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Developing a context specific climate smart aquaculture framework for improving food security in Tanzania

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DEVELOPING A CONTEXT SPECIFIC CLIMATE SMART AQUACULTURE FRAMEWORK FOR IMPROVING FOOD SECURITY IN TANZANIA

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A Thesis Submitted in Partial Fulfilment of the Requirements for the Degree of Doctor of Philosophy in Life Sciences of the Nelson Mandela African Institution of Science and Technology

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ABSTRACT

Aquaculture has great potential to improve global food and nutrition security. However, due to the effect of climate changes and poor fish farming practices in some countries, it favors the accumulation of chemicals and disease-causing pathogens including bacteria, viruses, and parasites. This study, therefore, aimed to assess the adequacy of existing aquaculture practices and evaluated the levels of heavy metals and the prevalence of parasites and bacteria pathogens. The information was used to design a context-specific climate-smart fish pond and fish feed for improving fish production in Tanzania. The study was conducted in Arusha and Morogoro regions, five sites from each region were selected for the study. A total of 130 fish farmers each with one fish pond were selected for interview and sample collection. The questionnaire was used to gather information on the existing aquaculture management practices. Pond water, sediments, fish feed, and fish samples were collected for the analysis of heavy metals, parasites, and bacteria pathogens. Polarized energy-dispersive x-ray fluorescence spectrometer was used for heavy metal analysis, a microscope was used to observe parasites present in fish samples, and analytical profile index test kits were used to identify bacteria pathogens. The results showed that farmers lacked proper knowledge of formulating high-quality fish feed and/or safe pond water management; these have a huge impact on overall fish health and consumer safety. The most prevalent parasites in Arusha and Morogoro were *Acanthocephalus* sp (49.2 & 50.7%) and *Diplostomum* sp (36.9 & 38. 4%). *Aeromonas sobria* was the most prevalent fish bacteria found in Arusha (35.3%) and Morogoro (49.2%). Chromium was the most accumulated heavy metal in the fish muscles sampled in Arusha (4.61 – 9.50 mg/kg) and Morogoro (2.53 – 5.57 mg/kg). In this study, we designed and constructed a climate-smart pond. The pond has the potential to support food security while reducing vulnerability to long-term climate change impacts. Google Sheets Program was used to formulate a higher quality insect-based fish feed. The quality and efficiency of the formulated feed were quantified by measuring growth performance and feed utilization. Fish were observed to have growth improvement and feed conversion efficiency throughout the experimental period. The use of the climate-smart fish pond and formulated feed is recommended in improving fish production and ensuring consumer safety.

DECLARATION

I, Frida Albinusi Nyamete, do hereby declare to the Senate of the Nelson Mandela African Institution of Science and Technology that this dissertation is my work and that it has neither been submitted nor being concurrently submitted for degree award in any other institution

_____

Frida Albinusi Nyamete   **Date**

Name and signature of the candidate

The above declaration is confirmed

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Dr. Jofrey Raymond   **Date**

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CERTIFICATION

The undersigned certify that they have read and hereby recommend for acceptance for the dissertation entitled *"Developing a Context-specific Climate-smart Aquaculture Framework for Improving Food Security in Tanzania"* In partial fulfillment of the Award of Doctor of Philosophy in Life Sciences at the Nelson Mandela African Institution of Science and Technology.

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DEDICATION

This dissertation is dedicated to my beloved uncle; Mr. Alex Mgaya who raised, cared, supported me, and laid down the foundation of my education; and to the loving memory of my parents, the late Mr. and Mrs. Albinusi Nyamete. May God rest their souls in peace.

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LIST OF ABBREVIATIONS AND SYMBOLS

ADWG	Average Daily Weight Gain
ANOVA	Analysis of variance
AOAC	Association of Official Agricultural Chemists
API	Analytical Profile Index
As	Arsenic
AUC	African Union Commission
BFAR	Bureau of Fisheries and Aquatic Resources
BHI	Brain Heat Infusion
BOD	Biological Oxygen Demand
BW	Body Weight
Cd	Cadmium
CR	Carcinogenic Risk
Cr	Chromium
CSA	Climate Smart Agriculture
CSF	Carcinogenic Slope Factor
DESA	Development of Economic and Social Affairs
DFC	Daily Fish Consumption
DO	Dissolved Oxygen
EAA	Ecosystem Approach to Aquaculture
EAF	Ecosystem Approach to Fisheries
EDI	Estimated Daily Intakes
ESP	Economic Stimulus Programme
FAO	Food and Agriculture Organization
FI	Feed Intake
GAP	Good Aquaculture Practices
GDP	Gross Domestic Product (GDP)
GHD	Green House gases (GHGs)
GIT	Gastro Intestinal tract
Hg	Mercury
HI	Hazard index
IPCC	Intergovernmental Panel on Climate Change
KNBS	Kenya National Bureau of Statistics
LIC	low-income countries
MC	Mean Concentration

MHA	Mueller-Hinton Agar
MLFD	Ministry of Livestock and Fisheries Development
MWG	Mean Weight Gain
NEPAD	New Partnership for Africa's Development
NGO	Non-Government Organization
NIMR	National Institute for Medical Research
NTU	Nephelometric Turbidity Units
OECD	Organization for Economic Co-operation and Development
Pb	Lead
PBS	Phosphate-Buffered Saline
PCA	Principle Component Analysis
PEC	Probable Effect Concentration
PED	Polarized Energy Dispersive
PER	Protein Efficiency Ratio
PI	Protein Intake
PVC	Polyvinyl Chloride
RDA	Redundancy Analysis
RDA	Redundancy Analysis
RfD	Reference Dose
SADC	Southern African Development Community
SD	Standard Deviation
SEAT	Sustaining Ethical Aquaculture Trade
SSA	Salmonella Shigella Agar
SUA	Sokoine University of Agriculture
TCBS	Thiosulphate Citrate Bile Salt Sucrose
TFAR	Tanzania Fisheries Annual Statistics Report
THQ	Target Hazard Quotient
TSA	Trypticase Soy Agar
UN	United Nations
URT	United Republic of Tanzania
USA	United State of America
USEPA	United State Environmental Protection Agency
WHO	World Health Organization
XRF	X-Ray Fluorescence

CHAPTER ONE

INTRODUCTION

1.1   Background of the Problem

Aquaculture is the predominant source of fish protein (Golden *et al.,* 2017) and currently contributes 47% of global fish production globally, with 5.8% annual growth registered during the period 2001– 2016 (Food and Agriculture Organization [FAO], 2018). The aquaculture industry is growing significantly in Africa (Waite *et al.,* 2014) and accounts for 17% of total fish production in the continent (FAO, 2018; Chan *et al.,* 2019). In Tanzania, inland aquaculture is dominated by small-scale fish farmers (average pond size of 150 m^2), producing fish mainly for both domestic consumption and export trade and contributing to poverty alleviation in the country (Watengere, 2010). Freshwater fish farming using small-sized earthen or concrete ponds is the most commonly practiced form of aquaculture system in Tanzania (Ministry of Livestock and Fisheries Development [MLFD], 2015). Most farmers in Tanzania own an average of one fish pond largely for subsistence purposes (Mdegela *et al.,* 2011). The country has over 20 000 freshwater fish ponds scattered across the mainland (Rukanda, 2018); the majorities are located in Ruvuma, Iringa, Mbeya and Kilimanjaro regions (MLFD, 2015). Like the rest of Sub-Saharan Africa, factors such as availability of water, the suitability of land for fish farming and economic potential in fish farming determine the distribution of fish ponds in Tanzania (United Republic of Tanzania [URT], 2015). Nile tilapia (*Oreochromis niloticus*) production is most preferred over African catfish (*Clarias gariepinus*) for its superior growth characteristics (De Graaf, 2004).

The opportunities afforded by fish farming to developing countries that suffer chronic food and nutritional insecurity are huge. This is still much the case in Africa. Fishers in these regions are mostly small-scale for subsistence and to satisfy the local markets and are heavily dependent on coastal and inland fisheries and so are particularly vulnerable to climate change. Subsistence fish farmers in these regions have no veterinarian supervision, regulation and consumer protection control (Rutaisire *et al.*, 2009) on potentially harmful substances and contaminants such as antibiotics, pesticides, chemicals and even heavy metals (Wamala *et al.*, 2018). Even so, small-scale fisheries and aquaculture provide jobs for approximately 12.5 million people directly engaged in fishing and another 34.5 million engaged in post-harvest activities (Intergovernmental Panel on Climate Change [IPCC], 2014). The high population densities in the tropics where the bulk of aquaculture production occurs make the sector especially vulnerable to climate change (De Silva & Soto, 2009).

Already, there is evidence that climate-related changes such as rising temperature and flooding of waterways affect the ecological functions of aquatic environments. Flooding of ponds can bring in nutrients from sewage or agricultural fertilizer, thus reduce dissolved oxygen levels and kill the fish as the result of algal blooms (Weatherdon *et al.*, 2016). When such changes happen in large water bodies such as oceans, most marine animals are forced to follow their ideal feeding and breeding habitat conditions (e.g. water temperature, oxygen concentration, carbon uptake and acidification, changes in salinity and freshwater content, etc.) (Cochrane *et al.*, 2009). Indeed, climate change and climate variability affect the suitability of some geographical locations for aquaculture systems. In farmed fish production, for example, changes in water temperature significantly impact yield and disease control, among other issues.

To manage and ensure yield, virtually all aquaculture systems use antibiotics to kill or inhibit bacteria growth. However, the accumulation of antibiotics in aquatic environments can cause ecological and public health effects (Kostich & Lazorchak, 2008). Fish farmers also use pesticides as indispensable inputs to treat and prevent diseases and to improve the water quality of their ponds. Pesticide residues pose a great health risk to the consumers and the environment (Burridge *et al.*, 2010). Other health risks such as heavy metal contamination come from agricultural runoff and other economic activities such as mining and industrial effluent (Mark *et al.*, 2019; Nnodum *et al.*, 2018; Choi *et al.*, 2016). Additionally, the use of an integrated fish farming system (i.e. using animal waste and excreta to supplement fish feed in ponds) in developing countries (Elsaidy *et al.*, 2015) could harbor pathogenic microorganisms (Mo *et al.*, 2018) and may threaten human health by causing food-borne illnesses. Thus, there is an urgent need for quantification of human exposure from aquaculture contaminants in low-income countries (LICs). This emphasizes the need for developing a resilient and robust aquaculture framework for ensuring sustainable fish production, safety, and nutritious farmed fish for public health protection. Therefore, a range of actions is required to make fisheries and aquaculture systems climate-smart considering the effect of climate change on food security, especially on the poor economies having the low capacity to adapt to change.

In fact, one of the biggest concerns is how the growing trade in fish and fisheries products also contributes to the increasing carbon dioxide emissions into the atmosphere. In addition to human factors, these emissions substantially change the aquatic ecosystems and affect the important services they provide for maintaining food security and livelihoods (FAO, 2016b). As such, the role of fisheries and aquaculture in supporting the reduction of emissions and natural removal of greenhouse gases cannot be underestimated (Nellemann *et al.*, 2009).

Of course, the impacts of climate change and adaptation options will vary by region. Local context-specific, climate-smart aquaculture strategies are required to guide the sector toward a sustainable

future. Therefore, the aquaculture sector needs to prioritize promoting the development of productive, climate-resilient, and low-carbon capture fisheries and aquaculture systems.

Globally, there have been efforts to innovate methods for intensifying and enhancing aquacultural productivity (Joffre *et al.,* 2017; Henriksson *et al.,* 2018) through environmentally friendly and sustainable climate-smart approaches (Kumar *et al.,* 2018) and the adoption of new technologies to improve husbandry and production processes (Kumar & Engle 2016; Kumar *et al.,* 2018). For Africa, innovative aquaculture production systems such as climate-smart ponds and utilization of insect proteins in fish feed formulation are some of the suggested context-specific approaches for increasing resource efficiency in the sector and reducing carbon imprint into the atmosphere (Tomberlin *et al.,* 2015).

Overall, aquaculture continues to experience increasing scarcity of critical inputs such as land, freshwater, and energy as well as poor value addition and low capacity in disease diagnostics and biosecurity, significantly impacting location, productivity, and scalability of the sector's production systems (FAO & World Bank, 2015). As such, local context-specific, climate-smart aquaculture strategies are required to enable the sector to prepare for a sustainable future, especially through the development of climate-resilient and low-carbon aquaculture systems (Gatonye *et al.,* 2020).

1.2   Statement of the Problem

There is little information on existing fish farming practices and their adequacy in ensuring sustainable fish production, safety, and quality of fish for human consumption in Tanzania. Researchers have reported the presence of pathogenic bacteria, parasites, and unacceptable levels of heavy metals in the aquatic environment and their associated health risks in some of the developing countries in Africa (Abdallah *et al*., 2013; Wamala *et al*., 2018). Tanzania is not unique in that respect and we suspect similar food concerns to happen in the aquaculture industry just as reported in crop farming and livestock sectors in the country (Kurwijila *et al*., 2006; Nonga *et al*., 2009).

Fish consumption is increasing rapidly in developing countries. Most people are consuming fish over red meat to avoid health problems associated with the continuous consumption of red meat (Shoko *et al*., 2011). Fish is a nutrient-rich and widely accepted animal source food for homestead consumption and sales. Fish farming is increasing taking an important place in the national economy as it enhances export revenue and creates employment opportunities. Considering the importance of aquaculture in improving food and nutrition security, it was important to assess the current management practices, analyse the possible contaminants, and design and develop a context-specific

climate-smart framework that can ensure sustainable fish production, safety, and nutrition of farmed fish for improving food security and protection of public health.

This study, therefore, assessed the adequacy of existing aquaculture practices, profile bacteria pathogens, and parasites that cause diseases in farmed fish, and quantitatively evaluated the levels of heavy metals in aquatic ecosystem. The information was used to design a context-specific climate-smart aquaculture framework that is culturally acceptable and economically viable for increasing fish production and ensuring food safety and nutrition in Tanzania.

1.3   Rationale of the Study

As, it is the predominant source of fish protein, aquaculture accounts for 47% of global fish production. Besides, aquaculture in Tanzania particularly mainmland is reported in contributing to poverty alleviation and for subsistence purposes. However, in Tanzania the aquaculture industry is free from veterinarian supervision, regulation and consumer protection control (Rutaisire *et al.,* 2009) subsequently, leading consumers to the exposure of   potentially harmful substances and contaminants such as antibiotics, pesticides, chemicals and even heavy metals (Wamala *et al.,* 2018). Furthermore, the industry is heavily dependent on coastal and inland fisheries and so are particularly vulnerable to climate change. To manage and ensure yield, the industry have been using antibiotic to kill or inhibit bacteria, however, this has been accounted for as the main source of accumulation of antibiotics in aquatic environments, which with pesticide residues pose a great health risk to the consumers and the environment (Burridge *et al.,* 2010). Thus, there is an urgent need for quantification of human exposure from aquaculture contaminants. This emphasizes the need for developing a resilient and robust aquaculture framework for ensuring sustainable fish production, safety, and nutritious farmed fish for public health protection. Therefore, a range of actions is required to make fisheries and aquaculture systems climate-smart considering the effect of climate change on food security, especially on the poor economies having the low capacity to adapt to change.

1.4   Research Objectives

4.1.1   General Objective

To design a context-specific climate-smart aquaculture framework for improving food security in Tanzania.

4.1.2   Specific Objectives

(i)   To examine the quality of existing aquaculture practices in Tanzania.

(ii)   To profile bacteria pathogens and parasites which cause diseases in farmed fish in Tanzania.

(iii)   To determine a distribution pattern and bioaccumulation of heavy metals in aquaculture.

(iv)   To develop a context-specific climate-smart aquaculture framework for proper fish farming.

1.5   Research Hypotheses

(i)   Existing aquaculture practices do not favor sustainable fish production, safety and nutrition of farmed fish in Tanzania.

(ii)   A context-specific climate-smart aquaculture framework has the potential to reduce greenhouse gas emission, increase productivity and improve food security.

1.6   Significance of the Study

Evaluation of existing aquaculture practices and examining the contaminants present in the aquaculture ecosystem served as a baseline for developing a resilient climate-smart aquaculture framework to ensure sustainable food fish production, safety, and nutrition in Tanzania. Furthermore, the framework will enable fish farmers to achieve the united nations sustainable development goals as it provides the opportunity to improve their income and alleviate hunger and poverty. Besides, the framework will sustainably improve fish production; provide resilient stability to climate variability, and reduce the emission of greenhouse gases. The outcome of this research has been translated into effective communication tools and disseminating those to a particular audience. These include peer-reviewed papers and guidelines. Also; the finding will be shared with policymakers and various stakeholders including the ministry of agriculture, livestock, and fisheries for appropriate actions.

1.7   Delineation of the Study

This research was conducted to establish the quality of existing aquaculture management practices as well as evaluation of bacteria, parasite, and heavy metals present in the aquaculture ecosystem. The outcome helped to develop a context-specific climate-smart framework to improve   fish production and ensuring the health and safety of the consumers. The framework includes designing and constructing an an environmentally-friendly fish pond that has the potential to increase   fish production with less labour and land among the rural communities while improving freshwater utilization, ensuring fish quality and safety by eliminating water runoff and other contaminants from

entering the pond and minimizing aquaculture effluent discharge into the environment. The framework also includes the formulation of inexpensive and relatively abundant nutrient-rich insect based fish feed for the purpose of improving fish production and reducing green house gas emission to the environment.

Although results from this study are encouraging, the study was conducted in only two regions in the country which could limit the generalization of the findings. Moreover, the study involves a small sample size that resulted in wider confidence intervals in some of the associations among variables which may reduce the precision of the study due to distribution effect. However, the study is still useful as it evaluated the quality of current aquaculture management practices and came up with the sustainable solution which could be adopted national wide by fish farmers and contribute toward improving food security in the country.

The study faced biases on data collection where some participants did not know bacteria and parasites diseases sypmptoms hence it was difficults for them to answer weather they experience disease occurance. Hence, to avoid this biasness, samples were collected and fish diseases were examined in the laboratory. Additionally, potential confounders between dependent and independent variables during data analysis were identified including farm characteristics, reported diseases and treatment used for desieases. These confounders were controlled by running multivariate analysis to reduce their effects on associations among variables.

CHAPTER TWO

LITERATURE REVIEW

2.1   An Overview

Globally, aquaculture has become the predominant source of fish protein (Golden *et al.,* 2017), doubling its production every decade for the past 50 years (Bostock & Seixas, 2015). In Africa, aquaculture has contributed immensely to the total amount of fish produced over the past decade, supporting rising fish demand and improving incomes, food and nutrition security of the growing populations in the region (Mwima *et al.,* 2012). The level of fish consumption in Africa during 2015–2017 was 9.9 kg but it's projected to decline to 9.6 kg by 2027 and ultimately to 7.7 kg by 2050 despite the steady population growth (Organization for Economic Co-operation and Development [OECD], 2018; Chan *et al.,* 2019) and increasing demand for fish (Kobayashi *et al.,* 2015). East African region, second only to Southern Asia, had the highest food insecure population in 2016, compared to the rest of the world (FAO 2018d; African Union Commission- New Partnership for Africa's Development [AUC-NEPAD], 2014a). As such, Africa is projected to continue heavily relying on imported frozen fish due to its ease of availability, steady supply, and price to meet the demand gap and its nutritional needs (OECD 2018; Tran *et al.,* 2019). As revealed by IMPACT Model projections, Africa's total fish output will be significantly low in the next decade because of the expected slow growth of capture fisheries and aquaculture in the region (Chan *et al.,* 2019).

More than 200 million people in Africa are reportedly regular fish consumers (Béné & Heck, 2005), especially tilapia―the most commonly cultured fish species in the region (De San, 2013). The top aquaculture producers in Africa are Egypt (~1.37 million tonnes) and Nigeria (~ 306 727 tonnes) (FAO, 2018a), mostly driven by increased investment by the private and public sector interventions as the continent strives to achieve food and nutrition secure nations (Mwima *et al.,* 2012). The FAO estimates that fish accounts for more than 20% of animal protein supplies in about 20 African countries (FAO, 2017a). The sector is now rapidly responding to this market demand for fish with an average annual growth rate of 21% in Sub-Saharan Africa alone (Satia, 2017), especially in Ghana, Kenya, Nigeria, South Africa, Uganda and Zambia (Asiedu *et al.*, 2015; Kaminski *et al.*, 2017; Satia, 2017).

Climate change and climate variability have the greatest impact on fisheries and aquaculture sector in Africa, especially on productivity and sustainability. Unfortunately, since the impact of climate change varies by region; it could likely worsen food insecurity in Africa as a whole by disrupting

the distribution of available resources in the continent's aquatic environments (Challinor *et al.*, 2007). It's projected that by 2050 if the current production and consumption trends in Africa continue, more than half of the fish consumed in Africa will be imported (Chan *et al.*, 2019). Hence, sustainable climate-smart growth of the aquaculture sector in Africa is required to meet the demand of an increasingly growing population.

Around 1.74 million tonnes of global aquaculture production comes from Africa, contributing about 2% of total global production. Nile perch (43.6%), African catfish (11.9%), and common carp (10.5%) are some of the most commonly consumed fish species in Africa (James, 2018). However, the growth and development of the aquaculture sector in Africa still require more effort to ensure the consistent production of safe and nutritious fish as well as smart aquaculture practices for a sustainable future (James, 2018). Indeed, a recent study on impacts of climate variability and adaptation options in Africa found that Egypt and Nigeria, two countries with high fish consumption per capita, were relatively vulnerable to the effects of climate change including temperature (Adeleke *et al.,* 2018).

