

**EFFECTS OF COFFEE PROCESSING PLANT EFFLUENT ON
BENTHIC MACRO INVERTABRATES DIVERSITY AND WATER
QUALITY OF RUKKISA RIVER IN ALETA WONDO WOREDA,
SOUTHERN ETHIOPIA**

MSc THESIS

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**Effects of Coffee Processing Plant Effluent on Benthic Macro Invertebrates
Diversity and Water Quality of Rukkisa River in Aleta Wondo, Southern
Ethiopia**

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The author Aster Yasa was born in 1992 in Aleta Wondo district. She attended her primary education in Gowadamo Primary School from 1999 to 2001. then she changed her school to Ewuket Fana Primary School from 2001 to 2006. Then she continued her secondary education at Aleta Wondo Secondary School from 2007 to 2008, She attended her preparatory education at Aleta Wondo Preparatory School from 2009 to 2011. After passing the university entrance exam, she joined Arba Minch University and studied her first degree education from 2013 to 2014. After graduation with BSc degree in Biology, she joined directly postgraduate program of Haramaya University to pursue her MSc degree in Applied Biology in 2017.

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ACRONOMYS AND ABBREVIATION

ANOVA	Analysis of Variance
APHA	American Public Health Association
BI	Biotic Index
BMI	Benthic Macro invertebrates
BOD	Biological Oxygen Demand
CLI	Community Loss Index
COD	Chemical Oxygen Demand
DO	Dissolved Oxygen
E	Evenness of the Species
EPA	Environmental Protection Authority
GFF	Glass Fiber Filters
H	Shannon-Weaver Diversity Index
H-FBI	Hilsenhoff Family Biotic Index
MBI	Macro Invertebrate Biotic Index Scores
NPS	Nonpoint Sources
NTU	Nephelometers Turbidity Unity
PS	Point Sources
UNET	United Nation Environmental Program
USEPA	United States Environmental Protection Agency
WHO	World Health Organization

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Effects of Coffee Processing Plant Effluent on Benthic Macro Invertebrate Diversity and Water Quality of Rukkisa River in Aleta Wondo, Southern Ethiopia

ABSTRACT

The use of benthic macro invertebrates together with physico- chemical parameters was considered as the reliable method for water quality assessment. This study was conducted to assess the level of pollution in the Rukkisa River water using biological and physico chemical parameters. Three sampling sites were selected, one site was not affected by the coffee processing plant effluent used as reference site and two affected sites in the outlet and downstream were used to collect the water samples. A total of three water samples were collected from the three sampling sites. Each water sample was subjected to various physico chemical and biological analyses. Five physico chemical parameters pH, Temperature, Total Alkalinity Turbidity and BOD were analyzed. In addition, identification and quantification of benthic macro invertebrates was carried out for each sampling sites. There were a significant differences ($p < 0.05$) in the physico-chemical parameters and macro invertebrates. Higher value of temperature, turbidity, total alkalinity and BOD was recorded at affected sites (outlet & downstream) than reference site (upstream). Most of the physico-chemical parameters were out of World Health Organization and Environmental protection Agency water quality standard. Totally 1149 specimens were identified which were belonged to 26 different species. 334 benthic macro invertebrates were collected from upstream, 431 and 384 benthic macro

invertebrates were collected from outlet and downstream, respectively. The numbers of taxa were higher in upstream than impacted sites. 24 different types of species were identified from upstream 20 and 21 species from impacted (outlet & downstream), respectively. Categorical physico chemical parameters associated with benthic macro invertebrates indices. The study showed the effectiveness of the benthic macro invertebrates' protocol in assessing River pollution condition. Therefore, environmental agencies and researchers have a good option of using macro invertebrates with the objective in assessing and monitoring programs of Rivers.

KEY WORDS – *biotic index, effluent, Macro- invertebrates, physico-chemical parameter and taxa.*

1. INTRODUCTION

Two-thirds or about 70% of the earth's surface is covered by water. From the 70% of water coverage only less than 1% of the water on the planet is readily available for drinking or for most agriculture. About 97% water on the earth is salt water stored in the oceans; only 3% of water on the earth is fresh water. From all of the freshwater on earth, 68% is in the icecaps of Antarctica and Greenland, 30% of it is fresh water in the ground, and only 0.3% covers surface waters, such as lakes and rivers (Shakhashiri, 2011).

Freshwater ecosystems represent 0.01% of all water ecosystems on the planet. This covers a total of 0.8% of the earth's surface (Jackson *et al.*, 2001). Yet this tiny fraction of global water supports at least 100,000 species, out of approximately 1.75 million (Gibert and Deharveng, 2002). But these freshwater ecosystems have been altered by human disturbances such as agriculture, urban development, impoundment, canalization, mining, forest fire suppression, road construction, species introduction and subsequent invasion (LaBonte *et al.*, 2001),

Water pollution happens when the water becomes overloaded with too much of one thing and the aquatic organisms cannot keep up with their cleaning responsibilities. Some organisms may die and others may grow too fast. There are two major water pollution sources; they are point water pollution and nonpoint water pollution sources (Dozier, 1987).

Nonpoint source of pollution (NPS) comes from many mixed or diffuse sources rather than from an identifiable specific point. This originated from urban or rural environments such as yards in neighborhoods or from agricultural production areas and crop fields. Point source pollution (PS) is from known specific source like an industrial or sewage outfall pipe, and associated with manufacturing processes discharges from water treatment plants and large animal feeding operations (Dozier, 1987).

Effluent from coffee processing plant is one of the point source of water pollution. It is one of the major agro-based industries which is responsible for fluctuating freshwater ecosystems (Salvamurugan *et al.*, 2010). The process discharges enormous amounts of effluents which have the probability to hurt the environment. (Campos *et al.*, 2010).

Coffee processing discharge are the main source of organic pollution in environment where concentrated coffee processing is practiced without suitable management systems. Environments that are affected by coffee processing plants display change in terms of their physical, biological and chemical behavior. Coffee waste water composed sugars and pectin and hence it is amenable to rapid biodegradations. Coffee waste water is concentrated with suspended solids, dissolved solids and higher nutrient. Mucilage and coffee pulp are other components which change water quality. Coffee pulp components are responsible for pollution of nearby water bodies and receiving environment. In addition to coffee pulp, mucilage also plays a great role in water pollution (Asrat *et al*, 2014).

Several taxa contribute to biodiversity in fresh water ecosystems. Aquatic macro invertebrates play a central ecological role in many running water ecosystems (Boulton, 2003) and are among the most ubiquitous (Voelz and McArthur, 2000) and diverse organisms in fresh waters. Despite the important roles played by macro invertebrates in aquatic ecosystems, it is apparent that macro invertebrate research (their diversity and anthropogenic impacts on them) in Ethiopia is limited (Strayer, 2006).

There are massive rivers in Ethiopia that move long distance through the towns and used as major source of water for consumption and other activity but they are industrial residues dumping area. The effluents from some industries are discharged into these rivers. There are also some other source of pollution of water supply in Ethiopia. Some pollutants released dangerous chemical products and others settled with different waste materials. People around the area use the water for domestic consumption and for other activities like irrigation of garden areas (Ademe and Alemayehu, 2014).

In general there was water pollution problem from coffee processing plant effluent on the water quality of discharge water body. In addition to water quality, the richness of benthic macro invertebrate's community composition was used to monitoring the water pollution. Moreover, in the study area (Rukisa River) has wet coffee processing plants which discharge their wastes directly into the river or stored the waste around river. So this study was done to assess the diversity of benthic macro invertebrate's community and the impact of the effluent on their biodiversity. The water quality of the river after the discharge and before discharge was

changed and the number of benthic macro invertebrates changed which causes various health problem, odor change, as well as effect on domestic animal. The parameters of the river were changed after the effluent of coffee processing plant discharged. People residing in the surrounding area of this plant were utilizing this stream water for domestic uses and were suffering from severe health problems. There was no documented data on effect of coffee processing plant effluent on benthic macro invertebrate's diversity and water quality in selected area. As a result the present study designed to asses water quality and diversity of benthic microorganisms of wastewater discharged from wet coffee processing site in Rukisa River.

General objective

To evaluate the effect of coffee processing plant effluent on benthic macro invertebrate's diversity and water quality of Rukisa River in Aleta Wondo, Southern Ethiopia

Specific objectives

1. To analyze Physico chemical parameters of the Rukisa river.
2. To identify the benthic macro invertebrates species in the Rukisa river.
3. To determine the diversity of the benthic macro invertebrates in the Rukisa river.
4. To associate water quality parameters with biological characteristics of Rukisa river.

2. LITERATURE REVIEW

2.1 Water Resources in Ethiopia

Total Ethiopia has 12 major rivers, and over 12 major swamps. The 123.25 billion m³ of water flow annual from these 12 rivers and can say 'Ethiopia is the water tower of East Africa'. The water tower of east Africa said in terms of receiving and donating of the sufficient water with adjacent countries. On the other hand not sufficient water willingly accessible for drinking use in the country (Tenalem, 2009).

Since Major River formed deep gorge in the country and move to near country this utilizes water resource of the country. The spring and streams exist only a high land area occupies 40%. There is no surface runoff areas blow 1500meters above sea level which cover 55% of the country. Ethiopia has equally distributed ground water that occupies 2.6 billion m³ in low land areas of the country. Due to the financial and employment problem water resource of the Ethiopia densely reflected as cause for environmental degradation. (Mekiso, 2004.).

2.2 Causes of Water Pollution

Both natural and manmade activity influences the quality of surface water. Water has dissolved and undissolved substances and living organisms natural, which is necessary to good water quality. Major source of water pollution is from human activities such as industrial effluent, domestic waste and agricultural activities. This all change Physico chemical characteristics of the water (EPA 2011).

“Water quality was affected by a wide range of natural and human influences” (Bartram, 1996). The natural impacts are geological, hydrological and climatic, since these affect the quantity and the quality of water availability. Natural ecosystem corresponds with water quality when the water quality changed the ecosystem interrupted or changed. Pollution of water by human faces is attributable to only one source. The human activities affect water qualities by both widespread and diverse in the degree. But the reasons for this type of pollution, its impacts are varied on water quality and the necessary corrective actions. (Bartram, 1996).

“There are different source of water pollution in which some of the pollutant may exposed by human beings and other lives to series problem” (Katko 1989). Direct release of industrial waste into waterways pollutes the water and also industrialization has encouraged relationship with increases of organic water pollution. The rapid growth of industry increase water pollution (Reddytvk 1987).

In urban areas careless dumping of industrial wastes and other wastes add significance effect on water quality. In the developing country the most urban rivers are end point of wastes discharged from the industries, while industrial production can affect water quality. Industrial facility to find a new source of water may force Poor water quality, halt production and it may decrease the quality of the product” (phiri *et al* 2005).

The Worldwide estimate 300-400 million tons of heavy metals, solvents, toxic sludge, and other industrial waste dump into water sources in each year (UNEP, 2010). The industrial pollution change water quality characteristics such as temperature, acidity, salinity and turbidity of receiving waters, leading to changed biomes and higher frequency of water-borne diseases. The effects can be sensitive by the synergistic mixture of pollutants affecting species communities and deprivation of other environmental facilities (Ademe and Alemayehu, 2014).

Organic matter is the major pollutant in water. Traditionally organic matter has been measured as Biological Oxygen Demand and Chemical Oxygen Demand. The Chemical Oxygen Demand analysis is ‘quick and dirty’ (if mercury is used). Biological Oxygen Demand is slow and cumbersome due to the need for dilution series (Henze and Comeau, 2008).

