4. OBJECTIVES OF THE STUDY

1.4.1. General Objective

The general objective of this study was to evaluate the bacteriological and physicochemical quality of drinking water source in Seka chekorsa town and its surroundings.

1.4.2. Specific Objectives

- To evaluate the people's level of awareness on water borne diseases and the care being taken in the study area.
- ✓ To determine the bacterial load of water sources being used for drinking in and around Seka town.
- ✓ To assess the contamination level of drinking water sources using total coliforms and fecal coliforms as pollution indicators.
- ✓ To analyze Physico-chemical quality of water samples, including Temp, turbidity, pH, total suspended solids (TSS), total dissolved solids (TDS), electric conductivity, biological oxygen demand (BOD), nitrate and phosphate levels.

2. Literature review

2.1. History of drinking water regulations

Drinking water supplies have a long history of being infected by a wide spectrum of microbes. Therefore, the primary goal of water quality management from health perspective is to prevent consumers from exposure to pathogens that cause disease. Disinfection and protection of water sources have greatly reduced the incidence of this disease in developed countries .Therefore, testing the source of water is necessary, especially when there is no water treatment. This is useful as result of the failure of treatment process or as a part of an investigation of serious water borne disease outbreak WHO (2008).

Safe Drinking Water Act (SDWA) was established for the first in America. American congress passed an act that brought change in America's drinking water system .Since the SDWA of 1974 established, water system have encountered many new regulations, such as meeting specific water quality standards, monitoring for contaminants and submitting water quality reports. For the first time, the 1974 acts the authorized the U.S. Environmental protection Agency (USEPA) set standards for any contaminant in public water systems that adversely affects public health (United States Environmental Protection Agency,2012). Water resources are used in various ways including direct consumption, agricultural irrigation, fisheries, hydropower, industrial production, reaction, navigation, environmental protection, the disposal treatment of sewage and industrial effluents(United States Environmental Protection Agency,2012).

2.2. Water Quality

Water quality is a technical term that is based upon the characteristics of water in relation to guideline values of what is suitable for human consumption and for all usual domestic purposes, including personal hygiene. Components of water quality include microbial or biological, chemical, and physical aspects (EPA, 2012).

2.3. Bacteriological Quality of water

The presence of certain microorganisms in water is used as an indicator of possible contamination and an index of water quality (Hurst *et al.*, 2002). Indicator organisms are selected to demonstrate the

presence of human and animal wastes and hence the potential presence of pathogens in drinking water. Indicator organisms are usually of intestinal origin from humans and animals (Hurst *et al.*, 2002). The most common and widespread health risk associated with drinking water is contamination, either directly or indirectly, by human or animal excreta, and with the micro-organisms contained in faces. Monitoring of specific bacterial, viral and protozoan pathogens is usually complex, expensive, and time consuming, and may fail to detect their presence. In monitoring for microbiological quality, reliance is therefore placed on relatively rapid and simple tests for the presence of indicator organisms. The three common organisms used as microbial indicators are total coliforms (TC), thermo-tolerant coliforms (TTC) or alternatively *E. coli* and Enterococcus (ADWG, 2001).

2.3.1. Total Coliform

Coliforms are aerobic or facultative anaerobic, Gram negative non-endosperm forming rod shaped bacteria that ferment lactose with the production of acid and gas within 48 hours of being placed in a medium at 35°C. Total coliform (TC) bacteria comprise many members of the family Entrobacteriaceae. TC bacteria are those that can grow in selective media at 35°C and ferment lactose or possess a β -galactosidase enzyme, as an indicator of fecal contamination. they are not useful as an index of fecal pathogens, but they can be used as an indicator of treatment effectiveness and to assess the cleanliness and integrity of distribution systems and the potential presence of bio films (WHO, 2008).On the one hand, the Total Coliform group of bacteria is unreliable indicators of fecal contamination because many members are capable of growth and long term persistence (having a nonfecal origin) in many environments, including water distribution systems. On the other hand, there are more TC bacteria in untreated fecal waste than any of the other fecal indicators or indicator groups, making the TC test the most sensitive of all indicator tests. Because of this sensitivity, the TCR (total coli form rule) relies on the TC bacteria test as the initial test to detect the possible presence of fecal contamination in delivered water, as well as to assess water treatment effectiveness and the integrity of the distribution system. Water from a distribution system that is free of TC bacteria should have no or minimal levels of pathogens.

2.3.2 Fecal Coli form

Under the TCR, if the TC test result is positive, that sample is then further tested for the presence of fecal coliform (FC) bacteria. Since it is difficult to monitor disease carrying microorganisms directly

we use the count of FC bacteria as a standard measure and indicator of disease potential. The presence of FC bacteria in water indicates that fecal material from mammals or birds is present, so organisms that cause water borne diseases may be present as well. The FC group of organisms is a subset of the TC group that can grow in selective media at 44.5°C and ferment lactose, majority of FC bacteria are E. coli (United Nations Children's Fund, 2011).

2.3.3. Enterococcus

Enterococcus is facultative organisms, *i.e.*, they are capable of cellular respiration in both oxygen-rich and oxygen-poor environments. Though they are not capable of forming spores, Enterococcus is tolerant of a wide range of environmental conditions: extreme temperature (10-45°C), pH (4.5-10.0) and high sodium chloride concentrations (Pelletier, 2003).

2.4 Physico-Chemical Water Quality Parameters

Drinking water quality acceptability is governed by limits of microbiological and Physico-chemical parameters. Because changes in water chemistry tends to be longer-term, chemical testing is not undertaken as frequently as microbiological analysis. However, some of the Physico-chemical parameters essential in water quality investigation are discussed here in after (Adejuwon *et al.*, 2011).

2.4.1 Turbidity

It is the optical property of a water sample that causes light to be scattered and absorbed rather than transmitted in straight lines through the sample. In simple terms, turbidity answers the question, how cloudy is the water? Moreover light's ability to pass through water depends on how much suspended material is present. Turbidity may be caused when light is blocked by large amounts of silt, microorganisms, plant fibers, sawdust, wood ashes, chemicals and coal dust. Any substance that makes water cloudy will cause turbidity. Additionally, the most frequent causes of turbidity in lakes and rivers are plankton and soil erosion from logging, mining, and dredging operations (EPA, 1999). *Electronic turbid-meter*, a device that used for the most accurate way of determining water's turbidity, has a light source and a photoelectric cell that accurately measures the light scattered by suspended particles in a water sample. The results are then reported in units called Nephelo-metric Turbidity Units or NTUs (Alanc *et al.*, 2000).

2.4.2. Color

Color is derived from the back scatter of light passing through the water, and is influenced by the dissolved or suspended constituents in the water. Color can be the result of natural factors (e.g., dissolution of iron from iron-rich minerals, and dissolved humic materials) or factors that result from human-based activities such as effluent discharge from industrial activities (Aftab, 2005).

2.4.3. Taste and Odor

Taste and odor problems in source waters are primarily an aesthetic concern; however, they can undermine consumer confidence in water supplies, and result in millions of dollars annually in treatment costs to the water industry (Atnaf, 2006).

2.4.4. Temperature

Temperature is measure of the kinetic energy of the particle. The amount of dissolved oxygen in water system is dependent on the water temperature .When temperature of the water increases the solubility of minerals, salts, metals, cat ions or anions in water increases. Water will heat up more rapidly and hold more heat; this, in turn, might adversely affect aquatic life that has adapted to a lower temperature. Temperature of the water could be determined by analytical methods such as mercury thermometer and multi parameter probe. For instance, (Kerketta et al., 2013) utilized mercury thermometer to measure the temperature of drinking water from different sources and obtained the temperature of the water in the range of 24 to 31°C. Temperature affects both biological and chemical functions. Chemical equilibrium constants, solubility's, and the rates of chemical reactions are all temperature-dependent. High water temperature enhances the growth of microorganisms and may increase taste, odor, and color problems of drinking water .Temperature also affects the concentration of dissolved oxygen and can influence the activity of bacteria in water bodies (Olson ,2004).

2.4.5. Nitrate

Nitrates get into water ways from lawn fertilizer run-off, leaking septic tanks and cesspools, manure from farm livestock, animal wastes (including fish and birds), and discharges from car exhausts. Nitrates can be reduced to toxic nitrites in the human intestine, and many babies have been seriously poisoned by well water containing high levels of nitrate, in order to check the concentration of nitrate

it can be measured at Point of entry, Reservoir inlets/outlets the end of distribution network (WHO, 2004).

2.4.6. Chlorine residual

Disinfection is a process designed for the deliberate reduction of the number of pathogenic microorganisms. While other water treatment processes, such as filtration, coagulation, flocculation and sedimentation, may achieve pathogen reduction, this is not generally their primary goal. A variety of chemical or physical agents may be used to carry out disinfection. Chlorine may be used as a disinfectant in the form of compressed gas under pressure which is dissolved in water at the point of application, solutions of sodium hypochlorite, or solid calcium hypochlorite (APHA, 2004).

2.4.7. pH

PH is an important parameter which is evaluating the acid-base balance of water .Also it is the indicator of acidic or alkaline condition of water status. The balance of positive hydrogen ions (H+) and negative hydroxide ions (OH-) in water determines how acidic or basic the water is. In pure water, the concentration of positive hydrogen ions is in equilibrium with the concentration of negative hydroxide ions, and the pH measures exactly 7 (Genet ,2008). Basically, the pH is determined by the amount of dissolved carbon dioxide [CO2], which forms carbonic acid in water (Genet, 2008).

2.4.8. Total suspended solids (TSSs) and total dissolved solids (TDSs).

Total suspended solids (TSS) - refer to small solid particles, which remain in suspension in water *as a colloid (or are any particles/substances that are neither dissolved nor settled in the water)* (WHO, 2004). TSSs including the volatile fraction of suspended solid (VFSS), are commonly monitored to evaluate the degree of pollution of natural waters and serve as a key parameter in the quality of drinking water. It is also used as one indicator of water quality (Cheesbrough, 2006). Suspended solid is a pollutant that may carry pathogens on its surface. The smaller the particle size, the greater the surface area, and so the greater the pollutant load that is likely to be carried.