Fish produced can be for domestic consumption, export trade, or both. Inland aquaculture is dominated by small-scale fish farmers, mainly for subsistence and to satisfy the growing local markets. Subsequently, a supply response has been observed in some countries such as Zambia whereby medium to large scale farms have begun to upgrade operations to increase their aquaculture output (Tweddle *et al.*, 2015; Kaminski *et al.*, 2017). Indeed, Zambia has recently emerged as the largest producer of farmed fish in the Southern African Development Community (SADC) region (Genschick *et al.*, 2017), while Kenya is the fourth in the whole of Africa, having grown exponentially to peak its fish production at 24 096 tonnes in 2014, driven largely by an ambitious Economic Stimulus Programme (ESP) aquaculture subsidy programme commissioned from 2009– 2013 (Kenya National Bureau of Statistics [KNBS], 2018; Obiero *et al.,* 2019c). The ESP focused on critical aquacultural infrastructures such as pond design and construction, fish feeds, fingerlings supply, as well as post-harvest management and human resource institution to assist fish farmers (Obiero *et al.,* 2019c).

Despite these incredible interventions to prioritize the development of the sector in Africa, subsistence fish farming has limited supervision from veterinarians and extension officers in the region. Sadly, consumer protection control and other regulations and legislations are not fully enforced in the aquaculture sector in some of the African countries (Rutaisire *et al.*, 2009) which increases the likelihood of contamination from harmful compounds such as antibiotics, heavy metals and pesticides from anthropogenic sources (Wamala *et al.*, 2018; Kostich & Lazorchak, 2008; Burridge *et al.*, 2010; Mark *et al.*, 2019; Nnodum *et al.*, 2018). Country-specific monitoring

programmes are in place to monitor such contamination to ensure food safety. However, such monitoring cannot completely prevent or eliminate the supply of contaminated aquaculture products to consumers.

Integrated fish farming, that is, using animal waste and excreta to supplement fish feed in ponds, is still the most commonly practiced form of aquaculture system (Elsaidy *et al.*, 2015). Of course, this disregards the importance of human health protection by intentionally introducing pathogenic microorganisms into the aquaculture facility which could lead to food-borne illnesses. Therefore, there is a need to establish a more suitable aquaculture system to ensure safety for the protection of public health. A range of actions is required to make the aquaculture system climate-smart considering the effect of climate change on food security, especially on the African economies that often have a low capacity to adapt to change.

More importantly, one of the biggest concerns is how climate-induced changes in productivity and availability of aquatic resources have led to the expansion of capture fisheries and commercial aquaculture to meet the growing market demand for fish and contributed to the increasing carbon imprint into the atmosphere. As such, a shift to local context-specific, climate-smart aquaculture strategies is required to enable the sector to prepare for a sustainable future, especially through the development of climate-resilient and low-carbon capture fisheries and aquaculture systems. Therefore, this review highlights the prospects for climate-smart aquaculture development in Africa considering climate change impacts on aquatic systems and more broadly, the potential reduction of vulnerability within the communities that depend on fisheries and aquaculture.

2.2 Climate-related Changes that Affect Ecological Functions

Climate-related changes include physical and chemical processes known to increase greenhouse gas emissions, much of which is absorbed by aquatic systems, leading to substantial changes in aquatic ecosystems (FAO, 2016b). The devastating effects of climate change and climate variability on aquatic environments include changes in the abundance and distribution of fisheries resources and the overall suitability of some regions for aquaculture systems (FAO, 2016a). This impacts their ability to provide food security and livelihoods to populations dependent on aquaculture (FAO, 2016b; Kareko *et al.,* 2011). Effects of climate change on aquaculture systems include changes in salinity and freshwater content, oxygen concentration, water acidification and temperature, storm systems, rainfall, and river flows (Cochrane *et al*., 2009; FAO, 2016b). Oceans and coastal areas are particularly vulnerable to these changes (FAO, 2016b). For example, changes in carbon chemistry can affect shell development in marine shellfish whereas temperature changes can increase the sensitivity of some species to pathogens (FAO, 2016b).

Productivity potential and species distribution in aquaculture systems dependent on inland water bodies such as dams, rivers, and lakes may be affected by extreme weather events including changes in air and water temperatures as well as levels of precipitation (International Plant Protection Convention [IPPC], 2014; FAO, 2016b). Climate change may also have significant stress on post-harvest activities. For instance, the availability of adequate water for processing may be challenging, especially if the same water source is required for other farming practices such as irrigation. Of course, climate-induced changes often occur simultaneously and their effects are cumulative, thus their impact on natural resources, food security, and social stability is huge (IPPC, 2014).

2.3   The growing Demand for Fish and other Aquatic Products in Africa

Worldwide, aquaculture as well as marine and freshwater-capture fisheries have contributed to the growth of fish production to meet the global demand, rising from 19 million metric tonnes (MT) in 1950 to 171 million MT in 2016 (FAO, 2018b). Fish is the most accessible and affordable source of animal protein, especially for 'poor' socioeconomic classes (Béné *et al.,* 2015). Fish production is crucial for over 3 billion people in developing countries since fish contribute 17% of animal protein and 7% of all proteins consumed (FAO, 2018b).

In Africa, many factors drive fish preferences and consumption including affordability (average of US$ 2/kg), rising population growth, increasing income levels, accessibility, as well as awareness of health benefits and the nutritional value of fish (Darko *et al.,* 2016; Githukia *et al.,* 2014). Nutritionally, fish provides docosahexaenoic and eicosapentaenoic omega-3 fatty acids, high-quality essential amino acids, minerals and vitamins, which are necessary for improved health (Golden *et al.,* 2016; Beveridge *et al.,* 2013; Kris-Etherton *et al*., 2002; Béné *et al.,* 2015). As such, fish is a high-value food that supports the nutritional wellbeing of poor communities (FAO, 2017b; Golden *et al.,* 2016; Beveridge *et al.,* 2013; Béné *et al.,* 2015) considering that several African countries have significant numbers of undernourished and malnourished populations (FAO, 2018c; FAO, 2018d). For instance, the high malnutrition incidences in the East African region, especially in Ethiopia, Kenya, Tanzania and Uganda, has been shown to correspond to a significantly lower quantity of fish consumed/capita (average of 5.3 kg compared to the rest of Africa (10.1 kg), and global level of 19.8 kg (Obiero *et al.,* 2019; Cai & Leung, 2017).

Roughly 200 million people in Africa consume fish as the main animal protein source and micronutrition (AUC-NEPAD 2014b). Africa's population is expected to double by 2050 (United Nations Department of Economic and Social Affairs [UN-DESA], 2017), but unfortunately, the continent's contribution to the amount of fish produced, consumed, and traded globally is so small. In 2016, for example, the aquaculture sector only contributed about 2.5% of global fish production

(FAO, 2018b). Thus, with overfishing and overexploitation in the capture fisheries sector (FAO, 2018b), aquaculture is expected to meet the increased fish demand in Africa (Chan *et al*., 2019), and continue supplying animal protein to the poor and food-insecure populations (Kobayashi *et al.,* 2015; Golden *et al.,* 2017).

Rising urbanization, increased incomes, and awareness of the health benefits associated with consuming fish have contributed largely to the increased global fish consumption rates (Anderson *et al.,* 2017). Capture fisheries and aquaculture resources have improved the economic security of farmers through domestic and international trade of wild and farmed fish, employment, and other livelihood support services (De Graaf & Garibaldi, 2014; Cai *et al.,* 2019). Global capture fisheries production peaked in 1996 at around 96 million MT, whereas aquaculture production has continued to grow for the past 50 years to produce 80 million MT of fish in 2016 (FAO, 2018b). As such, aquaculture alone is on-trend to produce 195 MT of fish by 2027 and contribute immensely to the future expansion of fish as food (Economic Co-operation Development- Food and Agriculture Organization [OECD-FAO], 2018). No doubt, the increasing demand for fish in Africa and the ongoing transformations in fish supply have led to the gradual growth and development of aquaculture in the continent (Kobayashi *et al.,* 2015), bringing an estimated US$ 3 billion annually (De Graaf & Garibaldi, 2014). The New Partnership for Africa's Development (NEPAD) estimates that about 1.6 million tonnes of fishery production in Africa come from aquaculture (African Union Commission- New Partnership for Africa's Development [AUC-NEPAD], 2014b). The sector also employs about 12.3 million people in the areas of fishing, processing, equipment manufacturing, and fish farming (De Graaf & Garibaldi, 2014), generating about 1.26% of gross domestic product (GDP) (AUC-NEPAD 2014b).

The growth of fish production in Africa is not immune to problems as competition for land, water, energy, and feed resources intensify. Combined with the potential impacts of climate change on ecosystems, the aquaculture sector faces significant challenges as it tries to satisfy the gap between capture fisheries.

2.4   Aquaculture Practices in Africa and its Challenges

Aquaculture in Africa is dominated by men. This is probably due to strong cultural norms that explicitly define men as heads of households and women as caretakers of chores in the homestead (Akrofi, 2002). However, women's roles in aquaculture production activities are significant, ranging from processing and transportation to marketing and sale (Kruijssen *et al.,* 2018; Akrofi, 2002).

Aquaculture accounts for 17% of total fish production in Africa (Chan *et al.,* 2019; FAO, 2018b; Obiero *et al.,* 2019a). Fish and fisheries products in Africa mostly come from two production techniques, namely aquaculture and wild-catch. In general, there are three types of aquaculture practiced around the world, namely, land-based commercial, water-based commercial, and small-scale production. These production techniques use different sets of inputs. For instance, the aquaculture sectors use five types of inputs, namely, seed, feed, labor, fuel, and sector-specific inputs such as capital investment in the facility. On the other hand, wild-catch (capture fisheries) sectors only use labor, fuel, and other sector-specific inputs. Regardless of the production techniques, there is evidence that climate-related changes such as rising temperature and flooding of waterways affect the ecological functions of the aquatic environments and impact overall yield. Flooding, for instance, may introduce pollutants from sewage into the ponds, thereby reducing dissolved oxygen levels and destroying the fish as a result of algal bloom (Weatherdon *et al.*, 2016).

Generally, the aquaculture sector in Africa has expanded its production capacity to include other mariculture species (Oyinlola *et al.,* 2018) through innovation and intensification of production systems (Joffre *et al.,* 2017), adoption of new technologies (Kumar *et al.,* 2018) and improvement in resource efficiency and utilization (Waite *et al.,* 2014). Indeed, studies indicate that the aquaculture sector has generally benefited from the adoption of new technologies in aquaculture production, breeding systems, nutrition and feed formulations, genetic selection programs, labor-saving equipment, development of vaccines, investment in management practices as well as improved regulatory frameworks and control (Joffre *et al.,* 2017; Kumar *et al.,* 2018). Despite the recent growth in Africa's aquaculture sector, the industry isn't technologically advanced and is largely constrained by lack of good-quality seed and feed, poor market access and value addition, lack of credit/capital, insufficient extension services, and programs, poor management systems, low capacity in disease diagnostics, training and biosecurity, and disadvantageous competition from cheaper imported fish products from established markets such as China (Mwima *et al.,* 2012; Kaminski *et al.,* 2017), thus its full potential in contributing to the sustainable food supply in the region is unknown (Brummett *et al.,* 2008; Obiero *et al.,* 2019c).

Like in other animal production systems, feed is the most expensive input in aquaculture. In Africa, most farmers prefer to use farm-made feeds formulated with grains (Amankwah *et al.,* 2016) either alone or with animal waste and excreta as a supplemental nutrient source, mainly to benefit from the synergistic effects of inter-related farm activities and off-set high production cost (Petersen *et al.*, 2002; Elsaidy *et al.*, 2015). Commercially manufactured feeds, either locally-made or imported, are often used by larger aquaculture operators with access to sufficient credit and markets, and often can tolerate risks associated with declining prices. The fish feeds are mostly sourced from privately-

or government-owned hatcheries (Opiyo *et al.*, 2018) and small-scale semi-commercial feed manufacturers (Obiero *et al.,* 2019b). However, the cost of high quality imported fish feeds is often beyond the budget of many small-scale fish farmers, to the extent that some of the farmers would switch to risk management strategy to stabilize their incomes or abandon production altogether to minimize losses when production cost increases or competition with increasing fish imports become unsustainable.

Unfortunately, the fish feed sector has an unreliable supply chain, lacks proper quality monitoring and standards management strategy, compromising on production performance, consistency, and food safety (Obiero *et al.,* 2019b). Nevertheless, several other factors play an important role in determining the actual production capacity of a fish farm and its sustainability. These include but are not limited to (a) technological shift that reduces the environmental impacts of aquaculture (Troell *et al.,* 2009); (b) the diversification strategies to maximize on space and input (Rapsomanikis, 2015); (c) quality of governance and access to advisory and extension support services (Kuehne *et al.,* 2017); and (d) promotion and adoption of sustainable aquaculture practices (Engle, 2017; Kumar *et al.,* 2018). Some of these are either unavailable or inaccessible to many small- and large-scale fish producers and traders in Africa, suggesting that investments by the private sectors are critical to sustain innovation, increase growth, improve production efficiency, and reduce production costs to stay in business.

2.4.1   Contaminants Present in the Aquaculture Environment

(i)   Antibiotics and Antibiotic Residues

Antibiotics can either be natural or synthetic compounds intending to kill or inhibit bacteria growth, in some cases, antibiotics are also used as a growth promoter (Holmstrom *et al*., 2003: Cabello *et al*., 2013). Oxytetracycline, chloramphenicol and oxolinic acid, sarafloxacin, and sulfadimidine are among the most common antibiotics reported being used in aquaculture (Liu *et al*., 2017). Despite the intensive use of antibiotics in the aquatic environment, limited data are available on the specific types and amount of antibiotics uses in aquaculture in African countries. Majority of documented data are from developed countries while aquaculture production largely takes place in developing countries where regulatory guideline are limited or barely exist (Sapkota *et al*., 2008). Even to those few countries with antibiotics usage data, the same antibiotics are often marketed under a different name and in some cases, the active ingredients are not listed (Sapkota *et al*., 2008). This makes it difficult in keeping the uniform record and to compare antibiotics usage from one country to another. Furthermore, there is a lack of information and education with regard to the antibiotics uses which

could potentially result in abuse and/or misuse that could end up unreported (Holmstrom *et al.*, 2003). The FAO has documented the list of antibiotics that are potentially used in aquaculture throughout the world; however, the documented data lack specific and actual antibiotics usage patterns (Sapkota *et al.*, 2008). The absence of any data for some countries particularly in African countries is not necessarily indicative of a lack of antibiotics usage, but rather might be due to the lack of information available in those countries.

Among the negative impact of antibiotics use in aquaculture is the accumulation of antibiotics residues in aquaculture products, ponds, sediments and surrounding environment that are impacted by aquaculture facilities (Dalsgaard *et al.*, 2000; Holmstrom *et al.*, 2003; Hoa *et al.*, 2011). Antibiotic residues of trimethoprim, sulfamethoxazole, norfloxacin, and oxolinic acid exceeding the minimum allowed level have been reported from aquatic environment in African countries (Holmstrom *et al.*, 2003). Other researchers have also reported the presence of a substantial concentration of oxalic acid in fish plasma, liver and muscle tissue (Fry *et al.*, 2016). Accumulation of antibiotic residues in fish and aquatic environments can cause ecological and public health effects. Low-level exposures to these residues present in fish food are not likely to cause acute toxic effects among the consumers; however, chronic effects are expected (Kostich *et al.*, 2008). These chemicals have the potential to gradually accumulate in the body and cause a certain organ or system malfunction. Many studies documented the effect of accumulating these chemical residues in the human body and among the reported health problems are cancer, immunological and nerve problem (Kostich *et al.*, 2008).

Many farmers from developing countries who routinely come into contact with antibiotics during its application lack appropriate protective gear and are also unaware of the potential health risk associated with antibiotics exposure. Thus, this is a significant risk of exposure through inhalation and skin contact (Holmstrom *et al.*, 2003). An antibiotic such as chloramphenicol which is a grouped as a potential human carcinogen has been linked with increased risk of aplastic anemia and leukemia in humans. Therefore, establishing the level of antibiotic residue in aquaculture in all emerging developing countries is of paramount importance. The information will help to determine antibiotic residue prevalence and develop appropriate mitigation strategies.

(ii)   Pesticides and Pesticide Residues

The use of pesticides in aquaculture has become indispensable inputs to treat and prevent diseases and to improve water quality. Its use has contributed to the productivity and growth of the aquaculture sector but has also attracted criticism as the result of potential food safety   and health hazard concerns upon consumption of fish with higher pesticides residue levels (Sapkota *et*

al., 2008). Endrin, Dichlorodiphenyltrichloroethane, Polychlorinated biphenyls and Pentachlorobenzene are among the pesticide residues reported in African countries (Omwenga *et al*., 2016). Researchers have reported that intensive use of pesticides in aquaculture has potential effects on the health of fish, consumers as well as the environment (Burridge *et al*., 2010). Accumulation of pesticides in the human body has been reported to cause mutagenic and carcinogenic effects. Human exposure to pesticides can occur through direct consumption of contaminated fish, consumption of food crops irrigated with contaminated wastewater from the fish pond or by drinking surface/groundwater contaminated with pesticides from aquaculture facilities. Some countries, particularly from the developed world including Canada, United State of America and some countries in the European Union do require testing for pesticides and other chemicals in imported fish. Aquaculture products with the higher level of pesticides exceeding the allowable concentration have been reported to be rejected by these countries (Windle *et al*., 2008). However, in most African countries this type of testing is limited to only imported aquatic products. Additionally, the majority of developing countries, particularly in sub-Saharan Africa, produce aquaculture products for local consumption which does not go through any quality control and assurance (Windle *et al*., 2008). The current information on the use of pesticides applied by farmers from African countries is very limited or even unavailable for most aquaculture producing countries (Sapkota *et al*., 2008). Research is needed to address the gap and provide detailed information on the use of pesticides in developing countries. The information is of crucial importance for evaluating the potential risks for aquatic products, human health and for the environment in these countries.

(iii)   Heavy Metals

Heavy metals are naturally present in the environment and make their way to the aquatic environments through various processes including, agricultural runoff, mining and industrial process (Jaishankar *et al*., 2014). Heavy metals such as mercury, lead, cadmium, and mercury are among the common reported metals in aquaculture industry in African countries (Farombi *et al*., 2007). They are also of the highest food safety and human health concern and much attention have been focused on these metals (Jaishankar *et al*., 2014). Neurotoxic, reproductive, carcinogenic and immune system effects are among the adverse human health effect associated with exposure to heavy metals as the result of farmed fish consumption (Mahaffey, 2004). Another major cause of heavy metal contamination in fish farming is Polychlorinated biphenyls (PCBs). They are industrial pollutants that find their way into freshwater and are then absorbed by aquatic animals (Mahaffey, 2004). The contamination by these heavy metals is more pronounced during the rainy season where heavy rain and flooding wash away industrial wastes and other contaminants which eventually end up in the fish ponds. High level of arsenic obtained in farm raised salmon samples was reported

(Mahaffey, 2004). A similar finding was reported by other researchers (Farombi *et al*., 2007). In another study, authors evaluated the concentration of mercury in farmed fish and the results indicated a high concentration of methyl mercury (Choi *et al.,* 2016).

Fish consumers from African countries are more susceptible to heavy metal contamination because the majority of fish farmers are operated in a subsistence scale and farming practice is characterized by lack of appropriate knowledge, veterinarian supervision, regulation and consumer protection control (Shoko, 2009). There have been little or no surveys on the presence of heavy metals and associate in most African countries; therefore studies are required to address the knowledge gap and to clearly determine the contamination with heavy metals and the levels of residue present at the local and national scale.

(iv)   Wastewater and Excreta

 Integrated fish farming systems are still a common practice in African countries whereby animal waste and excreta are used. The waste is directly consumed by fish and in some case provides nutrients for the growth of photosynthetic organisms which then become a source of food for the fish (Elsaidy *et al*., 2015). This kind of farming system has been reported to provide high fish yields at a very low cost since little or no addition of formulated feeds are needed (Elsaidy *et al*., 2015). However, this farming system could have negative impacts on fish safety and consumer health. Fish raised through this system could harbor pathogenic microorganisms (Fry *et al*., 2016). Microorganisms such as bacteria, virus, fungi, and parasites have been reported in the aquaculture environment fed with wastewater (Mo *et al*., 2018). In addition, another researcher found antibiotic-resistant bacteria including *Enterococcus* spp and *Acinetobacter* spp in samples isolated from an integrated fish farming system (Mo *et al*., 2018). *Enterococcus* spp is also a human pathogen, so if they are resistant and consumer get infected with it, then they cannot be treated with antibiotics. All these contaminants can cause health problems in humans upon consuming contaminated fish, yet limited epidemiologic research has investigated the degree of contamination from integrated fish farming in African countries and the specific disease outcome associated with fish fed with excreta and wastewater.

2.5   The role of Aquaculture in Greenhouse Gas Emissions

The major greenhouse gases (GHGs) associated with aquaculture production are: (a) Nitrous oxide (N_2O), emitted as a result of the microbial nitrification and denitrification of nitrogenous compounds in the ponds (e.g. fertilizers, manures, uneaten feed and excreted N), (b) Carbon dioxide (CO_2), from energy and fuel consumption associated with farm management such as pumping water, lighting

and powering vehicles, (c) Methane (CH_4), arising during fish farm waste management, and (d) Fluorinated gases (F-gases) leaking from cooling systems used on and off the farm (MacLeod *et al.,* 2019).

The application of feeds in aquaculture is the leading contributing factor in greenhouse gases (GHG) emission in the sector (Naylor *et al.*, 2000). Aquafeeds increase nutrient loadings in the water bodies and pond sediments in feed-based aquaculture production systems (Boyd *et al.,* 2010; Chatvijitkul *et al.,* 2017). Approximately 75% of the nitrogen consumed by fish from the feeds is excreted into the water as ammonia, while the remainder is converted into biomass (Hu *et al.,* 2012). Additionally, the carbon from the feeds can be transformed into carbon dioxide and methane by animals and microbes in the water (Boyd & Tucker, 2014) while a great amount of unconsumed feed become deposited in the pond sediments together with feaces (Boyd *et al.,* 2010), where they continue to provide carbon for submerged macrophytes (Yuan *et al.,* 2019). Approximately 39.9 million tons of aquafeeds were used in global aquaculture in 2016 alone, leading to about 10.9 teragram carbon and 1.82 teragrams nitrogen discharged into the environment (Alltech, 2017).