2.3 Industrial Waste Water

Industrial effluents are the major source of pollution of water and air in the environment. Based on the types of industry, various levels of pollutant can be discharged into the environment directly or indirectly through public sewer line. The operation and processing of many industries generate Waste water. Depending on the industry and their water use, the waste water contain suspended solids, both degradable and non-degradable organic matter, heavy metal ion, dissolved inorganic acids and coloring compounds (kanu *et al.*, 2011)

The industrial waste water is significantly different in both movement and contamination strength. Also the waste waters from industrial discharge may contain suspended, colloidal and dissolved organic mineral. Industrial waste water is excessively acidic or basic and also comprises high or low concentrations of bleached matter. It also affected by inert, organic, toxic materials and perhaps by pathogenic bacteria. These wastes discharged into the drain system without effective treatment and have undesirable effects on the drain system. The solid content, color, odor and temperature are physical characteristics of industrial waste water. The solid in waste water contain the insoluble, suspended solids and soluble compounds dissolved in water. Color is a qualitative characteristic used to assess the general condition of industrial waste water (Alturkmani, 2014.).

Waste water with light brown color is less decomposition, light-to-medium grey color is characterized by some degree of decomposition and when the color is dark grey or black, the waste water is characteristically septic. The odor of fresh waste water is usually not offensive, but a variety of odorous compounds are released when waste water is decomposed biologically under anaerobic conditions (Alturkmani, 2014.).

The biochemical oxygen demand (BOD), chemical oxygen demand (COD) and entire organic carbon (TOC) are the component of industrial waste water quality determination. Specific organic compounds are determined to assess the presence of priority pollutants". The BOD, COD and TOC are gross measure of organic content and do not reflect the response of the industrial waste water to various types of biological treatment technologies (Metcalf and Eddy, 1991).

2.4 Coffee Processing

Coffee processing steps grouped into primary, secondary and tertiary steps. Primary coffee processing is cutting of coffee beans from coffee tree (also called green beans). The next stage refers to harvesting, roasting and grinding. Tertiary processing involves making of prepared coffee or other value addition operations. Primary coffee processing is carried out within coffee plantations (Henze and Comeau, 2008).

Primary processing produces green beans from the coffee fruit. The coffee fruit consists of colored exocarp (skin), fleshy yellowish-white mesocarp (pulp), mucilage layers (covering the two beans joined together along the flat sides and made up mainly of pectin) and two coats (first a thin fibrous textured parchment and second a fine membrane, silver skin). Primary processing is done in two major ways, the dry and wet methods. In the 'dry' method, the fruits are picked and laid out to dry in the sun for 3-4 weeks and processed (Chanakya and Alwis 2004).

The 'wet' method or 'washed' coffee methods are used in locations with plentiful supplies of fresh water. It is carried out in two steps, pulping and washing. The distribution of wet and dry processing in the producing countries follow both methods and the rest desire to change to wet processing if possible. Ethiopia is one of the countries that produce wet processed coffee. The coffee fruit is squeezed between two serrated metal plates and the skin of the pulp are detached from the seed. The mucilage-coated seeds and fruit skins (with pulp) are separated into different streams. The skin and pulp are carried away in a stream of flowing water (Devi and Alemayehu, 2007).

In the receiving tank the skin with the pulp is separated out (solid waste) from the waste water (pulp water). The pulp water most often joins the waste water stream. However, there is an acute shortage of fresh water (as in many plantations today), this water recovered and recycled for 2–5 days in the pulping process. Freshly pulped coffee seeds are covered with a highly slippery mucilaginous layer approximately 1.5 mm thick and translucent. This slippery mucilaginous layer cover washed after fermentation by machine called a 'washer. After that the clean coffee beans goes other direction and the waste water with the slippery mucilaginous dump in one place and discharge into water body direct and indirect way this pollute water body(Chanakya and Alwis 2004).

2.5 Water Quality

Water quality is a technical term based on the characteristics of water in relation to guideline values of suitability 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, 1999).

Water quality is the term used to express the suitability of water to sustain several uses or processes. All kind of uses have certain requirements for the physical, chemical and biological characteristics of water. Water quality is a variable which limit water uses. It also comprises between the quality and quantity demand by different users to improve or maintain quality. The recognition of natural ecosystems increases the appropriate consideration option for water quality management. This is indicator of changes or decline on in overall water quality as long as useful physical, chemical and other evidence (Bartram *et al*, 1996).

Waste water quality can be well-defined by physical, chemical, and biological characteristics. Physical parameters include color, odor, temperature, solids, turbidity, oil, and fat. Chemical parameters associated with the organic components of waste water contain the biological and chemical oxygen demand , entire organic carbon (TOC), and overall oxygen demand (TOD). Inorganic chemical parameters include pH, acidity, nutrients and the like (Manahan, 2001).

Quality of the surface and ground water is influenced by both natural and human activities. Natural water quality would be determined by weathering of bed rock minerals. The atmospheric process of evapotranspiration, the deposition of dust and salt by wind is another natural activity. The natural leaching of organic matter and nutrients from soil, hydrological factors that lead to runoff and biological processes within the aquatic environment that can alter the physical and chemical composition of water. As a result, water in the natural environment contains many dissolved substances and non-dissolved particulate matter (Stark *et al.*, 2000).

Physico-chemical parameters are very important to test the water before it is used for drinking, domestic, agricultural and industrial purpose. The selection of these parameters for testing of water is only depend upon for what purpose to use that water and what extent we need it. Some physical test should be performed for testing of its physical appearance include temperature, color, odor, pH, turbidity, total dissolved solid etc. while chemical tests should be perform for its biological oxygen demand, chemical oxygen demand, dissolved oxygen,

alkalinity, hardness and other characters. It is obvious that drinking water should pass these entire tests and it should contain required amount of mineral level. Only in the developed countries all these criteria's are strictly monitored (Sawyer *et al*, 2003).

Different physicochemical parameters are tested regularly for monitoring of water quality. Physico chemical parameter study is very important to get exact idea about the quality of water (Patil *et al* .,2012).

The Physico chemical analysis of the waste water generated from the coffee processing plant pollutes water highly with organic load, nutrients. This organic load is measured in terms of chemical oxygen demand, biological oxygen demand and nutrients in the term of phosphate and nitrate. The values of this organic load, nutrients and different dissolved matter measured after the effluent is very high (Devi and Alemayehu, 2007).

The availability of water and its physical, chemical, and biological composition affect the ability of aquatic environments to sustain healthy ecosystems: as water quality and quantity are eroded, organisms suffer and ecosystem services lost. Typically, water quality is determined by comparing the physical and chemical characteristics of water with water quality guidelines or standards (Carr, and Neary, 2006).

2.6 Physico-Chemical Parameters

Oxygen that is dissolved in the water column is one of the most important components of aquatic systems. Oxygen is often used as an indicator of water quality, such that high concentrations of oxygen usually indicate good water quality (Carr, and Neary, 2006).

The biological oxygen demand (BOD) is oxygen required for breaking down of organic matter and its value increases when the water is polluted Devaraju *et al* (2005) has made Comparable observations in Maddur Lake with Garg *et al*, (2010) likewise the observation in Ramsagar reservoir.

High biological oxygen demand (BOD) value is unflavored by zooplankton. High value of biological oxygen demand is unfavorable for aquatic small animal. The waste water has high

oxygen-demanding there is biodegradable material in water. It is useful measure for the strength of effluent and its pollution potential (Asrat, *et al* 2014).

Biological oxygen demand reflects organic material contamination in water. Biological oxygen demand is the amount of dissolved oxygen required for the biochemical decomposition of organic compounds and the oxidation of certain inorganic materials (Patil,*et al* 2012).

Biochemical oxygen demand represents the amount of oxygen consumed by bacteria and other microorganisms while they decompose organic matter under aerobic conditions at a specified temperature (Delzer and McKenzie, 2013).

Biochemical oxygen demand can be used as gauge of the effectiveness of waste water treatment plants (Sawyer *et al*, 2003; Patil.*et al* 2012). The chemical oxygen demand content is useful for indication in relation to the design of treatment processes and subdivided in to fractions. The suspended and soluble chemical oxygen demand measurement is very useful fractions (Henze and Comeau, 2008).

The temperature of reservoir water varies in different seasons. The variations are high at every area (Sharma *et al* 2000). Temperature affects the concentration of dissolved oxygen and influence the activity of bacteria in water bodies. The biological functions as well as chemical functions affected by temperature. All chemical reactions depend on temperature (Atnaf, 2006, Olson, 2004).

Temperature affects the speed of chemical reactions, the rate at which algae and aquatic plants photosynthesize, the metabolic rate of other organisms, as well as how pollutants, parasites, and other pathogens interact with aquatic residents (Atnaf, 2006). Temperature is important in aquatic systems because it can cause mortality and it can influence the solubility of dissolved oxygen (DO) and other materials in the water column. Thermal pollution comes in the form of direct impacts, such as the discharge of industrial cooling water into aquatic receiving bodies, or indirectly through human activities such as the removal of shading stream bank vegetation or the construction of impoundments. urbanization, forestry, agriculture, impoundments, and industrial effluents can cause changes in surface water temperatures (Carr, and Neary, 2006).

The balance between positive hydrogen ions (H^+) and negative hydroxide ions (OH^-) in water regulate basicity and acidity of water. The determination of pH is determination of ions inversely. When pH value is " 0" there is high concentration of hydrogen ions, or strongly acidic. When pH value is " 14" there is high concentration of hydroxide ions or strongly basic (Gaur, 2008).

The pH value increase due to attributed sewage discharged by surrounding city and agricultural fields. The high value of pH is significant for aquatic plant growth according (Chisty, 2002; Umavathi *et al*, 2007).

The coffee waste water is acidic therefore discharging the waste directly to the water causes pollution. Volatile solids from pulp juice and mucilage increase the acidic properties of receiving water bodies. The increasing of pH values shows the wastes have high potential to pollute water bodies because the organic components are degradable. The value of the organic components delivers food for microorganisms and possible vectors example mosquito. therefore, coffee waste water has potential to increases odors and subsequently increases attraction of vectors(Asrat *et al*,2014).

During the coffee processing, the fermentation process in the effluents from pulp, fermentation tanks and mechanical mucilage removers, sugars fermented in the presence of yeasts to alcohol and CO_2 .However, in this situation the alcohol is quickly converted to vinegar or acetic acid in the fermented pulping water (Enden and Calvert 2002).

The effluent discharged from coffee processing plant into the water body cause water turbidity. The algal or aquatic weeds growth and breakdown of them increase water turbidity. Turbidity provides the food and shelter for microorganisms which may be pathogen to human. The higher the turbidity the higher cloudiness then the water gives bad smell and waterborne disease outbreak (Sharma *et al* 2000). The value of turbidity was one of the criteria of water quality parameters. The Turbidity of household water samples measured in Addis Ababa merkato higher than the turbidity of tap water source water and tank (selamawit, 2012).

The water turbidity measured by the cloudiness that indicates the growth of microorganisms which may pollute the water (Olson, 2004). During rainy season the water has high turbidity.

The results supported by Dagaonkar and Saksena (1992). This high turbidity during rainy season is combination of silt, clay and other suspended particles add turbidity values.

“Too much algae or sediment in lakes and streams can make them unsuitable for recreation and aquatic life” (Minnesota Pollution Control Agency, 2008).

Turbidity is produced by particles suspended or mixing in water that cut light transparency making the water appear gloomy or dark. Sediment like as clay and silt, fine biological and chemical matter, soluble colored organic mixtures, algae, and extra minuscule creatures. Algae that grow with sustenance from nutrients inflowing the tributary through leaf decay or other naturally happening decay processes can also be a basis of turbidity (Minnesota Pollution Control Agency, 2008).

2.7 Macro invertebrates and Water Quality

There are many macro invertebrates found with different taxa and grouped into five groups such as Platy helminthes, Annelida, Mollusca, Crustacean and insect. The macro invertebrate at the upper Stream was categorized water quality with a high species richness dominated like Ephemeroptera, odonata, Diptera and Coleopteran. The clean streams have a higher proportion of Gammarus and polluted streams have a higher proportion of Asellus (insect larvae). Asellus was a good indicator of organic pollution. The Asellus community was dominated by insect larvae and the substrate flow at both sites limit the abundance of the general fauna (Kelly-Quinn *et al*, 2003).

