



EFFECT OF HUMAN DISTURBANCE ON BENTHIC MACROINVERTABRATE ASSEMBLAGES  
AND PHYSICOCHEMICAL WATER QUALITY PARAMETERS OF TEMSA RIVER, AGARO  
TOWN, SOUTH WEST ETHIOPIA

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Jimma, Ethiopia

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TOWN, SOUTH WEST ETHIOPIA

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November, 2019

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## Approval sheet

As thesis research advisors, we hereby certify that we have read and evaluated this thesis prepared under our guidance by Desta Hordofa entitled as “ Effect of human disturbance on benthic macroinvertebrate assemblages and physico chemical water quality parameters of Temsa River, Agaro town, South west Ethiopia”. We recommended that it could be submitted as fulfilling the thesis requirement.

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As member of the board of examiners of the Msc. thesis open defense examination, we certify that we have read and evaluated the thesis prepared by Desta Hordofa and examined the candidate. We recommend that the thesis be accepted as fulfilling the thesis requirement for the degree of Master of Science in Environmental Science and Technology.

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**External Examiner**

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**Date**

## **Declaration**

I declare that this research paper is my own original work and has not been presented for a degree or other award in any other university and that all sources of materials used for the research paper have been correctly acknowledged.

### **Declared by:**

Name: Desta Hordofa

Signature: \_\_\_\_\_ Date: \_\_\_\_\_

## **ABSTRACT**

**Background:** Rivers are the most productive ecosystems in the world and have values for the fulfillment of human needs and ecological functions. However, different human activities taking place near to the river is posing risk (deterioration) to the ecological and physicochemical quality of the river. Though, increasing impact of human disturbances on the freshwater bodies of Ethiopian rivers calls for efficient and continuous monitoring based on physicochemical river quality parameters and ecological health assessment.

**Objective:** To assess the effect of human disturbances on benthic macroinvertebrate assemblages and water quality of Temsa river, Agaro town, south west Ethiopia.

**Methods:** A cross-sectional study was carried out on April, 2019 in the dry season. Macroinvertebrates and water samples were collected from thirteen sampling sites along the river. The samples were collected in a polyethylene bottles and transported to the laboratory by using ice box. Samples were analyzed for different physicochemical parameters and various biological metrics. Multivariate data analysis was used to examine the overall relationship among physicochemical parameters and macroinvertebrate assemblages with Principal component analysis, using software program Paleontological Statistics software package for education and data analysis (Past 3.18) version 1.0.0.

**Findings:** A total of 603 macro-invertebrates belonging to 41 families and 12 orders were recorded. From biological metrics Biological Monitoring Working Party score, Shannon diversity index, Ephemeroptera, Odonata and Trichoptera family richness and total family richness portrayed a clear pattern of decreasing with increasing in human disturbance; whereas, family biotic index score, which is an indicator of organic pollution, increased with increasing in human disturbance. Among the physicochemical variables, dissolved oxygen, electrical conductivity, chemical oxygen demand, phosphate and nitrate show significant variation between sampling sites ( $p < 0.05$ ). Macroinvertebrate metrics, biotic indices biological monitoring working parties and family biotic index showed significant variation at all sampling sites.

**Conclusions:** In conclusion, human activities in and around the river such as farming, solid waste dumping and effluent discharges contribute to the degradation of water quality and decreasing in the macroinvertebrate richness and diversity along the course of the river. Physicochemical parameters (nitrate, phosphate, ammonia) and human disturbances predominantly affecting the macroinvertebrate assemblages in the river study.

**Key words:** Benthic macroinvertebrates, human disturbance, water quality, metrics

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## **Abbreviations and acronyms**

ANOVA	Analysis of variance
APHA	American Public Health Association
BIBI	Biological Index of Biotic Integrity
BMI	Benthic Macro Invertebrates
BMWP	Biological Monitoring Working Party
CCA	Canonical Correspondence Analysis
COD	Chemical Oxygen Demand
EOT	Ephemeroptera, Odonata and Trichoptera
FBI	Family level Biotic Index
FFG	Functional Feeding Groups
PCA	Principal Correspondence Analysis
PAST	Paleontological Statistics software
TDS	Total dissolved solids
TH	Total Hardness
TP	Total Phosphate
UNESCO	United Nations Educational, Scientific and Cultural Organization
WHO	World Health Organization

## UNIT ONE: INTRODUCTION

### 1.1. Background of the study

Water is a fundamental and life-sustaining natural resource which is critical for the survival of all living organisms, food production and economic development (Berihun *et al.*, 2017). Water is essential for the development and maintenance of the dynamics of every aspect of the society (UNCSD, 2012). Water can be obtained from two principal natural sources: Surface water such as fresh water, lakes, rivers, streams, etc. and ground water, such as bore hole water and well water (Tadesse *et al.*, 2018). The benefits of rivers are not limited to the supply of water; they are also used for other purposes such as recreation and sport, fishing, navigation, irrigation, hydropower generation, transportation, waste disposal, and even sand mining (Mohamed *et al.*, 2015). Rivers ecosystem has a great ecological value, but their special typology makes them fragile and exposed to environmental changes, especially those related to disturbances of anthropogenic origin, which often implies irreversible degradation of their biota (Beasley &Kneale, 2003, Dahl *et al.*, 2004).

River ecosystems maintain diverse fauna communities, with species that are highly adapted, but also very vulnerable to changes in the river, both water quality and river structure. Even though they have a major role in sustaining the lives and livelihoods of many people, freshwater ecosystems are the most threatened ecosystems in the world (Dudgeon *et al.*, 2006). Furthermore, freshwater systems are a habitat to about 6% of the world's total known species, 40% of the global fish species and 25% of all vertebrate species (Dudgeon *et al.*, 2006; Dudgeon, 2010). In Ethiopia, stream and river waters are used for domestic consumption, agriculture production, industrial purposes, generating electricity, recreation, fish production and birds of great tourism attraction as well as several other species (Amare, 2017).

Ethiopia is gifted with many streams and rivers that comprise diverse aquatic ecosystems of great scientific interest and economic importance. The rapid population growth, expansion of urban and suburban areas, industrialization, land use change, and removal of riparian vegetation have resulted serious ecological problems on surface water resources (Aschalew, 2014). However, variety of stress (e.g. Water and sand abstraction, catchment and river bank degradation, reservoir flushing, diversion, etc.) cannot be detected and water management decisions may suffer under too little knowledge of the environmental consequences. Getachew Beneberu,

(2013) showed that degradation of streams and rivers in urban areas is increasing at an alarming rate because of rapid human population increase and associated waste production. Rapid population growth, urbanization and industrial development have been adversely degrading the environment by their effect through loss of biodiversity and pollution from wastes. Aquila *et al.*, (2011) and Aschalew (2014) stated that deforestation in the upstream of rivers, erosion, sedimentation, different agricultural activities; industrial and domestic waste, diversion and water abstraction are described as the threats for Ethiopian rivers and streams. These activities cause a detrimental impact on the total ecosystem ranging from deteriorating water quality to the partial or total destruction of river biota.

In order to understand the status of water quality and reduce pollution rate of waterways (streams and rivers), the knowledge about the health status of aquatic environment including their biodiversity is important. This can be done using various established bioindicators of water quality (Ojija and Laizer, 2016). Kripa *et al.* (2013) define bioindicators as a species or group of species that readily reflects the biotic or biotic state of an environment, represents the impact of environmental change on a habitat, community or ecosystem, or is indicative of the diversity of a subset of taxa, or the whole diversity, within an area. Among these bioindicators, the most frequently used are the benthic macroinvertebrates (Dickens and Graham, 2002; Elias *et al.*, 2014; Kaaya *et al.*, 2015).

Macroinvertebrates are organisms that are large enough to be seen with the naked eye and without vertebral column, which are most abundant and diverse group of animals found in fresh water, which includes flies, snails, mussels, worms, nematodes and crustaceans. Mac Neil *et al.* (2002) have recognized the concept that macro invertebrate families are very diverse, sensitive and suitable for assessment of severity of contamination of water pollution. Benthic macro invertebrates are the most preferred group in biomonitoring studies of fresh waters. This preference is due to their limited habitat and less moving ability; consequently, they cannot change their habitats quickly. Their life cycles are also long enough to understand what the differences are in their habitats before and after the pollution. All these reasons make the benthic macro invertebrates most favorable as biomonitors among the other group (Rosenberg & Resh, 1993).

An increase or decrease of macroinvertebrate population in a water body indicates pollution, the presence of stress factors and damage to the ecosystem (Ganguly *et al.*, 2018). Hardoy *et al.*, (2001) indicated that river pollution from city-based industries and untreated sewage can lead to serious health problems in settlements downstream. Benthic macro invertebrates are among the most diverse and abundant organisms in freshwater systems and are key for aquatic ecosystem functioning (Dalu *et al.*, 2013; Nhiwatiwa *et al.*, 2017). Benthic macro invertebrates are considered effective indicators of quality of rivers worldwide (Aura *et al.*, 2011).

Freshwater macroinvertebrate species are therefore at higher risk of loss due to habitat degradation following overwhelming human activities (invasive industrialization, agriculture, and urban development) near rivers (Elias *et al.*, 2014). Excessive loading of both industrial and domestic waste into rivers can alter the physical, chemical and biological characteristics of the aquatic system beyond their natural self-purification capacity (Shimba *et al.*, 2018). The key aim of the present study is to identify and describe the composition and the diversity of benthic macro invertebrates in Temsa and to determine the human disturbances that influence the macroinvertebrates distribution and water physicochemical parameters.

## **1.2. Statement of the problem**

In most developed countries pollution of rivers has resulted in strict enforcement of waste disposal legislation, where wastewater is to be disposed of only after the quality meets certain criteria; in contrast to most developing countries where sewage goes untreated (Mrutu *et al.*, 2013). However, with the growth of cities, the amount of waste disposed into rivers often grows beyond their self-purifying ability (Mbuligwe, 2009). Some rivers in urban areas are affected by municipal waste which washes into the river. In areas with poor sanitation or lack of wastewater treatment facilities, these runoffs are highly polluted and they make their way into the rivers, they also serve as a major pollutant in the river (Ihunwo *et al.*, 2018).

Many cities in Africa are disposing untreated liquid and solid wastes to nearby rivers including, Addis Ababa, the capital city of Ethiopia and the seat for African Union is a very good example (Ambelu, 2009). The sewage system is not complete and the waste collection system is very poor. Research has shown that urban effluent into water bodies have the ability to change the physical and chemical conditions of a river, thereby leading to a contaminated state (J. Zhang *et*

*al.*, 2017).

Recently, Agaro town had no waste treatment systems; most of the manufacturing firms from the town discharge their effluents directly into the nearby streams without any form of wastewater treatment. In addition, oil pollution of rivers from waste discharge from car wash and garages is very common situation of Temsa River. Most of the activities such as hotels, garages, totals, car washes, coffee processing, etc were established near water bodies for water consumption during production process and dumping the finished wastes; however, there are unwise use agricultural activities through the catchment of the river can be mentioned as one of the major stressors to the aquatic ecosystems through sedimentation, increasing the nutrient level from fertilizers and pesticides. Thus to fill these gaps, this study intended to determine the effect of human disturbance on the assemblage of macroinvertebrates and physicochemical water parameters of Temsa River, Agaro town, South west Ethiopia.

### **1.3. Research questions**

- ❖ What are the effects of human disturbances on water quality and macroinvertebrate diversity of Temsa River?
- ❖ Is the water quality of Temsa River is good for aquatic organisms?
- ❖ What are the factors that affecting water quality parameter and macroinvertebrates assemblages?



## 1.4. Significance

All life, including human beings depends on water. Rivers are the most important freshwater ecosystems being used for a variety of life sustaining purposes. Rivers supply water for: domestic consumption, agriculture production, industrial purposes, generating electricity, recreation, fish production and birds of great tourism attraction as well as several other species. However, in appropriate management of waste which was produced by human and industrial activities leads to negative effect to human health and environment as pollution on river water. Therefore, this study tried to determine the effect of human disturbance on physicochemical water quality and benthic macroinvertebrates assemblages; one of the critical issues in determining assessment outcome. The major findings of this study had been used as a baseline in the study area to initiate and promote for effective means of improving assessing bioindicators for natural rivers. Few disciplines exist in which the study of nature can offer so much direct benefit toward the preservation and protection of the very habitat being studied.

