PHYSICOCHEMICAL CHARACTERIZATION AND APPLICABILITY OF THE MINI STREAM ASSESSMENT SCORING SYSTEM AND THE SOUTH AFRICAN SCORING SYSTEM VERSION 5 IN THE UPPER AWASH BASIN, ETHIOPIA

By

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1 PREFACE

The research contained in this dissertation was completed by Zizile Yoliswa Jele while based in the Discipline of Hydrology, School of Agricultural, Earth and Environmental Sciences of the College of Agriculture, Engineering and Science, University of KwaZulu-Natal, Pietermaritzburg campus, South Africa. The REACH programme financially supported the research.

The contents of this work have not been submitted in any form to another university, and except where the work of others is acknowledged in the text, the results reported are due to investigations by the candidate.

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ABSTRACT

The Sustainable Development Goal (SDG) 6, introduced by the United Nations in 2015, focus on ensuring the availability and sustainable management of water, also encompassing the quality and sustainability of freshwater resources. Central to achieving SDG 6 is target 6.3, which aims to improve water quality by 2030. An essential indicator for this goal is SDG 6.3.2, tracking the proportion of water bodies with good ambient water quality. SDG 6.3.2 requires monitoring programmes based on relevant and measurable parameters. However, Africa faces challenges in implementing conventional physico-chemical water quality monitoring due to limitations such as scarce testing facilities, resource constraints, and expensive logistics. Therefore, a complementary approach is needed and in the context of this study, Biomonitoring is regarded as a necessary strategy. Biomonitoring involves assessing river health using aquatic macroinvertebrates. Research indicates successful applications of Biomonitoring in various countries, particularly in South Africa, where Biomonitoring tools like the South African Scoring System version 5 (SASS5) and the simplified Stream Assessment Scoring System (miniSASS) have been developed, tested and used. SASS5 is a standardized method that evaluates the presence and richness of macroinvertebrates, acting as sensitive indicators of water quality. MiniSASS is designed for non-experts, supports citizen science initiatives and aligns with SDG 6. b. The study aims to test the applicability of SASS5 and miniSASS in intermittent rivers and ephemeral streams in the Upper Awash River in Ethiopia. This region, situated in the Ethiopian highlands, represents a tropical area with high biodiversity and prominent water quality challenges. The research site choice is strategic, considering tropical rivers and more especially intermittent rivers and ephemeral streams which are often overlooked in water quality assessments. The study employs both Biomonitoring and physicochemical monitoring, including heavy metals. Principal Component Analysis (PCA) results highlight an inverse correlation between Biomonitoring metrics and heavy metals. The clustering of heavy metals in opposite end of the cluster of Biomonitoring scores reveals that Biomonitoring could be a good index for water quality even in relation of pollution with heavy metals. This research contributes to advancing scientific knowledge and addresses the applicability of South African Biomonitoring tools in diverse environmental contexts beyond their country of origin.

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LIST OF ABBREVIATIONS

ASPT	Average Score Per Taxa
Ave	Average
Cd	Cadmium
Cr	Chromium
DO	Dissolved Oxygen
DS	Dry Season
EC	Electrical Conductivity
ETH EPA	Ethiopia Environmental Protection Authority
Fe	Iron
Hg	Mercury
K	Potassium
Mg	Magnesium
MiniSASS	Streams Assessment Scoring System
Mn	Manganese
Pb	Lead

PCA	Principal Component Analysis
SASS5	South African Scoring System version 5
SDGs	Sustainable Development Goals
TDS	Total Dissolved Solids
Temp	Temperature
WS	Wet Season
Zn	Zinc

1. INTRODUCTION

1.1 Background

As Africa's population continues to grow and industrial and agricultural sectors advance, the foreboding trend is the decreasing quality of its rivers. As a result, the Sustainable Development Goal (SDG) 6 which was set forward by the United Nations in 2015 to ensure the availability and sustainable managements of water and sanitation, was set to restore the good quality status of freshwater resources (Kirschke *et al*. 2020). Central to achieving SDG 6 is goal 6.3 which aims to improve water quality by 2030 (Kirschke *et al*., 2020). To achieve such a target, SDG indicator 6.3.2 is crucial. "This indicator tracks the percentage of water bodies in a country with good ambient water quality" (UN Water, 2018). Therefore, water quality monitoring programmes based on relevant and measurable parameters are required (Kirschke *et al*. 2020). However, water quality testing facilities and capabilities are limited in Africa, making it difficult to monitor water quality (Chen *et al*., 2022). In addition, the lack of personnel, equipment, and logistical resources required to operate a water quality testing program in low-income settings pose additional challenges (Peletz *et al*., 2018). Therefore, a complementary approach to physicochemical analysis has been in effect, which is known as Biomonitoring monitoring (Mangadze *et al*., 2019). Assessing the status of water quality through Biomonitoring refers to the use of biomarkers/ bioindicators to monitor the health of rivers. In the context of this study, macroinvertebrates are regarded as the bioindicators. Biomonitoring in Africa is an important aspect because it offers the opportunity to overcome capacity constraints in water physicochemical water quality monitoring challenges. It also offers the opportunity to evaluate the impact of pollutants on aquatic ecosystems and provides insights into potential health risks to human populations (Dalu and Froneman 2016).

Furthermore, Biomonitoring can measure environmental health by combining stressors over a long time (Obubu *et al*., 2021). Biomonitoring can assess all rivers due to the bioindicators being globally present and diverse; it is less expensive compared to physicochemical analysis as it does not require intensive equipment, chemicals and reagents, the results produced can be comparable when the same protocols are use (Obubu *et al*., 2021). Lastly it can be understood by the public (Obubu *et al*., 2021). Initially, most of the indicator species that have been used to develop Biomonitoring approaches were based on the following five indicators, phytoplankton, macrophytes, benthic diatoms, macroinvertebrates, and fishes (Abdelkarim 2020). According to Bere (2016), diatoms are one the indicators that have shown patterns in Zimbabwe.

However, in most African countries, there are limited, or insufficient data and physicochemical water quality monitoring initiatives undertaken by professional scientists and government agencies, which hinders Biomonitoring (Aura *et al.*, 2021). To assist with some of the shortcomings, Biomonitoring also consists of citizen science tools that allow the general public to participate. These tools contribute towards SDG 6, more specifically SDG 6.b, which promotes stakeholder participation and help provide data.

1.2 Problem Statement

According to Abdelkarim (2020), many poor countries are unable to achieve noticeable changes in their aquatic systems due to a lack of reliable and consistent monitoring procedures, and the activities they take are insufficient to produce strong and effective improvement programs. Insufficient financial and technical assistance for monitoring water quality introduces a higher level of uncertainty when creating water resource management strategies, which can lead to waste investment and an inability to implement useful pollution control measures (Abdelkarim, 2020). African countries are prime examples of such. Furthermore, in many African countries, citizen science is rarely used to monitor rivers which can aid in assisting with insufficient data about the river water quality environment (Masese et al., 2019).

Although rivers in tropical Africa are known to be more diverse than other areas, little is known about their macroinvertebrate taxonomy and ecology (Elias, 2021). The potential application of tropical macroinvertebrate species in the development of Biomonitoring method(s) that can precisely assess the health of riverine ecosystems has been hampered by this knowledge gap (Elias, 2021). Resulting in the adoption of biological monitoring programs from foreign, non-tropical regions to monitor rivers (Elias, 2021).

However, to confirm the existence of generic adoption principles among macroinvertebrate-based methodologies from other regions, macroinvertebrate taxa in temperate regions do not necessarily coincide with those in the tropics (Elias, 2021). Therefore, such taxonomic problems and contradicting features of adoption of tropical Biomonitoring methods, necessitates the need to continuously test the applicability of Biomonitoring methods developed in other regions (Elias, 2021). Additionally, it is commonly accepted that aquatic organisms are useful indicators for assessing various levels of human impact (Masese *et al*., 2013). This has been accomplished by carefully analysing biological and ecological responses along human disturbance gradients, identifying indicator assemblages and species among assemblages with known responses to human alterations, identifying driving variables acting on aquatic ecosystems, and improving statistical techniques and approaches to detect effects across different types of aquatic ecosystems and regions (Masese *et al*., 2013). Although most African countries lack in the development of Biomonitoring tools according to (Masese *et al*., 2021), South Africa is an exception. Scientists in South Africa have developed SASS5 and miniSASS (citizen science initiative) to assess the health of rivers. These tools, adjusted from temperate regions, have the potential to form the foundation for the development of Biomonitoring tools for different African countries and also serve as a standard aquatic Biomonitoring tool in Africa under different hydro-ecology settings. Additionally, Dessie *et al*. (2023) regard miniSASS as an attractive Biomonitoring tool due to its citizen science initiative, cost-effectiveness, and successful implementation in other countries.

Ethiopia is referred to as the water tower of East Africa mainly because of several large rivers that stem from the Ethiopian highlands and flow into neighboring countries (Awoke *et al*., 2016). However, the rivers' quality is declining (Dabessa *et al*., 2022). Among those rivers, the Awash basin suffers the most impairment because it is the most utilised basin in the county (Assegide *et al*., 2022). Akaki and Kebena rivers are the Awash River tributaries that flow through Addis Ababa and are examples of the most contaminated rivers in the country (Awoke *et al*., 2016; Yohannes and Elias, 2017). The leading causes of water quality degradation include agricultural fields that contaminate the basin through point and non-point sources, lack of sanitation infrastructures, untreated domestic discharge from the city and uncontrolled urbanisation, all due to rapid population growth (Assegide *et al*., 2022). Recent studies in Ethiopia indicated that surface water pollution is high, and there is little information about the current ecological status of the rivers and streams (Awoke, 2016). This may be due to the lack or absence of regular monitoring programs, especially in intermittent rivers and ephemeral streams, which are often overlooked (Miliša *et al*., 2022). Though physicochemical measurements commonly form the basis of monitoring, they cannot account for the impact of various environmental stressors (Emana and Dubie, 2021).

In Ethiopia, there is a need to involve citizen scientists in water quality monitoring, but there is a lack of empirical evidence on how citizen scientists address this urgent monitoring needs, accuracy of citizen science data, and strengths and weaknesses of citizen science approaches with cost-effective, quick, and easy-to-use methods (Babiso *et al*., 2023).

Therefore, this leads to the research aim based on the presumption that living organisms are suitable pointers of ecological quality: "When water no longer supports living things, it will no longer support human affairs" (Emana and Dubie, 2021). Biomonitoring, in the context of the study, is regarded as the complementary monitoring approach needed to assess the health of rivers. Furthermore, Feio *et al*. (2021) state that assessing river or stream conditions is, in theory, a first step toward rehabilitating rivers that do not fulfill the quality criteria. Dabessa *et al*. (2022) further state that water quality monitoring is necessary to identify the current state of water quality and improve environmental conditions and public health problems.
The study is therefore guided by the hypothesis, research questions, aims and objectives as explained in the following sections.

1.3 Hypothesis

It is hypothesised that the Biomonitoring metrics will positively identify water pollution and ecological effects in the study area. This is predicated on the existence of widespread macroinvertebrate species, which are expected to share environmental tolerances with South African systems.

1.4 Research Aim

To test the applicability of Biomonitoring tools, SASS5 and miniSASS, in intermittent rivers in the Upper Awash River in Ethiopia, supported by concurrent physicochemical monitoring and heavy metal and other metal concentrations.

1.5 Objectives

The specific objectives that will be used to achieve the aim are listed below:

- To assess physicochemical parameters (Temperature, pH, dissolved oxygen (DO), electrical conductivity (EC), and Clarity) as well as heavy metal concentrations and other metals in selected rivers/streams.
- To conduct a miniSASS and SASS5 test in the selected rivers/streams during the peak wet and dry seasons.
- To examine the health of the stream/river by identifying existing aquatic macroinvertebrates.
- To compare the ease of use and effectiveness of miniSASS, SASS5, and testing of physicochemical parameters and heavy metals.
- To identify macroinvertebrate species, given that they are not recognized in the current SASS5 and miniSASS score sheets.

1.6 Research Questions

- How effectively do the South African Scoring System SASS5 and miniSASS Biomonitoring tools capture the ecological health of intermittent rivers in tropical regions?

1.7 Anticipated Outcomes

- A lack of suitable biotopes and few existing macroinvertebrates are expected in the river due to changes in stream channel conditions (Intermittency of the rivers) and water scarcity during the peak dry season.
- During the wet season, identifying the macroinvertebrates will be challenging due to the large amounts of substrate in the samples.
- The macroinvertebrates identified in the river may differ from the current list proposed in the miniSASS/SASS5.

1.8 Structure of Thesis

Figure 1.1 below displays how the thesis is divided into six sections. Section one provides the introduction for the study and provides the background information, problem statement, hypothesis, research aims and objectives, research questions, and anticipated outcomes. The second section provides a detailed examination of the literature relevant to the study. Section three describes the methodological approach undertaken in the study, the chosen study area, and the data sources used to carry out the study. Section four presents the results obtained, and Section five provides an analysis and discussion. Lastly, section 6 presents a summary of the research, recommendations, and concluding remarks.

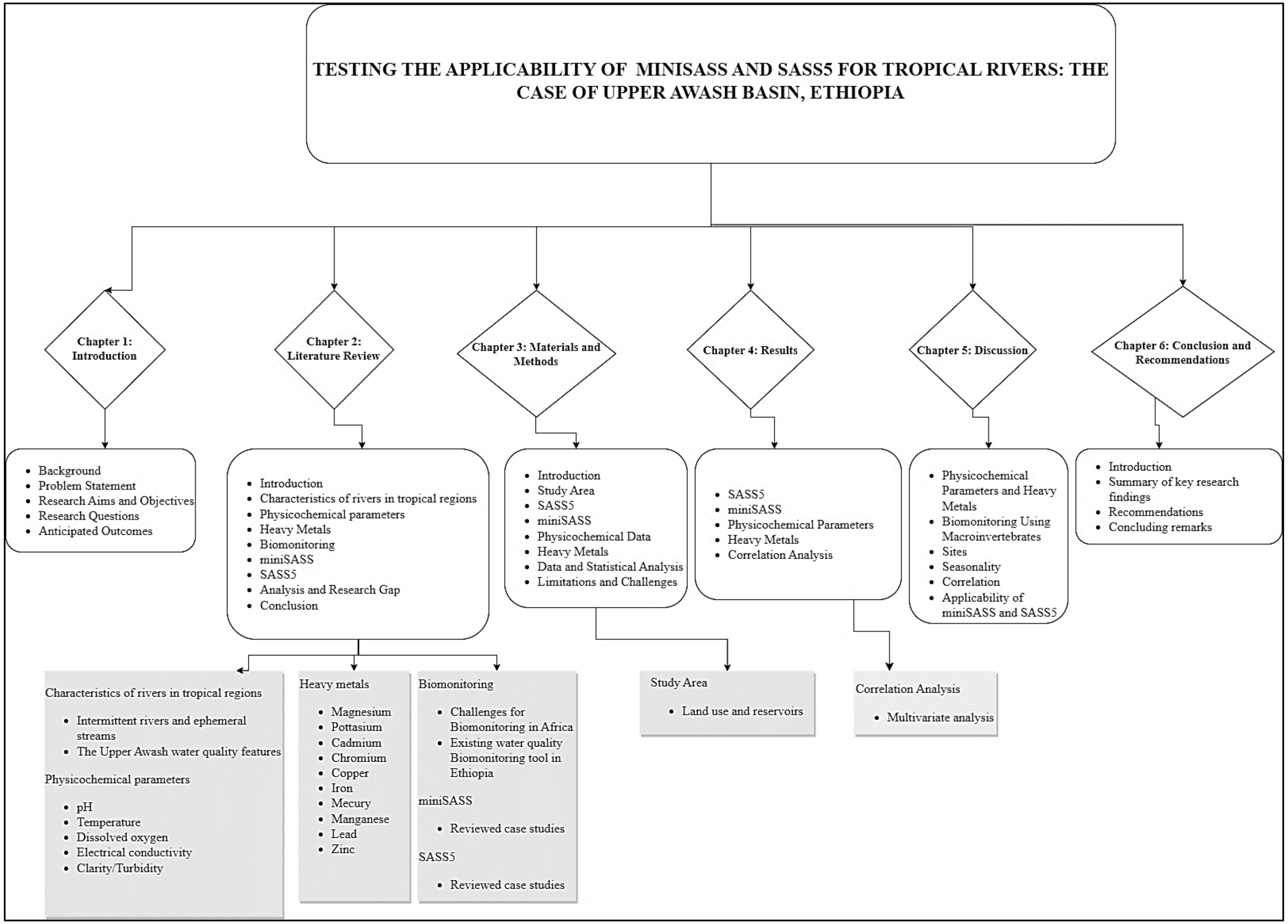


Figure 1.1 Structure of the thesis

2 LITERATURE REVIEW

2.1 Introduction

This section provides an overview of the Biomonitoring tools that may be beneficial to assess surface water quality. Primarily, an overview of the characteristics of rivers in tropical regions is given, followed by information on intermittent rivers and ephemeral streams and their prevalence is given, then followed by Ethiopian rivers' water quality changes, after that the conventional use of physicochemical parameters and heavy metals to assess the water quality of rivers. Then followed by the introduction to the concept of Biomonitoring to monitor rivers and streams and the challenges of Biomonitoring in African areas as well as Biomonitoring tools that have been applied in Ethiopia. Lastly, a review of biological tools, miniSASS and SASS5, is provided.

2.2 Characteristics of Rivers in Tropical Regions

In tropical African areas, there can be notable differences in river flow during the wet and dry seasons. Due to the movement of the Intertropical Convergence Zone (ITCZ) and the seasonal monsoon winds, many regions in Africa experience distinct wet and dry seasons, which are characteristics of their tropical climate (Syvitski *et al*., 2014). In the context of the study, Ethiopia’s rainfall patterns, both temporally and spatially, are significantly influenced by the passage of air masses connected to the InterTropical Convergence Zone (ITCZ) (Lemman *et al*., 2017). An outline of the normal seasonal variations in the flow of these rivers is as follows (Syvitski *et al*., 2014):

The wet season is frequently marked by heavy and continuous rainfall. This time is primarily associated with the arrival of the ITCZ, which brings moist air and causes the convergence of air masses, resulting in extensive precipitation. As a result, rainfall and runoff from saturated soils provide a considerable influx of water to the rivers. This results in a significant increase in river discharge, and many rivers experience higher water levels. As a result, some rivers overflow their banks, flooding surrounding areas. Floodplains and low-lying areas may become affected during this period. The increased flow of water during the wet season aids in the transportation of sediments downstream. Rivers may have a larger concentration of suspended particles and debris, which affects water clarity.

The dry season is distinguished by reduced rainfall. The ITCZ shifts away from the area, causing drier conditions. Certain locations may endure drought-like conditions. River discharge falls drastically when precipitation and runoff decrease. Many rivers experience decreasing water levels, and some may even dry up in sections. As water volume reduces, rivers' flow may become more confined, resulting in narrower channels. The reduced flow also decreases sediment transfer. In certain situations, isolated pools of water may occur in specific portions of the river, providing essential water sources for aquatic life throughout the dry season. It is worth to noting that specific changes in river flow might differ depending on the area, local climate, and the peculiarities of specific river systems.

Changes in river flow patterns, the physicochemical parameters of tropical rivers vary significantly between the wet and dry seasons. During the wet season, more rainfall causes higher river discharge and dilutes chemical load in the water. This encompasses nutrients, organic matter, and contaminants. Heavy rainfall can cause soil erosion and sediment transport, resulting in higher turbidity (suspended load) levels in rivers and streams. The suspended particles may cause the water to appear cloudier. Despite warmer air temperatures in the wet season, increasing cloud cover and rainfall can result in slightly colder water temperatures than in the dry season. Higher flow rates also generate increased turbulence and aeration, which benefits aquatic species by raising dissolved oxygen levels in the water. pH levels can fluctuate due to various causes such as rainfall, organic matter breakdown, and ion release from soils.

Water temperatures may rise during the dry season due to lower cloud cover and flow rates. Solutes, such as fertilizers and contaminants, accumulate in the remaining water. This can result in higher nutrient levels and potentially more pollution. Turbidity levels are decreased as a result of reduced runoff and sediment transport. As suspended particles settle down, the water becomes clearer. However, decreased flow rates and higher water temperatures can reduce dissolved oxygen levels. This can be difficult for aquatic organisms, particularly in areas with intense biological activity. pH levels may also change during the dry season as a result of less diffusion and greater effect from local natural ecological processes.

2.2.1 Intermittent rivers and ephemeral streams

Intermittent and ephemeral streams, according to Miliša *et al*. (2022), are those that stop flowing from time to time and are more prone to create pools or be dry. Levick *et al*. (2008) explain that numerous watersheds in dry, arid, and semi-arid regions are intermittent and ephemeral. Miliša *et al*. (2022) further state that intermittent rivers are intricate varieties of flowing, motionless, and terrestrial habitats that fluctuate in location and time, allowing them to support high biodiversity and ecosystem processes that produce ecosystem services. Therefore, this study will use the words intermittent and ephemeral interchangeably.

Intermittent rivers in tropical regions usually have a significantly different wet and dry season. During the wet season, greater rainfall and runoff contribute to the flow of water in the river, whilst the dry season sees little or no flow (Bourke *et al*., 2021). During the dry season, intermittent rivers might leave behind temporary ponds or isolated water bodies in some areas. These pools can be essential habitats for aquatic life and provide water for animals in the surrounding area. Intermittent rivers are important to the dynamics of tropical ecosystems. They provide habitat for a diverse range of plant and animal species that have evolved to cope with seasonal patterns in water availability. They also help to promote nitrogen cycling and biodiversity in the surrounding area. Human communities located in various tropical regions rely on intermittent rivers for water supply, agriculture, and other subsistence activities. Understanding and managing seasonal water resources is critical to sustainable development.

Historically, Miliša *et al*. (2022) state that monitoring, management, and water resource assessment programs have overlooked intermittent rivers. This arises from the recognition that these water sources are finite, subject to unpredictable renewal, and are contested resources sought after by both local communities and essential for wildlife (Day *et al*., 2019). However, international efforts to create effectively intermittent rivers-specific ecological state assessment tools are motivated by recent awareness of their prevalence and importance (Miliša *et al*., 2022). Climate change is causing these ecosystems to expand in many locations, necessitating effective management measures (Miliša *et al*.,2022).

2.2.2 The Upper Awash flow and water quality features

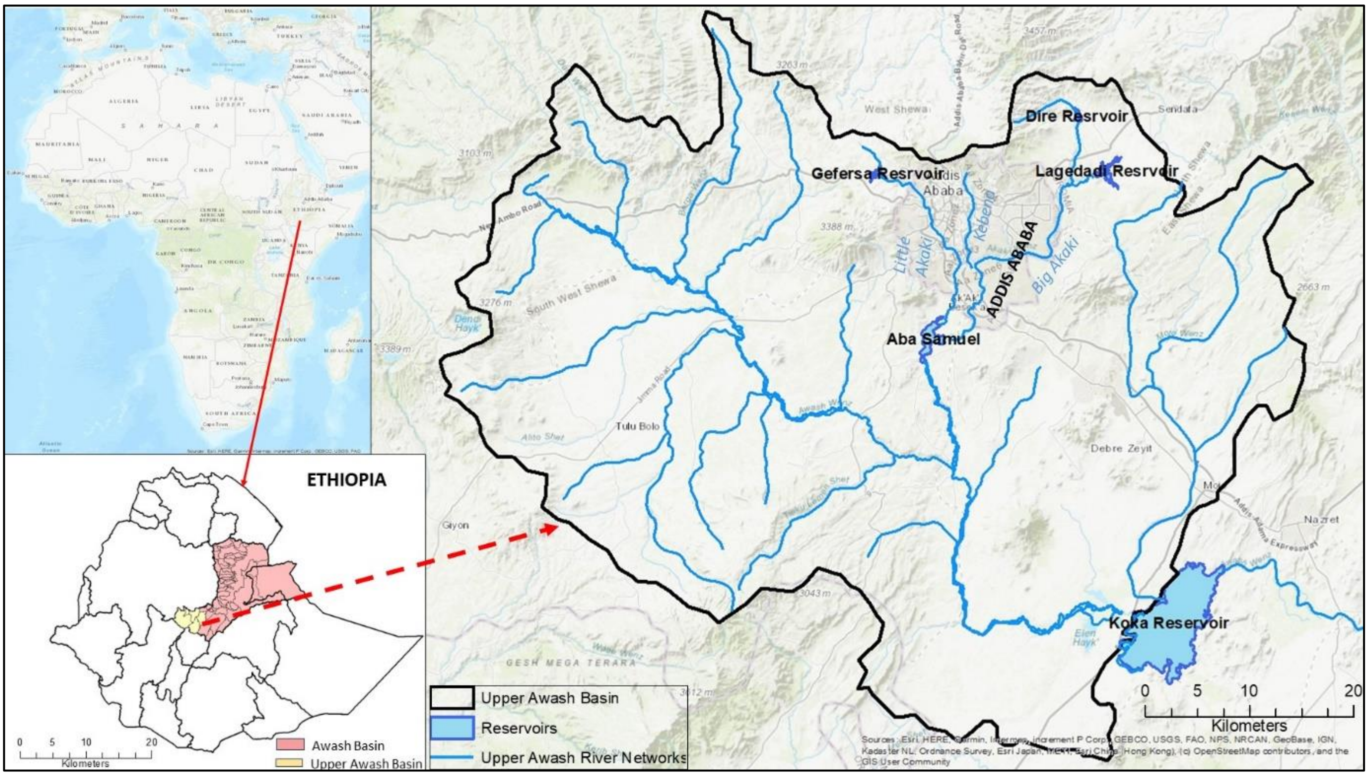


Figure 2.1 Map displaying the Upper Awash River basin and its tributaries

Climate variability, such as the El Niño Southern Oscillation (ENSO), has a significant impact on precipitation in Ethiopia's highlands (Chan *et al*., 2020). El Niño phases are linked to lower-than-average summer rainfall in the area and have been a major contributor to previous droughts (Chan *et al*., 2020). According to Heyi *et al*. (2020), rainfall in the Upper Awash basin averages about 1200 mm in the highlands, around 1050 mm along the escarpment, and less than 900 mm in the rift valley. Similar to rainfall, flows are highest during the wettest months of July, August, and September.

According to Dabessa *et al*. (2021), water is a critical resource that delivers a variety of provisioning, regulating, and cultural ecosystem functions. Ethiopia's river quality is declining, which has negative health consequences, higher water treatment costs, and lower fishery yields (Dabessa *et al*. (2021). Gule *et al*. (2023) state that waste from the city (Addis Ababa) is simply dumped into the rivers. River waste is mainly derived from municipal solid and liquid waste, toilets, and open urination (Gule *et al*., 2023). Anthropogenic changes due to population growth, lack of sanitation infrastructure and uncontrolled urbanisation threaten surface waters, causing severe quality degradation in physical and chemical freshwater habitats (e.g., temperature, pH, dissolved oxygen, conductivity, total phosphorus, and nitrates) and may have subtle to severe biological effects (Yohannes and Ellas, 2017; Dabessa *et al*., 2021). These changes indicate that the ecosystem and its organisms are under pressure (Dabessa *et al*. (2021). These changes in the aquatic environment can signify pollution levels in water bodies and potential risks to human health (Assefa *et al*., 2020). Each aquatic organism has specific requirements for the biological, chemical, and physical conditions of its habitat (Assefa *et al*., 2020). Therefore, disturbances of these conditions lead to a reduction in the number of species, a change in species dominance, or a complete loss of sensitive species (Assefa *et al*., 2020). Therefore, monitoring these activities' impacts on aquatic ecosystems is necessary, using cost-effective monitoring tools (Assefa *et al*., 2020). Although surface water monitoring is emerging in Ethiopia, poor technical and financial constraints cause delays (Assegide *et al*., 2022).

A Review by Yohannes and Elias (2017) shows the trends and current status of rivers and reservoirs in and around Addis Ababa. The Akaki River is recognized as a river that is heavily contaminated in the country, with studies indicating deteriorated water quality at assessed sites. The pollution stems from various sources, including industrial discharges, urban wastewater, and hospital wastes, and agricultural runoff. Industries along the Little Akaki River and its tributaries, such as tanneries, breweries, wineries, and pharmaceutical factories, contribute significantly to the pollution. The water quality is classified as very poor, with major threats including high concentrations of ions, heavy metals, and faecal coliforms, impacting uses like irrigation, swimming, and aquatic ecosystem preservation. Specifically, the Little Akaki River is more polluted than the Big Akaki River, exhibiting greenish-dark colours, pitch-dark sediment, and a pungent odour.

On the other hand, the Kebena River, a tributary of the Awash River, shows lower concentrations of some parameters, such as water pH, sodium, calcium, magnesium, COD, and manganese upstream. However, certain parameters increase downstream of Kebena river, including sulphate, nitrate, and arsenic. The upstream Kebena River experiences fewer anthropogenic pollutants and is relatively protected. Despite having poor quality, it is comparatively better than the Akaki River. The Kebena River faces organic pollution from various sources, primarily residential, commercial, agricultural, and institutional wastes, leading to habitat degradation and loss of habitat in local rivers.

Within the Upper Awash basin lies the Koka hydropower dam, which was constructed in the 1960s and is regarded as the oldest dam in Ethiopia (Getachew *et al*., 2023). Aba Samuel and Koka reservoirs were constructed for hydropower generation. Other reservoirs, such as the Dire and Legedadi, were constructed to supply Addis Ababa with water. This created a concern because reservoirs tend to alter the natural flow and morphodynamical patterns of rivers, which affects biodiversity and ecosystem services (Getachew *et al*., 2023).

2.3 Physicochemical Parameters

Indicator SDG 6.3.2 relies on on-site measurements and sample analyses from both surface and groundwater to assess water quality (UN Water, 2018). This evaluation considers core physical and chemical parameters reflecting natural water quality influenced by climate, geology, and major impacts (UN Water, 2018). Recorded values are used to classify water quality as satisfactory or unsatisfactory (UN Water, 2018). The growing significance of global on-site water quality data is expected in the future, crucial for validating and optimizing the broader spatial coverage achievable with earth observation technology (UN Water, 2018). In situ measurements are also essential for validating water quality models (UN Water, 2018). The monitoring plan for indicator 6.3.2 has two levels: Level 1 uses a water quality index with core physicochemical parameters, and Level 2 involves monitoring additional factors like biological, microbiological, or earth observation approaches (UN Water, 2018). The core physicochemical parameters essential for monitoring rivers include DO, EC, pH, total oxidised nitrogen and orthophosphate (UN Water, 2018).

Natural factors such as geology, hydrology, climate and weather, as well as anthropogenic activities, such as agriculture, wastewater discharges from domestic and industrial facilities, and environmental alterations affect the physicochemical conditions of aquatic ecosystems (Odume, 2017). Therefore, to control pollution and manage water quality, physicochemical variables are typically analysed (Odume, 2017). This assists water resource managers in assessing pollution levels, determining their fate and transport, and determining their persistence in aquatic environments (Odume, 2017). Tadesse *et al*. (2018) state that physicochemical parameters form part of water quality indicators. Many rivers in Ethiopia are monitored for water quality using conventional physicochemical parameters (Dabessa *et al*., 2021). Therefore, the study focuses on the following physicochemical parameters: pH, temperature, dissolved oxygen, electrical conductivity, and clarity.

2.3.1 Temperature

One of the main abiotic elements affecting the structure and functioning of aquatic ecosystems is water temperature, and variations in it may have a significant effect on biotic population (Bonacina *et al*., 2023). For many organisms, temperature signals the timing of spawning, migration, or emergence in the life cycles (Ethiopian EPA, 2003). Climate change can disrupt temperature signals, which can affect normal development (Ethiopian EPA, 2003). According to Bonacina *et al*. (2023), tropical macroinvertebrates typically have a smaller thermal range and are more sensitive to temperature changes compared to temperate macroinvertebrates.

Changes in temperature can have profound effects on the physical and chemical properties of water, impacting the overall health and dynamics of aquatic ecosystems. Temperature has an inverse relationship with dissolved oxygen whereby as temperature increases, the solubility of oxygen in water generally decreases (Ethiopian EPA, 2003). Warmer water holds less dissolved oxygen, making it a crucial factor in the oxygen balance of aquatic ecosystems. Lower oxygen levels can affect the respiration and survival of aquatic organisms.

While temperature itself may not directly influence pH, it can affect other factors which, in turn, impact pH. For example, temperature influences the solubility of gases like carbon dioxide, which can affect the carbonic acid equilibrium and subsequently alter pH levels.

Human disturbances can have a major impact on water temperature in aquatic ecosystems. Many industries release heated water into bodies of water as part of their processes, resulting in thermal pollution that alters local water temperatures. This change can affect aquatic organisms' thermal preferences and, in extreme circumstances, cause stress or death. Land use changes, such as clearing vegetation along riverbanks, can also have an impact on riparian zones' shade and temperature moderating. This may contribute to temperature increases in aquatic ecosystems. Agricultural runoff can inject extra nutrients into aquatic basins. This can promote algae growth, and the decomposition of organic materials by bacteria can cause rising temperatures, particularly in nutrient-rich conditions.

2.3.2 pH

pH is considered one of the essential parameters of water quality. pH indicates the strength of a basic or an acid solution ranging from 0 to 14, with 7 being neutral and less than 7 showing acidity, whereas a pH higher than 7 indicates a base. Pollution can change the pH of water, which can cause harm to aquatic animals (Omer, 2019). For example, water with extremely low pH can be lethal; only a few macroinvertebrates can endure water below a pH 3. A low pH also can make heavy metals such as cadmium, lead and chromium more soluble and bioavailable, thus more toxic (Omer, 2019). pH can affect the metabolic processes of aquatic macroinvertebrates. Extreme pH levels, either acidic or alkaline, can disrupt enzymatic activities and metabolic pathways, leading to reduced growth, development, and reproduction. pH alterations can lead to shifts in the composition and abundance of macroinvertebrate communities. Species that are well-adapted to a particular pH range may dominate, while others may decline or disappear.

Under natural conditions, seasonal variations in precipitation and runoff can affect pH through dilution of water and changes in nutrient inputs. Heavy rainfall can lead to runoff that may carry organic matter, nutrients, and pollutants into water bodies, influencing pH levels. Moreover, biological activities, such as photosynthesis, respiration, and decomposition, are often seasonally influenced. For example, during periods of high biological productivity in the spring and summer, photosynthesis by aquatic plants may lead to increased dissolved oxygen levels and higher pH. In contrast, in the fall and winter, decomposition processes may dominate, influencing pH differently.

2.3.3 Dissolved oxygen

The oxygen in water that supports fish, invertebrates, and other aquatic life is known as dissolved (DO) (Bozorg-Hadded et al., 2021). For example, fish cannot survive in water with less than 5 mg/L of dissolved oxygen. The majority of aquatic life depends on DO to thrive (Bozorg-Haddad et al., 2021). According to USEPA (1998) and WHO (2008), a water body is deemed healthy and suitable for aquatic life if its DO level is within the range of 5-14.6 mg/L. The demand for oxygen by organisms in the water affects the overall dissolved oxygen concentration, with potential implications for the health of the aquatic community.

The low level of DO in water is a sign of contamination and is an important factor in determining water quality, pollution control, and treatment process (Bozorg-Haddad *et al*., 2021). The maintenance of adequate dissolved oxygen (DO) concentrations is critical for the survival and functioning of the aquatic biota because it is required for the respiration of all aerobic organisms (Water Standards Ethiopia, 2003). DO is referred to as the most critical test of water pollution because a high concentration represents good water quality. DO levels are critical for defining the quality of aquatic habitats. Many species have specific oxygen requirements, and variations in DO can influence the distribution and abundance of aquatic organisms. DO levels can also influence the occurrence and intensity of algal blooms. High nutrient levels, coupled with sufficient DO, may stimulate algal growth. However, under low oxygen conditions, certain algal species may dominate, producing harmful effects. The toxicity of certain metals and chemicals is often influenced by the presence of DO. Adequate oxygen levels can mitigate the toxic effects of substances, while low oxygen conditions may exacerbate toxicity.

DO is dependent on temperature, salinity, and pressure. DO decreases as temperature rises (inverse relationship) (Omer, 2019). DO levels can also affect the pH of water. In the process of respiration and photosynthesis, aquatic organisms release or consume oxygen and carbon dioxide, influencing the acid-base equilibrium and, consequently, the pH of the water.

2.3.4 Electrical conductivity

According to Omero *et al*. (2019), conductivity measurements are a good way to determine the amount of salt in a body of water because they show the presence of ions in river water. EC is also described as a substitute for Total Dissolved Solids (TDS) and is used more commonly than TDS because of the availability of good, accurate, inexpensive EC meters. Electrical conductivity and predominant ions serve as proxies for assessing the influence of land use changes or shifts in geology on the physicochemical characteristics of water at the catchment scale (Masese *et al*., 2023).

Aquatic organisms, regulate water and ion levels through osmoregulation. Therefore, increased EC, which indicates increased dissolved salts, can impair osmoregulation in macroinvertebrates that are accustomed to lower salinity environments. High EC levels may reduce macroinvertebrate reproductivity. Changes in conductivity can affect egg development, hatching success, and the survival of larvae. Some species may exhibit preferences for specific for conductivity levels during reproductive activities. EC also influences nutrient cycling and the availability of food resources in aquatic ecosystems. Macroinvertebrates often rely on organic matter and detritus as food sources, and changes in conductivity may affect the breakdown and availability of these resources.

Moreover, EC can influence the habitat selection of macroinvertebrates. Species adapted to low-conductivity environments may avoid areas with high conductivity, impacting their distribution within aquatic ecosystems.

Electrical conductivity changes can influence a variety of physicochemical characteristics in water. In areas with elevated EC due to salinity, reduced oxygen solubility may affect aquatic organisms. While EC itself does not directly affect pH, changes in ion concentrations associated with variations in EC can influence the pH of water. For example, the presence of carbonate ions may act as a buffer, stabilizing pH levels.