Total dissolved solids (TDSs) - refer to any substance including minerals, salts, metals, cat ions or anions that are dissolved in water. It includes anything present in water other than the pure water molecule and suspended solids. TDSs come from organic sources such as leaves, silt, plankton, and

industrial waste and sewage. In general, TDS concentration is the sum of the cat ions (positively charged) and anions (negatively charged) in the water. Other sources of TDS are runoff from urban areas, fertilizers and pesticides that are used on farms⁻ Dissolved solids also come from inorganic materials such as rocks and air that may contain CaHCO₃, N, P, Fe, S and other minerals. Many of these materials form salts, which usually dissolve in water forming ions (WHO, 2004).

2.4.9. Electric conductivity

The Electric conductivity (EC) of the water sample is its ability to conduct electricity. The conductivity of water is more or less linear function of the concentration of dissolved ions. It is used to give information about the levels of inorganic substances including Ca, HCO_3^- , N, P, Fe, Cu, S and others in the water. Most these dissolved inorganic substances are present in water in their ionized forms and hence contribute to conductance. Sudden increase in the conductivity of water indicates the availability of the sources of dissolved ions in the vicinity of the water. EC is measured by analytical methods such as digital conductivity meter and multi parameter probe (Cheesbrough, 2006).

2.5 WHO and Ethiopian standards of drinking water quality

Water is essential to sustain life, and a satisfactory (adequate, safe and accessible) supply must be available to all. Improving access to safe drinking-water can result in tangible benefits to health. Every effort should be made to achieve drinking-water that is as safe as practicable. Safe drinking water, as defined by the Guidelines, does not represent any significant risk to health over a lifetime of consumption, including different sensitivities that may occur between life stages. In the development and implementation of standards it is essential to consider the current or planned legislation related to water, health and local government and the capacity of regulators in the country. For this study, WHO and Ethiopian guidelines values for drinking water are presented in Table 1.

Number	Parameter	WHO (1993) standard	Ethiopian standard
			(1998)
1	рН	6.5-8	6.5-8
2	Turbidity(NTU)	<5 at disinfection point	<5
3	Free chlorine residual(mg/L)	0.2-0.5 at distribution point	0.1-0.2
4	Fecal coli form(CFU/100mL)	0	0
5	Total coliform (CFU/100mL)	_	0
6	Nitrate (mg/L)	0.5	0.5
7	Ammonia (mg/L)	-	0.5

Table1. WHO and Ethiopian guide line values of drinking water (http://www.lenntech.com)

2.5.1 Health Risks Associated with Water

Water is a basic necessity for life. Unfortunately, not all water helps human to survive. Water from contaminated sources causes numerous diseases and untimely deaths. The fact that a human needs water and cannot live without it forces him to use it even for drinking purposes, from any source, whether pure or contaminated (Zeyede and Tesfaye , 2004). Usage of quality deteriorated water may be a cause for the existence of water born, water washed, water based and water related diseases. The term water associated disease is used to describe all infections whose causing agents are carried by water (OECD, 2005). These are cholera, bacillary dysentery, Escherichia Coli (E.coli), viral hepatitis A, shigellosis, typhoid fever, cryptosporidiosis, giardiasis (WHO, 2004). Generally, waterborne disease outbreaks usually involve, source contamination and the breakdown of the treatment systems, contamination of the distribution systems and the use of untreated water (WHO, 2004). Water-associated disease can be defined as a disease in relation to water supply and sanitation. There are four categories this are: - Waterborne disease, Water-washed disease, Water-based disease and Water-related disease (Joseph *et al.*, 2003).

2.5.2 Waterborne diseases

Several infections enteric or intestinal diseases of man are transmitted through water contamination by faecal matter. Pathogens excreted in water by an infected person include all major categories such as bacteria, viruses, protozoa and parasitic warms. In this category water acts as a passive vehicle for the infectious agent. Some water born disease and their disease causing micro-organisms are presented in Table 2

Diseases	Disease causing organism	Source
Typhoid	Salmonellae typhus	Human feces
Cholera	Vibrio cholera	Human feces
Amoebic dysentery	Entamoeba histolitica	Human feces
Giardiasis	Giardia lamblia	Human or animal feces

Table 2. Waterborne diseases and their disease causing organism (etiological agents)

2.6 Link between Disease and Environment

Unlike genetic diseases, which individuals are predisposed to, acquiring water, sanitation and hygiene related diseases are controllable and preventable. The spread of these diseases depends on environmental conditions and behavior in the household and community. The majority of these preventive measures are related to environmental conditions: appropriate shelter and site planning, clean water, good sanitation, vector control, personal protection (such as insecticide-treated nets), personal hygiene and health promotion. These measures address conditions in the environment, known as 'risk factors' because they can cause disease. It is important to understand the relationship between disease and environmental risk factors because interventions must target risk factors properly (USEPA, 2012).

Table 3: Overview of diseases and environmental risk factors (Public Health Guide for Emergencies,2000)

Disease	Symptoms	Environmental risk factors	Health hazards
upper Respiratory tract infections	All symptoms of the common cold, fever and heavy coughing	Crowding, poor hygiene	Influenza and pneumonia
Diarrhea	Watery stools with or without blood or slime and vomiting	Contaminated drinking water, food, poor sanitation	Dehydration, especially in children
Cholera	Modest fever, severe, Liquid diarrhea ,abdominal spasms and vomiting	As for diarrhea	As for diarrhea
Meningococcal meningitis	No symptoms for a considerable time. When an epidemic is in progress, headache, fever and general malaise will suggest the diagnosis,	Crowding	Often fatal if untreated at an early stage
Shigella dysentery	Diarrhea with blood in the stools, fever, vomiting and abdominal cramps	Contaminated drinking water or food, or poor sanitation, poor hygiene	Case fatality rate may be High
Typhoid fever	Starts like malaria, sometimes with diarrhea, prolonged fever, occasionally with delirium	As for diarrhea, and contaminated Foods	Without appropriate medical care, includingantibio
Measles	A disease of early childhood, characterized by fever and catarrhal symptoms, followed by maculopapular rash in mouth	Crowding, poor hygiene Very contagious	Severe constitutional symptoms, high case fatality rate
Viral hepatitis A	Nausea, slight fever, palecoloured stools, dark colored urine, jaundiced eye whites and skin	Poor hygiene, contaminated foods and water	Long-term disabling effects
Diphtheria	Inflamed and painful throat, coughing	Crowding, poor hygiene	Secretion is Deposited in the respiratory tract.

2.7 Health Effects of Drinking Water Contaminants

Chemicals in drinking water which are toxic may cause either acute or chronic health effects. An acute effect usually occurs almost immediately and it is easy to obtain the source as well as the possible solution. Top ten diseases occurred in 2003 E.c are presented in Table 4

Table 4 .Top 10 water borne diseases recorded in Addis Ababa sub-city (Addis Ababa Sub-city HealthOffice report, 2003)

Number	Disease		Number of people	
		male	female	total
1	cholera	15,944	21,480	37,424
2	Typhoid	5,968	8,652	14,620
3	Diarrhea	5,620	4,892	10,512
4	Amoebic dysentery	2,976	6,560	9,536
5	Dysentery	3,276	5,912	9,188
6	Cryptosporidiosis	4,284	4,492	8,776
7	Giardiasis	2,900	3,720	6,620
8	Traveler's diarrhea	1,876	2,184	4,060
9	Dracunculiasis	2,088	1,444	3,532
10	Attitus	1,320	1,452	2,772

2.8. Distribution System Water Quality Deterioration Factors

A distribution system's pipes and storage facilities constitute a complex network of uncontrolled physical, chemical, and biological reactions that can produce significant variations in water quality. The principal factors that affect water quality during distribution are the system's structure, its operation, and a number of water quality factors (Gundry *et al.*, 2006).

2.8.1. Water Quality Factors

Some of the factors that provide optimal conditions for microorganisms to multiply include water-stay long period of times in tanks and pipes, adequate nutrient levels, and warm temperatures. In addition, the level of biodegradable organic matter in the distribution system strongly affects bacterial re-growth and harbors opportunistic pathogens. An opportunistic pathogen can be any disease-causing organism, bacterium, virus, helminthes, or protozoan that slips through the treatment processes or enters the distribution system during pressure loss and finds the opportunity or favorable circumstances to lodge or reproduce in organic material, bacterial slime, or other material that it finds attractive. A number of other conditions also can affect water quality. For example, disinfectants may react with organic and inorganic compounds and cause taste and odor problems or form disinfection by-products. Also, particulate re-suspension may cause increased turbidity according to national environmental service center, rural development service, water supply and health, (2011). Diagnostic contamination risk in water distribution systems is a difficult task due to the following:-

- ✓ Water distribution system may comprise (depending on the size of the water utility) thousands of kilometers of pipes of different ages and materials;
- ✓ Operational and environmental conditions, under which these pipes function, may vary significantly depending on the location of the pipes within the system;
- ✓ Since the pipes are not visible, it is relatively difficult and expensive to collect data on their performance and deterioration, and therefore indeed little field data are available;
- \checkmark some factors and processes affecting pipe performance are not completely understood; and
- ✓ It is often difficult to determine or validate an exact cause for water contamination or waterborne disease outbreak because such episodes are often investigated after the occurrence has ended.