This study aimed to:

- ❖ The findings of this study can be used by stakeholders concerning to plan, regulate and manage rivers
- ❖ May provide a baseline data on macroinvertebrate and water quality of area that may give on the way for future research

## UNIT TWO: LITERATURE REVIEW

### 2.1. Overview

Freshwater ecosystems are the most threatened ecosystems in the world (Dudgeon *et al.*, 2006). Fresh water systems are a habitat to about 6% of the world's total known species, 40% of the global fish species and 25% of all vertebrate species (Dudgeon *et al.*, 2006; Dudgeon, 2010). Some uses of water, e.g. for domestic purposes, agricultural production, industrial production, mining, power generation and forestry practices, cause deterioration in ecosystem, and also access to safe drinking water for human consumption.

They play a very important part in the water cycle, acting as drainage channels for surface water. Rivers drain nearly 75% of the earth's land surface. Rivers provide excellent habitat and food for many of the earth's organisms. Many rare plants and trees grow by rivers. Ducks, voles, otters and beavers make their homes on the riverbanks. Reeds and other plants like bulrushes grow along the river banks. Other animals use the river for food and drink. Birds such as kingfishers eat small fish from the river. In Africa, animals such as antelopes, lions and elephants go to rivers for water to drink. Other animals such as bears catch fish from rivers. River deltas have many different species of wildlife. Insects, mammals and birds use the delta for their homes and for food.

Rivers provide travel routes for exploration, commerce and recreation. Water quality is defined in terms of the chemical, physical and biological contents of water. Water quality Guidelines provides basic scientific information about water quality parameters and ecologically relevant toxicological threshold values to protect specific water uses. Most of the rivers in the urban areas of the developing countries are the ends of effluents discharged from the industries. African countries and Asian countries experiencing rapid industrial growth and this are making environmental conservation a difficult task.

River water can be polluted by hazardous substances coming into contact with this surface water, dissolving or mixing physically, chemically or biologically with water can be called surface water disturbance (Fawell, 2016). Surface water is heavily affected by human impact and their pristine state is no more recognizable in many temperate regions, due to the long history of anthropogenic influence (Comiti, 2012). Moreover, surface water is considered the most

threatened ecosystems of the world .The discharge of effluent following sewage is one of the major factors affecting the biological quality of surface waters (Narciset *al.*,2013).There are hundreds, perhaps thousands of pollutants whose effects are of actual and potential concern. Pollutants have been classified according to their mode of occurrence into physical and chemical. Rivers have a natural, though limited, capacity to restore water quality to pre-pollution levels, through dilution, die-off, sedimentation and biological processes (Anyona *et al.*, 2014).

## **2.2. Factors that affect water quality and macroinvertebrates assemblages in rivers**

The human impact on water resources takes different forms. It includes physical alteration and pollution from industries and residential areas. Also, it includes changes in riparian vegetation and stream morphology, sedimentation, nutrient additions, organic enrichment and pesticide contamination from agricultural land uses (Whiles *et al.*, 2000). Anthropogenic influences are known sources of water pollution and include urban, industrial and agricultural activities increasing exploitation of water resources as well as natural processes, such as precipitation inputs, erosion and weathering of crustal materials degrade surface waters and damage their use for drinking water, recreational and other purposes (Irfan R. Shakil, 2012). In Ethiopia land degradation, urban sanitation, industrial and chemical pollution are the major environmental problems (Zinabu and Zerihun, 2002) that cause adverse impact on aquatic resources of the country.

Most rivers in urban areas are affected by municipal waste, which washes into the river. In areas with poor sanitation or lack of wastewater treatment facilities, these runoffs are highly polluted and when they make their way into the rivers, they serve as a major pollutant in the river. Research has shown that urban effluent into water bodies have the ability to change the physical and chemical conditions of a river, thereby leading to a contaminated state (O.Ihunwo *et al.*, 2017).

It is estimated that industry is responsible for dumping 300-400 million tons of heavy metals, solvents, toxic sludge, and other waste into waters each year worldwide (UNEP, 1991). Industrial effluent can alter the physical, chemical and biological nature of the receiving water body leading to deterioration in water quality and quantity that causes adverse impact on the water chemistry and biological elements (Carr and Neary, 2008).

Rivers and streams exposed to agricultural activities, reservoir flushing and paper mill waste are generally compared to samples from stream flowing through natural forest with similar underlying habitat and eco-region. Agriculture is one of the major human activities responsible for nonpoint-source of pollution in streams of Ethiopia. Agricultural practices such as crop cultivation adjacent to streams can lead to soil erosion and subsequent runoff of fine sediments, nutrients and pesticides (Lowrance *et al.*, 1984; Cooper *et al.*, 1986).

Moreover, the unwise agricultural activities through the catchment of rivers and streams can be mentioned as one of the major stressors to the aquatic ecosystems through sedimentation, increasing the nutrient level from fertilizers and pesticides. The health of these water bodies are increasingly deteriorating, since there is no continuous monitoring related to the high cost incurred to physicochemical parameter and absence of bio assessment based water quality assessment policy for mitigation and control measures. Above all, most rivers and streams in Ethiopia are not sufficiently studied and there is limited knowledge on ecological health for proper management to develop a systematic overall picture of the status of these lotic environments.

African countries are known for the highest fertility rate in the world. Due to this, the population increment is faster than in any other part of the world. As the population increases, the land for agriculture and other fixed resources, like water, are becoming scarce. The per capita cultivated land has fallen by 40% (0.5 to about 0.3 hectare per person) since 1995 (Franzel *et al.*, 2004), which has resulted in land constraints. Because of this reason, people started to cultivate forests and wetlands and others migrated to the urban areas seeking better life and other job opportunities. This had result in the generation of extra wastes, which are in most cases disposed haphazardly in the environment.

To feed the growing human population of the region, different methods to increase crop productivity are practiced. One of these techniques is the application of fertilizers and pesticides on farm lands. Since soil fertility has been largely reduced, application of organic and inorganic fertilizer to farm lands is necessary to feed the growing human population (Franzel *et al.*, 2004). The second technique is the use of surface waters to irrigate farmlands to increase the productivity by increasing the frequency of crop harvesting. This needs diversion and damming of rivers and draining of lakes.

Chemical surface water disturbance is generally atoms or molecules, which have been discharged into natural water bodies, usually by the activities of humans. Common examples of such chemical surface water pollutants are mercury emanating from mining activity, certain nitrogen compounds used in agriculture, chlorinated organic molecules arising from sewage or water treatment plants or various acids which are the externalities of various manufacturing activities (Fawell, 2016).

### **2.3. Anthropogenic activities and environmental degradation**

The majority of the Ethiopian surface water resources have faced a serious quality deterioration that mainly resulted from increasing anthropogenic activities. The alarming human population growth has demanded intensified agricultural activities resulting in more forest clearings, irrigation, fertilizers and pesticides application and overgrazing, which are becoming major surface water pollution sources (EPAE, 2008). Industrialization and urbanization are other major threats in the deterioration of surface water quality. In fact, the deterioration of the water quality was already detected some time ago (Zinabu and Elias, 1989).

Agricultural activities, deforestation, industrial activities, fires and overgrazing have been mentioned as major threats to the biological diversity and the environment in Ethiopia. For example, the regional government of the south-western part of Ethiopia has documented that land degradation is a major environmental deterioration (Bizuayehu *et al.*, 2002). This land degradation resulted from erosion of soil due to the topography of the area, the lack of vegetation cover, poor land use and management practices, rainfall and wind (Barber, 1984). It has been observed that, like in the other parts of Ethiopia, the vegetation in this watershed is under high pressure due to agricultural activities and waste discharges (Mulat, 2001).

The few available reports are showing that there is an increasing discharge of liquid and solid waste into the nearby rivers. Studies done on a limited number of sites of a few rivers have indicated that water quality of rivers crossing urban environment are getting degraded due to municipal and industrial discharges (Berhe *et al.*, 1989; Hailu and Mulat, 1997; Beyene *et al.*, 2009a). In general, pollution coming from point and diffuse sources are major threats resulting in a continuous decline of the water quality. It is therefore imperative to have a decision support tool for monitoring and management of surface waters in Ethiopia.

## **2.4. Benthic macroinvertebrate assemblages**

Benthic Macroinvertebrates valuable indicators of the health of aquatic environments in part because they are benthic, meaning they are typically found on the bottom of a stream or lake and do not move over large distances. Therefore, they cannot easily or quickly migrate away from pollution or environmental stress. Because different species of macro invertebrates react differently to environmental stress like pollution, sediment loading and habitat changes, quantifying the diversity and density of different macro invertebrates at a given site can create a picture of the environmental conditions of that body of water.

Roy *et al.* (2009) suggested that the diversity of benthic macroinvertebrates in natural rivers was measured and the patterns compared with the information of relative human activities like urbanization in order to test the biodiversity changes with human development. The observation of these species is particularly useful in testing rivers water quality because most of them will be a good indicator species. These reasons are why most benthic macroinvertebrates assemblages are such excellent indicator species. They were important members in the food web of a river ecosystem, and a pollutant enters the rivers or some damage disturbance, they were inevitably being affected. This studies on the potential use of benthic macro invertebrates as bio indicators for rivers ecosystems were major finding literatures.

## **2.5. Biomonitoring**

Biomonitoring is generally defined as the systematic use of living organisms or their responses to determine the condition of an environment. LiL *et al.*, (2010), indicates that biomonitoring is a method of observing the impact of external factors on ecosystems and their development over a period or it is an ecological exercise where various kinds of biota are considered in determining the extent of pollution in a water body (Sharma,2010). Biological monitoring based on various aquatic biotas may be more effective than measuring water physicochemical alone, because the organisms integrate the chemical and physical properties of streams over time (Yung-Chul Jun, 2012).

Biomonitoring techniques are best used for detecting aquatic life impairments and assessing their relative severity. According to Barbour *et al.*, (1999), once impairment is detected, additional

ecological data such as chemical and biological testing is helpful to identify the causative agent, its source, and to implement appropriate mitigation. Integrating information from these data types as well as from habitat assessments, hydrological investigations, and knowledge of land use is helpful to provide a comprehensive diagnostic assessment of impacts from different principal factors for description of water quality, habitat structure, energy source, flow regime, and biotic interaction factors (Barbour *et al.*, 1999).

## **2.6. Bioindicators**

Bioindicators are any biological species or group of species whose function, population, or status can be used to determine ecosystem or environmental integrity. An example of such a group is the copepod and other small water crustaceans water bodies. Such organisms are monitored for changes (biochemical, physiological or behavioral) may indicate a problem within their ecosystem. According to Tingey *et al.*, (2009), biological indicators are species used to monitor the health of an environment or ecosystem. LiL *et al.*, (2010), indicates that are bioindicators are an ideal indicator at least should have taxonomic soundness (easy to be recognized by non-specialist); wide distribution; low mobility (local indication); well-known ecological characteristics; numerical abundance; suitability for laboratory experiments; high sensitivity to environmental stressors; and high ability for quantification and standardization.

Biological indicators can tell us about the cumulative effects of different pollutants on the ecosystem and about how long a problem may have been present which physical and chemical testing cannot. Shailendra *et al.*, (2006), suggested that macroinvertebrates are animals that lack a backbone and generally are visible with the naked eyes. They live in the lower areas of the streams underneath rocks. They include larval forms of many common insects such as Dragon flies, Damselflies and Crane flies. Macroinvertebrates reveal low mobility, long life-span and high diversity with respect to pollution tolerance that make them useful bioindicators. Several investigators have been describing the use of microinvertebrates for water quality bio indicators (Adakole, 2001; Ogeibu & Ezeunara 2002).