2.3.5 Clarity

Clarity refers to the transparency and ability of light to penetrate the water, while turbidity focuses on the presence and concentration of suspended particles and total dissolved solids (TDS) focuses on the overall concentration of dissolved substances in water. All three of the parameters are interconnected and can influence one another. In the context of the study, clarity is used as a proxy for turbidity. The amount of solid matter present in the suspended state determines how turbid the water is (Meride and Ayenew, 2016). Water clarity can vary throughout the year in tropical regions with distinct wet and dry seasons. Runoff can lead to murky water in the wet season, while the water may be clearer in the dry season. Additionally, sediment particles serve as a base for other pollutants, primarily metals and bacteria, and because of this, turbidity measurements are reliable predictors of possible water body pollution (Agboola *et al*., 2019).

These methods can detect stressor effects, but they cannot account for the impact of various stressors, such as biochemical changes in the system over time (Dabessa *et al*., 2021; Mezgebu, 2022). Therefore, water management decisions may be affected by a lack of environmental knowledge (Dabessa *et al*., 2021). Chikodzi *et al*. (2017) state that due to the limitations presented by physicochemical assessments on water quality, biological species have been used to assess the impacts of human-caused activities in aquatic ecosystems, resulting in the development of biological monitoring in ecology.

In the context of the Upper Awash basin, a study by Gule *et al*. (2023) revealed that Legedadie reservoir water samples recorded high turbidity values (NTU), which means that the water was very turbid and dangerous to drink, which may be caused by nearby pollution. Water clarity plays a role in influencing the behavior, ecology, and abundance of macroinvertebrates. Many aquatic macroinvertebrates, especially those visual predators, for example, notonectidae or those relying on vision for activities such as feeding, may prefer clear water habitats. Clear water allows for better visibility and detection of predators and prey. Clear water allows for better light penetration, supporting photosynthetic activities of aquatic plants and algae. This photosynthetic activity contributes to oxygen production, which can be crucial for macroinvertebrates with certain oxygen concentration requirements. Changes in water clarity can indicate pollution or environmental disruptions. Macroinvertebrates may be susceptible to changes in clarity caused by increased sedimentation, fertiliser runoff, and other contaminants. Clear water conditions can help to regulate temperature more effectively. Macroinvertebrates may have more stable and appropriate temperature regimes, which affects their growth, metabolism, and overall physiological functioning. Moreover, heavy metals, such as lead, copper, and zinc, can bind to suspended particles in turbid water. The transport of these metal-laden particles can result in the contamination of downstream areas, impacting aquatic ecosystems and potentially posing risks to human health.

2.4 Heavy Metals And Other Metals

In developing countries that are experiencing rapid population growth, urbanisation, and industrialisation growth, heavy metal pollution of aquatic ecosystems from point and nonpoint anthropogenic sources is a major concern (Oremo *et al*., 2019). According to Tadesse *et al.* (2018), heavy metals are metallic elements with a moderate density and a high potential for toxicity or poisonous effects at low concentrations. The primary causes of heavy metal accumulation in rivers are diverse and include sources like raw wastewater from industries, mining activities, sewage, and leaching of agricultural chemicals (Eliku and Leta, 2018). Mercury (Hg), Cadmium (Cd), Arsenic (As), Chromium (Cr), Thallium (TI), Copper (Cu), Manganese (Mn), Zinc (Zn), and Nickel (Ni) are among the most commonly studied heavy metals in environmental science (Tadesse *et al*., 2018). Heavy metals severely affect people and the environment despite their pollution being less visible. However, heavy metals like iron, copper, nickel and other trace elements are essential for functioning biological systems (Prabu *et al*., 2009).

Yohannes and Elias (2017) highlight the Akaki River in Addis Ababa, Ethiopia, as one of the most contaminated rivers among those affected by heavy metals. Industrial waste has been identified as a significant contributor to heavy metal contamination, posing a substantial threat to water quality within the city (Yohannes and Elias, 2017). Additionally, the contamination of rivers raises concerns about the composition and physiology of macroinvertebrates (Yohannes and Elias, 2017).

Rivers carry heavy metals with them as they flow through impacted areas (Assegide *et al*., 2021). This may result in changes to the heavy metals' spatial distribution patterns in sediments and surface waters (Assegide *et al*., 2021). The existence, movement, and end-of-life of organic molecules and hazardous and persistent heavy metals in water bodies are major global concerns (Assegide *et al*., 2021). Most heavy metals are very hazardous due to their water solubility, extended biological half-lives, and non-biodegradable nature (Assegide *et al*., 2021). River basins with high concentrations of Pb, Cd, Cr, Ni, and Zn tend to accumulate more in places with high industrial and commercial activity (Assegide *et al*., 2021). Therefore, physicochemical water analysis is valuable in assessing water quality.

2.4.1 Other metal - Magnesium (Mg)

Magnesium, an alkaline earth metal, is a naturally occurring element in water and the eighth most abundant element in the earth's crust (Meride and Ayenew, 2016). Magnesium affects both aquatic macroinvertebrates and several physicochemical parameters in water. Magnesium affects macroinvertebrates' habitat choices. Some species may have preferences for water with specific magnesium concentrations, which affects their dispersion in aquatic settings. Macroinvertebrates use osmoregulation to maintain normal internal salt concentrations. Magnesium is one of the ions involved in this process, and its availability has an impact on macroinvertebrate osmotic equilibrium. Magnesium also influences macroinvertebrates' reproductive success. Adequate magnesium levels are required for the normal operation of reproductive processes such as egg formation and hatching success.

Furthermore, magnesium can act as a buffer in water, helping to maintain pH levels. Its presence can help to keep the pH in an acceptable range for aquatic life, preventing fast changes.

Magnesium concentration changes can have an impact on overall ionic strength (a measure of the concentration of ions in a solution), which affects chemical reactions and interactions in aquatic environments. Magnesium can participate in precipitation reactions alongside other ions in water. These reactions can produce solid precipitates, which affect water clarity and sedimentation processes. Magnesium can affect the thermal characteristics of water. Changes in magnesium concentrations may impact the water's ability to hold or release heat, helping to regulate temperature in aquatic habitats.

2.4.2 Other metal Potassium (K)

While potassium is generally essential for the health and functioning of aquatic ecosystems, excessively high concentrations can be detrimental. Potassium is an alkali metal that is silver-white and very reactive to water (Meride and Ayenew, 2016). Elevated potassium levels may result from anthropogenic activities such as fertilizer runoff or discharges from certain industrial processes. Potassium is involved in osmoregulation, the process by which organisms regulate water balance and ion concentrations within their bodies. Aquatic macroinvertebrates need to maintain proper osmotic balance, and potassium plays a role in this physiological process. Adequate potassium levels are necessary for the formation of tissues and the overall development of larvae and juveniles.

Potassium can act as a buffer in water, helping to regulate pH levels. Its presence can contribute to maintaining the pH within a suitable range for aquatic life, preventing extreme fluctuations that may negatively impact organisms. The availability of potassium can influence the solubility of oxygen in water. However, changes in potassium concentrations may impact the oxygen saturation levels, affecting the respiratory processes of aquatic organisms.

2.4.3 Cadmium (Cd)

Through mechanisms like bioaccumulation, cadmium may contribute to acidity variations, influencing the availability of other metals and the overall buffering capacity of the water. Cadmium pollution can reduce dissolved oxygen levels in water by disrupting oxygen transport processes, which can be detrimental to the survival of aquatic organisms, including macroinvertebrates. Cadmium has the potential to harm organisms even at low quantities (Temesgen and Shewamolto, 2022). Cd build up in the tissues of aquatic species, especially macroinvertebrates. Higher up the food chain species may have greater cadmium contents because of this. Aquatic macroinvertebrates are extremely sensitive to cadmium, and exposure to high doses can be fatal. The species, life stage, and length of exposure all affect how harmful the effects are.

Cadmium may contribute to changes in acidity, potentially impacting the availability of other metals and affecting the overall buffering capacity of the water. Cadmium contamination can contribute to reduced dissolved oxygen levels in water. This is often associated with the disruption of oxygen transfer mechanisms and can negatively impact the survival of aquatic organisms, including macroinvertebrates. The standard water quality maximum requirement for Cd in surface water is 5 µg/ℓ (Water Standards Ethiopia, 2003).

2.4.4 Chromium (Cr)

Chromium is found naturally in the earth and is a common environmental element (Jeong *et al*., 2023). Metal processing, chromate manufacture, tannery operations, stainless steel welding, chromate pigment production, texture dyes, and catalysts are among the industries that significantly release chromium into the environment (Jeong *et al*., 2023). Although the concentration of chromium in the environment has been declining for decades, it remains high in certain regions, like the area surrounding the Ethiopia Tannery Share Company (Jeong *et al*., 2023).

Chromium exists in a variety of oxidative forms, ranging from divalent to hexavalent, but the trivalent (Cr III) and hexavalent (Cr VI) forms are the most frequent and are often stable biologically and ecologically (Jeong *et al*., 2023). Trivalent chromium is a crucial element in biological systems because it contributes to glucose, lipid, and protein metabolism (Jeong *et al*., 2023). Trivalent chromium's poor membrane permeability contributes to its low toxicity (Jeong *et al*., 2023). In contrast, hexavalent chromium can pass through biological membranes via a sulfate/phosphate anion transporter before being reduced to lower oxidation forms like pentavalent, tetravalent, and trivalent chromium (Jeong *et al*., 2023). Reactive oxygen species (ROS), including hydroxyl radicals are molecules produced by Haber-Weiss or Fenton-like reactions, can have a variety of harmful effects throughout the reduction process (Jeong *et al*., 2023). Thus, chromium contamination can be lethal to aquatic creatures (Jeong *et al*., 2023). Chromium can influence pH levels in water. The impact depends on the oxidation state of chromium, with Cr(III) typically having less effect on pH than Cr(VI). Changes in pH can affect the solubility of other metals and the overall water chemistry. Chromium contamination can contribute to reduced dissolved oxygen levels in water. This is often associated with the disruption of oxygen transfer mechanisms and can negatively impact the survival of aquatic organisms, including macroinvertebrates. The standard water quality maximum requirement for Cr in surface water is 50 µg/ℓ (Water Standards Ethiopia, 2003)

2.4.5 Copper (Cu)

Copper is a relatively scarce metal in nature, but it is frequently found as a contaminant because of anthropogenic sources such as industrial discharges, agriculture, and stormwater runoff (Jeong *et al*., 2023). Regulatory measures, effective monitoring, and pollution control practices are essential to mitigate the impact of copper on water quality and the health of aquatic ecosystems. Controlling the sources of copper contamination and implementing measures to reduce its release into water bodies is crucial for preserving the integrity of aquatic environments. Copper is considered nontoxic if found in low concentrations (Omer, 2019). However, copper is toxic to aquatic macroinvertebrates at higher concentrations, and exposure to elevated concentrations can lead to adverse effects. This includes mortality, reduced reproductive success, and impaired growth and development. It may lead to abnormalities in egg development, larval growth, and overall reproductive output.

Copper contamination can contribute to reduced dissolved oxygen levels in water. This is often associated with the disruption of oxygen transfer mechanisms and can negatively impact the survival of aquatic organisms, including macroinvertebrates. Copper can influence microbial communities in sediments and water. Changes in microbial activity may have cascading effects on nutrient cycling and overall ecosystem functioning.

2.4.6 Mercury (Hg)

Mercury (Hg) is a naturally occurring element found in the earth's crust, but human activities like coal and gold mining, as well as coal and fuel combustion, have led to increased concentrations in the environment (van Rooyen *et al*., 2023). Burning coal releases small amounts of Hg into the air, allowing it to travel and settle in surface water, soils, and sediments (van Rooyen *et al*., 2023). Numerous studies indicate elevated Hg levels in freshwater ecosystems and organisms, even in areas far from direct sources, highlighting the significant impact of long-range atmospheric transport from human-related origins (van Rooyen *et al*., 2023). The consequences of Hg on freshwater systems, fish, and human health have been extensively researched globally, revealing harmful effects on both ecosystems and human well-being (van Rooyen *et al*., 2023).

The chemical forms of Hg in the environment play a crucial role in its behaviour, fate, transport, bioaccumulation, and toxicity (van Rooyen *et al*., 2023). Additionally, the form of Hg in aquatic systems is strongly influenced by factors like pH, redox conditions, inorganic hydroxide, sulphide ligands, and chlorides (van Rooyen *et al*., 2023). The water quality standard for Hg in surface water is 1 µg/ℓ (Water Standards for Ethiopia, 2003). In aquatic environments, Hg attaches to suspended particles, playing a vital role in its movement, and settling onto sediments (van Rooyen *et al*., 2023).

Mercury tends to accumulate in living organisms through a process known as bioaccumulation. In aquatic environments, macroinvertebrates may take up mercury from water, sediment, or their food sources. As these organisms are consumed by predators higher up the food chain, the mercury concentration can increase.

2.4.7 Iron (Fe)

One of the most prevalent and necessary elements for all plants and animals is iron (Temesgen and Shewamolto, 2022). However, at elevated concentrations, iron can have both direct and indirect effects on aquatic macroinvertebrates and physicochemical parameters in water quality. High concentrations of iron can interfere with the respiratory processes of aquatic organisms, particularly macroinvertebrates. This may affect gill function, leading to reduced oxygen uptake and potential stress on organisms. While iron is a necessary nutrient, excessive concentrations can be toxic to aquatic macroinvertebrates. Prolonged exposure to high levels of iron may result in adverse effects such as impaired growth, decreased reproductive success, and even mortality.

Iron can influence pH levels in water. The oxidation and reduction of iron can contribute to changes in acidity or alkalinity, impacting the overall pH of the aquatic environment. Iron can contribute to water turbidity by forming suspended particles. High concentrations may lead to increased turbidity, potentially reducing light penetration and affecting the growth of aquatic plants. The standard water quality maximum requirement for iron in surface water is 1000 µg/ℓ (Water Standards Ethiopia, 2003)

2.4.8 Manganese (Mn)

It is noteworthy that the impact of manganese on water quality and macroinvertebrates can differ based on various factors, including the type of manganese (particulate or soluble), water chemistry, and the species sensitivity of the individual macroinvertebrate present. For aquatic macroinvertebrates, high manganese concentrations can be toxic. Negative consequences from prolonged exposure to high manganese levels can include stunted growth, decreased success in reproduction, and even death. Exposure to manganese can interfere with the respiratory processes of aquatic organisms, including macroinvertebrates. This may affect gill function, leading to reduced oxygen uptake and potential stress on organisms.

Manganese oxidation and reduction can cause changes in acidity or alkalinity, which affects the overall pH of the aquatic environment. Changes in manganese concentrations may affect oxygen saturation levels, thereby influencing aquatic organisms' respiratory processes. Manganese can form insoluble precipitates, which affect water clarity and sedimentation processes. These precipitates may have an impact on benthic habitats and the substrate in which macroinvertebrates live.

Manganese can interact with other metals in water, leading to complexation or precipitation reactions. These interactions can influence the mobility and bioavailability of manganese and other metals in the aquatic environment. The standard water quality maximum requirement for Mn in surface water is 300 µg/ℓ (Water Standards Ethiopia, 2003)

2.4.9 Lead (Pb)

To limit the impact of lead on water quality and aquatic ecosystem health, regulatory measures, effective monitoring, and pollution control strategies must be implemented. Addressing the causes of lead pollution and putting in place measures to prevent its flow into bodies of water are critical for maintaining aquatic ecosystem integrity.

Lead is very toxic to aquatic macroinvertebrates, and excessive amounts can cause mortality, decreased reproduction, and delayed growth and development. Lead may build up in the tissues of aquatic species, especially macroinvertebrates, by processes such as bioaccumulation. This can result in increased levels of lead in species further up the food chain. It might cause anomalies in the growth of the larvae and eggs, as well as a decrease in total reproductive production. as well as changes to behaviour like eating, swimming, and general activity levels.

Lead has an effect on the levels of physicochemical factors in water. Although it doesn't directly influence pH, its presence may interact with other ions and substances to change how alkaline or acidic a solution is. Lower dissolved oxygen concentrations in water can be caused by lead contamination. This is frequently linked to the disturbance of processes that transfer oxygen, which has an adverse effect on aquatic organisms' ability to survive, including macroinvertebrates. Over time, lead can accumulate in soil, and contaminated sediments can store lead exposure. Through this mechanism, macroinvertebrates that live in sediment may be exposed to lead for an extended period of time. Lead can react through complexation or precipitation with other metals in water. Lead and other metals' mobility and bioavailability in the aquatic environment may be impacted by these interactions.

2.4.10 Zinc (Zn)

Ouma *et al*. (2022) state that most aquatic organisms can tolerate Zn concentrations lower than 100 µgL−1. Nevertheless, Ouma *et al*. (2022) also observed that when Zn levels become elevated, it adversely impacts the transformation and emergence rates of mature stages of freshwater insect populations. In the study, Ouma *et al*. (2022) found that pollution-sensitive mayflies (Ephemeroptera) decreased in population due to the presence of Zn and toxicity in streams combined with two other heavy metals.

The presence of zinc in water with elevated concentrations, can have numerous effects on physicochemical parameters thus influencing river quality (Ouma *et al*., 2022). While zinc does not directly alter pH, its presence can interact with other ions and compounds in water, potentially contributing to shifts in acidity or alkalinity. With high concentrations of zinc, it may have adverse effects on the solubility of oxygen in water. This can lead to decreased dissolved oxygen levels, impacting the ability of aquatic organisms to respire and survive. Zinc is a conductive metal, and its presence in water can contribute to changes in electrical conductivity (Ethiopian EPA, 2003).

In terms of temperature, zinc has no direct effect. Its presence may, however, be associated with industrial discharges or other anthropogenic activities that contribute to thermal pollution. The presence of zinc particles or precipitates can also contribute to the turbidity of water. An increase in turbidity may hinder light penetration, which may have a negative impact on aquatic plants and the overall ecosystem. It is also possible for zinc to interact with other metals in water, leading to complexation or precipitation. As a result, zinc and other metals can have an impact on their mobility and bioavailability in aquatic environments.

2.5 Biomonitoring

SDG 6.3.2 monitoring plan has two levels (UN Water, 2018). However, according to the UN Water (2018), the main indicator in Level 1 doesn't capture all the aspects affecting water quality, so the more detailed Level 2 steps ensure a balance between global and national relevance. Level 1 therefore serves as the foundation for more specific monitoring programs (UN Water, 2018). In Level 2, monitoring can be enhanced by measuring more parameters or employing additional methods for assessing water quality. These additional methods include biological monitoring. A few African countries have modified and improved biological monitoring over the years such as SASS5 with the results are often combined with physicochemical measurements to attain overall assessment of water quality.

The study focuses on Biomonitoring using macroinvertebrates as a monitoring method for assessing water quality. The principle underlying Biomonitoring is that macroinvertebrates are pointers of the environment's health they live in (Chikodzi *et al*., 2017). The advantage of Biomonitoring is that physical, chemical, and biological effects caused by harmful doings on a river system can be noticed. Miliša *et al*. (2022) and Dabessa *et al*. (2021) state that because of their abundance, varying tolerances to water contamination and diversity, river macroinvertebrates are among the most extensively used markers for determining the condition of freshwater ecosystems. Furthermore, macroinvertebrates are widespread, prolific, simple to gather, respond quickly to pollution, have extended life spans, and indicate local conditions due to sedentary behaviour (Dabessa *et al*., 2021). Biological indicators such as macroinvertebrates and diatoms have recently become routine monitoring instruments, particularly in industrialised countries (Dabessa *et al*., 2021).

Feio *et al*. (2021) state that assessing river or stream conditions is, in theory, a first step toward rehabilitating rivers that do not fulfil the quality criteria. Dabessa *et al*. (2022) further state that water quality monitoring is necessary to identify the current state of water quality and improve environmental conditions and public health problems. However, limited research on macroinvertebrate indicators has been done to assess water pollution's specific impact on macroinvertebrates. Biomonitoring is comprehensive and includes a variety of sources of aquatic deterioration (Obubu *et al*., 2021). Obubu *et al*. (2021) list the importance of Biomonitoring as follows:

- demonstrates overall ecological integrity;
- provides a comprehensive picture of the state of the environment by combining stressors over time;
- is simple to understand by the general public once they are trained on their indicative value;
- can be carried out in all rivers due to the ubiquitous and diverse nature of bioindicators; and
- is considered less expensive than physical and chemical approaches because it does not require the use of chemicals,
- incorporates the assessment of various substances in the ecosystem, such as heavy metal deformities in chironomid mouthparts, and
- results are comparable if the same techniques are applied.

Complementary to the conventional monitoring parameters, Biomonitoring using macroinvertebrates is introduced in this section. These bioindicators respond to both hydro morphological variation and physicochemical changes as a result of human and natural stressors in streams, rivers and their surrounding areas (Jerves *et al*., 2020). Furthermore, macroinvertebrates continuously respond to environmental variations over a long period, as opposed to physicochemical samples that reflect the water quality at a particular point in time (Jerves *et al*., 2020). While many of the macroinvertebrate families utilized for constructing biotic indices in temperate regions exist in tropical regions, they may not inhabit identical ecological niches and may exhibit distinct sensitivities to water pollution (Ochieng *et al*., 2020)

2.5.1 Challenges for Biomonitoring in Africa

Biomonitoring presents several challenges in Africa. According to Obubu *et al*. (2021), the first challenge is correctly identifying bioindicators at the appropriate taxonomic level due to inadequate national, regional, and continental-based taxonomic identification keys, guides, or experts. African countries rely heavily on foreign, mainly temperate-based taxonomic guides and keys, often unreliable in tropical regions (Obubu *et al*.,2021). It is debatable whether macro-invertebrate indicators from temperate and other biogeographical zones are appropriate and reliable for evaluating pollution in tropical rivers (Ochieng *et al*., 2020). For example, the concept is often considered as non-rational. Elias (2021) argues that due to environmental variations between regions that affect the capability and reliability of the adopted method, tropical African regions have been hindered by geographical incompatibility when assessing water pollution in their rivers using nontropical Biomonitoring methods.

Secondly, macroinvertebrates are sometimes identified abroad due to the lack of local expertise, thus making the process very costly (Obubu *et al*., 2021). Also, early studies on macroinvertebrates in Africa were done by foreign researchers who focused primarily on taxa of medical significance, which led to the lack of broader identification guides covering macroinvertebrates (Obubu *et al*., 2021). During that time, molluscs, biting midges, and simulids (black flies) were the priority taxa due to the spread of Schistosomiasis (bilharzia) and Onchocerciasis (Obubu *et al*., 2021). Most non-disease-transmitting macroinvertebrates did not receive attention (Obubu *et al*., 2021).

2.5.2 Existing water quality Biomonitoring tools in Ethiopia

In Ethiopia, there has been limited research on benthic macroinvertebrates in streams and rivers. The earliest studies played a crucial role in exploring macroinvertebrates to assess the pollution levels in Ethiopian streams (Mehari, 2014). In 1988 a comprehensive examination of benthic macroinvertebrate communities in mountain streams across seven river systems, focusing on the biogeography of Afrotropical Mountain stream fauna, was conducted (Mehari, 2014). Sitotaw (2006) contributed to the field by introducing a Benthic Index of Biotic Integrity in a study involving nine Ethiopian rivers (Mehari, 2014). His findings revealed that widespread agricultural operations, as well as industrial and urban land-use, posed substantial challenges to Ethiopian river ecosystems. Recently, with the purpose of providing an easy-to-use Biomonitoring index for assessing the ecological state of streams in Ethiopia's highlands that is, ecoregions with forests and grasslands above a height of 1800 meters ETHbios was developed (Aschalew and Moog, 2015). It was formed on the Biological Monitoring Working Party (BMWP) strategy, removing taxa that do not occur in Ethiopia and including certain Ethiopian benthic fauna (Mezgebu, 2022).

A sensitivity score was assigned to 59 benthic macroinvertebrate taxa collected from 104 sites in the upper Awash, Rift Valley, Wabi-Shebele, and Genale basins, covering a total area of around 98,000 square kilometres (Mezgebu, 2022). Benthic invertebrates are sensitive to organic pollution, siltation, and some hydro morphological deterioration, according to ETHbios (Mezgebu, 2022). Scores were assigned based on calculating a guide score using experts' judgement. The taxa with low scores show high tolerance for stressors, and the taxa with high scores show low tolerance for stressors (Mezgebu, 2022).

The 'guide score' is generated using the Ethiopian multimetric index and the dispersal and number of benthic macroinvertebrate taxa across the five river quality classes (Mezgebu, 2022). To arrive at the final value, an agreement was reached among experts, quantified occurrence and abundance occurrences of each taxon, a reference score of each taxon based on the South African Scoring System (SASS) and Hindu Kush-Himalayan biotic score (HKHbios), and knowledge of the autecology of the benthic invertebrate group were taken into consideration (Mezgebu, 2022). Then ETHbios was determined as the total of each taxon's sensitivity score in a sample (Mezgebu, 2022). ETHbios was divided by the total number of taxa examined in the calculation to get the Average Score Per Taxon (ASPT) (Mezgebu, 2022). However, Obubu *et al*. (2021) state that ETHbios has not been accepted nationally, notably by the Ministry of Water for Biomonitoring, due to its shortcomings. A significant obstacle to its progress is a poor taxonomic list, the lack of Government support for research, and insufficient infrastructure to reach water bodies and take and process samples (Mezgebu, 2022). Instead, universities and other institutions use it for academic and research purposes (Obubu *et al*., 2021).

2.6 South African Scoring System Version 5 (SASS5)

A widely used biological monitoring system, the South African Scoring System (SASS), was created by Chutter (1998) and updated by Dickens and Graham (2002) to version 5 (SASS5). SASS5 is the outcome of the Biological Monitoring Working Party's adaptation to South African river systems. The tool has been used for various impact assessments, and locally, it is used by multiples institutions such as Umgeni Water, Umlaas Irrigation Boards, Mpumalanga Parks Board, the Department of Water Affairs and Forestry, CSIR, Cape Metro Council and countless companies (Dickens and Graham, 2002). Before the development of SASS5, there was SASS4 that had deficiencies and had to undergo extensive testing (Dickens and Graham, 2022).

The South African Scoring System is suitable for assessing river water quality and health. According to Dicken and Graham (2002), the tool can be used to evaluate the ecological state of aquatic ecosystems, assess the environmental condition in spatial and temporal trends, consider developing problems and lastly but not limited to predict changes due to developments.

The SASS5 scoring sheet, used for identifying macroinvertebrates after sampling, encompasses over ninety families, each assigned scores ranging from 1 to 15 based on their sensitivity to changes in water quality. Results are expressed as both an index score and an average score per recorded taxon (ASPT) value (Watson and Dallas, 2013).

Dickens and Graham (2002) state that it is unsuitable for wetlands, impoundments, estuaries, and other lentic ecosystems due to its design for moderate flow hydrology. Additionally, it has not been thoroughly tested in ephemeral rivers, and Dickens and Graham (2002) recommend cautious use in such cases. The technique generally performs best in various biotopes, such as riffles or rapids, but can still yield valuable results in poor habitats (Dickens and Graham, 2002). Interpretation of the data should consider factors like habitat quality, availability, diversity, and the ecoregion of origin (Dickens and Graham, 2002). Seasonal and yearly variations should also be considered during data evaluation (Dickens and Graham, 2002).

2.6.1 Reviewed case studies on SASS5

Several Southern African countries have adopted SASSA to monitor the quality of water and the ecological health of lotic systems (Bere and Nyamupingdza, 2014).

A case study conducted by Watson and Dallas (2013) focused on comparing SASS5 to the Macroinvertebrate Response Assessment (MIRAI) method to determine how the methods could be applied to temporary rivers and to evaluate the ability of the tools to determine the current ecological state category of macroinvertebrates. The study area was the Seekoei River, an ephemeral tributary of the Orange River in South Africa. Sampling was conducted every six weeks, starting from the month of March 2006 to the year 2007, October. The results showed that SASS5 implementation was hampered by lacking habitats, generalist taxa, and irregular flow/no-flow periods. Watson and Dallas (2013) suggested that the hydrological phase must be considered when interpreting data.

A research case study conducted by Bere and Nyamupingidza (2014) tested the applicability of SASS5 on streams in Chinhoyi town in Zimbabwe. The sites were selected to evaluate the impact of poor infrastructure maintenance and development (sewage discharge) on water quality and the distribution of macroinvertebrates. The study evaluated the connection between SASS5 indices (SASS5 score and SASS5 ASPT) and physicochemical water quality variables using statistical analysis, and the results revealed a positive connection; the results also indicated that the system is appropriate to the chosen area of interest due to the wide distribution of macroinvertebrates taxa that have similar environmental tolerance to those recorded for South Africa. However, the significant obstacle was the lack of proper taxonomic identification guides and the time-consuming identification necessitating specialisation.

A study conducted by Munyika *et al*. (2014) aimed to monitor the water quality of the Orange River in Namibia using SASS5 and physicochemical parameters and to evaluate the effects of land use changes on water quality using Landsat images. A total of six sites out of eight were sampled using the SASS5 techniques between February and March. However, sites lacked suitable biotopes for the application of SASS5. With the six sampled sites, SASS5 results reflected a clear relationship between water quality and river flow and how the diversity of macroinvertebrates responds to the changes in land use and water quality. The distribution of macroinvertebrates may be affected by the continuous dam releases, which alters the flow of water and the parameter concentrations downstream and, subsequently, the abundance of macroinvertebrates. Overall, the study concluded that SASS5 results and water quality parameters (pH, dissolved oxygen, and electrical conductivity) showed the Orange River to be in fairly good health.

A recent study by Deventer *et al*. (2021) applied SASS5 to assess the impact of various land use on the water quality and biological activity reflected by macroinvertebrates composition. The chosen study area was the upper uMgeni Catchment, KwaZulu-Natal, South Africa. The sampling sites were strategically selected to understand the status of water quality and ecological health in an undisturbed site, moving down to a site with settlements and, lastly, a site just before the river flows into the Midmar Dam. A total of nine sites were selected for physical and chemical monitoring and six were selected for Biomonitoring. Sampling took place at bimonthly intervals across 10 months to account for seasonal changes. Through statistical analysis, the shifts in macroinvertebrates positively correlated with seasonal shifts in water quality. SASS5 results also reflected the decline of water quality from upstream to downstream.

2.7 Streams Assessment Scoring System (MiniSASS)

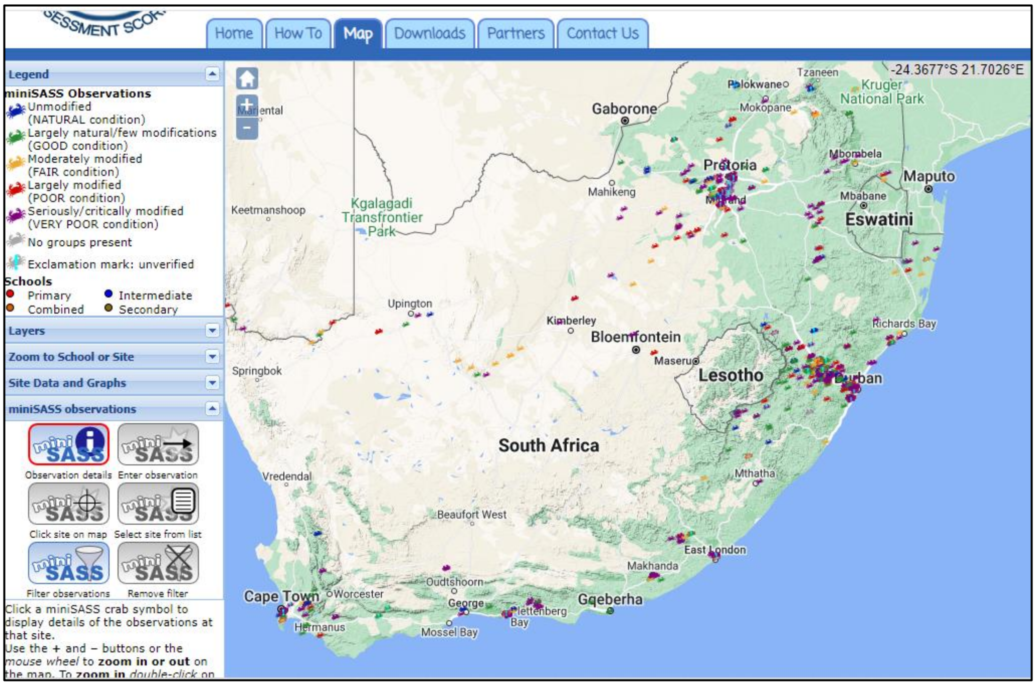
In dealing with complex challenges like water management and ecological infrastructure, considering the context and involving participants are crucial (Taylor *et al*., 2021). Addressing these issues often requires combining knowledge from natural and social sciences, along with economics, as there is usually no single solution (Taylor *et al*., 2021). Citizen science offers a cost-effective way to collect data, especially with high spatial and temporal resolution, as it's often more economical to support volunteers than to acquire data professionally (Babiso *et al*., 2023). A citizen scientist is a public participant who contributes time and effort to a scientific study, often in collaboration with professional scientists (Babiso *et al*., 2023).

According to Taylor *et al*. (2021) the democratization of science through citizen science, supported by accessible tools, is proving to be effective. Building collective capacity in the face of resource-based risks and uncertainty involves social learning, contributing to meaningful social change (Taylor *et al*., 2021). Therefore, in the study context, miniSASS is a crucial tool with potential global relevance. It mobilizes people, promotes the Sustainable Development Goals (SDGs), and provides accessible Biomonitoring data at virtually no cost, contributing to SDG indicator 6.3.2 (Taylor *et al*., 2021).

Taylor *et al*. (2021) describes miniSASS as an easy-to-use tool for monitoring the health of a river that anyone can use, whereby samples of macroinvertebrates are taken from natural rivers or streams. After that, based on the presence of each group, a river's health index can be determined (Taylor *et al*., 2021). Using the score obtained, Taylor *et al*. (2002) categorised the river's ecological condition into five groups, ranging from natural to very poor. A Google Earth layer (www.minisass.org) is generated from the results and added to the miniSASS website. This database is effectively a 'Living data' system, with new data regularly added (Figure 2). Originally, miniSASS was developed from the South African Scoring System (SASS), and since its introduction, the system has proven to be very effective. Many countries in southern Africa have successfully used it, and other countries like Brazil, India, Vietnam, Canada, and Germany have all successfully implemented this technique (Taylor *et a*., 2021). With the help of citizen science and crowdsourcing, miniSASS can generate large amounts of data. Even though these data may not be valid due to lack of formal training from the people who collect the data, they have their unique validity due to the sheer volume of data (Taylor *et al*., 2021). Furthermore, miniSASS is an essential enabling tool that can mobilise people to help meet the Sustainable Development Goals (SDGs) and offer available Biomonitoring data with minimal expenses (Taylor *et al*., 2021).
However, there are limitations to the tool that needs to be considered when applying miniSASS. These challenges are listed below:

- Participants need to identify a local stream or river and prepare to enter the water to locate organisms. (Taylor *et al*., 2021)
- In some cases, catching the organisms for identification is difficult (Taylor *et al*., 2021).

- It is necessary to identify the organisms once they have been collected. Often, this is not an easy task. Users can use a simple dichotomous key on the miniSASS website www.minisass.org (Taylor *et al*., 2021).
- Accuracy of the tool is a concern because of its use by non-professionals; however, training is provided through instructional videos on the website (Taylor *et al*., 2021).



Click a miniSASS crab symbol to display details of the observations at that site.
Use the + and – buttons or the *mouse wheel* to **zoom in or out** on
he map. To **zoom in** *double-click* on

Figure 2.2 South African map reflecting areas where miniSASS has been successfully applied (*www.miniSASS.org*, 2022)

2.7.1 Reviewed case studies on miniSASS Applications

As part of Ko et al. (2020)'s study, three rapid aquatic macroinvertebrate Biomonitoring tools, the Asian Foundation method, miniSASS and The Australian Waterwatch, were evaluated to assess their applicability and suitability, considering sufficient taxa groups for monitoring the health of rivers in Myanmar. The selected sampling sites were situated on the Chaungmagyi and Myitnge Rivers in Myanmar, Asia, chosen for their minimal impact from human activities and habitat conditions likely to support macroinvertebrates during low water levels. Through statistical analysis and taxonomic identification in the laboratory, The Asia Foundation index method emerged as the most suitable for Myanmar taxa, with slight differences noted in comparison to miniSASS and the Australian WaterWatch. The study's conclusion highlighted that miniSASS, among the two other tools, was less comprehensive, making it suitable for young scholars and the general public. The Australian WaterWatch method, while deemed appropriate for community use in monitoring, was found to be more complex due to the numerous taxa involved. Meanwhile, The Asia Foundation tool was considered more extensive and suitable for researchers and government organizations.

2.8 Analysis and Research Gap

In numerous developing nations, the evaluation of stream ecological health and water quality tends to focus on physical and chemical properties, neglecting biological monitoring (Bere and Nyamupingdza, 2013). Much of the literature emphasizes issues in flowing rivers and often overlooks intermittent/non-perennial streams. According to Messager et al. (2021), freshwater science has primarily concentrated on the protection of perennial water bodies, and the cessation of riverine flow has only recently become a research focus. Consequently, there is a scarcity of science-based management tools and protocols, such as Biomonitoring tools, for these unique ecosystems. Mezgebu (2022) notes that Biomonitoring in Ethiopia is in its early stages, lacking a system for decision-making and state-supported research.

Biomonitoring tools such as SASS5 and miniSASS have been tested and used in Southern Africa and are developed for rivers with perennial water flows; however, no known publications have attempted to test the application of the methods in intermittent rivers in Ethiopia. Ephemeral and intermittent streams in arid environments are characterised by distinct hydrologic and ecological regimes different from perennial streams. Because of the water scarcity in these intermittent stream channels, their ecosystems are less resilient to anthropogenic pressure and climate change and do not recover quickly. Moreover, literature on the application of Biomonitoring tools in African tropical rivers is inadequate.