In the Caver River, major chronological alterations were observed in the community arrangement of the macro invertebrate fauna among the seasons. The observed richness reduction showed that the macro invertebrate fauna become controlled by a few species during late summer. The abundance of fauna shows a generalized pattern among the studies low in the winter and then slowly rapid increase in summer. Gammarus, Asellus, Lymnea, Planorbis Baetis and Chironomidae were major indicators in the water. Asellus, Lymnea, Planorbis Baetis species are often dominated and common in weakly polluted water. This is definite by the

chemical outcomes having greater values of Ammonia nitrogen, nitrate, nitrite, Chemical oxygen Demand and Phosphate in the lower section (Duran, 2006).

Zooplankton is the community of invertebrates that is suspended in the water column, where as lake and river bottoms are inhabited by benthic macro invertebrates. Invertebrates have the ability to control the abundance of algae through grazing and they are important food source to organisms in higher trophic levels, such as fish and other predatory invertebrates.

Invertebrate assemblages are good indicators of localized conditions because many have limited migration patterns or are sessile (non-motile) (Carr, and Neary, 2006).

2.8 Correlation (r) between Water Quality and Benthic Macro invertebrates

The correlation coefficient (r) between parameter and benthic macro invertebrate's indices calculated by using their index values. Correlation coefficient (r) between parameters and macro invertebrates show association between two of the Lower Manair Reservoir water. Correlation coefficient (r) was the degree of line association between two of the water quality parameters (Thirupathaiah *et al*, 2012).

The quarter of components influenced by the physicochemical parameters is seasons and sections. The least components influenced by abundance of diversity, dissolved O₂ and hardness. Physico chemical parameters provided a strong discrimination of the five distinct groups. Also warm water and high value of some Ammonium ions might be limiting for many species and high nitrogen affects water quality and communities (Duran, 2006).

Decomposable organic matter results in consumption of DO in the receiving streams and does not differentiate between biologically degradable & nondegradable organic matter. Chemical oxygen demand is related to the biological oxygen demand (Marg and Khas, 1999).

3. MATERIAL AND METHODS

3.1 Description of the Study Area

The study was conducted in Aleta Wondo Woreda which is found in the Sidama Zone, Southern Nations, Nationalities and People's Regional State (Figure 1). The area located between 6°15' and 6°45' N latitude, and between 38°15' and 38°45' E longitudes. Aleta Wondo is bordered by Dale Woreda in the North, Darra Woreda in the South, Bursa Woreda in the West, Chukko Woreda in the East and Hulla Woreda in the South west. The administrative center is Aleta Wondo (Anteneh, 2010).

The study area spans over a total area of about 567.03 square kilometer and inhabited by 347,123 people with crude density of 612.2 persons per sq. km. A survey of the land in this woreda shows that 72% is arable or cultivable, 12.9% pasture, 7% forest, and the remaining 8% is considered swampy, degraded or otherwise unusable. Important cash crops include corn, wheat, barley, beans, local varieties of cabbage, and shallots. Coffee Arabica, enset (veterocuse) and fruit are perennial crops in the area and their productivity and production depends on the availability of the required amount of rain in addition to other required inputs. The country essentially coffee was produced in the Aleta Wondo woreda from the Southern region of Ethiopia (Anteneh, 2010).

The study area comprises varied thermal zones ranging from 'kola' (Tropical) to 'Dega'. There are three distinct agro-ecological zones; 12 % of the Woreda is classified as Dega (highlands), 71 % as Woinadega (midlands), and 17 % as dry Kolla (lowlands). The Woreda receive a mean annual rainfall varying from 801-1000mm in the western parts to 1401-1600mm in the central and northern parts (Anteneh, 2010).

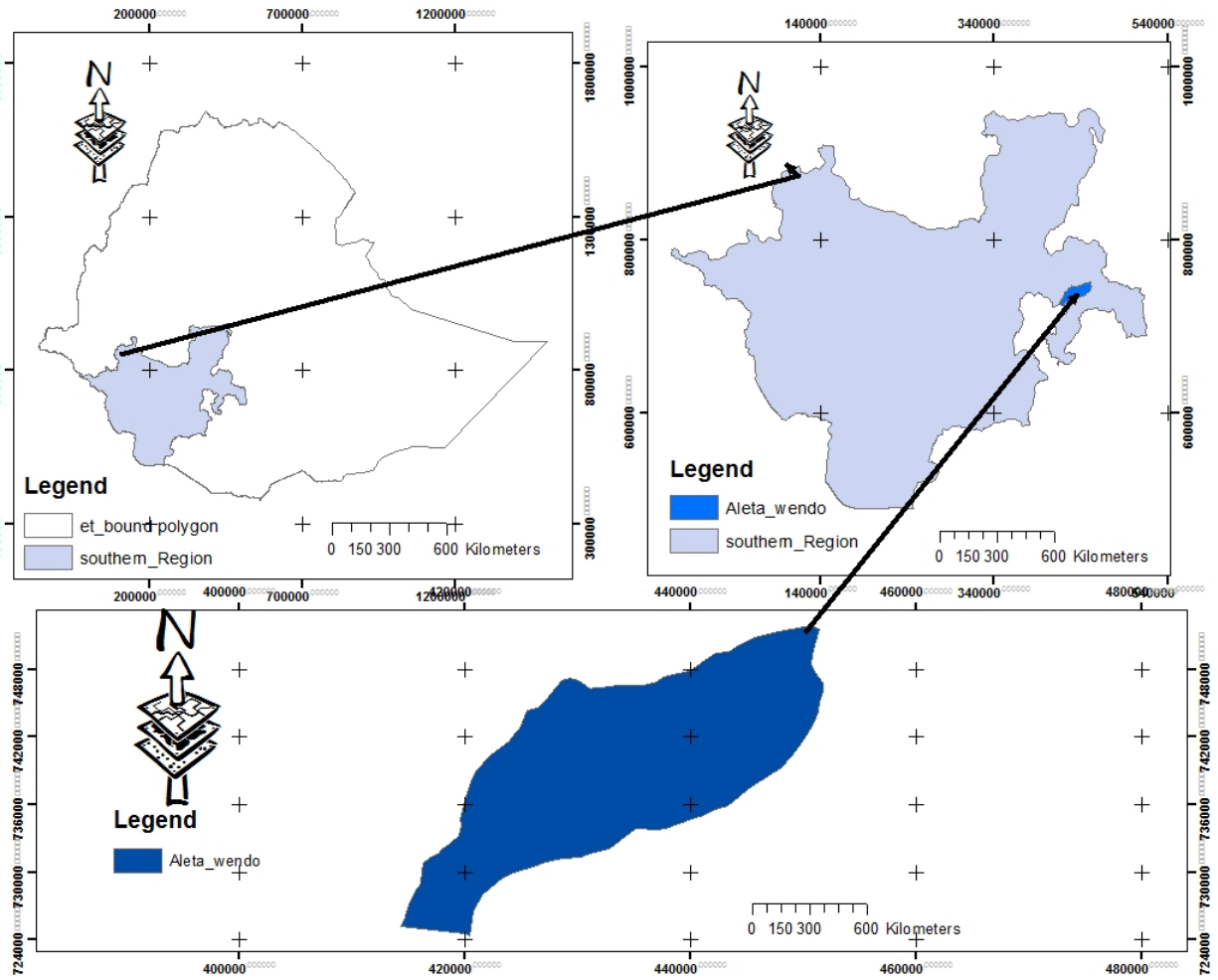


Figure 1. Geographical Location of the Study Area (GIS).

3.2. The Research Design and Sampling Methods

Laboratory based cross sectional study was conducted to assess the effect of coffee processing plant effluent on benthic macro invertebrate diversity and water quality of Rukkisa River using physico-chemical parameters and benthic macro invertebrates in Aleta Wondo, Southern Ethiopia .Wet coffee processing in Aleta Wondo usually begins at the end of September and proceeds until mid-December. Consequently, sample was collected at the end of November 2016 and the second round sampling of benthic macro invertebrates done in the beginning of December 2016. Although all the coffee processing plants are following the same wet coffee processing method and expected to produce waste water of similar composition. The sample was collected using the standard method (EPA 2012). This is a standard method for the examination of water and waste water.

The coffee processing plants were purposively selected to make the sampling representative. In addition, identification and quantification of benthic macro invertebrates (BMI) species as well as their diversity estimation was investigated for each water sampling sites. The design based on laboratory investigation was carried out by water samples from three sites using purposive sampling in two times for physico chemical parameters like temperature, turbidity, pH, total Alkalinity and biological oxygen demand.

Benthic macro invertebrates' collection was done by using D-net (net with 500 μm mesh size).To assess the influence of wet coffee processing waste water being discharged, water samples were taken from the rivers that receive waste water from coffee processing at upstream and downstream of the discharging points. A standard method was applied for all procedure of the set of experiments. All the chemical reagents used were analytical graded and their expiry dates was checked.

3.3 Sampling Sites and Collection of River Water Samples

Three sampling sites were selected based on purposely sampling methods; these were upstream, outlet of effluent and downstream. The reference condition collectively refers to the range of quantifiable ecological elements (chemistry and biology) that were found in minimally disturbed environments. Finding reference sites in streams and rivers are a difficult task, because no regions are entirely without areas of human disturbance.

Therefore, the reference site has to be selected based on minimally or least disturbed attributes (Gregory *et al.*, 1991). The reference site was identified based on the following criteria as a reference site that indicated by Jennifer *et al.*, (2003): the Same water body type, the same size, chemical characteristics as treated site, within same watershed as treated site, minimal application of aquatic pesticides within the last few years, and Limited anthropological inputs.

According to the above criteria, a reference site the upstream was selected to compare against the impacted sites, outlet of effluent and downstream. Chemical and biological measures taken in impacted sites were compared to reference site. The reference site was chosen because it was found in the upper stream of the river with good habitat quality and had minimal anthropogenic interventions.

The watershed of Rukkisa River is highly exposed to several anthropogenic interventions including- agricultural activities and waste water from coffee processing plant are the major industrial activities along the watersheds. There is no other industry along the watershed to affect the habitat qualities of the river. Agricultural activities (both farming and grazing) are known as source of pollution and a riparian vegetation removal. The sampling site selection was conducted which based on 100m length with riffle zone. This site was selected according to the USEPA Rapid Bio assessment protocol criteria (Barbour *et al.*, 1999). Three sites were sampled along the course of the river.

Upstream- is located at the upstream of the river .The area was covered by distinctive vegetation and grasses. This portion was clear, clean and was chosen as the control point on the

assumption that it was unpolluted. It was a reference site for bio assessment with less human activity, which was around 300 m upstream far from the factory.

Releasing point (outlet) of the effluent- is located at the point of effluent from coffee processing plant. It is outlet of the effluent and contact point of the effluent from the plant before the flow to downstream.

Downstream- is around 300m far from outlet and mainly covered by polluted water. Thus a composite sampling technique was employed to take water samples at 3 points across the width of rivers for chemical analysis (Bartram and Balance, 2001).

The samples were transported to laboratory within 1-6 hours for analysis and chemical analysis following the standard methods (EPA, 1998). A total nine samples of water were taken. One liter of water sample was taken from each site using a one liter plastic bottle and stored in ice box prior to analysis.