## **2.7. Physicochemical parameters**

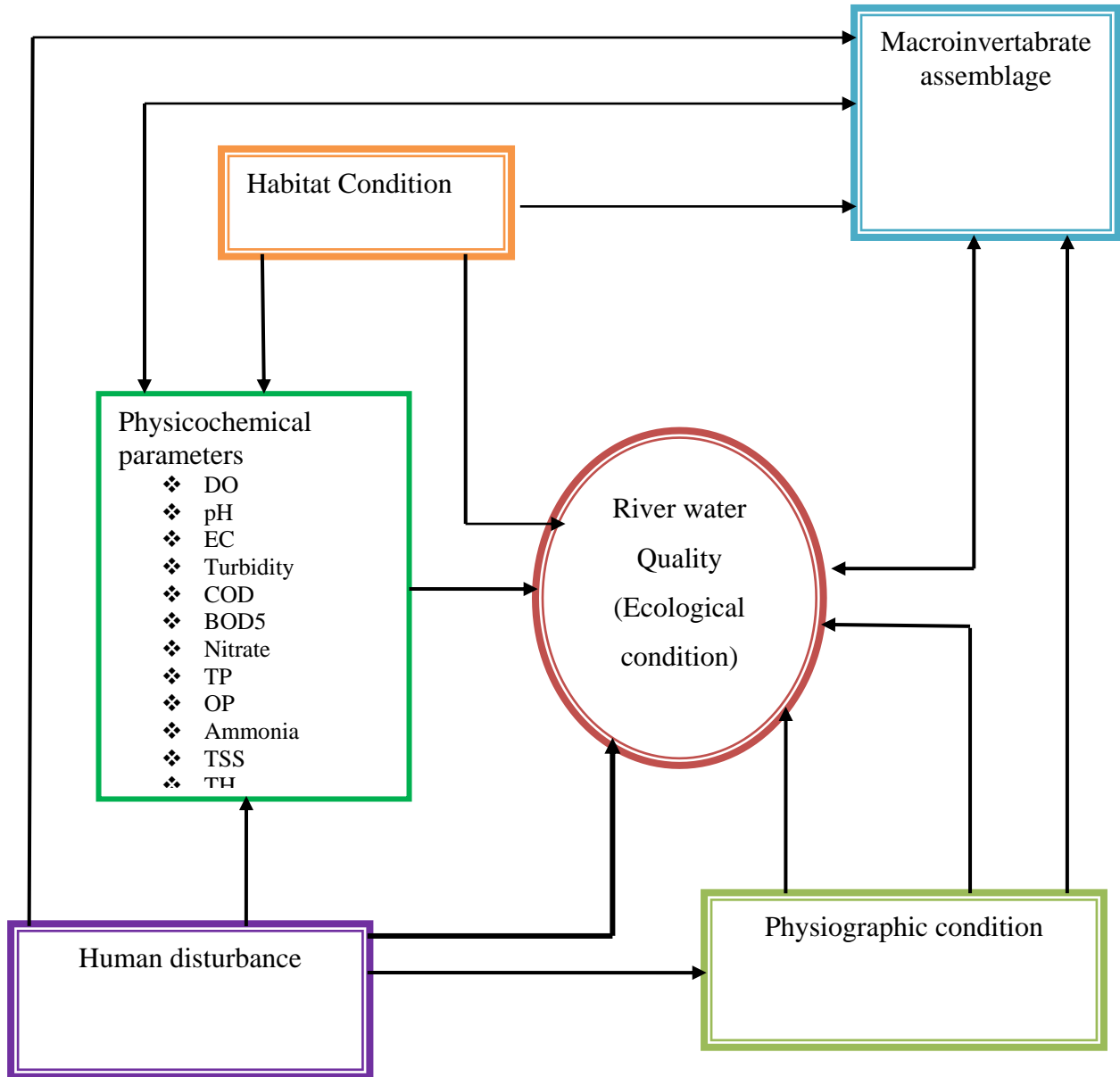
In any aquatic ecosystem, physicochemical parameters affect macroinvertebrates either positively

or negatively depending on their source. Excessive physicochemical parameters can cause long- or short-term shifts in invertebrate community richness, abundance and species composition. Sarkar *et al.* (2006) indicates an increase in nutrient, organic matter or contaminant concentrations in surface waters, sediments or food sources for instance, has been shown to result in low diversity of macro invertebrates, with an increase in the abundance of stress tolerant species.

Physicochemical parameter study is very important to get exact idea about the quality of water and we can compare results of different physicochemical parameter values with standard values. It is very essential and important to test the water before it is used for drinking, domestic, agricultural or industrial purpose. Physical and chemical properties are parameters that do not identify particular chemical species but are used as indicators of how water quality may affect water uses. These are temperature, electrical conductivity, total dissolved solids, chemical oxygen demand, hydrogen ion concentration (measured as pH), alkalinity, hardness, and total dissolved solids. Water must be tested with different physicochemical parameters. Selection of parameters for testing of water is only depends upon for what purpose we are going to use that water and what extent we need its quality and purity.



## 2.8. Conceptual framework



**Figure 1.** Conceptual frame work of factors affecting the river water quality

## **CHAPTER THREE: OBJECTIVES**

### **3.1. General objective**

To assess the effect of human disturbance on benthic macroinvertebrates assemblages and physicochemical water quality of Temsa River, Agaro Town, Southwestern Ethiopia

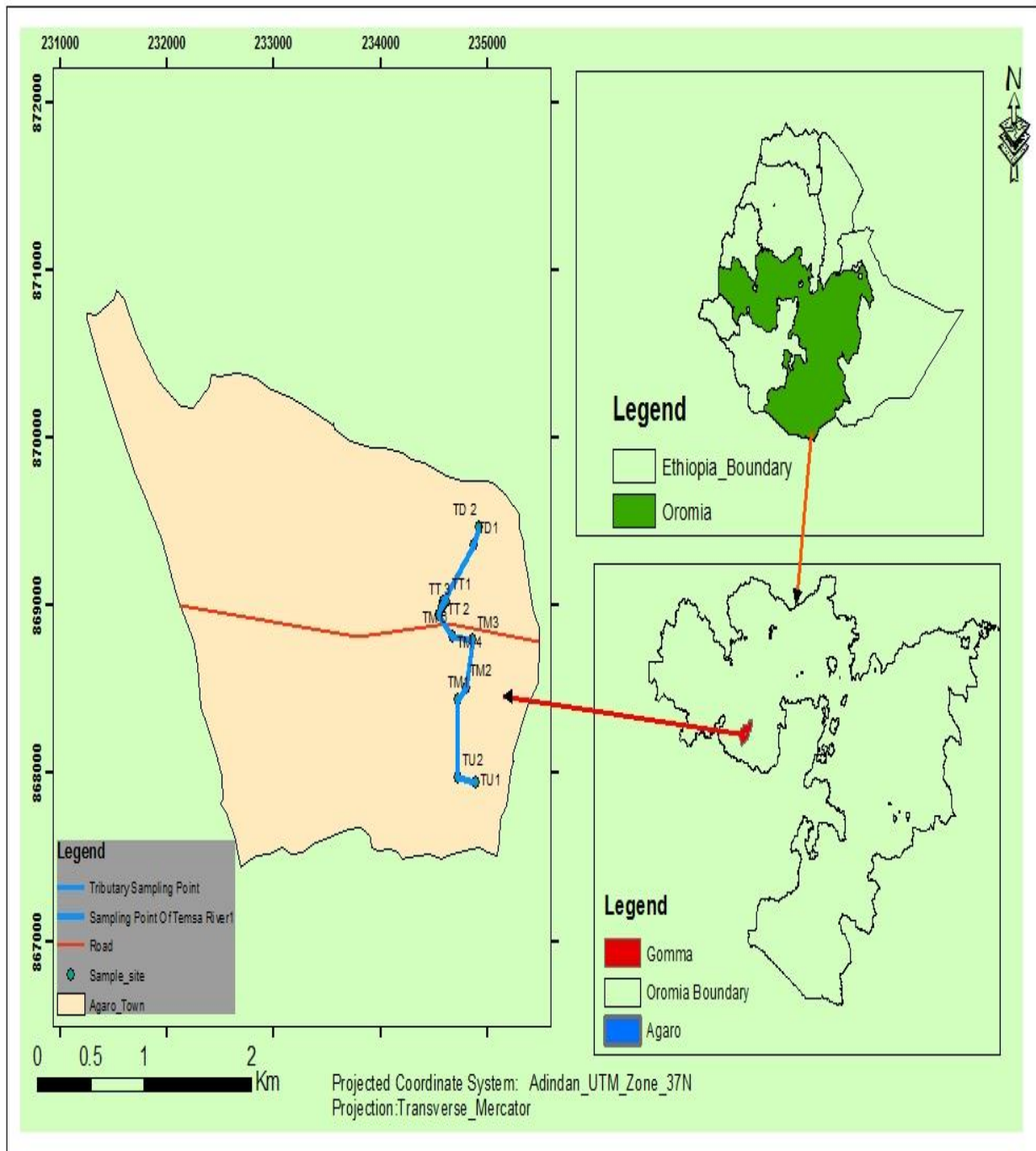
### **3.2. Specific objectives**

- To determine the assemblage of macroinvertebrate (abundance and diversity) of Temsa River
- To determine the physicochemical river water quality parameters of Toms River, Agra town
- To identify the relationships between macro invertebrates and water quality parameter
- To identify the major human disturbance activities that affect water quality of Temsa River

## CHAPTER FOUR: METHODS AND MATERIALS

### 4.1. Description of the study area

The study was conducted in Temsa River, Agaro town, located at a distance of about 400 km from Addis Ababa the capital city of Ethiopia, and around 50 km towards south west from Jimma town. The town's geographical coordinates are situated at latitude of 7°51'N and longitude 36°35'E with an altitude of 1560 m above sea level. The town has estimated total population of 92,458 of whom 44,938 are men and 47,520 are women. The river used for a variety of domestic and agricultural activities (e.g. crop cultivation and cattle farming) and these activities are some of the major stressor which contributes for the pollution of the river through the release of agrochemicals and organic waste. In addition, at upper sites it was used for intensive irrigation in the dry season mainly for growing a stimulant locally known as '*Chat*' or '*Khat*'(*Catha edulis*) and vegetable production. The river widely serves for various domestic activities (washing/bathing, drinking, dumping of domestic wastes) and dumping of the finished wastes. Due to the intense human activities taking place in the area there was devoid of riparian vegetation.



**Figure 2.** Location of study area and sampling sites of Temsa River Agaro town

## **4.2. Study design and period**

A cross-sectional study was used to assess the effect of human disturbances on macroinvertebrates assemblages and physicochemical water quality on Temsa River, Agaro town. The samples were collected on April 2019 at the dry season.

## **4.3. Site selection and sampling techniques**

Benthic macroinvertebrate assemblage and water samples were collected from a total of thirteen (13) sampling sites (ten from the Temsa River and three from its tributary) purposely along the river and the sites were divided into upstream, midstream, downstream and tributary. Two of the sampling sites (TU1, TU2) were located at upstream of the river before municipal waste and sewage discharge points and therefore serves as a referencing site. Six sampling sites were found at mid stream (TM1, TM2, TM3, TM4, TM5 and TM6) with a bridge crossing and high anthropogenic impacts, major activities such as dumping of wastes, washing clothes, vehicle washing, garage wastes disposal, wastes discharge from hotels, market and abattoir activities were at the points of different municipal waste discharges (midstream) takes place. While, two sites ( TD1, TD2) were along the sub-catchment (downstream) where wastes had already mixed thoroughly and high vegetation cover .In addition, three sampling sites (TT1, TT2 and TT3) were collected from the tributary which flows from the town and join the river between middle five and middle six sampling site. The sampling criterion was the activities impacts on study sites and depends on the bases of their size and the existing information.

## **4.4. Data collection**

The study was designed to investigate the water physicochemical and biological study of the river was done to determine the biological composition, water quality and human activities that more affect the river.