The use of foreign Biomonitoring tools raises concerns regarding the transfer of indices developed in one geographic region into another as there may be faunal differences among regions and potential environmental differences that may modify species' response to the environment (Bere and Nyamupingidza, 2014). Endemic macroinvertebrate taxa may also occur in different regions, necessitating development of region-specific indices (Bere and Nyamupingidza, 2014).

Assefa *et al*. (2020) state that previous literature reported that one of the difficulties faced by East African researchers is taxonomic difficulties with monitoring freshwater bodies using macroinvertebrates. Therefore, such information raises more concerns about classifying macroinvertebrates in intermittent and ephemeral streams. Furthermore, due to the Biomonitoring tools from South Africa, the use of keys for macroinvertebrate identification may limit the credibility of assessments yet to be conducted in Ethiopia. However, SASS5 has over 90 taxa that macroinvertebrates can be identified. Moreover, when considering the potential use of a Biomonitoring tool outside the region where it was developed, it is crucial to take into account if the river system is prevalent in the area, as well as the range of aquatic biotopes in the rivers there. And in regions with biotopes similar to the existing Biomonitoring methods, sampling protocols can be adapted without much change. (Dallas, 2021).

Reviewing miniSASS, it is evident that the application of the tool has been extensively used in South Africa and other countries as shown on the map in Figure 3, however, there are limited published papers that highlight the significance of miniSASS. The case study, where miniSASS was tested abroad, revealed the efficiency of the tool and its suitability to be used by the community. Although the accuracy may be in question, the data provided on the website is monitored by experts for quality assurance.

Reviewing the Biomonitoring tool, SASS5, It is evident that the system has shortcomings which deter the attempt to apply the technique in intermittent rivers. Some of the drawbacks are listed as follows by Dickens and Graham (2002).

- SASS5 should only be used under the conditions of its design. The model is inaccurate in wetlands, lentic areas, estuaries, or heavily flooded rivers. Samplers need to exercise caution in ephemeral rivers as well.
- There is a risk of misinterpreting results due to habitat variability and high flow rates.

In spite of the limitations linked with SASS5, the case studies outlined in this review clarify that SASS5 can be applied beyond its country of origin. Moreover, these case studies highlight numerous advantages that contribute to the effective monitoring of rivers. The outcomes derived from SASS5 have the capacity to depict the influence of seasonal variations on water quality, showcase variations in water quality from upstream to downstream, and offer prompt results. Additionally, the literature indicates a favourable correlation between certain physicochemical parameters, heavy metals, and the indices of SASS5 and miniSASS, enhancing the credibility of these Biomonitoring tools.

2.9 Conclusion

In conclusion, intermittent and ephemeral rivers require Biomonitoring attention because they serve an essential role in the ecosystem. Due to climate change, these ecosystems are anticipated to be expanding in many locations, which shows the need of developing effective intermittent and ephemeral river management measures. The use of miniSASS for river monitoring is simple, cost-effective, and requires basic training, offering a community engagement opportunity in water quality programs and awareness initiatives. Employing these tools in stream monitoring aids in identifying sources of pollution. Furthermore, miniSASS and SASS5 can enhance collaboration among diverse stakeholders engaged in river monitoring. Additionally, testing the applicability of SASS5 and miniSASS will contribute to the development of suitable Biomonitoring tools for assessing the health of intermittent rivers in tropical regions

3 MATERIALS AND METHODS

3.1 Introduction

This section details the methodology used to achieve this study's aims (Section 1.4) and objectives (Section 1.5). This section will first describe the chosen study area, Upper Awash River, and its tributaries within the Awash River basin, the river flowing through the city of Addis Ababa, Ethiopia. Next, an overview of the research methodology is provided, including the selection of sampling sites, data collection, and the techniques used for data analysis.

3.2 Study Area

The Awash River is found in the Dandi District of the West Shewa Zone, Oromia Region, Ethiopia. It lies at a latitude of 9°5′N and a longitude of 38°10′E. The capital district, Ginchi, is located 75 kilometres west of Addis Ababa, the Ethiopian capital (Dabessa *et al*. (2021). The section covers an area of 109,729 hectares (Dabessa *et al*. (2021). The population living in the Awash River Basin was estimated to be more than 18 million, with a population density greater than 6,452 persons/km2.

Based on physical and socio-economic factors, the Awash River Basin is divided into three sections: the Upper Awash encompasses upstream areas from Koka Dam (over 1500 meters); the Middle Awash encompasses areas between Koka Dam and Awash Station (between 1000 and 1500 meters); and finally, the Lower Awash encompasses areas between 500 and 1000 meters in the Eastern catchment (Tadese *et al*., 2020).

The Upper Awash includes Addis Ababa city which lies within the Akaki watershed, which flows into Ethiopia's Awash River (Tefera *et al*., 2023). In the highlands of Entoto Mountain and other highlands, there are three major rivers: Kebena, Little Akaki, and Big Akaki (Tefera *et al*., 2023), which are important freshwater sources in the Akaki watershed, supplying Addis Ababa with water. These rivers flow from north to south, ultimately reaching the Awash River (Tefera et al., 2023). Additionally, the upper watershed also includes the Dire and Legedadi Reservoirs, which contribute to the city’s water (Tefera et al., 2023).

This study focuses on the Upper Awash River (Figure 3.1), located in the central Ethiopian highlands, with a climate that ranges from semi-arid to sub-humid (Kebede *et al*., 2020). The watershed’s monthly average maximum temperature ranges from 23.45 to 27.75 °C. Similarly, the watershed’s average monthly low temperature ranges from 9.43 to 13.240 °C (Daba *et al*., 2022). The rainfall in the upper Awash Basin is unimodal with main rainfall from June to September and very little falling between February and May season (Gebremichael *et al*., 2022).

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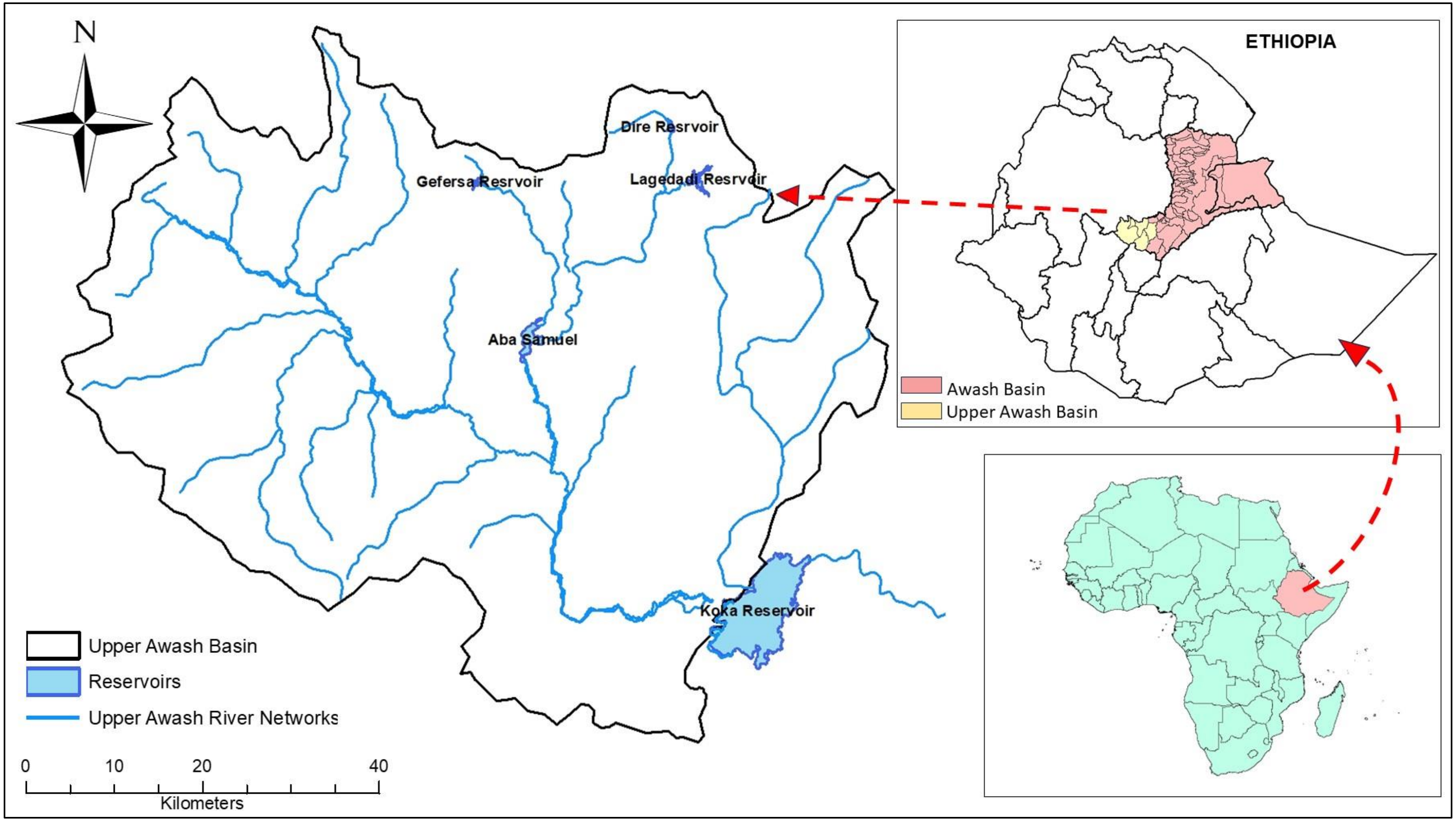


Figure 3.1 Map showing the location of the Upper Awash Basin and river networks

3.2.1 Land use and reservoirs

Making reference to Assegide et al. (2022), the Upper Awash basin is described as a basin that displays a variety of characteristics in terms of land uses. Primary land use involves agriculture supported by rain, which includes crops that rely on rainfall, shrubland, and grazing land. However, the basin also contains land that is irrigated, which the government mainly develops. The basin accounts for approximately 60% of the country's large-scale irrigated agriculture. Other land uses in the basin include industrial areas, forests, and reservoirs. Figure 3.2 displays the various land uses within the Upper Awash Basin.

Over 65% of Ethiopia's national industries are situated within the Awash basin, which is shared by the Afar, Amhara, Oromia, and Somali Regional States, as well as Addis Ababa and Dire Dawa City Administrations (Assegide et al., 2022). The main industrial concentration is found around the Little Akaki River and its primary tributaries in Addis Ababa (Kassegne et al., 2020). The Little and Tiliku Akaki Rivers serve as significant tributaries to the Awash River, merging into the Awash River system at Aba-Samuel Lake (Assegide et al., 2022).

One of the major cities, Addis Ababa, is located in the Awash basin. Addis Ababa records an annual average rainfall of approximately 1184 mm, with the wet season lasting from June to mid-September, peaking in July and August (Feyissa et al., 2018; Emna and Dubie, 2021). The dry period spans from October to May, with occasional rainfall observed between February and April (Emna and Dubie, 2021). March and May stand out as the warmest months in the area, while October and December mark the coldest (Emna and Dubie, 2021). The air temperatures vary between 10 and 29 degrees Celsius, encompassing the minimum and maximum temperatures (Emna and Dubie, 2021).

There are 4 major reservoirs within the Upper Awash basin. The Legedadi Reservoir, which is the major drinking water supply source for Addis Ababa (Habtemariam *et al*., 2021). Another important reservoir is the Dire reservoir which supplies drinking water to the city (Admasu *et al*., 2023). The Great Akaki River courses through the city's eastern section, comprising numerous southward-flowing tributaries that traverse residential and commercial hubs. Converging 37 km southwest of Addis Ababa, the two major rivers meet at the Aba Samuel Reservoir (Kassegne *et al*., 2020). This reservoir inadvertently functions as a repository for pollutants originating from both upstream Addis Ababa and agrochemicals discharged by various scales of agricultural operations, including irrigation farms and floriculture (Kassegne *et al*., 2020).

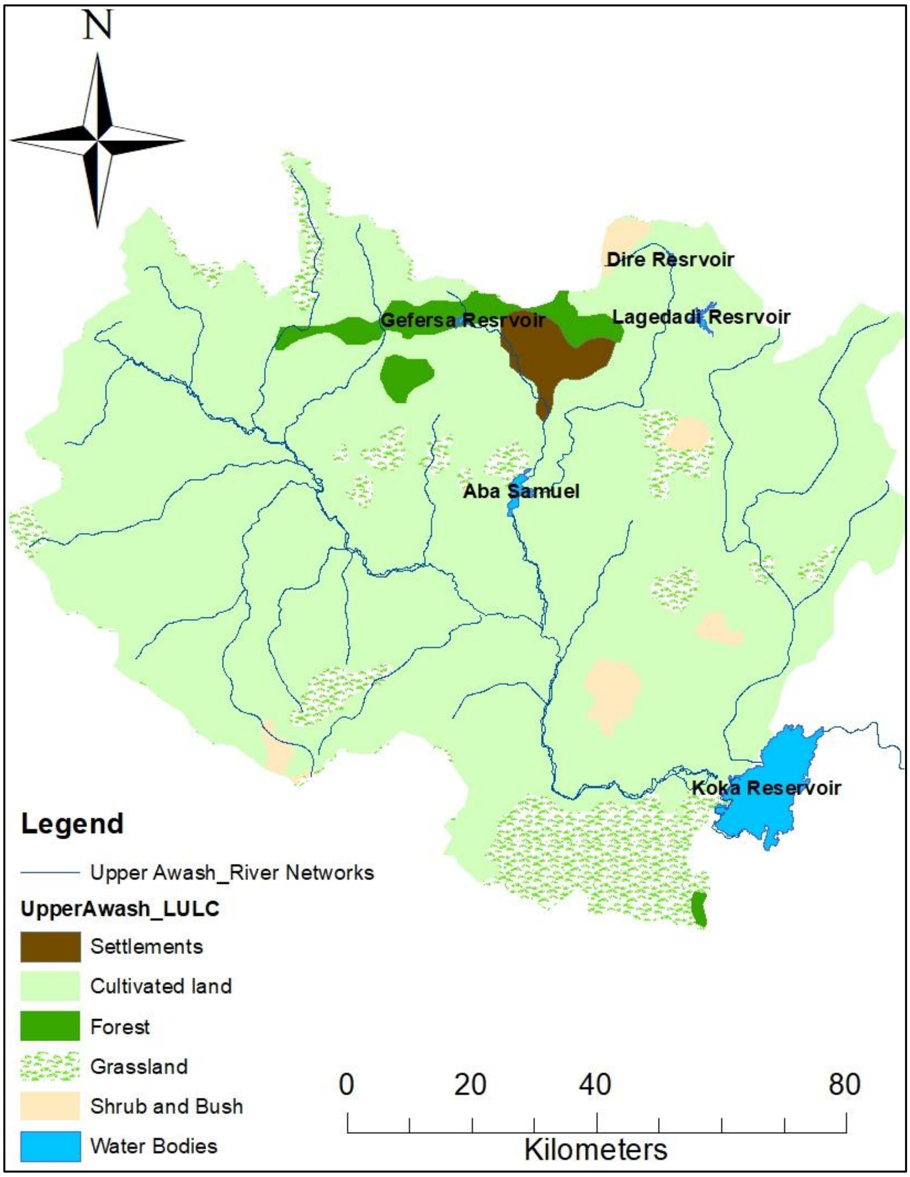


Figure 3.2 Various land uses within the Upper Awash Basin (Wolde,2022)

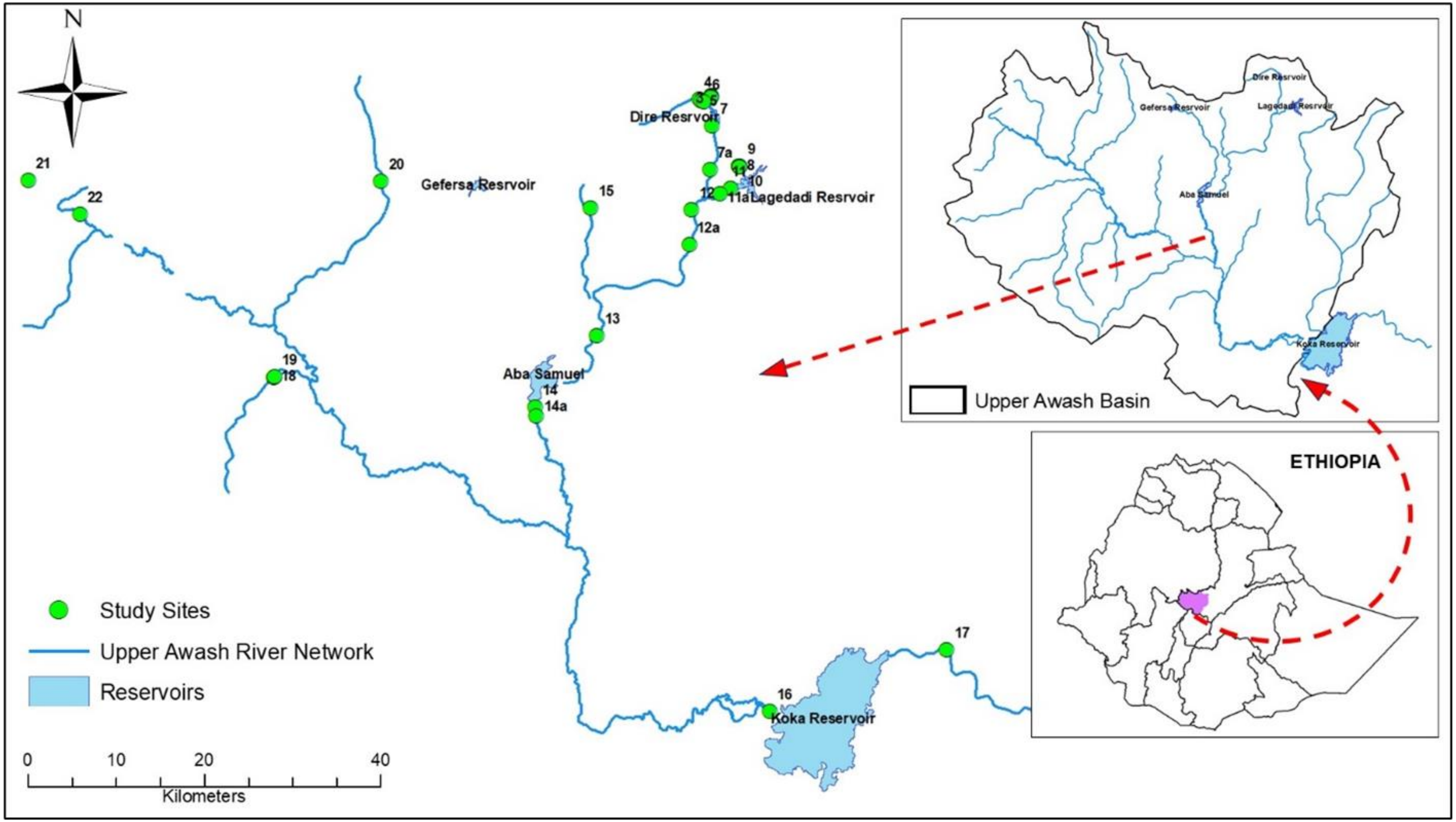


Figure 3.3 Map of Ethiopia and sampling sites along the tributaries of the Upper Awash River

Figure 3.4 Pictures taken above the Legedadi Reservoir during the wet (left) and dry (right) season (Author, 2023)

3.3 Methodology of Sampling

The study focused on the Upper Awash River and its tributaries (Figure 3.3) because of the land use diversity associated with the river. Sampling was conducted along a highland-lowland transect. A total of 17 sample sites were sampled during the wet (July 2022) and a total of 26 sample sites during the dry (mid-January to mid-February 2023) season (Figure 3.4) along the intermittent tributaries of the upper Awash River. Table 8.1 in Appendix A gives a description of each site. The standard aim for selecting the sampling sites was to determine the changes in water quality from upstream to downstream, whereby upstream in the highlands, the river quality is expected to be in good condition with less interference from humans. To also determine the impact of different land uses, such as agriculture, on the river quality. It is also to determine changes after the confluence of tributaries and to detect the degree of influence of reservoirs and urban/ rural settlements on water quality. Ethiopia's surface water bodies are seriously threatened by the country's recent fast urbanization and population increase, particularly in Addis Ababa (Dessie *et al*., 2024). The wastewater treatment system of the city has not kept up with the issues, resulting in a number of environmental threats. (Dessie *et al*., 2024). Therefore, sample sites were also located within the city to determine the urbanisation impact on the river quality.

The primary method used in this study followed a similar sampling technique as that of miniSASS and SASS5, where miniSASS and SASS5 tests were conducted along with physicochemical parameters in the Upper Awash intermittent tributaries. To avoid bias, miniSASS was conducted by an independent operator and SASS5 was conducted by the author. Sampling was conducted during the peak wet season and repeated during the dry season. Physicochemical parameters were measured in situ. The importance of sampling during the peak wet season was because intermittent rivers are expected to flow when there is adequate input of rainfall to sustain surface flow which then represents a lotic system that is suitable for conducting a SASS5 and miniSASS test. And then sampling during the peak dry season in intermittent rivers was able to help determine the challenges and sources of error associated with performing a miniSASS and SASS test in unsuitable conditions. During the dry season, taking macroinvertebrates is generally justified by the following: during a dry period, the traces of the greatest anthropogenic impacts are easier to determine, and the dry season is when there are the most macroinvertebrate species and species richness (Assefa *et al*., 2023). In the case of finding unfamiliar aquatic invertebrates, the specimens were sent to a specialist at Addis Ababa University for identification. Table 8.2 in Appendix B displays the macroinvertebrates that were found and identified using the SASS5 score sheet. Figures 8.2 and 8.3 in Appendix B show examples of macroinvertebrates that were found.

The type of data that was collected included:

- Physicochemical data and heavy metals
- Aquatic macroinvertebrates

The main features that were also considered included:

- Availability of suitable biotopes (Stones, Vegetation, Gravel, Sand and Mud)
- Depth/flow of water
- Grid reference (latitude and altitude- will be recorded using the Geographic Positioning System (GPS))
- Instream/ riparian disturbances
- Other biotas

3.3.1 Physicochemical Data

The physicochemical parameters were collected before disturbing the various biotopes in the targeted area. The following parameter measurements were recorded:

- pH
- temperature (°C)
- dissolved oxygen (mg/L)
- electrical conductivity (µs/cm)

using a handheld multiparameter (PCD650 meter) onsite whereby the required probes set to the required measurements mode were dipped into the sample site, fully immersed in water. In the water, the probes were gently moved in a stirring motion to create a homogenous sample until the reading stabilized to take the reading.

According to the SASS5/miniSASS scoring sheets, the following parameters were also tested in-situ before applying the SASS5 and miniSASS method. (i) A clarity test was conducted using a clarity tube. The tube was first rinsed three times before filling it with the water from the sample site. Then after the tube was filled to the brim with no air bubbles it was then closed with a lid. Then the tube was held up at 90 degrees perpendicular to the sun as shown in Figure 3.5. By observing through the view end of the tube, sliding the black magnet away from the eye until it stops being visible, and using the numbers outside the tube measured in centimetres, y1 was recorded. Then sliding the magnet from the back end until it appears, y2 was then recorded. The visual clarity was calculated using the average of the two distances.

$$\text{Visual clarity} = (y1+y2)/2 \tag{3.7}$$

Y1= Number observed from the front end

Y2= Number observed from the back end

Thereafter for turbidity, the water was rated from very low to high, the colour was also observed based on the SASS5 and miniSASS guides.

(ii) The flow of water was determined using the transparent velocity head rod.
After that, the results were recorded in both the SASS5 and miniSASS score sheets.

Figure 3.5 Transparent velocity head rod (left), Clarity tube (middle) and multiparameter PCD650 meter (right)

3.3.2 Heavy metals and other metals

Water samples were collected along the sample sites before disturbance, 30cm below the surface of the water, using one-litre polyethylene bottles; the water bottles were washed three times with the sample water before being filled. The collected samples were unfiltered, and one drop of nitric acid (1M) was added to the samples, which were then stored in an ice box before transporting to the laboratory. The tested sample matrix was liquid, total heavy metals and other metals, Zinc (Zn), Chromium (Cr), Magnesium (Mg), Mercury (Hg), Manganese (Mn), Lead (Pb), Copper (Cu), Iron (Fe), Potassium (K), and Cadmium (Cd). The samples were a portion of homogenised samples that were acid-digested and then analysed by ICP-OES (EPA Method 6010B). Inductively Coupled Plasma-Optical Emission Spectrometry, Revision 2, December 1996.

3.3.3 SASS5

Following the sample collection method from Dickens and Graham (2002), sampling were taken at the following biotopes, stones (S), vegetation (Veg) and gravel, sand, and mud (GSM). Samples were collected over a wide range to ensure diversity.

Depending on the type of biotopes that were found at a particular site, the first biotope sampled was the stones biotope, starting with stones in current (SIC). Stones in current consist of bedrock (boulders > 25cm) or loose stones, which are located where the flowing water does not allow fine silt to settle on the stones (Dickens and Graham, 2002). Stones in current were kicked and rubbed with boots (bedrock) to dislodge the macroinvertebrates into the net, placed closely but downstream of the stones. The technique of sampling was done for approximately two minutes.

The second biotope sampled was stones out of current (SOOC), which consisted of stones, and bedrock susceptible to fine sediments settling on their upper surface with minimal disturbance from flowing water. The SOOC were sampled for approximately one minute following the same sampling technique as SIC. After that, the samples collected from both biotopes formed into a stone’s biotope sample.

In sites with vegetation (marginal vegetation, usually found on the banks of the river, can be in and out of current, for example, reeds and aquatic vegetation, fully submerged, for instance, water hyacinth (Dickens and Graham, 2002)) SASS5 was applied. Marginal vegetation was sampled for a total length of two meters which is approximately equivalent to 6 nets distributed across the river's diverse marginal vegetation and different water flow velocities. Marginal vegetation was sampled by pushing the net into the vegetation in a continuous backwards and forward motion to dislodge macroinvertebrates into the net. For aquatic vegetation, macroinvertebrates were dislodged into the net by pushing it repeatedly against and through the vegetation for one square meter.

The third biotopes that samples were taken from was from gravel (stones less than 2cm), sand (sand particles less than 2mm in diameter) and mud (including silt and clay with particles less than 0.006mm in diameter). These biotopes were sampled for one minute by shuffling with boots to create a disturbance and dislodge macroinvertebrates into the net. In between sampling, macroinvertebrates that may have been missed using the sampling techniques were sampled by either hand-picking or visual observation and recorded on the SASS scoring sheet (Figure 3.5). In sites that formed pools, too shallow and dry, SASS5 was not applied.

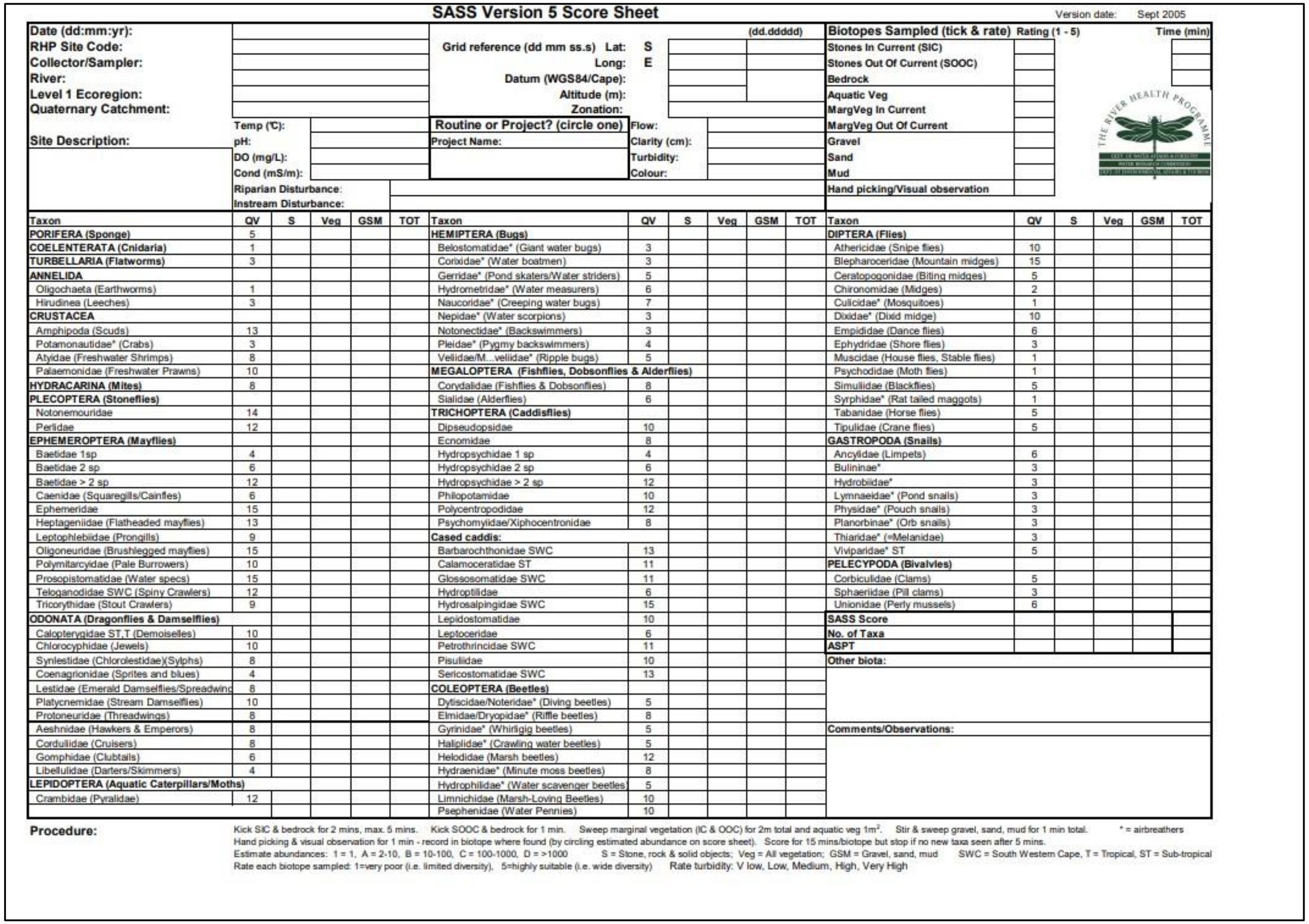
Next, the sampled macroinvertebrates were cleaned and placed in separate trays (indicating the different biotopes) and filled with clean water. All the large and small sediments, including debris, were removed and ensured clear identification.

Each biotope's macroinvertebrates were observed for a duration of 15 minutes, and the identified taxa were marked under the corresponding biotope on a SASS version 5 score sheet, as illustrated in Figure 3.5. Additional details needed for interpretation were also recorded on the SASS5 sheet, following the guidance provided by Dickens and Graham (2002).
Subsequently, the results were computed as follows:

SASS score = sum of quality value scores for each taxon ticked in the total column (3.1)

Number of Tax (No. Taxa) = The total number of taxa found (3.2)

ASTP = SASS score/ No. Taxa (3.3)



SASS Version 5 Score Sheet

Version date: Sept 2005

Date (dd:mm:yr):
RHP Site Code:
Collector/Sampler:
River:
Level 1 Ecoregion:
Quaternary Catchment:
Site Description:

Temp (℃):
pH:
DO (mg/L):
Cond (mS/m):
Riparian Disturbance:
Instream Disturbance:

(dd.ddddd)
Grid reference (dd mm ss.s) Lat: S
Long: E
Datum (WGS84/Cape):
Altitude (m):
Zonation:

Routine or Project? (circle one)
Project Name:

Flow:
Clarity (cm):
Turbidity:
Colour:

Biotopes Sampled (tick & rate)	Rating (1 - 5)	Time (min)
Stones In Current (SIC)		
Stones Out Of Current (SOOC)		
Bedrock		
Aquatic Veg		
MargVeg In Current		
MargVeg Out Of Current		
Gravel		
Sand		
Mud		
Hand picking/Visual observation		

Taxon	QV	S	Veg	GSM	TOT	Taxon	QV	S	Veg	GSM	TOT	Taxon	QV	S	Veg	GSM	TOT
PORIFERA (Sponge)	5					**HEMIPTERA (Bugs)**						**DIPTERA (Flies)**					
COELENTERATA (Cnidaria)	1					Belostomatidae* (Giant water bugs)	3					Athericidae (Snipe flies)	10				
TURBELLARIA (Flatworms)	3					Corixidae* (Water boatmen)	3					Blepharoceridae (Mountain midges)	15				
ANNELIDA						Gerridae* (Pond skaters/Water striders)	5					Ceratopogonidae (Biting midges)	5				
Oligochaeta (Earthworms)	1					Hydrometridae* (Water measurers)	6					Chironomidae (Midges)	2				
Hirudinea (Leeches)	3					Naucoridae* (Creeping water bugs)	7					Culicidae* (Mosquitoes)	1				
CRUSTACEA						Nepidae* (Water scorpions)	3					Dixidae* (Dixid midge)	10				
Amphipoda (Scuds)	13					Notonectidae* (Backswimmers)	3					Empididae (Dance flies)	6				
Potamonautidae* (Crabs)	3					Pleidae* (Pygmy backswimmers)	4					Ephydridae (Shore flies)	3				
Atyidae (Freshwater Shrimps)	8					Veliidae/M...veliidae* (Ripple bugs)	5					Muscidae (House flies, Stable flies)	1				
Palaemonidae (Freshwater Prawns)	10					**MEGALOPTERA (Fishflies, Dobsonflies & Alderflies)**						Psychodidae (Moth flies)	1				
HYDRACARINA (Mites)	8					Corydalidae (Fishflies & Dobsonflies)	8					Simuliidae (Blackflies)	5				
PLECOPTERA (Stoneflies)						Sialidae (Alderflies)	6					Syrphidae* (Rat tailed maggots)	1				
Notonemouridae	14					**TRICHOPTERA (Caddisflies)**						Tabanidae (Horse flies)	5				
Perlidae	12					Dipseudopsidae	10					Tipulidae (Crane flies)	5				
EPHEMEROPTERA (Mayflies)						Ecnomidae	8					**GASTROPODA (Snails)**					
Baetidae 1sp	4					Hydropsychidae 1 sp	4					Ancylidae (Limpets)	6				
Baetidae 2 sp	6					Hydropsychidae 2 sp	6					Bulininae*	3				
Baetidae > 2 sp	12					Hydropsychidae > 2 sp	12					Hydrobiidae*	3				
Caenidae (Squaregills/Cainfles)	6					Philopotamidae	10					Lymnaeidae* (Pond snails)	3				
Ephemeridae	15					Polycentropodidae	12					Physidae* (Pouch snails)	3				
Heptageniidae (Flatheaded mayflies)	13					Psychomyiidae/Xiphocentronidae	8					Planorbinae* (Orb snails)	3				
Leptophlebiidae (Prongills)	9					**Cased caddis:**						Thiaridae* (=Melanidae)	3				
Oligoneuridae (Brushlegged mayflies)	15					Barbarochthonidae SWC	13					Viviparidae* ST	5				
Polymitarcyidae (Pale Burrowers)	10					Calamoceratidae ST	11					**PELECYPODA (Bivalvles)**					
Prosopistomatidae (Water specs)	15					Glossosomatidae SWC	11					Corbiculidae (Clams)	5				
Teloganodidae SWC (Spiny Crawlers)	12					Hydroptilidae	6					Sphaeriidae (Pill clams)	3				
Tricorythidae (Stout Crawlers)	9					Hydrosalpingidae SWC	15					Unionidae (Perly mussels)	6				
ODONATA (Dragonflies & Damselflies)						Lepidostomatidae	10					**SASS Score**					
Calopterygidae ST,T (Demoiselles)	10					Leptoceridae	6					**No. of Taxa**					
Chlorocyphidae (Jewels)	10					Petrothrincidae SWC	11					**ASPT**					
Synlestidae (Chlorolestidae)(Sylphs)	8					Pisuliidae	10					**Other biota:**					
Coenagrionidae (Sprites and blues)	4					Sericostomatidae SWC	13										
Lestidae (Emerald Damselflies/Spreadwinc	8					**COLEOPTERA (Beetles)**											
Platycnemidae (Stream Damselflies)	10					Dytiscidae/Noteridae* (Diving beetles)	5										
Protoneuridae (Threadwings)	8					Elmidae/Dryopidae* (Riffle beetles)	8										
Aeshnidae (Hawkers & Emperors)	8					Gyrinidae* (Whirligig beetles)	5					**Comments/Observations:**					
Corduliidae (Cruisers)	8					Haliplidae* (Crawling water beetles)	5										
Gomphidae (Clubtails)	6					Helodidae (Marsh beetles)	12										
Libellulidae (Darters/Skimmers)	4					Hydraenidae* (Minute moss beetles)	8										
LEPIDOPTERA (Aquatic Caterpillars/Moths)						Hydrophilidae* (Water scavenger beetles)	5										
Crambidae (Pyralidae)	12					Limnichidae (Marsh-Loving Beetles)	10										
						Psephenidae (Water Pennies)	10										

Procedure:

Kick SIC & bedrock for 2 mins, max. 5 mins. Kick SOOC & bedrock for 1 min. Sweep marginal vegetation (IC & OOC) for 2m total and aquatic veg 1m^2. Stir & sweep gravel, sand, mud for 1 min total. * = airbreathers
Hand picking & visual observation for 1 min - record in biotope where found (by circling estimated abundance on score sheet). Score for 15 mins/biotope but stop if no new taxa seen after 5 mins.
Estimate abundances: 1 = 1, A = 2-10, B = 10-100, C = 100-1000, D = >1000 S = Stone, rock & solid objects; Veg = All vegetation; GSM = Gravel, sand, mud SWC = South Western Cape, T = Tropical, ST = Sub-tropical
Rate each biotope sampled: 1=very poor (i.e. limited diversity), 5=highly suitable (i.e. wide diversity) Rate turbidity: V low, Low, Medium, High, Very High

Figure 3.6 SASS5 Version 5 score sheet

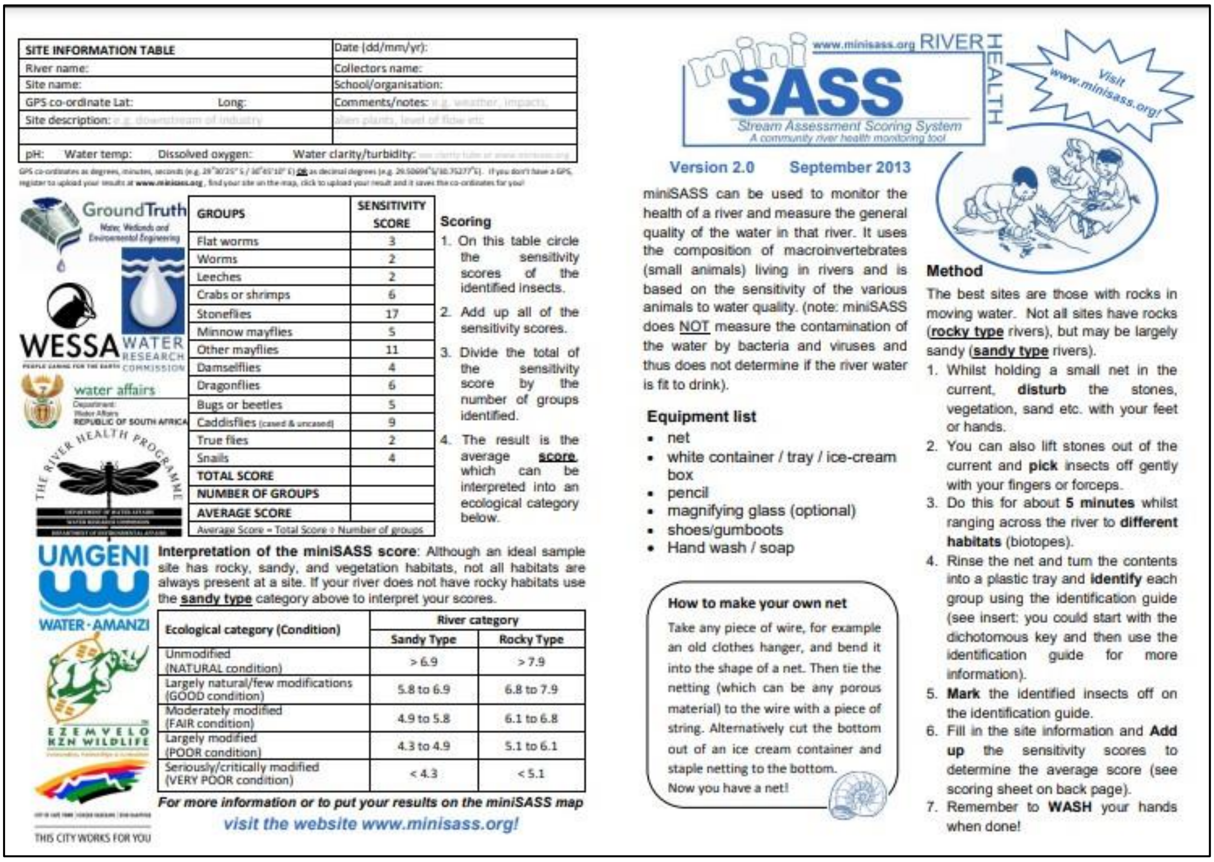
3.3.4 miniSASS

Following the guidelines from (www.minisass.org), samples were collected by another sampler to avoid biasness. Samples were collected from three biotopes, site dependent, a rocky biotope/ river, a vegetative biotope (with vegetation usually on the sides) and the GSM biotope. The three biotopes were sampled for five minutes combined. The sampling techniques of biotopes were similar to that of SASS5, whereby kicking, shuffling and pushing the net into vegetation are required. After that, the collected samples were prepared by being deposited into different trays according to their biota and filled with clean water.