3.4 Measurements of Physico-Chemical parameters

Physico-chemical variables were measure to assess the amount at pollution of Rukkisa River. The parameters considered, include pH, temperature, turbidity, total Alkalinity and BOD. Temperature was measured by digital thermometer which was carried out on site. pH of water was measured using portable digital pH meter and turbidity by digital turbididometer (USGS, 2003). Total alkalinity was measured by digital portable titrator. Titration was done by addition of small, precise quantities of sulfuric acid (the reagent) to the sample until the sample reaches a certain pH (the endpoint). The amount of acid used corresponds to the total alkalinity of the sample water. Digital titrator have counters that display numbers. A plunger was forced into a cartridge containing the reagent by turning a knob on the titrator. As the knob turns, the counter changes in proportion to the amount of reagent used. Alkalinity was then calculated based on the amount used (USGS, 2003).

$$\text{Alkalinity (as mg/l CaCO}_3\text{)} = (2a - b) \times 0.1$$

Where:

a = digits of titrant to reach pH 4.5

b = digits of titrant to reach pH 4.2 (including digits required to get to pH 4.5)

0.1 = digit multiplier for a 0.16 titration cartridge and a 100-ml sample

3.4.1 Biological Oxygen Demand (BOD)

In order to measure BOD, the water sample was taken according to the standard described in Table 1.

Table 1, Assumption of Simplified sample

BOD range (mg/L)	Sample volume (mg/L)
0 to 35	420
0 to 70	355
0 to 350	160
0 to 700	95

As shown in Table 1 by assuming the water sample BOD ranges from 0-350 (mg/L) 160 mL of water sample was taken and cooled the sample by 19 to 21 °C. The sample was homogenized and appropriate amount of sample was taken using a graduated cylinder. One contents of nutrient buffer pillow was added into the graduated cylinder then moved into BOD Track II bottle. Stir bar was put into the bottle and the bottle neck was sealed by seal cup. Two potassium hydroxide pellets was added to the seal cup by using spatula scoop. The bottle was putt on the BOD Trak II chassis and the applicable tubes were connected to the sample bottle and tighten the cap. The BOD Trak II chassis or instrument was taken into the Incubator with temperature 20 ± 1 °C. Then the power was on and the left and right arrows pushed at the same time to set up instrument menu, then the time and date was set. After the end of the 5 days display shows "END". The result was shown on the BOD Trak II display. Then the value was calculated.

BOD₅ is calculated by: $BOD_5 = (DO - D_5) / P$ or

$$BOD_5 = (DO - D_5) - (BO - B_5) f / p$$

Where:

D_0 is the dissolved oxygen (DO) of the diluted solution after preparation (mg/l)

D_5 is the DO of the diluted solution after 5 day incubation (mg/l)

P is the decimal dilution factor

B_0 is the DO of diluted seed sample after preparation (mg/l)

B_5 is the DO of diluted seed sample after 5 day incubation (mg/l)

f is the ratio of seed volume in dilution solution to seed volume in BOD test on seed.

3.5 Sampling of Benthic Macro invertebrates

Sampling of benthic macro invertebrates was done at the end of November 2016 and the second round of sampling of benthic macro invertebrates was done at the beginning of December 2016. The sample was collected in three sites upstream, at the point of outlet of effluent and downstream to make sample representative. The sample was collected using D-net (net with 500 μ m mesh size) with simple procedure for stream-dwelling macro invertebrates. It was used in riffle areas where the majority of the organisms prefer to live. The method was quite effective in determining relative stream health (Typical riffle) locate shallow, faster moving mud-free section of stream with a stream bed composed of material ranging in size from one-quarter inch gravel or sand to ten-inch cobbles. The depth of water was approximately two inches to a foot with moderate flow and water was not disturbed recently. At each site, qualitative macro invertebrates sample were collected from a River section of about 100 meter length, and finally preserved in 70% ethanol.

The debris and other confusing Detroit were removed. After that the macro invertebrates was picked out by forceps and taken to Hawassa University Fisher laboratory. Then the

identification of macro invertebrates was done using binocular compound microscope and various available keys and assigned taxonomic levels. All of the macro invertebrates were collected and identified to appropriate taxonomic level such as order, suborder, family and species (Mandaville, 2002). Biological Survey Data Sheet was used to calculate various metrics that assess stream integrity.

3.5.1 Numerical Identification of Benthic Macro invertebrates

The macro invertebrates' numerical identification represents the richness of taxa, taxa loss due to intolerant to the pollution, family biotic index. The criteria for metric selection were the ability to be associated with physical degradation, ability to provide unique information about the River and ability to discriminate reference site from impacted sites (Chihart, 2003).

Taxa richness measures the abundance of different types of organisms as determined by the total number of taxa represented in sample. Generally, taxa richness increases as water quality, habitat diversity and habitat suitability increase.

However, some pristine headwater streams naturally harbor few taxa, while the number of taxa can actually increase in polluted streams. Sample density estimates the total number of organisms collected from stream site after subsampling. Sample represents the total number of organisms collected (Hilsenhoff, 1988).

A) Macro invertebrates Biotic Index Scores (MBI)

Subsample estimated as sample density before, but the number of subsampled organisms is needed to calculate the Macro invertebrates Biotic Index. Nutrient-enriched water has a high density of organisms, while water polluted with toxic chemicals or silt or sand usually has a lower density (Hilsenhoff, 1977). The Macro invertebrates Biotic Index scores (MBI) and the percent composition of taxa in a stream determines the presence or absence of taxa which have a high pollution tolerance. MBI values reflect stream quality as follows:

- 1, Less than 6.0 = good water quality
2. 6.0 to 7.5 = fair water quality

3. 7.6 to 8.9 = poor water quality

4. Greater than or equal to 9.0 = very poor water quality, (Hilsenhoff, 1977).

To calculate biotic index score of identified macro invertebrates multiply the number of organisms identified from each taxon by its tolerance rating. The “Tolerance Rating (Ti)” is the value of the macro invertebrates tolerate to the organic pollution. The Greek letter Σ sigma is the symbol for “total. “Macro invertebrate Biotic Index” is the total tolerance value divided by the total number of organisms (Hilsenhoff, 1987).

$$MBI = \sum (Tv) \div \sum N$$

“Taxa Richness” is the total number of taxa that were identified -- $\sum \text{Taxa}$

“Sample Density” is the total number of indicator organisms collected or subsampled -- $\sum N$.

B) Percent Composition of Taxa (%C)

Percent Composition reflects the organisms were most prominent in the stream. The No. of Organisms (N) in each taxon was divided by its community density (“ $\sum N$ ”) and multiplied by 100 to obtain the percent composition.

$$\%C = (N) \div \sum N \times 100$$

The “% C” of each taxon obtained was subtotal percentage (“% subtotal”) was added to obtain percentage of organism. The “% subtotal” from 100% to obtain the percentage of other organisms in the sample was subtracted (Hilsenhoff, 1977). The percent composition (%C) of macro invertebrate taxa also reflects stream quality. Streams with high percentages of mayflies and stoneflies are considered to be in good health. Those that harbor a high percentage of midge larvae and aquatic worms are considered to be in poor health, since these organisms are tolerant to some types of pollution that reduce dissolved oxygen levels (Hilsenhoff, 1977).

C) Percent Dominance (%DF)

Percent Dominance (%DF) = The Percentage Contribution of Dominant Family or percent dominance (%DF) equals the abundance of the numerically dominant family relative to the total number of organisms in the sample. Percent dominance (%DF) is the sum of individuals in the three (3) most abundant taxa in each replicate, divided by the total number of individuals in that replicate, multiplied by 100. The percentages from the three replicates are then averaged for this metric. This indicates water quality with respect to pollution.

$$\%DF = \frac{\text{Total dominant species}}{\text{total macro organisms}} * 100$$

D) Ratio of EPT to Chironomidae

Total number of individuals in Orders Ephemeroptera, Placoptera, and Trichoptera divided by the total number of Chironomidae individuals.

$$\text{RatioEPT} = \frac{\sum \text{EPT}}{\sum \text{chiro.}}$$

The Ratio of EPT to Chironomidae abundance shows the number of individuals from sensitive orders compared to pollution tolerant or (Chironomidae). A high ratio indicates low levels of water pollution.

E) Dissimilarity of Taxa or Community Loss Index

Dissimilarity between taxa from site to site indicates loss of taxa or community loss index (CLI). Community Loss Index determines the loss of taxa in a study site with respect to a reference site.

Community loss index was calculated as:

Where, A- is the number of taxa common to both sites

D- Is the total number of taxa present in reference site

E- Total numbers of taxa present in the study sites.

F) Hilsenhoff Family Biotic Index (BI)

Hilsenhoff family Biotic Index (BI) based on categorizing macro invertebrates into categories depending on their response to organic pollution. The tolerance of various levels of dissolved oxygen, one of the most comprehensive of these indexes was the one proposed by the Hilsenhoff (1988) formula:

Where n_i is the number of specimens in each taxonomic group, a_i is the pollution tolerance score for that taxonomic group, and N is the total number of organisms in sample. Macro invertebrates are given a numerical pollution tolerance score (a_i) ranging from 0 to 10. The family biotic index estimation value was shown table 2 below.

Table 2. Modified Hilsenhoff Biotic Index (source Mandaville 2002).

Biotic Index	Water Quality	Degree of Organic Pollution
0.00-3.50	Excellent	No apparent organic pollution
3.51-4.50	Very good	Possible slight organic pollution
4.51-5.50	Good	Some organic pollution
5.51-6.50	Fair	Fairly significant organic pollution
6.51-7.50	Fair poor	Significant organic pollution
7.51-8.50	Poor	Very significant organic pollution
8.51-10.00	Very Poor	Severe organic pollution

G) Shannon-Weaver Diversity Index (H)

The Shannon-Weaver Diversity Index (H) was evaluating the abundance of benthic macro invertebrates among the sites. This was calculated as:

$$H = -(\log_2 P_i)$$

Shannon-weaver diversity index also indicates the diversity of the species in the area. The richness and evenness of diversity also indicates the diversity.

Shannon-weaver diversity index (H)

$$H = -\text{SUM} [(p_i) * \ln (p_i)]$$

Where,

SUM = Summation

p_i = Number of individuals of species i /total number of samples

S = Number of species or species richness

H_{\max} = Maximum diversity possible

(N) is number of the species

$$H_{\max} = \ln (N)$$

$$E = \text{Evenness} = H/H_{\max}$$

3.6 Data Analysis

SPSS and Microsoft Excel sheet on computer were used to analyze data obtained from physico-chemical and biological parameters. The data was analyzed by using statically model like as significant difference between physical, chemical and macro invertebrate among the site. For site comparisons, the physico-chemical variables and benthic macro invertebrate community structures were analyzed for all of the three sampling sites. For all statistical tests, a probability of $P < 0.05$ was considered significant. Relationship between the environmental and biological (macro invertebrates) data were assessed. The chi-square was used to associate the physico chemical parameters and benthic macro invertebrates of the river.

4. RESULTS AND DISCUSSION

4.1 The Physico- Chemical Parameters of Rukkisa River Water Samples

4.1.1 pH

The average pH value was 6.6, 4.5, and 5 for the upstream, outlet of the effluent and downstream respectively. The mean value of the pH was high at upstream, decrease at outlet point and slightly increases at downstream. The standard error of the pH was high at the outlet point, slightly increases in downstream and almost small at the upstream point as shown in figure 2. The pH values of the impacted sites (outlet of the effluent and downstream) were not

permissible according to EPA (2003) standards for surface water (6.0-9.0) and WHO drinking water quality standards and standard for surface water (6.5-8.5).

The pH of the water is important because it affects the solubility and availability of nutrients and how they can be utilized by aquatic organisms. It also alters the ionic and osmotic balance of individual organism and determines the type of the chemicals of numerous elements and molecules found in water. Aquatic organisms are very sensitive to the pH of the aquatic environment because most of metabolic activities were pH dependent (Wang *et. al.*, 2002).

In the present study only reference site or upstream was within EPA (2003) standards for surface water (6.0-9.0) and within WHO drinking water quality standards and ambient standard for surface water for pH (6.5-8.5).

Comparable study was done by Solomon Endris *et al* (2008) in Jimma with in 11 river sampling sites and has got similar result. The pH of the lower stream of those rivers was lower and it was below the WHO standard range of (6.5-8.5) for human use. As the organic waste oxidized, CO₂ released and increase acidic characters then pH decreased. This indicates the waste water discharges mainly from coffee processing plant decrease pH which directly affects water quality as well as the distribution of the benthos.