### **4.4.1. Water quality samples**

Water sampling was carried out by sampling from thirteen of water from each sampling site. For this study, the major water quality parameters determined were: temperature, pH, electrical conductivity, nitrate, total dissolved solids, turbidity, biological oxygen demand, Chemical oxygen demand and dissolved oxygen. On site water temperature, dissolved oxygen, pH,

electrical conductivity and turbidity were measured by using a multi-probe meter (HQ30d Single-input Multi-Parameter Digital Meter (APHA *et al.*, 1995). The Water samples were collected in 2-L polyethylene sampling bottles and 10 cm below the surface rinsed at least three times before sampling as indicated in APHA *et al.*, (1999), was stored in an icebox and transported to the Laboratory of Environmental Health Science and Technology, Jimma University. Ammonia was analyzed using direct nesslerization method (APHA, 1998). Total phosphorus samples were first digested in a block digester using ammonium per sulfate and sulphuric acid reagent (APHA, 1998) and measured with photometric kits (HACH LANGE) using a Hach DR5000 spectrophotometer. Five days biochemical oxygen demand, chemical oxygen demand, total hardness (TH), alkalinity, chloride, calcium hardness and magnesium hardness were analyzed in the laboratory according to APHA *et al.*, (1995).



**Figure 3.** Determination of physicochemical parameters water quality parameters in the laboratory

#### 4.4.2. Benthic macroinvertebrates samples

Benthic macroinvertebrates were sampled according to a standardized protocol using a triangular D-frame kick net with a 300 µm mesh size in all the available habitat types (multi habitat sampling procedure), such as riffles, macrophytes, pools and bedrock collectively for 3min up to 6 min per site within 10 m radius for each sampling sites (Kebede *et al.*, 2010 and Munyika *et al.*, 2014). The bottom sediment was disturbed by kicking with the feet during sampling in order to effectively collect benthic macro invertebrates. Macroinvertebrates were collected in the field, kept in vials containing 80% ethanol for later identification and enumeration. Macro invertebrates were identified to family level in the laboratory using a stereomicroscope (4 ×magnifications) and standard identification key of Bouchard (2004; 2012). Moreover, total family richness, abundance, richness of ET taxa, Biological Monitoring of Working Parties scores (BMWPs) and family biotic index (FBI) was calculated to characterize a sampling site.



**Figure 4.** Collection of macroinvertebrate taxa in Temsa River, April, 2019





**Figure 5.** Macroinvertebrates identifications by using stereo microscope and identification key of Bouchard (2004)

#### **4.5. Human disturbance determination**

At the beginning anthropogenic activities/human disturbance were observed and identified along the river by physical observation at each sampling sites. The checklist which was used for conducting those activities was prepared. During sample collection the human disturbance (anthropogenic activities) that were observed and identified were registered with checklist by present /absent response (for the presence of disturbance we used “✓” and if no we used “×”) (Appendix 7).

#### **4.6. The Study Variable**

##### A. Macroinvertebrate assemblages

- Abundance
- Richness
- Diversity
- Composition



## B. Water quality

- Dissolved Oxygen
- pH
- Turbidity
- BOD5
- COD
- Total dissolved solids
- Total Suspended Solids
- Water temperature
- Total phosphate
- Ortho phosphate
- Electrical conductivity
- Ammonia
- Nitrate
- Total hardness

## C. Human disturbance/anthropogenic activities

- Waste dumping
- Effluent discharge
- Agriculture
- Washing, clothing, grazing
- Removal of vegetation
- Open defecation
- Car wash
- Settlement
- Drainage

## **4.7. Data analysis**

### **4.7.1. Biological Metrics**

Metrics describe a sampled macroinvertebrate population in terms of its taxonomic composition, community structure, trophic structure and presence of tolerant and intolerant taxa. In this study, about ten macroinvertebrate metrics were calculated which are categorized as diversity measures, composition measures and biotic indices. A sensitive macroinvertebrate metric is able to discriminate sites according to the degree of perturbation (Davies and Jackson, 2006). Along with the degree of disturbance, macroinvertebrate metric may show increment or decrement.

#### ***4.7.1.1. Diversity measures***

Diversity measures are widely applied in the assessment of river ecology. Their major advantage is in condensing large amounts of biological data into numbers comprehensible and useful to people not immediately familiar with the specific biota. In that way, they are convenient for policy makers and river managers. Species richness emphasizes the number of taxa, usually at species or family level: the more species, the greater the diversity. Most diversity indices may be classified as either species diversity measures or dominance diversity measures.

#### **A. Family abundance and richness**

Abundance is the total number of individuals counted in a sample or a study site. It can be also used to express the abundance of sensitive taxa in a sample. Taxa richness expresses the number of distinct taxa in a sample or a study site and represents the diversity within a sample (Barbour *et al.*, 1999). Higher numbers of taxa are an indication of a better water quality.

#### **B. Shannon Diversity Index ( $H'$ ) (Shannon, 1948)**

It is the most used metric to measure heterogeneity. Shannon Diversity Index is interpreted as the probability of finding the same species in two organisms randomly selected from a sample (Balderas *et al.*, 2016). The values above 3.0 indicate that habitat structure is stable, high taxa and balanced and values under 1.0 indicate the presence of pollution and degradation of

habitat structure (Mengesha *et al.*, 2017). As a diversity index, it expresses the average degree of uncertainty of predicting the taxon of an individual picked at random from a community. Uncertainty increases both as the number of taxa increases and as the individuals are distributed more equally among the collected taxa. Unlike the Simpson index, it is sensitive to the addition or the loss of rare taxa. This index is calculated as:

$$H = -\sum [(n_i / N) \times \ln (n_i / N)] \dots \dots \dots \text{equation 1}$$

Where: H: Shannon Diversity Index

$n_i$ : Number of individuals belonging to  $i$  species

N: Total number of individuals.

As indicated in Mandeville (2002), this index frequently varies from 0 to 5; as the number and distribution of taxa (biotic diversity) within the community increases, so does the value of  $H'$

**C. Simpson index (D)**

It measures the probability that two individuals randomly selected from a sample was belong to the same taxon. This index is not sensitive to the loss or the addition of rare taxa (Simpson, 1949). This index is calculated as:

$$D = \frac{\sum n_i(n_i-1)}{N(N-1)} \dots \dots \dots \text{equation 2}$$

Where  $n_i$ = total number of individuals of a particular species.

N = total number of individuals of all species Simpson's index of diversity

**D. Evenness Index “J” (Pielou, 1975).**

$$J = \frac{H_{max}}{H} \dots \dots \dots \text{equation 3}$$

Where J: Pielou evenness index,

H: the observed value of Shannon index  $H_{max}$ :  $\ln S$ , S: total number of species.



## B. Biological monitoring working party (BMWP)

The BMWP score were calculated by adding the individual scores of all indicator organisms present at family level. The organisms were identified to the family level and then each family was allocated a score between 1 and 10 based on literature. Then the BMWP index value was calculated for each sampling site. The score each family gets reflected their perceived defenselessness to pollution, which is based on the principle that different aquatic invertebrates have different tolerances to pollutants (Paisley *et al.*, 2004). The presence of these high scoring families indicates a site with unpolluted water. As indicated in Zeybek *et al.*, (2014), the overall BMWP Score for a site is the sum of all of the scores of each family present at that site.

Table 2. Categories of water quality based on BMWP (Mandeville, 2002)

BMWP Score	Category	Interpretation
0-10	Very Poor	Heavily Polluted
11 – 40	Poor	polluted or Impacted
41 – 70	Moderate	Moderately Impacted
71 – 100	Good	Slightly Impacted
> 100	Very Good	Unpolluted

### 4.7.1.3. Composition measures

Composition measures were calculated for macroinvertebrate groups which are sensitive or tolerant and are calculated in terms of percentages or rations from the sample. The percentage of EPT and Chironomidae (Barbour *et al.*, 1997), were the composition measures used in this study.

### 4.7.1.4. Statistical analysis

Multivariate statistical analyses were used to analyze the existence of linear relationships between biological data and environmental variables. Principal component analysis (PCA) was performed to assess relationships among sampling sites using statistical software Paleontological Statistics software package for education and data analysis (Past 3.18) version 1.0.0. The

relationships of environmental variables, biological metrics among sampling sites were elucidated using correlation principal component analysis (PCA).

In this study, PCA was applied to reveal the relationship between macroinvertebrates and environmental data with sampling sites. The distributions of benthic macroinvertebrate taxa in relation to the sampling environmental variables were analyzed. Macroinvertebrate metrics and environmental data were nominalized and transformed  $\log^{(x+1)}$  prior to analysis to obtain homogeneity of variance. In order to evaluate water quality biological indices values of sensitivity to water contamination for the various macroinvertebrate families was done. Microsoft excel were used to calculate the metrics. Analysis of variance (ANOVA) was used compare the mean of macroinvertebrate metrics and environmental data obtained from thirteen sampling sites. One-way ANOVA was used to test whether there were significant differences ( $p < 0.05$ ) between water quality variables and biological metrics from all sampling sites. Mean and significance tests were analyzed by using SPSS version 20. All statistical analyses were carried out in the statistical software packages PAST, SPSS, and Excel

#### **4.8. Ethical consideration**

Ethical clearance was taken from Ethical and Research Committee of Jimma University, Institute of health to officially as certain that the research was relevant and approved by the college as well as by the Department of Environmental Health Science and Technology.

#### **4.9. Dissemination plan**

The final result of this study was presented to Jimma University, Institute of Health, Department of Environmental Health Science and Technology. Endeavors were made to so as to provide important information for biomonitoring of river ecosystem programs for the local community and international level as general. Publication in national and international level journals was also considered.

## CHAPTER FIVE: RESULTS

### 5.1. Macro invertebrate abundances and occurrence

A total of 652 individual macro invertebrates classified into 41 families and 12 orders were collected from Temsa River Agaro town during April, 2019. The most abundant families were Coenagrionidae, Chironomidae and Hydrophysycidae; which are accounted for 111 (17.02%), 89 (13.65%) and 47 (7.21%) respectively; While the most abundant order was Odonata consisting of eight families with a relative abundance of 31.6%. The second dominant order was Dipterans consisting of nine families with a relative abundance of 22.9%. Chironomidae had been the most frequently occurring family in the order Dipterans, which was found in the 59.7% of the study sites (Table 3).

Table 3. Relative abundance of macroinvertebrate assemblages at sampling sites

Macroinvertebrate s families	Sampling sites												
	TU1	TU 2	TM1	TM2	TM3	TM4	TM5	TM6	TD1	TD1	TT1	TT2	TT3
Aeshaidae	8	3	2	4	1	0	0	0	1	0	0	0	0
Baetidae	3	2	2	0	5	2	0	0	0	0	0	0	0
Belostomatidae	0	0	0	0	0	0	4	3	0	0	2	6	4
Caenidae	0	0	0	0	0	4	2	3	1	11	3	2	2
Calopterygidae	3	11	3	5	0	0	0	0	0	0	0	0	0
Ceratopogonidae	0	0	0	1	0	0	3	0	0	0	0	1	3
Chironomidae	0	0	0	3	8	10	14	9	5	7	7	11	15
Chlorocyphidae	4	2	2	1	0	0	0	0	0	0	0	0	0
Coenagrionidae	10	8	16	17	16	13	0	0	2	27	2	0	0
Corixidae	0	0	0	1	0	0	1	3	0	0	0	1	0
Culicidae	0	0	0	0	0	0	1	3	4	2	0	0	2
Dixidae	6	0	1	0	0	0	0	0	0	0	0	0	0
Dytiscidae	1	1	1	0	2	0	0	0	0	0	0	0	0