Next, the macroinvertebrates were identified using the identification guide and the Dichotomous key of macroinvertebrates in Figure 8.1 (Appendix B). After the identification, the macroinvertebrates were scored using the miniSASS scoring sheet Figure 3.7, thereafter the river's health was calculated as follows:

Total score = sum of the sensitivity scores of the identified macroinvertebrate (3.4)
Number of Groups = the number of groups found (3.5)
Average score = Total score/ number of groups (3.6)

After calculating the average scores, the river's ecological condition was then categorized across five categories ranging from blue, which represents natural conditions, to purple, which represents very poor conditions, as seen in Figure 3.7. Thereafter the miniSASS website Google Earth layer (www.minisass.org) was also used to record/ store the results.



SITE INFORMATION TABLE	Date (dd/mm/yr):
River name:	Collectors name:
Site name:	School/organisation:
GPS co-ordinate Lat: Long:	Comments/notes: e.g. weather, impacts,
Site description: e.g. downstream of industry	alien plants, level of flow etc
pH: Water temp: Dissolved oxygen: Water clarity/turbidity:	

GPS co-ordinates as degrees, minutes, seconds (e.g. 29°30'25" S / 30°45'10" E) OR as decimal degrees (e.g. 29.50694°S/30.75277°E). If you don't have a GPS, register to upload your results at **www.minisass.org**, find your site on the map, click to upload your result and it saves the co-ordinates for you!

GROUPS	SENSITIVITY SCORE
Flat worms	3
Worms	2
Leeches	2
Crabs or shrimps	6
Stoneflies	17
Minnow mayflies	5
Other mayflies	11
Damselflies	4
Dragonflies	6
Bugs or beetles	5
Caddisflies (cased & uncased)	9
True flies	2
Snails	4
TOTAL SCORE	
NUMBER OF GROUPS	
AVERAGE SCORE	
Average Score = Total Score ÷ Number of groups	

Scoring

1. On this table circle the sensitivity scores of the identified insects.
2. Add up all of the sensitivity scores.
3. Divide the total of the sensitivity score by the number of groups identified.
4. The result is the average **score** which can be interpreted into an ecological category below.

Interpretation of the miniSASS score: Although an ideal sample site has rocky, sandy, and vegetation habitats, not all habitats are always present at a site. If your river does not have rocky habitats use the **sandy type** category above to interpret your scores.

Ecological category (Condition)	River category	
	Sandy Type	Rocky Type
Unmodified (NATURAL condition)	> 6.9	> 7.9
Largely natural/few modifications (GOOD condition)	5.8 to 6.9	6.8 to 7.9
Moderately modified (FAIR condition)	4.9 to 5.8	6.1 to 6.8
Largely modified (POOR condition)	4.3 to 4.9	5.1 to 6.1
Seriously/critically modified (VERY POOR condition)	< 4.3	< 5.1

For more information or to put your results on the miniSASS map

visit the website www.minisass.org!

mini SASS www.minisass.org RIVER HEALTH
Stream Assessment Scoring System
A community river health monitoring tool

Version 2.0 September 2013

miniSASS can be used to monitor the health of a river and measure the general quality of the water in that river. It uses the composition of macroinvertebrates (small animals) living in rivers and is based on the sensitivity of the various animals to water quality. (note: miniSASS does NOT measure the contamination of the water by bacteria and viruses and thus does not determine if the river water is fit to drink).

Equipment list

- net
- white container / tray / ice-cream box
- pencil
- magnifying glass (optional)
- shoes/gumboots
- Hand wash / soap

How to make your own net

Take any piece of wire, for example an old clothes hanger, and bend it into the shape of a net. Then tie the netting (which can be any porous material) to the wire with a piece of string. Alternatively cut the bottom out of an ice cream container and staple netting to the bottom. Now you have a net!

Method

The best sites are those with rocks in moving water. Not all sites have rocks (**rocky type** rivers), but may be largely sandy (**sandy type** rivers).

1. Whilst holding a small net in the current, **disturb** the stones, vegetation, sand etc. with your feet or hands.
2. You can also lift stones out of the current and **pick** insects off gently with your fingers or forceps.
3. Do this for about **5 minutes** whilst ranging across the river to **different habitats** (biotopes).
4. Rinse the net and turn the contents into a plastic tray and **identify** each group using the identification guide (see insert: you could start with the dichotomous key and then use the identification guide for more information).
5. **Mark** the identified insects off on the identification guide.
6. Fill in the site information and **Add up** the sensitivity scores to determine the average score (see scoring sheet on back page).
7. Remember to **WASH** your hands when done!

Figure 3.7 miniSASS score and interpretation sheet version 2.0 (www.minisass.org, 2023)

3.4 Data and Statistical Analysis

Data was entered into Microsoft Excel 2021 for basic scatter plot graphs to show the data trends, which display the upstream and downstream variation of the various parameters influenced by different land uses during the wet and dry seasons. Thereafter, the data were analysed using the Correlation matrix (to assess the relationship between variables); then the multivariate analyses (analysis that considers multiple factors) were performed using Minitab 15 software to perform Principal Component Analysis (PCA) (statistical technique that simplifies large data sets) to reveal the relationship between the SASS5 and miniSASS metrics, physicochemical parameters, and heavy metals. The steps taken to perform PCA plots included using the size of the eigenvalue (Appendix D) to determine the number of principal components. The principal components that displayed the largest eigenvalues were retained. Thereafter, the scree plot was arranged from largest to smallest, where the components in the steep curve before the first point that starts the straight line were used (Appendix D).

A Hierarchical Cluster Analysis (HCA) (method used to group similar data points into clusters), based on correlation coefficient distance (complete linkage) to show the similarities between the sample sites has also been performed. These statistical methods have been commonly used as unbiased analysis in the study of water quality data, allowing for the extraction of significant findings (Kane and Lazo, 2015). They are used to evaluate and assess water quality, allowing for the examination of spatial and temporal fluctuations caused by both natural and man-made processes (Kane and Lazo, 2015). This multivariate data analysis has been shown to effectively reduce data without compromising original information (Kane and Lazo, 2015). Muniz and Oliveira-Filho (2023) also state that particularly, PCA has been widely applied as a method in the analysis river water quality. Furthermore, the HCA of variables was used to group the parameters based on the similarities and represented using the dendrogram, which is a diagram that provides visual observations of the clustering process. Additionally, physicochemical parameters and heavy metals has been compared to the Guideline Ambient Environment Standards for Ethiopia (Water Quality Standards) using spatial plots.

3.5 Limitations and Challenges

The selection of sample sites was conducted through the Google Earth engine. However, the main challenge with the number of sites chosen is the accessibility of sites. In some cases, permission needed to be granted prior to accessing the site, which delayed sampling sites consecutively. Addis Ababa is a populated city; therefore, issues of traffic affected the consistency of sampling at a fixed time. Sampling had to take place at different times of the day. Sampling in heavily polluted streams poses a health risk to samplers, therefore, influences the time taken to complete sampling in that particular site and also affects the urge and willingness to expand sample sites. The miniSASS and SASS5 do not have visuals of macroinvertebrates in their larvae stages, which makes it difficult to identify and requires experts to identify those species.

In relation to sampling, performing the miniSASS and SASS5 techniques in small and not-too-deep rivers was easier compared to wide and deep rivers, especially during the wet season. It is unsafe and challenging to collect samples of macroinvertebrates and environmental variables during the wet season for some of the rivers are deep and fast flowing. However, during the dry season, it was difficult to sample in sites that were too shallow and rocky. This affected the outcome of the results. Hence, in places where SASS5 could not be applied this contributed to missing values.

The selected water quality parameters, such as heavy metals, were influenced by the time it took to transport samples from Ethiopia to South Africa. Overall, the main concern with heavy metal data used in this study, was that some water samples were lost during transportation for certain sites. For this reason, total heavy metals were used in the study.

4 RESULTS

4.1 Physicochemical Parameters

The spatial and temporal variations of the five selected physicochemical parameters are shown in this section. The sample sites are numbered from 1 to 24, which represent the chosen tributaries of the Upper Awash basin. The parameters were observed over a period of two seasons (dry and wet season). The results from the different sites were measured to see if they exceed or fall within the Ethiopian EPA (Water Quality Standards- Surface Waters). Figure 4.1 shows the variation of temperature across the sample sites during the wet and dry season. The maximum temperature measured is 24.8 (°C) at site S16 during the dry season. And the minimum temperature measured is 13.7 at S1 during the wet season and S15 during the dry season.

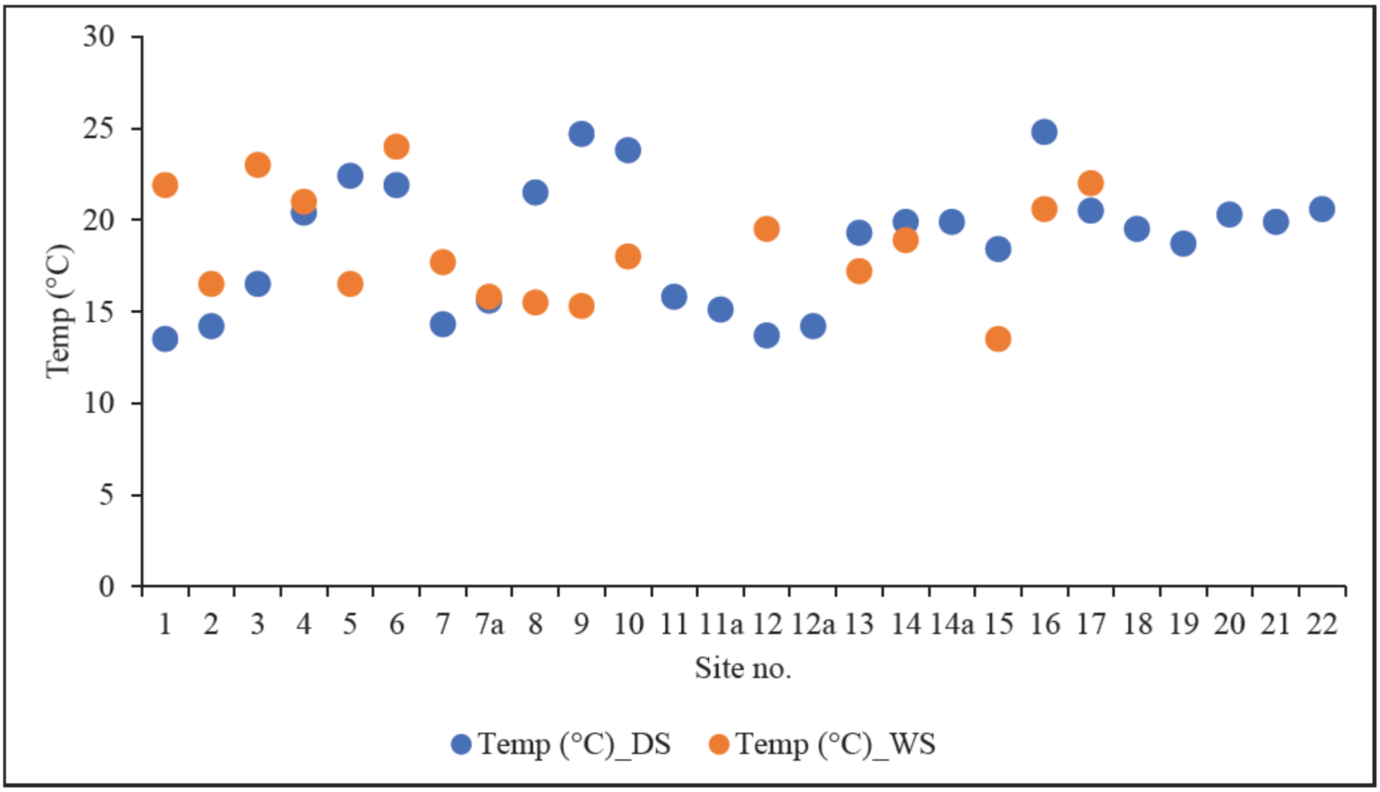


Figure 4.1 Measured temperature (°C) across the sample sites in the Upper Awash basin during the wet and dry season

Figure 4.2 shows pH variations amongst the sample sites during the wet and dry season. It is observed that majority of pH levels are slightly higher during the dry season than pH levels measured during the wet season. The pH levels lie between 7.4 and 9, which indicate that the sample sites are alkaline.

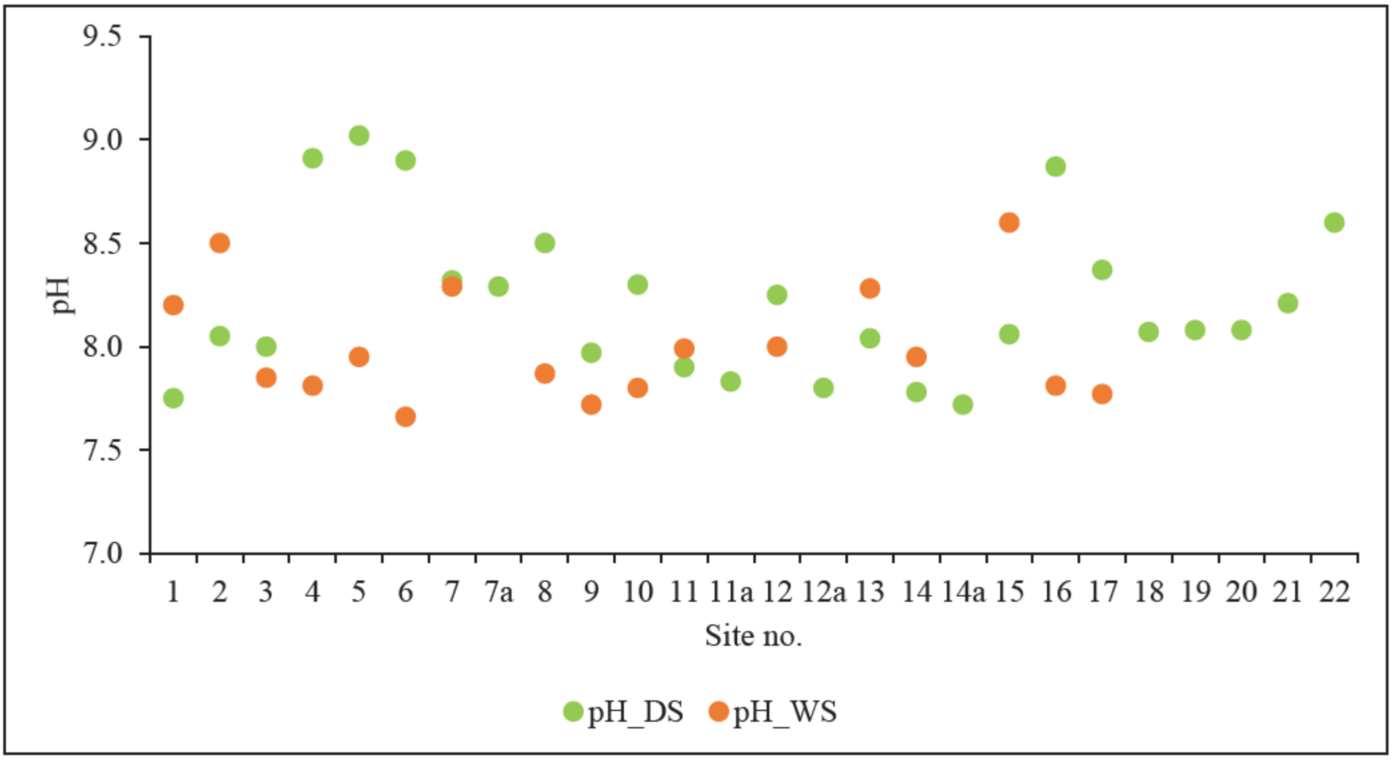


Figure 4.2 Measured pH across sample sites in the Upper Awash basin during the wet and dry season

Figure 4.3 shows the majority of DO (mg/L) values measured across the sample sites are above the Ethiopian EPA minimum for Course fish and below the Ethiopian EPA minimum for Game fish. S15 (Kebena) recorded the lowest DO during the dry season. From S1 to S6 (Above the Dire Reservoir), the DO levels during the dry season are higher compared to the wet season. For sites showing high levels of DO reflects that there is significant amount of oxygen dissolved in the water, which is crucial for the survival of aquatic organisms.

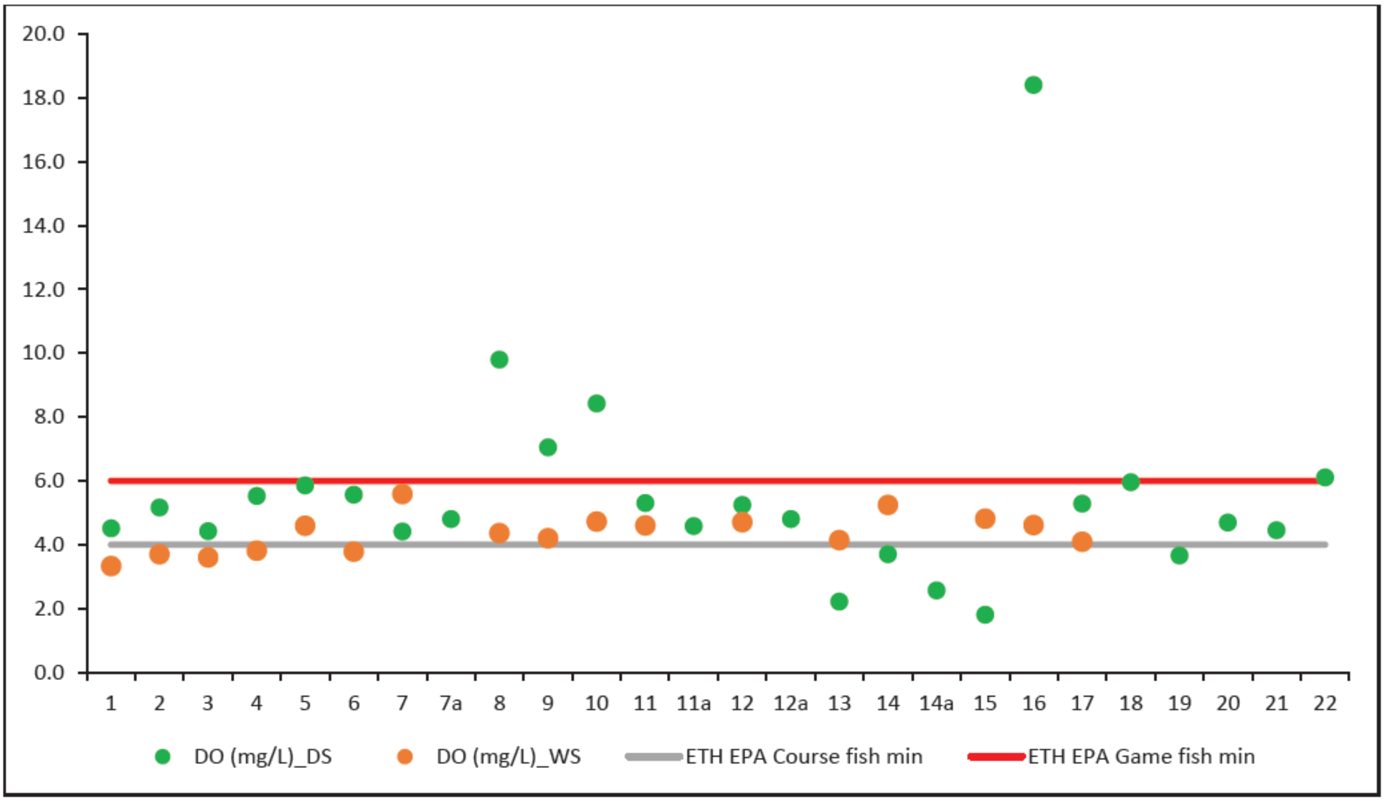


Figure 4.3 Measured DO values across the sample sites in the Upper Awash basin during the wet and dry season

The EC values for the dry seasons range from 171 (µs/cm) to 1094 (µs/cm) and during the wet season the EC values range from 146 (µs/cm) to 754 (µs/cm) as displayed in Figure 4.4. Majority of the sites during the dry season recorded EC values that are below the maximum Ethiopian EPA standards of 1000 µs/cm except for two sites, S14 and S14a located after the Aba Samuel Reservoir with EC values of 1090 and 1094 (µs/cm). During the wet season, all the sites recorded EC values below the Ethiopian EPA standard and the lowest recorded EC value is at S2 located upstream of Dire Reservoir. The graph also shows that the EC values for the wet season are relatively higher compared to the dry season.

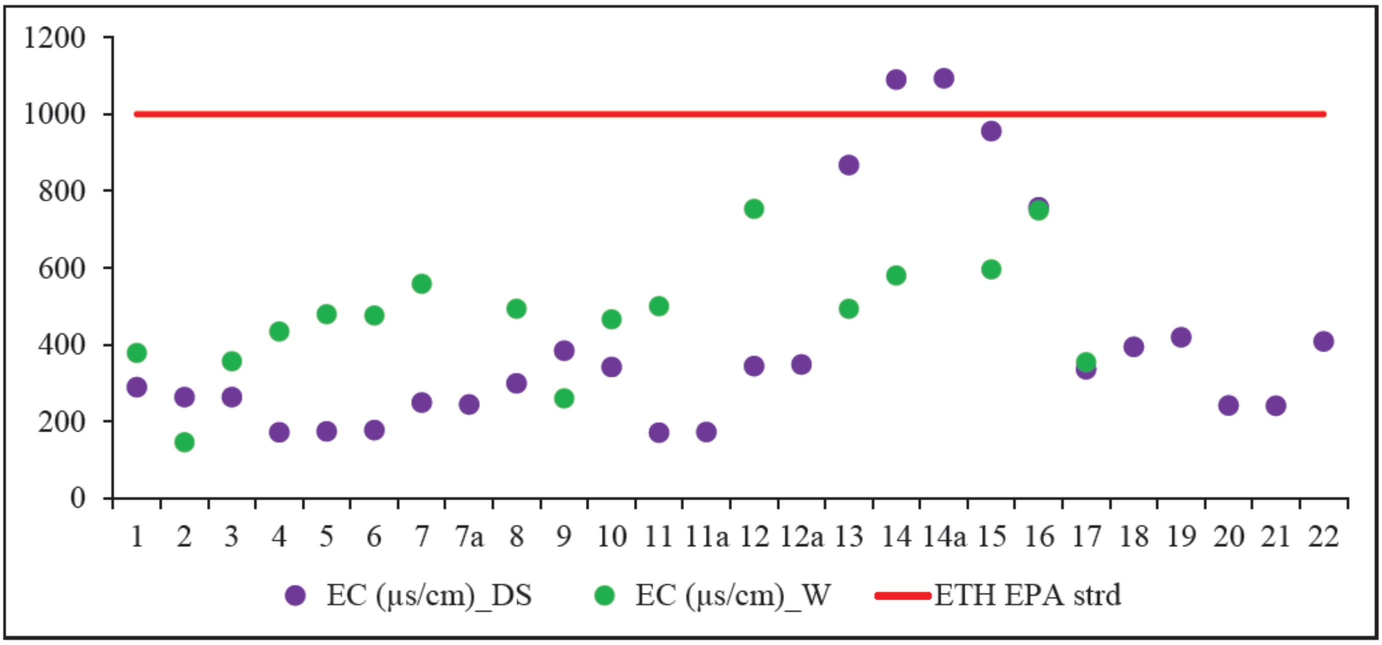


Figure 4.4 Measured EC (µs/cm) across the sample sites in the Upper Awash basin during the wet and dry season

Figure 4.5 shows the measured turbidity of water at the different sites. The majority of the sites recorded high clarity levels during the dry seasons compared to the wet season. Site 10 (Upstream of Legedadi) recorded the highest clarity level during the dry season and Site 6 recorded the highest during the wet season. The lowest observed clarity levels fall in site 11, 11a (Downstream of Legedadi), 12, 12a (Confluence) and 15 (Kebena) during the dry season.

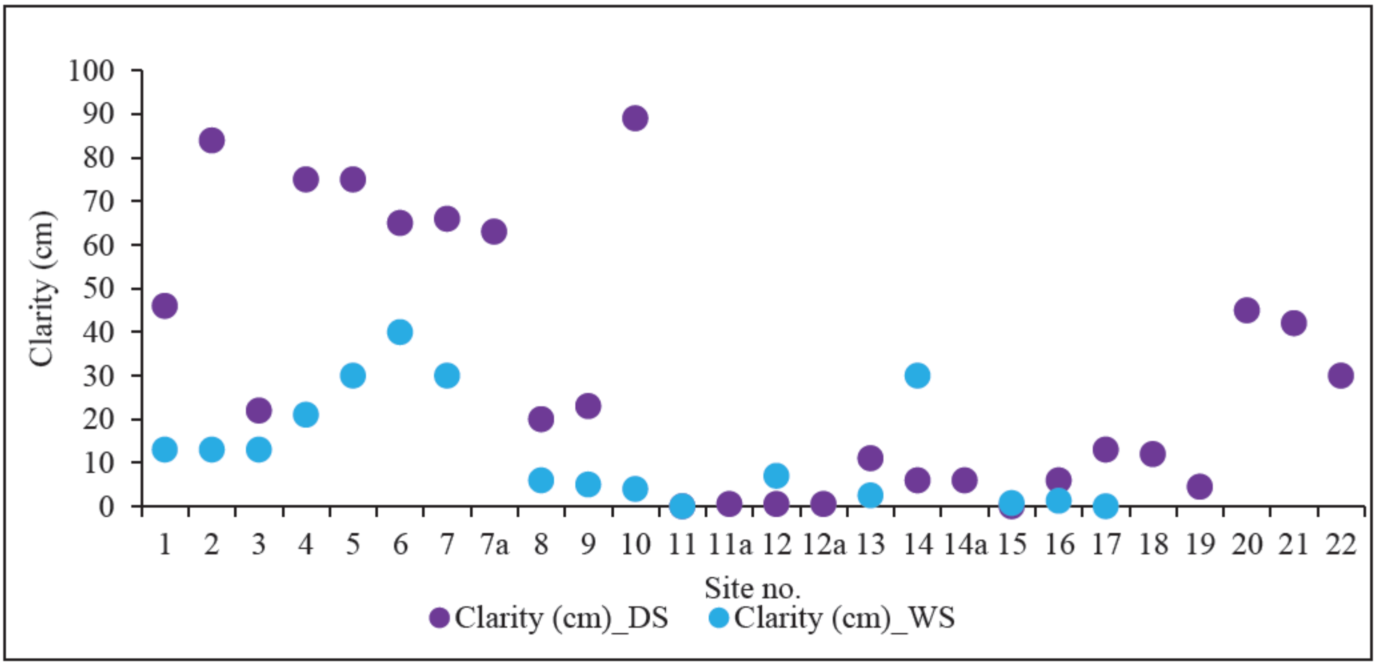


Figure 4.5 Measured Clarity (cm) across the sample sites in the Upper Awash basin during the wet and dry season

4.2 Heavy Metals

The spatial and temporal variations of the seven selected total heavy metals and two other metals are shown in this section. The sample sites are numbered from 1 to 22 which represent the chosen tributaries of the Upper Awash basin. The metals were observed over a period of two seasons (dry and wet season). The results from the different sites were measured to see if they exceeded or fell within the Ethiopian EPA (Water Quality Standards- Surface Waters). Figure 4.6 shows the variation of Magnesium (Mg) across the sample sites during the wet and dry seasons. High Mg measurements were observed in S11 and S11a. The lowest measured Mg was observed at site 12 after the confluence of the Dire and Legedadi rivers. Unfortunately, the Ethiopian EPA does not have a maximum or minimum requirement for magnesium.

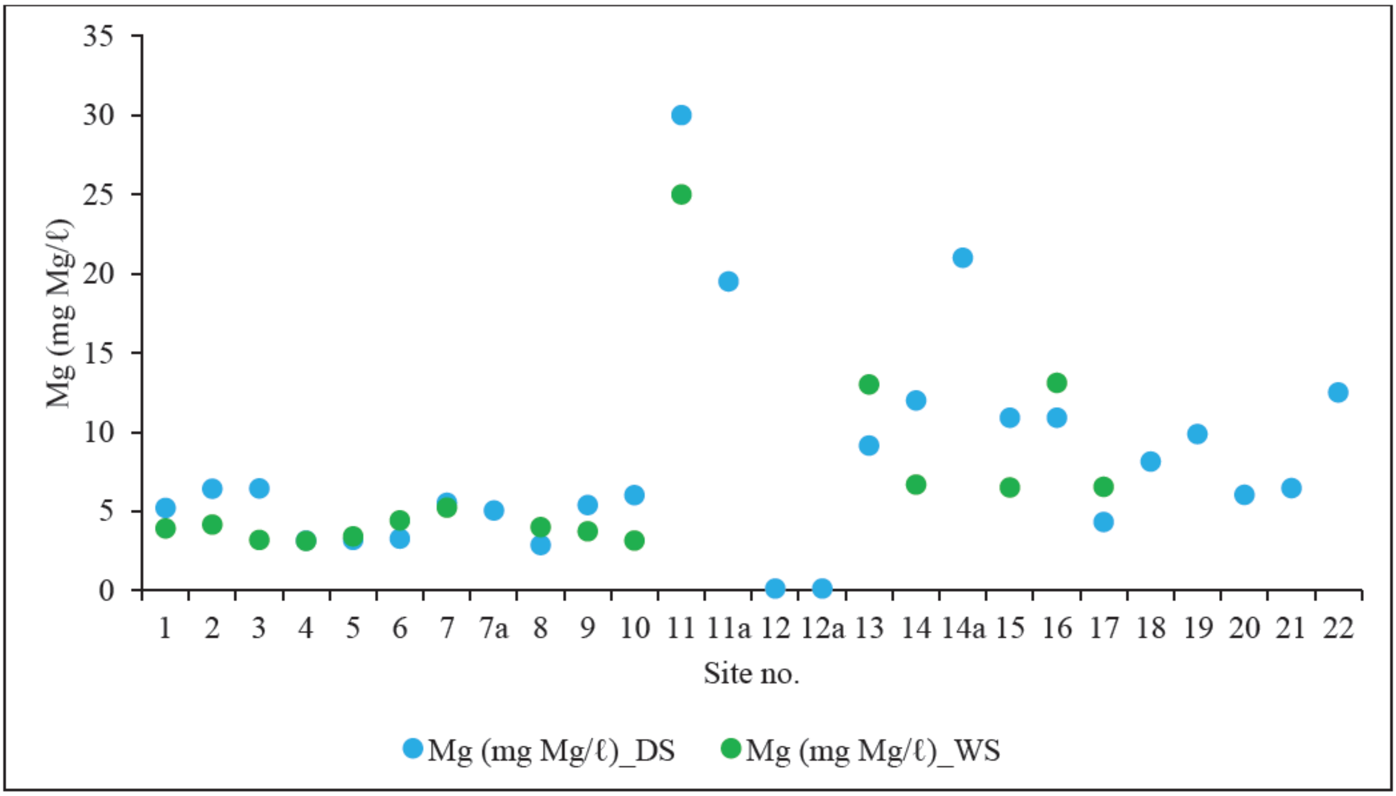


Figure 4.6 Measured Magnesium (Mg) across the sample sites in the Upper Awash basin during the wet and dry season

Figure 4.7 shows the variation of potassium across the sample sites. High potassium measurements were observed at S14a downstream of Aba Samuel reservoir. Upstream of the Dire reservoir in the high altitudes, low potassium measurements were observed for both seasons and started rising from upstream of the Legedadi reservoir, then decreased again at S12 and S12a at the confluence of the two rivers. In S13 (Akaki River), S14 (Aba Samuel), S15 (upstream of Koka), and S17 (downstream of Koka), measurements of Potassium were low during the wet season as compared to the dry season, which may show the dilution of the metal concentration due to high flows.

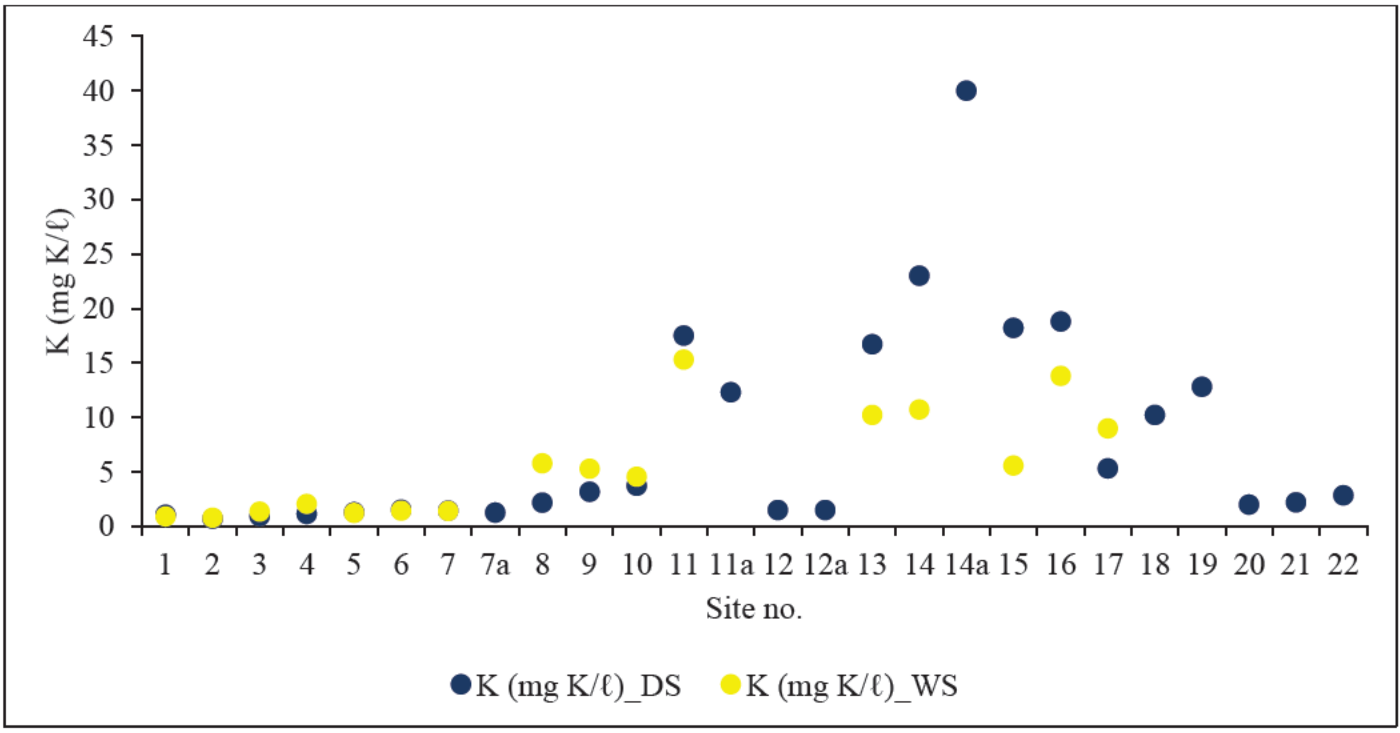


Figure 4.7 Measured Potassium (K) across the sample sites in the Upper Awash basin during the wet and dry season

Figure 4.8 shows the measurements of cadmium across the sites. The graph displays majority of the sites having low concentrations of cadmium below the ETH EPA standard except for sites 11 which displays concentrations of cadmium above the ETH EPA.