Similarly Abebe Beyene *et al* (2011) at Mana and Gomma Wereda in Jimma Zone reported that coffee waste is known to lower the pH, and make acidic water (pH=4.5) that coincide with the peak coffee-processing season.

The lower pH of the outlet point and downstream along the river create high acidic condition which was in turn likely to be used by microorganisms and this can pose severe health problems among the residents of nearby community who use the downstream water for different purposes and this is in agreement with the previous study reports made by Hadis and Devi(2007) in Jimma zone.

All of the pH values of the effluent during the sampling period were below the effluent discharge limits. The low pH value were due to acidic discharges (organic wastes) resulting

from coffee processing plant. Thus, the effluent was acidic and has the potential to acidify the river water.

Apart from high organic content of coffee processing plant effluent, spent wash generated from the fermentation step also contains nutrients. The result indicated that small variation in pH was seen during first round and second round data collections between upstream, outlet point and upstream. These consent an accumulation of discharge of organic waste in to the river which brings change in pH. Naturally occurring fresh waters have a pH range between 6 and 9 EPA (2003) the concentration range suitable for the existence of most biological life is quite narrow and critical. Most fresh waters are relatively well buffered and more or less neutral.

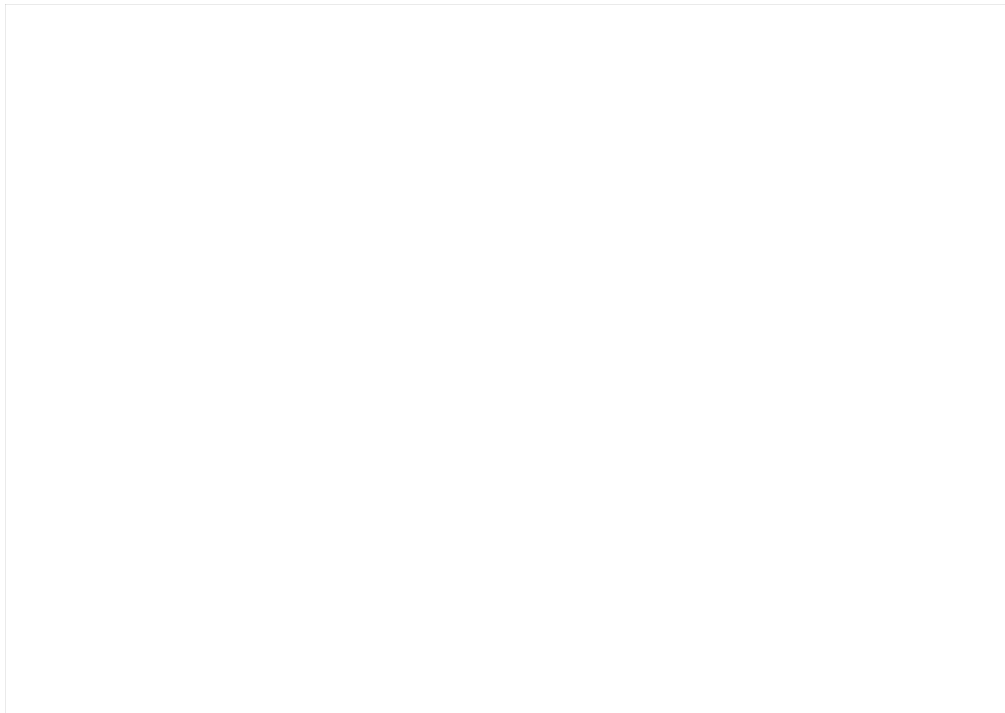


Figure 2. Mean pH value of rukkisa river water samples

4.1.2 Temperature

The result obtained was 22.05°C, 28°C and 27°C at upstream, outlet point and downstream respectively as shown in figure 3 and appendixes table 1. The mean temperature measured at the three sites upstream, outlet and downstream was not similar. The upstream site has lower temperature than that of the outlet point and downstream.

The downstream water temperature in particular (28°C and 27°C) was above the permissible range of WHO(2003) standard quality guidelines for discharging of effluents for irrigation purpose and allowed to enter into natural surface water bodies shown in the appendixes table 1. The results of temperature also disagree with the work of Hadis and Devi (2007) in Jimma zone. This may be happen because of climatic difference between the two rounds of sampling as well as selected area.

The mean value of temperature was low at the upstream, exceed at outlet and slightly decrease at downstream. The standard error of the temperature was almost little error at upstream exceeds at outlet and downstream (figure 3).



Figure 3. Mean temperatures of rukukisa river water samples

4.1.3 Biological Oxygen Demand (BOD)

The Figure 4 shows the average value and standard error of BOD. The BOD measured at the three sites upstream, at the outlet and downstream has variation. The BOD content of coffee processing plant effluent was 0.87 mg/l at the upstream, 290 mg/l at the outlet and 189 mg/l at the downstream, respectively. The mean value of the BOD was highly increased at the outlet and slightly decreases at downstream. The standard error of BOD was almost no error at upstream; the error highly exceeds at outlet and downstream.

The BOD result at the upstream was lower than that of the point of outlet and downstream. The outlet point of effluent had higher BOD than that of downstream this was happen because of the concentrated effluent discharge into the water. The result was similar to the work of Hadis and Devi (2007) in Jimma zone they observed BOD values up to 10,800.

The WHO (2003) standard for effluent discharges on land for irrigation and to receiving water has a limit value of 100 mg/l BOD, the maximum effluent concentration obtained from this analysis were higher than the acceptable limit and the reference samples, respectively indicating the pollution strength of the wastewaters as shown in appendix table 2. This indicates large amount of biological oxygen demanding substances in the effluent released from the coffee processing waste water released into the river. This also indicate there was low oxygen available for living organisms in the waste water when utilizing the organic matter present.

Comparable study was done in Jimma by Dejen Yemane (2015) at Gomma and Mana districts and got similar result 9048 mg/L. According to Tamrat Alemayehu *et al* (2006) the maximum BOD value recorded in the polluted river downstream from conventional wet coffee processing industries were 10,604 mg/L.

Alemayehu Haddis and Rani (2008), explained wet coffee processing effluents are complex furthermore; have several effects on water bodies and human health. The higher value were recorded at a point where conventional wet coffee processing effluents discharge into traditional waste water lagoon or pools. The value of the BOD observed was not permissible by WHO (2003) standard and has effect on living organisms.



Figure 4. Mean value of BOD of rukkisa river water sample

4.1.4. Turbidity

The Turbidity of the water samples among the upstream, outlet point and downstream was 5 NTU, 100 NTU and 84 NTU, respectively. The result indicates higher difference within sampling sites. Indicating the water quality deterioration in the river. The permissible value for drinking water was 5 NTU, where in line with water sampled from the reference sites or upstream. The other sites do not permissible limit of drinking water standard WHO (2003) shown the appendix table 2.

The mean value of the turbidity was highly increased at the outlet and slightly decreases at downstream. The standard error of turbidity was almost no error at upstream; the error highly exceeds at outlet and slightly decreases at downstream.

Coffee processing effluent solid causes the dissolved oxygen problem by sedimentation and forming oxygen demanding sludge deposit, which cause turbidity in the receiving water

and may alter the habitat of aquatic microorganisms. Similar work was done by Dejen Yemane *et al* (2015) in Jimma. The concentration of turbidity recorded by Tsigereda Kebede (2011) was similar with very high observed value of impacted sites. Turbidity of waste water indicates the quantity of suspended and colloidal material in the effluent (Tsigereda Kebede 2011).



Figure 5. Mean value of turbidity of rukkisa river water sample

4.1.5 Total Alkalinity

The mean value of total alkalinity measured at the upstream, outlet of effluent and downstream had different value (Figure 6). The value of Alkalinity found were 29 at upstream, 99 at the outlet point of effluent and 83 downstream respectively.

The mean values of the total alkalinity were highly increased at the outlet and downstream. The standard error of total alkalinity was almost no error at upstream; the error highly exceeds at outlet and downstream.

The Alkalinity result at the upstream was lower than that of the releasing point (outlet) and downstream. At the point of releasing the effluent has higher Alkalinity than that of downstream this was happen because of the concentrated effluent discharge into the water.

This consequence of effluent at the point of releasing was with low pH and increase Alkalinity. the maximum effluent concentration obtained from this analysis were higher than the acceptable limit and the reference sampling respectively indicating the pollution strength of the waste waters. This indicates the waste water discharges mainly from coffee processing plant increase total Alkalinity which directly affects water quality as well as the distribution of the benthos.

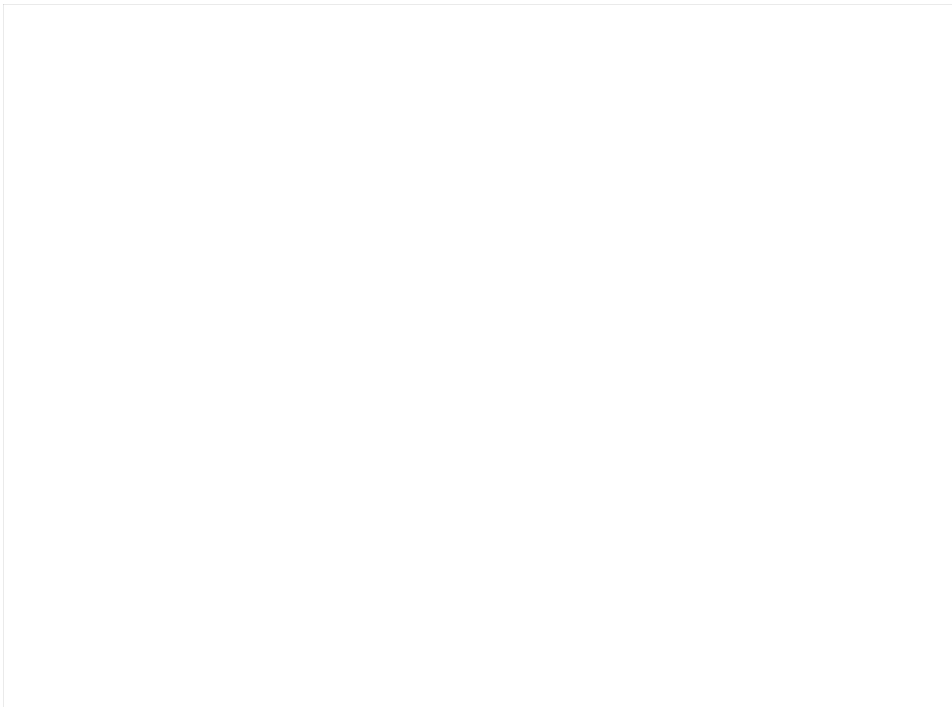


Figure 6. Mean value of total alkalinity of rukkisa river water samples

4.2 Identified Benthic Macro invertebrates Species in Rukkisa River Water Samples

In total, in the three sampling site of Rukkisa River there were 1144 benthic macro invertebrates collected and examined for detection of MBI, FBI, BI and H or diversity of the species. From these there were 13 orders, 19 suborder, 26 families, and 26 species. From those over all taxa 329 benthic macro invertebrates were found in upstream, 431 was at the point or outlet of effluent and 384 was in the downstream. The number and species identified was listed below with their order, suborder, family and species with their selected site upstream, outlet and downstream, respectively as shown in the appendix table 3.

A wide variation was observed in the abundance of MBI from site to site. The more diversity was observed in upstream were different benthos including, placopectera, Trichoptera, Megaloptera, nematode, Ephemeroptera and some clean water species of odonata were trapped. Odonata, Hemiptera, diptera and Coleoptera found in the wide range in the at the point of release and downstream (table 3).