Ecnomidae	0	0	0	0	1	0	0	0	0	3	0	0	0
Elmidae	0	0	0	0	0	0	0	2	0	0	0	0	2
Ephydriidae	0	0	0	0	0	0	2	0	0	0	1	0	0
Gerridae	0	0	0	0	0	1	4	1	1	0	2	0	0
Gomphidae	5	2	2	5	0	0	0	0	3	2	0	0	0
Gyrinidae	0	3	2	0	0	1	0	0	0	3	0	0	0
Helodidae	1	3	1	0	1	0	0	0	0	0	0	0	0
Heptageniidae	6	3	3	0	0	0	0	0	7	4	0	0	0
Hirudinae	0	0	0	0	0	0	3	1	4	0	0	2	6
Hyrophilidae	3	0	0	0	0	0	0	0	1	0	0	0	0
Hydropsychidae	0	11	3	0	4	0	11	2	13	3	0	0	0
Lestidae	0	0	0	0	0	0	6	0	0	1	3	3	0
Lymnaeidae	0	0	0	0	0	0	0	6	1	3	1	3	4
Libellulidae	0	0	0	0	0	0	0	3	0	2	0	0	3
Leptophlebiidae	5	3	0	0	0	0	0	0	3	0	0	0	0
Naucoridae	3	7	15	2	0	0	0	0	0	0	0	0	0
Nepidae	0	1	0	0	0	0	0	0	0	0	0	0	0
Notonectidae	2	2	4	1	0	0	0	0	0	0	0	0	0
Oligochaeta	0	0	0	0	0	1	0	0	0	0	0	0	0
Philopotamidae	0	0	0	2	0	1	0	0	0	2	0	0	0
Potamanautidae	0	0	0	0	0	0	1	0	0	0	0	0	0
Pyrilidae	2	3	0	0	0	0	0	0	0	0	0	0	0
Tabanidae	0	1	1	2	3	0	1	0	0	0	0	0	0
Tipulidae	5	0	1	0	0	0	0	0	0	0	0	0	0
Simulidae	0	0	0	0	0	1	0	2	0	0	0	0	0
Syrphidae	0	0	0	0	0	0	1	3	5	0	1	4	2
Sphaeriidae	0	0	0	0	0	0	0	1	0	2	0	0	0
Veliidae	0	0	0	0	0	0	0	9	6	0	5	3	1



## **5.2. Biological metrics**

### **5.2.1. Macroinvertebrate richness and diversity**

As shown in table 4, the average values of benthic macroinvertebrate metrics varied across sampling sites of the river. Upper stream had relatively higher family richness and diversity having the mean value of 14.5 and 2.58 respectively as compared to other sites, while low family richness and diversity was found at tributary site having the mean values of 10.33 and 2 respectively. From the upper sites TU2 had the highest family richness which consists of 17 families and TT3 had the least which consists of nine (9) families. In addition, BMWP score and family richness of sensitive taxa such as Ephemeroptera and Odonata (EOT family richness) were higher in upper stream having the mean value of 93.5 and 8.5, while the least BMWP and EOT family richness was found at tributary site having the mean value of 51.33 and 2.33 respectively. On the other hand, FBI score was higher in tributary and middle stream sites having the mean value of 7.99 and 6.22, in contrast the least mean value of the FBI was found at the upper stream site having the mean value of 4.6.

As can be seen % Chironomidae, the taxon was highly observed in tributary and the middle stream site having the mean value of 30.2 and 16.5 respectively. In contrast percent of Chironomidae was not observed in upper stream site and had the least in downstream having the mean of 9.2. High % EPT was observed in downstream and upper stream sites having the mean value of (26.5, 21.8) and lower in tributary and middle stream (9.07, 13.56). Shannon diversity index of macroinvertebrate communities from all sites showed 1-3 indexes, which shows moderate pollution (Table 4).

Table 4. Mean values of biometrics in Temsa River Agaro town, South West Ethiopia

Metrics	Upper stream (n=2)	Middle stream (n=6)	Downstream (n=2)	Tributary (n=3)
Abundance	66.5	47.17	64.5	35.67
Family Richness	16.5	12.5	14.5	10.33
Shannon index	2.58	2.01	2.27	2.0
Evenness	0.92	0.80	0.85	0.85
FBI	4.61	6.23	5.69	7.99
BMWP	93.5	63.67	85	51.3
Simpson index	0.08	0.16	0.14	0.13
EOT family richness	8.5	5	7.5	2.33
Dipterans family richness	1	3.17	3	3.33
% Chironomidae	0	16.54	9.25	30.18
%EPT	21.84	13.56	26.57	9.07

### 5.2.2. Macroinvertebrate biotic indices (BMWP and FBI)

The average values of the family biotic index in the studied area ranged from 4.61 to 7.99. According to the family biotic index, tributary sites were found to be very poor water quality which had severe organic pollution; middle stream, and downstream was fairly poor, which had significant organic pollution, and upper stream site had good water quality was slight pollution. BMWP showed an average value of 93.5 to 51.33; accordingly, the upper stream and downstream which had a mean value of 93.5 and 85 were comprised of good water quality (slightly impacted). The other site middle stream and tributary sites were found in the range of 41-70, which had moderate water quality or moderately impacted (Table 5).

Table 5. Evaluation of water quality and organic pollution in Temsa River with BMWP and FBI indices

Sites	FBI	water quality class	BMWP	Water quality class
Upper stream	4.61	Good	93.5	Good
Middle stream	6.23	Fairly poor	63.67	Moderate
Down stream	5.69	Fair	85	Good
Tributary	7.99	Very poor	51.33	Moderate

### 5.3. Physicochemical characteristics of water samples

The mean value of dissolved oxygen concentration at middle stream and tributary sites were below 5 mg/L (Table 6). The pH value of three sites (75%) was found between 6.5-9.0 except tributary having the mean value of 5.87; the chemical oxygen demand concentration at middle stream, downstream and tributary were above 30 mg/L except at the upper stream which had the value of 21.18 mg/L and the low electric conductivity with a range of (75.3 $\mu$ S/cm to 279 $\mu$ S/cm) was recorded at all sites (Table 6). The concentration of NO<sub>3</sub><sup>-</sup> between the permissible range < 50mg/L was recorded at all sites. The highest value of total suspended solids was 3.4 mg/L at TT3; whereas the highest value of TDS was 485.33 was recorded at tributary site and the highest value of Phosphates were 2.76 mg/L which was recorded at tributary site. The concentration of phosphate was not in permissible range, which was 1.79-3.03 mg/L. The measure of turbidity was ranged from 33 NTU at TU2 site to highly turbid conditions 288 NTU at TT3. The mean values of Chemical oxygen demand, nitrate and total phosphate, electrical conductivity, ammonia, turbidity, total dissolved solids and alkalinity concentrations were higher at tributary sites are probably indicative of high organic loads.

Table 6. Mean and standard deviation of physicochemical parameters in the study area (NTU =Nephelometric Turbidity Units, SD = standard deviation)

Physicochemical variables	Unit	Upstream (Mean ± SD)	Middle stream (Mean ±SD)	Downstream (Mean ±SD)	Tributary
WT	°C	26.75±0.636	22.82±3.78	22.05±1.06	20.47±1.96
pH		8.07±0.028	6.78±0.76	7.275±0.64	5.87±
DO	mg/L	7.39±0.269	4.54±0.89	5.89±0.27	3.67±0.48
EC	µS/cm	76.75±2.051	91.62±18.3	118.4±0.14	249.33±32.25
Turbidity	NTU	33.25±0.35	62.42±35.2	124±5.66	227.67±54.50
Nitrate	mg/L	1.39±0.028	2.24±1.25	4.87±0.17	7.47±1.56
TP	mg/L	1.1±0.127	1.41±0.354	2.175±0.30	2.76±0.26
OP	mg/L	0.46±0.17	0.45±0.18	0.535±0.19	1.12±0.17
Ammonia	mg/L	0.1±0.00	0.707±0.41	0.125±0.04	0.9±0.29
TDS	mg/L	98±8.49	119.33±26.58	162±2.83	485.33±317.5
TSS	mg/L	0.46±0.028	0.65±0.21	1.11±0.07	2.76±0.6
BOD5	mg/L	18.6±2.616	46.9±16.24	40.7±5.09	64.8±3.34
COD	mg/L	21.18±2.418	60.38±17.75	51.775±5.78	81.57±10.38
Cl <sup>-</sup>	mg/L	11±1.418	11.17±1.57	16±0	21.32±1.15
Alkalinity	mg/L	30±2.828	30.67±3.266	32±0.0	44±4
TH	mg/L	48±0.00	36.67±3.01	40±0	56±6.92

#### 5.4. Macroinvertebrate metrics and environmental variables relationships

The correlation between water quality variables and macroinvertebrate metrics is represented in (Appendix 6). Pearson's correlation represented some water quality parameters that were significant correlation with macro invertebrate metrics. pH and DO were significant correlation

with family richness; BMWP, FBI, EOT family richness and % Chironomidae ( $p < 0.05$ ). On the other hand DO have strong negatively significant correlation with FBI and % of Chironomidae. Electrical conductivity, total phosphate, nitrate and TDS were negatively correlated with EOT family richness, total family richness, Shannon diversity index, BMWP score and they were not statically significant ( $p > 0.05$ ). Ammonia had a strong negatively significant correlation with total family richness, EOT family richness and abundance. BOD5 and COD were a strong significant relationship with family richness, FBI and % Chironomidae ( $p < 0.05$ )

### 5.5. Human disturbance

During sample collection human disturbance at each sampling sites were identified by physical observation with checklist (Table 7).

Table 7. Summary of human disturbance that identified and registered at Temsa River, Agaro town, 2019

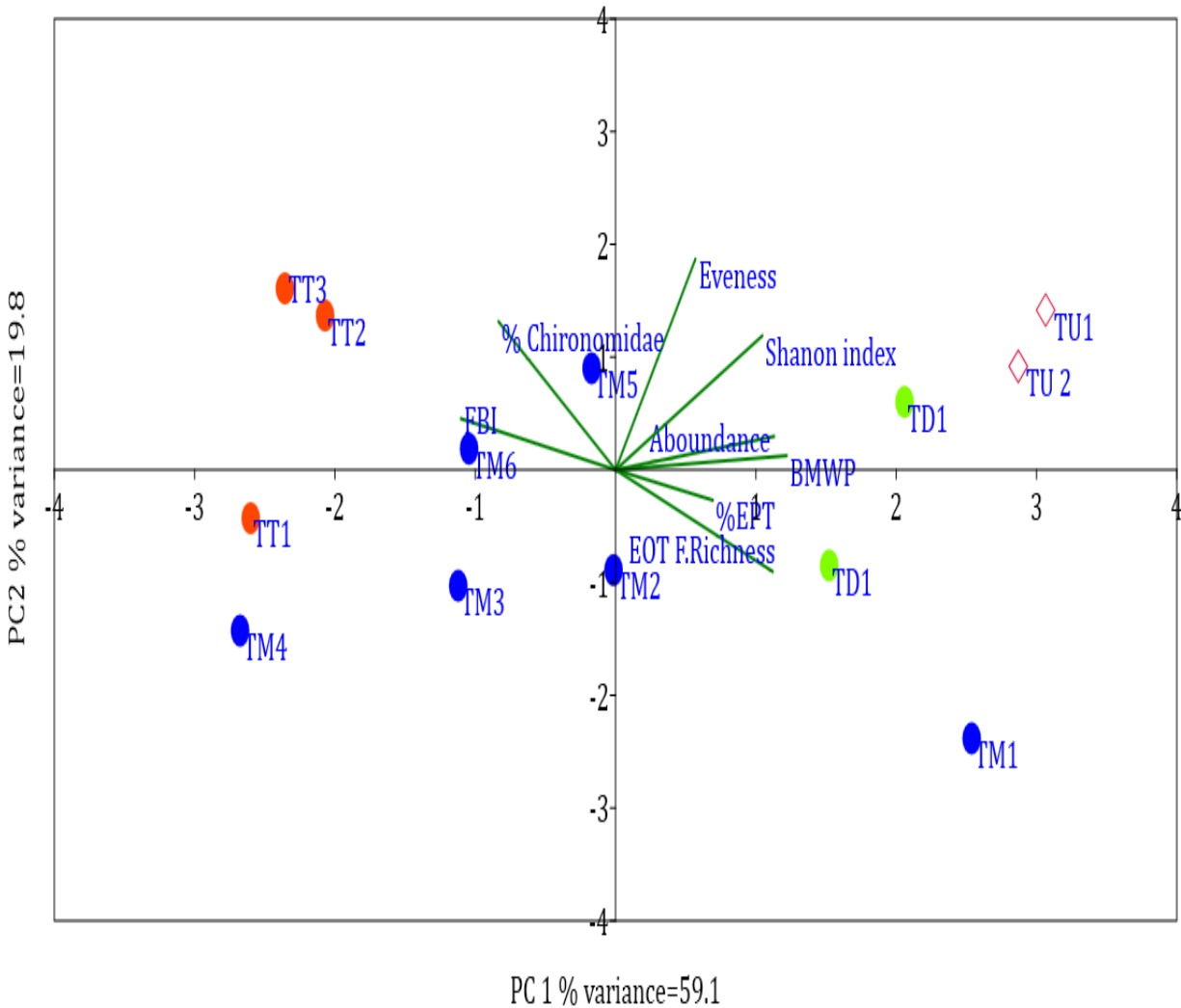
Sample sites	Major human disturbance at sampling sites
Upper stream	Minimal farming, minimal bathing, no visible impact
Middle stream	Solid waste dumping, disposal of domestic sewage, highly car washing, minimal farming, drainage, municipal waste discharges (from hotels, garages,market, from coffee processing) , open bathing, washing clothes, fuel station, grazing, bridge crossing, dumping of wastes, washing clothes, vehicle of washing
Downstream	Minimal farming, removal of vegetation, minimal bathing and washing of clothes,
Tributary	High disposal of domestic sewage( from hotels, markets, solid waste, coffee processing, Open defecation, mostly vehicle of washing (Bajaj, motors etc ) ,drainage, high settlement,