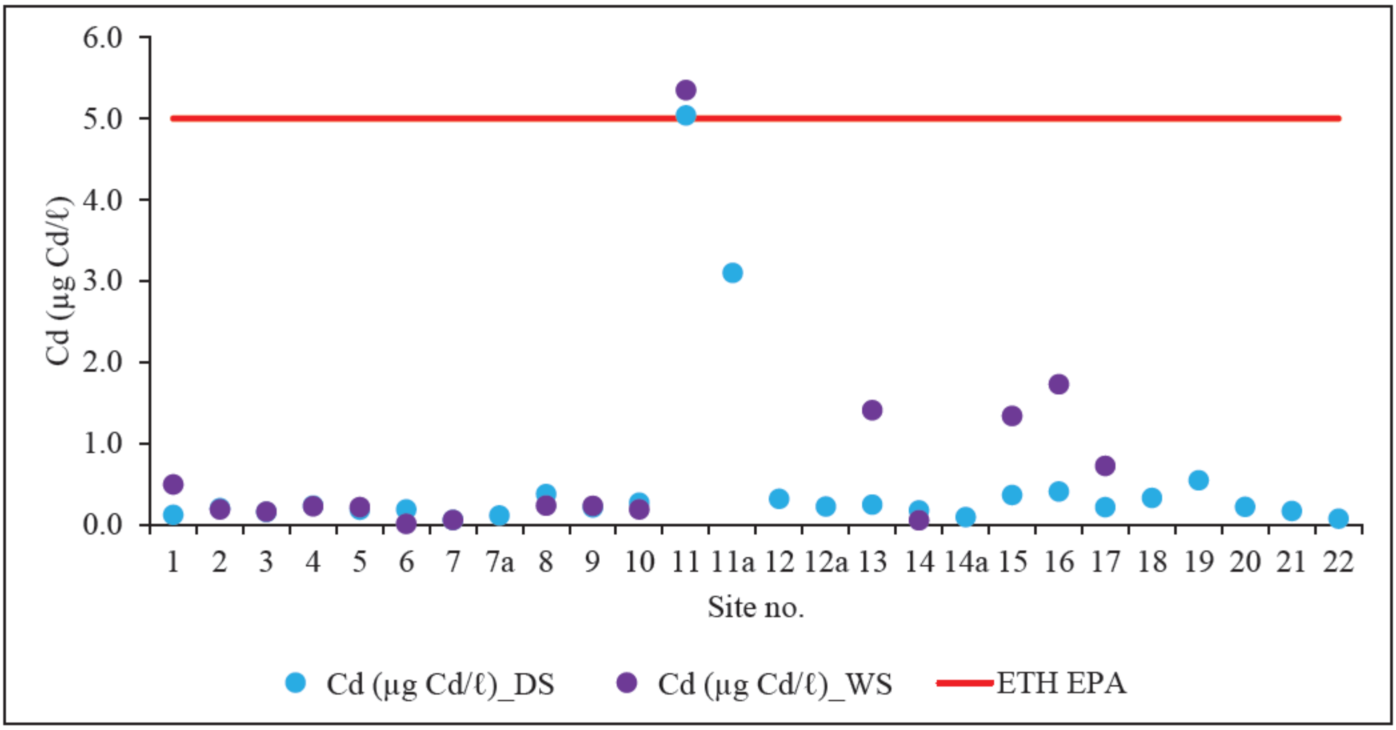


Figure 4.8 Measurements of Cadmium (Cd) across the sample sites in the Upper Awash basin during the wet and dry season

Figure 4.9 shows the measurements of chromium across the sample sites. The graph shows the majority of the sites having low concentrations of chromium during the wet and dry season which are below the ETH EPA standard. S11 measured very high concentrations of approximately 270 (µg/ℓ) which is above the ETH EPA standard. S13 and S16 measured concentrations above ETH EPA standards during the wet season, while S14a measured concentrations of 55 µg just above the ETH EPA standard during the dry season.

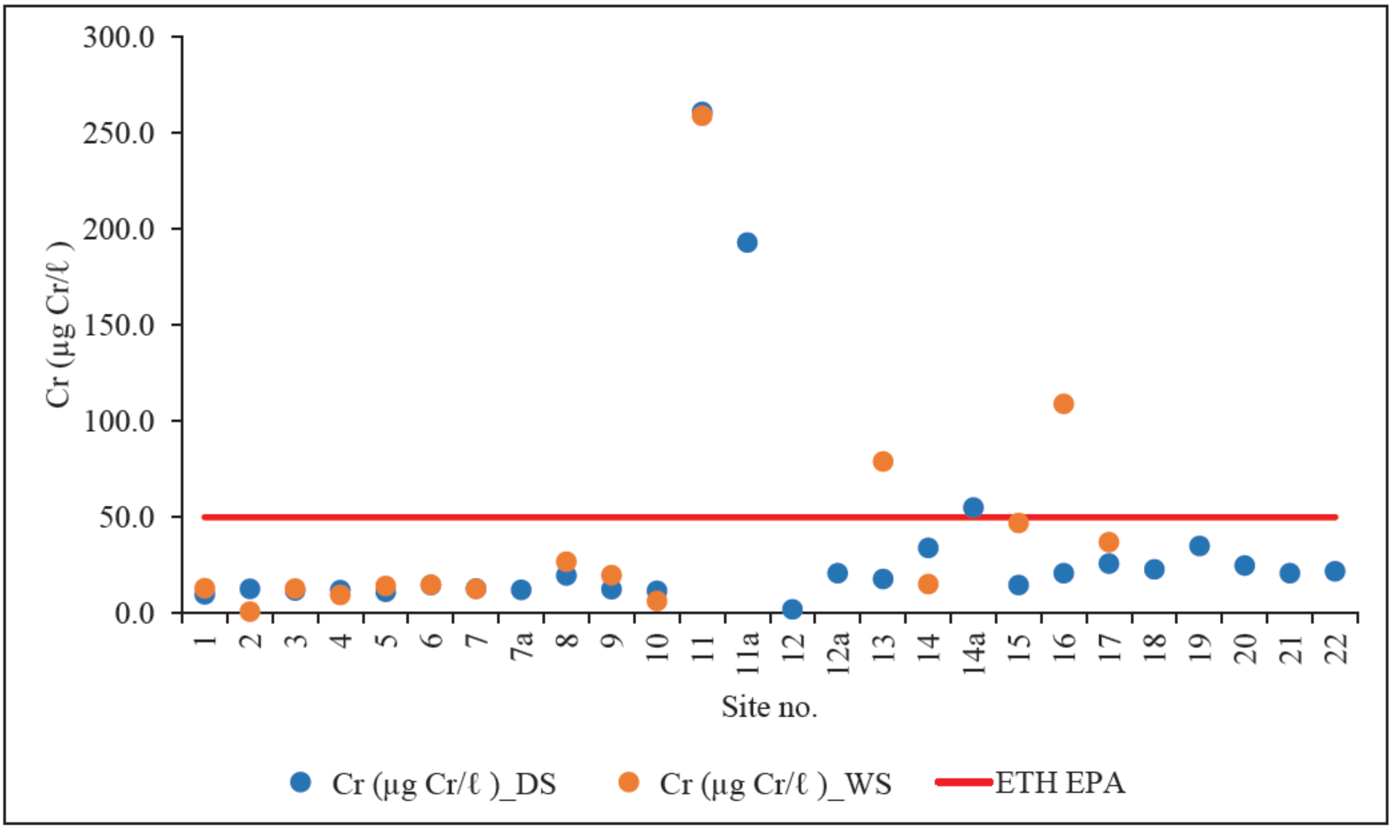


Figure 4.9 Measured Chromium (Cr) across the sample sites in the Upper Awash basin during the wet and dry season

Figure 4.10 shows the measured copper concentrations across the sample sites. S1, S9, SS11, 11a, S14, S14a, S17, and S18 measured high concentrations of copper above all ETH EPA standards during the dry season, while the majority of the sites showed low concentrations of copper during the dry season. S11 shows high levels of copper during the dry season as well.

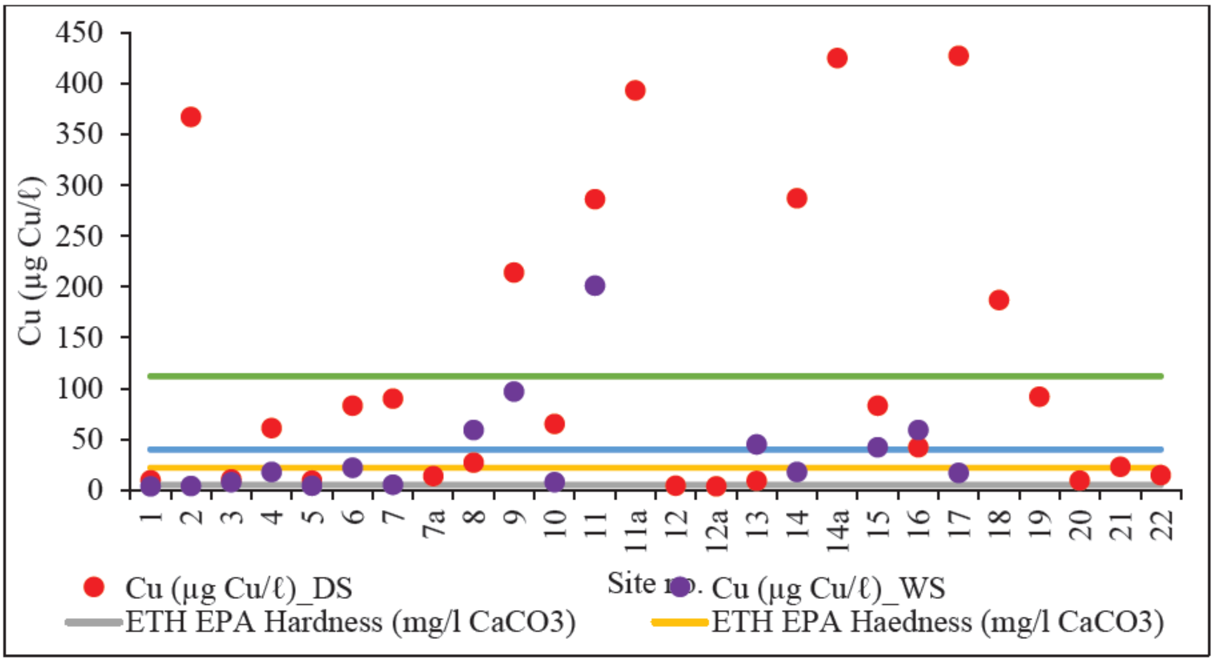


Figure 4.10 Measured copper (Cu) across the sample sites in the Upper Awash basin during the wet and dry season

Figure 4.11 shows the measured concentrations of mercury across the sample sites. All sites measured Hg concentrations above the ETH EPA standard.

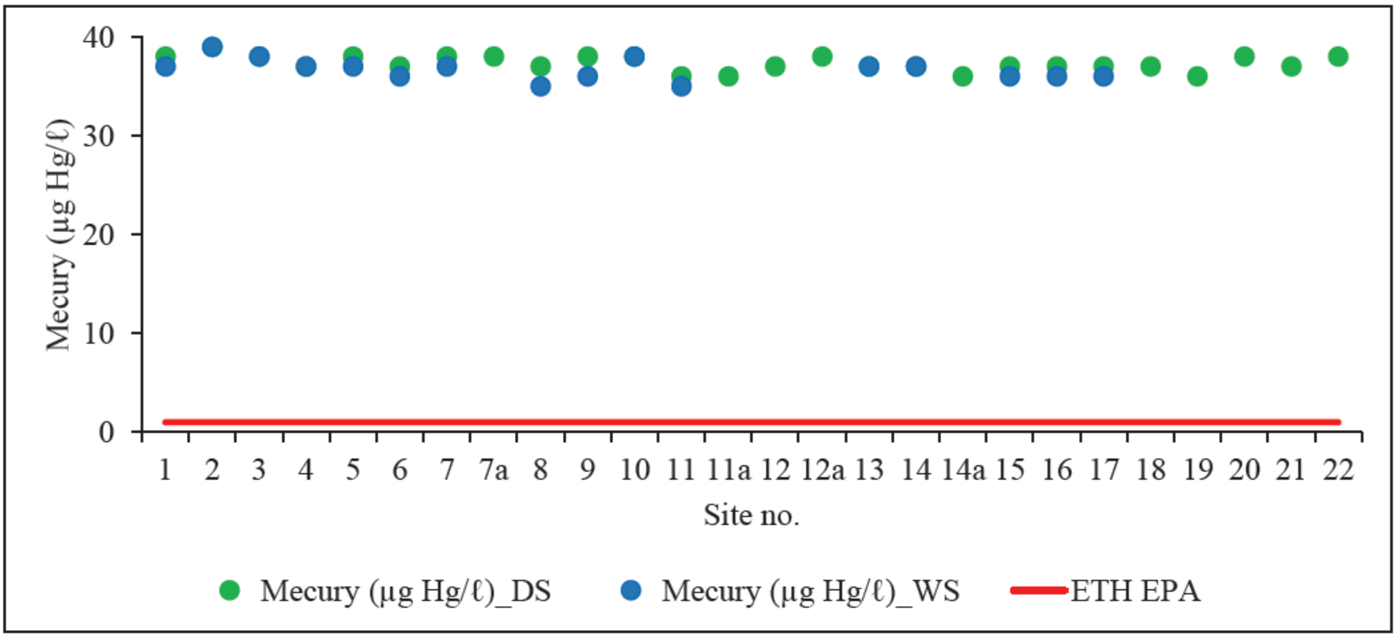


Figure 4.11 Measured Mercury (Hg) across the sample sites in the Upper Awash basin during the wet and dry season

Figure 4.12 shows the concentrations of iron across the sample sites. Majority of the sites show relatively low and slightly high concentration of iron against the ETH EPA standard. While site 11, 11a, 13, 15, 16 and 17 show high concentrations of iron during the wet season and site 11 shows extremely high concentrations of iron during the dry season.

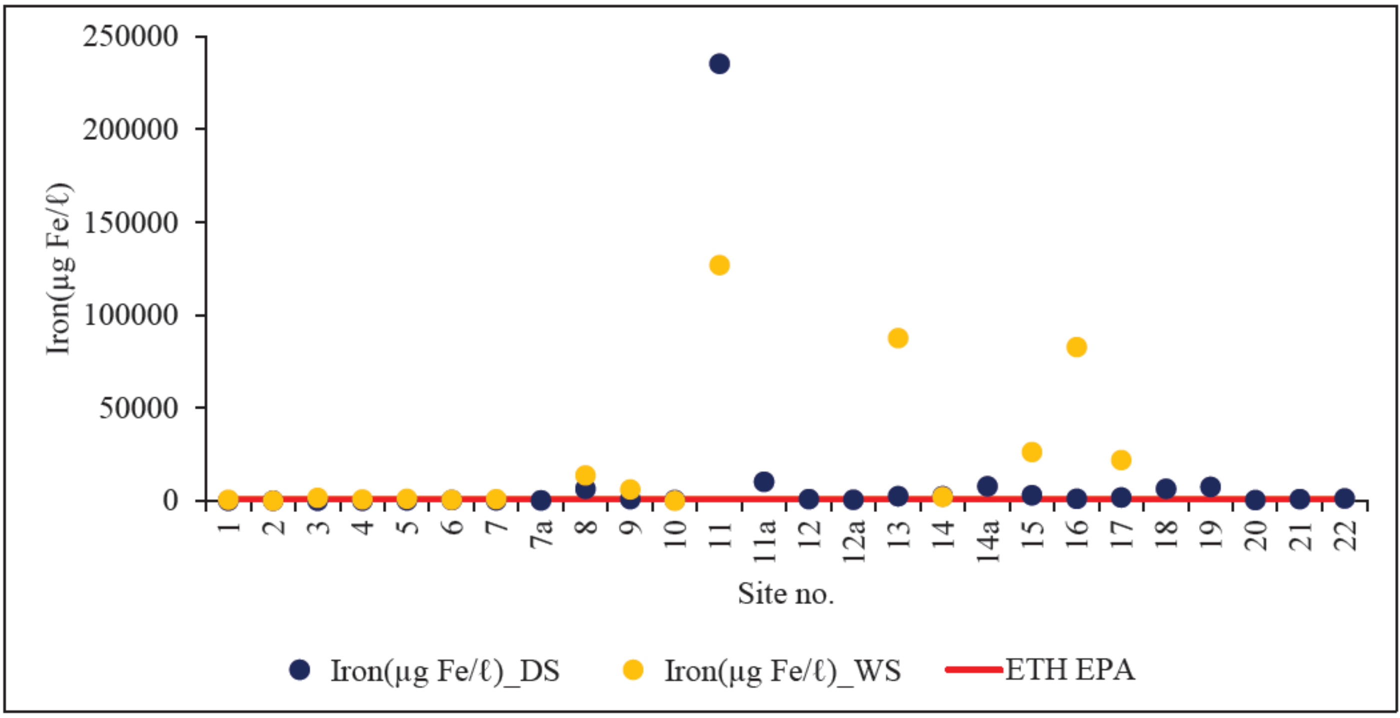


Figure 4.12 Measured Iron (Fe) across the sample sites in the Upper Awash basin during the wet and dry season

Figure 4.13 shows the measured concentrations of manganese across the sites. Sites above the Dire reservoir and below the reservoir in the highlands (site 1 to 7a) and sites in the west side on the city of Addis Ababa (site 20 to site 22) show low concentrations of manganese below the ETH EPA standards. Site 11 shows high concentrations of manganese for both seasons.

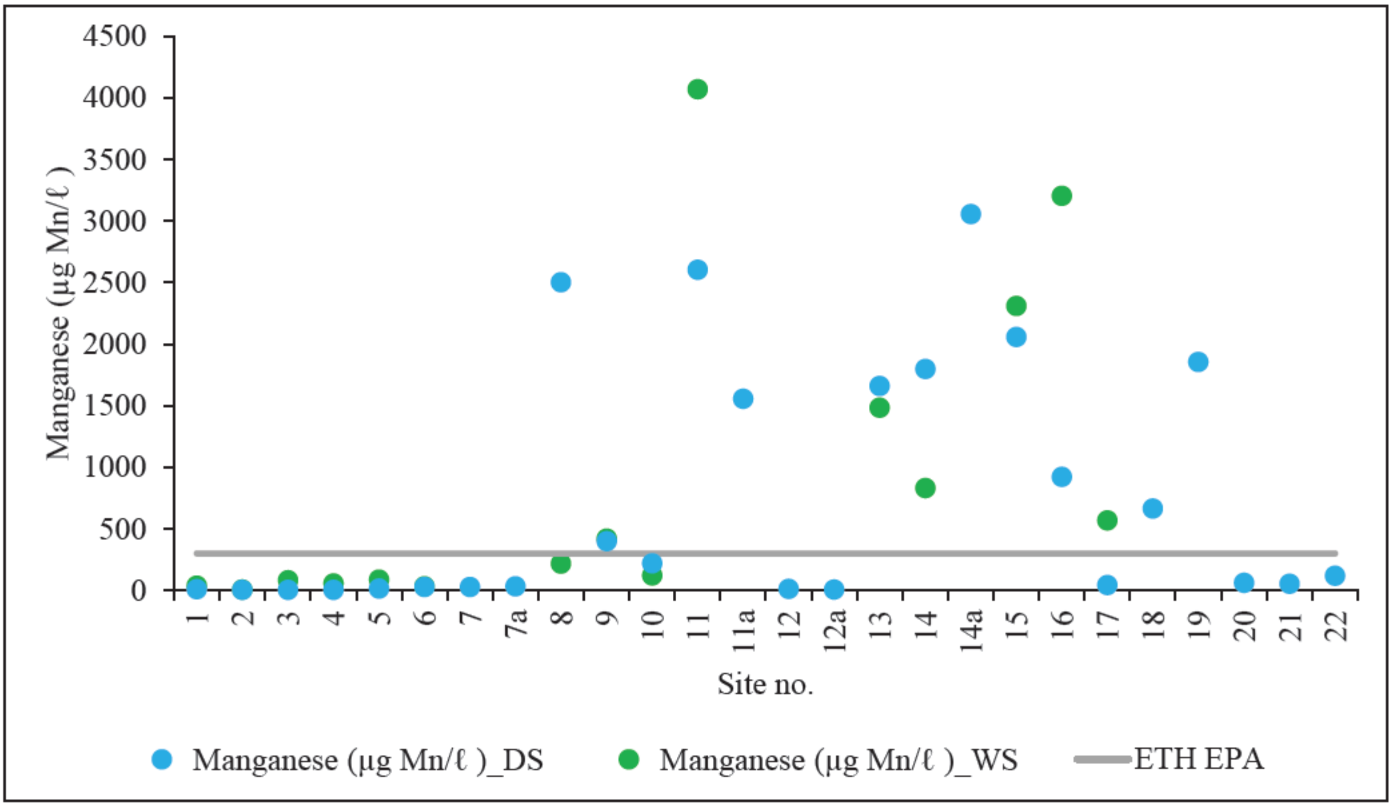


Figure 4.13 Measured Manganese (Mn) across the sample sites in the Upper Awash basin during the wet and dry season

Figure 4.14 shows the concentrations of lead across the sites. A majority of the sites show low concentrations of lead below the ETH EPA standard for both seasons. However, S11 shows high concentrations levels of lead above the ETH EPA standard for both seasons.

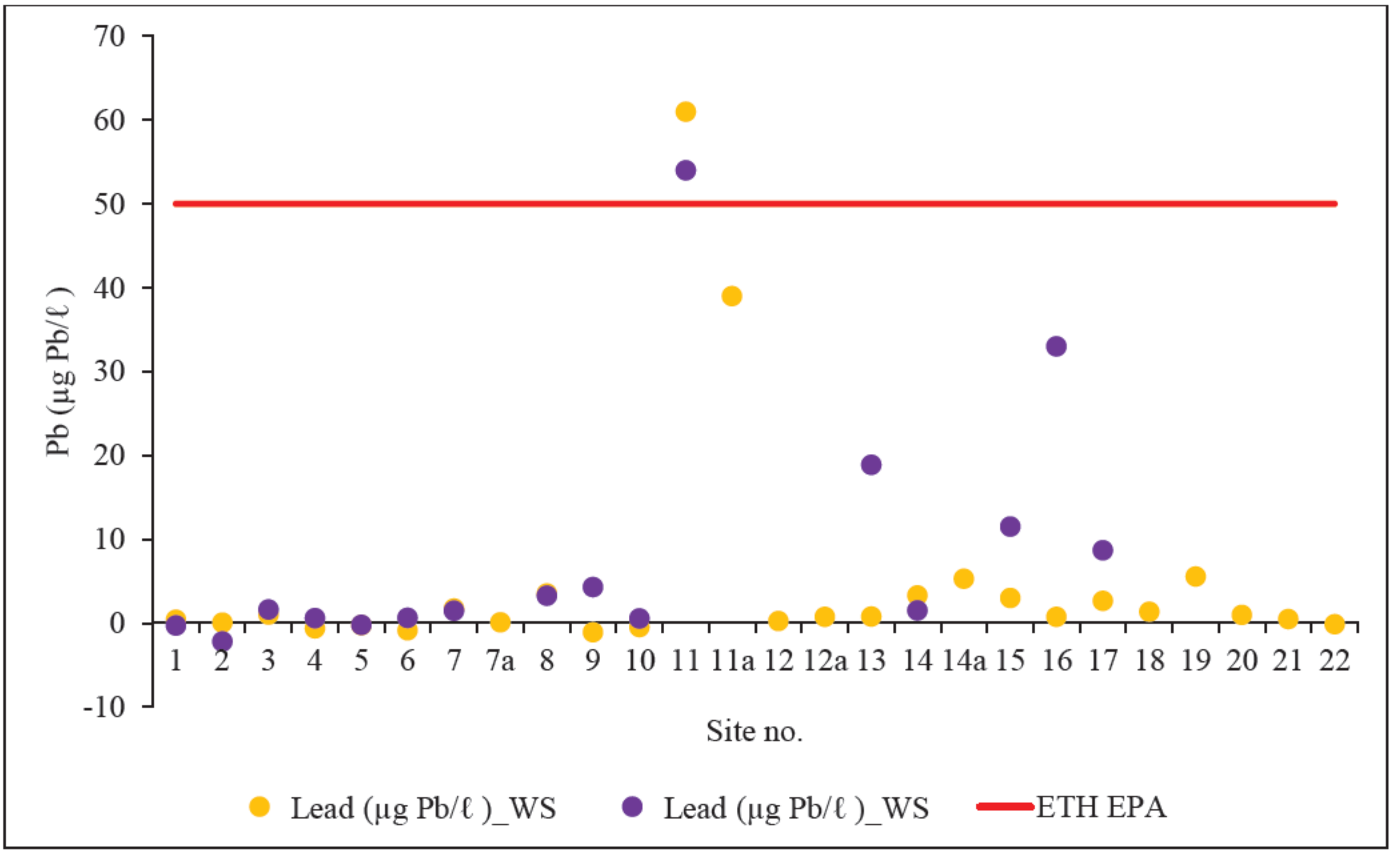


Figure 4.14 Measured Lead (Pb) across the sample sites in the Upper Awash basin during the wet and dry season

Figure 4.15 shows the concentration of zinc across the sites. From site 1 to site 8 and site 12, 20, 21, and 22 show low concentrations of zinc below all ETH EPA standards except for site 2 during the dry season. Site 11 and 11a show concentration levels higher than all ETH EPA.

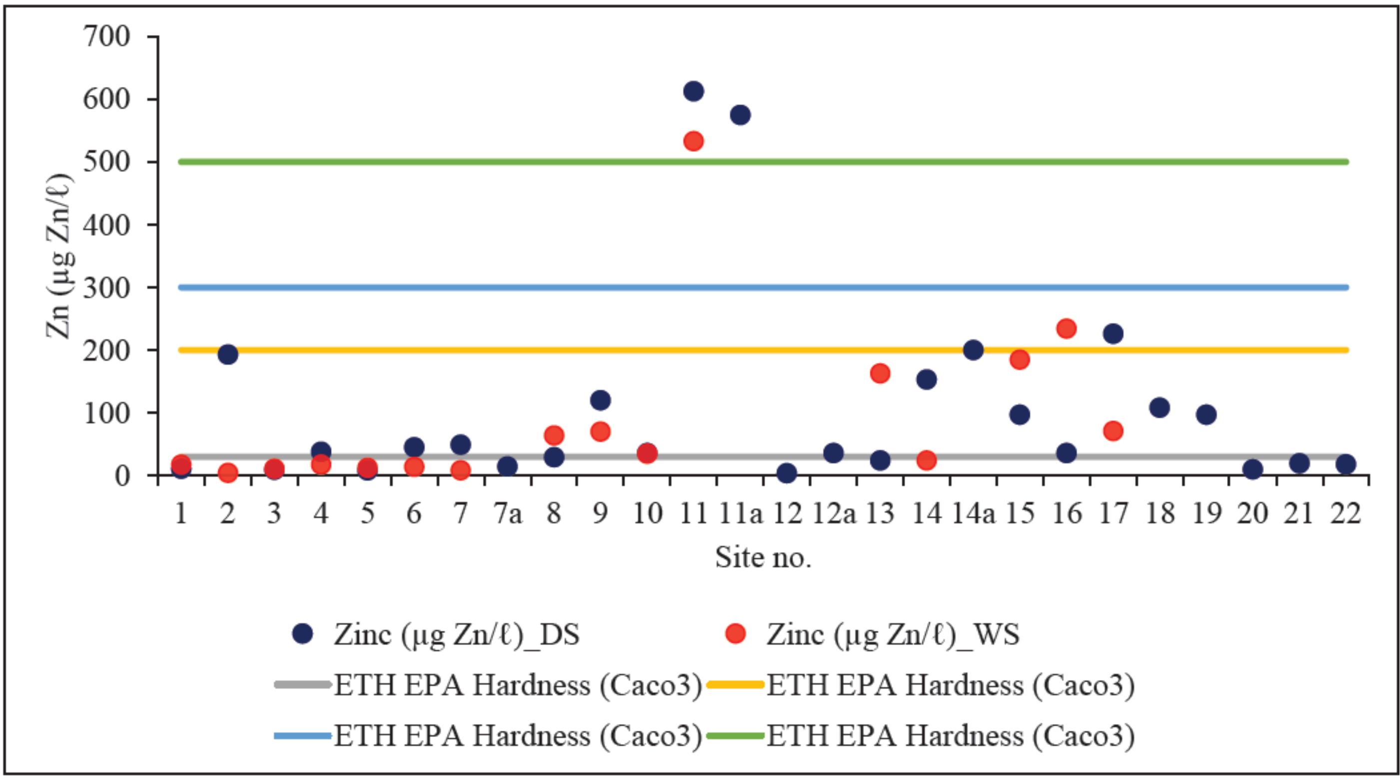


Figure 4.15 Measured Zinc Zn) across the sample sites in the Upper Awash basin during the wet and dry season

4.3 SASS5

This section displays the variation in the season of the SASS5 ASPT and the ecological conditions of the tested sample sites in the Upper Awash basin during the wet and dry seasons. SASS5 ASPT is the chosen measure as it is a more reliable measure of the health of good-quality rivers. The SASS5 ASPT results were interpreted using the approach demonstrated in Figure 4.16. From Figure 4.17 to Figure 4.19, the pie charts show the proportion of sensitive to tolerant taxa (Taxa found is shown in Table 8.2 and Figure 8.3 in the Appendix B).

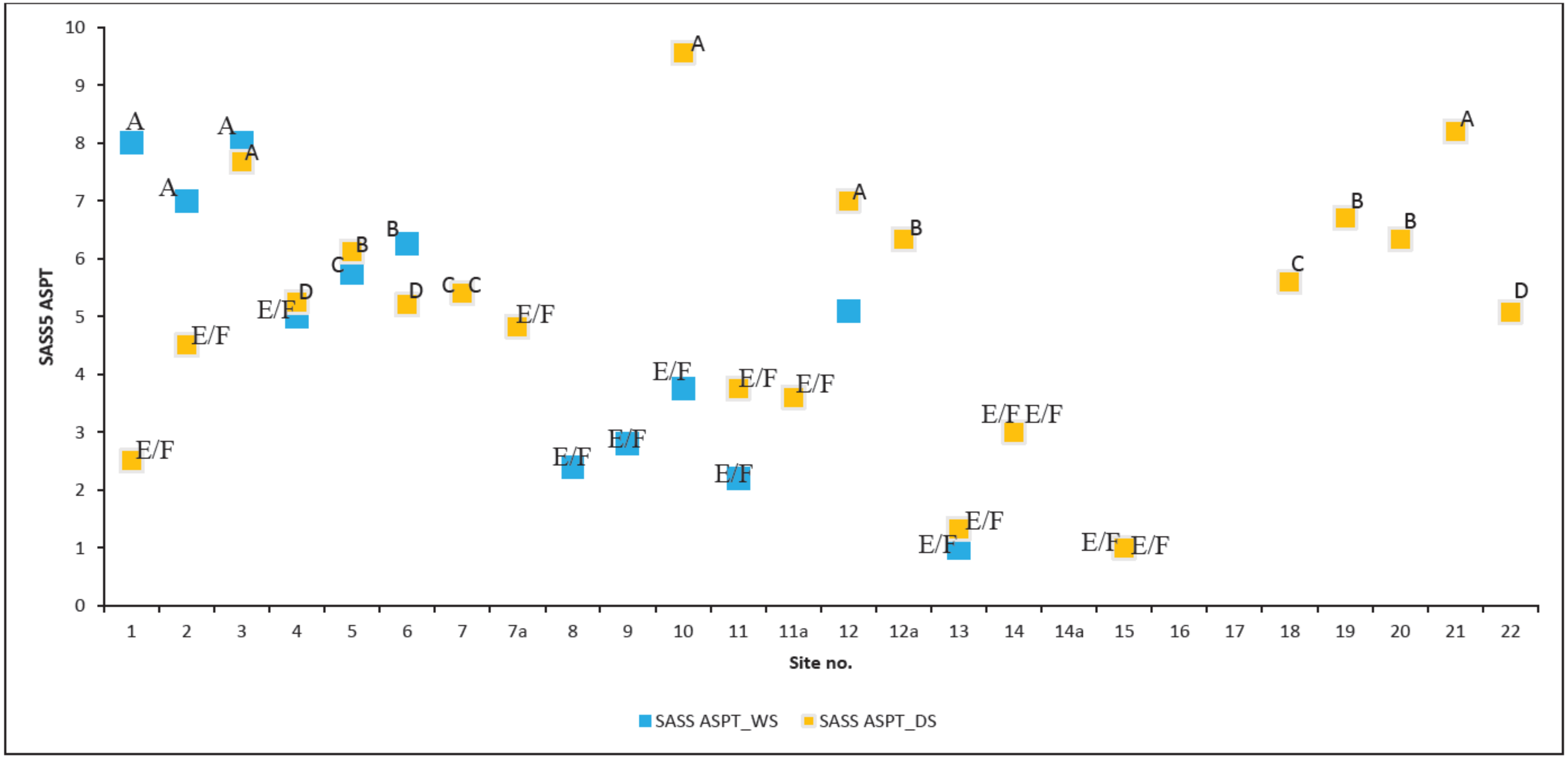


Figure 4.16 Seasonal variation of SASS ASPT for the different sites along the Upper Awash basin. The ecological categories: A (natural water quality), B (good water quality), C (fair water quality), D (poor water quality) and E/F (very poor water quality) are indicated on the bars (Dallas, 2007). (Refer to Table 8.3 and 8.4, Appendix C, SASS5 guidelines and interpretations).