Today, it is estimated that >40% of worldwide aquaculture production is carried out in earthen ponds around the world (Yuan *et al.,* 2019), contributing about 80.3% of the total methane emitted into the environment (Hu *et al.,* 2014). Since intensified systems with continuous aeration reportedly have the least emissions (Hu *et al.,* 2014), global adoption of aerated systems has been proposed to mitigate the significant rises in methane emissions from aquaculture sources (Yuan *et al.,* 2019). It has also been suggested that pond sediments can sequestrate carbon and contribute to mitigation (Boyd *et al.,* 2010), which had previously been shown to complicate quantification of greenhouse gas emission (Verdegem & Bosma, 2009). However, later studies such as the Sustaining Ethical Aquaculture Trade (SEAT) project determined that it was impossible to quantify the extent by which pond sediments can act as carbon sinks due to uncertainties over the sequestration rates and stability of the carbon storage (Henriksson *et al.,* 2014).

Fertilizers are used in inland aquaculture systems to stimulate phytoplankton production for supplemental nutrients for the fish (Green, 2015). However, such anthropogenic use of fertilizers has the potential to significantly increase methane and nitrous oxide emissions from aquaculture systems into the environment. For instance, in 2008 alone, a global nitrous oxide emission from the aquaculture sector was estimated as 0.08 teragram (Williams & Crutzen, 2010). Using the nitrous oxide emissions factor of influent nitrogen (EF_N = 1.80%) in sludge and wastewater treatment processes (Ahn *et al.,* 2010), nitrous oxide emission was later projected to increase to 0.60 teragrams by 2030 and account for 5.72% of global anthropogenic nitrous oxide emissions (Hu *et al.,* 2012).

Overall, approximately 0.45% of global anthropogenic greenhouse gas emissions in 2013 came from aquaculture sources, which is similar to the emission intensity from sheep production in the same period (MacLeod *et al.,* 2019). The modest emissions are largely because fish have a low feed conversion rate compared to terrestrial animals (Gjedrem *et al.,* 2012), do not produce methane via enteric fermentation (Hu *et al.,* 2012; MacLeod *et al.,* 2019), and lastly, their high fertility rate reduces breeding overhead (MacLeod *et al.,* 2019). These are three key determinants of fish emission intensity, considering that the greatest greenhouse gases in aquaculture come from aquafeeds (MacLeod *et al.,* 2019). Furthermore, unlike terrestrial mammals, fish (both finfish and shellfish) require less energy for physiological functions and excrete ammonia directly (MacLeod *et al.,* 2019). In aquaculture, shrimps and prawns have the highest emission intensity because they require energy usage for water aeration through the systems. On the other hand, bivalves have the lowest emission intensity since they source food from their environment and thus have no synthetic feed-related emissions (MacLeod *et al.,* 2019).

However, despite the low emissions from the sector, the contribution of aquaculture to the increasing global carbon footprint cannot be ignored. For example, carbon dioxide emission from energy usage in the post-harvesting and value addition activities such as drying, smoking, cold storage, and transportation, which are not included in the 0.45%, also has significant global warming potential. Additionally, aquafeed production use machines and equipment that require energy to grind and mix the raw materials or make and dry the pellets. The total energy used depends on local energy source and production efficiencies. The feed materials can be marine or terrestrial in origin and are often formulated to meet the nutritional needs of the fish depending on species and age. Poor feed quality may reduce fish performance and increase greenhouse gas emissions. Of course, the feed must eventually be transported to the farms for use, which requires energy utilization. Therefore, operations and processes that require high amounts of fuel and energy are among the highest greenhouse gas emitters.

2.6   Climate Change and its Potential Impacts on Aquaculture Systems in Africa

Food production systems are especially vulnerable to the impacts of climate change and associated risks (Handisyde *et al.,* 2017). As such, there's an urgent need to effectively respond to the threat of climate change, through mitigation and progressive adaptation strategies. On a global scale, climate change effects on the aquaculture and fisheries sector will lead to significant changes in the availability and trade of fish products, and for countries whose economies rely on this sector, create other geopolitical tensions (Barange *et al.,* 2018). In general, climate change is expected to affect fish and ecosystems, livelihoods, trade and economies (Allison *et al.,* 2009; Badjeck *et al.,* 2010;

Daw *et al.,* 2008; Brander, 2010). According to greenhouse gas emission scenario RCP 8.5, global marine catch potential is projected to decrease by 7.0 – 12.1% by 2050, resulting in shifts in the availability and distribution of species (Barange *et al.,* 2018). As such, adaptations to climate change, including institutional adaptations, are necessary and must consider the multifaceted nature of aquaculture and fisheries.

Freshwater is a valuable resource and is used in many sectors of human life ranging from human consumption to agriculture, aquaculture and recreation. Unfortunately, climate change is projected to result in a significant reduction in freshwater resources (Jimenez *et al.,* 2014). Competition for scarce freshwater resources seriously affects the sustainability of inland aquaculture and fisheries and adds stress to the already resource-stretched sector (Katikiro & Macusi, 2012). Today, Morocco is one of the African countries currently facing high stresses and is projected to become even dire in the future, while Papua New Guinea, the Congo, the Central African Republic and Gabon are under low stress at present and are projected to remain as such in the future (Barange *et al.,* 2018).

The physical and ecological impacts of climate change on global aquaculture and capture fisheries is well documented in the literature (Allison *et al.,* 2005; Allison *et al.,* 2007; Allison *et al.,* 2009; Barange & Perry, 2009; Daw *et al.,* 2008; Handisyde *et al.,* 2006). In general, the implications of climate change on aquaculture systems in individual countries and communities depend on their adaptive capacity (Aswani *et al.,* 2018). Climate change impacts on aquaculture may include losses of production, infrastructure, fish markets, or decreased safety of fishers at sea (Katikiro & Macusi, 2012; Barange *et al.,* 2018). For instance, the impact of precipitation on inland freshwater ecosystems has a significant effect on the supply and quality of freshwater lakes and rivers that support inland aquaculture and fisheries (Barange *et al.,* 2018).

Aquaculture systems are especially vulnerable to rising global temperatures, particularly production infrastructures in the tropics, where population densities are high (De Silva & Soto, 2009). In the last decade alone, global warming has produced weather events that were exceedingly rapid and extreme and differed from those of the past, adding more stress to the environment and aquatic systems and leading to changes to relative abundance, distribution, and productivity of fish in the water bodies (Cheung *et al.,* 2010). Changes in sea temperature are ultimately responsible for other impacts such as acidification, sea-level rise, increased frequency and intensity of storms, extreme winds, flooding, and erosion (De Silva & Sotto, 2009; Barange *et al.,* 2018), which may radically change the whole ecosystems and hence directly impact aquaculture-dependent communities and damage aquaculture infrastructure (Allison *et al.,* 2005). Furthermore, any losses to important coastal habitats such as the mangroves ecosystem which support numerous fish species (IPCC,

2007) as a result of climate change could lead to disruption of fishing patterns and behavior (Katikiro & Macusi, 2012).

In Africa, both marine and inland water bodies such as wetlands, floodplains, lakes, and rivers are all susceptible to climate change effects, especially precipitation and rising temperature (FAO, 2010a; Settele *et al.,* 2014). Of course, increase precipitation lead to the expansion of fish habitats, and fishers would be expected to adapt to new systems and fishing range to maximize success (Barange *et al.,* 2018). However, increased precipitation may also lead to extreme events such as floods which may introduce contaminants and pathogens via surface runoffs into ponds and waterways. Low precipitation or prolonged drought had a profound effect on Nigeria's aquaculture systems supported by Lake Chad and was feared could lead to the total collapse of fishery activities in the West African nation (Oyebande *et al.,* 2002; FAO, 2010a). Generally, reduced precipitation leads to increased competition for freshwater. Reduced levels of rainfall in inland catchments over time may make farmers in the agriculture sector to take on fishing to support their livelihoods (Katikiro & Macusi, 2012). On the other hand, increased precipitation in wetland and inland aquaculture systems may cause changes to the salinity of the water bodies, which could impact the survival of salinity-sensitive aquatic organisms including prawns (Katikiro & Macusi, 2012). In Africa, Uganda, Nigeria and Egypt are estimated to be the most vulnerable to climate change (Barange *et al.,* 2018).

Increasingly wet conditions also put at risk the traditional food processing techniques such as the drying of fish (IPCC, 2014). Moreover, incidences of food-borne illnesses, such as ciguatera fish poisoning, and other types of diseases, are likely to increase as a result of climate change (IPCC, 2014). Increased flooding may also cause displacement of communities, subsequent migration and/or conflict, and destruction of aquaculture infrastructure, thus small-scale fishers in countries that over-depend on aquaculture and fisheries are most likely to suffer the consequences of climate change (Barange *et al.,* 2014; FAO, 2015).

Changing water temperatures and associated phonologies affect fish physiological processes and their ecological fitness (Brander, 2007; Barange & Perry, 2009; IPPC, 2014). It has been observed that most fish species sensitivity to acidification and pathogens increases in habitats beyond their thermal ranges (FAO, 2016b). Therefore, short-term climate change impacts on aquaculture and fisheries systems can include increased risks of pathogens and parasites, arising from rising global temperatures that affect their growth, metabolism, and ability to fight pathogens and diseases (Ficke *et al.,* 2007; Allison *et al.,* 2007). Long-term impacts can include prolonged drought and a decline in aquaculture and fisheries production. At worse, climate-driven changes in global temperature,

precipitation levels, ocean acidification, changed monsoon cycles, sea-level rise, the length and frequency of hypoxia events, modified ocean circulation patterns, and the modified hydrological regimes (De Silva & Sotto, 2009; Katikiro & Macusi, 2012) are expected to have long-term impacts in the aquaculture sector to varying magnitudes (Barange *et al.,* 2018).

In Africa, Egypt's brackish water production and Madagascar's marine aquaculture are considered to be highly vulnerable to climate change (Barange *et al.,* 2018; Handisyde *et al.,* 2017). In the case of brackish water production, Senegal, Côte d'Ivoire, Tanzania, Madagascar and Papua New Guinea are the countries with the lowest adaptive capacity to cope with the impacts of climate change; while for marine aquaculture, Mozambique, Madagascar, Senegal and Papua New Guinea were found to have the particularly low adaptive capacity (Barange *et al.,* 2018; Handisyde *et al.,* 2017).

In the past, fishing communities in Africa have been able to cope with the rare weather events such as flooding by being geographically mobile and creating alternative livelihoods (Boko *et al.,* 2007). However, today, progressive adaptation strategies and resilience building are required since increasing population growth and administrative barriers make age-old tactics inapplicable. Of course, small-scale and artisanal fisheries and fishers are particularly vulnerable to the impacts of climate change (Barange *et al.,* 2018). They often consist of commercial boat-based, or a small-scale, beach-based line- and net-fishery, which are labor-intensive and mainly exploit the species in estuaries and near-shore waters (Barange *et al.,* 2018). Therefore, the adaptation options provided in the FAO guidelines (FAO, 2012; FAO, 2015) are particularly designed for this cohort and could be useful for promoting sustainable aquaculture development in Africa. Additionally, community-based approaches to fisheries governance would be essential in improving the economic stability of small-scale fishers in the region, considering the increasing likelihood of extreme weather incidences in the decades to come (Barange *et al.,* 2018).

Lastly, the impacts of climate change do not respect administrative borders, even though each country has unique risks and vulnerabilities as well as institutional and socio-economic differences. Inevitably, climate-induced implications on marine stock availability, distributions, and assemblage (Barange & Perry, 2009) can lead to transboundary conflict at both regional and international levels (Barange *et al.,* 2018). Many species could migrate towards deeper ocean waters to find their ideal habitat conditions such as temperature and oxygen levels. Some commercial species may migrate offshore, further away from traditional fishing grounds (IPCC, 2007), permitting other species that are tolerant of higher temperatures and changes in the salinity of coastal waters to move into the void (Roy *et al.,* 2007; FAO, 2016b), negatively impacting fishery and profitability (Fairweather *et al.,* 2006). In Africa, the impacts of climate change are of greatest concern in the South Western

region, especially the fishing communities that depend on coastal and inland fisheries due to the high exposure of the low-latitude regions to the impacts of global warming (Barange *et al.,* 2014) and limited capacity to adapt to associated risks and opportunities (IPCC, 2014).

2.7   Climate-smart Approaches in Aquaculture Systems and their Challenges

The FAO's ecosystem-based climate-smart approaches in aquaculture and fisheries consider (a) sustainable increase in productivity and efficiency, considering environmental and socio-economic aspects of the sector, (b) reducing vulnerability and increasing resilience to enable the sector to cope with impacts of climate change, and (c) mitigating greenhouse gases throughout the entire value chain. The suggested climate-smart approaches capable of achieving these objectives are the Ecosystem Approach to Fisheries (EAF) and the Ecosystem Approach to Aquaculture (EAA) (FAO, 2016a).

According to FAO, some of the benefits of implementing the ecosystem approach to fisheries and aquaculture include (a) improving the general resilience of fisheries and aquaculture systems, including promoting the consumption of a greater diversity of fish species, to minimize vulnerability to the impacts of climate change and climate variability on resources, (b) adoption of context-specific and community-based adaptation strategies, and (c) stabilization of income for communities that rely on capture fisheries and aquaculture for their livelihoods (Chomo & Seggel, 2017; FAO, 2016a).

2.7.1   Sustainably Increasing Productivity and Efficiency in Aquaculture

For aquaculture, fully integrated systems, proper watershed management, water planning, improved feed efficiency, better disease diagnosis, and treatment can help increase productivity and efficiency in the aquatic systems (De Silva & Soto, 2009; Troell *et al.,* 2014a), without compromising the nutritional quality and safety of the fish (Beveridge *et al.,* 2013). Some developed economies use innovative technologies such as hyperspectral imaging (HSI) to check diseases and microbial contamination in fish products (Vejarano *et al.,* 2017). Additionally, emerging biotechnologies such as the development of transgenic fish, for example, salmon in the United States and Canada (Aerni *et al*., 2004) have enabled the production of fish with greater tolerance to temperature, salinity, and susceptibility to disease (Wakchaure *et al.,* 2015).

In terms of feed formulation, the aquaculture sector has been over-reliant on fishmeal and fish oil (Tacon & Metian, 2008); this has significantly constrained growth in the sector (Little *et al.,* 2016). Other constraints include increased competition from the agricultural sector for the available land

and water resources (Troell *et al.,* 2014b), which could significantly impact location, productivity, and scalability of the aquaculture production systems (FAO & World Bank, 2015). As such, aquaponics (the symbiotic relationship between aquaculture and hydroponics) has been suggested as a potential climate-smart option for increasing efficiency and address these constraints (Martins *et al.*, 2010). Of course, hydroponics (the cultivation of plants in water without soil) can be combined with aquaculture in a closed recirculation system. The roots of the plants (or crops) floating on water can assimilate the nutrients metabolized by the bacteria, and then the purified water is often returned to the tanks/ponds for fish to use (FAO, 2016a; Chomo & Seggel, 2017).

Therefore, aquaponics (integrated agriculture/aquaculture technique) is a climate-smart approach for increasing productivity and efficiency in food production (FAO, 2016a). The utilization of plants and vegetables in aquaponics helps minimize fish waste discharge and reduces watershed pollution by eliminating the need for mineral fertilizers and pesticides in agriculture (Chomo & Seggel, 2017). With increasing competition for freshwater resources, aquaponics has the potential to sustain high productivity with less labour and land while maximizing nutrient utilization and minimizing water usage (FAO, 2014; FAO, 2016a; Chomo & Seggel, 2017).

Aquaponics has other benefits too. Since it's a controlled system, it provides a level of biosecurity that reduces the risk of disease or infestation, while solving challenges found in traditional agriculture such as soil degradation, erosion, mineral fertilizer requirement and irrigation. It's believed that aquaponics generates fewer greenhouse gas emissions to produce the same amount of product in a relatively small space by eliminating the energy requirement for tilling the land or no application of mineral fertilizers (FAO, 2014; FAO, 2016a).

Large-scale commercial aquaponics requires substantial capital investment and a ready market for the often premium-priced pesticide-free vegetables and may be too expensive to small and medium-scale farmers (Chomo & Seggel 2017). However, for a start, FAO has prioritized supporting Small-scale Aquaponic Food Production efforts (FAO, 2014) and has invested in conducting training workshops in Eastern and Northern Africa and building demonstration sites in the Caribbean countries (FAO, 2016a; Chomo & Seggel, 2017).

Despite the high capital expenditure and technical requirement for Climate, Smart Aquaculture approaches such as transgenic fish production, hyperspectral imaging for disease control, and aquaponics/hydroponics, these technologies have the potential to support economic development and enhance food security and nutrition in Africa. Unfortunately, these Climate Smart Agriculture (CSA) technologies may not be easily adopted in Africa because they are not context-specific in terms of regional cultures and economies. For instance, transgenic fish may not be entrepreneurially

feasible in Africa because of country-specific cultural norms and unknown long-term environmental and human health consequences (Aerni *et al.*, 2004). Additionally, expensive high-tech technologies such as hyperspectral imaging equipment for disease control may be inaccessible and unaffordable for many small and large scale farmers in the region. As such, contextualized technology suitable for Africa's multi-cultural situations, economic realities, and political challenges is crucial for the adoption of CSA technologies to ensure food security for the region.

2.7.2   Reducing Vulnerability and Increasing Resilience to Climate Change Impacts

The Democratic Republic of the Congo is the second most vulnerable national economy globally to climate change-driven impacts on fisheries since its nutritionally dependent on fish (45% of animal protein being derived from fish) (Allison *et al.,* 2009). For such an economy, climate-smart disaster risk reduction and management strategies are valuable because climate change and climate variability can cause reduced yields from aquaculture farms arising from global warming, acidification and pathogens (FAO, 2016a). Culture-based aquaculture (a stock enhancement process) is a smart way to improve resilience and increase fish production and diversification for food (Amarasinghe & Nguyen, 2009) using limited resources such as freshwater (De Silva, 2003). Culture-based aquaculture is very relevant for species whose breeding grounds have been affected by climate change, such as: (a) mussels, (b) shrimp, especially *Penaeus monodon* and freshwater prawns, (c) tuna and (d) some high-value marine finfish (Barange *et al.,* 2018). Bivalves and seaweeds, of course, require no additional feed input.

Culture-based aquaculture is less costly, environmentally friendly, and does not consume external feed resources, thus has no greenhouse gas emission related to feeding (FAO, 2016a). Financially, this would be suitable for semi-intensive to extensive aquaculture systems in Africa to ensure food security, especially in the rural communities that often share communal waterbodies. Regardless, the system is vulnerable to the unpredictability of precipitation resulting from climate change, which is beyond human control, and thus, can have a significant impact on the productivity of the system. To adapt, stocking in culture-based aquaculture can be done during the rainy season, and harvesting can take place at the onset of the dry season. Also, indigenous fish species from well-managed broodstock could be utilized to avoid genetic introgression and disease from wild stock (FAO, 2016a).

Another practical option would be to introduce marine and euryhaline species (with wide salinity tolerance) or shift to coastal aquaculture-based fisheries in response to water circulation changes, water stress, and drought conditions (Daw *et al.,* 2009; De Silva & Soto, 2009). Building such resilient livelihoods in Africa would equip communities with tools to withstand damage from

climate change, recover quickly as well as adapt to change (IPCC, 2014). Lastly, the resilience of the aquaculture sector to climate change impacts may need adaptation efforts focused on enhancing the sustainability of aquaculture resources as well as constructing climate-resilient infrastructure such as deeper ponds, among others.

2.7.3   Mitigating Greenhouse Gas Emission

Generally, aquacultures play significant roles in reducing and/or supporting the natural removal of emissions as well as providing alternative energy sources. In aquaculture food production, feed, and fertilizer and the primary and secondary contributors to greenhouse gas emissions, respectively (FAO, 2016c). It was estimated that 385 million tonnes of CO_2 equivalent (CO_2) were emitted in 2010 from the aquaculture sector, amounting to approximately 7% of those from agriculture (Hall *et al.,* 2011). Emissions (methane and nitrous oxide) from sediments and water systems tend to increase from the extensive system (no treatment and/or only partial fertilization) to semi-intensive (uses fertilizers and/or partial feeding) or intensive systems (fully dependent of feeds). In fact, intensive production of finfish and crustaceans is the greatest emitter for greenhouse gases because it is heavily reliant on feeds as well as energy for water aeration (Hasan & Soto, 2017; Robb *et al.,* 2017). Comparatively, the farming of molluscs produces relatively low greenhouse gas emissions (Bonaglia *et al.,* 2017).

Additionally, energy sources (e.g. fuel) for machines and equipment (e.g. water pumps and vehicles, etc.) used in aquaculture production processes also generate greenhouse gases (FAO, 2016a; Hasan & Soto, 2017; Robb *et al.,* 2017). Energy-intensive post-harvest processing such as smoking, drying, packaging, storage, and transportation contribute to greenhouse gas emission. Newer and more efficient machines and equipment can save fuel compared to old engines. Renewable energy (e.g. wind, solar and hydropower) could eliminate the need for diesel for hydraulics, refrigeration, heating, cooling, lighting, pumps, etc. required in aquaculture operations (Thomas *et al.,* 2010).

Models indicate that using better technologies, renewable energy, improving feed conversion rates, and formulating fish feed with crop-based ingredients instead of marine-based ingredients would greatly reduce greenhouse gas emissions in aquaculture (Waite *et al.,* 2014). Using 2010 as the baseline year, these efforts together are projected to increase global aquaculture production by 133 percent by 2050 while reducing greenhouse gas emissions by 21% in CO_2 emission per tonne of fish produced (Waite *et al.,* 2014). It's also been reported that integrated food production systems, for example, rearing fish in rice paddies would maximize food production and energy utilization sustainably while mitigating greenhouse gas emission from rice fields (Lipper *et al.,* 2017).