## **5.6. Multivariate analysis**

### **5.6.1. Macro-invertebrate metrics in relation to river sites**

The variability existing of macroinvertebrates assemblages for PC1: explained 59.1% variation of macroinvertebrate observed among sites and PC2: explained 19.8% variation of macroinvertebrate observed among sites. PCA therefore revealed that PC1 and 2 could explain 78.9% of the variation among measured macroinvertebrate metrics which is an indication of good ordination. On PC1 metrics such as abundance, Shannon index, BMWP, EOT family richness and % EPT were a strong positive relationship with upper and downstream sites.

FBI and % Chironomidae were a strong relationship with tributary and some middle sites. Tributary and some middle sites (TM6, TM5 and TM4) had low diversity and richness of macroinvertebrate was an indication of deteriorating water quality at these sites, which may be attributed to organic loads, urban effluents and other anthropogenic activities.

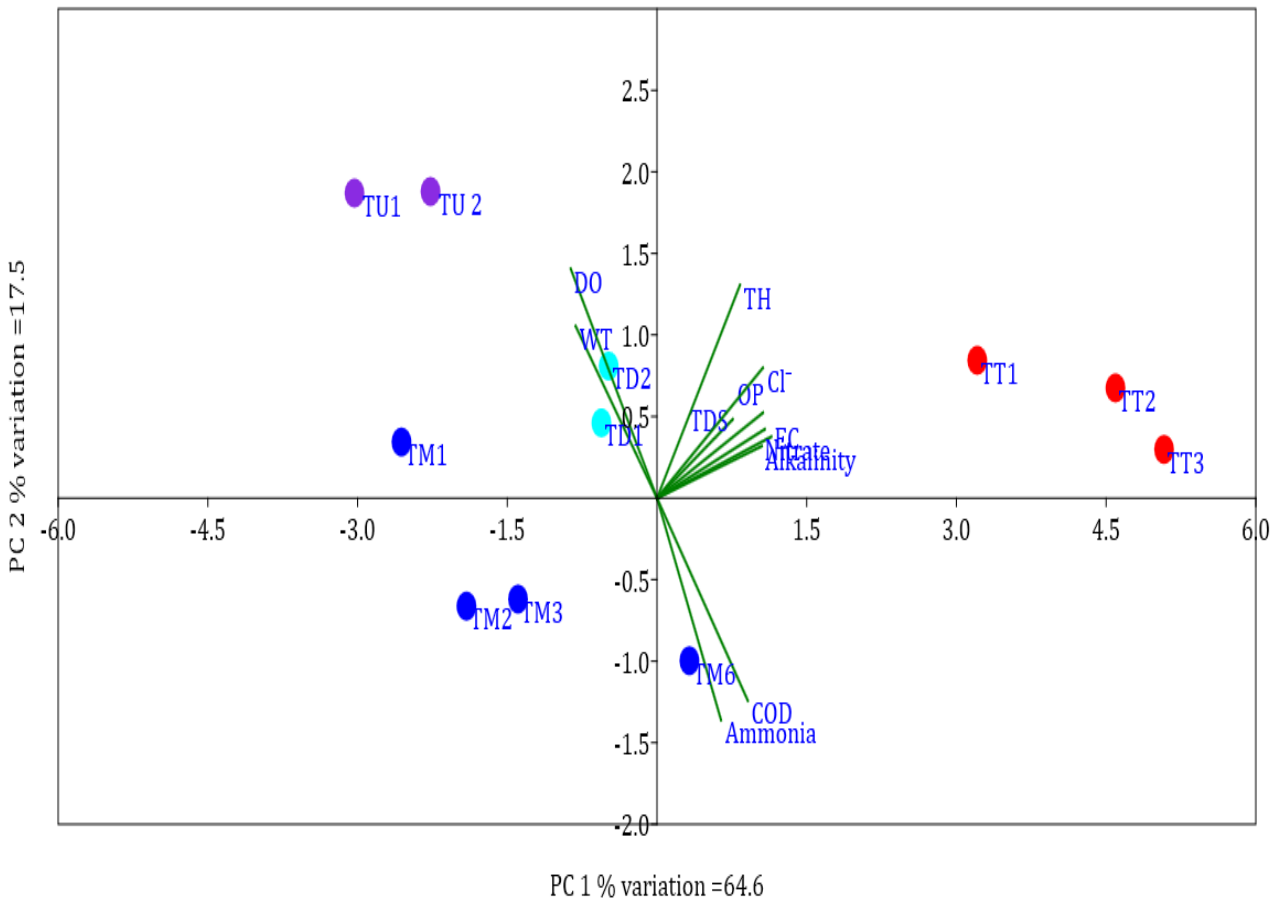


**Figure 6.** Principal Component Analysis (PCA) of sampling sites and benthic macroinvertebrate families.

### 5.6.2. Physico chemical parameters in relation to sampling sites

The variability existing in the abundance data of environmental variables and PC1 explained 64.6% of environmental variables, variation observed among sites, PC2 explained 17.5% of environmental variables variation among sites. The PC1 primarily described the nutrient with positive loadings for phosphate, nitrate, chloride, EC, TDS and alkalinity. PC2 had a strong

positive relationship with DO, water temperature and TH and negative relationship with ammonia and COD. The results of physicochemical analysis of sampling sites at each sampling station were upper stream had the highest DO concentration relative to the rest sites of river; this indicates they were heavily polluted by DO whereas; high concentration of nitrate, EC, TSS was found at tributary sites. Accordingly, PCA result of physicochemical analysis of tributary sites had poor and polluted water quality as compared to other three sites of the river, because of high discharge of wastes, dumping of solid wastes and most anthropogenic impacts takes place at the sites.



**Figure 7.** PCA bi plot of environmental parameters and sampling sites based on the first two components.



## CHAPTER SIX: DISCUSSION

### 6.1. Physicochemical parameters

In the present study, most physicochemical variables, dissolved oxygen (DO), COD, OP, EC, turbidity, alkalinity and nitrate show significant variation among sampling sites ( $p < 0.05$ ). Dissolved Oxygen is an important measure of the extent of pollution, the lower its value, the higher the pollution concentration and vice versa (Mgbemena and Okwunodulu, 2015). It is an important parameter in assessing water quality because of its influence on the organisms living within a body of water (Selvanayagam & Abril 2016). It should be available in sufficient amount as it is essential for good water quality. Therefore, DO levels are an indicator of a water body's ability to support aquatic life.

According to U.S. Environmental Protection Agency, and Alakananda *et al.* (2011)  $DO > 5$  mg/l is considered favorable for growth and activity of most aquatic life; but in the present study about 53.84% of the study sites were below the recommended guideline of USEPA i.e. more than half sites were with  $DO < 5$ mg/L. This may be due to high amount of organic pollution in the river that comes from different sources of discharges from town and high decomposition process. Derso *et al.*, (2015) has reported that organic pollution from animal excrements and sewage discharges from the town is responsible for high turbidity concentration and low DO values. As a result, the current study and Derso *et al.*, (2015) report shows similarity, Oxygen concentration is identified as one of the most important predictors of benthic macroinvertebrate assemblages, where low oxygen concentration leads to the reduction of taxa richness, loss of sensitive taxa and increase of tolerant taxa (Sunderman *et al.*, 2013).

The concentration of phosphate was higher in the tributary sites than in the upper stream sites. Phosphorus is essential for the growth of aquatic plants in a stream. However, phosphate encourages the growth of eutrophication in rivers (Jordan, *et al.*, 2017). As Jordan showed that, the phosphate increase in a fluvial system can be attributed to discharge from both urban, such as wastewater and sewage effluent and agricultural sources from phosphorus-enriched soils. From the sampling sites, phosphate concentration was statistically significant ( $p < 0.05$ )

High values of BOD and COD in streams indicate organic contamination probably originated from domestic sewage (Souto *et al.*, 2011). According to Alakananda *et al.*, (2011) COD concentration is within the permissible limit if it is below 30 mg/L. Study in Ethiopia by Getachew *et al.*, (2011) showed that the nutrients and organic materials as a result of human activities associated with agriculture, deforestation and waste dumping are the major causes of water quality deterioration in streams. In the present study the concentration of Chemical oxygen demand and BOD<sub>5</sub> were > 30 mg/L except at the upper sites, this is due to pollutants coming by runoff from agricultural fields and from urban effluent discharges. However, COD and ammonia showed there was statically significant difference between the four sampling sites ( $P < 0.05$ ).

## 6.2. Macroinvertebrates

The increase in diversity correlates with increasing the health of the assemblage and suggests that niche space, habitat, and food source are adequate to support survival and proliferation of many species (Barbour *et al.*, 1999). In other words, undisturbed habitats are characterized by high diversity of species. While the pollution is increasing, the number of tolerant species increases and sensitive species begin to disappear (Türkmen and Kazanci, 2010). Diversity of taxa is a good indication of the ability of the ecosystem to support varied taxa (Barbour *et al.*, 1999).

The values of the Shannon-Wiener index above 3.0 indicate the habitat structure is stable and balanced; the values below 1.0 indicate there are pollution and degradation of habitat structure (Turkmen and Kazanci, 2010). The index value, usually lies between 1.5 and 3.5 for ecological data and rarely exceeds 4.0 (Seaby and Heanderson, 2007). In the present study, Shannon diversity index revealed that all sampling sites were between 1 to 3 index values indicating the presence of moderate pollution.

Moreover, highest family richness were observed at upper stream and downstream sites having an average value of 16.5 and 14.5 families respectively and the least family richness was observed in the tributary sites having an average value of 10 families. Despite higher sensitive taxa (EOT) at some sites TM1, TU2 and TD2 comprising of 10 and 9 families respectively, there was a little EOT family of other sites such as TT2, TT3, and TM5. This indicates there may be

disturbances at these sites.

One important macroinvertebrate community indicator was the %EPT, or the total number of Ephemeroptera (mayfly), Plecoptera (Stonefly) and Trichoptera (caddisfly) taxa in a sample. An increasing %EPT value correlates with increasing water quality (Rothrock *et al.*, 1998) and many studies have indicated that Ephemeroptera, Plecoptera, and Trichoptera show a strong negative response to anthropogenic disturbances in aquatic ecosystems (Ode *et al.*, 2005). The absence of EPT (Ephemeroptera, Plecoptera, and Trichoptera) throughout this study period is an indication that the stream was polluted and of low biological water quality. Ephemeroptera and Trichoptera comprise a group of organisms, highly sensitive to pollution, requiring clean and well oxygenated waters for their survival. Thus, the occurrence of these taxa is an indication of good water quality (Souto *et al.*, 2011). In our study % EPT were higher in upper stream and downstream. In contrast, tributary site and middle stream of the river had lower % EPT and was positively correlated with dissolved oxygen (DO), pH and negatively correlated with Electrical conductivity (EC) and chemical oxygen demand (COD).