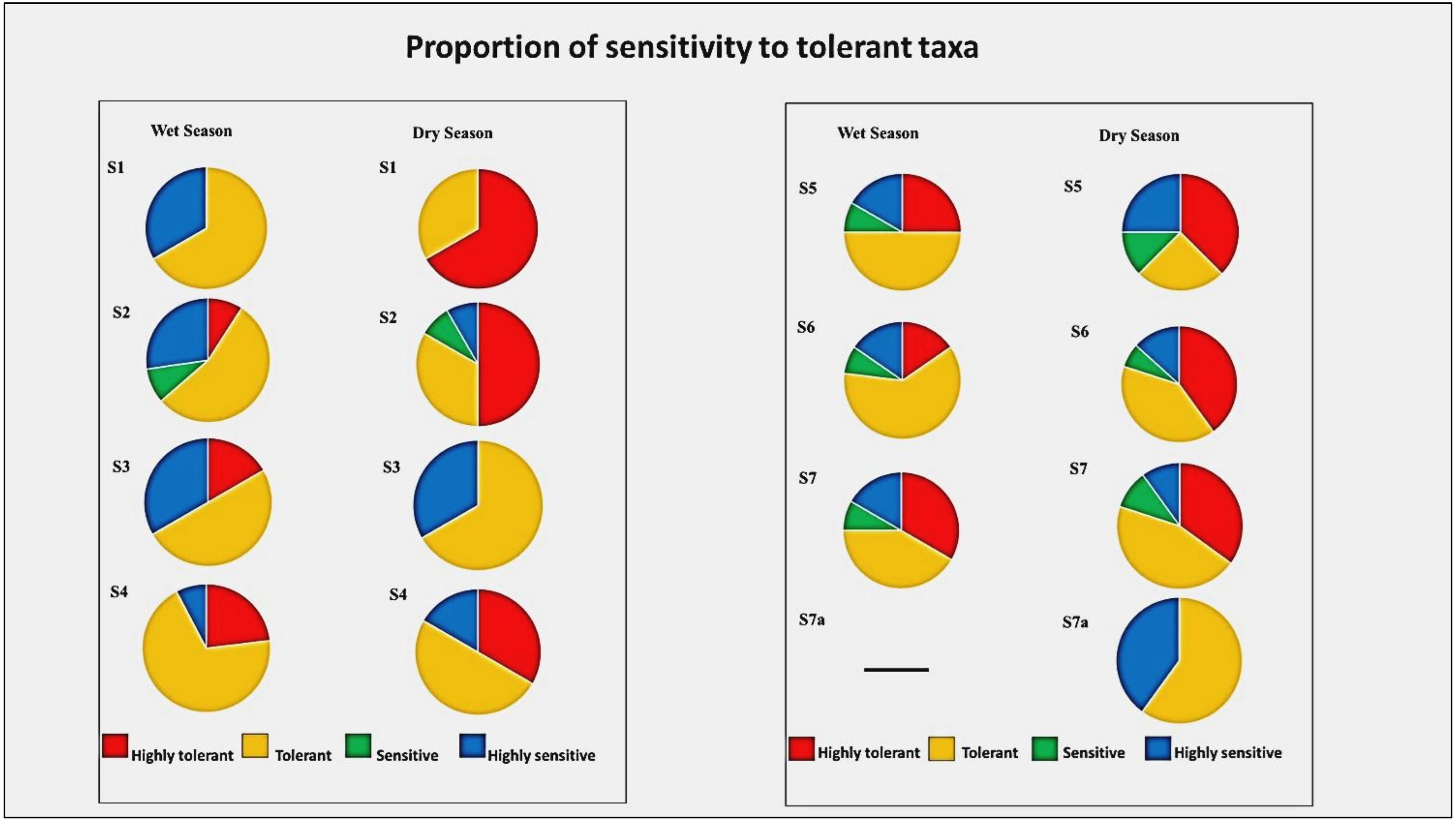


Figure 4.17 Proportion of sensitivity to tolerant taxa (Proportions based on sensitivity weightings as follows: Highly tolerant (1-3), Tolerant (4-7), Sensitive (8-11) and Highly sensitive (12-15)

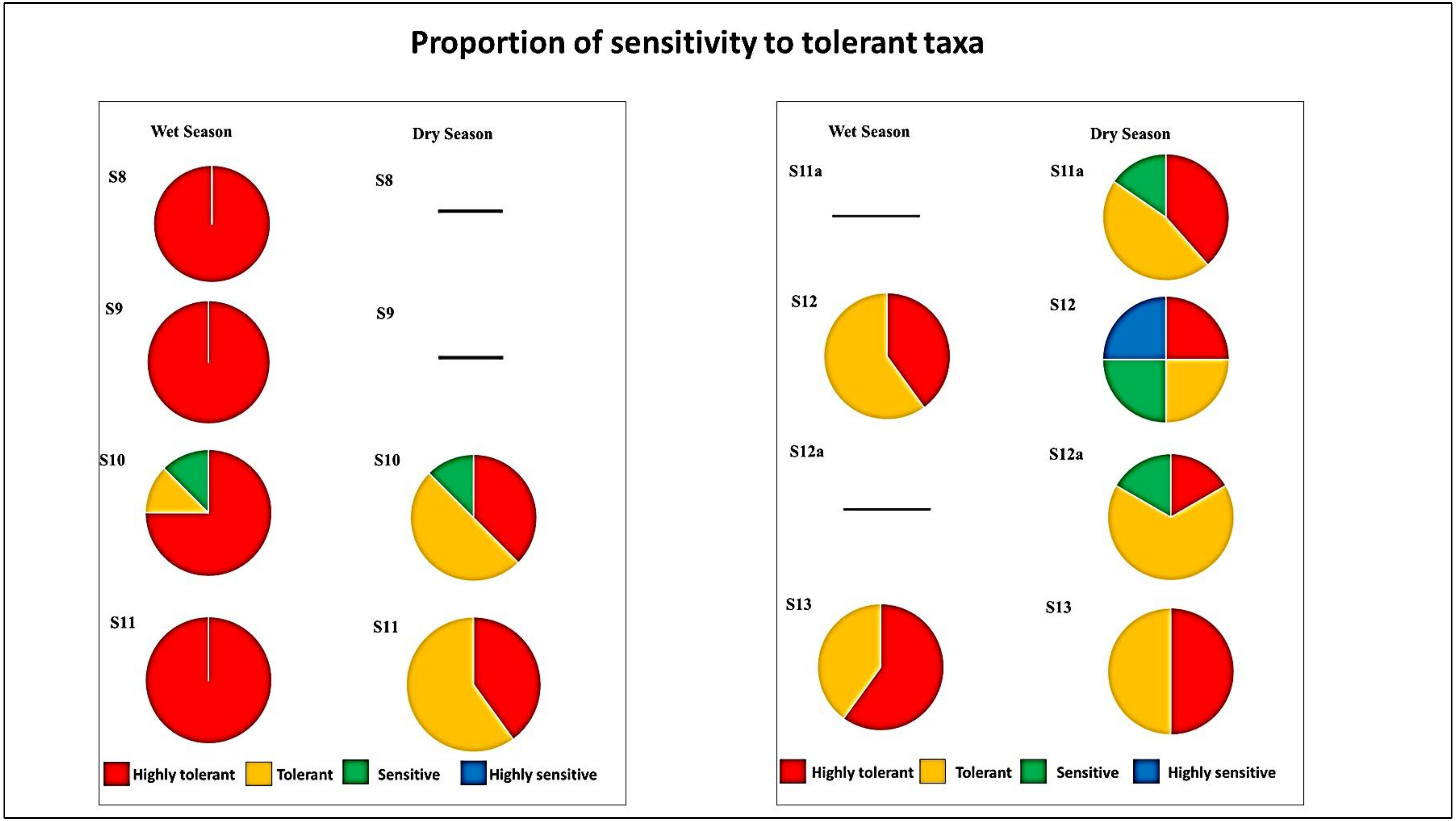


Figure 4.18 Proportion of sensitivity to tolerant taxa (Proportions based on sensitivity weightings as follows: Highly tolerant (1-3), Tolerant (4-7), Sensitive (8-11) and Highly sensitive (12-15)

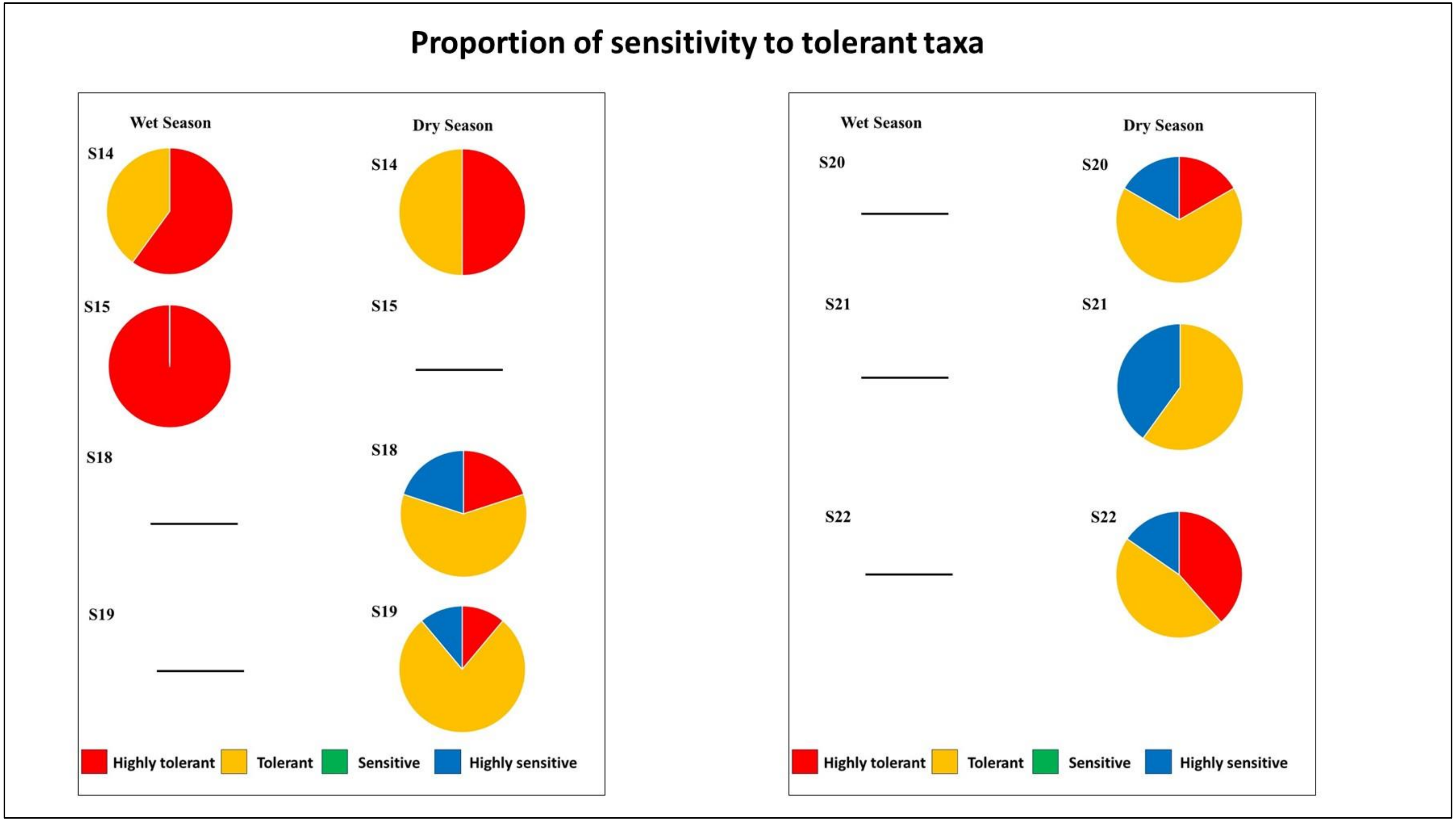


Figure 4.19 Figure 3 Proportion of sensitivity to tolerant taxa (Proportions based on sensitivity weightings as follows: Highly tolerant (1-3), Tolerant (4-7), Sensitive (8-11) and Highly sensitive (12-15)

4.4 MiniSASS

This section displays the ecological conditions of the tested sample sites in the Upper Awash basin. Figure 4.20 displays the miniSASS results during the wet season, and Figure 4.21 displays the results for the dry season. When looking at the overall ecological condition of the study area during the wet season the results reflect poor conditions of the sites.

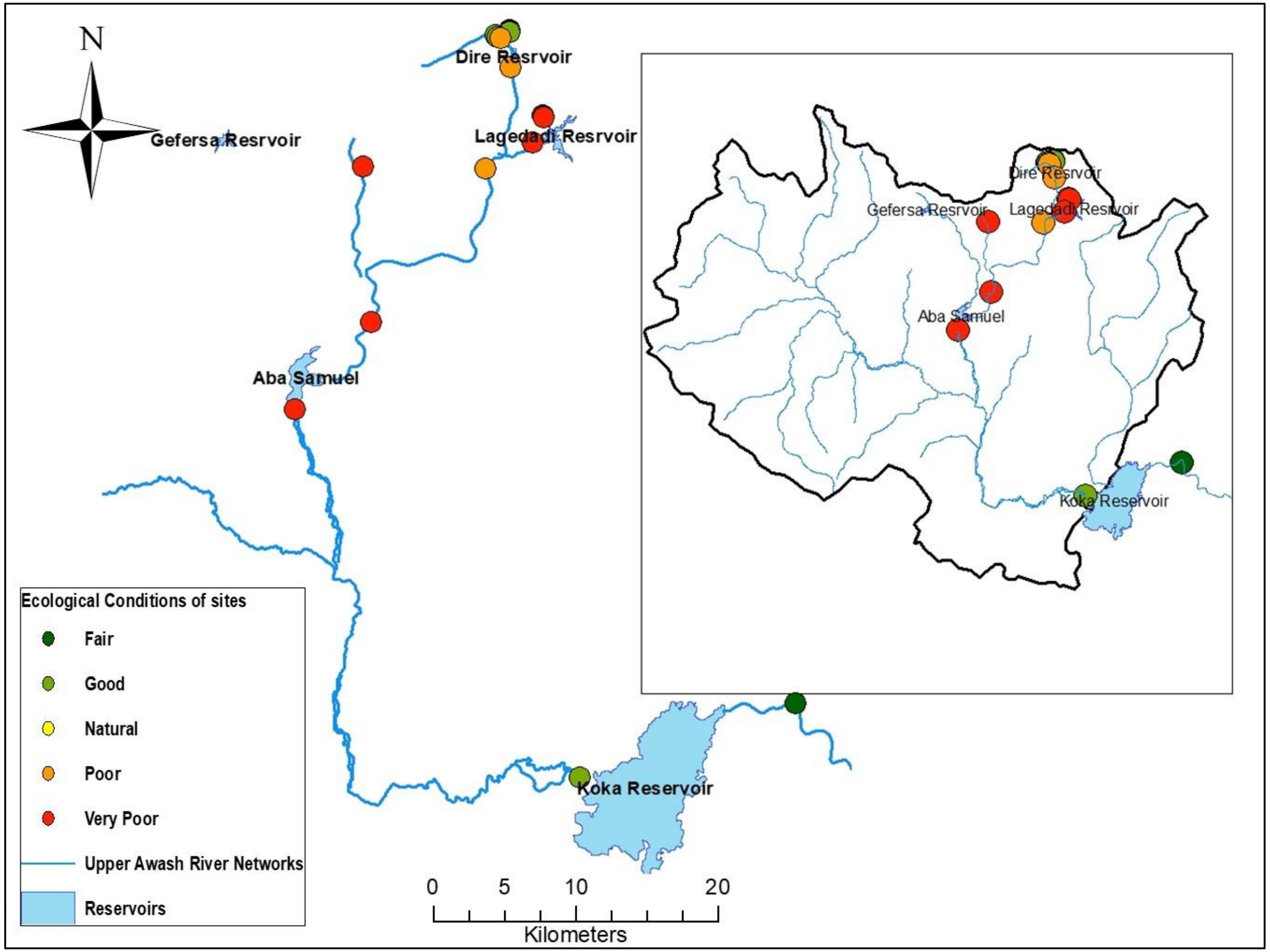


Figure 4.20 Map of the Upper Awash River basin showing the miniSASS ecological conditions of the different sites from upstream, city centre of Addis Ababa and downstream during the wet season (Refer to Table 8.5 in Appendix C)

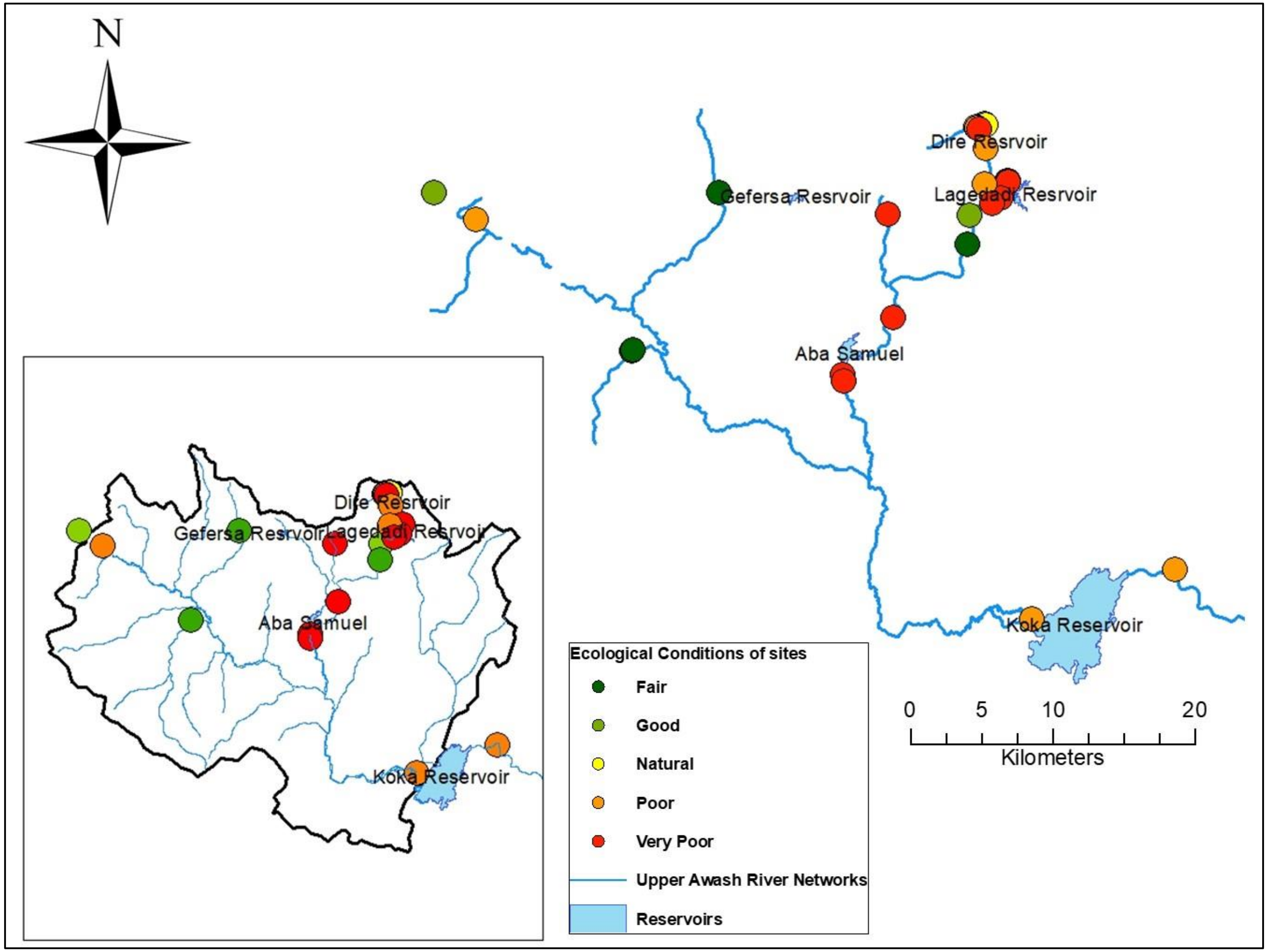


Figure 4.21 Map of the Upper Awash River basin showing the miniSASS ecological conditions of the different sites from upstream, city centre of Addis Ababa and downstream during the dry season (Refer to Table 8.5 in Appendix C)

4.5 Correlation Analysis

Table 4.1 shows the correlation matrix between the Biomonitoring metrics, physicochemical parameters, and heavy metals and other metals during the wet season. MiniSASS Ave scores of the different sites displayed strong positive correlations with SASS5 ASPT and moderate positive correlations with temperature, pH, and clarity. Strong negative correlations were observed between miniSASS Ave and DO and K, while moderate negative correlations were observed between miniSASS Ave and EC, Mg, Cd, Cr, Cu, Fe, Mn, Pb, and Zn. SASS5 ASPT displayed strong negative correlations with K, Cu, Fe, Pb, and Zn.

Table 4.1 Correlation matrix table showing the correlation between SASS5 ASPT, miniSASS Ave, physicochemical parameters and heavy metals during the wet season

	Temp (°C)	pH	DO (mg/L)	EC (µs/cm)	Clarity (cm)	miniSASS Ave	SASS5 ASPT	(mg Mg/ℓ)	(mg K/ℓ)	(µg Cd/ℓ)	(µg Cr/ℓ)	(µg Cu/ℓ)	(µg Fe/ℓ)	(µg Hg/ℓ)	(µg Mn/ℓ)	(µg Pb/ℓ)	(µg Zn/ℓ)
Temp (°C)	1.00																
pH	-0.46	1.00															
DO (mg/L)	-0.48	0.14	1.00														
EC (µs/cm)	0.01	-0.04	0.61	1.00													
Clarity (cm)	0.33	-0.13	0.08	-0.02	1.00												
miniSASS Ave	0.46	0.03	-0.54	-0.42	0.38	1.00											
SASS5 ASPT	0.66	-0.12	-0.50	-0.37	0.49	0.90	1.00										
Mg (mg Mg/ℓ)	-0.04	0.08	0.22	0.37	-0.41	-0.31	-0.48	1.00									
K (mg K/ℓ)	-0.09	-0.13	0.36	0.50	-0.53	-0.61	-0.76	0.82	1.00								
Cd (µg Cd/ℓ)	-0.09	0.11	0.13	0.27	-0.49	-0.22	-0.42	0.95	0.72	1.00							
Cr (µg Cr/ℓ)	-0.05	0.01	0.17	0.34	-0.46	-0.27	-0.45	0.98	0.78	0.99	1.00						
Cu (µg Cu/ℓ)	-0.22	-0.12	0.14	0.14	-0.46	-0.38	-0.54	0.81	0.68	0.86	0.88	1.00					
(µg Fe/ℓ)	-0.07	0.08	0.14	0.40	-0.54	-0.37	-0.57	0.96	0.83	0.90	0.93	0.77	1.00				
(µg Hg/ℓ)	0.02	0.30	-0.21	-0.45	0.23	0.52	0.56	-0.47	-0.53	-0.49	-0.54	-0.66	-0.47	1.00			
(µg Mn/ℓ)	-0.16	0.14	0.31	0.54	-0.52	-0.43	-0.63	0.90	0.84	0.89	0.90	0.76	0.90	-0.52	1.00		
(µg Pb/ℓ)	-0.04	0.00	0.20	0.42	-0.52	-0.33	-0.51	0.97	0.83	0.95	0.98	0.84	0.96	-0.54	0.95	1.00	
(µg Zn/ℓ)	-0.17	0.08	0.21	0.36	-0.55	-0.36	-0.55	0.95	0.78	0.98	0.98	0.89	0.92	-0.57	0.94	0.97	1.00

Table 4.2 shows the correlation between the Biomonitoring metrics, physicochemical parameters, and heavy metals and other metals during the dry season. MiniSASS Ave shows positive correlations with pH, DO, clarity, and Hg, while it shows moderate positive correlations with SASS5 ASPT. SASS5 ASPT shows strong positive correlations with DO. Low negative correlations were observed between SASS5 ASPT and temperature, Mg, Cd, Cr, Cu, Fe, Pb, and Zn, with moderate negative correlations with Mn and EC.

Table 4.2 Correlation matrix table showing the correlation between SASS5 ASPT, miniSASS ave, physicochemical parameters and heavy metals during the dry season

	Temp (°C)	pH	DO (mg/L)	EC (µs/cm)	Clarity (cm)	miniSASS Ave	SASS5 ASPT	(mg Mg/ℓ)	(mg K/ℓ)	(µg Cd/ℓ)	(µg Cr/ℓ)	(µg Cu/ℓ)	(µg Fe/ℓ)	(µg Hg/ℓ)	(µg Mn/ℓ)	(µg Pb/ℓ)	(µg Zn/ℓ)
Temp (°C)	1.0																
pH	0.5	1.0															
DO (mg/L)	0.5	0.5	1.0														
EC (µs/cm)	0.2	-0.3	-0.1	1.0													
Clarity (cm)	0.1	0.4	0.1	-0.5	1.0												
miniSASS Ave	-0.4	0.1	0.1	-0.5	0.2	1.0											
SASS5 ASPT	0.3	0.3	0.7	-0.5	0.3	0.5	1.0										
(mg Mg/ℓ)	-0.1	-0.4	-0.1	0.3	-0.4	-0.4	-0.4	1.0									
(mg K/ℓ)	0.2	-0.3	0.0	0.8	-0.5	-0.5	-0.6	0.7	1.0								
(µg Cd/ℓ)	-0.2	-0.2	0.0	-0.2	-0.3	-0.2	-0.2	0.7	0.2	1.0							
(µg Cr/ℓ)	-0.2	-0.3	-0.1	-0.1	-0.4	-0.3	-0.2	0.8	0.4	1.0	1.0						
(µg Cu/ℓ)	0.0	-0.4	-0.2	0.2	-0.2	-0.3	-0.3	0.5	0.5	0.4	0.5	1.0					
(µg Fe/ℓ)	-0.2	-0.1	0.0	-0.2	-0.2	-0.2	-0.2	0.7	0.2	0.9	0.8	0.2	1.0				
(µg Hg/ℓ)	-0.1	0.1	0.1	-0.3	0.6	0.3	0.2	-0.6	-0.7	-0.5	-0.6	-0.4	-0.4	1.0			
(µg Mn/ℓ)	0.1	-0.3	-0.1	0.6	-0.6	-0.6	-0.5	0.7	0.8	0.5	0.5	0.4	0.4	-0.7	1.0		
(µg Pb/ℓ)	-0.3	-0.2	-0.1	-0.2	-0.4	-0.3	-0.2	0.8	0.3	1.0	1.0	0.4	0.9	-0.5	0.5	1.0	
(µg Zn/ℓ)	-0.2	-0.3	-0.1	0.0	-0.3	-0.3	-0.3	0.8	0.4	0.9	0.9	0.8	0.7	-0.5	0.5	0.9	1.0

4.5.1 Multivariate analysis

Figure 4.22 displays the principal component analysis (PCA) biplot, which shows the correlations between the physicochemical parameters, Biomonitoring metrics, and heavy metals for the different sites. The components used have eigenvalues greater than 1 (refer to appendix). Therefore, the components explain 90.4% of the variation in the data. In the results, the first principal component has large positive associations with heavy metals (except Hg), DO, and EC.

The second component has large positive associations with SASS5 ASPT, miniSASS Ave, clarity, temperature, and Hg; therefore, this component measures the aquatic ecosystem's health. The parameters also show that they have a positive correlation with each other. PCA also shows the weak influence of clarity on the second component.

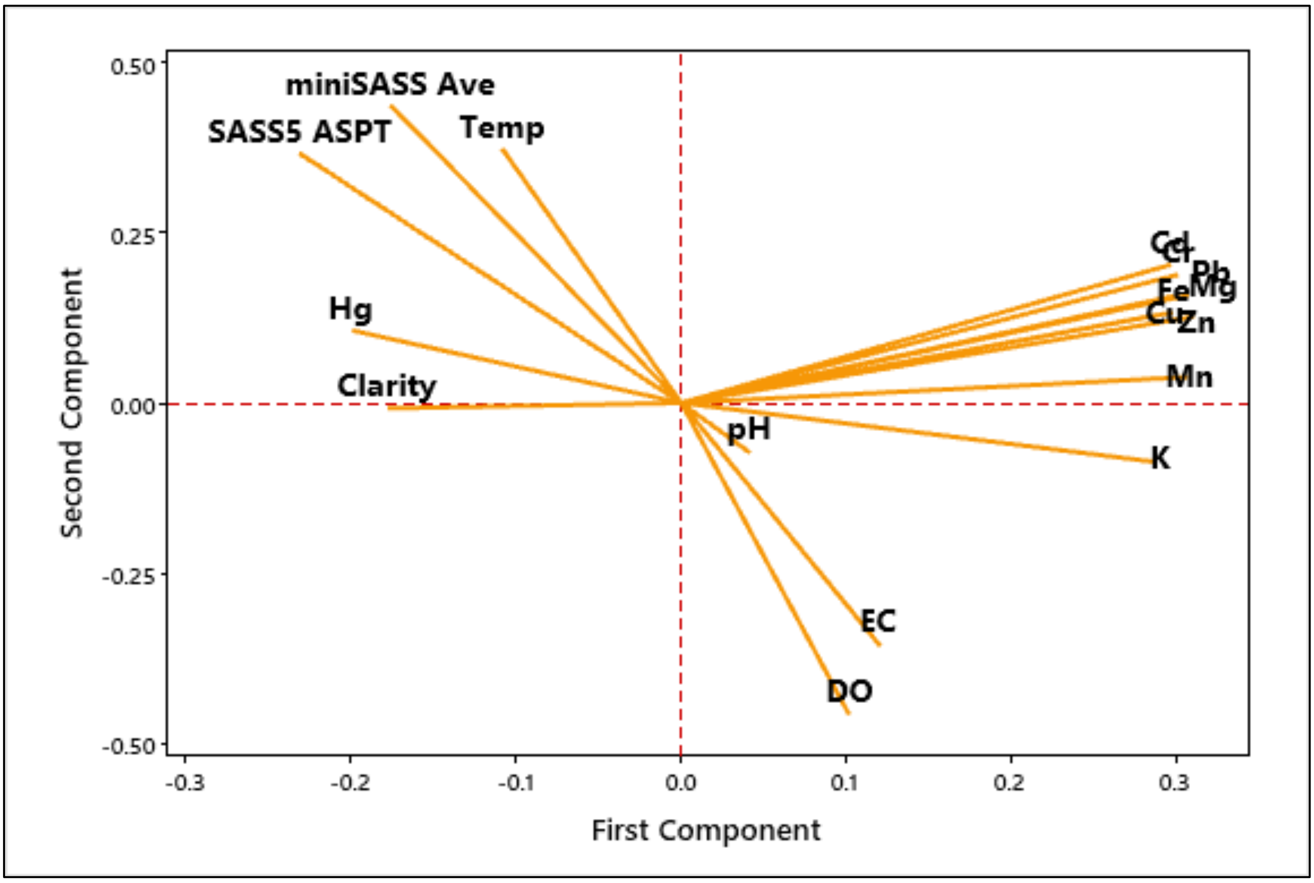


Figure 4.22 PCA biplot showing the correlation between physicochemical parameters, Biomonitoring metrics and heavy metals in the different sites during the wet season

Figure 4.23 displays the PCA score plot that shows the clustering of sample sites based on their similarity. Site 8, 9,11, 13,14 and 15 loaded positively to the first component with heavy metals, DO, pH and EC (Figure 15). The first component was influenced by sites located in and around the city with water treatment and hydropower plants and particularly S11 located downstream of a water treatment plant. While Sites 1, 2, 3, 4, 5, 6, 7, and 10 loaded negatively in the first component together with miniSASS, SASS5 ASPT, clarity and Hg influenced by sites located upstream of Dire reservoir located in a rural environment.

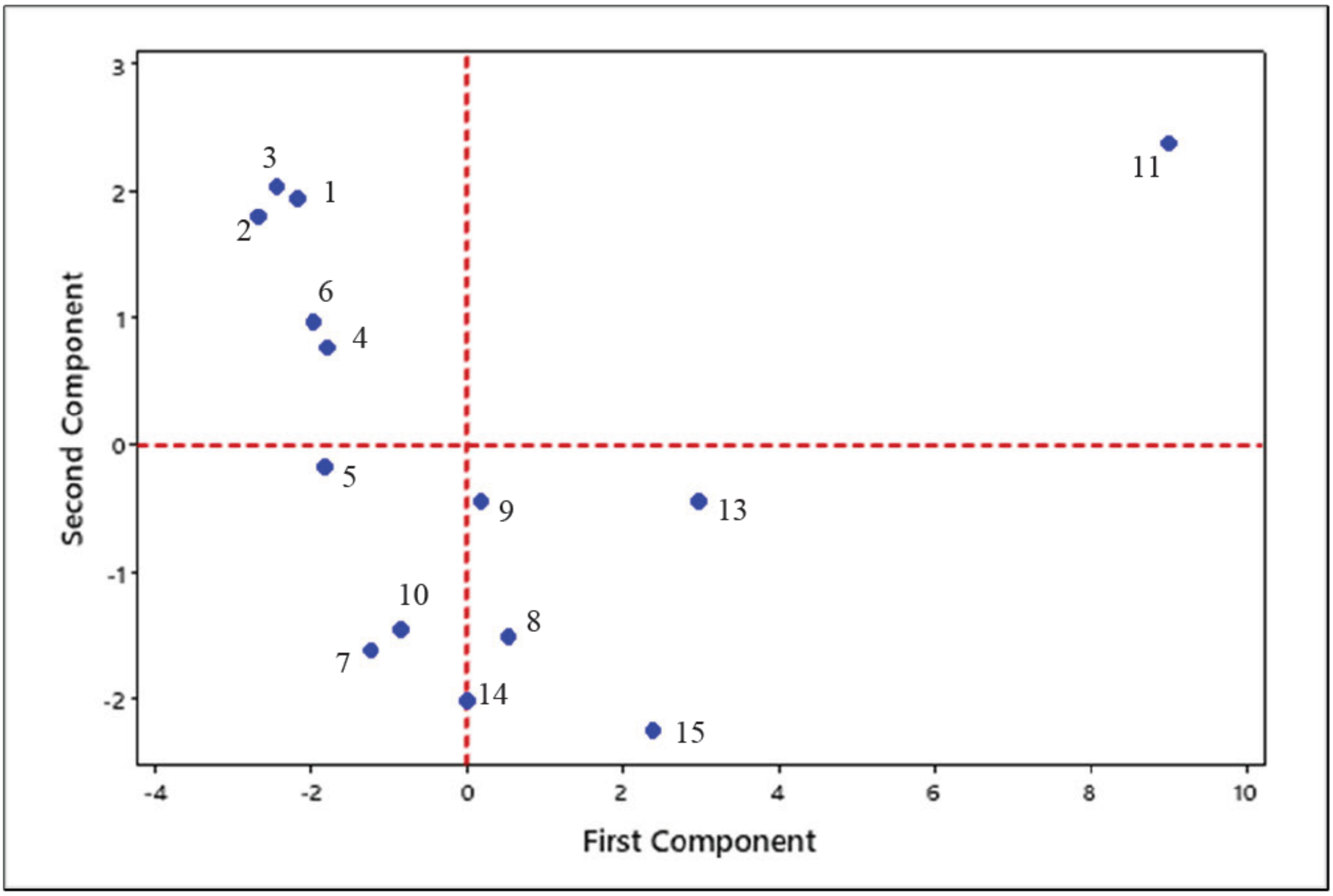


Figure 4.23 PCA score plot showing the clustering of sites based on similarities for the wet season

Figure 4.24 displays the dendrogram of physicochemical parameters, Biomonitoring metrics and heavy metals. The dendrogram was created using a final partition of two clusters, which occur at similarity level of 43. The first cluster (blue) is composed of five observations (temperature, clarity, miniSASS Ave, SASS5 ASPT and mercury) with a similarity level higher than 50% while miniSASS Ave and SASS5 ASPT shows a similarity level of about 94% with Hg, temperature and clarity. The second cluster is composed of twelve observations (pH,dissolved oxygen, electrical conductivity, Mg, Fe, Cd,Zn, Pb, Mn, Cu and K) with similarity level of 43%. While Mg, Fe, Cd, Zn, Cr, Pb, Mn, Cu, K represent a similarity level of about 87% higher than DO and EC.

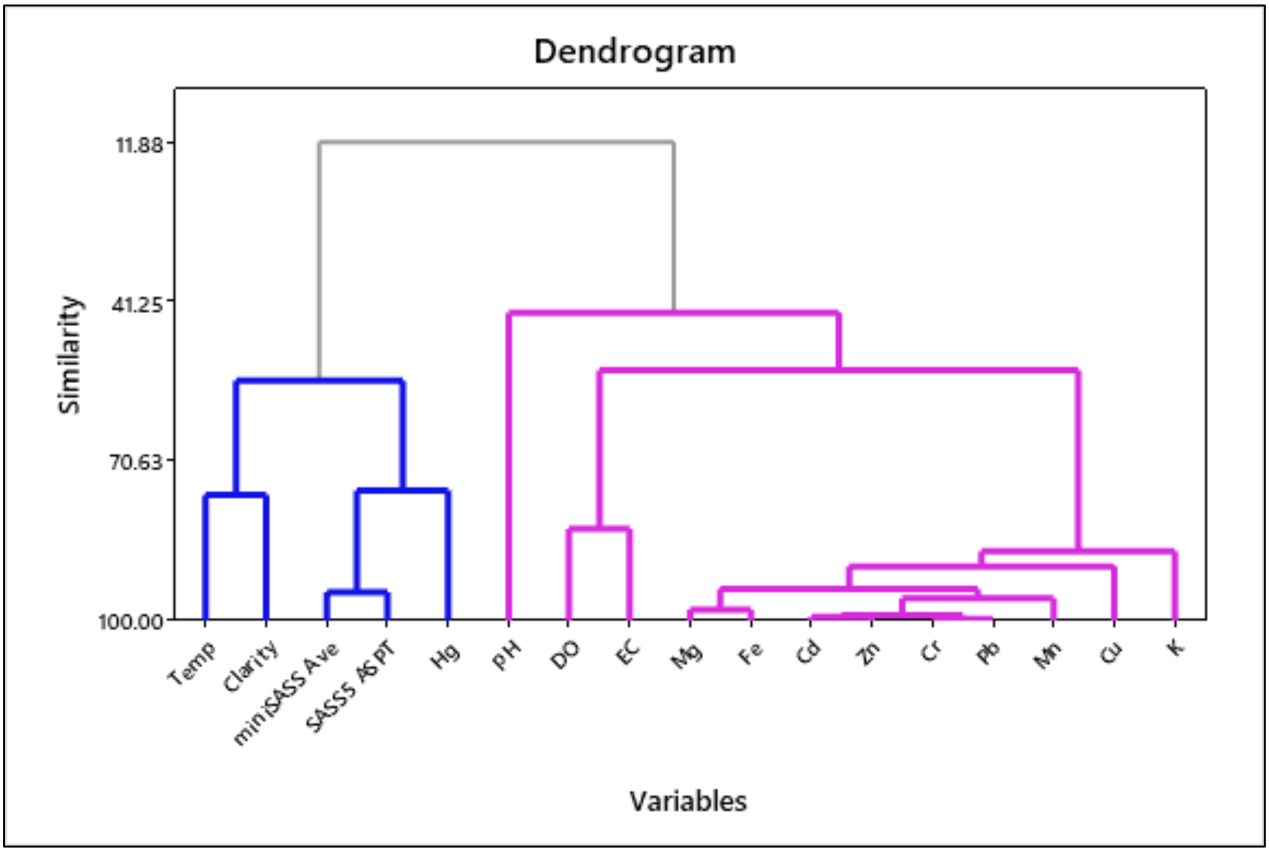


Figure 4.24 Cluster analysis of physicochemical, Biomonitoring metrics and heavy metals for the wet season

Figure 4.25 displays the dendrogram of sample sites. The dendrogram was created using a final partition of four clusters, which occur at similarity level of 99,92%. The first cluster (blue) is composed of four observations (S1, S3,S9 and S17) with a similarity level of 99,97% while S2 clustered in yellow shows a similarity level of about 99,92% The third cluster (green) is composed of five observations (S4, S5, S11, S6, S7 and S14) with similarity level of 99,96%. Cluster four (purple) consist of six observations (S8, S10, S11, S12, S13, S15 and S16) represent a similarity level of about 99,99%.