For these reasons, high uncertainties are inherent in any risk measure that may be assigned to the distribution system (National research council, 2012). The deterioration of drinking water distribution infrastructure is among the main causes for the loss of quality and quantity of drinking water at the consumers tap, well and spring water. However, since a major portion of the distribution infrastructure is underground, its deterioration does not present the same visual urgency as other visible infrastructure. Since deterioration of the distribution infrastructure adversely impacts the water quality, public water systems cannot justify the costs and efforts of treating water to potable levels and then transmitting them through deteriorating distribution systems (Gundry *et al.*, 2006). The quality of drinking water may be controlled through a combination of: - Protecting of water sources, Controlling of treatment processes, Management of transmission and distribution systems and Handling of water at household level (Gundry *et al.*, 2006).

3. Methods and materials

3.1. Socio-demographic data collection

Structured and pre-tested questionnaires were used to gather pertinent information on sociodemographic characteristics of the study population and their level of awareness about waterborne diseases. In this survey data were collected through village to village survey, by interviewing the head of the householders or members above 15 years old. A Systematic random sampling technique was used to address representative households during data collection (Castillo, 2009).

3.2. Study design and sampling technique

A cross-sectional study design was carried out to assess microbiological and physicochemical quality of drinking water of different sources, including well water, tap water and spring water. A systematic random sampling technique was employed to address representative households to collect data related to the respondent's socio-demographic characteristics and their water handling practices (Annex I). The laboratory investigation was carried from June, 2017 to September, 2017.

3.3. Description of the Study area

3.3.1. Study site and period

The study was conducted in Seka chekorsa town, and it surroundings, Jimma zone, South west Ethiopia between June, 2017 to September, 2017. Seka chekorsa town is located about 372 km south west of Addis Ababa, and 20 km from Jimma town, the Zonal capital .Its location between latitudes $7^{0}35'$ - 8^{0} 00'N, longitude between $36^{0}33'$ - $37^{0}14$ 'E and altitudes between 1,740-2,660 m above sea level. It is the administrative center of Seka Chekorsa Woreda /district (Fig. 1).

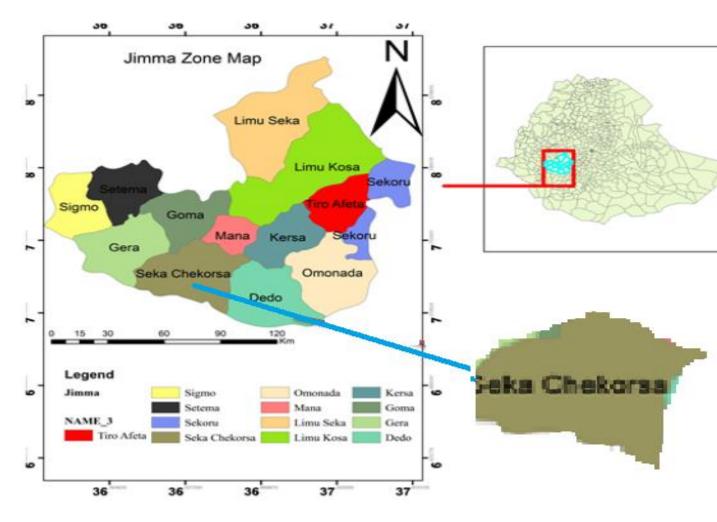


Figure 1. Map of the study area

3.3.2. Study population

Seka chekorsa town and its surrounding had three kebele and 9 large villages with total population of 7,284 of them 3,544 were males and 3,740 were females based on data from the Central Statistical Agency of Ethiopia (CSA, 2006). The town had three spring water sources (Bature, chore and oddo) currently used for drinking. All the three areas were selected for the study. The total populations of these three sites were about 3284 with total numbers of 1420 households. Therefore in this study, 60 households were included in three randomly selected sites of resident population.

3.3.3. Sample size determination

A total of 60 drinking water samples from three different randomly selected sites of seka chekorsa town and its surrounding were collected: 25 samples from well water source, 25 from tap water source and 10 samples from spring water sources.

3.3.4. Water sample collection and processing

The sample collection was done according to Standard Methods for the Examination of Water and Wastewater; (WHO, 2001).Total of 60 water samples was collected from three different water sources including well water (n=25), spring water (n=10) and tap water (n=25).The water sample from three selective location (Bature, chore and oddo), one sampling site was used in each study area; and three types of water sources are used in each study sites. Therefore in two rounds of sampling, triplicate samples of water was collected from each type of water sources in each study area and sampling site and analyzed during June, 2017 to September, 2017. Samples were aseptically collected from each sampling site in sterile glass bottles and transported to laboratory in ice box and analyzed within 6 h of sample collection according to WHO guidelines for sample collection (WHO, 2003). For the chlorinated water samples, about 2.5 ml sodium thiosulphate was added into each sampling bottle to stop the chlorination process during transportation.















d



e

Figure2. Representative water sample collection sources of the study site: spring water (a.b.c) tap water (d) and well water (e)

3.5. Bacteriological analysis

3.5.1. Isolation and enumeration

Ten ml of the water samples were separately transferred into 90 ml sterile peptone water. After thorough mixing and appropriate serial dilutions, 0.1 ml aliquot of each diluted sample was inoculated onto appropriate pre-sterilized and solidified growth medium in duplicates and spread plated on the surface of the solid agar media using a sterile bent glass rod. Inoculated plates were incubated at 37°C for 24 h. Discrete colonies that developed on each plate were counted on plate count agar as total heterotrophic aerobic mesophilic microbes and average values recorded as cfu/ml.

3.5.2. Count of Entrobacteriaceae

From the appropriate serial dilution 0.1 ml aliquots was aseptically inoculated in duplicate on presolidified surface of MacConkey agar (Oxoid) and incubated at 30-35^oc for 18-24hs.Then pink to redpurple colonies were counted as member of family Entrobacteriaceae.

3.5.3. Enumeration of total and faecal coliforms

For determination of total and fecal coliforms, most probable number (MPN) method was employed using multiple fermentation tubes (Oparaocha *et al.*, 2010).

3.5.4. Presumptive test (count of coliform bacteria)

Water sample is aseptically added to each of five tubes containing sterile MacConkey broth. According to WHO(2011), for partially treated and protected drinking water sources,10 ml of sample should be added to 10 ml of (double-strength medium), 1 ml sample to 10 ml and 0.1 ml of sample to 10 ml of single strength medium ,respectively. In this study, the same method was used for the analysis of well, tap and spring water sources. In all test tubes, inverted Durham tubes were inserted and sterilized in autoclave. All the tubes were incubated at 37^oC for 48 hours. The tubes which showed acid and gas production were considered positive for coliforms. From the distribution of these positive tubes, most probable number (MPN) of total coliforms was determined by referring to standard probability table for estimation of total coliforms (National research council, 2012).

3.5.5. Confirmation test (count of fecal coliform bacteria)

Total coliforms were confirmed by transferring a loopful of culture from positive tube from presumptive test in to a tube of Brilliant Green Lactose Bile (BGLB) broth with Durham tubes and incubated at 37° c for 24 to 48 hours. To determine the presence of faecal coliforms the entire tubes positive for total coliforms were sub cultured in to 10 ml of single strength brilliant green bile broth with inverted Durham tubes and incubated at 44.5° C for 24-48 hours. The tubes showing acid and gas productions were considered as positive for faecal coliforms. From the number of these positive tubes, MPN of faecal coliforms were calculated by referring to the table as for total coliforms.

3.5.6. Completed test (confirmation of *E. coli*)

Completed test was carried out by streaking loopful of broth from appositive tube for *E. coli* test on to Eosine Methylene Blue (EMB) agar. The plates were incubated at 37^oC for 24-48 hours. The presence of *Escherichia coli* was confirmed by the formation of metallic sheen color (Adetunde and Glover, 2011).

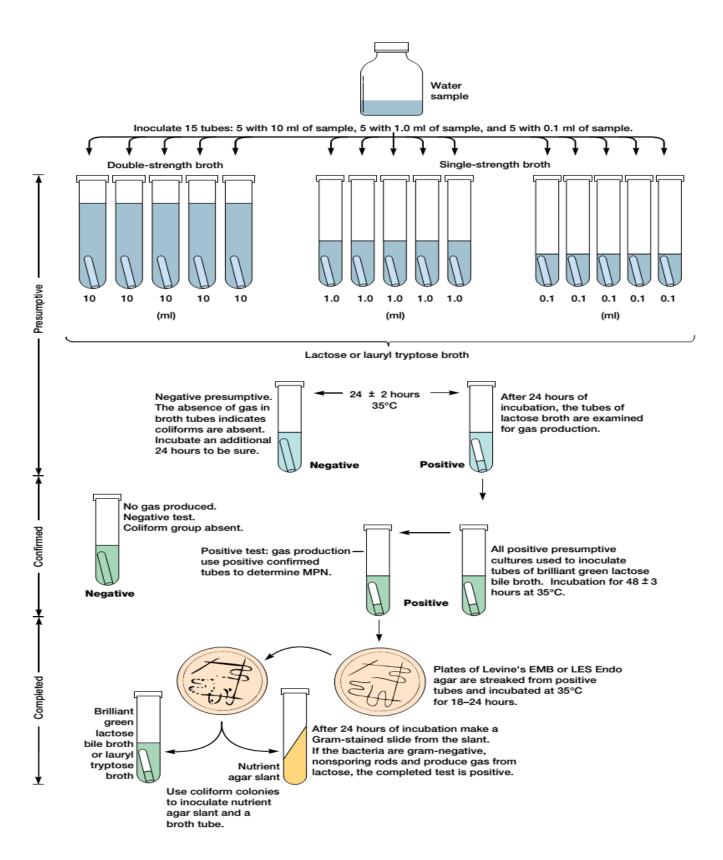


Figure 3. The Most Probable Number (MPN) method Procedure of Water Examination for the Presence of Coliforms by the Presumptive, Confirmed, and Completed Tests.

3.6. Isolation and Characterization

About 10–15 colonies were randomly picked from countable plates of PCA and MacConkey agar and inoculated into 5 ml nutrient broth tubes followed by incubation at 30–35 °C for 24 h. Cultures were purified by repeated plating on nutrient agar and characterized to the genus level using biochemical tests listed below(Zvidzai et al., 2007).