Additionally, integrated mangrove-shrimp cultivation can substantially reduce blue carbon emissions (carbon sequestered, stored and released in coastal mangroves, seagrass and salt marshes) (Ahmed *et al.,* 2017; McLeod *et al.,* 2011). Mangroves are one of the most threatened tropical ecosystems (Donato *et al.,* 2011) yet they store carbon better than other tropical upland forests (Alongi, 2014). Destruction of mangrove forests has led to an increase in emissions of blue carbon (Alongi, 2014). It's estimated that an area covered by 50% mangrove forest and integrated with shrimp culture can sequester 0.86–1.04 million tones of carbon per year and reduce overall greenhouse gas emissions (Ahmed *et al.,* 2017). According to Naturland organic aquaculture standards, integrated mangrove-shrimp farming can also be certified as organic aquaculture (Naturland, 2019).

Lastly, seaweed (microalgae) aquaculture in the deep seas can act as a CO_2 sink, and if used for biofuel (bio-ethanol and biodiesels) production, has the potential to reduce greenhouse gas emissions from fossil fuels required for energy production (Duarte *et al.,* 2017). Biofuel production is an already established enterprise; it's efficiency and yields are increasing. Seaweed aquaculture can also improve soil quality and significantly eliminate the need for synthetic fertilizers in agricultural production, and when used in animal feed, help lower methane emission from cattle (Duarte *et al.,* 2017). However, inland seaweed aquaculture, grown in conventional culture systems, face challenges such as lack of suitable areas for the practice, resource competition, high capital for infrastructure installation to cope with extreme climate change impact on off-shore environments, and ready market demand for seaweed products (Duarte *et al.,* 2017; Barange *et al.,* 2018).

In Africa, reducing greenhouse gas emissions from aquaculture systems can be achieved in ways that are cost-effective and socially efficient. Some measures with the potential to improve the physical performance of fish and reduce greenhouse gas emissions include: (a) Breeding for improved feed conversion ratio (FCR) (Thoa *et al.,* 2016); (b) Vaccination for *streptococcosis* which is likely to improve animal welfare by reducing mortality rates as well as a decrease in antibiotic use (Liu *et al.,* 2016); and (c) adding phytase to the ration to improve nutrient utilization and bioavailability (Adeoye *et al.,* 2016).

2.8 The Need for Context-specific Climate-smart Aquaculture Framework for Africa

According to FAO, the goal of CSA is to support food and nutrition security while considering the mitigation of greenhouse gas emission as well as adaptation to a changing climate (FAO, 2013). In Africa, aquaculture-based communities are particularly vulnerable to impacts of climate change on the natural resources required for productivity and survivability of fish and other aquatic invertebrates. As such, CSA addresses challenges regarding aquaculture infrastructure development

for protecting and improving production capacities and the supply chain while minimizing their potential negative trade-offs (FAO, 2013; FAO, 2016a).

Additionally, CSA is targeted at building resilience to climate change impacts on aquaculture, thus enhancing FAO's achievement of national food security and sustainable development goals (FAO, 2013), and which will require: (a) Improving natural resource utilization efficiency e.g. aquaponics/hydroponics; (b) Reducing vulnerability and increasing resilience at the local level to support aquaculture-dependent communities; and (c) Reducing and removing greenhouse gases, mainly carbon dioxide (CO_2), methane (CH_4) and nitrous oxide (N_2O) (Barange *et al.,* 2018). Fish farmers in Africa could help the sector achieve this by adapting to an aquaculture system that ensures increased production efficiency through improved feeding (lower feed conversion ratio), proper disease diagnosis and management, use of renewable energy, and reduction of postharvest and production losses (FAO, 2016a; De Silva & Soto, 2009; Daw *et al.,* 2009). These strategies will require institutional and human capital, the involvement of private and public sectors, as well as participation at regional and national levels, to ensure the aquaculture sector is climate-smart even as the sector tries to expand economic and trade opportunities across countries.

It would be expected that regional policymakers and other stakeholders should be able to develop and implement appropriate responses to climate change in their respective regions through inclusive dialogue with neighboring countries and proper analysis of scientific data. The success of CSA in the African region will require that climate-smart approaches are locally relevant, economically sustainable, culturally appropriate, and environmentally friendly. Even so, adaptation to the impacts of climate change and climate variability as well as mitigation strategies to reduce or limit greenhouse gas emissions must consider the use of aquaculture practices that adhere to the FAO Code of Conduct for Responsible Fisheries (FAO, 1995) and whose implementation is facilitated by the ecosystem approach to fisheries and aquaculture (EAF/EAA) (FAO, 2003; FAO, 2009; FAO, 2013; Chomo & Seggel, 2017). Already, Nigeria has responded by adopting integrated aquaculture to encourage increased food production; treatment of fish wastewater to minimize pollution of surrounding water bodies; adopting the use of tarpaulin ponds during dry weather; and erecting shades over the pond to control water temperature and reduce evaporation losses (Thaddeus *et al.,* 2012).

Constraint to CSA in Africa may include high adaptation costs that negatively impact production and profits; unclear trade and value-added opportunities; lack of awareness, preparedness, and appropriate skills; political interactions at national and regional levels; competitiveness of exports and world trade patterns; and local social, economic and policy measures of greenhouse gas impact.

Therefore, context-specific climate-smart aquaculture processes and actions may reduce the impacts of climate change and climate variability on aquaculture systems, improve the sector's mitigation potential, build value chain resiliency and promote sustainable production and consumption while ensuring societal and environmental sustainability.

Community-based capacity building in basic aquatic resource management will ensure underlying resilience in the face of climate variability and change. Of course, community-based adaptation strategies will improve the management of farms and the choice of farmed species by facilitating understanding, and the use of inclusive devolved approaches involving local stakeholders. Adaptation measures to climate change that could be appropriate in Africa's context may include: proper zoning, planning and site selection for aquaculture through risk analysis (Cattermoul *et al.,* 2014; FAO, 2017d); adoption of environmental monitoring systems to track weather events that can trigger disease outbreak and water movements that cause toxic algal blooms (Barange *et al.,* 2018); provision of access to affordable credit and insurance for recovery from climate-change-induced damages (Karim *et al.,* 2014; FAO, 2016d, 2017e); better management practices that improve the environmental performance, productivity and profitability of aquatic farms (Barange *et al.,* 2018); technological innovations that reduce susceptibility to climate change such as aquaponics/hydroponics (Somerville *et al.,* 2014); aquaculture diversification strategies that's compatible with local ecosystems (Harvey *et al.,* 2017); and integrated agri-aquaculture production systems to maximize resource utilization and reduce greenhouse gas emission (Crespi & Lovatelli, 2011; Shelton, 2014; Barange *et al.,* 2018). Nonetheless, to facilitate the mainstreaming of CSA in Africa, efforts are needed to integrate aquaculture into climate change adaptation and food security policies at every level of governance in each country to build synergies in local institutions and ensure their incorporation into development planning.

CHAPTER THREE

MATERIALS AND METHODS

3.1   Description of the Study Regions

This study was carried out in fish ponds located in 10 sites within Arusha (3.3869° S, 36.6830° E) and Morogoro (6.8278° S, 37.6591° E), which lie in the northern and eastern regions of Tanzania, respectively.

Arusha Region is among Tanzania administrative regions with an estimated population of 1 694 310 according to the 2012 national census and a total area of 37 576 km^2. The region is bordered by Kajiado and Narok County in Kenya to the North, Kilimanjaro Region to the East, Manyara and Singida regions to the South, Mara and Simiyu Regions to the West. Arusha City Council is the capital of the Arusha region which is bordered to the South, West, and North by Arusha Rural District and the East by Meru District.

According to the 2012 national census, the Morogoro region had a population of 2 218 492. The total area of the region is reported to be 70 624 km^2. The region is bordered to the north by the Tanga Region, to the east by the Pwani and Lindi Regions, to the south by the Ruvuma region, and the west by the Iringa and Dodoma Regions. Morogoro town is the capital of the Morogoro region with a population of 315 866 (2012 census) located in the eastern part of Tanzania, 196 kilometers (122 mi) west of Dar es Salaam, the country's largest city and commercial centre, and 260 kilometers (160 mi) east of Dodoma, the country's capital city.

(a) Study locations in Arusha (b) Map of Tanzania showing administrative boundaries (c) Study locations in Morogoro
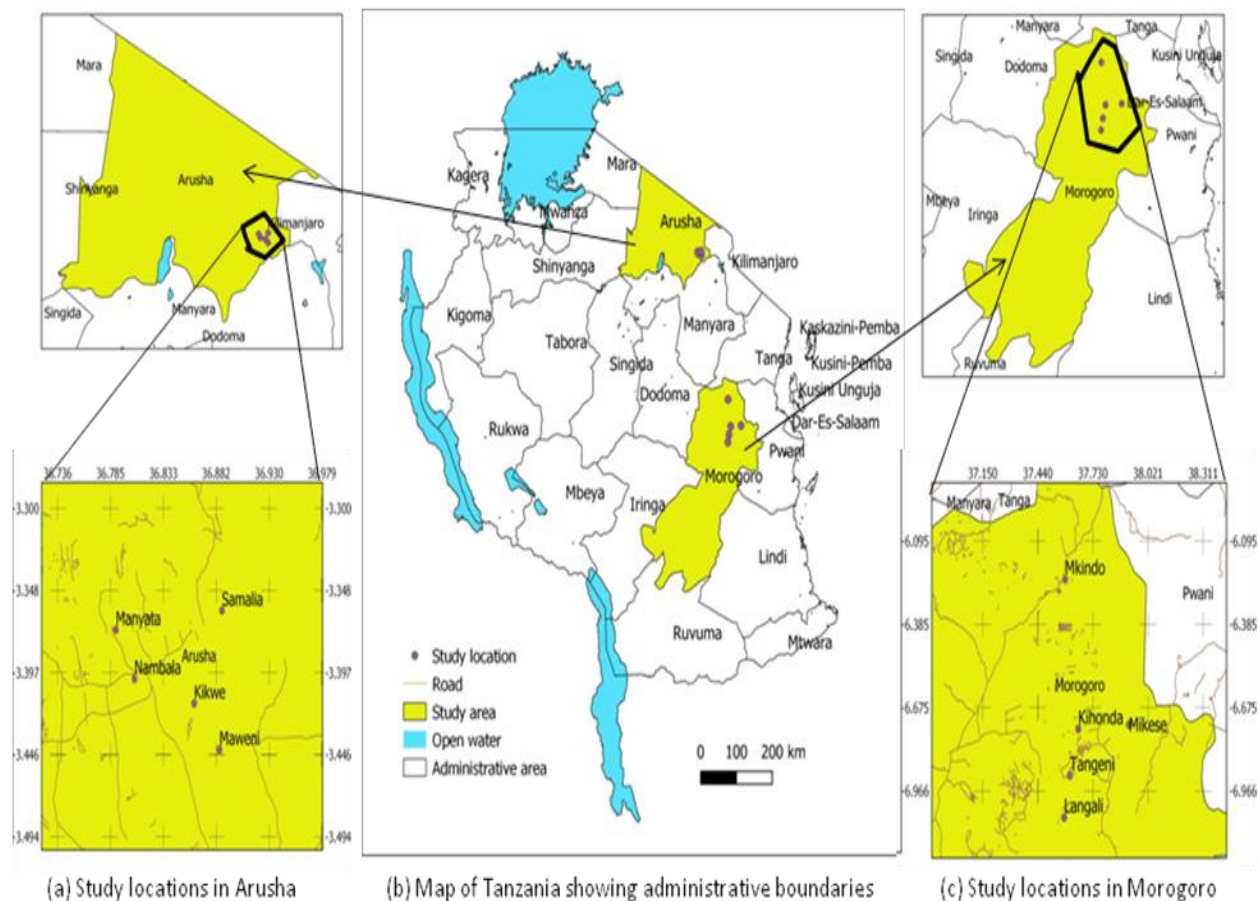
Figure 1: Tanzania map indicating the study regions (Arusha and Morogoro) and sites

3.2   Study Design

A stratified sampling technique was used to capture data from emerging aquaculture producing regions, namely, Arusha and Morogoro. Both regions are emerging as reliable aquaculture resources in the country, thus could illuminate current aquaculture practices in Tanzania. Five villages from each region were selected for this study (Fig. 1). Kihonda, Langali, Mikese, Mkindo and Tangeni villages from Morogoro region; Kikwe, Nambala, Maweni, Manyata and Somalia villages from Arusha region. A total of 130 fish ponds were randomly selected with 65 fish ponds for each region and were found to use either integrated, non-integrated, or semi-integrated fish farming methods.

3.3   The Study Population

3.3.1   Inclusion Criteria

The study included fish farmers from selected sites in each of the two regions. An adult (>18) fish farmer or farm manager who was willing to participate and capable of answering the questionnaire was considered fit to participate in the research.

3.3.2 Exclusion Criteria

The study excluded all the fish farmers below the age of 18 and all farmers who were mentally unable to pursue the study. All the farmers who were unwilling to participate or provide consent were excluded from the study.

3.4 Sample Size Determination

Eligible fish farmers were selected with the assistance of the local authority representative -in-charge at each participating site until a total of 130 fish farmers (130 fish ponds) were recruited to the study. The sample size was calculated using a formula described by Fischer *et al.* (1991) as follows:

$$n=Z^2pq/d^2$$

Where; n = the desired sample size

Z = standard normal deviation set at 1.96 correspondings to 90% CI

q = 1.0 – p

d = degree of accuracy desired (0.05)

p= proportion in the target population with certain characteristics (types of existing fish farming systems such as earth and cage ponds with and without integrated farming practices).

3.5 Sample Collection

Samples for lab work were collected from 130 fish ponds. Each region contributed half of the total fish ponds. Care was taken to ensure that we obtain representative samples from each fish farming system (cage and earth ponds with and without integrated farming practices). Samples collected from these ponds include fish muscles, fish feeds, and pond water. Fish and feed samples were collected and packaged in sterile polyethylene zip bags. Water and sediment samples were collected from the surface and the bottom of the fish ponds using a sterile bott 100 mL bottles.

3.6 Data Collection and Laboratory Analysis

3.6.1 To Examine the Quality of Existing Aquaculture Practices in Tanzania

On-site interviews and a set of pre-tested structured questionnaires were used to collect information from a sample of fish farm owners and managers in each region. The interviews were conducted by the author of the questionnaire, two trained interviewers, and a local authority representative. The information collected captured the following: aquaculture knowledge background of the respondents, their farm infrastructure (e.g. pond area, volume and stocking density), details of the

antimicrobials, parasiticides, and disinfectants usage during the last crop, and whether they were used for disease prevention or treatment. The interviews also probed for types and frequency of diseases and respondents' understanding of the clinical symptoms of these diseases. In some farms, direct observation and description of feed medication practices were recorded. Types and dosages of the applied antimicrobials and disinfectants were collected from the farmers' records. The farmers also provided registration of the chemicals used in the farms and the rationale for the choice of each chemical was explored.

To triangulate the data on types and doses of chemical use reported by farmers, data were cross-checked by comparing with supplier product label information from the shops selling chemicals for aquaculture.

3.6.2 Prevalence of Fish Parasites in Nile Tilapia and African Catfish and Physicochemical Characteristics of Pond Water

(i) Collection of Fish Samples and Freshwater Samples

Fishing was done using 1×1 m seine net following the method by Mdegela *et al.* (2011). Fish were randomly captured from each of the 130 fish ponds studied in both Arusha and Morogoro regions and then transferred in oxygen-filled polyethylene bags to keep the fish alive. Water samples for physicochemical analyses were collected aseptically from the pond's water surface (maximum 1 feet depth) using sterile cap bottles and then transported to the laboratory in an ice-packed container. All samples were transported to the Microbiology Laboratory at the Department of Microbiology, Parasitology, and Biotechnology in the College of Veterinary Medicine and Biomedical Sciences, Sokoine University of Agriculture (SUA), Morogoro.

(ii) Parasitological Examination and Identification

The weight and standard length of each fish were measured and recorded. Live fish were stunned with a single blow to the back of the head and pithed to separate the central nervous system from the spinal cord. Gross examination of the skin was done for ectoparasites. Wet mounts of the skin scrapings and gill filaments were collected on slides with saline and examined under the microscope for ectoparasites. The eyes were removed and contents were expressed on a slide and examined for eye flukes. Post mortem of the fish was performed as described by Noga (2010). For endoparasite identification, the dissection plate was used to lay every fish on its correct side with the mid-region coordinated towards the dissector. The body cavity was opened into two cuts by using a pair of scissors. A dark shaded plate was used to put the removed gastrointestinal tract (GIT). The GIT was

then opened and the internal content was removed. Around 100 mL of distilled water was added, delicately shaken to clean out the digestive organs, and analyzed for parasites utilizing laboratory lamps with the aid of a magnifying lens. Any parasite found was carefully transferred using plastic forceps into bottles containing 15 mL of 70% alcohol and stored for identification.

Stainless steel sieve (W.S. Tyler Incorp. Mentor, OH, USA) with approximately 212 µm pore size was used to filter the remaining contents. After filtration, the blend was transferred to a Petri dish for additional assessment of parasites under a stereo microscope following the method of Mdegela *et al.* (2011). Briefly, the filtrates were centrifuged at 425.6G force for five minutes using a centrifuge machine (Sigma, USA). After discarding the supernatants, a drop of the collected sediments was placed on a microscope glass slide followed by two drops of saline, carefully smeared, and covered using glass coverslip. A light microscope (10× and 40× magnification) was used to examine the content for the presence of worm eggs, adult parasites, and coccidian oocyst. Additionally, wet smears of the intestinal mucosa were prepared by sampling the contents with a glass slide at multiple areas and the parasites observed identified using standard fish parasite identification keys as described by Paperna (1996). A stereo microscope (20x amplification) with a side lamp was used to observe any parasites present in the samples previously stored in 70% alcohol and smeared on a magnifying instrument glass slide with a drop of lactophenol before placing a coverslip.

(iii)   Analysis of Water Quality in Pond Using Physico-Chemical Parameters

Physicochemical water quality parameter analysis (e.g. nitrate, ammonia, alkalinity and hardness, turbidity, and Biological Oxygen Demand (BOD) were done using Water test kits (Tetra Gmbh, Germany) and according to the manufacturer's instructions. Water Ph, Dissolved Oxygen (DO) and the temperature was measured using a portable handheld multi-parameter probe (Eco pond supply, USA).

3.6.3   Occurrence of Fish Bacteria Pathogens Isolated from Farmed Nile Tilapia and African Catfish

(i)   Fish, Pond Water, and Feed Sample Collection

A total of 130 fish were collected from the 10 study sites in Arusha and Morogoro regions. Every site had 13 different fish ponds sampled. A set of 100 mL sterile bottles were used to collect water samples. The bottles were inverted into the water to about a foot deep to avoid entrapping any air bubbles and then capped under the water. A total of 130 fish feed samples (100 g each) were also

collected from the study locations and placed in acid-cleaned polyethene bottles. All samples were immediately transported in cool boxes to the Microbiology Laboratory at the Department of Microbiology, Parasitology, and Biotechnology in the College of Veterinary Medicine and Biomedical Sciences, Sokoine University of Agriculture, Morogoro.

(ii)   Sample Preparation

All the water, fish feed, and fish samples were assayed for bacteria contamination. The sampled fish were first inspected and then assumed to be clinically normal because none of them had a gross lesion. The fish were humanely killed by the physical destruction of the brain using a sharp blow to the head. The fish surfaces were disinfected using 70% ethanol solution and then aseptically dissected to obtain tissue samples of the skin, gills, liver, kidney and intestines. Skin samples were obtained by aseptically macerating 1 cm^2 of skin in 10 mL of water following the method described by Wamala *et al*. (2018). All other tissue samples were obtained by Uddin and Al-harbi (2012). The tissue samples from individual fish were then pooled and homogenized in sterile phosphate-buffered saline (PBS) using mortar and pestle technique.

(iii)   Bacteria Isolation

The fish feed samples (45-g dry weight) were homogenized with a 15-mL solution whose mass concentration of substances (in g/100 mL water) was NaCl, 9; $Na_4P_2O_7$, 0.1; polyoxyethylene ether w-l, 0.1. The homogenate was then diluted with 0.1% peptone water for analysis. Water, fish feed, and fish samples homogenate were inoculated in Blood Agar, Brain Heat Infusion (BHI), and Trypticase Soy Agar (TSA). Additionally, some selective nutrient agar were used including Mueller-Hinton Agar (MHA) for *Pseudomonas* spp. Salmonella Shigella Agar (SSA) was used to enumerate *Salmonella* spp and *Shigella* spp For pathogenic *Vibrio* spp. Thiosulphate Citrate Bile Salt Sucrose (TCBS) agar was used, all from Sigma Aldrich, USA. The plates were incubated at 30 °C for 48 h upon which sub-culturing was done to obtain pure cultures.

(iv)   Identification and Characterization of the Isolated Bacteria

The isolated bacteria were identified and then characterized using previously described morphological and biochemical test procedures (MacFaddin, 2000). First, colony morphology (shape, colour, pigmentation, hemolytic activity, size, edges and elevation) were determined, and isolates grouped accordingly for each region. Then from each group, three representative isolates were subjected to further tests including Gram staining, Mobility test, oxidase test, catalase, oxidative fermentation, methyl red, nitrate reduction, citrate and urea slants. Analytical Profile Index

(API) test kits 20E, 20NE and API Staph from Biomerieux were used to further confirm isolates identification, following the manufacturer's recommended protocol.