The presence of some intolerant species in the upstream which include Trichoptera, Megaloptera, Placopectera and Nematode demonstrates the level of pollution. The existence of Coleoptera suborder Dytiscids species *Agabetes* in the upstream was that indicate the species live in all kind of water. However, the abundance blood red Chironomidae species in the downstream indicate the water was highly polluted Barbour *et al.*, (1999). Chironomidae species is very tolerant to the pollution this was the hemoglobin pigment that transports dissolved oxygen in the polluted water (Tyokumbur *et al*, 2002).

Similarly, Chironomidae species are among the tolerant groups of Diptera taxa with the red blood being more tolerant. Higher tolerance of the blood red Chironomidae is due to its pigment that helps the organism to get oxygen from the atmosphere hence the name “blood red” (Barbour *et al.*, 1999; Bouchard, 2004).

Another species Psychodidae order Diptera (rat tail maggot *Eristalis*) larvae which have tolerance value 10 was highly abundant in the releasing point and downstream indicates the water is polluted. These species use atmospheric oxygen with its long tubed tail. Because of the

shortage of the oxygen in the polluted water, these species send its tail out of the water and get air from the outside and sustain its life (Bouchard, 2004).

Table 3. Family biotic index, percentages and diversity Shannon value of macro invertebrates

No	Family	Species	T.value	upstream	At R. point	Downstream
1	Dragonfly nymph	Lestidae	5	2	53	34
2	Gomphidae	Ophiogomphus	1	42	-	-
3	Libellula	Libellulidae	9	-	21	18
4	Damselfly nymph	Ischnura 1	9	-	22	19
5	Diptera larvae	Simulium aureum	7	1	36	31
6	Diptera sails	Simulidae	7	2	34	42
7	Chironomidae larvae	Tribe Chironomidae	8	1	8	11
8	Chironomidae	Blood red Chironomidae	10	1	76	69
9	Psychoda	Syrphidae	10	1	5	4
10	Coleoptera larvae	Agabetes	5	12	71	57
11	Dytiscids	Agabetes	5	33	14	13
12	Hydrophilidae	Laccophilus	5	6	18	20
13	Bdellidae	Hirudinae	10	2	17	12
14	Micronectae	-	-	12	-	4
15	Nepidae	-	5	3	12	8

16	Notonectidae	-	5	2	14	18
17	Corixidae	Hesperocorixa	6	1	12	8
18	Anura	Rana angolensis	-	6	1	2
19	Nephan	Nectidae	5	4	1	1
20	Ephemerebellidae	Attenella attenuata	1	43	-	-
21	Dannella	Ameletus	0	64	-	-
22	Glossiphoniidae	Haplotaxidae	5	6	16	11
23	Dugesia	Planariidae	1	24	-	-
24	Taeniopterygidae	Taeniopteryx	2	16	-	-
25	Philopotamidae	Dolophilodes	0	28	-	-
26	Chauliodes	Corydalidae	0	22	-	-
Total		26	26	334	431	384
Total number of macroinvertebrates in whole sites						1149
BI				1.6108	6.8283	6.8802
%C				29	37.6	33.4
H				3.158	2.409	2.544

Where BI represent biotic index %, percentages H Shannon-Weaver Diversity Index

4.3 Distribution of Benthic Macro invertebrates of the Rukkisa River

4.3.1 The Benthic Macro invertebrates Metrics

The diversity and distribution of the benthic macro invertebrate species were estimated by calculating the Benthic Macro Invertebrate index such as Shannon weaver diversity index, Family biotic index, community loss index and evenness of species, MBIS, percent dominance and Ratio of EPT to Chironomidae (table 5).

According to Shannon –weaver index the result obtained in upstream, at the outlet point and downstream shows difference on water quality (table 3). The Shannon –weaver index obtained at the upstream was higher value than the other. The result obtained was (3.16) at upstream (2.409) at the point of outlet and (2.54) at the downstream (table 3). Gerritsen *et al.*, (1998) reported that the number and distribution of taxa (biotic diversity) within the community increases, so does the value of “H”).

The result of the study showed that the water body at the upstream had higher number of taxa than the other. The higher taxa distribution implies the community was within stable life which indicates the water was in the higher quality. Similarly Richard A. *et al* (2003) stated that diversity values may vary directly with water quality and low diversity may indicate an unstable community. headwater streams have high diversity and still represent excellent water quality. Mandaville (2002) said that the healthier the community is, the greater the number of taxa found within that community.

The Shannon-weaver index obtained from outlet point of effluent and downstream was lower than upstream. This suggested that instability of the community as explained by Richard A. *et al* (2003) that the diversity values may vary directly with water quality and low diversity may indicate an unstable of community. Homogeneity of diversity type in the impacted streams due to pollution, would lead to lower taxa accumulation, while taxa accumulation would increase in non-impacted sites with more diverse range of habitats (Plafkin *et al.*, 1989). The upstream taxa of the Rukisa River were very stable and have no disturbance from the industrial pollution.

Domination of similar taxa was high at the point of release (outlet) of effluent and downstream. The dominant taxa can survive with the stress the other taxa which was intolerant to the stress cannot survive and their number was decreased. Therefore the accumulation of the similar species at the outlet and downstream indicate the water of the river was with low quality.

Plafkin *et al.*, (1989) stated that the homogeneity of habitat type in the affected streams due to pollution, would lead to lower taxa accumulation, while taxa accumulation would increase in non-impacted sites with a far more diverse range of habitats. The “H” value of the river at the releasing point (outlet) was higher than that of the downstream because of high pollution level. The “H” value of the downstream was lower because of the dilution of river with the stream and taxa numbers slightly increase.

The richness of taxa also indicates water quality. The species richness (Hmax) of the Rukkisa River was 3.18,3 and 3.1 at upstream, point releasing (outlet) of the effluent and downstream respectively (Table 5). According to Richard A. *et al* (2002) high taxa richness was generally

associated with good water quality, low taxa richness does not necessarily indicate poor water quality. The Hmax of the upstream was greater than the other, suggesting the water quality at upstream is good quality as per the suggestion of Richard A. *et al* (2002).

The outlet of the effluent and downstream had low taxa richness, which indicates the poor water quality of the river in the site. The quality of water at the outlet of the effluent was poorer than the three. Which is associated to high effluent discharges occurred .The downstream had higher taxa richness than the outlet .That could be due to dilution of water body.

The evenness of the taxa indicates diversity and water quality. The result obtained was 0.99 at the upstream, 0.8 at the outlet of effluent and 0.82 at the downstream. The value obtained was decrease from the upstream to downstream. This indicates the evenness of the diversity decrease with water quality decrease.

The Macro invertebrates Biotic Index scores (MBI) also indicates the taxa richness of the stream. Nutrient-enriched water has a high density of organisms, while water polluted with toxic chemicals or silt or sand usually has a lower density (Hilsenhoff, 1988).

The Macro invertebrates Biotic Index scores (MBI) of taxa in a stream determines the presence or absence of taxa which have a high pollution tolerance. MBI values reflect stream quality (Hilsenhoff, 1977). The MBI value of the water quality was indicated as.

- 1, Less than 6.0 = good water qualityupstream.
2. 6.0 to 7.5 = fair water quality.....downstream.
3. 7.6 to 8.9 = poor water qualityreleasing point.
4. Greater than or equal to 9.0 = very poor water quality

The MBI values obtained from the Rukkisa river was as follow .The MBI of upstream was 5, 7.9 for outlet and 7.4 and downstream. The result indicated that MBI value of the upstream was less than 6 which indicates water at the upstream was in a good quality. However the value of the MBI of the releasing point (outlet) of effluent was 7.9 showing poor water qualities.

Likewise the MBI value of the downstream was 7.4 and this demonstrated the fair water quality related to poor water quality. The MBI value of Rukkisa River was increase from the upstream to downstream but the quality of water from the upstream to downstream be kept on decreasing (Table 5). This phenomenon is mainly related to the increasing number of the most tolerant taxa.

The FBI of the Rukkisa River was 1.61 for the upstream, 6.83 at the point of outlet of effluent and 6.88 at the downstream. Based on the values obtained indicated that the family biotic index of the upstream was low (table 5).

Hilsenhoff Family Level Biotic Index (H-FBI) was originally formulated to detect organic load of pollution (Hilsenhoff, 1988). The family-level index has been modified for the rapid bioassessment protocol II to include organisms other than just arthropods using the genus and species-level tolerance values adopted by the State of New York (Bode *et al.*, 1991, 1996, 2002). Although the FBI may be applicable for toxic pollutants, but it has only been evaluated for organic pollutants. The Biotic Index was subsequently modified to the family-level with tolerance values ranging from 0 (very intolerant) to 10 (highly tolerant) based on their tolerance to organic pollution (Mandaville, S.M. 2002).

This indicates the water at the upstream of the river was at high quality and there was no apparent load of organic pollution. The value of FBI at the outlet point and downstream was very high. It indicated that the water had fair poor quality and significant organic load of pollution. Presence of numerous families of highly tolerant organisms usually indicates poor water quality (Hynes, 1998). These results occurred because the more intolerant genera and species in each family predominate in clean streams, while the more tolerant genera and species predominate in polluted streams.

Table 4. Hilsenhoff Biotic Index with respect to Degree of Organic Pollution

Biotic Index	Water Quality	Degree of Organic Pollution	
0.00-3.50	Excellent	No apparent organic pollution	Upstream
3.51-4.50	Very good	Possible slight organic pollution	

4.51-5.50	Good	Some organic pollution	
5.51-6.50	Fair	Fairly significant organic pollution	
6.51-7.50	Fair poor	Significant organic pollution	Outlet and downstream
7.51-8.50	Poor	Very significant organic pollution	
8.51-10.00	Very Poor	Severe organic pollution	

The percent composition EPT Index or the Ephemeroptera, Placoptera, and Trichoptera (EPT) index shows the taxa richness within these groups which are considered to be sensitive to pollution. Their number should increase with increasing water quality (Mandaville, 2002). Initially developed for species-level identifications; this index was valid for use at the family-level (Plafkin *et al.*, 1989).

The percentage composition of Ephemeroptera, Placoptera and Trichoptera at the upstream constituted 35.2% of all macroinvertebrates of upstream. This result showed that the water had high water quality. These three macro invertebrates have low tolerance value between 1 and 0 (Bode *et al.* 1996). So the presence of these three species indicates there was no water pollution at the upstream of the Rukkisa River.

While the number of those three (Ephemeroptera, Placoptera and Trichoptera) macro invertebrates in the downstream and at the point of releasing was lower than upstream. They compose the 6.69% at the outlet point and 1% at the downstream.

This was happening these three species (Ephemeroptera, Placoptera and Trichoptera) was intolerant to water pollution. The water at the point of release and downstream was polluted or low water quality. The absences of those three (Ephemeroptera, Placoptera and Trichoptera) species also indicate the water was highly polluted by effluent shown (Table 5).

In the current study the ratio of EPT to Chironomidae was 25, 0.03 and 0.025 at the upstream, outlet point and downstream, respectively (Table 5). The high ratio at the upstream of the river indicates low levels of water pollution. Ephemeroptera, Placoptera and Trichoptera (EPT) were present only at the upstream. Members of EPT are considered very sensitive to environmental stress, their abundance at upstream sites shows comparatively clean

environment (Armitage *et al.* 1983), hence EPT showed to be the possible bioindicator of unpolluted ecosystem.

The Ratio of EPT to Chironomidae abundance shows the number of individuals from sensitive orders compared to pollution tolerant (Chironomidae) species. The abundance of EPT and Chironomidae indicates the balance of the community, since EPT are considered to be more sensitive and Chironomidae to be less sensitive to environmental stress. A community considered to be in good biotic condition will display an even distribution among these four groups, while communities with disproportionately high numbers of Chironomidae may indicate environmental stress (Plafkin *et al.*, 1989).