3.6.1. KOH test (test for lipopolysaccharide)

Gram reaction was determined using KOH test (test for lipopolysaccharide), the rapid method recommended by Gregerson (Al-Tomi, (2007) to differentiate whether the organism is gram negative or gram positive. Briefly, about 2mm loop of bacterial growth, obtained from a pure culture was taken and stirred in a circular motion in few drops of the KOH solution. The loop was occasionally raised 1 to 2cm from the surface of the slide. The KOH solutions characteristically become very viscous and mucoid with gram –negative bacteria. A string of the mixture would follow the loop when it was raised. The KOH test was considered positive only when the viscosity occurred within the first few seconds of mixing the bacteria in the KOH solution. Gram positive bacteria suspended in the KOH solution generally do not show such kind of reaction (Al-Tomi, (2007).

3.6.2. Catalase test

Whether the organism produce Catalase enzyme or not was checked by adding few drops of 3 % H_2O_2 on an overnight grown culture plate for production of air bubbles. Formation of bubbles was considered as positive for production of Catalase enzyme (WHO, 2008).

3.6.3. Cytochrome oxidase test

Cytochrome oxidase test was done according to Kovac's (Kovacs, 1956) to identify bacteria that produce Cytochrome c oxidase, an enzyme of the bacterial electron transport chain. (Note: All bacteria that are oxidase positive are aerobic, and can use oxygen as a terminal electron acceptor in respiration. A small pieces of filter paper was soaked in 1% Kovac's oxidase reagent (N, N, N, N, -tetra methyl phenylenediamine di hydrochloride) and let dry. Using a loop a well-isolated colony was picked from a fresh (18 to 24h culture) bacterial plate and rubbed on to treated filter paper. Oxidase positive was confirmed by the formation of dark purple color with 5 to 10 seconds.

3.7. Detection of Salmonella

To test for the presence of *Salmonella*, 1 ml of each sample was aseptically inoculated into 10 ml of lactose broth (LB) and incubated at 37 °C for 24 h for recovery and proliferation of cells. After the pre-enrichment, 1 ml culture was transferred into 10 ml of secondary enrichment broth (selenite cysteine broth) and incubated at 42 °C for 48 h. After the secondary enrichment, 1ml was transferred in to 10 ml of Rappaport-Vassiliadis salmonella enrichment and incubates at 41.5°c for 24h. Loopful of culture from Rappaport-Vassiliadis broth was streaked onto Salmonella–Shigella agar (SSA), Xylose Lysine Deoxycholate agar (XLDA) and modified Brilliant Green agar (BGA) followed by incubation at 37 °C for 18 h. Characteristic colonies are picked, further purified and tested biochemically (WHO , 2001).

3.8. Biochemical Tests for Salmonella and Shigella identification

Suspected non-lactose fermenting bacterial colonies were further characterized having inoculated into the following biochemical tubes: Triple Sugar Iron (TSI) agar, Simon's Citrate agar, Sulfur Indole motility (SIM) medium, Lysine Iron agar, Urea agar, and fermentation tubes of glucose, sucrose and Mannitol. Finally, the proportions of *Salmonella* positive samples were determined based on the above biochemical results (Basheir M. and Elhassan, 2006).

3.8.1. Triple sugar iron agar

Triple sugar iron agar was inoculated with suspected colony of salmonella and Shigella spp. The butt was stab and the slant was streaked and incubated at 37^oC for 24 hrs to detect fermentation of glucose, sucrose and lactose as well as production of H2S. The presence of alkaline (red) slant and acid (yellow) butt, with or without production of H2S was considered as presumptive for Salmonella spp (WHO, 2001).

3.8.2. Simmons citrate agar

Simmons citrate agar was inoculated with suspected colony of salmonella and Shigella spp. The slant was streaked and the tube was incubated at 37 0 C for 24 hrs to determine citrate utilization as a sole source of carbon. The presence of growth and color change from green to blue was considered as presumptive for Salmonella spp (WHO, 2001).

3.8.3. Sulfide Indole motility (SIM) medium

A motility medium was prepared using a test tube. A purified broth culture was taken by a sterile needle and stabbed straight vertically into a test tube containing motility medium to the bottom of the tube and incubated at 37^{0} C for 24 h. A positive motility test was indicated by a red turbid area diffusing away from the line of inoculation and a negative test was indicated by red growth along the inoculation line only but no further (APHA, 2008). The presence of motility was considered as positive for salmonella but non motile were considered as presumptive for Shigella (Al-Tomi, (2007).

3.8.4. Lysine iron agar

Lysine sugar iron agar was inoculated with the suspected colony of salmonella and Shigella spp. The butt was stabbed and the slant was streaked and incubated at 37^{0} C for 24 hrs. Then, the production of an alkaline reaction (purple color) throughout the medium was presumptive for Salmonella spp. But all Shigella do not spp (Basheir M. and Elhassan, 2006).

3.8.5. Urease Agar (Oxoid)

Urease Agar (Oxoid) was inoculated with suspected colony of *Salmonella* and *Shigella* spp. The slant was streaked and the tube was incubated at 37^oC for 24 hrs to assess the hydrolysis of urea. No color change was considered as negative and thus presumptive for Salmonella spp. (WHO, 2004).

3.8.6. Mannitol and glucose/sucrose fermentation

Mannitol, glucose or sucrose fermentation by Salmonella and Shigella was checked by using a fermentation broth base containing ingredients of peptone,10g; NaCl, 5g; phenol red,0.024g; distilled water,1000ml; pH ,7.2. An amount of 10 gram each of Mannitol, glucose or sucrose was separately added to the broth base. Fermentation tubes were contained inverted Durham tubes to detect gas production. To a certain sterility of the media for biochemical test, all tubes with media were pre-incubated at 37 $^{\circ}$ c for 18-24hrs. The broths were inoculated with young culture of suspected salmonella and Shigella and incubated at 37 $^{\circ}$ c for 18-24 hrs. Organisms which fermented glucose and Mannitol and produced gas were considered as positive for salmonella, but in the case of Shigella no Mannitol fermentation and gas production (Ibrahim, 2005).

3.9. Physico-chemical analysis

Physico-chemical parameters including pH, temperature, electric conductivity and dissolved oxygen were measured instruments HQ 40d multi parameter (HQ 40d, HACH Company). Turbidity was measured using Wag tech International Turbidity Meter (Wag-WT3020, Halima PLC Company, Whereas the remaining Total suspended substances (TSS) and total dissolved substances (TDS)and biological oxygen demand determined according to standard method 2540D, standard method 5210C and standard method 2540B, respectively. Nitrate and phosphate concentrations were measured by using reagent kit method (LCK 339 and LCK 349) respectively (WHO, 2011).

3.10. Data analysis

Data was analyzed using SPSS statistical software (version 20). Results of physico-chemical analysis and mean bacterial counts of the investigated water samples were compared with the set standards (WHO guide lines for drinking water quality) and interpreted as acceptable or unacceptable. The significances of differences within samples are determined based on calculated coefficient of variation (% CV). Mean separation between samples categories was computed using one-way ANOVA. The parameters are correlated against each other to determine their relationship using Pearson's correlation. In all cases, significance was considered at 95 % confidence interval (P<0.05).

3.11. Ethical consideration

Sampling of water was carried out with full consent of the head of the households. By respecting their beliefs and culture, the respondent was informed about the objective of the study and their agreement was taken before interaction. Permission was obtained from municipality of the town for public water source samples and consent from private water source owners

4. RESULTS

4.1. Socio-demographic characteristics of the study population

Table 5 Socio-demographic characteristics of the study population in seka chekorsa, 2017

Socio-demograph	ic characteristics	Frequency	Percent (%)
Sex	М	21	35
	F	39	65
Age	15-30	40	66.66
	31-65	20	33.33
Education status	Grade 1-7	21	35
	Grade8-12	19	31.66
	Diploma	8	13.33
	Degree	12	20
Ethnicity	Oromo	45	75
	SNNPE	11	18.33
	Tigrie	4	6.66
Occupation	Civil servant	20	33.33
	Student	21	35
	Merchant	3	5
	Farmer	6	10
Religion	Muslim	27	45
	Orthodox	11	18.33
	Protestant	22	36.66

A total of 60 respondents were included in the current study. Among them, 21 (35%) were male and 39 (65%) were females. Most of them 40 (66.6%) were found within age group ranging between 15 and 30 and about 45 (75%) of were Oromo by ethnicity. Majority of them were followers of Muslim religion.

were obtained before water sample collection.

4.2. Awareness on waterborne diseases and hygienic practices by the local community of the study area

Table 6. Knowledge of the respondents on water borne diseases and the care being taken, seka chekorsa town and its surroundings, 2017

Characteristics		No of respondent (N=60)	Percent (%)
Source of drinking	Well water	11	18.33
Water	Tap water	11	18.33
	Spring water	38	63.33
Quality of water	Very good	12	20
	Good	16	26.66
	Satisfactory	32	53.33
Problem of drinking water accessibility in the area	Yes	38	63
in the area	No	22	37
Types of container used to collect water	Bucket	9	15
	Plastic spot	49	81.66
	Clay spot	2	3.33
Method of purification of water before use	Directly	56	93.33
use	Boiling	2	3.33
	Using Agar	2	3.33
Waste material disposal method	Bury in pit	11	18.33
	Open field	34	56.6
	Burning	13	21.6
Distance of water	5-10	5	8.33
source from latrine (in meter)	10-15	7	11.66
	15-20	13	21.66
	>20	35	58.33

Based on the information obtained from the respondents, majority 38 (63.33%) of the population are using spring water as main source of drinking water while about a quarter of them 11 (18.33%, each) were relying on wells and tap water (Table 6). Only12 (20%) and 16 (26.66%) of the respondents think that their drinking water source is very good and good, respectively, whereas the majority (53.33%) rated it as satisfactory. Although most of the people get their drinking water from spring and wells, only few of them (3.33%, each) were responded that they use their water after boiling and using disinfection and the majority (93.33%) use it directly without any further treatment. Plastic pots are the most favored (81.66%) material for water storage, making the heat treatment of the storage facilities unlikely. About 65% of the water sources were found at a distance of more than 20m from latrine (58.3%) and (42.66%) of them were located in lower elevation with respect to the nearby toilet rooms. Waste management practices of the localities was found poor as more than 56.6% of the respondents dispose waste materials on open field.