3.6.4 Bioaccumulation and Distribution Pattern of Heavy Metals in Aquaculture Systems

(i)   Sample Collection and Preparation

Fish

In fish, only the muscle, which is the main part of the fish being consumed, was used to assess the quality of fish for human consumption. Representative fish were caught from each of the 130 ponds studied using small gill net and sealed in zip lock bags. Iceboxes were used to transport the fish samples immediately to the laboratory and kept at –30 °C. In the laboratory, fish samples were first thawed and rinsed thoroughly with deionized water before obtaining the muscle tissues using a stainless steel scalpel. The muscle samples, approximately 100 g, were collected from just below the dorsal fin, above the midline, and then dried up on the oven at 60 °C for 24 h, ground into a fine powder, homogenized, and stored in desiccators before analysis.

Fish Feed

A total of 130 fish feed samples were collected on-site from the farmers and placed in acid-cleaned polyethene bottles and transported to the laboratory. In the laboratory, fish feed samples were oven-dried at 60 °C for 12 h in aluminum trays, ground into a fine powder, homogenized and stored in a desiccator before analysis.

Sediments

Sediment samples were collected from the bottom (upper layer ~0–10 cm) of each of the 13 fish ponds studied in every location following previously established principles (Kodom *et al*., 2010; Kodom *et al*., 2012), but with some modifications. Briefly, the sediments were sampled using a stainless steel core sampler and immediately transferred into appropriately labeled clean polythene bags, sealed and placed in iceboxes to prevent oxidation. All the samples were then taken to the laboratory and stored at 4 °C until analysis. For analysis, samples were prepared for drying by spreading them evenly on aluminum trays, taking care to avoid sample cross-contamination or contamination from any external sources. Samples were oven-dried at 60 °C until a constant weight (Mean Concentration [MC] <20%) was attained, then ground and stored before spectrometrically analyzing them in replicates. Moisture content above 20% interferes with the X-Ray Fluorescence (XRF) analysis (Kodom *et al*., 2010).

(ii)   X-ray Fluorescence Instrumentation and Analysis of Heavy Metals

Heavy metals analysis in pond sediment, fish feed, and fish muscle samples were carried out at the Tanzania Atomic Energy Commission Laboratory in Arusha, Tanzania. Following previously established protocols for X-ray fluorescence (XRF) spectrometer analysis (Kodom *et al.*, 2012), the finely ground dry samples were first sieved to achieve approximately 75 μm particle sizes by using Retsch aluminum test-sieves with a vibratory electronic sieve shaker, which were then pulverized (ground into finer loose powder state) to further reduce the particle size to <60 μm. This is because the XRF spectrometers only analyze the surface layer of pellets, and the smaller the particle size the better the homogeneity of the sample (Kodom *et al.*, 2010). To make pellets for analysis, 0.9 g of Hoechst wax (a mixture of starch, cellulose, and polyvinyl alcohol binder) was added to 4.0 g of each finely ground sample and thoroughly mixed using Retsch Mixer Mills (MM301) before manually pressing into pellets of 32 mm diameter and 3 mm thickness by using Specac hydraulic press machine of 15 tons (or 15 000 kg) maximum pressure limit, following Protocal by Kodom *et al.* (2010).

Heavy metals concentrations in the pellets were quantitatively measured using a polarized energy dispersive X-ray fluorescence (PED-XRF) spectrometer following protocols described by Kodom *et al.* (2012). The PED-XRF instrument was fitted out with an Rh anode X-ray tube, 0.5 mm Be side window, and a circular rotating position sample changer inside a sample chamber with a holding capacity of up to 20 sample holder disks of 32 mm diameter for sequential sample analyses. A computer-based multichannel analyzer containing a menu-based Spectro X-LAB Pro Software Package (Turboquant) was utilized in controlling sample analysis using a pre-set method, made up of a series of tasks (Guthrie & Ferguson, 2012). Due to the high sensitivity of the spectrometer, great care was taken to avoid putting fingerprints on the pellets' surface layer. The results (spectra) collected were reported as a mass fraction in parts-per-million (mg/kg) and represented in terms of mg/kg. Detection in the PED-XRF instrument was achieved with a SPECTRO X-LAB 2000 instrument equipped with a Si(Li) detector of 10–30 mm^2 active surface, 3–4 mm effective thickness, and a maximum energy resolution of 150 eV at 5.9 keV and a count rate of 1000 cps.

(iii)   Estimated Daily Intakes

The estimated daily intakes (EDI) for the examined metals were obtained by multiplying the heavy metal's respective mean concentration (MC) in the targeted edible fish muscle samples by the average fish weight consumed by an adult person (60 kg) in Tanzania per day (URT, 2015). The fish consumption data for Tanzania was obtained from the Tanzania Annual Fisheries Statistics Report of 2015-2016 (URT, 2015).

EDI was calculated by using the following Equation:

$$EDI = DFC \times MC \hspace{4cm} 1$$

Where EDI = estimated daily intakes; DFC = daily fish consumption; MC = mean concentration of metal in the fish sample. On a fresh basis, the DFC rate for a 60 kg Tanzanian adult was 22.1 g on average (URT, 2015).

(iv)   Non-carcinogenic Risk

The non-carcinogenic risk levels associated with the consumption of heavy metals were estimated by calculating Target Hazard Quotient (THQ). The THQ values for each metal analyzed were calculated following a modified equation established by Wang *et al*. (2005) while using the standard assumption for an integrated United State Environmental Protection Agency (USEPA) risk analysis (United State Environmental Protection Agency [USEPA], 2000).

$$THQ = EDI /(RfD \times BW) \hspace{4cm} 2$$

where EDI stands for Estimated Daily Intakes for the analyzed heavy metals; BW = average body weight (60 kg) of a Tanzanian adult; RfD is the oral reference dose (mg/kg/day); RfDs are based on 0.003, 0.004, 0.0005, 0.001, 0.0015 mg/kg-bw/day for As, Pb, Hg, Cd and Cr, respectively (USEPA, 2000). The THQ value of greater than 1 indicates that there is a potential health risk to the exposed population (Hallenbeck, 2018).

(v)   Hazard Index

Hazard index (HI) is the total risk assessment of the cumulative or interactive effect of two or more pollutants (USEPA, 2020). The total health risk was measured by summing up THQ value of each metal analyzed in this study per USEPA risk-based concentration table (USEPA, 2020).

$$HI = \sum THQ = THQ \text{ (toxicant 1)} + THQ \text{ (toxicant 2)} + \ldots + (THQ \text{ (toxicant 3)} \hspace{1cm} 3$$

A higher HI value indicates a greater risk level and corrective measures must be considered (Wang, 2005).

(vi)   Carcinogenic Risk

Cancer develops over time. The cumulative likelihood of a person developing cancer over a lifetime due to exposure to a potential carcinogen is expressed mathematically as a carcinogenic risk (CR) using the equation below (USEPA, 2000).

$$CR = EDI \times CSF \times 10^{-3} \hspace{4cm} 4$$

Where, CR = carcinogenic risk; EDI = estimated daily intake of heavy metals; and CSF = cancer slope factor set by USEPA (USEPA, 2000). The carcinogenic slope factor (CSF) of As, Pb, Cd and Cr used in this study were 1.500, 0.0085, 0.38 and 0.5 mg/kg/day, respectively (WHO, 1976). Acceptable risk of cancer progression in the course of human lifespan range from 10^{-4} (risk of developing cancer over a human lifetime is 1 in 10 000) to 10^{-6} (risk of developing cancer over a human lifetime is 1 in 1 000 000) (WHO, 1976).

3.6.5   Design of a Context-specific Climate-smart Sustainable Fish pond

(i)   Key Elements of Climate-smart Designed Pond

The Model

The conceptual climate-smart pond design considered both eliminating discharge of wastewater into the environment and enhancing resilience to climate change and disasters. The structure consisted of two production units each measuring 2 m height × 2 m width × 1.5 m depth, constructed with iron vertical columns and horizontal bars welded together to form a supporting frame onto which the polyethylene membrane (pond liner) was attached (Fig. 2a, & 3a & b). For comparison purposes, the first pond had no filter but rather a drain valve to empty the pond before replacing the water biweekly. The second pond had a filter for stripping and recycling the water within the system (Fig. 3c). For this pond, once it was filled at the beginning of the production cycle, no water was added except by precipitation. A solar-powered pump was used to channel the used water into the filtration unit in oxygen-filled polyethylene bags every week for recycling through the system (Fig. 2b & 3c).

(ii)   Construction of the Fish Pond

The climate-smart pond was constructed using locally source materials. The iron support columns, polyethylene membrane (pond liner), piping, valves, bolts, and solar-powered water pump were purchased from hardware stores. The filter was constructed using a plastic food-grade 55-gallon barrel whose inside was stratified with a layer of cotton at the bottom followed by activated charcoal and four inches of clean fine sand sandwiched between two layers of gravel (Fig. 2b & 3c). A special

feature of the climate-smart pond was that the pump pushed used water through the filter starting from the cotton layer at the bottom and then upwards toward the top fine sand layer. The clean filtered water was then channeled back into the pond via a plastic piping system at the top section of the filter (Fig. 2b).

(a) Top view of the pond

(b) Front elevation, cross-section X–X view of the pond
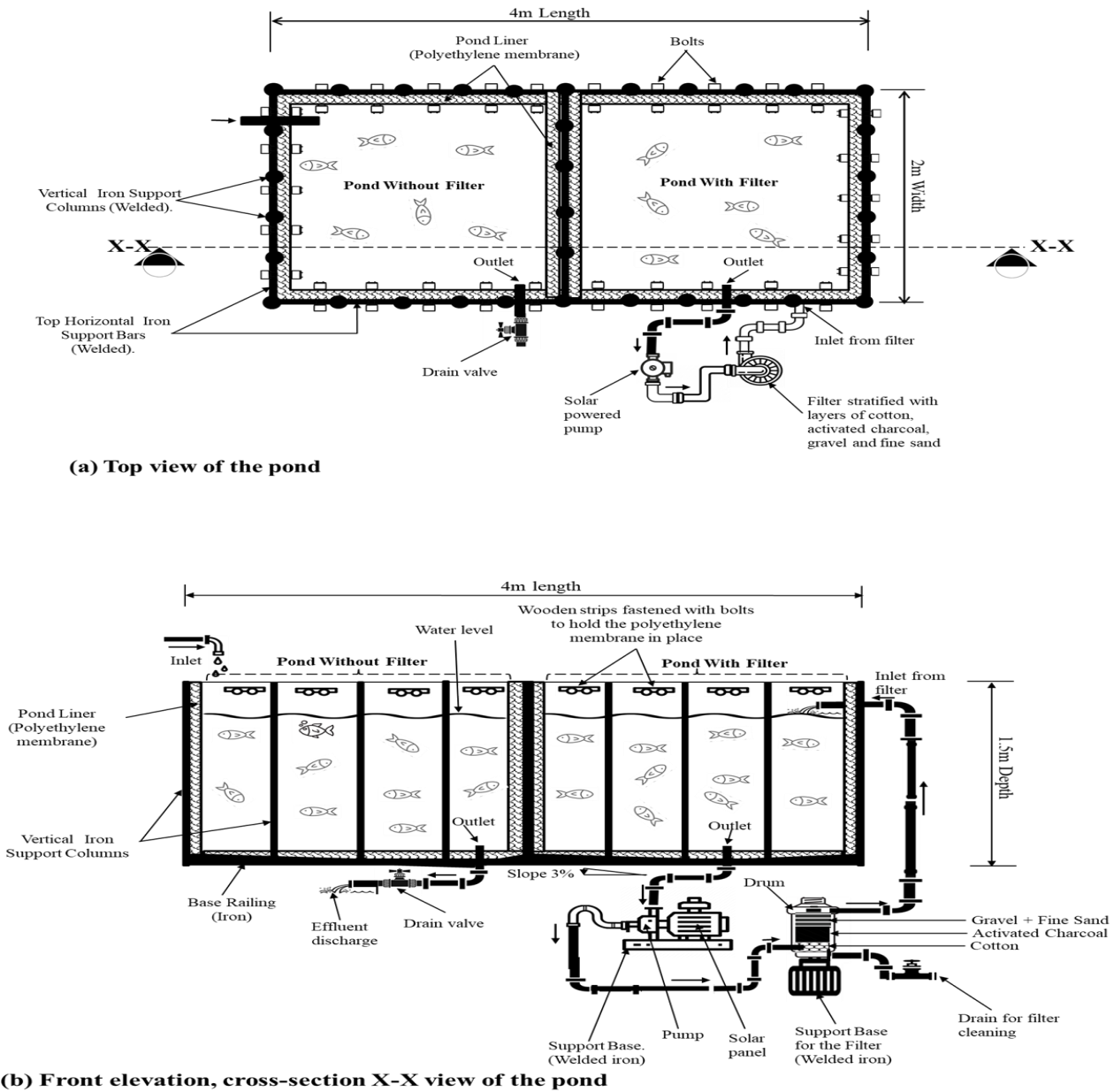
Figure 2: Conceptual climate-smart pond structural design

(a) Iron frame structure set above ground

(b) Pond liner attached to the supporting frame

(c) Pond with filtration system, showing water pump and 55-gallon barrel with cotton, charcoal, gravel and fine sand
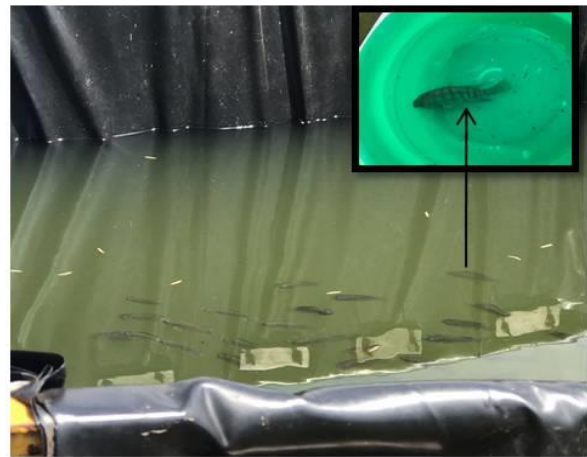
(d) Nile tilapia in the pond

Figure 3: Prototype climate-smart pond design in Morogoro

(iii)   Fish Growth Evaluation

Fish Stocking

In this study, Nile tilapia was used for the growth performance evaluation in the new climate-smart pond prototype. Three hundred fish seeds were obtained from a government Hatchery Center located at Kingolwira, Morogoro region in February 2020. Immediate transportation of the fish seeds in oxygen-filled polythene bags to SUA ensured a 100% survival rate. In SUA, a small amount of pond water was added to the oxygen-filled bags to help the fish seed acclimatize before releasing them into the water. Both the newly designed climate-smart ponds without filter and with filter each received about one hundred fish seeds and the remainder was given to a farmer whose pond was used as the control and was asked to raise the fish following aquaculture practices he normally uses with his other stock. The fish were cultured for six months, during which water quality parameters

were recorded from all ponds every seven days. The feed formulation used in all 3 ponds was similar (a home-made mixture of grain and insect-protein based fish feed), and the feeding was adjusted biweekly at the rate of 3% of fish body weight as they grew.

Analysis of Water Quality in Pond Using Physico-Chemical Parameters

Physicochemical water quality parameter analysis (e.g. nitrate, ammonia, alkalinity, Biological Oxygen Demand (BOD), and hardness) were done using Water test kits (Tetra Gmbh, Germany) and according to the manufacturer's instructions. Water pH, temperature, and Dissolved Oxygen (DO) were measured using a multi-parameter probe (Eco pond supply, USA).

3.6.6 Fish Feed Formulation

(i) Study Location

This study was carried out between in Morogoro, Tanzania, at the Department of Food Technology, Human Nutrition, Sokoine University of Agriculture (SUA). A climate-smart pond with water filtration mechanism was constructed and stocked with Nile tilapia while the control was a conventional concrete fish pond owned by a farmer located in Mikese, Morogoro. Morogoro region is located in the eastern zone of Tanzania (6.8278° S, 37.6591° E).

(ii) Rearing and Harvesting of Black Soldier Fly Larva

The substrate was a mixture of kitchen waste comprising of cooked corn meal, sweet potatoes, cassava, rice, fish bones, fruit and vegetable pieces such as ripe tomatoes, pawpaw, orange peels, jackfruit rind, cabbage and collard greens. These were placed in three separate 55-gallon plastic containers, each holding 10 kg of waste material. The moisture content of the substrate was deemed sufficient for optimal larva growth. The containers were covered with a lid provided with a single ventilation hole and the waste left to ferment, decompose and attract flies. The eggs laid by female black soldier flies hatched into larvae within two days. On hot sunny days, the containers had average temperature of 27 ± 1 °C and a relative humidity of $85 \pm 3\%$. Fresh kitchen waste material was added into each container after three days and the larva harvested six days after the first one had appeared. Some larvae were left in the same container and provided with fresh kitchen waste substrate, and the above procedure repeated five times with 6-day intervals. The collected larvae from each container were pooled, washed with tap water and oven-dried until constant weigh, then milled into a meal and stored in an air-tight container at room temperature prior to use.

(iii)   Moringa Oleifera and Basil leaf Powders

Both Moringa oleifera and basil leaf powders were obtained according to moringa flour production method of Hèdji *et al.* (2014) but with modifications. Briefly, fresh moringa leaves and basil leaves were collected from a farm in Pangawe, Morogoro, washed with tap water, sun-dried, separately ground into fine powders using an electric coffee grinder (Krups Inc., New Jersey, USA), sieved, transferred into clean airtight containers and stored at room temperature until use

(iv)   Feed Formulation

Google Sheets Program, a web-based software office suite offered by Google, integrated with the USDA National Nutrient Database for Standard Reference (Table 17), was used in developing the fish feed formulations (Table 18). Nutrasheets^{TM}, a recipe formulation add-on for Google Sheets, was used to custom batches, add ingredients such as black soldier fly larva missing in the USDA database and compute nutrition facts and formulation costs. Ingredients such as rice bran, corn bran and sunflower seedcake were purchased from local flour mills and ground into fine powder using a coffee grinder (Krups Inc., New Jersey, USA). Flour sifter helped discard coarse materials. Spirulina powder and vital wheat gluten were sourced from the local stores; while black soldier fly larva meal, moringa leaf powder and basil leaf powder were home-made as described above and used in the feed formulations. Bench-top feed preparation was done by combining these ingredients to form diets (Table 18) meeting nutrient requirement of fish as computed using the Google Sheets Program (Table 19). Ranaya Fish Pellet was purchased from animal feed store in Morogoro and used as the control diet.

(v)   Preparation of Feeds

For the experimental diet (Table 18), the dry ingredient powders were individually weighed (Digital Kitchen Scales, USA) into plastic bowls and then blended together in a saucepan for 3 minutes to obtain a homogeneous powder to which warm tap water was added and kneaded for 5 minutes to obtain a dough of about 35% moisture. Feed was extruded using a bench-top pasta roller (Kitchen Aid, USA) to form 3-mm "spaghetti" strands and then cut into about 3-mm pellets using a knife. The pellets were placed on a cookie sheet and then sun-dried to achieve approximately 10% moisture prior to storage in labeled air-tight plastic containers at room temperature until fed to the fish. Pellet stability in water was measured using methods described by Webster *et al.* (1994). Growth performace and feed utilization of the formulated diet were evaluated in Nile tilapia over 3-month period (Table 20).

(vi) Ponds and Water Quality

The feeding trial was conducted in a newly designed climate-smart prototype fish pond having a vertical water filtration and recycling mechanism stratified with layers of cotton, activated charcoal, gravel and fine sand for solids removal. It was set up at Sokoine University of Agriculture (SUA), Morogoro, Tanzania, in February 2020, prior to the start of the experiment. Water was circulated back to the pond through the filter at a rate of 530 liter per hour at three feet of head. Water aeration was aided by a 1-foot drop and turbulence upon return into the pond. Uneaten diet and feces trapped at the bottom of the filter were removed through a discharge valve.

Water quality parameters reported, that is, nitrate, ammonia, alkalinity, Biological Oxygen Demand (BOD), and hardness) were done using Water test kits (Tetra Gmbh, Germany). Water pH and Dissolved Oxygen (DO) was measured using a probe (Eco pond supply, USA) while temperature was measured using a thermometer. Water quality parameters measured in the climate-smart fish pond for the duration of the study averaged: temperature (27.4 ± 0.50 °C), pH (6.6 ± 0.28), nitrate (1.6 ± 0.34 mg/L), ammonia (0.2 ± 0.11 mg/L), DO (7.6 ± 0.77 mg/L), BOD (4.3 ± 0.46 mg/L), alkalinity (23.2 ± 1.89 mg/L), hardness (105 ± 3.09 mg/L), and turbidity (3.9 ± 0.58 Nephlometric Turbidity Units [NTU]), and were optimal for growth and survival of Nile tilapia (Djissou *et al.,* 2017).

A local farmer's concrete pond was used as the control; the fish were fed using a commercial diet. Water quality parameters of the control pond measured for the duration of the trial averaged: Temperature (27.2 ± 0.43 °C), pH (9.7 ± 1.94), nitrate (14.7 ± 3.23 mg/L), ammonia (1.6 ± 0.71 mg/L), DO (4.3 ± 1.75 mg/L), BOD (29.6 ± 4.94 mg/L), alkalinity (57.4 ± 10.36 mg/L), hardness (264 ± 16.92 mg/L), and turbidity (38.0 ± 5.08 NTU).

(vii) Fish Stocking and Feeding Trial

About 100 Nile tilapia (*Oreochromis niloticus*) fish seeds were obtained from Hatchery Center, a government hatchery located at Kingolwira, Morogoro region in February 2020 and acclimated to the water conditions of the climate-smart experimental pond (volume 4 m^3) by gradually introducing small quantities of the pond water into the hatchery water containing the fish seeds so that environmental changes did not exceed 1°C and 1 ppt salinity every 30 min. To establish a baseline and ensure that all the fish seeds were nutritionally equivalent, an introductory diet was fed to the fish twice a day for 10 days prior to the start of the study. Once acclimated, the initial average weight of the Nile tilapia seeds were determined as 3.40 ± 0.97 g and feeding started using test diet on Table

18. The feeding regimen was: 7% of their body weight for six weeks and 5% of their body weight from 7 to 12 weeks using the experimental BSFL diet.