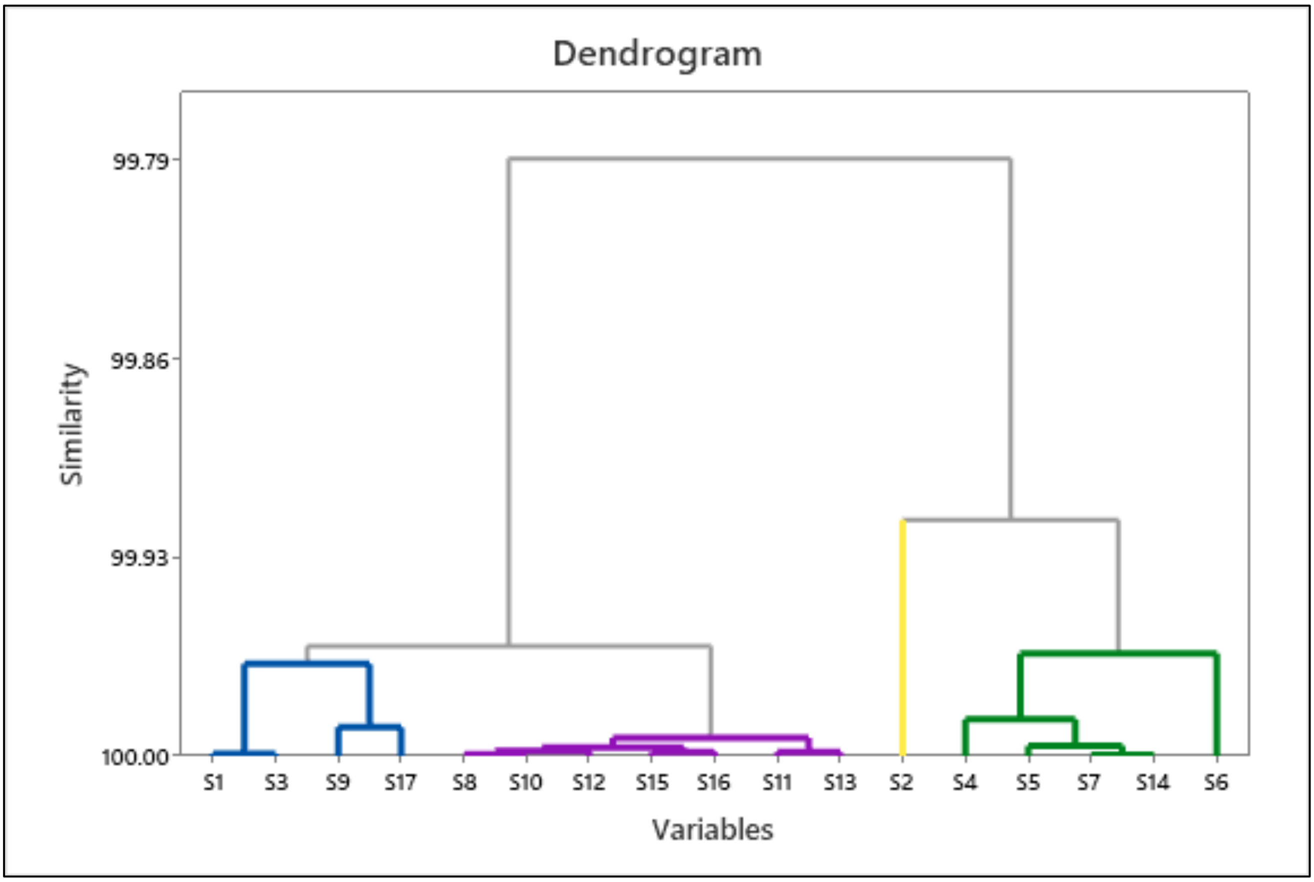


Figure 4.25 Cluster analysis of sampling sites for the wet season

Figure 4.26 displays the principal component analysis (PCA) biplot which shows the correlations between the physicochemical parameters, Biomonitoring metrics and heavy metals in the different sites. The components used have eigenvalues greater than 1, therefore the components explain 86.4% of the variation in the data. In the results the first principal component has large positive associations with heavy metals (except Hg) and EC, therefore this component primarily measures the geological structure of the river and the anthropogenic input. The second component has large negative associations with DO, SASS5 ASPT, miniSASS Ave, clarity and pH.. The parameters also show that they have a positive correlation amongst each other. PCA also shows the weak influence of temperature and Hg on the second component.

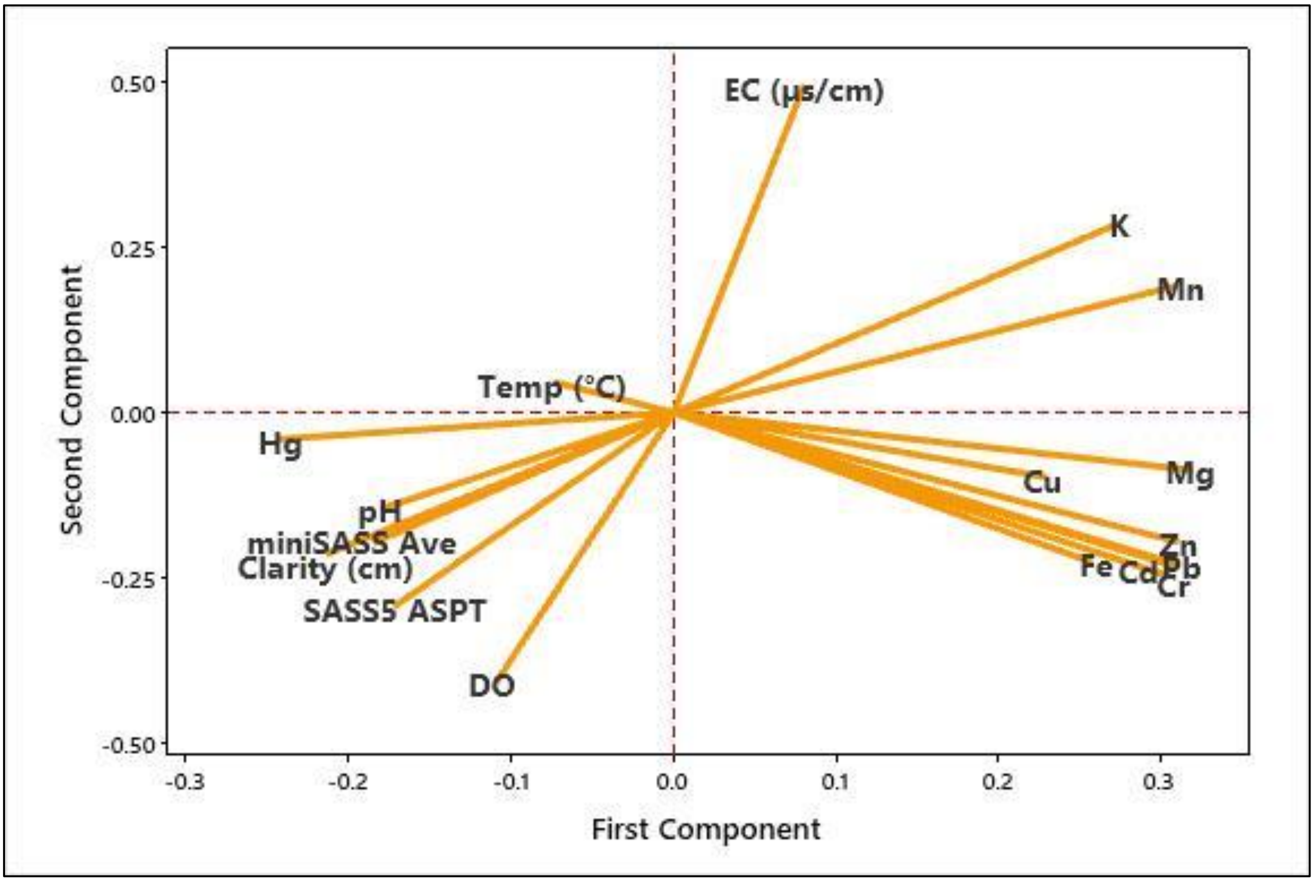


Figure 4.26 PCA biplot showing the correlation between physicochemical parameters, Biomonitoring metrics and heavy metals in the different sites during the dry season

Figure 4.27 displays the PCA score plot that shows the clustering of sample sites based on their similarity. The first component has positive associations with Sites 13, 15, 14, 18, 19, 11 and 11a with heavy metals and EC. Sites 1, 2, 3, 4, 5, 6, 7, 7a, 8, 9, 10, 12, 12a, 20, and 22 are loaded negatively to the first component with miniSASS, SASS5, DO, temperature, pH, Hg, and clarity. The grouping of these sites was influenced by the land use around with sites from 1 to 6 are located upstream of Dire reservoir, sites 8 to 10 located upstream of Legedadie reservoir.

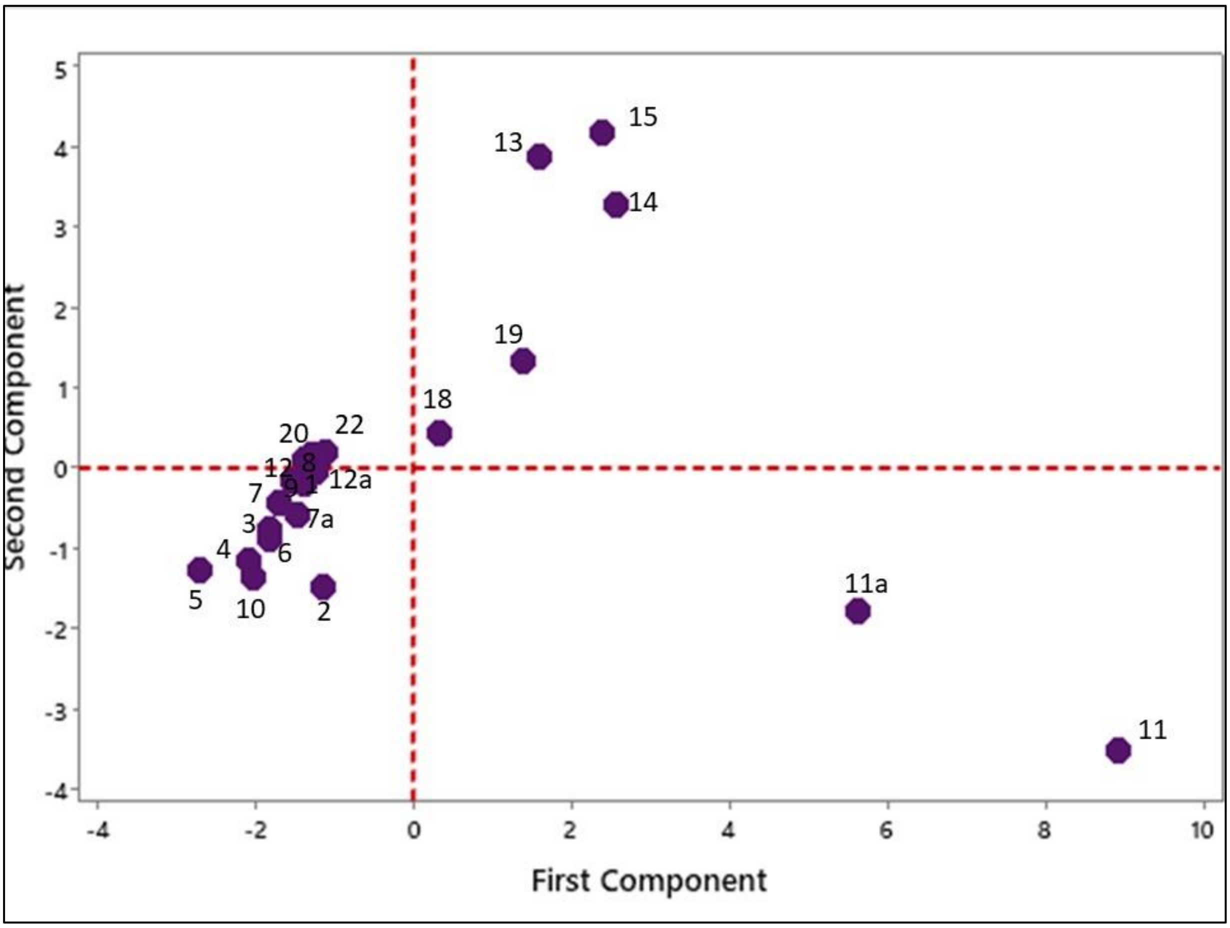


Figure 4.27 PCA score plot showing the clustering of sites based on similarities for the dry season

Figure 4.28 displays the dendrogram of physicochemical parameters, Biomonitoring metrics and heavy metals. The dendrogram was created using a final partition of four clusters, which occur at similarity level of 56%. The first cluster (blue) is composed of four observations (temperature, pH, DO and SASS5 ASPT) with a similarity level higher than 62% while miniSASS Ave, Hg and clarity shows a similarity level of about 56% in the second cluster (green). The third cluster is composed of two observations (electrical conductivity, K and Mn) with similarity level of 77%. While Mg, Fe, Cd, Zn, Cr, Pb, and Cu represent a similarity level of about 68%.

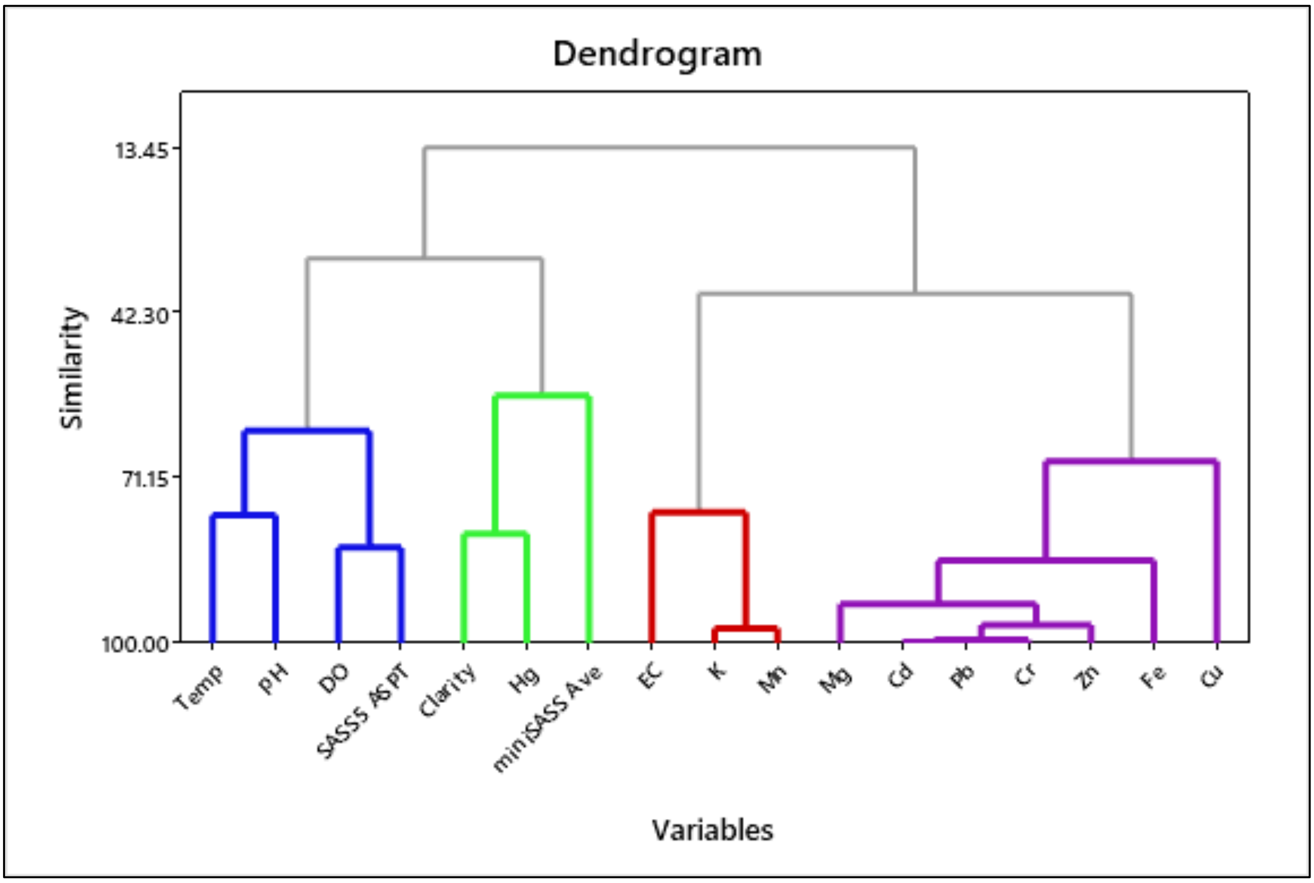


Figure 4.28 Cluster analysis of physicochemical, Biomonitoring metrics and heavy metals for the dry season

Figure 4.29 displays the dendrogram of sample sites. The dendrogram was created using a final partition of four clusters, which occur at similarity level of 69%. The first cluster (blue) is composed of eight observations (S1, S3, S4, S5, S7, S7a, and S10) with a similarity level higher than 88% while S2 clustered in yellow shows a similarity level of about 69% The third cluster is composed of thirteen observations (S6, S8, S11, S11a, S12, S12a, S14a, S17, S18, S19, S20, S21, and S22) with similarity level of 90%. Cluster four consist of six observations (S9, S13, S14, S15, and S16) represent a similarity level of about 96%.

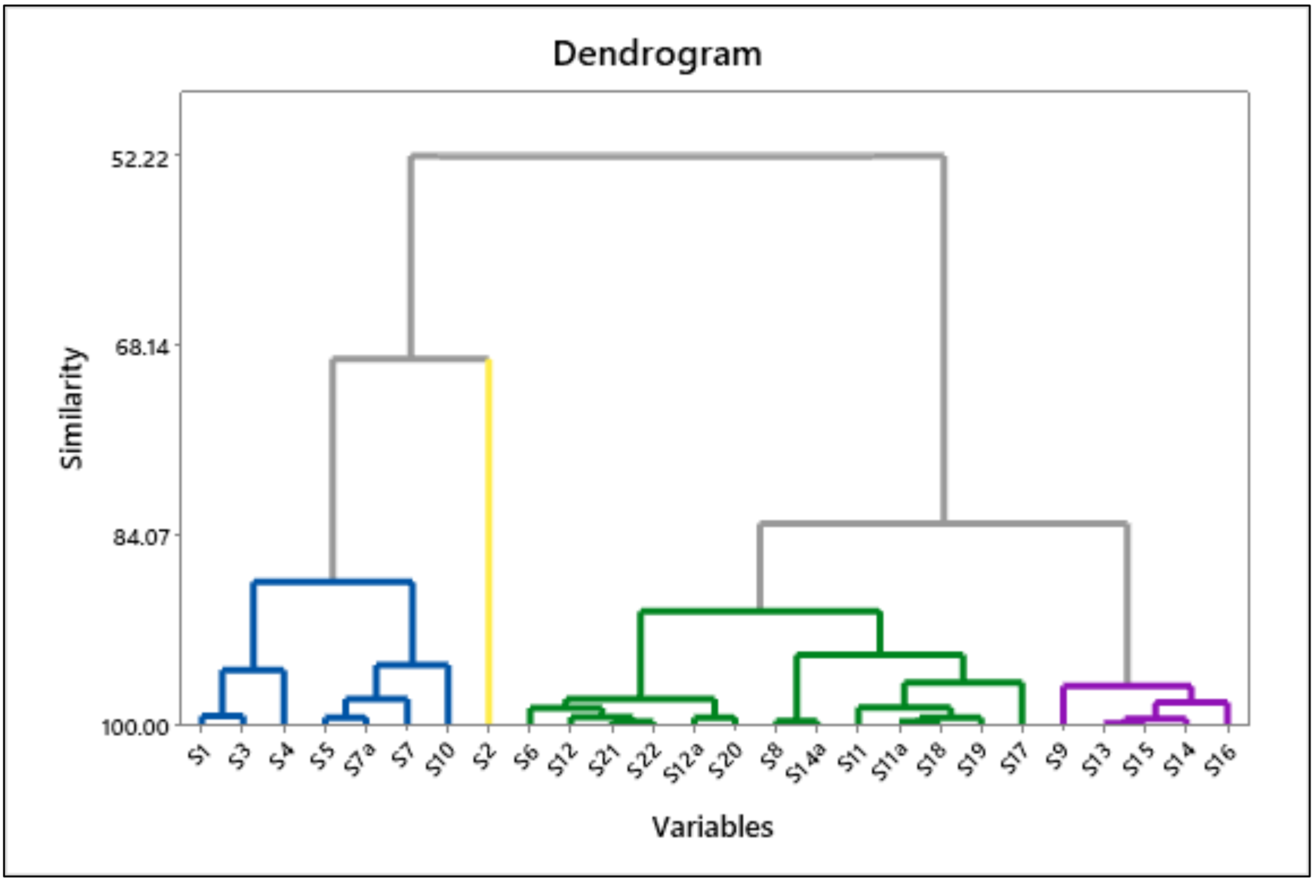


Figure 4.29 Cluster analysis of sample sites for the dry season

5 DISCUSSION

5.1 Physicochemical Parameters and Heavy Metals

A habitat that exhibits optimal physicochemical parameters is the ideal habitat for aquatic macroinvertebrates. These environments have dissolved oxygen levels generally above 5 mg/L and cool temperatures, which are essential to maintaining dissolved oxygen levels. To support a diverse macroinvertebrate population, the water should ideally be neutral to slightly alkaline in pH and low in pollutants such as heavy metals, pesticides and organic waste. A comprehensive water quality standard was developed by the Ethiopian EPA to make sure macroinvertebrates can thrive in an ideal habitat. Therefore, the study analysed the changes in physicochemical parameters and heavy metal status along the Upper Awash and across the dry and wet seasons were assessed and compared with the Ethiopian EPA standards.

Temperature results in Figure 4.1 varied across the sample sites during the wet and dry seasons, and this may be attributed to the time and weather of sampling. Site 1 measured the lowest temperature, which may be attributed to the fact that Site 1 is located at a high altitude. Site 15 measured the lowest temperature during the wet season, and this may be attributed to the fact that the site had more shade due to the steep wall of the bank of the river. The highest temperature recorded was 24.8°C at site S16 during the dry season. These temperatures fall within the acceptable range for aquatic life, although the higher temperatures in the dry season may be approaching stress thresholds for some macroinvertebrates.

The pH levels at the sample sites ranged from 7.4 to 9, indicating alkaline conditions. It was observed that pH levels were generally higher during the dry season compared to the wet season. These values fall within the Ethiopian EPA standards for surface waters, ensuring the pH is suitable for most aquatic organisms, including macroinvertebrates.

Dissolved oxygen levels varied across the sample sites, with most values meeting the minimum requirements for coarse fish but falling below the standards for game fish. Dissolved oxygen was the lowest in site 15, which is located in the river that passes through the city of Addis Ababa, where many industrial and domestic wastes in Addis Ababa are discharged directly into rivers with little or no treatment (Yohannes and Elias, 2017). Interestingly, sites S1 to S6 (above the Dire Reservoir) had higher DO levels during the dry season compared to the wet season, likely due to lower water temperatures and reduced biological activity.

EC values in Figure 4.4 ranged from 171 µs/cm to 1094 µs/cm during the dry season and from 146 µs/cm to 754 µs/cm during the wet season. Most sites recorded EC values below the Ethiopian EPA standard of 1000 µs/cm, except for sites S14 and S14a after the Aba Samuel Reservoir during the dry season. During the wet season, all sites were within acceptable limits. Higher EC values during the dry season may indicate increased concentration of dissolved salts due to lower water volumes, potentially stressing macroinvertebrate communities.

Water clarity was generally higher during the dry season whereas sites with low clarity measurements were relatively predominant during the wet season. According to Masese *et al*. (2023), runoff during the wet season can increase the delivery of sediments into streams and rivers and elevate turbidity. Site 10 (upstream of Legedadi) had the highest clarity during the dry season, while Site 6 had the highest clarity during the wet season. Conversely, sites 11, 11a (downstream of Legedadi), 12, 12a (Confluence), and 15 (Kebena) showed the lowest clarity levels during the dry season. High turbidity during the wet season can negatively impact macroinvertebrates by smothering habitats and reducing light penetration necessary for photosynthesis.

From the nine selected total heavy metals, the analysis focused on whether the concentrations of these metals exceeded the Ethiopian EPA Water Quality Standards. High concentrations of magnesium were observed at sites 11 and 11a, while the lowest concentration was measured at site 12 after the confluence of the Dire and Legedadi rivers. This variation indicates localized sources of magnesium pollution, particularly downstream of certain areas, which could impact aquatic life differently across sites.

Potassium levels peaked at site 14a downstream of the Aba Samuel reservoir. In contrast, sites upstream of the Dire reservoir showed low potassium levels in both seasons, with a general rise upstream of the Legedadi reservoir before decreasing at sites 12 and 12a. During the wet season, potassium concentrations were lower at sites 13, 14, 15, and 17 compared to the dry season, suggesting dilution effects from higher water flows.

Most sites recorded cadmium concentrations below the Ethiopian EPA standard, except for site 11, which consistently displayed elevated levels. This indicates a significant pollution source at site 11, potentially posing a risk to aquatic ecosystems and necessitating targeted remediation efforts.

Chromium concentrations were generally low across the majority of sites and within acceptable limits, except for site 11, which recorded very high levels (approximately 270 µg/L) exceeding the standards. Sites 13 and 16 also showed elevated chromium during the wet season, while site 14a exceeded the standard during the dry season, indicating seasonal and site-specific chromium pollution sources.

Copper concentrations in Figure 4.10 exceeded Ethiopian EPA standards at sites 1, 9, 11, 11a, 14, 14a, 17, and 18 during the dry season, with site 11 showing high levels in both seasons. This suggests significant copper pollution in these areas, likely from anthropogenic activities, which could harm local macroinvertebrate populations and other aquatic life.

Mercury levels were above the Ethiopian EPA standard at all sites during both seasons, highlighting widespread mercury contamination throughout the basin. This pervasive pollution poses severe risks to both aquatic ecosystems and human health due to mercury's high toxicity and bio accumulative nature.

Iron concentrations varied, with most sites showing low to slightly elevated levels compared to the Ethiopian EPA standard. However, sites 11, 11a, 13, 15, 16, and 17 exhibited high iron levels during the wet season, with site 11 having extremely high concentrations during the dry season. These findings indicate significant iron pollution at these locations, particularly site 11.

Sites above and below the Dire reservoir and those on the west side of Addis Ababa (sites 1 to 7a and 20 to 22) showed low manganese concentrations within acceptable limits. However, site 11 consistently recorded high manganese levels in both seasons, pointing to a localized pollution source requiring attention.

Most sites exhibited lead concentrations below the Ethiopian EPA standard during both seasons, except for site 11, which showed elevated levels year-round. This persistent lead contamination at site 11 indicates a significant pollution source that poses a threat to the local aquatic environment and potentially human health.

Zinc concentrations were generally low across most sites, remaining below the Ethiopian EPA standards. However, sites 11 and 11a displayed significantly higher zinc levels, exceeding the standards, indicating localized zinc pollution sources that could impact aquatic life adversely.

The majority of the heavy metals were in low concentration in sites located upstream of Dire reservoir in the high altitudes and the western tributaries originating from the high altitudes, where there are no industry inputs. Notably, sites 11 and 11a downstream of Legedadi were observed to reflect high concentrations of most heavy metals compared to the other sites, and both sites measured 0cm using the clarity tube test, which means there are high amounts of sediments in the river. This may be attributed to the sediments in the river released from the water treatment plant, that these sediments carry the heavy metals.

When observing upstream (S16) and downstream (S17) of Koka reservoir, for most physicochemical parameters and heavy metals, S16 records the highest concentrations, which may imply that the pollution from upstream deposits in S16 then within the reservoir, the dilution process takes place, resulting in S17 recording lower concentrations of heavy metals and physicochemical parameters. However, this creates confounding efforts to assign impacts directly to the presence of the dam.

When observing upstream (Akaki tributary S13) and downstream (S14 and 14a) of Aba Samuel reservoir, the concentration of physicochemical parameters and heavy metals varied across the season. Some parameters were recorded to be below the ETH EPA standards and for some were above. This signifies the local contributions of different contaminations from riparian disturbances.

Overall, it was noted that the use of conventional methods to assess the health of rivers using heavy metals and physicochemical reflects the environmental changes and sources of pollution can be identified from the selected sites.

5.2 Biomonitoring Using Macroinvertebrates

Macroinvertebrates are adapted to a range of structural and environmental characteristics that support their needs for eating, breeding, shelter, and survival. An ideal habitat consists of a variety of substrates like gravel, sand, leaves, rocks, and organic debris that provide essential surfaces for attachment and hiding. A mixture of fast-flowing riffles and slow-moving pools creates a range of microhabitats. Abundant aquatic vegetation, including submerged plants, emergent plants, and algae, also provides food, shelter, and breeding sites. Macroinvertebrates can thrive in diverse environments due to their ability to adjust to varying environmental changes.

Although there might be concerns about using foreign Biomonitoring program from one area to another (Bere and Nyamupingidza, 2014), it's worth noting that all the macroinvertebrates identified in this research were common species that are used in SASS5 and miniSASS. Table 8.2 in Appedix B shows the macroinvertebrates that were identified using the SASS5 identification chart. Figure 8.2 and 8.3 shows examples of the macroinvertebrates that were found. Site 7 (downstream of Dire reservoir) recorded the most macroinvertebrates/ highest diversity (Naucoridae, Gerridae, Psephenidae, Leptoceridae, Planorbidae, Hydrophilidae, Ceratopogonidae, Heptageniidae, Hydropsychidae, Gyrinidae, Nepidae, Baetidae >2sp, Coenagrionidae, Chironomidae/midges, Aeshnidae, Planaria, Caenidae, Lymnaeidae, Physidae, Culicidae pupa) during the dry season, however SASS5 and miniSASS interpreted the site to be in a poor condition. This may be due to the availability of habitat and low flows compared to the wet season. The availability of sources of nourishment and microhabitats determines the macroinvertebrate taxa's spread within a river system (Raphahlelo *et al*., 2022). Therefore, observing the results in Figure 4.2 to Figure 4.4 which show the the proportion of sensitivity to tolerant taxa observed varied amongst tolerant, highly tolerant, sensitive, and highly sensitive macroinvertebrates in the different sites during the dry and wet season may be influenced by the habitat and availability of food. Ochieng *et al*. (2020) state that although macroinvertebrates used for temperate regions may also be found in tropical regions however, they may not share similar sensitivity to water pollution and may not occupy similar habitats. This indicates that tolerant taxa may also be found in sites with good conditions. Literature indicated that at most polluted sites, there are less diverse or absent macroinvertebrates which is also reflected by low DO, especially in the Akaki River (Dessie et al., 2024); this was indeed observed within the study (S13) and within other sites that were categorized as very poor/poor.

5.3 Sites

The ability of PCA score plots to group sites based on their similarity demonstrates that there is a good correlation (negative and positive) between physicochemical parameters, heavy metals and biometrics. The good correlations (negative or positive), therefore, imply that the water quality variables are influenced by environmental changes, which demonstrates the applicability of using multivariate tools in assessing river quality. However, it was noted that SASS5 and miniSASS reflect changes within a specific site within the study rather than showing a general trend from upstream to downstream. This may be accounted for by the variable land uses observed at the sites, the dilution by tributary streams and the different habitats, and, as anticipated, the lack of suitable biotopes of sites. The distribution of biotopes in a river varies with its geomorphological characteristics, with stones dominating upper reaches (Dallas, 2021). The majority of the sample sites along the Upper Awash River were dominated by rocks. Therefore, it is a challenge to account for trends of changes from upstream to downstream. Also, to determine the changes from upstream and downstream of reservoirs to account for the possible restorations done by the reservoirs on the quality of the river is a challenge because these Biomonitoring metrics will reflect the land uses above and below the reservoir.

5.4 Seasonality

In a study by Masese *et al*. (2023) in an Afrotropic river, seasonality had a major impact on runoff, erosion, and solute leaching, all of which affected water quality characteristics in the river. Which is also demonstrated in the study, PCA, showed expected correlations during the dry season as compared to the wet season. Also, the issue of high flows during the wet season and low flows during the dry season is a concern for comparison purposes; for example (S8) can only be measured during the wet season however, if flows are extremely high, the site cannot be sampled therefore, optimum conditions for sampling needs to be addressed. Overall, as expected, sampling during the wet season is also a challenge due to the large substrates in the samples.

5.5 Correlation

In Figure 4.22 PCA shows the large positive association of the first component with heavy metals and EC and Figure 4.25 the first principal component has large positive associations with heavy metals except for Hg and EC, which reflects the measurement of the geological structure of the river and anthropogenic input primarily form the industrial and agricultural sectors located within the basin. Agboola *et al*. (2019) state that sediments carry pollutants such as metals therefore, according to Smal *et al*. (2022) the number of heavy metals present in the sediments reflects both human activities and geological structure of the river. The PCA plots shows positive correlation amongst miniSASS Ave, SASS5 ASPT, Hg, clarity and Temp in Figure 4.22 and in Figure 4.25 positive correlation is shown amongst miniSASS Ave, SASS5 ASPT, Hg, clarity, pH and DO which reflects the measurement of the aquatic ecosystem health in the study. However, if the pH levels increase, and reach higher alkaline levels, the bioindicators would reduce. DO is consistent with other studies which reflect that DO is one parameter used to assess water quality and aquatic ecosystems health. According to Tampo *et al*. (2021) the health of rivers is reflected in the aquatic community, which combines structural and functional features. PCA also shows the weak association of temperature with the components during the dry season which is also reflected in a study by Fasil Degafu *et al*. (2013) conducted in the Upper Awash River where temperature may have not been a key parameter in defining the water quality among sampling sites. The positive correlation between Biomonitoring metrics and mercury is unexpected as high concentrations of mercury can lead to direct mortality in aquatic macroinvertebrates, however, the results may be due to long exposure of mercury that the macroinvertebrates have become more resistant to its effects. Essentially since mercury tends to accumulate in living organisms, it can induce behavioural changes in macroinvertebrates. Overall, the PCA plots displayed an inverse relationship between the Biomonitoring metrics (miniSASS Ave and SASS5 ASPT) and heavy metals (except Hg) which shows that when the Biomonitoring metrics reflect high scores, the heavy metals are low in concentration. Moreover, the plots also show an inverse relationship between the Biomonitoring metrics and some physicochemical parameters.

5.6 Applicability of miniSASS and SASS5

The noteworthy correlations between miniSASS average score and SASS5 ASPT, heavy metals and physicochemical parameters of sites recorded in this study reflected the success of SASS5 and miniSASS that the tools may be used to reflect environmental changes in water quality and ecological health of the Upper Awash tropical rivers. The ability of SASS5 ASPT and miniSASS Ave to reflect site specific environmental changes showed that these tools can identify the source of pollution. SASS5 and miniSASS tools are able to reflect what has been observed visually (including riparian disturbances). However, the lack of reference (natural rivers) sites in the Upper Awash River and tributaries poses a challenge in fully determining the applicability of SASS5 and miniSASS.

SASS5 was developed for temperate regions therefore, following the exact guidelines for sampling posed a challenged especially because tropical rivers are most likely to form pools and become dry during the dry season, therefore, SASS5 could not be applied in such areas. Moreover, SASS5 could not be applied in rivers that are deep during the wet season. Therefore, the results may be influenced by missing data. The interpretation guidelines for SASS5 data also poses a challenge as they are based on South Africa’s ecoregion-geomorphological zone.

However, in the case of miniSASS, following the simple guidelines, the tool was applicable to all sites, the guidelines were also applicable to the study sites because the sites reflected habitats that are mentioned in the miniSASS guidelines. Using miniSASS to identify macroinvertebrates is fairly easy. However, the identification guide needs to be modified to cater for macroinvertebrates which are in larvae stages.

SASS5 has the potential to be used as a foundation to develop Biomonitoring tools that are region specific with few modifications. As for miniSASS, the tool can be applicable in the tested region because of it’s simplicity, however, it can also be further developed to make it more user friendly especially in identifying macroinvertebrates that are in larvae stages.

6 CONCLUSION AND RECOMMENDATIONS

6.1 Introduction

This chapter contains a summary of the study's important findings, recommendations, and concluding notes. The important findings are given in accordance with the objectives of the study, research questions, and overall goal.

6.2 Summary of noteworthy findings

The study aimed to assess the applicability of SASS5 and miniSASS under different conditions than that the tools were developed for. The significance of the study was to introduce these Biomonitoring tools as a complementary approach to convention monitoring tools that uses physicochemical parameters and heavy metals which are a challenge to African countries due to challenges such as limited testing facilities, lack of adequate staff and expensive resources. Therefore, below are the key findings of the research in relation to the aim, hypothesis, objectives and research questions.

6.2.1 Assessing physicochemical parameters and heavy metals

Variations were observed along the sample's sites. Some of the physicochemical parameters and heavy metals were above the set ETH EPA standards for surface water and this may have been due to the anthropogenic activities, industries located within the basin and geological composition of the river. Overall, the physicochemical parameters and heavy metals were able to reflect what was observed in SASS5 and miniSASS.

6.2.2 Conducting miniSASS and SASS5 during the wet and dry season

Seasonality plays a crucial role in sampling especially in intermittent and ephemeral streams under tropical regions as it influences the flow and depth of rivers. Sampling in shallow rivers using the techniques is a challenge as that of sampling in high flow which affects the number of macroinvertebrates found. In the study, results showed that sampling during the dry season reflects greater anthropogenic effects as compared to the wet season.

6.2.3 Aquatic macroinvertebrates

The use of foreign score sheets to identify the macroinvertebrates for both miniSASS and SASS5 was a concern, however, for this study all macroinvertebrates found were present in the score sheets. Macroinvertebrates found in their larvae stages were challenging to identify especially for miniSASS which may have an influence in the scores. The distribution of macroinvertebrates also varied amongst sites and between the season and this may be due to the habitat and availability of food.