4.3. Bacteriological analysis of water source

Table 7. Mean bacterial counts (CFU/ml or CFU/100 ml) of drinking water samples of different sources (N = 60), Seka Cchekorsa town and its surroundings, 2017

Microbial groups	Parameter	Water Sample so	irces		
		Well water	Tap water	Spring water	P-value
	Mean	157	140.8	122	0.78
Total heterotrophic bacteria	SD	45.7	43.70	37.11	
	%CV	29.10	31.03	30.41	
	Max	245	224	184	
	Min	88	74	60	
Entrobacteriaceae	Mean	14.44	15	9.2	0.39
	SD	5.45	5.66	2.89	
	%CV	37.74	37.73	31.41	
	Max	27	32	14	_
	Min	8	7	4	_
	Mean	49.36	31.96	32.1	0.70
Total coliform	SD	29.73	20.29	17.722	
(CFU/100 ml)	%CV	60.23	63.48	55.20	—
	Max	94	79	79	_
	Min	17	11	17	_
	Mean	4.08	3.4	5.3	0.76
Fecal coliform	SD	4.310	3.674	3.433	
(CFU/100 ml)	%CV	105.63	107.94	64.77	
	Max	17	11	9	
	Min	Below detectable level	Below detectable level	Below detectable level	

SD=standard divisionCV= coefficient of varianceMax=maximumMin = minimumFC= fecal coliformTC= total coliformCFU= colony form unit

Bacteriological analysis of the water samples indicated that all well water, tap water and spring water samples were found contaminated with heterotrophic bacteria, members of Enterobacteriaceae, and Total coliforms. The mean heterotrophic bacterial count (CFU/ml) of well water, tap water and springs were 157 ± 45.7 , 140.8 ± 43.704 and 122 ± 37.118 , respectively. Likewise mean counts (CFU/ml) of Enterobacteriaceae were 14.44 ± 5.45 , 15 ± 5.66 and 9.2 ± 2.898 in the same order. The highest total coliform count (CFU/100ml) was recorded for well water (49.36±29.73) and the highest mean fecal coliform count (CFU/100ml) was encountered in spring water samples (5.3 ± 3.43) (Table xx). About 66.67 % of tap water samples were found to be negative for FC and *E.coli* were not detected in all the tap water samples. The entire samples from both wells and springs were positive for indicator organisms. Among the 25 well water samples analyzed, only 12(20 %) had bacterial count below 10 CFU/100 ml and 10 (16.67 %) were negative for fecal coliforms. Result of analysis of mean separation (both One-Way ANOVA and Pearson Correlation) in counts of the four microbial groups along the three water types showed no statistically significant difference (P>0.05) (Table 8). Analysis for degree of variability in counts of different microbial groups within all well water, tap water and spring water revealed that the mean counts of heterotrophic bacteria are significantly different from that of entrobacteriaceae, total coliform and fecal coliforms (P=0.00). However, there were no significant differences among mean counts of entrobacteriaceae, total coliform and fecal coliforms (P>0.05).

4.4. Comparison of Microbial load between water sources in different sites

Table 8 Comparison of mean bacterial counts (CFU/ml or CFU/100 ml) of well, Tap and Spring water source, Seka Chekorsa, 2016/17

Microbial			ple sourc	e source						
Group	param	Well water			Tap water			Spring water		
Group	eter	Bature	Chore	Oddo	Bature	Chor	Oddo	Bature	Chore	Oddo
		site	site	site	site	e site	Site	site	site	site
Total	Mean	155.2	166.12	150.44	127.33	87.66	146.11	129.2	87.66	143.75
heterotrophic	SD	43.29	46.99	50.675	46.151	33.78	50.690	13.79	25.42	41.684
bacterial	%CV	27.88	28.29	33.68	36.24	38.53	34.69	10.67	28.99	28.99
(CFU/ml)	Max	245	222	224	143	110	184	224	207	74
	Min	115	102	88	117	60	88	88	100	222
Entrobacteria	Mean	15.37	15.75	12.44	7.333	9.666	10.25	17.37	15.62	12.333
ceae	SD	5.806	6.713	3.609	7.501	4.373	4.031	3.055	2.081	3.304
(CFU/ml)	%CV	37.77	42.62	29.01	102.29	45.24	39.32	17.58	13.31	26.78
	Max	23	27	19	10	12	14	32	24	18
	Min	9	8	8	4	8	6	9	11	7
	Mean	42.25	40.666	33.75	28.375	33.72	32	27.33	31	36.5
TC	SD	28.23	22.107	19.174	21.639	22.27	22.360	6.110	3.464	29.320
(CFU/ml)	%CV	65.19	89.48	92.98	83.50	96.40	118.72	42.20	61.39	41.36
	Max	94	79	79	79	94	79	34	33	33
	Min	17	22	17	14	14	11	22	27	17
	Mean	3.25	3.777	5.25	4.125	33.72	2.375	4	0.5	4
FC	SD	5.922	3.113	3.882	5.536	5.802	3.502	3.559	1	3.605
(CFU/ml)	%CV	182.21	82.41	73.94	134.20	17.20	147.45	88.97	200	90.125
	Max	17	9	11	9	11	9	7	2	7
	Min	Below	Below	Below	Below	Below	Below	Below	Below	Below
		detectable	detectabl	detectable	detectable	detect	detectabl	detecta	detecta	detectable
		level	e level	level	level	able	e level	ble	ble	level
						level		level	level	

SD=standard division CV= coefficient of variance Max=maximum Min = minimum

FC= fecal coliform

TC= total coliform CFU= colony form unit

The highest mean heterotrophic bacterial count (CFU/ml) of 166.12 ± 46.99 was recorded from well water of chore site and the lowest mean heterotrophic bacterial count (CFU/ml) of 87.66 ± 33.780 was observed in tap and spring water of chore site. Well water sources had overall mean TC and FC counts of 42.25 and 3.25 CFU/100 ml, 40.66 and 3.77 CFU/100 ml, 33.75 and 3.882 CFU/100 ml, at Bature, chore and oddo sites, respectively. Whereas, the three sites revealed an overall mean TC and FC counts of, respectively, (28.375 and 4.125 CFU/100 ml), (33.727 and 33.727 CFU/100 ml) and (32 and 2.375 CFU/100 ml) from tap water and (27.33 and 4 CFU/100 ml), (31 and 7 CFU/100 ml), (36.5 and 4 CFU/100 ml) from spring water samples (Table 8). There were significant differences in the mean counts of heterotrophic bacteria and other microbial groups within and among water types (P=0.00).

Isolation and Characterization

The dominant microbes isolated from the water samples were characterized to at least group/genus levels using different biochemical tests. Accordingly, the isolates were found dominated by Enterobacteriaceae (34 %), *Bacillus* (26.4 %) and *Pseudomonas* (17 %), followed by *Micrococcus* (6.9 %) and *Staphylococcus* (6.0 %). Unidentified Gram negative cocci (4.7 %) and Gram positive rods (5 %) were among the least encountered in the water samples (Table 10).

Water	Dominant bacterial groups and their frequencies of isolation							
samples	Bacillus	Entrobacteri	Pseudomo	Staphyloc	Microco	Other	Other	
	spp (%)	aceae (%)	nas (%)	occus (%)	ccus (%)	Gram +ve	Gram –ve	
						rods	cocci	
Well	12.2	14	7	4	2.6	2	1.3	
Тар	6	9	4	-	1.	2	1	
Spring	8.2	11	6	2	3.3	1	2.4	
Total	26.4	34	17	6	6.9	5	4.7	

Table 9 Frequency of isolation of bacterial groups from different water sources, Seka Chekorsa, 2017

4.5.1. Prevalence of salmonella and Shigella spp.

Table10. The Prevalence of Salmonella	and Shigella in differ	ent water samples, Seka	chekorsa, 2017

water source	No of samples	Salmonella positive		Shigella positive		
		No	%	No	%	
Well water	25	1	1.66	0	0	
Tap water	25	0	0	0	0	
Spring water	10	1	1.66	0	0	
Total	60	2	3.33	0	0	

From a total of 60 water samples examined, only 2 samples (one from well and the other from springs) were found positive for *Salmonella* spp., but all samples were negative for *Shigella*. Thus, despite high counts of Entrobacteriaceae and coliforms in some of the water samples, both *Salmonella* and *Shigella species* were less prevalent (Table 10).