(viii) Proximate Analyses

The determination of moisture, crude ash, crude proteins, crude lipids (ether extract), and crude fiber in both the experimental and commercial diets were carried out in triplicate according to the Association of Official Agricultural Chemists (AOAC) (1990) standard methods. Dry matter (DM) was determined by drying 2 g of triplicate samples to constant weight in an oven at 105 ℃ overnight (12 h). Crude ash was determined by incineration at 550 °C for 4 h in a combustion oven (International Organization for Standardization [ISO], 2002). Nitrogen content was determined by the standard Kjeldahl nitrogen method following Dumas principle (ISO, 2008). Crude protein content in experimental and commercial diets were calculated by multiplying total N by 6.25 (Finke, 2007). Crude lipid (CL) content (ether extract, EE) was quantitatively determined after extraction with diethyl ether with a Soxhlet system (ST 243 SoxtecTM, Hilleroed, Denmark) (ISO, 1999). Crude fiber (CF) content was determined in triplicate according to the AOAC (1990) standard method 962.09. Finally, the nitrogen-free extract (NFE) content was computed by subtracting the sum of crude ash, CP, CL, and CF from the respective DM values.

(ix) Growth Performance and Feed Efficiency

During the study, fish weight gain and length increase were measured every two weeks using a digital balance and meter rule, respectively, and used to compute growth and nutrient utilization response parameters below following Olvera-Novoa *et al.* (1990) methods.

Mean weight gain (g) = Final mean weight (g) – Initial mean weight (g).

Average daily weight gain (g) = Mean weight gain/length of feeding trial.

Feed intake (g) = Amount of feed throughout the experiment.

Protein intake (g) = Total feed consumed/% crude protein in feed.

Specific growth rates: $\text{SGR}\% = 100 \times \frac{\ln[\text{FBW}] - \ln[\text{IBW}]}{t}$

Where t = time in days and IBW and FBW are Initial Body Weight and Final Body Weight, respectively.

Feed conversion ratio: $FCR = \dfrac{\text{dry feed intake (g)}}{\text{final weight gain (g)}}$

Protein efficiency ratio: $PER = \dfrac{\text{Net body weight gain (g)}}{\text{Amout of protein intake (g)}}$

3.7   Statistical Analysis

Descriptive statistics were used to give summaries such as the mean and standard deviation of weight and length of fish and other nutrient utilization parameters. Analysis of variance (ANOVA) was used to test the study hypotheses. The R software was used to statistically analyze the the parameters.

3.7.1   Farmer's Knowledge of Aquaculture Management Practices Data

Data were entered into R-software for descriptive analysis (mean and SD). For comparative purposes, the type and frequency of reported diseases and chemical use were presented as ratios. The chemical and biological products were grouped into antimicrobials, disinfectants, and parasiticides.

Multivariate analyses were used to evaluate correlations between (1) respondents, farm characteristics (independent variables), and reported diseases (dependent variable) in the farms and, (2) reported diseases (independent variables), and the chemical treatments used (dependent variable). Redundancy Analysis (RDA) was used to test for the significance of any correlation between the independent variables and the variance in the dependent variable dataset. The correlation of the tested independent variable was considered significant when $p \leq 0.05$. Individual bi-plots were constructed only for those independent variables that showed significance at $p \leq 0.05$.

3.7.2   Prevalence of Fish Parasites and Physicochemical Characteristics of Pond Water

Simple descriptive statistics were used to give summaries such as the mean and standard deviation of weight and length of fish and other physicochemical parameters. Analysis of variance (ANOVA) was used to test the study hypotheses. The R software was used to statistically analyze the prevalence of fish parasite infestation in the two regions.

3.7.3   Occurrence of Fish bacteria Pathogens

The obtained data were entered and organized in Ms. Excel sheets and exported to R-statistical software for analysis using the Chi-square test ($p = 0.05$). Multivariate analysis was used to evaluate the correlation between independent variables (sampling sites, production system, farmed species, and feeding types) and bacteria occurrence (dependent variable).

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3.7.4 Bioaccumulation and Distribution Pattern of Heavy Metals

All metal concentrations were determined on a dry weight basis. The heavy metals concentration in the study locations were analyzed using R statistics software, Analysis of Variance (ANOVA) was applied and the means were compared using Duncan's multiple range test at a 5% significance level.

3.7.5 Design of a Context-specific Climate-smart Sustainable Fish Pond

Simple descriptive statistics using R- software were used to give summaries such as the mean and standard deviation of weight and length of fish and other physicochemical parameters.

3.7.6 Fish Feed Formulation

All growth data were subjected to analysis of variance (ANOVA). The significance ofdifference between means was determined by Duncan's multiple range test ($P<0.05$) using R-software. Values are expressed as means $\pm$ SE.

3.8 Ethical Consideration

The study was approved by the Tanzania National Institute for Medical Research (NIMR) and was given an ethical clearance certificate with a reference number KNCHREC00025 (Appendix 3). Fish farmers signed an informed consent which clearly explained the aim, procedure, benefits of the study (Appendix 2). Anonymity was ensured using numbers to represent the names of an individual fish farmer during questionnaire handling.

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 Results

4.1.1 Farmer's Knowledge of Aquaculture Management Practices

(i) Farmers' Background Information and Farm Characteristics

The majority of the respondents surveyed in both regions were farm owners (75%) while the rest (25%) were farm managers. According to the data collected, 43% of the farmers surveyed in Arusha region had some university degrees compared to only 23% of those from the Morogoro region. Likewise, the high number of farmers with degree holders in Arusha region also correlated with a high number (76%) of those with aquaculture knowledge compared to those from Morogoro region (9%). Those farmers with aquaculture knowledge also reported having attended some training, short courses, and/or workshops organized by various stakeholders such as universities, Non-Government Organization (NGO), and fish feed companies to supplement their knowledge.

Fish production practices in both regions were also taken into consideration. About 74% of Morogoro fish farmers raise their fish in concrete ponds whereas 40% of Arusha farmers use earth ponds. Farmers in both regions, however, seemingly use the monoculture production method. According to the findings, tilapia and catfish are the only fish species farmed in both regions (Table 1). Among the farmers surveyed in Arusha region, the production of tilapia and catfish is 83% and 17%, respectively; whereas their numbers in Morogoro region are found to be 71% and 29%, respectively, suggesting the farmers mostly preferred tilapia farming in both regions.

Pond sizes also varied in both regions. The majority (80%) of Morogoro region farmers own pond size ranging from 20 to about 100 M^2 with an average capacity of 100-1000 fingerlings. None of the farmers surveyed had ponds larger than 200 M^2 in size (Table 1). However, some Arusha region farmers own ponds ranging from 100 to over 200 M^2 with average initial fingerlings ranging from 100-30 000, indicating water volume impact stocking capacity.

In this research, it was found that the majority (82%) of fish farmers in both regions produced their fish feed. To reduce cost, they used single or a mixture of locally available feed ingredients such as maize bran, sardines, wheat bran, cassava meal, and sunflower seedcake.

Unfortunately, most of the fish farmers surveyed had no formal training on fish feed formulation, processing, handling, and storage techniques. Many had little knowledge or understanding of

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restrictive feeding techniques and break feeding schedules. The lack of know-how of fish feed requirements could be the reason why most farmers realized undersize fish despite the regular feeding of their stock.

Semi-intensive feeding was the most common type of feeding practised. All the fish farmers incorporated manure to increase the production of natural food organisms such as phytoplankton, zooplankton, and insects to supplement the fish diet.

Table 1: Farmers' background information and farm characteristics

	Variable	Morogoro region N=65	Arusha region N=65	Mean	χ^2	F-test
Respondent characteristics	Role in the farm[a]	O(85); M(15)	O(66); M(34)	O(75); M(25)		
	General education level[b]	NO(22); PS(25); SS(31); U(23)	NO(8); PS(22); SS(28); U(4)	NO(15); PS(23); SS(29); U(33)		
	Aquaculture knowledge[c]	NO(8); UE(83) TA (9)	NO(4); UE(21); TA(76)	NO(6); UE(52); TA(42)	1.5472**	
Farm characteristics	Aquaculture type[d]	C(74); EP(26)	C(60); EP(40)	C(56); EP(44)	2.8249*	
	Production practices[e]	M(89); P(11)	M(90); P(10)	M(90); P(10)	-	
	Farmed species[f]	B(11); C(29); T(61)	B(9); C(5); T(87)	B(10); C(17); T(74)	1.4126ns	
	Pond size(M^2)[g]	20-100(80); 101-150(12);151-200(8); >200(0)	20-100(45); 101-150(26);151-200(5); >200(25)	20-100(62); 101-150(19); 151-200(6); >200(12)		1.305ns
Production	Initial number of fishes[h]	<100(1); 101-1000(80); 1001-2000(17); 2001-3000(2); >3001(0)	<100(0); 101-1000(55); 1001-2000(36); 2001-3000(5); >30001(5)	<100(0.5); 101-1000(67.5); 1001-2000(26.5); 2001-3000(3.5);>3000(2.5)		0.3137ns
	Total Fish harvest[i]	<65(2); 66-1000(82); 1001-2000(14); 2001-3000(2); >3000(0)	<100(0); 101-1000(56); 1001-2000(36); 2001-3000(5); >3000(3)			0.2735ns
	Feeding[k]	C (14); FS 9); HM (77)	C (12); FS (2); HM (86)	C (13); FS (5); HM (82)	2.2391ns	

Note: Statistically significant at *P < 0.05, **P < 0.01, ***P < 0.001; ns = not significant.

All information showed in this table was collected during the chemical use interviews

Numbers in parentheses is the percentages

[a] O: Owner; M: Manager.

[b]NO: None; PS: Primary school; SS: Secondary School; U: University.

[c]NO: None; UE: Untrained with aquaculture experience; TA: Trained in aquaculture.

[d]C: Cage; EP: Earth pond.

[e]M: Monoculture; P: Polyculture.

[f]C: Catfish; T: Tilapia; B: Both catfish species.

[k]C: Commercial; FS: Food scraps; HM: Homemade.

(ii)   Farm Water Management

Figure 4 (a-d) shows the result of water resources and management efforts in Arusha and Morogoro regions. According to the findings, most of the fish farmers depend on three main sources of water. Majority (approximately 45%) of the farms used boreholes as their major source of water followed by tap water (40%) (Fig. 4a).

All aspects of water treatment play a significant role in intensive fish production. Unfortunately, most of the fish farmers didn't have technical knowledge and equipment and lacked basic knowledge for testing the quality of their water supply. Consequently, about 87% of all farmers surveyed did not treat their water prior to stocking (Fig. 4b) and only about 2% of them changed their pond water regularly (Fig. 4c).

In this research, it was found that the fish farmers from both regions had poor wastewater management. Sadly, about 95% of the fish farmers released untreated wastewater freely into the environment (Fig. 4d).

A
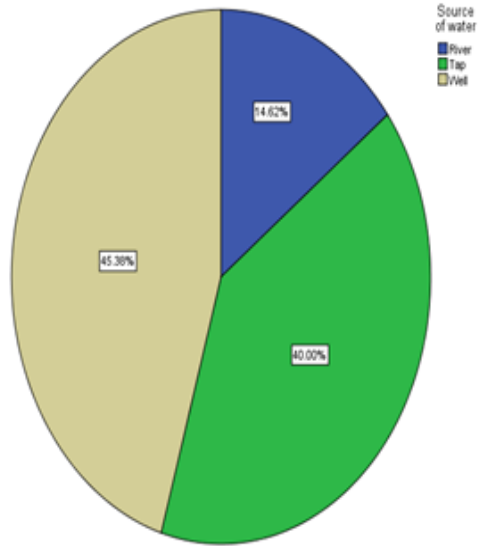
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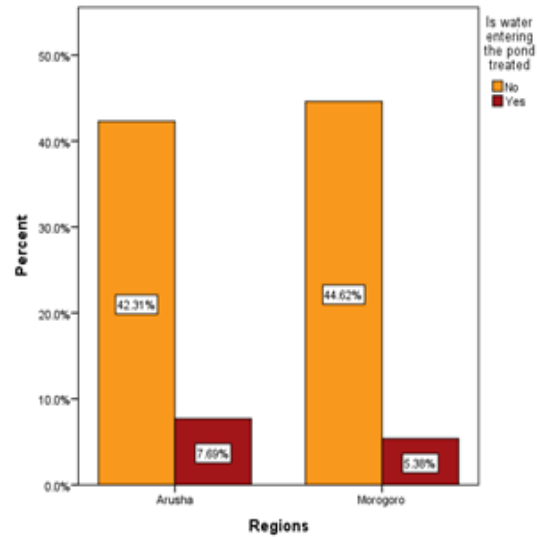
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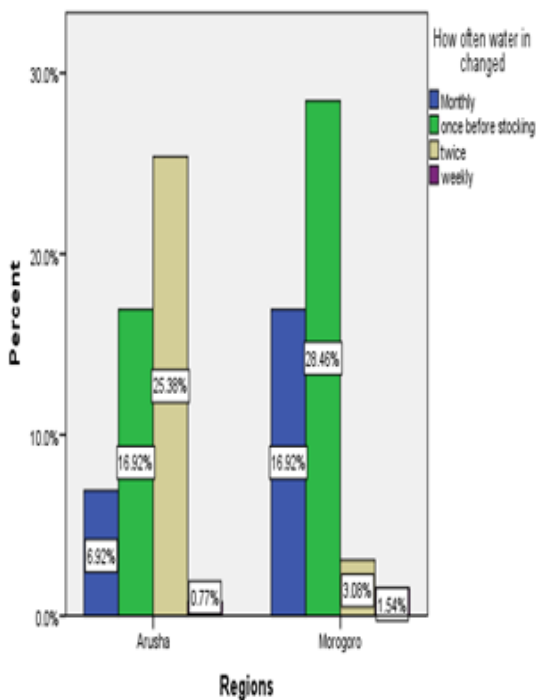
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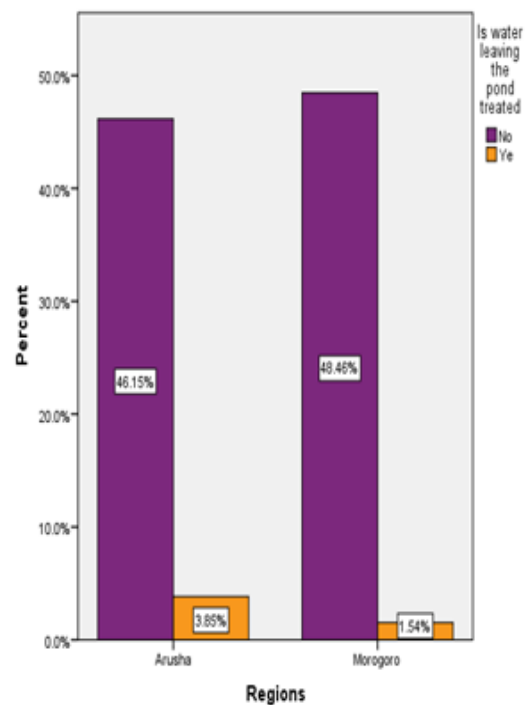
Figure 4: Water management in Arusha and Morogoro fish Farms

(iii)   Disease Occurrence and Diagnostic Capacity

Disease occurrence was reported by fish farmers from both regions (Fig. 5), which could lead to significant production losses. Unfortunately, the majority of the fish farmers had neither disease diagnostic equipment nor health management plan for preventing and treating diseases in case of outbreaks. It observed that less than 21% of the surveyed farmers kept written records on the initial

number of fingerlings stocking, final fish harvested, water management, diseases diagnosed, chemicals applied, and purpose of such application.

There was a statistical correlation between aquaculture knowledge and disease occurrence, suggesting proper aquaculture education, training, and application of good aquaculture management practices can result in the ultimate health protection of fish in aquaculture (Fig. 6). There was no significant correlation between pond size, pond structure, stocking density, feeding types, and general education level. Those farmers with aquaculture knowledge were also more likely to adhere to veterinarians and/or fish technicians on chemical usage.
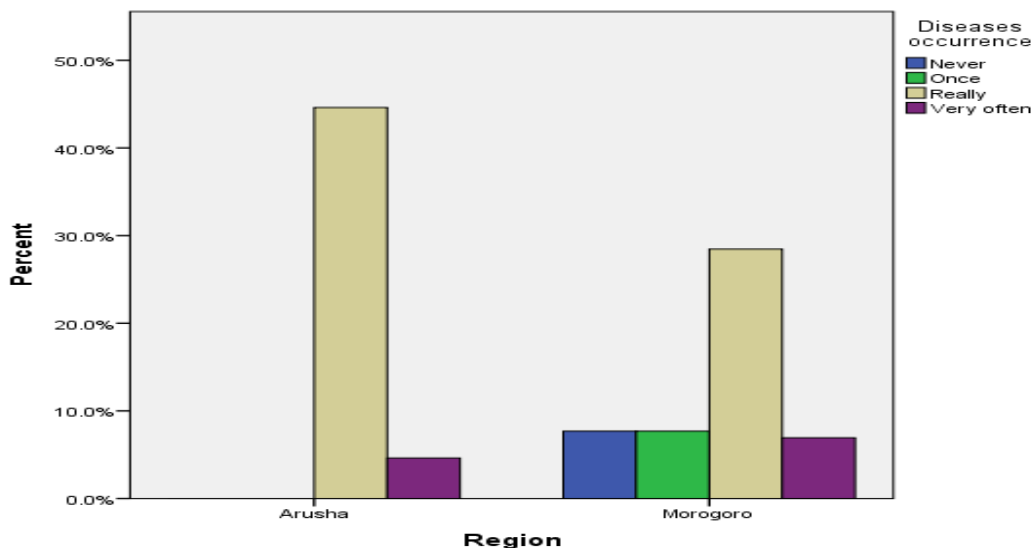
Figure 5: Frequency of disease occurrence in Arusha and Morogoro surveyed fish farms
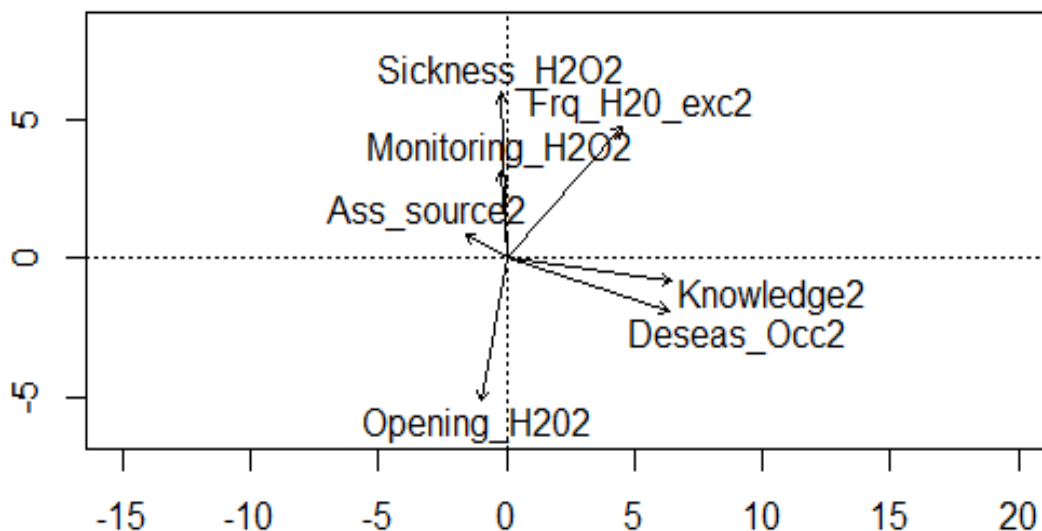
Figure 6: Ordination diagram (redundancy analysis; RDA)

(iv)   Disease Treatment Practices

In this study, the number of chemical agents used by the farmers for disease control in Arusha and Morogoro fish ponds was found to vary considerably (Table 2). However, farmers' intentions were similar: disease prevention. Overall, antibiotics for disease control were used heavily by fish farmers in both regions (Table 2). Farmers in Morogoro region most preferred oxytetracycline, sulfadiazine, and trimethoprim (Fig. 7) while those in Arusha region preferred oxytetracycline, gentamycin, and florfenicol (Fig. 8). In general, oxytetracycline was the most preferred antibiotic in both regions (Fig. 7 & 8).

There are several disinfectants fish farmers can use in treating their ponds. In this study, it was found that chlorine, formaldehyde, hydrogen peroxide, and iodine solutions were the most common disinfectants used by fish farmers in Morogoro region (Fig. 7a) while those in Arusha region mostly preferred iodine solutions (Fig. 8) for water treatment before stocking and throughout production.

Sanitation of equipment is also essential in preventing the introduction of pathogens to aquaculture facilities. Pesticides such as calcium hypochlorite were used to disinfect farmers' protective gear including boots and other farm equipment.

The fish farmers surveyed in both regions reported that to control internal parasites, they predominantly used a parasiticide called mebendazole (Fig. 7 & 8). The other commonly used parasiticides in both regions were copper sulfate and trichlorfon but to a lesser degree compared to mebendazole, while azadirachtin was only used by the fish farmers in Arusha.

In this study, no fish farmer in any region reported using the internationally banned antimicrobials such as chloramphenicol, fluoroquinolones, nitrofurans, and quinolones classes of antibiotics. It was also found that the choice of antibiotics used by most of the fish farmers was based on experience. Unfortunately, most of the fish farmers reported not to follow the dosage recommendation provided by the suppliers of the chemical agents.