The presences of Dipteran specially the Chironomidae are the indicator of organic pollution (Selvanayagam & Abril, 2016). These tolerant taxa were observed at most sites in this study (Table 6). The reason for this may be the number of sensitive taxa become limited due to their lower tolerance to disturbances; as a result these tolerant taxa became proliferated in number. Chironomids are regarded as highly tolerant families among aquatic macroinvertebrates because of their ability to survive in oxygen depleted environments (Fouche and Vlok, 2010). This characteristic could have contributed to their dominance at the tributary and middle stream sites with high human disturbances and organic loads. In addition, most chironomids feed on fine particulate organic matter, particularly algae, and their increase may be related to high nutrients levels.

Hilsenhoff FBI and BMWP indices were used to assess the organic pollution status of the river using families of macroinvertebrate assemblages and the results were presented in (Table 5). Accordingly upper stream shows FBI with good water quality, or some organic pollution, middle stream were fairly poor or significant organic pollution, downstream were fair or fairly significant organic pollution and tributary sites were very poor water quality or severe organic

pollution. About 38.4 % of the sampling sites are classified as a very poor water quality, which were likely to severe organic pollution because of high human disturbance were recorded and about 46.15% of sampling sites were classified as good and fair water quality that was some organic pollution and had a low human disturbance were recorded at a site.

BMWP is the measure of organic pollution; in calculating these metrics organisms were identified to family level and then each family is allocated a score between 1 and 10 based on literature. The higher the score value, the higher is sensitivity; here according to our finding BMWP was negatively correlated with nitrates, phosphates, electrical conductivity and COD (Appendix 6). This may be due to the sensitive taxa scoring higher value in BMWP resulting in macroinvertebrates sensitive to nutrient pollution and disturbances. In disturbed systems the number of intolerant taxa typically decreases and the proportion of tolerant individuals increase (USEPA, 2002).

According to BMWP only one site show a very good or unpolluted, four sites were good or slightly impacted and eight sites where moderate or moderately impacted. The average value of upper stream and downstream sites of BMWP showed a good water quality or slightly impacted; whereas the middle stream and tributary sites showed a moderate water quality or moderately impacted. The source of this organic pollution may be organic matter/pollution coming from human disturbance activities such as effluent discharge from the town, dumping of wastes, agricultural runoff and vegetation break down (litter decomposition).

### **6.3. Multivariate analysis**

As compared and seen from the PCA (Figure 6) the diversity index of upper stream sites was significantly different from middle stream sites and tributary sites. In other words tributary and middle stream had poor water quality as compared to upper stream sites. Here, this site had higher value relatively and it was polluted may be by pollutants coming by runoff from urban effluent, grazing and agricultural fields. According to Kyriakeas and Watzin (2006), macroinvertebrate communities in Vermont, USA, were highly affected by agricultural activities in the nearby river system. However, there was no significant difference among the sampling sites ( $p > 0.05$ ).

The increase in diversity correlates with increasing the health of the assemblage and suggests that niche space, habitat, and food source are adequate to support survival and proliferation of many species (Barbour *et al.*, 1999). In other words, undisturbed habitats are characterized by high diversity of species. While the pollution is increasing, the number of tolerant species increases and sensitive species begin to disappear (Turkmen and Kazanci, 2010). The results of physicochemical analysis of Temsa thirteen sites at each sampling station were Upper stream had the highest DO concentration relative to the rest sites of river; this indicates they were heavily polluted by DO whereas high concentration of nitrates EC, TSS was found at tributary sites. Accordingly to PCA, the result of physicochemical analysis of tributary sites had poor and heavily polluted water quality as compared to other three sites of the river.

The analysis indicated that metrics such as abundance, family richness, Shannon diversity index, and BMWP and EOT richness were positively correlated with dissolved oxygen, water temperature and negatively correlated with EC, turbidity, COD, ammonia, nitrate and orthophosphate. A similar result was obtained by Orwa *et al.* (2013) in Kenya that there were significant negative correlations of macroinvertebrates abundance and diversity with phosphate and nitrate in Nyando river .i.e. stations with high macroinvertebrate diversity recorded low levels of phosphate and nitrate; the Shannon diversity index also indicates a negative relationship between macroinvertebrate abundance and diversity with nutrient levels. Also (Mereta *et al.*, 2013) obtained a result supporting this result that chemical oxygen demand concentration was negatively related to EOT richness, family richness and filterer-collectors which were core metrics in the development of multimetric index. These all could imply that high nutrient levels, which are indicators of pollution, have a negative impact on macroinvertebrate ecology and diversity.

Percentage of Chironomidae and FBI was strongly correlated positively with nitrate, COD and orthophosphate and negatively correlated with DO saturation and water temperature. The major anthropogenic sources of nitrate in aquatic ecosystem are sewage, fertilizers, and waste from domesticated animals (Ambelu *et al.*, 2013). The source of nutrients may be from discharges from town, fertilizer application in the surrounding agricultural land that comes from runoff to the river; dung droppings by cattle during the dry season that flooded in rain period and riparian

litter break down of oxygen demanding organisms. Decreases in pH may be caused by high organic loads (black water) bacterial process (e.g nitrification or sulfate reduction or oxidation of sulfide sediments) (Baldwin *et al.*, 2005).

## **CHAPTER SEVEN: CONCLUSIONS AND RECOMMENDATION**

### **7.1. Conclusions**

The result suggested that domestic, agricultural and solid waste dumping discharged from urban activities are posing significant effect on the water quality parameters in river. Temsa River deteriorates and decreased as the river flows down from the upstream to downstream due to the entrance (mixture) of tributary which carries different pollutants from the town. Comparing the values of water quality parameters for the Temsa River, it can be concluded that water quality of the river is deviating from the standard with dissolved oxygen, COD, TSS and turbidity at the middle stream where the tributary enters to the river and at the tributary sites itself. Initially the physicochemical water quality of the river was good, but after the mixing of the tributary its quality becomes deviate from the standard (poor) and far from the tributary at the down-stream most physicochemical parameter reverts to the standard except COD.

On the other hand, fluctuations in various macroinvertebrate index parameters were observed at up-stream, mid-stream and down-stream site of the river. From the values of FBI and BMWP it can be concluded that the mid-stream and tributary sampling sites showed the river water quality is under fairly poor water quality and slightly impacted. Additionally, from the taxa richness and macroinvertebrate abundance it can be concluded that the up-stream river site showed good water quality with high EPT taxa, but due to the mixing of tributary the abundance reduced significantly. The river sites with low disturbance (upstream sites) support a higher diversity of macroinvertebrates than highly disturbed sites (midstream sites).

Furthermore, it can be concluded that relatively at tributary sites, there were a high human disturbance, the water quality was very poor, and the degree of pollutant were severely organic pollution. Nutrients such as ammonia, nitrate, orthophosphate and pollutants were the predominantly affecting variables to the river macroinvertebrate assemblages in this study. In general, this study showed that tributary and middle stream was relatively more polluted due to high human disturbance major activities such as farming; effluent discharge, waste dumping and domestic activities had a great role in determining the abundance, diversity and richness of macroinvertebrate assemblages.

## 7.2. Recommendations

Tamsa River is found to be under slightly impact (fairly poor) and is impaired. On the other hand, the river water is used for a variety of purposes such as irrigation, cattle drinking and domestic purposes without prior treatment.

- ❖ For sustainable management of this water resource, environmental protection agencies at different levels and other concerned administrative and/or nongovernmental bodies should take strict as well as technical measures in order for tackle the upcoming impacts in the river and not to further affect the river.
- ❖ In addition to this continuous monitoring of the river using parameters such as those used in this study should be employed to assess timely status of the river.
- ❖ The Agaro town municipality should strictly regulate the activities of human to minimize the impact of human disturbance on the river.
- ❖ The community also has to see the river as their assets and combat any human influence or impact on the river to minimize the human disturbance.
- ❖ All stakeholders (mainly the Agaro town municipality) have to plan, to protect, regulate and manage the river in the study area in order to tackle the impact.
- ❖ Further study should be done especially on seasonal variation and microbial contamination on study the area



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## Appendixes

### Appendix 1. Sampling protocol

#### General information

1. DD/MM/YYYY-----Time-----
  2. Name of River-----Sampling station-----
  3. Altitude (m) -----coordinates-----
  4. Weather condition -----
  5. Previous day rain history -----
  7. Size of site under assessment (ha) -----
  8. Size of total (ha)-----
- Physico-Chemical parameters (Field)
9. Water temperature (0c) -----DO (mg/l)-----EC( $\mu$ S/cm)-----pH-----
  10. Turbidity (NTU) -----

#### Physicochemical parameters (laboratory)

11. COD-----NO<sub>2</sub>-----
12. NH<sub>4</sub>-----TP-----
13. TSS-----PO<sub>4</sub><sup>3-</sup>-----
  
14. BOD<sub>5</sub>-----NO<sub>3</sub>-----

#### Land use

15. Adjacent land use pattern
  - a. Agriculture tilled----- e. road-----
  - b. Pasture----- f. commercial-----
  - c. Native vegetation----- g. industrial-----
  - d. Residential area----- h. recreational-----
16. Anthropogenic activities
  - b. Tree removal -----
  - c. Tree plantation -----
  - d. grazing -----

- e. Grass cutting -----
- f. Car washing -----
- g. Clay mining/pottery -----
- h. Waste dumping -----
- i. swimming -----
- j. Other potential threats
  - a. Agricultural biocides-----
  - b. Point source pollution-----
- 18. Wetland ecological state
  - a. Unmodified, natural-----
  - b. Largely natural with few modification-----
  - c. Moderately modified-----
  - d. Largely modified-----
  - e. Seriously modified-----
  - f. Critically/extremely modified-----
- 19. Any additional comments -----

Appendix 2. Data of Macroinvertebrate assemblages collected at each sampling sites

Macroinvertebrates families	Sampling sites												
	TU1	TU 2	TM1	TM2	TM3	TM4	TM5	TM6	TD1	TD1	TT1	TT2	TT3
Aeshaidae	8	3	2	4	1	0	0	0	1	0	0	0	0
Baetidae	3	2	2	0	5	2	0	0	0	0	0	0	0
Belostomatidae	0	0	0	0	0	0	4	3	0	0	2	6	4
Caenidae	0	0	0	0	0	4	2	3	1	11	3	2	2
Calopterygidae	3	11	3	5	0	0	0	0	0	0	0	0	0
Ceratopogonidae	0	0	0	1	0	0	3	0	0	0	0	1	3
Chironomidae	0	0	0	3	8	10	14	9	5	7	7	11	15
Chlorocyphidae	4	2	2	1	0	0	0	0	0	0	0	0	0
Coenagrionidae	10	8	16	17	16	13	0	0	2	27	2	0	0
Corixidae	0	0	0	1	0	0	1	3	0	0	0	1	0
Culicidae	0	0	0	0	0	0	1	3	4	2	0	0	2
Dixidae	6	0	1	0	0	0	0	0	0	0	0	0	0
Dytiscidae	1	1	1	0	2	0	0	0	0	0	0	0	0
Ecnomidae	0	0	0	0	1	0	0	0	0	3	0	0	0
Elmidae	0	0	0	0	0	0	0	2	0	0	0	0	2
Ephydriidae	0	0	0	0	0	0	2	0	0	0	1	0	0
Gerridae	0	0	0	0	0	1	4	1	1	0	2	0	0
Gomphidae	5	2	2	5	0	0	0	0	3	2	0	0	0
Gyrinidae	0	3	2	0	0	1	0	0	0	3	0	0	0
Helodidae	1	3	1	0	1	0	0	0	0	0	0	0	0