4.6. Physico-chemical analysis of drinking water sources

Summarized below are the average values of the Physico-chemical parameters including, Temp, pH, electric conductivity, dissolved oxygen, turbidity, TSS, TDS, concentrations of phosphate and Nitrate, and biological oxygen demand of drinking water samples were given in the following table (Table 12):

Sample source		Temp	pН	EC	DO	Turbidity	TSS	TDS	PO ₄ -	NO ₃ ⁻	BOD
		(⁰ C)		(µs/cm)	(mg/L)	(NTU)	(mg/L)	(mg/L)	3(mg/L)	(mg/L)	(mg/L)
Well water	Mean	22.5	6.78	63.68	2.31	14.38	15.44	232.4	0.106	0.535	3.60
water	SD	1.15	0.35	24.53	0.79	13.62	2.916	40.05	0.191	0.496	1.40
	%CV	5.09	7.10	38.52	34.19	94.71	18.84	12.44	180.18	92.71	36.32
	Max	24.4	7.75	121.3	3.95	55.08	21	331	1.004	1.898	6.8
	Min	21	5.85	43.1	1.01	5.21	8	163	0.034	0.145	2
	P-value	0.05	0.06	0.05	0.28	0.45	0.06	0.09	0.33	0.09	0.05
Тар	Mean	22.3	7.16	320.8	5.15	5.524	47.4	175.4	0.518	5.933	4.50
water	SD	0.75	0.51	106.2	0.788	1.660	19.06	17.17	0.293	4.478	1.24
	%CV	3.35	5.02	33.10	15.14	30.07	40.21	9.78	56.56	75.47	30.11
	Max	24.1	8.56	407	6.15	7.64	68	220	0.963	16.01	7
	Min	21.4	6.75	107.5	3.68	2.15	9	146	0.054	3.05	3
	P-value	0.05	0.25	0.06	0.06	0.25	0.27	0.05	0.37	0.09	0.07
Spring	Mean	21.96	6.80	144.4	9.22	2.279	11.2	159.2	0.071	14.764	4.52
water	SD	0.64	0.45	6.528	1.36	0.539	3.190	13.55	0.009	1.545	1.525
	%CV	2.94	6.61	4.51	14.75	23.34	28.48	8.51	16.66	10,46	27.67
	Max	13.4	6.78	122.8	11	3.02	16	177	0.085	17.015	7.1
	Min	20.2	6.20	105.1	6.97	1.53	8	136	0.058	12.168	2.3
	P-value	0.37	0.08	0.21	0.24	0.05	0.26	0.15	0.23	0.09	0.05

Table 11 Physico-chemical analysis of drinking water sources, seka chekorsa town and its surroundings, 2017 (N=60)

SD= standard division CV= coefficient of variance Max=maximum Min = minimum EC= electron conductivity DO= dissolved oxygen TSS= total suspended solids TDS= total dissolved solids BOD= biological oxygen demand

4.6.1. Temperature

The recorded mean temperature of the water samples were $22.55 \pm 1.15^{\circ}$ c, $22.35 \pm 0.75^{\circ}$ c and $21.96 \pm 0.65 \,^{\circ}$ C for wells, tap and spring s, respectively (Table 11). Of the total water samples (n = 60). The maximum temperature (24.4 $^{\circ}$ C) was recorded for well water and the minimum (20.2 $^{\circ}$ C) for springs. There were no observable significant variations both within the samples (CV = 4.52–9.24 %) and among water samples collected from the three different sources (P = 0.05, 0.05 and 0.37) recorded in well, tap and spring respectively.

4.6.2. Hydrogen ion concentration (pH)

The mean pH of wells, taps and springs were 7.18 ± 0.51 , 6.96 ± 0.35 and 6.80 ± 0.45 , respectively. Well water samples had mean pH value around neutrality (pH = 7.18) ranging between 7.4 and 8.14. There was no significant differences in pH within and between water samples collected from different sources despite variation in temperature differences within same sample source (CV < 10 %, P=0.06, 0.25 and 0.08), respectively (Table 11)

4.6.3. Electric conductivity

Mean electric conductivity (μ s/cm) for well water, tap water and springs were 63.68 ± 24.53, 320.8 ± 106.2 and 144.4 ± 6.52, respectively (Table 11). There was no statistically significant difference (P = 0.05, 0.06 and 0.21) among mean electric conductivities of different water samples but there was significant difference observed within samples (CV >10% (except for spring water).

4.6.4. Dissolved oxygen (DO)

The mean values of dissolved oxygen (DO) (mg/l) for wells, taps, and springs were 5.48 ± 0.76 , 5.15 ± 0.78 and 3.92 ± 0.15 , respectively (Table 11). There was no significant differences (P = 0.28, 0.06 and 0.24) in DO among the assessed water samples.

4.6.5. Turbidity

The mean turbidity value of water samples was the highest (10.66 NTU) for wells and the least (2.27 NTU) for spring water. The mean turbidity observed in well and tap water sources are above the

maximum permissible level recommended by WHO standards (5 NTU). Variations were statistically significant within samples (CV> 10 %) (Table11).

4.6.6. Phosphate

Mean phosphate concentration (mg/L) level recorded for well water ,tap water and springs were 0.106 \pm 0.191, 0.518 \pm 0.293 and 0.071 \pm 0.009, respectively (Table 11). Phosphate concentration values of 0.07 \pm 0.009 mg/l was measured as the lowest mean value from spring and 0.518 \pm 0.293 mg/l was the highest from tap sources. The values did not show significant variations (p= 0.33, 0.37 and 0.23) among water samples although highly variable within samples (CV > 10 %).

4.6.7. Nitrate

Mean nitrate concentration (mg/l) values of 0.535 ± 0.496 , 5.933 ± 4.478 and 14.764 ± 1.545 were recorded, respectively, for well water, taps and springs (Table 11). The maximum mean nitrate value of 14.764 ± 1.545 mg/l was recorded from spring and the minimum from wells 0.535 ± 0.496 . Variations were not statistically significant among means of different water samples (P = 0.09, 0.09 and 0.09).

4.6.8 Total suspended solids (TSS)

The mean TSS (mg/l) of wells, taps and springs were 15.44 ± 2.91 , 47.4 ± 19.06 and 11.2 ± 3.19 , respectively (Table 11), the highest mean concentration being in tap and the least in spring water. Statistically significant variations were not observed among mean values s of different water sampling sources (P = 0.06, 0.27 and 0.26) but within all samples of the same source.

4.6.9. Total dissolved solids (TDS)

High TDS level (175.44 \pm 17.17) was observed in tap water samples while relatively lowest level (148.4 \pm 18.47) was encountered in well water. Variation in TDS within samples was not significant (% CV < 10). Significant variations were noted among the different water sample sources (P = 0.09, 0.05 and 0.15) and variation within sample was not significant for tap water.

4.6.10. Biological oxygen demand (BOD)

The observed BOD value (mg/l) was the highest in tap water (2.59 ± 0.78) followed by well water and the lowest mean BOD value recorded from spring water samples (1.12 ± 0.31) (Table 11). There were statistically significant variations in BOD values among different water samples collected from the three sources (P =0.05, 0.07 and 0.05), respectively.

5. Ddiscussion

Access to safe potable water is one of the major challenges to humanity, especially in developing countries. Majority of population, therefore, are reinforced to rely on all available options to secure the daily demands for water. However, not all water sources are microbiologically and chemically safe for consumption. In the current study, The high mean heterotrophic bacterial count (157 CFU/ml) observed in well water and the lowest mean count (122 CFU/ml) recorded from spring water samples documented in this study, with about 70 % of the water samples having aerobic mesophilic bacteria counts, is in agreement with the earlier report from Nigeria (Shamsuddeen *et al.*, 2010). Although the observed contamination level with regards to aerobic mesophilic bacteria was not significantly high, their very detection by itself is an indication of high vulnerability of the water sources to microbial contamination, including potential pathogens.

The predominant bacterial groups identified in the water samples were members of the family Entrobacteriaceae, *Pseudomonas* spp. and *Bacillus* spp. Similarly, other scholars (Zvidzai et al., 2011), reported that the most prevalent bacterial species in well water sources from Rural Areas of Zimbabwe were members Gram negative, non-spore forming bacilli belonging the family entrobacteriaceae. Bacillus species were the second dominant bacterial groups in the current study. Few of the Bacillus species, including strains of Bacillus cereus, are pathogenic to humans and animals being responsible for food poisoning (A bed and Alwakeel, 2007). The incidence of *Pseudomonas* spp. as the third dominant bacteria in the current study was in agreement with report made elsewhere (Geldreich, 1996) With 100 and 80 % detection rates of TC and thermo tolerant coliforms, respectively, about 76.67 % of the samples had TC bacterial count beyond the Canadian acceptable level for drinking water (10 CFU/100 ml) (Anon, 2002) with all water samples having microbial counts above WHO recommendation (0 CFU/100 ml) (Chan et al., 2007). According to WHO guidelines, E. coli or thermo-tolerant coliform bacteria should not be detectable in any water intended for drinking(WHO,1997). Results of this study were in agreement with the reported detection of coliforms from 75 % of unprotected well and spring samples from North-Gondar, Ethiopia (Mengesha et al., 2004) and the 90 % detection of the same microbial groups from protected spring samples of Uganda (Haruna et al., 2005) Similarly, 87.5 % of the water samples collected from other six protected wells and eighteen unprotected wells of Serbo town (Mohammed, 2012) revealed TC count above the permissible limits for drinking water. About 80 % of the water samples were positive

for fecal coliforms (FC) and the highest observed mean coliform count was 266.67 CFU/100 ml. In contrary to our report, significantly high counts (1100 CFU/100 ml) of FC bacteria were reported from water samples collected from rural areas of Iran (Ghaderpoori *et al.*, 2009) and unprotected springs of central High lands of Ethiopia (741.7 CFU/100 ml) (Birhanu , 2008).

The prevalence of *Salmonella* was very low in the current study, with only one positive sample from wells and one from spring water samples. In a related study, (Shittu *et al.*, 2008) reported absence of *Salmonella* and *Shigella* in all well water samples examined in Nigeria although stream samples were positive. However, as long as the counts of fecal coliforms are high in most of the water samples examined for microbial load and safety, the absence of any *Salmonella* and *Shigella* in many of the samples could not qualify the water sources' safety.