Table 2: Summary data on the use of antibiotics, disinfectants, and parasiticides in the surveyed farms: total number of recorded compounds (n) and percentage of farms that use them (% use)

		Arusha region	Morogoro region
Antibiotics	Total number of recorded compounds (n)	5	5
	Percentage of farms that use them (%)	30.76	15.38
Disinfectants	Total number of recorded compounds (n)	5	4
	Percentage of farms that use them (%)	18.46	12.30
Parasiticides	Total number of recorded compounds (n)	4	3
	Percentage of farms that use them (%)	9.23	10.76
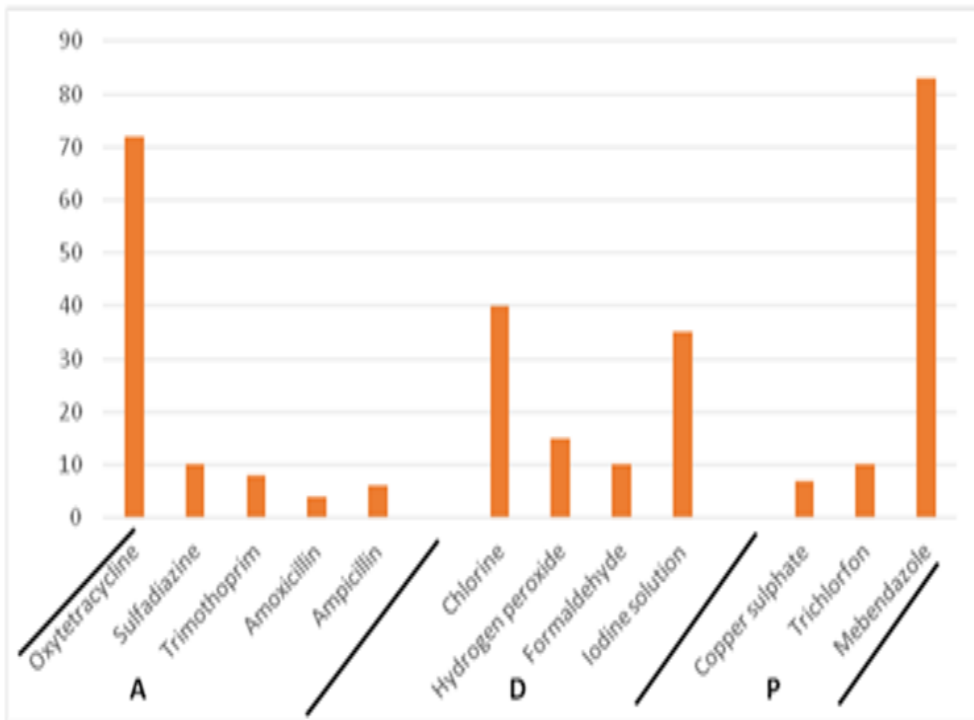
Figure 7: **Percentage of farmers using antibiotics (A), Disnfectants (D) and Parasiticides in Arusha region studied farm groups**
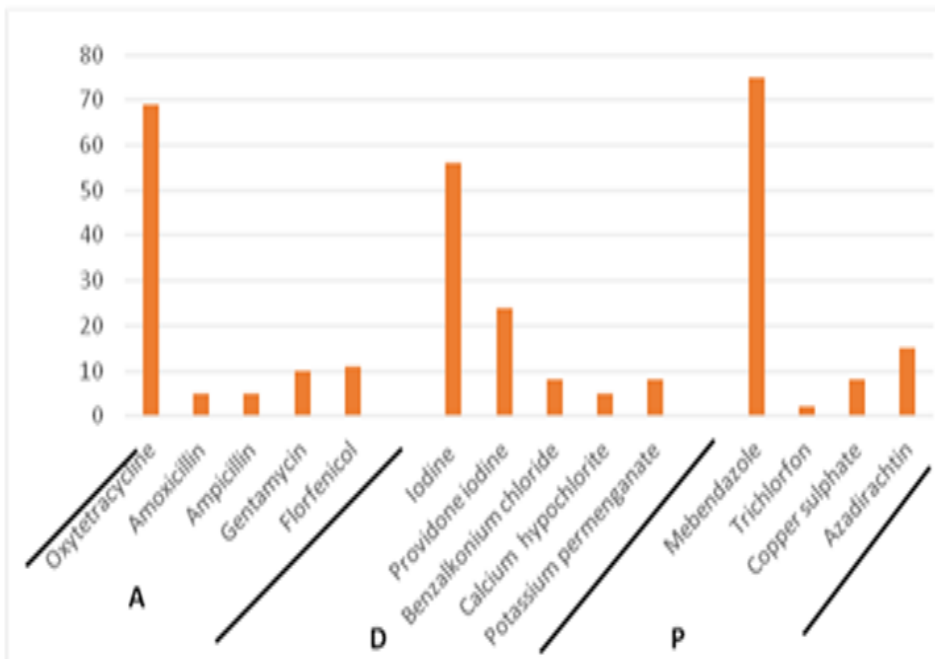
Figure 8: Percentage of farmers using antibiotics (A), Disinfectants (D), and Parasiticides (P) in Morogoro region studied farm groups

4.1.2 Prevalence of Fish Parasites in Nile Tilapia (*Oreochromis niloticus*) and African Catfish (*Clarias gariepinus*) and Physicochemical Characteristics of Pond Water

(i)   Physicochemical Analysis of Pond Waters

The mean water temperature in Arusha and Morogoro regions was 25.9 ± 0.2 °C and 27.1 ± 0.2 °C, respectively (Table 3). The mean pH of pond water sampled in Arusha and Morogoro regions were 9.8 ± 1.4 and 7.2 ± 0.8, respectively, and were highest in all sites studied in Arusha (8.6 ± 0.9 to 11.2 ± 0.8) compared to the latter (6.4 ± 0.1 to 8.3 ± 03) (Table 3).

The mean alkalinity of the pond water sampled from all sites studied in Arusha and Morogoro regions was 48.6 ± 22.3 mg/L and 44.6 ± 14.6 mg/L, respectively, and were highest in Nambala (79.8 ± 11.1 mg/L) and Maweni (59.1 ± 15.0 mg/L) in Arusha region as well as Mikese (55.5 ± 8.2 mg/L) and Langali (53.6 ± 15.0 mg/L) in Morogoro region (Table 3).

The mean nitrate levels reported in pond water in Arusha and Morogoro regions were 8.4 ± 5.2 mg/L and 4.9 ± 2.4 mg/L, respectively, and were highest in Samalia (13.4 ± 2.8 mg/L), Manyata (12.8 ± 2.4 mg/L) and Maweni (10.7 ± 1.8 mg/L) in Arusha followed by Tangeni (8.9 ± 2.1 mg/L) and Langali (4.8 ± 0.9 mg/L) in Morogoro region (Table 3). The mean ammonia levels in pond water samples studied from Arusha and Morogoro regions were 1.3 ± 0.9 mg/L and 1.2 ± 0.9 mg/L, respectively, and were highest in Samalia (1.9 ± 1.0 mg/L) and Nambala (1.6 ± 0.9 mg/L) in Arusha as well as Mikese (1.6 ± 1.0 mg/L) in Morogoro region (Table 3).

The mean DO report in Arusha and Morogoro regions were 4.0 ± 1.3 mg/L and 6.0 ± 1.93 mg/L, respectively (Table 3). Sites with ponds that had DO levels above 5 mg/L were Mkindo (9.1 ± 4.1 mg/L), Mikese (6.1 ± 2.7 mg/L), Langali (5.9 ± 2.3 mg/L), and Tangeni (5.3 ± 2.3 mg/L), all located in Morogoro region (Table 3).

On the other hand, Biological Oxygen Demand (BOD) indicates biodegradable organic content in the pond water. The recommended BOD level in pond water is 20 mg/L (Boyd, 2003). In this study, it was found that the mean BOD levels in Arusha and Morogoro regions were 19.4 ± 6.3 mg/L and 29.8 ± 6.0 mg/L, respectively (Table 3). All sites studied in Morogoro region had BOD levels above the recommended 20 mg/L concentration.

In this study, turbidity varied considerably from site to site and ranged from 3.2 ± 1.5 to 15.8 ± 5.5 NTU and 12.3 ± 3.9 to 25.7 ± 9.8 NTU in sites sampled in Arusha and Morogoro region, respectively (Table 3). These levels are ideal for tilapia and catfish farming. Ponds with clear

waters were mainly found in Manyata (3.2 ± 1.5 NTU) followed by Maweni (4.6 ± 3.3 NTU), both in Arusha region (Table 3).

Finally, water hardness was measured since it indicates calcium concentration in pond water. In our study, water hardness varied significantly from site to site and ranged from 60 – 260 mg/L and 189 – 300 mg/L in Arusha and Morogoro region, respectively (Table 3). The highest and lowest hardness values were found in Tangeni (300 ± 57.3 mg/L) in Morogoro and Samalia (60 ± 22.7 mg/L) in Arusha region.

Table 3: Mean physicochemical water parameters in fish ponds in sites within Arusha and Morogoro regions

Region		Arusha (n = 65)						Morogoro (n = 65)					
		Kikwe	Nambala	Maweni	Manyata	Samalia	Overall Arusha mean	Kihonda	Langali	Mikese	Mkindo	Tangeni	Overall Morogoro mean
Village (site)		n for each site = 13						n for each site = 13					
Water quality parameter	Temperature (°C)	25.6±0.1	26.0±0.1	25.9±0.1	25.8±0.1	26.0±0.1	25.9±0.2	27.2±0.4	26.9±0.1	27.0±0.1	27.2±0.2	27.0±0.1	27.1±0.2
	pH	9.2±0.8	8.6±0.9	11.2±0.8	9.9±1.6	10.3±1.3	9.8±1.4	6.4±0.1	6.9±0.1	8.3±0.3	7.5±0.7	6.8±0.4	7.2±0.8
	Nitrate (mg/L)	3.2±0.9	2.1±0.7	10.7±1.8	12.8±2.4	13.4±2.8	8.4±5.2	3.9±0.8	4.8±0.9	3.7±0.7	3.1±0.7	8.9±2.1	4.9 ± 2.4
	Ammonia (mg/L)	1.2±0.7	1.6±0.9	1.0±0.7	1.0±0.6	1.9±1.0	1.3±0.9	1.0±0.5	1.3±0.8	1.6±1.0	1.0±0.8	1.0±1.1	1.2±0.9
	DO (mg/L)	3.7±1.1	4.3±1.8	4.3±1.5	3.6±0.8	4.2±1.1	4.0±1.3	3.8±1.4	5.9±2.3	6.1±2.7	9.1±4.1	5.3±2.3	6.0±3.1
	BOD (mg/L)	12.7±2.7	16.8±5.5	19.3±5.4	22.4±4.3	25.6±4.4	19.4±6.3	34.2±4.3	29.4±5.0	32.6±5.2	29.8±4.4	23.1±4.8	29.8±6.0
	Alkalinity (mg/L CaCO₃)	35.8±8.7	79.8±11.1	59.1±15.0	45.7±9.1	22.4±7.3	48.6±22.3	36.2±7.0	53.6±15.0	55.5±8.2	48.7±11.2	29.2±10.3	44.6±14.6
	Turbidity (NTU)	8.9±4.3	15.8±5.5	4.6±3.3	3.2±1.5	15.8±8.5	9.7±7.4	19.8±6.4	25.7±5.7	12.3±3.9	17.8±6.4	25.6±9.8	20.2±8.3
	Hardness (mg/L CaCO₃)	125±26.1	260±77.9	140±32.7	78.0±23.5	60.0±22.7	133±81.6	200±33.0	195±46.8	220±41.7	189±73.3	300±57.3	221±65.3
Fish measureme	Fish length (cm)	15.7±5.5	17.5±5.8	15.4±6.0	13.7±6.1	13.1±4.8	15.1±5.7	12.5±4.4	13.5±6.0	14.1±6.7	14.8±5.5	13.2±5.5	13.6±5.6
	Fish weight (g)	267.1±97	277.0±133	243.5±131	199.4±145	212.2±135	239.8±129	218.7±116	216.4±119	206.8±135	217.0±118	211.4±122	214.1±119

Values are mean ± standard deviation; n is the number of ponds; DO = Dissolved Oxygen; BOD = Biological Oxygen Demand; mg/L = concentration expressed in milligrams per liter; NTU = Nephelometric Turbidity Units, a turbidimeter (nephelometer) measurement of light intensity as a beam of light passes through a water sample at 90 degrees; cm = centimeter; g = grams.

(ii)   Prevalence of Parasites in Nile Tilapia and African Catfish in Arusha and Morogoro

The standard length of fish captured in this study varied because ponds were at different production cycles and ranged from 8 cm to 23 cm (mean 15.1 ± 5.7 cm) and 6 cm to 22 cm (mean 13.6 ± 5.6 cm) in Arusha and Morogoro regions, respectively; while their weights in those two regions ranged from 46 g to 412 g (mean = 239.8 ± 129 g) and 30 g to 395 g (mean = 214.1 ± 119 g), respectively (Table 3).

Overall, seven parasite species were recovered in both Nile tilapia and African catfish samples studied in both regions. The thorny headed worm *Acanthocephala* sp was the most prevalent (Arusha (49.2%); Morogoro (50.7%)); while leeches were the least (Arusha (4.6%); Morogoro (7.6%)) occurring parasites in both Nile tilapia and African catfish samples studied (Table 4). Under light microscopy, *Acanthocephala* sp is small, bilateral symmetrical worms with a retractable spined proboscis. Leeches had body segmentation, with an anterior and a rear sucker differentiating them from common free-living annelids.

The overall prevalence of digenean trematode *Diplostomum* sp (eye flukes), recovered from the vitreous humour of the eyes of fish, were found as 36.9% and 38.4% in Arusha and Morogoro regions, respectively, especially in African catfish samples from Arusha (72.7%). *Diplostomum* sp had a cup-shaped front structure with suckers, with immature gonads contained in a cylindrical hind body.

The overall prevalence of nematode *Contracaecum* sp recovered from the intestines of fish, were found as 49.2% and 41.8% in Arusha and Morogoro regions, respectively; again, with the highest occurrence in African catfish samples (Table 4). Visible to the naked eye, these worms are round, with a solid cuticle.

The overall prevalence of Ciliophora *Trichodina* sp a protozoan recovered from the skins of fish, was 44.6% and 41.5% in Arusha and Morogoro regions, respectively. Under light microscopy, *Trichodina* sp. had hooked ring-like denticles, appeared circular when observed dorsally, with a cup-shaped structure.

Finally, monogenean trematodes *Dactylogyrus* sp and *Gyrodactylus* sp were also recovered from the gills and skin of fish. The overall prevalence of *Dactylogyrus* sp. in Arusha and Morogoro regions was 47.6% and 32.3%; while the occurrence for *Gyrodactylus* sp in the fish was 36.9% and 47.6% in Arusha and Morogoro regions (Table 4). Under light microscopy, *Dactylogyrus* sp contained a scalloped head with anteriorly eye spots while *Gyrodactylus* sp had a V-shaped head, an opisthohaptor at the back end, and no eyespots. Statistically, there was no significant difference

($P < 0.05$) in the prevalence of the parasites recovered on both Nile tilapia and African catfish in the sites studied, except for *Diplostomum* sp in Arusha region and *Trichodina* sp. and *Diplostomum* sp in Morogoro region (Table 4).

Table 4: The occurrence of parasites on Nile tilapia and African catfish in Arusha and Morogoro

Parasites	Parasite occurrence, n (%)							χ^2
	Arusha				Morogoro			
	Nile tilapia ($n = 54$)	African catfish ($n = 11$)	Total ($n = 65$)	χ^2	Nile tilapia ($n = 46$)	African catfish ($n = 19$)	Total ($n = 65$)	
Endoparasites								
Acanthocephala sp.	26 (48.1)	6 (54.5)	32 (49.2)	0.301^{ns}	25 (54.3)	8 (42.1)	33 (50.7)	0.631^{ns}
Diplostomum sp.	16 (29.0)	8 (72.7)	24 (36.9)	2.306^{*}	22 (47.8)	3 (15.7)	25 (38.4)	1.426^{*}
Contracecum sp.	26 (48.1)	6 (54.5)	32 (49.2)	0.985^{ns}	17 (36.9)	10 (52.6)	27 (41.8)	1.011^{ns}
Ectoparasites								
Trichodina sp.	25 (46.2)	4 (36.3)	29 (44.6)	0.253^{ns}	19 (19.5)	8 (42.1)	27 (41.5)	1.73^{*}
Dactylogyrus sp.	26 (48.1)	5 (45.4)	31 (47.6)	0.868^{ns}	16 (34.7)	5 (26.3)	21 (32.3)	0.611^{ns}
Gyrodactylus sp.	20 (37.0)	4 (36.0)	24 (36.9)	0.023^{ns}	23 (50.0)	8 (42.1)	31 (47.6)	0.913^{ns}
Leeches	3 (5.5)	0 (0)	3 (4.6)	0.836^{ns}	4 (8.6)	1 (5.2)	5 (7.6)	0.920^{ns}

Legend:

n is the number of fish samples

Numbers in parentheses are the percentages (%) of detected bacteria from two different fish species.

Statistically significant at *$P < 0.05$; ns = not significant

(iii)   Prevalence of Fish Parasites Infesting Farmed Nile Tilapia and African Catfish in Select Sites within Arusha and Morogoro

The endoparasites recovered in the fish studied were *Acanthocephala* sp, *Diplostomum* sp and *Contracecum* sp. The highest occurrence of *Acanthocephala* sp. in Arusha and Morogoro region was found in Nambala (76.9%) and Tangeni (76.9%). From our results, this parasite least occurred in Samalia (23.0%) in Arusha region. Statistically, there was a significant ($P < 0.05$) difference in the occurrence of *Acanthocephala* sp between the individual sites sampled in both regions (Table 5). The highest occurrence of *Diplostomum* sp in Arusha region was found in ponds within Kikwe (53.8%) and Maweni (53.8%); while in Morogoro region, this parasite mostly occurred in Tangeni (53.8%) and Langali (46.1%). There was a significant ($P < 0.05$) difference in the occurrence of this parasite across the ponds studied in Arusha (Table 5). The highest occurrence of *Contracecum* sp in Arusha region were found in Maweni (76.9%), followed by Kikwe (69.2%); while in Morogoro

region, the most occurrence happened in Tangeni (76.9%), followed by Langali (38.4%), and Mikese (38.4%). There was a significant (P < 0.01) difference in the occurrence of *Contracecum* sp. across the ponds sampled in Arusha region, which was not observed in sites from Morogoro region (Table 5). Overall, the highest occurrences of endoparasites in this study were found in Kikwe, Nambala and Maweni in Arusha region as well as Tangeni in Morogoro region.

The four ectoparasites recovered in this study were *Trichodina* sp, *Dactylogyrus* sp, *Gyrodactylus* sp, and leeches. The highest occurrence of *Trichodina* sp in Arusha was found in Kikwe (76.9%) followed by Maweni (46.1%); while in Morogoro region, this parasite mostly occurred in Tangeni (61.5%), followed by Kihonda (53.8%). Prevalence of *Trichodina* sp significantly (P < 0.05) differed across sites studied in Arusha region, which wasn't the case in Morogoro region (Table 5). Furthermore, the results show that there was a higher *Dactylogyrus* sp infection in Arusha region compared to Morogoro, with the highest occurrence in ponds within Maweni (69.2%). Furthermore, it was found that the occurrence of *Gyrodactylus* sp, the other monogenean trematode, was higher in Maweni (84.6%) in Arusha region and Tangeni (76.9%) in Morogoro region, with no significant difference in their prevalence across all the sites studied (Table 5).

Lastly, leeches rarely occurred in the fish samples, and none existed on fish sampled from ponds within Kikwe, Maweni, Manyata, Kihonda, Langali and Mikese (Table 5). Overall, the highest occurrences of ectoparasites found in fish sampled in this study were from ponds within Kikwe and Maweni in Arusha region and Tangeni in Morogoro region.

Table 5: Prevalence of endoparasites and ectoparasites in farmed fish in study sites within Arusha and Morogoro

Bacteria species	Parasite occurrence, n (%)													χ^2
	Arusha (*n* for each site = 13)							Morogoro (*n* for each site= 13)						
	Kikwe	Nambala	Maweni	Manyata	Samalia	Total	χ^2	Kihonda	Langali	Mikese	Mkindo	Tangeni	Total	
Endoparasites														
Acanthocephala sp.	8 (61.5)	10 (76.9)	5 (38.4)	6 (46.1)	3 (23.0)	**32 (49.2)**	9.824*	7 (53.8)	4 (30.7)	7 (53.8)	5 (38.4)	10 (76.9)	**33 (50.7)**	15.758*
Diplostomum sp.	7 (53.8)	5 (38.4)	7 (53.8)	1 (7.6)	4 (30.7)	**24 (36.9)**	4.727*	5 (38.4)	6 (46.1)	3 (23.0)	4 (30.7)	7 (53.8)	**25 (38.4)**	8.254??
Contracecum sp.	9 (69.2)	5 (38.4)	10 (76.9)	4 (30.7)	4 (30.7)	**32 (49.2)**	12.123**	4 (30.7)	5 (38.4)	5 (38.4)	3 (23.0)	10 (76.9)	**27 (41.5)**	4.701[ns]
Ectoparasites														
Trichodina sp.	10 (76.9)	4 (30.7)	6 (46.1)	4 (30.7)	5 (38.4)	**29 (44.6)**	15.255*	7 (53.8)	5 (38.4)	4 (30.7)	3 (23.0)	8 (61.5)	**27 (41.5)**	8.098[ns]
Dactylogyrus sp.	8 (61.5)	5 (38.4)	9 (69.2)	6 (46.1)	3 (23.0)	**31 (47.6)**	7.58[ns]	3 (23.0)	4 (30.7)	2 (15.3)	6 (46.1)	6 (46.1)	**21 (32.3)**	6.500[ns]
Gyrodactylus sp.	5 (38.4)	3 (23.0)	11 (84.6)	3 (23.0)	2 (15.3)	**24 (36.9)**	5.591[ns]	6 (46.1)	5 (38.4)	6 (46.1)	4 (30.7)	10 (76.9)	**31 (47.6)**	6.243[ns]
Leeches	0 (0)	2 (15.3)	0 (0)	0 (0)	1 (7.6)	**3 (4.6)**	3.259[ns]	0 (0)	0 (0)	0 (0)	4 (30.7)	1 (7.6)	**5 (7.6)**	6.429*

Legend: *n* is the number of fish samples

Numbers in parentheses are the percentages of detected bacteria from two different fish species

Statistically significant at *$P < 0.05$, **$P < 0.01$; ns = not significant

4.1.3 Occurrence of Fish Bacteria Pathogens Isolated from Farmed Nile Tilapia (*Oreochromis niloticus*) and African catfish (*Clarias gariepinus*)

(i) The Occurrence of Bacteria in Nile Tilapia and African Catfish

All the bacteria genera isolated in this study occurred in both Nile tilapia and African catfish. Overall, the most common bacteria found in Arusha farms were *Aeromonas sobria* (35.5%), *Pseudomonas aeruginosa (26.1%), Edwardsiella tarda* (24.6%), and *Enterococcus faecalis* (24.6%) while those found in Morogoro were *Aeromonas sobria* (49.2%), *Comamonas testosteroni* (21.5%), and *Vibrio cholera* (20%) (Table 6). On the other hand, *Staphylococcus aureus*, a human pathogen was not detected in Arusha fish samples, and *Chryseobacterium indoligenes* were very scanty in Arusha, occurring at only 4.6%, in both Nile tilapia and catfish. However, the least prevalent bacteria pathogens in Morogoro were *Streptococcus* spp and *Staphylococcus aureus,* both occurring at 3.0%, and were found in both fish types (Table 6). Overall, there was a significant difference ($p < 0.05$) in the occurrence of *Chryseobacterium indoligenes* between the two types of fish in Arusha and *Flavobacterium spp* in Morogoro.