The Ratio of EPT to Chironomidae at the point of effluent release was lower than the Ratio of EPT to Chironomidae at the upstream. Typical Ephemeroptera species of headwater stream in the Vosges Mountains were fully eradicated when the pH dropped (Francois Guerold *et al.*, 2000). Similarly in the current study the decreasing of those species in the outlet point and downstream could be associated with the drop of pH value in these two points Rukkisa river. The Ratio of EPT to Chironomidae at the downstream also low and indicate the water has low quality (Table 5). The finding showed that the water at the releasing point and downstream at low quality.

The percent dominance of Chironomidae obtained from the Rukkisa river was 0.6%, 19.5 and 20.8% at the upstream, outlet of effluent and downstream respectively. The blood red Chironomidae species has tolerance value of 10 (Bode *et al.*, 2002).

The lower the domination of Chironomidae at the upstream indicates the higher quality of the river. The domination of Chironomidae at the releasing point was increase due to the increasing of the pollution but not increase as the downstream.

The percent dominance (%DF) of Chironomidae and Diptera family indicates the abundance of those species. The Percent Contribution of Dominant Family (%DF) equals the abundance of the numerically dominant family relative to the total number of organisms in the sample. According to Plafkin *et al.*, (1989) community dominated by relatively few families would have

a high %DF value, thus indicating the community is under the influence of environmental stress.

Chironomidae are also considered to be capable of building up huge population quickly and tolerating sudden changes in the habitat conditions (Solimini *et al.* 2003). The domination of Chironomidae at the downstream was increase due to the increasing pollution. According to Klemm, D.J *et al* (2002) high degrees of dominance by few tolerant taxa imply lowered diversity and increased disturbance in the water.

The domination of diptera was 1.7% at upstream which indicates the water is in high water quality. While, domination of diptera at the releasing point of effluent was 23% implying the increasing in the pollution level. The percent of domination of diptera at the downstream was 41%. This indicates increasing pollution at the downstream. The percent dominance (%DF) of diptera shows water pollution, because the tolerance value of the diptera is high (Plafkin *et al.*, 1989).

The percent composition of order Odonata was 13% at the upstream, 22.3% at the point of releasing effluent 18.5% at the downstream. There were four odonata species found; damselfly, dragonfly, Libellulidae and Gomphidae within two suborder Anisoptera and Zygoptera. From those four only Gomphidae has its low tolerance value. The percent composition of Odonata at the upstream was lower than the two remaining points indicates upstream has high water quality. While, percent composition of the Odonata species was increase from upstream. The Odonata species has high tolerance value and can survive stress (Plafkin *et al.*, 1989).

The percent composition of the Odonata species increased at upstream and had slight deference b/n the point of release and downstream. This phenomenon indicates the water quality deteriorates as moved from upstream to downstream of the river.

To determine the degree of dissimilarity as well as loss of benthic taxa, community loss index was used. The degree of dissimilarity was 1.14 at the releasing point of the effluent and 1.12 at the downstream (Table 5). Dissimilarity between taxa from site to site indicates loss of taxa or community loss index (CLI). Community loss index determines the loss of taxa in a study site with respect to a reference site. The Community loss index of the upstream was not calculated,

since of the upstream was the reference site for comparison and estimated as no community loss.

According to Mandaville, (2002) Community Loss Index tends to decrease with increasing water quality to maintain healthy aquatic ecosystem. Lower scores at the downstream were associated with high CLI which indicates higher ecosystem disorder at the downstream and the point of outlet of effluent. Due to the difference in their tolerance to pollution, the presence or absence of certain macro invertebrates can provide valuable information on stream quality. Therefore, the water quality at the outlet point and downstream was low quality or polluted.

Table 5. Biological Indexes value obtained from the rukkisa river water quality manage

Indexes	Upstream	Outlet(releasing)	Downstream	Towards water quality
FBI	1.6108	6.8283	6.8802	Decrease
H	3.158	2.409	2.544	Increase
MBI	5	7.9	7.4	Decrease
%DF	0.6	19.5	20.8	Decrease
Chironomidae				
% DF diptera	1.7	23	41	Decrease
% CEPT	35.2	0.69	1	Increase
% odonata	13	22.3	18.5	Decrease
RatioEPT	25	0.03	0.025	Increase
		1.14	1.12	Decrease
Hmax	3.18	3	3.1	Increase
E	0.99	0.8	0.82	Increase

4.4 Correlation Analysis between Physico-Chemical Parameters and Benthic Macro invertebrate's Indexes

The relationship between the physico-chemical parameters and benthic macro invertebrates indexes of Rukkisa River was examined using the Pearson's correlation analysis, and the results are shown in the correlation matrix (Table 6).

The BOD value was correlated with temperature turbidity total alkalinity and macro invertebrates indexes, the value observed was negatively correlated with pH, Shannon-weaver diversity index, %CEPT percent composition of Ephemeroptera ,Placoptera and Trichoptera,

rEPT ratio of Ephemeroptera ,Placoptera and Trichoptera ,Hmax diversity richness and E evenness with ‘r’ (r=-.989),(r=-.774),(r=-.938),(r=-.935),(r=-.975) and(r=-.897) respectively.

The pH value was negatively correlated with all benthic macro invertebrate’s indices and only positively correlated with Shannon-weaver diversity index , percent composition of Ephemeroptera ,Placoptera and Trichoptera, ratio of Ephemeroptera ,Placoptera and Trichoptera ,diversity richness and evenness with ‘r’ value r=.670,r=.980, r=.978,r=.930 and r=.953 respectively.

Total alkalinity correlated positively with macro invertebrate’s biotic index score, percent domination of Chironomidae, percent domination of diptera, percent composition of odonata and community loss index with ‘r’ value r=.998,r=.962,r=.768,r=.980 and r=.979 respectively.

Total alkalinity correlated negatively with all other value. Turbidity negatively correlated with Shannon-weaver diversity index, percent composition of Ephemeroptera, placoptera, ratio of Ephemeroptera, placoptera and Trichoptera diversity richness and evenness with ‘r’value r=-.707,r=-.968,r=-.966,r=-.947 and r=-.937 respectively (Table 6).

Temperature correlated positively with biotic index score, percent domination of Chironomidae, percent domination of diptera, percent composition of odonata and community loss index with ‘r’ value r=.942,r=.846,r=.557,r=.996 and r=.883 respectively.

Shannon-weaver diversity index correlate significantly with only pH ‘r’value r=.670 from physico chemical parameters otherwise negatively (Table 6).

It also correlated positively with percent composition of Ephemeroptera, placoptera and Trichoptera, ratio of Ephemeroptera, placoptera and Trichoptera, diversity richness and evenness with ‘r’ value r=.507, r=.500, r=.896 and r=.415 respectively otherwise negatively correlate with others.

The diversity richness and evenness insignificantly correlated with pH, Shannon-weaver diversity index percent composition of Ephemeroptera, placoptera and Trichoptera and ratio of Ephemeroptera, placoptera and Trichoptera only. Community loss index negatively correlated with pH, Shannon-weaver diversity index, percent composition of Ephemeroptera, placoptera

and Trichoptera and ratio of Ephemeroptera, placopectera and Trichoptera only. With 'r' value $r=-.981$, $r=-.513$, $r=-.1$ and $r=-1$ respectively (Table 6).

Table 6. Pearson's Correlation (r Values) between the Physico-Chemical Parameters and benthic macro invertebrates 'biotic index

Parameters	BOD	pH	T.A	Turb	Temp	FBI	H	MB	%D	%	%	%	R.	
				b	p			I	F	DF	CE	odo	EP	
									Ch	dip	PT		T	
BOD	1													
Ph	-.989	1												
T.A	.990	-1	1											
Turb	.995	-.999	.999	1										
Temp	.999	-.957	.960	.971	1									
FBI	.932	-.976	.974	.964	.871	1								
H	-.774	.670	-.677	-.707	-.856	-.493	1							
MBI	.980	-.999	.998	.995	.942	.985	-.633	1						
%DF Chiro	.913	-.964	.962	.950	.846	.999	-.449	.976	1					
% DF dip	.670	-.774	.768	.741	.557	.893	-.048	.804	.914	1				
% CEPT	-.938	.980	-.978	-.968	-.879	-1.	.507	-.988	-.998	-.886	1			
% odo	.998	-.987	.980	.988	.996	.910	-.809	.967	.889	.627	-.917	1		
R.EPT	-.935	.978	-.976	-.966	-.875	-1.	.500	-.987	-.998	-.889	1.	-.914	1	
	.941	-.981	.979	.970	.883	1.	-.513	.989	.997	.882	-1.	.920	-1.	1
Hmax	-.975	.930	-.933	-.947	-.997	-.827	.896	-.911	-.799	-.486	.837	-.986	.832	-.841
E	-.897	.953	-.951	-.937	-.825	-.996	.415	-.967	-.999	-.929	.995	-.871	.995	-.994

4.4.1 Pearson's significant Differences (P) Value between Physico-Chemical Parameters Benthic Macro invertebrates

There was significant difference between the value of pH obtain from the rukkisa river and Shannon-weaver diversity index with $p=0.05$. The value of pH was decrease with increasing pollution and also Shannon-weaver diversity index decrease with increasing pollution. The value of family biotic index was significantly different with community loss index and percent composition of Chironomidae with $p =0.05$.

The value of pH was insignificantly different with macro invertebrate biotic index score with $p=0.05$. The pH was insignificantly different with Turbidity with $p=0.01$ and has no association. Family biotic index insignificant deference with ratio of the Ephemeroptera, placoptera and Trichoptera with $p=0.01$ there was no association. The value of family biotic index was insignificant with percent composition Ephemeroptera, placoptera and Trichoptera with $p=0.01$.

Community loss index was insignificant deferent with percent composition Ephemeroptera, placoptera and Trichoptera and ratio of the Ephemeroptera, placoptera and Trichoptera with $p=0.01$ without association. Evenness of the macro invertebrates insignificant deference with percent composition of Chironomidae with $p =0.05$ and Biological oxygen demand significantly deferent with percent composition of odonata.

Total alkalinity was significantly different with macro invertebrate biotic index score and Turbidity with $p =0.05$. Percent composition of Chironomidae insignificant deference with percent composition Ephemeroptera, Placoptera and Trichoptera with $p =0.05$. Ratio of the Ephemeroptera, Placoptera and Trichoptera was insignificant deferent with community loss index and evenness of the macro invertebrates with $p =0.05$.

Physico chemical parameters highly associated with the pollution tolerant species but with pollution sensitive species it associate low. The family biotic index value obtained from rukkisa river was significantly associated with all physico chemical parameters obtained from rukkisa

river with $p > 0.05$. Shannon-weaver diversity index highly associated with all physico chemical parameters obtained from Rukkisa River with $p > 0.05$. The value of macro invertebrate biotic index score was associated with BOD, turbidity and temperature but not associated with pH and total alkalinity.

Percent composition of Chironomidae and percent composition of diptera was highly associated with all physico chemical parameters obtained from Rukkisa River. But the percent composition Ephemeroptera, Placoptera and Trichoptera slightly associated with physico chemical parameters obtained from Rukkisa River. Percent composition of odonata associated with physico chemical parameters obtained from Rukkisa River except BOD. Ratio of the Ephemeroptera, Placoptera and Trichoptera, community loss index, richness of taxa and evenness associated with all physico chemical parameters obtained from rukkisa river. Temperature was highly associated with community loss index, ratio of the Ephemeroptera, Placoptera and Trichoptera and evenness taxa.

5. SUMMARY, CONCLUSIONS AND RECOMMENDATION

5.1 Summary

This study focused on the effects of coffee processing plant effluent discharge on Rukkisa River and water quality. Rukkisa River was addressed one of the coffee processing plant Effluent discharged poor water quality that does not meet the predetermined minimum requirement for discharge in to surface water. The quality of this river, the reference site was found at the upper stream location, the second site was located at the discharged point (outlet) and the third site was found downstream location. Benthic macro invertebrate's species were collected by using D-net; they were identified by using compound binocular microscope and sorted at species level.