Temperature is one of the Physico-chemical parameters used to evaluate quality of potable water. It affects many phenomena including the rate of chemical reactions in the water body, reduction in solubility of gases and amplifications of tastes and colors of water (Olajire and Imeokparia, 2001). The highest (24.4 °C) and lowest (20.2 °C) temperature recorded from tap water and spring, respectively, were related to the 28 °C reported from different water source of Nigeria (Oparaocha *et al.*, 2010) but higher than the study conducted in Bahir Dar town (15–20 °C) (Milkiyas *et al.*, 2011). Almost all the recorded water temperatures were above the WHO recommended level (<15 °C) and within temperature optima of some aerobic mesophilic bacteria and fungi. The variations in temperature of the samples may be attributed to sampling locations as some of the water sources were collected from underground (including well water) while others were found partly on the surface exposed to direct sunlight. Richness in organic matter, hence microbial activities, could also contribute besides the geographic location of the study area (tropical zone). It is desirable to have the temperature of drinking water not exceeding 15 °C as the palatability of water is enhanced by its coolness (WHO, 2003).

With an overall mean pH value of 6.98 that ranged between (5.85 and 8.56), about 66 % of pH of water samples fall within WHO standard (6.5–8.5) (WHO, 1996). Except tap water, the majority of other drinking water sources were slightly acidic (below pH of 7), whereas tap water sources had pH value greater than 7 (slightly alkaline). The pH values in most of the samples were found within the recommended standards of European Commission and WHO (ranges from 6.5 to 8.5) for potable

waters. According to (Byamukama et al., 1999) the low pH values observed in most wells and springs could be associated with carbon dioxide saturation in the groundwater. In fact, the Physico-chemical nature of the soil of sampling sites could partly contribute to the final pH of the samples. In related development, the pH of water samples collected and analyzed from Katanga, North of Kampala city, were found to be acidic (Nshekanabo and Wozei ,1997) contributing to the final low pH of water samples analyzed from the same sites.

In this study, about 60 % of the samples had turbidity level above 5 NTU (beyond the acceptable standard) although all well water and 70 % of the spring had values below 5 NTU. High turbidity is often associated with higher levels of suspended organic matter and microorganisms including bacteria and other parasites. Usually, the acceptable turbidity level is 5 NTU although it could vary with local circumstances (WHO, 1997). The consumption of highly turbid water may constitute a health risk as excessive turbidity can protect pathogenic microorganisms from the effect of disinfectants, and also stimulate the growth of bacteria (Zvikomborero, 2005).

The highest conductivity recorded from tap water sources could be due to the corrosion of metals that led to the accumulation of heavy metals. Even though conductivity values in the water samples ranged from 43.1 to 407 μ s/cm , more than 93.33 % of the samples had electric conductivity (EC) value below 399 μ s/cm, with the lowest conductivity values recorded from well and springs. Actually all mean EC values were within WHO maximum recommended limit (1500 mg/l). Related results were reported from well water samples in Nigeria (Oparaocha et al., 2010), where the EC levels ranged from 22 to 315 μ s/cm. To the contrary, EC values greater than our finding was reported from ground water sources of Turkey, where the lowest and highest conductivities were 463 and 1460 μ s/cm, respectively, (Aydin, 2006).

The lowest total dissolved solid (TDS) (136 mg/l), recorded from spring and the highest value (331 mg/l) recorded from well water were below the maximum allowable limit (1000 mg/l) recommended by WHO, (2001). Total dissolved solid (TDS) are measures of the general nature of water quality (Shittu *et al.*, 2008). The TDS include carbonate, bicarbonate, chloride, sulphate, phosphate, nitrate, calcium, magnesium, sodium, organic ions and other ions. TDS affect the taste of drinking water if present at levels below the WHO recommended level. Accordingly, the TDS values recorded in this study could be considered tolerable.

On the other hands, the overall mean total soluble substances (TSS) recorded in the study ranged between 11.2 and 47.4mg/l with the lowest and highest measurements being observed in spring water and well water samples. The variability or range in the recoded TSS data was significantly high as compared to the earlier report (10–32.4 mg/l) made from Southern Rajasthan, India from hand pump water sources (Sharma et al., 2008) and the 210.0 \pm 127.7 mg/l from untreated tap water of Jimma town, Ethiopia (Israel ,2007). Although there is no set guideline for the maximum permissible limit of TSS in drinking water, the TSS value recommended for fisheries and aquatic life in Ethiopia (25 mg/l) could be used as reference for this purpose (Israel ,2007). Accordingly, the concentrations of TSS obtained from all wells, most of tap (85.2 %) and spring (80.0 %) water sources were above even the tolerable limits for maintenance of aquatic life and fisheries. The higher concentration of TSS in the water samples could be due to poor sanitation practice with possibility of contamination of the water sources with municipal wastes and plant debris.

The different water samples revealed mean dissolved oxygen (DO) values ranging between 3.93 and 5.49 mg/l although there were significant variations both within and among samples. About 93.3 % of the samples had mean DO ranging between 3.68 and 6.21 mg/l. As compared to the WHO acceptable standards for dissolved oxygen in fresh water (10–12 mg/l), the observed results were partly acceptable although significant number of individual records fall out of the range. Related observation was reported by Tenagne (2009) from drinking water in Bahir Dar, Ethiopia, in which the mean DO concentration of the water samples were between 0.45 and 5.27 mg/l. (Purushottam *et al.*2010), also reported DO values ranging from 1.2 to 4.6 mg/l from different lake water samples. Dissolved oxygen with only lesser oxygen dissolved in warm water than cold water (Tenagne, 2009). Therefore, high temperature of the water sources could be one of the factors for low DO values recorded in the current study.

The mean BOD after 5 days (BOD5) was found within the range of 2–36mg/l. Although no guideline set for the maximum tolerable limit of BOD in drinking water, for fisheries and aquatic life, European Union and Ethiopia recommend 3–6 mg/l and less than 5 mg/l, respectively (Israel, 2007). This suggests that drinking water sources were highly polluted by organic matter. In a related development, detection of phosphate in water sources (0.09–1.04 mg/l) usually indicates contamination of the water sources by run-off from agricultural farms using inorganic fertilizers (Taha and Younis, 2009).

Related result (0.27–1.41 mg/l) was also recorded from underground water samples from Ondo State, in the western part of Nigeria (Ololade *et al.*, 2009). All the water samples assessed in this study were observed to have concentration of phosphate ions below the maximum permissible level (5 mg/l) set by European commission and WHO. The high phosphate concentrations in some of the water samples could be due to the presence of agricultural activities near the water sources, as most of the people in the study area were practicing farming. These observations indicate that the water from these sources could not be stored for long in open containers, as the presence of phosphate encourages the growth of algae and consequently cause adverse changes at least in color and taste of the water sources (Agunwamba, 2000).

The highest mean nitrate concentration in the samples 14.67mg/l were recorded in the current study. Accordingly, most of the water samples fall within the permissible limit (50 mg/l) set by the European commission for drinkable water except for two of the wells with concentration above 50 mg/l. Study done on the quality of packaged water analyzed in Nigeria reported concentrations of 0.0-40.0 mg/l nitrate ions (Ajayi et al., 2008), while analysis on well water samples from the same country revealed nitrate concentration of about 50.6 mg/l. Higher nitrate levels (>50 mg/l) were also previously reported (Eed Lafi, 2009., Riemann et al., 2003). These reports have conformity with the present findings. Similar observations have been reported from groundwater sources in Iganga, eastern Uganda, with nitrate levels ranged between 21 and 145 mg/l in protected springs. In another study done in Tanzania, nitrate levels ranging between 0 and 90.28 mg/l was recorded from different drinking water sources (Napacho and Manyele, 2010). However, lower nitrate concentration was also reported from northeastern region of Buenos Aries Province, Argentina (Galindo et al., 2007). This variation may be explained by the differences in hydro-geological regimes and likely contaminant entry point. While nitrogen is a vital nutrient for plant growth, high concentrations are harmful to people and nature. The agricultural use of nitrates in organic and chemical fertilizers has been a major source of water pollution in Europe (Ajayi et al., 2008). Generally, farming remains responsible for over 50 % of the total nitrogen discharge into surface waters. Thus, excessive nitrate concentrations in water are mainly related to pollution (with agriculture as the main source). Lifetime exposure to nitrite and nitrate at levels above the maximum acceptable concentration could cause such problems as diuresis, increased starch deposits and hemorrhaging of the spleen (Omoigberale and Ogbeibu, 2005) because of their high toxicity to humans and aquatic life.

6. Conclusion

Bacteriological quality of most water samples analyzed in the current study did not meet the standards set for drinking water. From the quality and sanitary risk evaluation points of view, the studied water sources could be classified as grossly polluted and only very few of them had reasonable quality. Most of the Physico-chemical data indicated marginally tolerable quality with respect to pH, EC and TDS but poor quality in relation to turbidity, temperature, conductivity, and BOD and nitrate concentration with values much in excess of the permissible standards. Excessive nitrate concentrations recoded from some water samples are mainly related to pollution (with agriculture as the main source). Lifetime exposure to nitrite and nitrate at levels above the maximum acceptable concentration could cause many health problems including increased starch deposits and hemorrhaging of the spleen.

Recommendations

- As indicator bacterial counts in most sampled water sites have exceeded the guidelines set for drinking water, an urgent measure should be taken to develop safe water supplies and basic sanitation in the area.
- Protection of water sources accompanied by sanitation and hygiene promotion programs can improve the hygiene quality of rural water sources, where disinfection is not feasible.
- Thus, with the current high dependence on alternative water sources other than tap water, it calls for awareness development on hygienic handling of wells and springs besides designing protections and regular purification strategies by the concerned bodies.
- Regular disinfection of water with chlorine, maintenance of water distribution system, proper sanitation and drainage network system in the town is also very important.
- This study is limited to few Physico-chemical and bacteriological parameters and sampling frequency. Therefore, year round sampling (to know seasonal variation) and analysis of additional water quality parameters better undertaken.

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Annex I.

Interview questions on the assessment of bacteriological quality of drinking water in Seka Chekorsa town and its surroundings, Jimma Zone, south west Ethiopia.