6.2.4 Comparing miniSASS, SASS5, physicochemical parameters and heavy metals

Results showed the largest inverse relationship between the Biomonitoring metrics (miniSASS Ave and SASS5 ASPT), physicochemical parameters, and the heavy metals except for mercury (Hg). Which indicate that when SASS5 ASPT and miniSASS reflect good conditions of a site, the heavy metals are most likely to be in low or within accepted concentrations required for the survival of macroinvertebrates. Noteworthy, turbidity measured as clarity showeda positive correlation with the miniSASS Ave and SASS5 ASPT.

6.2.5 Modify the SASS5/miniSASS identification guidelines

The study demonstrated that miniSASS and SASS5 can be applicable under different conditions however, more studies can be done to validate this study. Therefore, for the purpose of the study no modifications were done in the identification guide. However, it is recommended that further studies test these tools in conjunction with ETHBios and develop interpretation guidelines in line with the Ethiopian ecoregion-geomorphological zones.

6.2.6. Water management implication of the findings

- Better management of wastewater discharge into the rivers
- Protection of tributary streams from further pollution
- Keep more water in the streams during the dry season so as to increase dilution capacity (by water retention in the wet season in upstream sites)

6.3 Recommendations

It is recommended that further studies incorporate more parameters such as BOD, COD as the study was limited in the measuring of the parameters. Further investigation can be done on the relationship between Biomonitoring metrics and mercury. And when choosing sampling sites located downstream of a dam/reservoir, the time of water release and volume of water released should be considered as it may influence the distribution of macroinvertebrates. For further research, community participation can be incorporated for miniSASS, to test the acceptance and ease of use from the perspective of non-expert in Biomonitoring in the basin.

Overall, capacity-building programs are critical for improving Biomonitoring in Africa. Local scientists and technicians need to be trained in monitoring procedures, data analysis, and interpretation. Collaboration among academic institutions, government agencies, non-governmental organizations, and local communities is also essential for facilitating data sharing, establishing monitoring networks, and developing comprehensive water management programs. Biomonitoring is frequently used in conjunction with physical and chemical water quality evaluations to provide a comprehensive picture of ecosystem health therefore, integrating data from numerous sources provides for more accurate evaluations and aids in identifying pollution sources and impacts. Biomonitoring data can also be used to guide policy decisions and management activities.

Policymakers can implement suitable actions to minimize pollution and maintain aquatic ecosystems by identifying water quality issues and their causes. Effective water management methods can encourage long-term water use, protect public health, and maintain biodiversity. It is crucial to highlight that the specific procedures and methods utilized in Biomonitoring can differ based on the region, available resources, and monitoring program objectives. However, miniSASS and SAS5 can form the basis for Biomonitoring programmes best fit for that particular region. Local and regional environmental organizations, research institutes, and international organizations all play important roles in supporting Biomonitoring and water resource management in Africa.

6.4 Concluding remarks

Assessing the health of rivers in tropical regions cannot solely depend on conventional methods. Therefore, miniSASS and SASS5 can aid in monitoring rivers concurrent with conventional methods. miniSASS and SASS5 are inexpensive, easy-to-use tools that can reflect the ecological status of rivers under different conditions.

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8 APPENDICES

Appendix A

Table 8.1 Description of sampling sites in the Upper Awash basin

			Description	
Site no.	**Coordinates Latitude and Longitude**		**Wet season**	**Dry season**
Upstream of Dire reservoir				
S1	9.183155	38.931178	The river was observed to be dominated by stones and bedrock with marginal vegetation on the banks of the river. The river had rapids. On the side of the river is a steep mountain with vegetation and falling rock. On the other riverbank, there was a tree plantation taking place.	The river had no rapids and was fairly shallow.
S2	9.181692	38.931672	The river was observed as dominated by stones and bedrock, with marginal vegetation on the banks of the river. The river is shallow with mud	The river was observed to have less mud on the side. Riparian disturbances continue from people

			on the side. Riparian disturbances occur from people crossing and herding the cattle across the river. The river had a variety of flows, rapids and fairly consistent flows.	crossing the river and herding cattle. The river was shallow with slow flows.
S3	9.180724	38.931399	The river forms a meander in between the sample sites. The sample site was observed to be dominated by bedrock and stones. Riparian disturbances are from the farming activities which cause stones to fall into the river, cattle graze along the sides of the site and drink from the river.	The river was observed to be shallow with algae on the rocks. Less disturbances on the day of sampling.
Upstream of Dire Reservoir				
S4	9.177717	38.916594	The sample site was observed to be dominated by bedrock and stones with marginal vegetation on the banks of the river. People use water for domestic purposes. Riparian disturbances include cattle grazing nearby and drinking from the river.	The sample site was observed to have less water flowing, exposing more bedrock with algae.
S5	9.175686	38.919047	The site is dominated by bedrock and stones. Marginal vegetation is present on the riverbanks. The water is shallow. Riparian disturbances	The river was clear in colour, exposing the algae on the bedrock. The river showed no flow which created pools of water.

			include cattle crossing, people using the water for washing and other domestic uses.	
S6	9.174863	38.92154	The river is dominated by bedrock and stones, with marginal vegetation on the riverbanks. The river is shallow then forms a pool which is deep and then continues.	More bedrock was exposed due to less flow and amount of water. The river was shallow. With cattle crossing in the river.
Downstream of Dire reservoir				
S7	9.1436111	38.9325	The site is dominated by bedrock and stones. On the sides of the river, the land is used for agricultural purposes. Riparian disturbances include cattle grazing and falling sand and rocks due to farming.	Disturbances observed on site remain the same as those observed during the wet season. Algae on the rocks was observed.
S7a	9.0884233	38.9300253	-	The site was observed to be dominated by rocks and bedrock. The river was relatively shallow with low flows.
Upstream of Legedadi Reservoir				
S8	9.0935071	38.9662717	Site dominated by bed rocks and stones with rapid flows. Riparian disturbances include cattle. Marginal vegetation on the banks of the river.	The rocks and stones were more exposed as the river was relatively dry forming pools of water covered in algae.

S9	9.0928014	38.9674111	Site dominated by rocks and stones with rapid flows. Riparian disturbances included trucks crossing through the river.	The site showed no flow of water, the site was covered in rocks and vegetation (grass).
S10	9.0917778	38.9678181	The site had marginal vegetation and deep in depth with relatively high flows. The river was narrower compared to site 8 and site 9.	The site was relatively shallow, exposing the riverbed (sand) covered in algae.
Downstream of Legedadi Reservoir				
S11	9.0649083	38.95612	The sample site is located downstream of the Legedadi Reservoir and water treatment plant. The river was observed to be fairly shallow and dominated by bedrock and stones. The river has a high content of sedimentation. The riverbanks have marginal vegetation and also the vegetation forms an island in the river. There are also forest trees along the slope of the mountain. Riparian disturbances are minimal. a small	The site was relatively shallow with high content of sedimentation. However, the river covered less of the riverbed.
S11a	9.0576505	38.9420706	-	Site is located downstream of S11. The river was observed to be dominated with stones and rocks. Maintaining the same brown colour as that of site 11.
After the confluence of the Dire and Legedadi rivers				

S12	9.0380556	38.9063889	The sample site is located after the confluence of the Dire and Legedadi rivers. The river is dominated by bedrock and stones. Pools of water form along the sides of the river. With relatively high flows. The river has a low diversity of marginal vegetation. Riparian disturbances include people washing and crossing the river.	The sample site was muddier with low flows.
S12a	8.9931844	38.9032558	-	The sample site is located downstream of S12. The river is dominated by bedrockrock and stones with minimal vegetation and very high turbidity.
Above Aba Samuel Reservoir-Akaki River				
S13	8.8777445	38.7858067	Brown river water, full of sand. Riparian disturbances include human disturbances (collection of sand) and Cattle drinking water.	Dark brown river water with pollution (papers, plastic bottles, clothes). The flow of water is not too fast nor too slow. Riparian disturbances include construction of a bridge. Cows drink from the river and people continue to collect sand in the river. Foul smell.
Below Aba Samuel Reservoir				
S14	8.7869444	38.7063889	Overflow of Aba Samuel dam, high flows with rapids. River dominated by stones and bedrock and marginal vegetation.	No overflow was observed from the dam. Natural release of water. Water is too shallow near the banks of the river.

S14a	8.7762124	38.7077764	-	The site was located downstream of the Aba Samuel Reservoir. The site was deep with slow flows and dominated with rocks.
City River-Kebena				
S15	9.0401248	38.7778385	The sample site is in the city river called Kebena. The river feeds into the Bulbula river. Site dominated by bedrock and stones and sand. Riparian disturbances include toilets on the banks of the river, and people using the river as a sanitary place. The colour of the river was brown.	The river was fairly shallow and has a foul smell. The river was observed as black in colour.
Upstream of Koka				
S16	8.4013751	39.0058716	Wide, deep river. Riparian disturbances include fishing. Difficult to access, requires a boat. Thiaridae (Snails) found in the riverbank in abundance.	Similar observations were seen as that of the wet season except for the colour which changed from brown to green.
Downstream of Koka				
S17	8.4794199	39.2320254	Wide, deep river with slow flow. Relatively polluted with plastics and surrounded by sugarcane farming.	Similar observations were seen as that of the wet season.
S18	8.82464	38.372924	-	Sandy river, fairly deep with marginal vegetation.

S19	8.8253314	38.374117	-	Sandy river, with light brown water, with no marginal vegetation.
S20	9.07379	38.510017	-	Sandy river, with light brown water. Fairly deep with slow flow of water. Riparian disturbances include kids using the river for swimming.
S21	9.073976	38.059131	-	Shallow river with clear water. Full of rocks and stones. Slow flow of water.
S22	9.031177	38.125398	-	Sandy river with clear water. Sandy river bed with a steep bank on one side and a flat floodplain on the other side. Slow flow of water and a narrow channel.

Appendix B

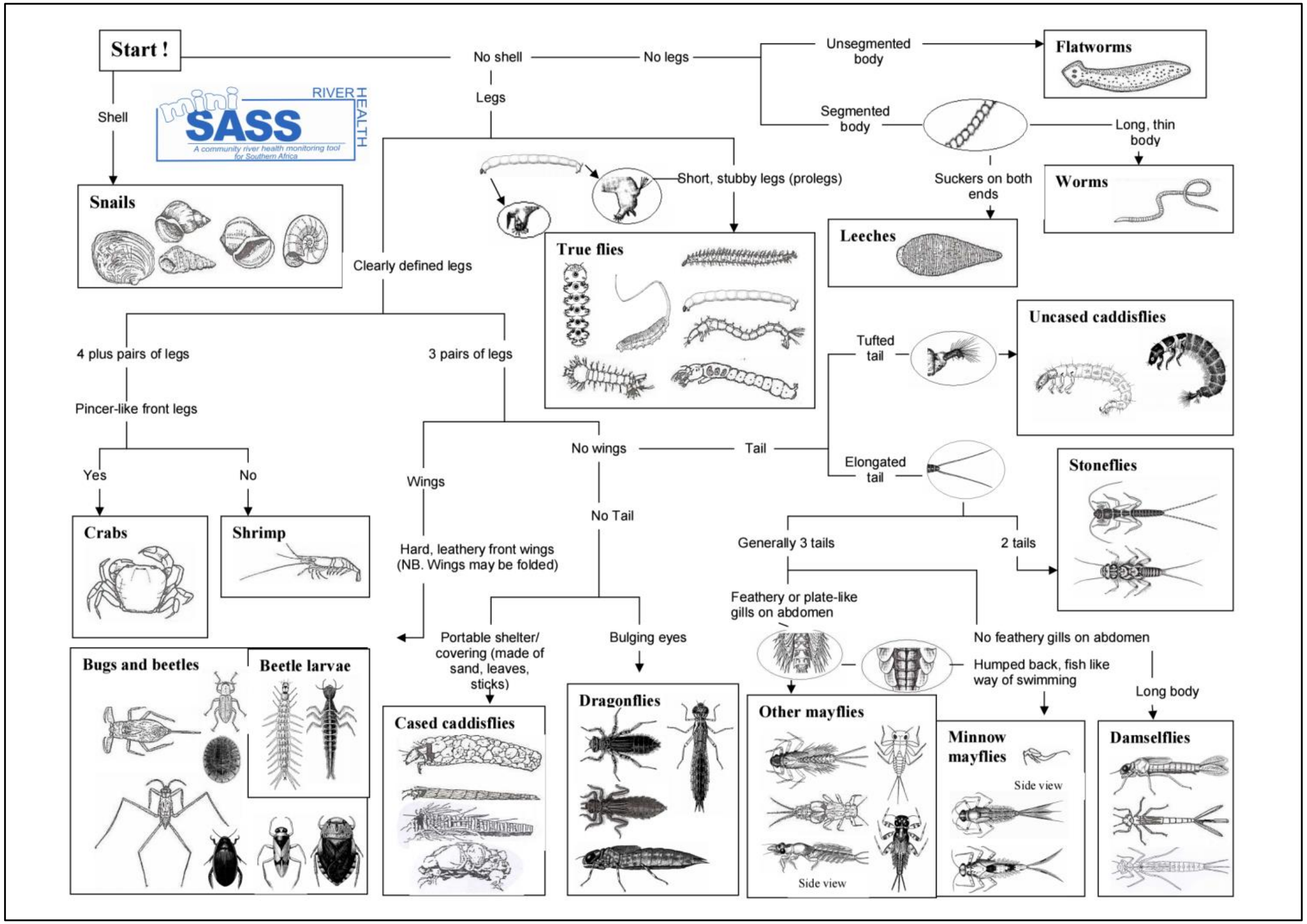

Figure 8.1 miniSASS Dichotomous key (*www.miniSASS.org*, 2022)

Table 8.2 Macroinvertebrates found during the dry and wet season using the SASS5 Identification guide

Site	**Wet season**	**Dry season**
1	Baetidae 1sp, teloganodidae, naucoridae	Chironomidae, caenidae, muscidae pupa, oligochaete, libellulidae, Culicidae pupa
2	Baetidae 1sp, Baetidae 2p, Baetidae>2p, Simuliidae, Heptageniidae, Teloganodidae, Hydrometridae, Veliidae, Naucoridae, Elmidae, Psychodidae (moth flies)	Corixidae, Annelida Hirudinea, Gyrinidae, Baetidae>2p, Chironomidae, Oligochaeta, Simuliidae, Ceratopogonidae, Aeshnidae, Planaria, Caenidae, Culicidae pupa
3	Hydropsychidae 1sp, Naucoridae, Baetidae 1sp, Baetidae>2p, Teloganodidae, Physidae	Hydropsychidae, 1sp, Naucoridae, Baetidae>2p
4	Baetidae 2p, Baetidae >2p, Naucoridae, Turbellaria, Coenagrionidae, Gomphidae, Gerridae, Notonectidae, Pleidae, Hydrophilidae, Tabanidae	Corixidae, Baetidae >2p, Chironomidae, Naucoridae, Planaria, Caenidae, Hydropsychidae 1sp, Dytiscidae

5	Baetidae 1sp, Baetidae >2sp, Aeshnidae, Hydropsychidae 1sp, Caenidae, Oligochaeta, Teloganodidae, Coenagrionidae, Corixidae, Naucoridae, Nepidae, Hydrophilidae	Baetidae >2sp, Chironomidae/midges, Muscidae, Aeshnidae, Heptageniidae, Hydropsychidae 1sp, Planaria, Caenidae
6	Notonectidae, Planorbidae, Heptageniidae, Hydropsychidae 1sp, Baetidae 2sp, Baetidae >2sp, Caenidae, Synlestidae, Coenagrionidae, Dytiscidae, Gyrinidae, Hydrophilidae	Naucoridae, Gerridae, Tipulidae, Notonectidae, Planorbidae, Muscidae, Heptageniidae, Hydropsychidae 1sp, Gyrinidae, Baetidae 2sp, Baetidae >2sp, Chironomidae/midges, Aeshnidae, Planaria, Caenidae Culicidae pupa
7	Naucoridae, Gerridae, Planorbidae, Heptageniidae, Baetidae >2sp, Coenagrionidae, Lymnaeidae, Culicidae pupa, Turbellaria, Hirudinea, Synlestidae, Libellulidae, Pleidae	Naucoridae, Gerridae, Psephenidae, Leptoceridae, Planorbidae, Hydrophilidae, Ceratopogonidae, Heptageniidae, Hydropsychidae, Gyrinidae, Nepidae, Baetidae >2sp, Coenagrionidae, Chironomidae/midges, Aeshnidae, Planaria, Caenidae, Lymnaeidae, Physidae, Culicidae pupa
7a	-	Naucoridae, Gerridae, Oligochaeta, Leptoceridae, Planorbidae, Ceratopogonidae, Heptageniidae, Hydropsychidae, Baetidae, Coenagrionidae, Chironomidae/midges, Simuliidae, Muscidae, Caenidae, Pleidae, Physidae, Culicidae pupa

8	Oligochaeta, Chironomidae, Lymnaeidae, Physidae	-
9	Turbellaria, Chironomidae, Lymnaeidae, Physidae, Sphaeriidae (Pill clams)	-
10	Naucoridae, Turbellaria, Oligochaeta, Hyacarina, Chironomidae, Lymnaeidae, Physidae, Planorbinae	Naucoridae, Nepidae, Coenagrionidae, Planaria, Notonectidae, Gerridae, Dytiscidae, Hydrachnellae
11	Chironomidae, Turbellaria, Oligochaeta, Sphaeriidae	Gerridae, Dytiscidae, Chironomidae, Planaria, Ceratopogonidae
11a	-	Planaria, Baetidae 1sp, Dytiscidae, Corixidae
12	Oligochaeta, Baetidae 1sp, Caenidae, Simuliidae, Physidae	Aeshnidae, Hydropsychidae, Planaria, Heptageniidae

13	Oligochaeta	Chironomidae, Oligochaeta, Culicidae pupa
14	Simuliidae, Chironomidae, Oligochaeta, Baetidae 1sp, Physidae	Simuliidae, Hydropsychidae, Chironomidae, Oligochaeta
14a		
15	Psychodidae	Psychodidae
16	-	-
17	-	-

18	-	Baetidae >2sp, Hydropsychidae, Naucoridae, Caenidae, Hydrophilidae, Hirudinea
19	-	Baetidae >2sp, Naucoridae, Caenidae, chironomidae
20	-	Baetidae >2sp, Hydropsychidae, Naucoridae, Caenidae, Hydrophilidae, Hirudinea
21	-	Baetidae, Hydropsychidae, Naucoridae, Heptageniidae, Tabanidae
22	-	Baetidae, Hydropsychidae, Naucoridae, Caenidae, Heptageniidae, Simuliidae, Ceratopogonidae, Chironomidae, Culicidae, Coenogrionidae, Physidae, Corixidae, Oligochaeta

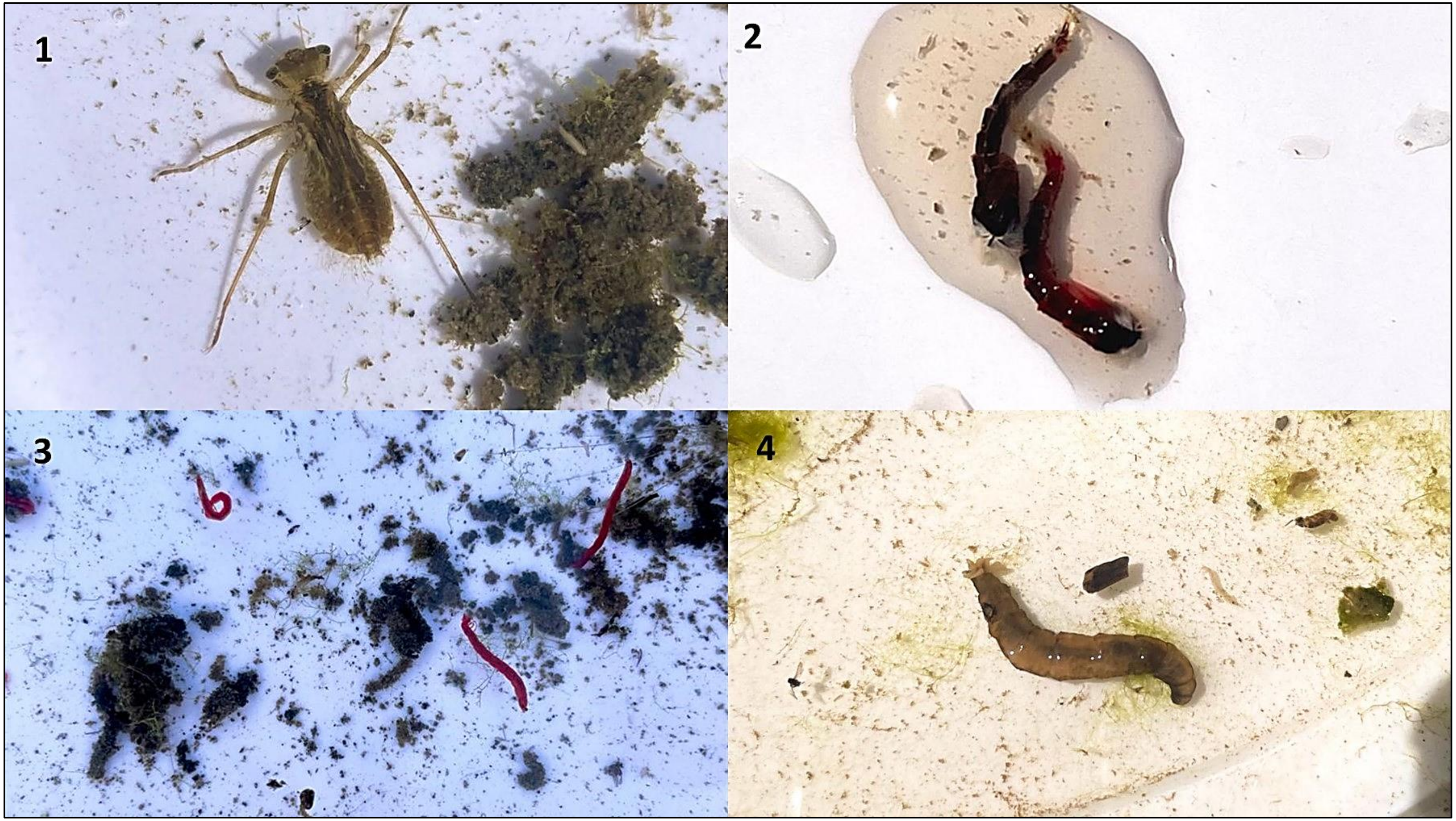

Figure 8.2 Examples of macroinvertebrates found at different sites during the wet season, libellulidae (dragonfly-1), Culicidae pupa (trueflies-2), Chironomidae (trueflies-3) and tipulidae (truefly-4) (Author, 2023)

Figure 8.3 Examples of macroinvertebrates found at different sites during the dry season, thiaridae (snail-right), aeshnidae (dragonfly-middle) and nepidae (bugs-left) (Author, 2022)

Appendix C

Table 8.3 Guideline SASS5 scores and ASPT values for interpreting SASS5 data in relation to river health for the Southern folded mountains lower zone (Dallas, 2007)

Ecological Category	SASS5 Score	ASPT Score	Ecological Name	Description
E/F	0-42	0-4.2	Critically modified	Critically modified
D	>42-53	>4.2- 4.5	Poor	Largely modified
C	>53-71	>4.5- 4.8	Fair	Moderately modified
B	>71-104	>4.8- 5.8	Good	Largely natural with few modifications
A	>104-180	>5.8- 8	Natural	Unmodified

Table 8.4 SASS5 calculated ASPT scores and interpretations

	Wet season	Dry season
Site 1		
measured	8	2.5
interpretation	Natural condition	Critically modified condition
Site 2		

measured	7	4.5
interpretation	Natural condition	Poor condition
Site 3		
measured	8	7.7
interpretation	Natural condition	Natural condition
Site 4		
	5	5.3
	Good condition	Poor condition
Site 5		
measured	5.75	6.1
interpretation	Good condition	Natural condition
Site 6		
measured	6.25	5.2
interpretation	Poor condition	Very poor condition
Site 7		
measured	5.38	5.4
interpretation	Poor condition	Poor condition
Site 7a		
measured	-	4.8
interpretation	-	Good condition
Site 8		

measured	2.4	-
interpretation	Critically modified	
Site 9		
measured	2.8	-
interpretation	Critically modified	
Site 10		
measured	3.75	9.6
interpretation	Critically modified	Natural
Site 11		
measured	2.2	3.8
interpretation	Critically modified	Critically modified
Site 11a		
measured	-	3.6
interpretation	-	Critically modified
Site 12		
measured	5.1	7.0
interpretation	Good condition	Natural condition
Site 12a		
measured	-	6.3
interpretation	-	Natural condition
Site 13		

measured	1.0	1.3
interpretation	Critically modified	Critically modified
Site 14		
measured	3.0	3.0
interpretation	Critically modified	Critically modified
Site 15		
measured	1.0	1.0
interpretation	Critically modified	Critically modified
Site 18		
measured	-	5.6
interpretation	-	Good condition
Site 19		
measured	-	6.7
interpretation		Natural condition
Site 20		
measured	-	6.3
interpretation	-	Natural condition
Site 21		
measured	-	8.2
interpretation	-	Natural condition
Site 22		

measured	-	5.1
interpretation	-	Good condition

Table 8.5 miniSASS calculated average scores and interpretation during the wet and dry season

	Wet season	**Dry season**
Site 1		
measured	7.5	7.0
interpretation	Good condition	Good condition
Site 2		
measured	8.75	5.8
interpretation	Natural condition	Poor condition
Site 3		
measured	7.25	8.3
interpretation	Good condition	Natural condition
Site 4		
	6.67	6.0
	Good condition	Poor condition

Site 5		
measured	7.75	6.0
interpretation	Good condition	Poor condition
Site 6		
measured	5.50	5.0
interpretation	Poor condition	Very poor condition
Site 7		
measured	4.75	5.4
interpretation	Poor condition	Poor condition
Site 7a		
measured	-	5.2
interpretation	-	Poor condition
Site 8		
measured	2.67	3.7
interpretation	Very poor	Very poor condition
Site 9		
measured	3.0	3.5
interpretation	Very poor	Very poor condition
Site 10		
measured	3.00	3.0
interpretation	Very poor	Very poor condition

Site 11		
measured	4.20	3.3
interpretation	Very poor	Very poor condition
Site 11a		
measured	-	4.25
interpretation	-	Very poor condition
Site 12		
measured	5.43	7.3
interpretation	Poor condition	Good condition
Site 12a		
measured	-	6.4
interpretation	-	Fair condition
Site 13		
measured	2.0	2.0
interpretation	Poor condition	Very poor condition
Site 14		
measured	2.67	4.3
interpretation	Very poor	Very poor condition
Site 14a		
measured	-	4.0
interpretation	-	Very poor condition

Site 15		
measured	2.0	2.0
interpretation	Very poor	Very poor condition
Site 16		
measured	4.0	5.0
interpretation	Good condition	Poor condition
Site 17		
measured	5.0	4.7
interpretation	Fair condition	Poor condition
Site 18		
measured	-	5.3
interpretation	-	Fair condition
Site 19		
measured	-	5.8
interpretation		Fair condition
Site 20		
measured	-	5.4
interpretation	-	Fair condition
Site 21		
measured	-	6.0
interpretation	-	Good condition

Site 22		
measured	-	5.3
interpretation	-	Poor condition

Appendix D

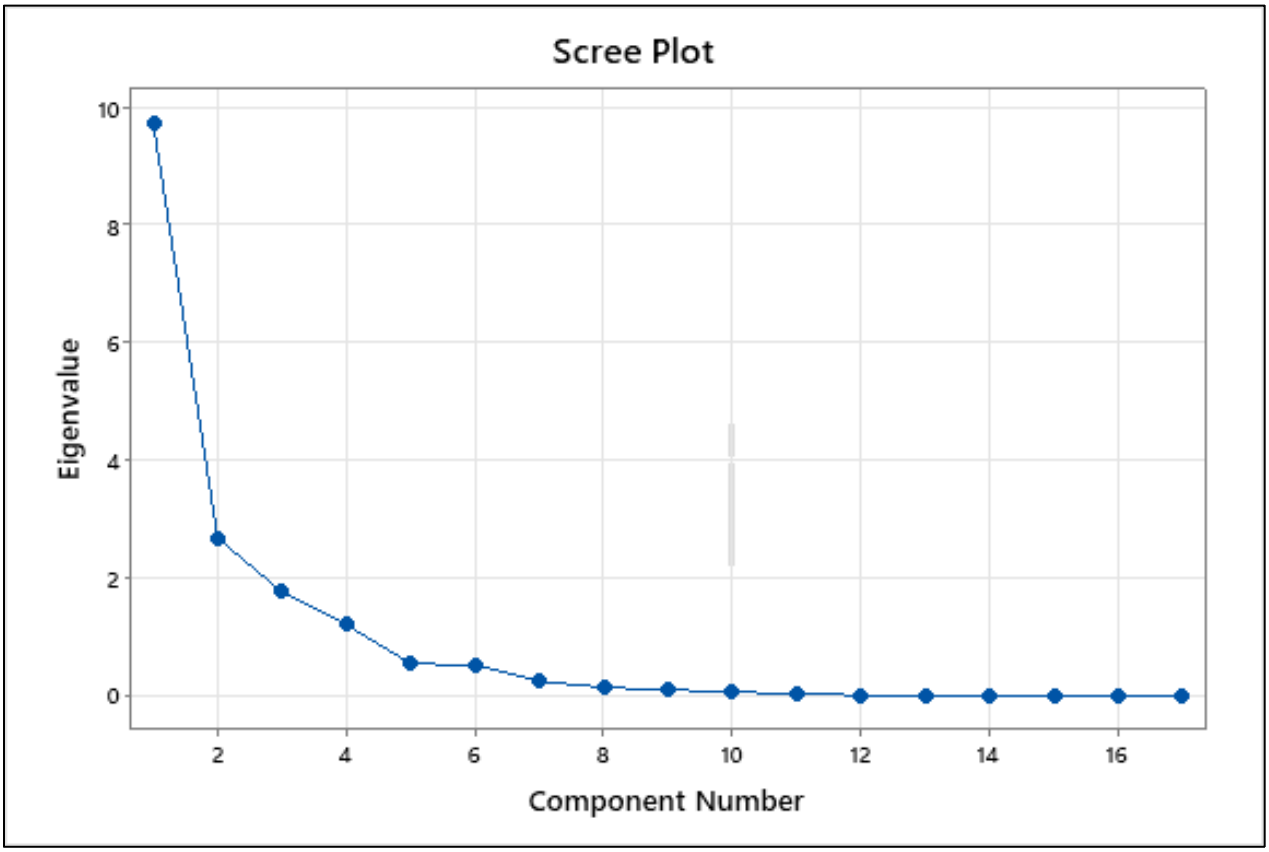

Figure 8.4 Scree plot for the wet season

Table 8.6 Eigenanalysis of the correlation matrix for the wet season

Eigenanalysis of the Correlation Matrix										
Eigenvalue	9.7277	2.677	1.7644	1.2051	0.5402	0.5165	0.2431	0.1371	0.0955	0.0654
Proportion	0.572	0.157	0.104	0.071	0.032	0.03	0.014	0.008	0.006	0.004
Cumulative	0.572	0.73	0.833	0.904	0.936	0.967	0.981	0.989	0.995	0.998
Eigenvalue	0.019	0.008	0.0009	0	0	0	0			
Proportion	0.001	0	0	0	0	0	0			
Cumulative	0.999	1	1	1	1	1	1			
14 cases used, 3 cases contain missing values										

Table 8.7 Amalgamation Steps to determine the final partition of the cluster for the wet season

Step	Number of clusters	Similarity level	Distance level	Clusters joined		New cluster	Number of obs. in new cluster
1	16	99.7263	0.00547	11	16	11	2
2	15	99.2646	0.01471	10	17	10	2
3	14	99.0198	0.01960	10	11	10	4
4	13	98.0325	0.03935	8	13	8	2
5	12	95.8611	0.08278	10	15	10	5
6	11	94.7594	0.10481	6	7	6	2
7	10	94.1663	0.11667	8	10	8	7
8	9	89.9874	0.20025	8	12	8	8
9	8	87.2431	0.25514	8	9	8	9
10	7	83.1118	0.33776	3	4	3	2
11	6	76.8532	0.46294	1	5	1	2
12	5	76.0477	0.47905	6	14	6	3
13	4	55.7618	0.88476	1	6	1	5
14	3	53.8222	0.92356	3	8	3	11
15	2	43.3292	1.13342	2	3	2	12
16	1	11.8753	1.76249	1	2	1	17

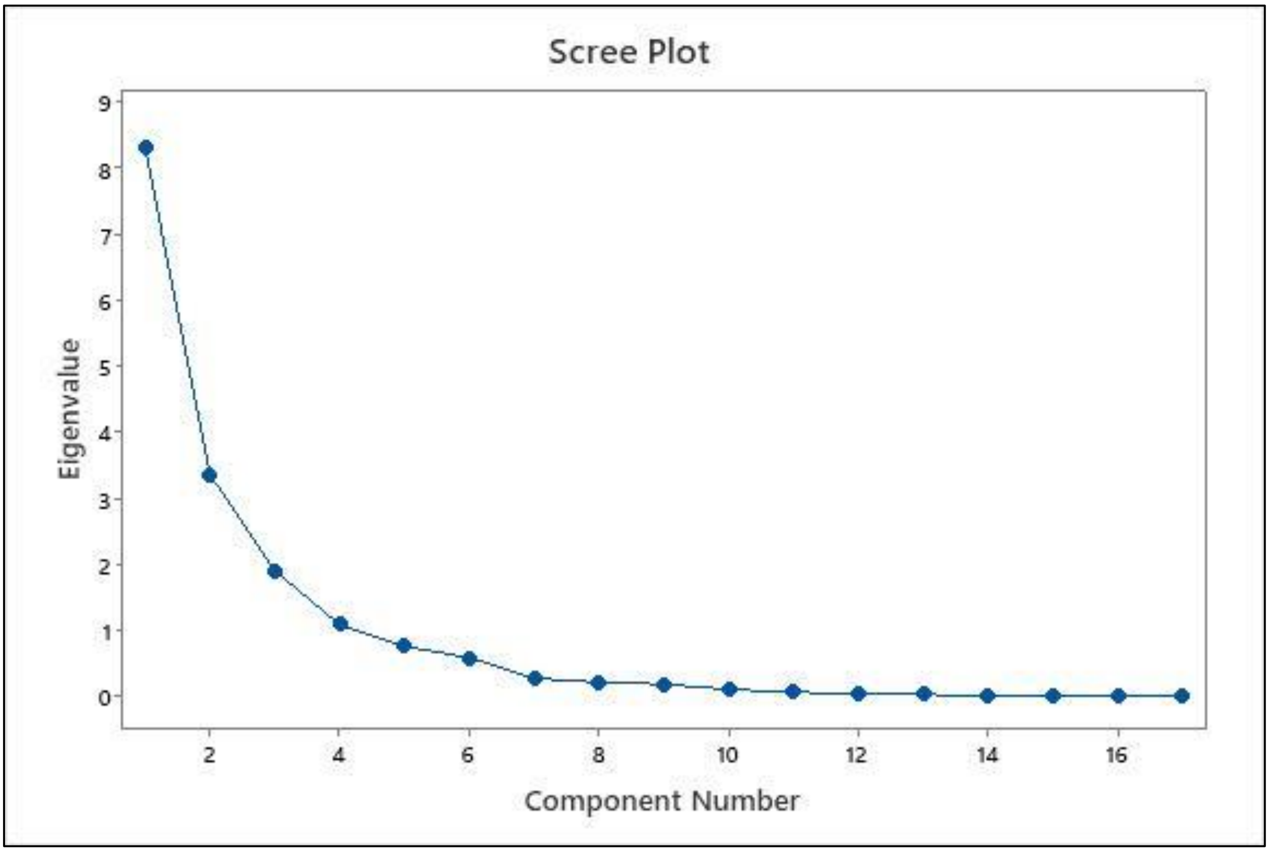

Figure 8.5 Scree plot for the dry season

Table 8.8 Eigenanalysis of the correlation matrix for the dry season

Eigen analysis of the Correlation Matrix										
Eigenvalue	8.3172	3.368	1.9065	1.0941	0.7613	0.5925	0.2796	0.2175	0.19	0.11
Proportion	0.489	0.198	0.112	0.064	0.045	0.035	0.016	0.013	0.011	0.006
Cumulative	0.489	0.687	0.8	0.864	0.909	0.944	0.96	0.973	0.984	0.99
Eigenvalue	0.0704	0.0452	0.0357	0.0083	0.0029	0.0006	0.0002			
Proportion	0.004	0.003	0.002	0	0	0	0			
Cumulative	0.995	0.997	0.999	1	1	1	1			
21 cases used, 7 cases contain missing values										

Table 8.9 Amalgamation Steps to determine the final partition of the cluster for the dry season

Step	Number of clusters	Similarity level	Distance level	Clusters joined		New cluster	Number of obs. in new cluster
1	16	99.8245	0.00351	10	16	10	2
2	15	99.4289	0.01142	10	11	10	3
3	14	97.4225	0.05155	9	15	9	2
4	13	96.8520	0.06296	10	17	10	4
5	12	93.1932	0.13614	8	10	8	5
6	11	85.5820	0.28836	8	13	8	6
7	10	83.2597	0.33481	3	7	3	2
8	9	80.9108	0.38178	5	14	5	2
9	8	77.6692	0.44662	1	2	1	2
10	7	77.1144	0.45771	4	9	4	3
11	6	68.2696	0.63461	8	12	8	7
12	5	62.9498	0.74100	1	3	1	4
13	4	56.7456	0.86509	5	6	5	3
14	3	38.9638	1.22072	4	8	4	10
15	2	32.8569	1.34286	1	5	1	7
16	1	13.4524	1.73095	1	4	1	17