Totally 1149 individual organisms were identified 334 benthic macro invertebrates were collected from upstream, 431 and 384 benthic macro invertebrates were collected from outlet and downstream respectively. Abundance and distribution of species were measured from the three study sites. This was done by using Indices like, Shannon -Weaver diversity indices (H), Family biotic indices (FBI) , community loss indices (CLI), macro invertebrates biotic index score (MBI),percent composition of taxa (%C), percent domination of dominant species (%DF), richness of taxa(Hmax) and Evenness of taxa (E). The abundance and distribution of benthic macro invertebrates species were vary among upstream and impacted (outlet & downstream). The numbers of taxa were higher in upstream than impacted sites. Twenty four different types of species were identified from upstream twenty and twenty one species from impacted (outlet & downstream).

Physico chemical parameter used to measure the qualities of Rukkisa River they were; pH, Temperature, Total alkalinity, Turbidity and BOD. The physical characteristics like, PH, Temperature and turbidity of the River were measured in the study site where as the BOD of the River were measured in the Laboratory. Most of the physico-chemical parameter were higher in the impacted site (outlet & downstream) than reference site (upstream) and are significantly correlated with Biological parameters.

5.2 Conclusion

Coffee processing industries have constructive impacts on economic development of the country and the neighboring group of people. However they have unenthusiastic impact on the aquatic environment: macro invertebrates' depletion or degradation of biodiversity and disappearance of sensitive taxa in the downstream was broadened. Although coffee processing is one of the seasonal agricultural activities performed immediately after the big rainy season during the months of September to December, its impact on nearby fresh waters was more persistent. This was due to the disposal of waste water to the nearby rivers. Waste water discharged to the nearby rivers has enormous effect on the degradation of the ecosystem. Both physicochemical and biological results exposed ecological destruction of outlet and downstream sites of river due to direct discharge of high organic waste from coffee processing industries into the rivers. Physico-chemical parameters measurements showed that the concentrations of oxidizable organic materials from coffee waste caused nearly complete deoxygenating the rivers and swept out the pollution-sensitive taxa. This improves what is indicated in the results of macro invertebrates, which show the effect. The diversity indices were able to capture water quality impairment during the peak coffee processing season.

In the coffee processing time, macro invertebrate diversity was significantly different between impacted and unimpacted sites. The ratio of pollution sensitive taxa was decrease as coffee processing time. The study showed the effectiveness of the benthic macro invertebrate protocol in assessing River pollution condition. Therefore, environmental agencies and researchers have a good option of using macro invertebrate with the objective in assessment and monitoring programs of Rivers.

5.3. Recommendations

- The authority should be designed and constructed to decrease the points pollution of Rukkisa river water.
- Environmental practices should approve and encouraged in the coffee-growing regions of Ethiopia.
- Local authorities should take critical actions to improve the ecological quality of these rivers as part as the efforts to restore their ecology and relieve public health risks.
- Proper disposal of waste water should be improved. The waste water should be disposed after treatment or the pollution effect is reduced.
- Waste water treatment (pond system) has to be treated before it is discharged to the nearby Rivers such as ponding systems using wetlands. The Waste water treatment should be constructed.
- Many approaches have been tested in different coffee producing countries to get the pollution load under control. These approaches should be improved in the coffee-growing regions of Ethiopia.

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7. APPENDICES

Appendix Table 1. Statistical analyzed mean value of physico-chemical parameters temperature (0C) and turbidity (NTU) all others measured in mg/L.

Parameters	Upstream	Releasing (outlet) point	Downstream	WHO max(2008)	EPA standard (2003)
PH	6.6± 0.20	4.55±1.00	5.00 ±0.10	6.5-8.5	6-9
Temperature	22.05±0.45	28.15±0.75	25.2 ±0.30	18-25 ⁰ c	20-25 ⁰ c
Turbidity	5.5±0.25	392±3.00	288.5±2.50	-	-
Total Alkalinity	29.0± 1.00	99±1.00	83±1.00	-	-

BOD	0.73±0.10	290±1.00	186±0.75	50-100	100
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Appendix Table 2. WHO (1995) permissible limits for the treated effluents to be discharged for irrigation

S.N	Parameters	WHO permissible limits
1	pH	6.5–9
2	Temperature(°C)	20-25
3	Total Alkalinity	-
4	BOD(mg/l)	100
5	Turbidity	300
6	code(mg/l)	300

Haddis, R. Devi (2008).

Appendix table 3. Identification of Benthic Macroinvertebrates Species in the Rukisa River at the Three Sites

No	Order	Suborder	Family	Species	T.valu e	Upstream	Relea point
1	Odonata	Anisoptera	Dragonfly	Calopterygidae	5	2	53
			Gomphidae	Ophiogomphus	1	42	-
			Libellula	Libellulidae	9	-	21
			damfly	Ischnura 1	9	-	22
2	Diptera	Diptera Simulidae	Diptera larvae	Simulium aureum	7	1	36

			Diptera sails	Simulium	7	2	34
		Blood-red Chironomidae	Chironomidae larvae	Tribe Chironomini	8	1	8
			Cyronomedae	Chironomus	10	1	76
		Syrphidae	Psychodidae	Syrphidae	10	1	5
3	Coleoptera	Dytiscids	Coleoptera larvae	Agabetes	5	12	71
			Dytiscids	Agabetes	5	33	14
			Hydrophilidae	Laccophilus	5	6	18
4	Annelida	Hirudinea	Leech	Bdellidae	10	2	17
5	Hemiptera	Heteroptera	Water measurer		-	12	-
		Ranatra	Nepidae		5	3	12
		Notonecta	Notonectidae		5	2	14
		Corixidae	True bugs	Hesperocorixa	6	1	12
6	Anura	-	-	Rana angolensis	-	6	1
7	Gadiformes	Guttigadus	-	-	-	4	1
8	Ephemeroptera	Drunella	Ephemerellidae	Attenella attenuata	1	43	-
			Mayflies	Ameletus	0	64	-
9	Oligochaeta	Haplotaxidae	Aquatic Worms	Haplotaxidae	5	6	16
10	Nematode	Trichodida	Dugesia	Planariidae	1	24	-
11	Placoptera	Stonefly	Taeniopterygidae	Taeniopteryx	2	16	-
12	Trichoptera	Caddisflies larvae	Philopotamidae	Dolophilodes	0	28	-
13	Megaloptera	Fishflies (hellgrammites)	Dobsonfly larvae	Corydalidae	0	22	-
Total	13	19	26	26			
Total number of macro invertebrates in each sites						334	431
Total number of macroinvertebrates in whole sites						1149	

Appendix table 4. Tolerance Values for macroinvertebrates for application in the Modified Family Biotic Index and other metrics

Bode *et al.*, (1996); Hauer and Lamberti, (1996); Hilsenhoff, (1988); Plafkin *et al.*, (1989)

macro invertebrates	T. Values	macro invertebrates	T. Values
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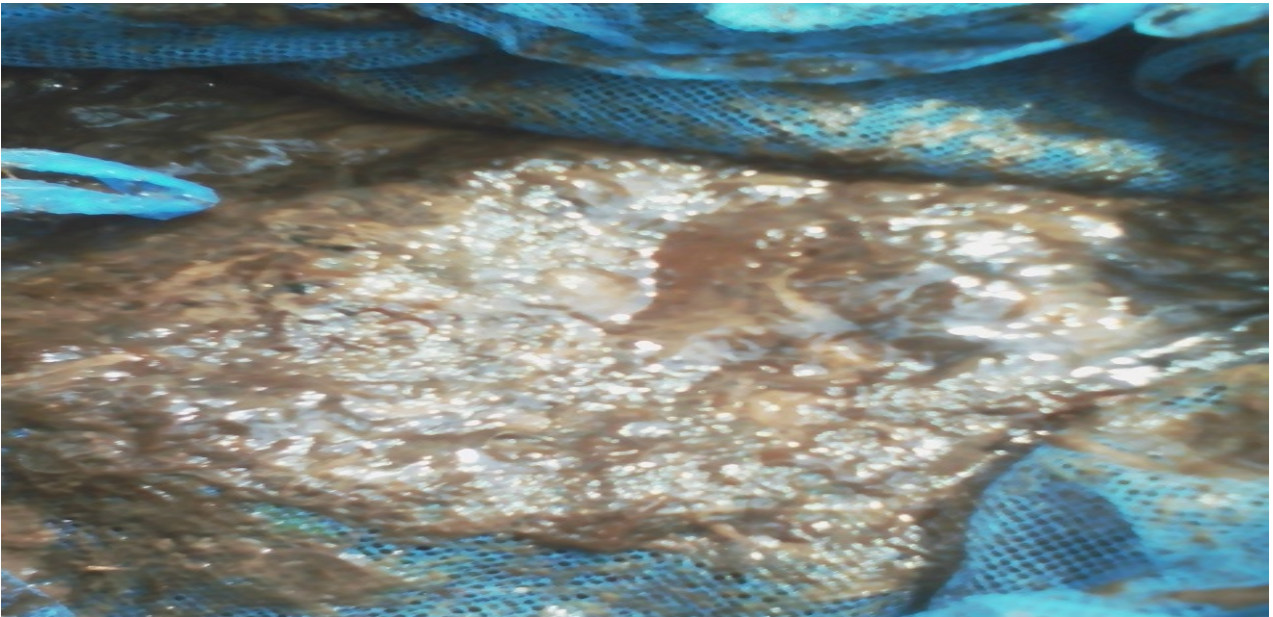
Ephemeroptera (Mayflies)	0	TRICHOPTERA (caddisflies)	3
Ameletidae	0	Psychomyiidae	2
Ephemerellidae	1	Dipseudopsidae	5
Dannella sp.	2	Philopotamidae	0
		Dolophilodes	0
		Glossosomatidae	0
ODONATA(dragonflies and damselflies)		DIPTERA(Two-winged or” true flies”)	
Gomphidae	0	Tipulidae (crane flies)	3
Ophiogomphus sp	1	Psychodidae (moth flies)	10
Libellulidae	9	Dixidae (dixid midges)	1
Dragonfly nymph	5	Diptera simulidae	7
dam fly nymph	9	Tribe Chironomini	8
Lestidae sp.	9	Blood-red Chironomidae	10
		Syrphidae	10
		Simuliidae (black flies)	6
		Tabanidae (horse and deer flies)	6
HEMIPTERA (water or true bugs)		Annelida (true worms)	
Corixidae	5	Hirudinea	10
Nepidae(Water scorpions)	5	Helobdella	10
Notonectidae	5	Sabellidae	6
Micronectae	5	Lumbricina	6
True bugs	6	Tubificidae	10
PLECOPTERA (stoneflies)		Anura (Amphibian)	
Taeniopterygidae	2	Tadpole	
Leuctra ferruginea	0	Gammaridae	4
Nemouridae	2	Hyaellidae	8
Chloroperlidae	1	Talitridae	8
Perlodidae	2		
COLEOPTERA (beetles)		Gadiformes	
Dytiscids	5	Guttigadus (Frog)	
Hydrophilidae	5		
Coleoptera	5		
Agabetae	5		
Gyrinidae	4		
Elmidae	4		
MEGALOPTERA(Corydalidae (fishflies, dobsonflies)	0	Oligochaeta(Aquatic Worms)	8
Sialidae (alderflies)	4	Haplotaxidae	5
Chauliodes sp	4	Undetermined Haplotaxidae	10
Nematode Planariidae	1	Glossiphoniidae	8
Tubellaria	6	Gastropoda(snails and limpets)	7



Appendix Figures 1. Upper shed of Rukkisa River.



Appendix Figures 2. Sampling of benthic macro invertebrates.



Appendix Figures 3. Pollution of downstream water and it's grubby.



Appendix Figures 4. Dumping area of the sluge.



Appendix Figures 5. Sorting the benthic macroinvertebrates in Hawassa fisher lab.