Purpose of the interview

This study is aimed to assess the Physico-chemical and bacteriological quality of drinking water in Seka Chekorsa town and its surroundings. Thus, you are kindly requested to respond to the following questions. Please fill the space provided with appropriate information. Put "x" mark on your choice inside the box

Keble----- House No-----

Part I Socio-demographic survey

C	DDE	-							
1.	Sex	Male	Femal						
2.	Age	15-30	31-65	Above 65					
3.	education status	grade 1-7	grade 8-12	degree	Diploma				
4.	Ethnicity	oromo	amhara	SNNE SNNE	Tigire				
5.	Occupation	civil servant	merchant	student	Farmer				
6.	Religion	Muslim	Orthodoox	protestan	t others				
Part I	Part II Water handling practice (collection and transport)								
1. Wha	t kind of water you	use for drinking?	Tap sp	oring well	others (specify)				
2. Wha	t is the distance of	your water source fro	om latrine (in meter)?	•					
	5	-10 10-15	□ ₁₅₋₂₀ □	□ _{>20}					

3. Is there any contact between drinking water pipes	with solid or liquid	wastes? Yes	No No
4. Are any tap shared with other house hold(s)?	No No	Yes	

5. Is the water fetched from spring and tap transported to your house with covered containers?
Yes No
6. In your house, is the water for drinking is stored in a separate container from water intended for other
purposes? Yes No
7. Do you know there are water born agents that cause health problem?
Yes No
8. Do you have drinking water accessibility problem in your area?
Yes No
9. Do you think that the water you fetch from your spring or tap is safe?
Yes No
10. Do you have experience of boiling of water before using it for drink purpose?
yes No
11. Do you think that water can be polluted in the distribution system?
12. What type of container do you use to collect water from water sources?
Plastic spot clay spot Bucket others(specify)
13. What is your opinion on the quality of water (color and odor) being used in your locality?
very good good satisfactory poor
14. How do you dispose waste material?
15. Did you face any water borne diseases in recent years in your home? Yes No 16. If yes to Q 15,, what type of appropriate measures you think to resolve it?
17. If yes to Q 15, in what frequency did you take action to solve the problem?

Data collector's name ______ signature _____ date _____

Annex 2: Output of Statistical analysis

Mean separation of water samples (well, tap and spring, in general)

ANOVA								
	Sum of Squares	Df	Mean Square	F	Sig.			
Heto Between Groups	1077.403	2	538.701	.241	.788			
Within Groups	49128.597	22	2233.118					
Total	50206.000	24						
Entro Between Groups	58.518	2	29.259	.982	.391			
Within Groups	655.722	22	29.806					
Total	714.240	24						
TCC Between Groups	395.585	2	197.793	.363	.700			
Within Groups	11997.375	22	545.335					
Total	12392.960	24						
FCC Between Groups	8.236	2	4.118	.271	.765			
Within Groups	333.764	22	15.171	u l				
Total	342.000	24						

Descriptive

	-					95% Confidence Interval for			
						Mean			
				Std.			Upper		
		Ν	Mean	Deviation	Std. Error	Lower Bound	Bound	Minimum	Maximum
Hetro	1	8	1.5525E2	43.29880	15.30844	119.0513	191.4487	115.00	245.00
	2	8	1.6612E2	46.99677	16.61587	126.8347	205.4153	102.00	222.00
	3	9	1.5044E2	50.67571	16.89190	111.4916	189.3972	88.00	224.00
	Total	25	1.5700E2	45.73748	9.14750	138.1205	175.8795	88.00	245.00
Entro	1	8	15.5000	5.80640	2.05287	10.6457	20.3543	9.00	23.00
	2	8	15.7500	6.71353	2.37359	10.1374	21.3626	8.00	27.00
	3	9	12.4444	3.60940	1.20313	9.6700	15.2189	8.00	19.00
	Total	25	14.4800	5.45527	1.09105	12.2282	16.7318	8.00	27.00
тсс	1	8	42.2500	28.23245	9.98168	18.6471	65.8529	17.00	94.00
	2	8	41.6250	23.43342	8.28496	22.0342	61.2158	22.00	79.00
	3	9	33.6667	17.93739	5.97913	19.8788	47.4546	17.00	79.00
	Total	25	38.9600	22.72385	4.54477	29.5801	48.3399	17.00	94.00
FCC	1	8	2.8750	4.15546	1.46918	5991	6.3491	.00	9.00
	2	8	3.5000	4.17475	1.47600	.0098	6.9902	.00	11.00
	3	9	2.1111	3.37062	1.12354	4798	4.7020	.00	9.00
	Total	25	2.8000	3.77492	.75498	1.2418	4.3582	.00	11.00

Multiple Comparisons

sVAR00006

LSD

(I)	(J)VAR	Mean Difference			95% Confidence Interval			
VAR00007	00007	(I-J)	Std. Error	Sig.	Lower Bound	Upper Bound		
1hetero	2entro	141.80556 [*]	12.49685	.000	116.2069	167.4042		
	3TCC	109.39286 [*]	13.31049	.000	82.1276	136.6582		
	4FCC	152.00000*	12.85915	.000	125.6592	178.3408		
2	1	-141.80556 [*]	12.49685	.000	-167.4042	-116.2069		
	3	-32.41270 [*]	12.96081	.019	-58.9617	-5.8637		
	4	10.19444	12.49685	.422	-15.4042	35.7931		
3	1	-109.39286 [*]	13.31049	.000	-136.6582	-82.1276		
	2	32.41270 [*]	12.96081	.019	5.8637	58.9617		
	4	42.60714 [*]	13.31049	.003	15.3418	69.8724		
4	1	-152.00000 [*]	12.85915	.000	-178.3408	-125.6592		
	2	-10.19444	12.49685	.422	-35.7931	15.4042		
	3	-42.60714 [*]	13.31049	.003	-69.8724	-15.3418		

*. The mean difference is significant at the 0.05 level.

ANOVA										
		Sum of Squares	df	Mean Square	F	Sig.				
Well water	Between Groups	117873.796	3	39291.265	59.403	.000				
	Within Groups	18520.079	28	661.431						
	Total	136393.875	31							
WCH	Between Groups	138314.849	3	46104.950	64.045	.000				
	Within Groups	20156.651	28	719.880						
	Total	158471.500	31							
WOD	Between Groups	102423.824	3	34141.275	42.275	.000				
	Within Groups	22612.645	28	807.594						
	Total	125036.469	31							

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Multiple Comparisons

	(I)	(J)				95% Confidence Interval		
Dependent	VAR00	VAR00	Mean Difference					
Variable	007	007	(I-J)	Std. Error	Sig.	Lower Bound	Upper Bound	
Well	1	2	141.80556 [*]	12.49685	.000	116.2069	167.4042	
		3	109.39286 [*]	13.31049	.000	82.1276	136.6582	
		4	152.00000*	12.85915	.000	125.6592	178.3408	
	2	1	-141.80556*	12.49685	.000	-167.4042	-116.2069	
		3	-32.41270 [*]	12.96081	.019	-58.9617	-5.8637	
		4	10.19444	12.49685	.422	-15.4042	35.7931	
	3	1	-109.39286*	13.31049	.000	-136.6582	-82.1276	
		2	32.41270 [*]	12.96081	.019	5.8637	58.9617	
		4	42.60714 [*]	13.31049	.003	15.3418	69.8724	
	4	1	-152.00000*	12.85915	.000	-178.3408	-125.6592	
		2	-10.19444	12.49685	.422	-35.7931	15.4042	
		3	-42.60714 [*]	13.31049	.003	-69.8724	-15.3418	
WCH	1	2	153.30556 [*]	13.03732	.000	126.5998	180.0113	
		3	128.03571 [*]	13.88615	.000	99.5912	156.4802	
		4	163.00000*	13.41529	.000	135.5200	190.4800	
	2	1	-153.30556 [*]	13.03732	.000	-180.0113	-126.5998	
		3	-25.26984	13.52134	.072	-52.9671	2.4274	
		4	9.69444	13.03732	.463	-17.0113	36.4002	
	3	1	-128.03571 [*]	13.88615	.000	-156.4802	-99.5912	
		2	25.26984	13.52134	.072	-2.4274	52.9671	
		4	34.96429*	13.88615	.018	6.5198	63.4088	
	4	1	-163.00000*	13.41529	.000	-190.4800	-135.5200	
		2	-9.69444	13.03732	.463	-36.4002	17.0113	
		3	-34.96429*	13.88615	.018	-63.4088	-6.5198	
WOD	1	2	132.52778 [*]	13.80877	.000	104.2418	160.8138	
		3	111.32143 [*]	14.70782	.000	81.1938	141.4490	
		4	139.37500 [*]	14.20910	.000	110.2690	168.4810	

	1	-132.52778 [*]	13.80877	.000	-160.8138	-104.2418
	3	-21.20635	14.32143	.150	-50.5425	8.1298
	4					
		6.84722	13.80877	.624	-21.4388	35.1332
2						
3	1	-111.32143 [*]	14.70782	.000	-141.4490	-81.1938
	2	21.20635	14.32143	.150	-8.1298	50.5425
	4	28.05357	14.70782	.067	-2.0740	58.1812
4	1	-139.37500 [*]	14.20910	.000	-168.4810	-110.2690
	2	-6.84722	13.80877	.624	-35.1332	21.4388
	3	-28.05357	14.70782	.067	-58.1812	2.0740

*. The mean difference is significant at the 0.05 level.

Declaration

I, the undersigned, declared that this M.Sc. thesis is my original work, has not been presented for a degree in this or any other university and that all sources of materials used for the thesis have been fully acknowledged.

Name: Abebe Ayiza

Signature: _____

Date of submission: _____

Approval advisors

This thesis has been submitted for examination with my approval as University advisor:

Advisor name: Ketema Bacha (PhD, Professor)

Signature _____

Date: _____