Heptageniidae	6	3	3	0	0	0	0	0	7	4	0	0	0
Hirudinae	0	0	0	0	0	0	3	1	4	0	0	2	6
Hyrophilidae	3	0	0	0	0	0	0	0	1	0	0	0	0
Hydropsychidae	0	11	3	0	4	0	11	2	13	3	0	0	0
Lestidae	0	0	0	0	0	0	6	0	0	1	3	3	0
Lymnaeidae	0	0	0	0	0	0	0	6	1	3	1	3	4
Libellulidae	0	0	0	0	0	0	0	3	0	2	0	0	3
Leptophlebiidae	5	3	0	0	0	0	0	0	3	0	0	0	0
Naucoridae	3	7	15	2	0	0	0	0	0	0	0	0	0
Nepidae	0	1	0	0	0	0	0	0	0	0	0	0	0
Notonectidae	2	2	4	1	0	0	0	0	0	0	0	0	0
Oligochaeta	0	0	0	0	0	1	0	0	0	0	0	0	0
Philopotamidae	0	0	0	2	0	1	0	0	0	2	0	0	0
Potamanautidae	0	0	0	0	0	0	1	0	0	0	0	0	0
Pyralidae	2	3	0	0	0	0	0	0	0	0	0	0	0
Tabanidae	0	1	1	2	3	0	1	0	0	0	0	0	0
Tipulidae	5	0	1	0	0	0	0	0	0	0	0	0	0
Simulidae	0	0	0	0	0	1	0	2	0	0	0	0	0
Syrphidae	0	0	0	0	0	0	1	3	5	0	1	4	2
Sphaeriidae	0	0	0	0	0	0	0	1	0	2	0	0	0
Veliidae	0	0	0	0	0	0	0	9	6	0	5	3	1

Appendix 3. List of Macroinvertebrate taxa tolerance value: Orders and Families encountered in this study with indication of their Functional feeding group (FFG) and Tolerance value

Order	Family	Feeding group	Ts	Referance
Odonata	Aeshnidae	Predators	5	Bode et al., 1996
	Calopterygidae	Predators	8	Hauer and Lamberti, 1996
	Coenagrionidae	Predators	6	Plafkin et al., 1989
	Gomphidae	Predators	1	Plafkin et al., 1989
	Libellulidae	Predators	9	Hauer and Lamberti, 1996
	Chlorocyphidae	Predator	2	Mandaville, 2002
	Lestidae	Predator	9	Plafkin et al., 1989
Ephemeroptera	Baetidae	Gathers-Collectors	4	Hauer&Lamberti, 1996
	Heptagenidae	Scrapers	4	Hauer&Lamberti, 1996
	Leptophlebiidae	Gatherer-coll.	2	Plafkin et al., 1989
	Caenidae	Gatherer-collector	7	Plafkin et al., 1989
Diptera	Culicidae	Collectors-filters	8	Bode et al., 2002
	Dixidae	Gathers-Collectors	1	Mandaville, 2002
	Ephydriidae	Gathers-Collectors	6	Hauer&Lamberti, 1996
	Ceratopogonidae	Predator	6	Hauer and Lamberti, 1996
	Chironomidae	Predator,Gathcolle	8	Bode et al., 1996
	Simuliidae	Filterer-collector	6	Bode et al., 1996
	Syrphidae	Gatherer-collector	10	Hauer and Lamberti, 1996
	Tabanidae	Predator	6	Hauer and Lamberti, 1996
	Tipulidae	Shredder/Gatherer	3	Hauer and Lamberti, 1996
Coleoptera	Dytiscidae	Predators	5	Babour et al., 1999
	Elmidae	Predators	5	Hauer&Lamberti, 1996
	Gyrinidae	Predators	4	Babour et al., 2002

	Helodidae	Predators	10	Mandaville, 2002
	Hydrophilidae	Gathers- Collectors	5	Hauer&Lamberti, 1996
Hemiptera	Belostomatidae	Predators	10	SPS, 1999
	Corixidae	Collectors	9	Barbour et al., 1999
	Gerridae	Filters	8	Deemool&Prommi, 2015
	Naucoridae	Predators	5	SPS, 1999
	Veliidae	Predator	6	Bode et al., 1996
	Nepidae	Predator	5	Hauer and Lamberti, 1996
Trichoptera	Ecnomidae	Filterer -collector	6	SPS, 1999
	Hydropsychidae	Filterer-collector	4	Hauer and Lamberti, 1996
	Notonectidae	Predators	5	Plafkin et al., 1989
	Philopotamidae	Filterer-collector	3	Hauer and Lamberti, 1996
Lipoptera	Pyralidae	Scrapers	5	Hauer&Lamberti, 1996
Hirudinae	Hirudinea	Predators	10	Barbour et al., 1999
Gastropoda	Lymnaeidae	Scraper	6	Barbour et al., 1996
Decapoda	Potamonautidae	Predators	7	Plafkin et al., 1989
Bivalvia/ Pelecypoda	Sphaeriidae	Filterer-collector	8	Barbour et al., 1999
Oligochaeta	oligochaeta	Gatherer-collector	8	Bode et al., 2002



Appendix 4. Biological Monitoring working party (BMWP) tolerance score of benthic macroinvertebrates.

BMW score	Macroinvertebrate families
8	Aeshnidae ,calopterygidae, coenagrionidae, Gomphidae, Lestidae Libellulidae, Philopotamidae, Potamanautidae ,corixidae
4	Baetidae
6	Belostomatidae, Ceratopogonidae
2	Chironomidae, Chlorocyphidae, Syrphidae
5	Culicidae, Dytiscidae, Elmidae, Ephydriidae, Gerridae Hydrophilidae, Hydropsychidae, Naucoridae, Nepidae Notonectidae, Simuliidae Gyrinidae, Helodidae, Hirudinae, Tipulidae, Veliidae
10	Dixidae, Ecnomidae, Heptageniidae, Leptoceridae, Leptophlebiidae, Pyralidae
3	Sphaeriidae, Lymnaeidae
1	Oligochaeta
0	Tabanidae

Appendix 5.Physicochemical parameter of water samples of the thirteen sampling sites of the river

parameter s	Sites												
	TU1	TU 2	TM1	TM2	TM3	TM4	TM5	TM6	TD1	TD1	TT1	TT2	TT3
WT	27.2	26.3	25.6	27.1	25	22.4	17.6	19.2	21.3	22.8	22.3	20.7	18.4
PH	8.05	8.09	7.53	7.47	7.32	6.29	5.74	6.3	6.82	7.73	5.78	6.8	4.9
DO	7.58	7.2	5.7	5.31	4.52	4.47	3.23	4	6.08	5.7	4.19	3.33	3.39
EC	78.2	75.3	78.8	77	80.5	83.3	113	117.4	118.5	118.3	215	254	279
Turbidity	33.5	33	42.3	44.3	41.4	48.7	65.8	132	120	128	182	213	288
Nitrate	1.41	1.37	1.5	1.46	1.8	1.86	2.06	4.75	4.99	4.75	6.3	6.94	9.25
Nitrite	0.02	0.02	0.03	0.08	0.04	0.06	0.04	0.21	0.18	0.16	0.3	0.69	1.43
TP	1.19	1.01	1.16	1.16	1.29	1.19	1.59	2.05	1.96	2.39	2.51	2.75	3.03
OP	0.34	0.58	0.34	0.37	0.4	0.55	0.28	0.77	0.4	0.67	0.92	1.19	1.25
Ammonia	0.1	0.1	0.12	0.62	0.82	1.38	0.72	0.55	0.15	0.1	0.58	1.15	0.98
TDS	104	92	104	80	120	128	124	160	164	160	844	372	240
TSS	0.48	0.44	0.54	0.592	0.456	0.584	0.68	1.056	1.156	1.064	2.192	2.684	3.404
BOD	16.75	20.45	23	43.6	38.4	53.4	71.4	51.6	44.3	37.1	61.8	64.2	68.4
COD	19.47	22.89	35.3	64.84	43.55	68.46	83.7	66.49	55.86	47.69	80.47	71.78	92.47
Cl <sup>-</sup>	10	12	11.5	10	11.5	10	10	14	16	16	19.99	21.99	21.99
Alkalinity	28	32	28	32	32	28	36	28	32	32	40	48	44
TH	48	48	36	36	40	32	36	40	40	40	48	60	60
Ca <sup>2+</sup>	28	28	20	20	24	24	24	28	32	28	40	36	44
Mg <sup>2+</sup>	20	20	16	16	16	8	12	12	8	12	8	24	16

Appendix 6. Pearson's correlation of macroinvertebrate metrics and environmental variables using

	Abundance	Family Richness	Shannon index	Evenness	FBI	BMWP	Simpson index	EOT FR	Diptera n FR	% Chir.	% of Ephemera	% of Trich.	%EPT
WT	.718	.872	.867	.679	-.884	.788	-.773	.789	-.953*	-.876	.518	.416	.469
pH	.948	.995**	.917	.594	-.994**	.969*	-.663	.978*	-.841	-.998**	.785	.768	.805
DO	.944	.992**	.976*	.730	-.953*	.979*	-.778	.960*	-.888	-.973*	.853	.717	.808
EC	-.778	-.815	-.593	-.134	.912	-.759	.244	-.839	.565	.875	-.458	-.747	-.640
Turbidity	-.696	-.792	-.612	-.220	.892	-.705	.344	-.778	.666	.849	-.357	-.587	-.497
Nitrate	-.620	-.742	-.577	-.219	.847	-.639	.349	-.712	.673	.800	-.270	-.488	-.396
TP	-.592	-.732	-.590	-.265	.832	-.622	.396	-.688	.709	.786	-.248	-.430	-.352
NH3	-.992**	-.947	-.886	-.581	.896	-.986*	.598	-.971*	.678	.927	-.958*	-.888	-.963*
TDS	-.790	-.808	-.573	-.097	.905	-.761	.203	-.844	.522	.868	-.476	-.785	-.672
TSS	-.758	-.806	-.587	-.137	.906	-.742	.251	-.823	.577	.866	-.429	-.715	-.607
BOD	-.899	-.980*	-.931	-.651	.979*	-.937	.727	-.941	.902	.981*	-.729	-.671	-.720
COD	-.890	-.977*	-.943	-.684	.969*	-.934	.757	-.931	.920	.974*	-.732	-.646	-.707
Cl <sup>-</sup>	-.601	-.701	-.500	-.104	.823	-.604	.235	-.691	.580	.770	-.233	-.522	-.400
Alkalinity	-.793	-.806	-.568	-.088	.902	-.762	.192	-.845	.509	.867	-.483	-.796	-.682
TH	-.418	-.327	.013	.520	.473	-.318	-.441	-.446	-.123	.419	-.115	-.679	-.450
Ca	-.523	-.555	-.271	.202	.706	-.481	-.078	-.598	.292	.644	-.140	-.593	-.402
Mg	.014	.252	.477	.728	-.205	.149	-.772	.083	-.736	-.212	-.002	-.405	-.252

\*. Correlation is significant at the 0.05 level (2-tailed).\*\*. Correlation is significant at the 0.01 level (2-tailed).

Appendix 7. Summary of Human disturbances identified at the sampling sites

Human disturbance activities	Sampling sites												
	TU1	TU 2	TM1	TM2	TM3	TM4	TM5	TM6	TD1	TD1	TT1	TT2	TT3
Liquid waste discharge	x	x	x	x	√	√	√	√	x	x	√	√	√
Car washing	x	x	x	√	√	√	√	x	x	x	√	√	√
Drainage	√	√	√	√	√	√	√	√	√	√	√	√	√
Effluent discharge	x	x	x	√	√	√	√	√	√	√	√	√	√
Agriculture	√	√	√	x	x	x	x	x	√	x	x	x	x
Fuel station	x	x	x	x	x	√	√	√	x	x	√	√	√
Grazing	√	√	√	√	x	√	√	√	√	√	x	x	√
Irrigation	x	x	√	√	√	x	x	x	x	x	x	x	x
Open defecation	x	x	x	x	√	√	√	√	√	√	√	√	√
Settlement	x	x	x	√	√	√	√	√	x	x	√	√	√
Solid waste dumping	x	x	x	√	√	√	√	√	√	√	√	√	√
Swimming	x	x	√	√	x	x	x	x	√	√	x	x	x
Vegetation Removal	x	√	x	√	x	x	x	x	√	√	x	x	√
Washing	x	√	√	√	√	√	√	√	√	√	√	√	√