Table 6: The occurrence of bacteria species on Nile tilapia and African catfish

Bacteria species	Arusha			χ^2	Morogoro			χ^2
	Tilapia (n = 54)	Catfish (n = 11)	Total (n = 65)		Tilapia (n = 46)	Catfish (n = 19)	Total (n = 65)	
Aeromonas sobria	22 (40.7)	1 (9.0)	23 (35.3)	0.201^{ns}	21(45.6)	11(57.8)	32 (49.2)	0.831^{ns}
Edwardsiella tarda	15 (27.7)	1 (9.0)	16 (24.6)	0.000^{ns}	7(15.2)	4(21.0)	11(16.9)	0.730^{ns}
Flavobacterium spp.	11 (20.3)	0 (0)	11 (16.9)	0.868^{ns}	1 (2.1)	4(21.0)	5 (7.6)	3.611^{*}
Streptococcus spp.	9 (16.6)	1 (9.0)	10 (6.1)	0.303^{ns}	2 (4.3)	0 (0)	2 (3.0)	1.376^{ns}
Plesiomonas shigelloides	9(16.6)	0(0)	9 (13.8)	0.685^{ns}	5 (10.8)	7(36.8)	12(18.4)	2.061^{ns}
Chryseobacterium indoligenes	2 (3.7)	1 (9.0)	3(4.6)	4.023^{*}	2 (4.3)	4(21.0)	6 (9.2)	1.959^{ns}
Pseudomonas fluorescens	6 (11.1)	2 (18.1)	8 (12.3)	0.636^{ns}	5 (10.8)	1(5.2)	6 (9.2)	1.500^{ns}
Pseudomonas aeruginosa	15 (27.7)	2 (18.1)	17 (26.1)	1.510^{ns}	4 (8.6)	5 (26.3)	9 (13.8)	1.053^{ns}
Vibrio cholerae	8 (14.8)	0 (0)	8 (12.3)	0.598^{ns}	9 (19.5)	4 (21.0)	13 (20)	0.577^{ns}
Proteus spp.	6 (11.1)	0 (0)	6 (9.2)	0.433^{ns}	3 (6.5)	1 (5.2)	4 (6.1)	0.400^{ns}
Klebsiella spp.	13 (24.0)	2 (18.1)	15(23.0)	1.740^{ns}	5 (10.8)	3 (15.7)	8 (12.3)	0.024^{ns}
Serratia marcescens	13(24.0)	1 (9.0)	14(21.5)	0.030^{ns}	7 (15.2)	5 (26.3)	12 (18.4)	0.017^{ns}
Burkholderia cepacia	4 (7.4)	0 (0)	4 (6.1)	0.279^{ns}	5 (10.8)	4 (21.0)	9 (13.8)	0.086^{ns}
Comamonas testosteroni	13(24.0)	1(9.0)	14 (21.5)	0.030^{ns}	10 (21.7)	4 (21.0)	14 (21.5)	0.971^{ns}
Escherichia coli	8 (14.8)	0 (0)	8 (9.2)	0.598^{ns}	4 (8.6)	5 (26.3)	9 (13.8)	1.053^{ns}
Shigella dynteriae	5 (9.25)	0 (0)	5 (7.6)	0.355^{ns}	8 (17.3)	4 (21.0)	12 (18.4)	0.273^{ns}
Staphylococcus aureus	0 (0)	0 (0)	0 (0.0)	-	0 (0.0	2 (10.5)	2 (3.0)	3.095^{ns}
Enterococcus faecalis	15(27.7)	1(9.0)	16 (24.6)	0.000^{ns}	7 (15.2)	1 (5.2)	8 (12.3)	2.875^{ns}
Salmonella typhi	5 (9.25)	1 (9.0)	6 (9.23)	1.265^{ns}	2 (4.3)	3 (15.7)	5(7.6)	0.903^{ns}

Legend: n is the number of fish samples

Numbers in parentheses are the percentages of detected bacteria from two different fish species

Statistically significant at $*P < 0.05$; ns = not significant; ⁻ no statistics were computed because of the absence of bacteria species in analysed samples.

(ii)   The occurrence of Bacteria in Fish Samples from Each Site

Of the 18 genera found, at least 19 different species of bacteria were isolated and identified in this study, and their corresponding prevalence at farm level are reported in Table 7. *Aeromonas sobria* was the most occurring bacteria species found in the Arusha region (35.3%) especially in Kikwe location, followed by *Pseudomonas aeruginosa* (26.1%), *Enterococcus faecalis* (24.6%), *Edwardsiella tarda* (24.6%) *Serratia marcescens (21.5%)*, and *Comamonas testosteroni* (21.5%) (Table 2). In Morogoro region, the prevalence of *Aeromonas sobria* was still the highest (49.2%), especially in Tangeni location, followed by *Comamonas testosterone* (21.5%), *Vibrio cholera* (20.0%), *Plesiomonas shigelloides* (18.4%), *Serratia marcescens* (18.4%) and *Shigella dynteriae* (18.4 %).

A total of 7 human bacteria pathogens were identified in the fish samples, namely *Vibrio cholera, Escherichia coli, Pseudomonas aeruginosa, Shigella dynteriae, Staphylococcus aureus, Enterococcus faecalis,* and *Salmonella typhi*. In Arusha, the most prevalent human pathogen was *Pseudomonas aeruginosa* (26.1%), mostly found in Kikwe and Maweni locations; as well as *Enterococcus faecalis* (24.6%), mostly found in Maweni, Nambala, and Samalia. Even though *Staphylococcus aureus*, was not found in fish from all the five locations studied in Arusha (Table 7), it's potential to contaminate fish at any one point in time cannot be entirely ruled out since it was isolated from pond water samples in Arusha region (Table 9). However, in Morogoro, *Staphylococcus aureus* was present in two ponds from within only one location (Kihonda) (Table 2). The most occurring human bacterial pathogen in fish sampled in Morogoro was *Vibrio cholerae* (20.0%), mostly found in Mikese and Tangeni sites; followed by *Shigella dynteriae* (18.4%), mostly found in Mkindo (Table 7). There were significant differences (p<0.05) in the occurrence of different bacteria species between sampling sites. The principal component analysis biplot shows that PC1 accounts for 8.92% variability in bacteria occurrence in fish samples while PC2 accounts for 8.51%. The PCA result indicates the existence of a correlation between the observed bacteria pathogens in fish samples with sampling sites, production types, and farmed species (Fig. 9).

Table 7: Bacteria pathogen in fish samples

Bacteria species	Arusha (n for each site = 13)						χ^2	Morogoro (n for each site= 13)						χ^2
	Kikwe	Nambala	Maweni	Manyata	Samalia	Total		Kihonda	Langali	Mikese	Mkindo	Tangeni	Total	
Aeromonas sobria	8 (61.5)	3 (23.0)	6 (46.1)	5 (38.4)	1(7.6)	23 (35.3)	9.824^*	7 (53.8)	1 (7.6)	9 (69.2)	5 (38.4)	10 (76.9)	32 (49.2)	15.758^*
Edwardsiella tarda	4 (30.7)	2 (15.3)	8 (61.5)	2 (15.3)	0 (0)	16 (24.6)	15.255^{**}	0 (0)	2 (15.3)	1 (7.6)	3 (23.0)	5 (38.4)	11 (16.9)	8.098^{ns}
Flavobacterium spp.	0 (0)	0 (0)	4 (30.7)	6 (46.1)	1 (7.6)	11 (16.9)	15.758^{**}	1 (7.6)	0 (0)	0 (0)	1 (7.6)	3 (23.0)	5 (7.6)	6.500^{ns}
Streptococcus spp.	3 (23.0)	0 (0)	2 (15.3)	4 (30.7)	1 (7.6)	10 (15.3)	4.727^{ns}	0 (0)	0 (0)	0 (0)	0 (0)	2 (15.3)	2 (3.0)	8.254^{ns}
Plesiomonas shigelloides	3 (23.0)	5 (38.4)	1 (7.6)	0 (0)	0 (0)	9 (13.8)	12.123^{**}	4 (30.7)	2 (15.3)	1 (7.6)	4 (30.7)	1 (7.6)	12 (18.4)	4.701^{ns}
Chryseobacterium indoligenes	0 (0)	0 (0)	0 (0)	2 (15.3)	1 (7.6)	3 (4.6)	5.591^{ns}	3 (23.0)	1 (7.6)	2 (15.3)	0 (0)	0 (0)	6 (9.2)	6.243^{ns}
Pseudomonas fluorescens	1 (7.6)	4 (30.7)	2(15.3)	0 (0)	1 (7.6)	8 (12.3)	6.557^{ns}	0 (0)	0 (0)	0 (0)	1 (7.6)	5 (38.4)	6 (9.2)	17.260^*
Pseudomonas aeruginosa	8 (61.5)	3 (23.0)	5 (38.4)	1 (7.6)	0 (0)	17 (26.1)	16.409^*	2 (15.3)	2 (15.3)	4 (30.7)	1 (7.6)	0 (0)	9 (13.8)	5.675^{ns}
Vibrio cholerae	2 (15.3)	0 (0)	5 (38.4)	0 (0)	1 (7.6)	8 (12.3)	12.259^*	1 (7.6)	1 (7.6)	5 (38.4)	2 (15.3)	4 (30.7)	13 (20.0)	6.346^{ns}
Proteus spp.	1 (7.6)	1 (7.6)	2 (15.3)	2 (15.3)	0 (0)	6 (9.2)	2.571^{ns}	3 (23.0)	0 (0)	0 (0)	0 (0)	1 (7.6)	4 (6.1)	9.057^{ns}
Klebsiella spp.	2 (15.3)	0 (0.00)	3 (23.0)	5 (38.4)	5 (38.4)	15 (23.0)	7.800^{ns}	2 (15.3)	0 (0.00)	4 (30.7)	1 (7.6)	1 (7.6)	8 (12.3)	6.557^{ns}
Serratia marcescens	3 (23.0)	1 (7.6)	7 (53.8)	2 (15.3)	1 (7.6)	14 (21.5)	11.289^*	3 (23.0)	1 (7.6)	6 (46.1)	1 (7.6)	1 (7.6)	12 (18.4)	9.811^*
Burkholderia cepacia	0 (0)	0 (0)	3 (23.0)	1 (7.6)	0 (0)	4 (6.1)	9.057^{ns}	4 (30.7)	1 (7.6)	4 (30.7)	0 (0)	0 (0)	9 (13.8)	10.833^*
Comamonas testosteroni	4 (30.7)	6 (46.1)	1 (7.6)	3 (23.0)	0 (0)	14 (21.5)	10.378^*	0 (0)	0 (0)	5 (38.4)	4 (30.7)	5 (38.4)	14 (21.5)	12.199^*
Escherichia coli	2 (15.3)	5 (38.4)	0 (0)	1 (7.6)	0 (0)	8 (12.3)	12.259^*	2 (15.3)	1 (7.6)	1 (7.6)	0 (0)	5 (38.4)	9 (13.8)	9.544^*
Shigella dynteriae	0 (0)	0 (0)	0 (0)	4 (30.7)	1 (7.6)	5 (7.6)	13.000^*	3 (23.0)	2 (15.3)	2 (15.3)	4 (30.7)	1 (7.6)	12 (18.4)	2.657^{ns}
Staphylococcus aureus	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0.00)	-	2 (15.3)	0 (0)	0 (0)	0 (0)	0 (0)	2 (3.0)	8.254^{ns}
Enterococcus faecalis	1 (7.6)	5 (38.4)	6 (46.1)	1 (7.6)	3 (23.0)	16 (24.6)	8.622^{ns}	0 (0)	1 (7.6)	1 (7.6)	4 (30.7)	2 (15.3)	8 (12.3)	6.557^{ns}
Salmonella typhi	2 (15.3)	0 (0)	0 (0)	0 (0)	4 (30.7)	6 (9.2)	11.751^*	4 (30.7)	0 (0)	0 (0)	1 (7.6)	0 (0)	5 (7.6)	13^*

Legend: n is the number of fish samples

Numbers in parentheses are the percentages of detected bacteria from individual sample

Statistically significant at $^*P < 0.05$, $^{**}P < 0.01$; ns = not significant; no statistics were computed because of the absence of bacteria species in analysed samples
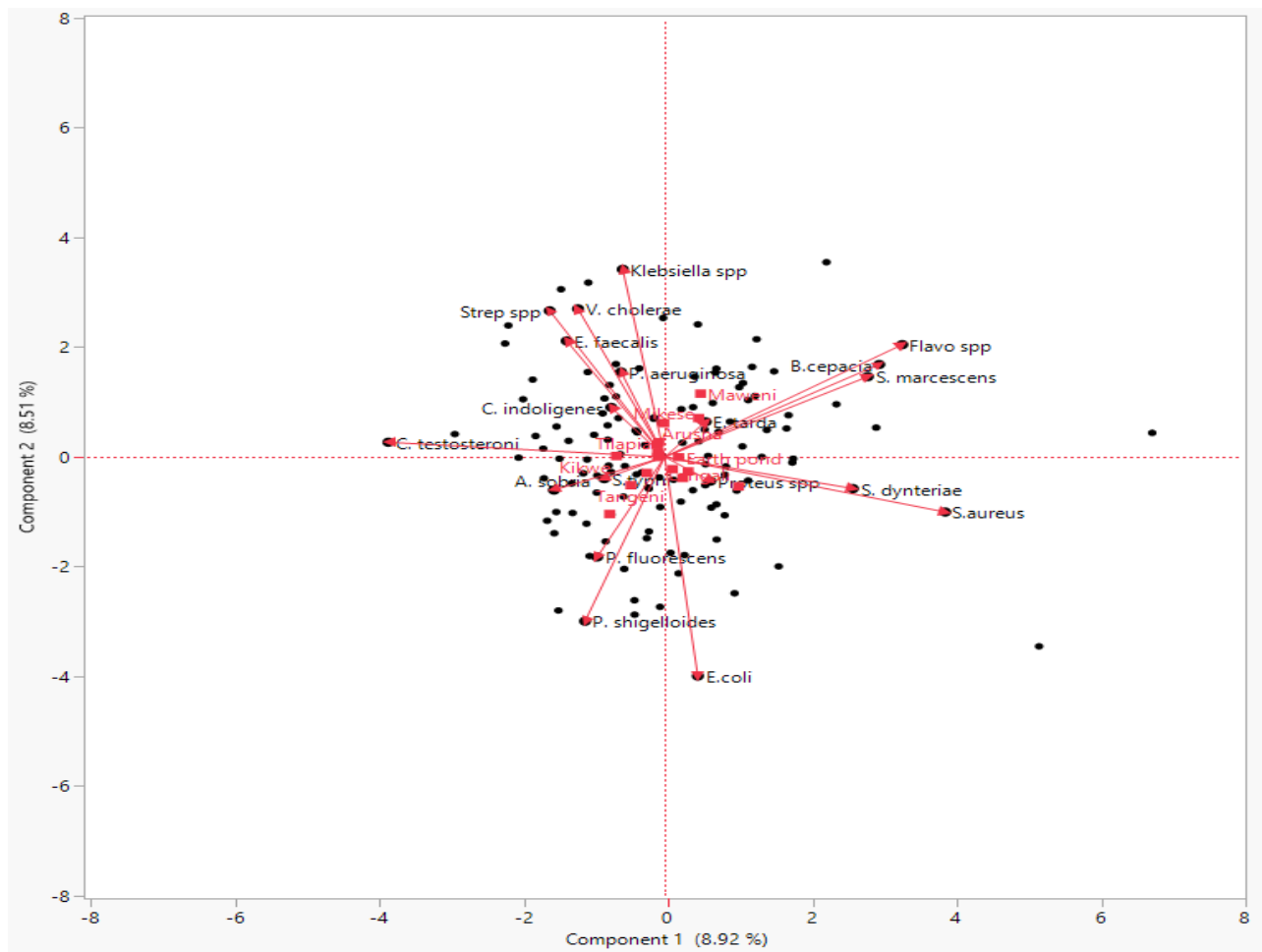
Figure 9: Principal component analysis (PCA) showing the relationship between bacteria pathogen occurrence and independent variables (sites, aquaculture types, and farmed species) that emanated insignificant effects (p<0.05)

(iii)   The Occurrence of Fish Bacteria in Different Fish Production

Concrete ponds were the most used fish production system in all sites studied. Both earthen and concrete ponds from multiple sites in Arusha and Morogoro regions were found to have at least s16 or more of the 19 bacteria species identified in this study (Table 8). In Arusha, the only bacteria not found in the earthen ponds were *Staphylococcus aureus* while the concrete ponds had no *Burkholderia cepacia.* On the other hand, in Morogoro, only 3 bacteria pathogens, namely, *Streptococcus* spp, *Proteus* spp and *Klebsiella* spp were not found in the earthen ponds while the concrete ponds had the least occurrence of *Staphylococcus aureus* (2%) (Table 8). Between the two types of production systems, *Aeromonas sobria* was the most common pathogen in both regions. A significant difference (p<0.05) was observed in the occurrence of different bacteria between two types of production systems. Overall, based on the total number of sampled ponds per production system in both regions, more bacterial pathogens occurred in the earthen ponds compared to the concrete ponds.

Table 8: Occurrence of fish bacteria in the different fish production systems (earthen and concrete ponds)

Bacteria species	Bacteria occurrence, n (%)							
	Arusha				Morogoro			
	Earthen pond (n = 26)	Concrete pond (n = 39)	Total (n = 65)	χ^2	Earthen pond (n = 17)	Concrete pond (n = 48)	Total (n = 65)	χ^2
Aeromonas sobria	16(61.5)	7(17.9)	23 (35.3)	12.964^{***}	8 (47.0)	24 (50.0)	32 (49.2)	0.43^{ns}
Edwardsiella tarda	9 (34.6)	7 (17.9)	16(24.6)	2.335^{ns}	4 (23.5)	7 (14.5)	11 (16.9)	0.715^{ns}
Flavobacterium spp.	5 (19.2)	6 (15.3)	11(16.9)	0.165^{ns}	1 (5.8)	4 (8.3)	5 (7.6)	0.106^{ns}
Streptococcus spp.	5 (19.2)	5 (12.8)	10 (15.3)	0.492^{ns}	0 (0.00)	2(4.1)	2 (3.0)	0.731^{ns}
Plesiomonas shigelloides	3 (11.5)	6 (15.3)	9 (13.8)	0.193^{ns}	3 (17.6)	9 (18.7)	12 (18.4)	0.010^{ns}
Chryseobacterium indoligenes	0 (0.0)	3 (7.6)	3(4.6)	2.097^{ns}	1 (5.8)	5 (10.4)	6 (9.2)	0.308^{ns}
Pseudomonas fluorescens	3 (11.5)	5 (10.2)	8 (12.3)	0.024^{ns}	1 (5.8)	5 (10.4)	6 (9.2)	0.308^{ns}
Pseudomonas aeruginosa	9(34.6)	8 (20.5)	17 (26.1)	1.606^{ns}	4 (23.5)	5 (10.4)	9 (13.8)	1.810^{ns}
Vibrio cholerae	2(7.6)	6 (15.3)	8 (12.3)	0.855^{ns}	4 (23.5)	9 (18.7)	13 (20.0)	0.179^{ns}
Proteus spp.	3 (11.5)	2 (5.1)	6 (9.2)	0.041^{ns}	0 (0.0)	4 (8.3)	4 (6.1)	1.510^{ns}
Klebsiella spp.	7 (26.9)	5 (12.8)	15 (23.0)	2.438^{ns}	0 (0.0)	8 (16.6)	8 (12.3)	3.231^{ns}
Serratia marcescens	10 (38.4)	6 (15.3)	14(21.5)	1.737^{ns}	6 (35.2)	6 (12.5)	12 (18.4)	4.333^{*}
Burkholderia cepacia	2 (7.6)	0 (0.00)	4 (6.1)	1.585^{ns}	4 (23.5)	5 (10.4)	9(13.8)	1.810^{ns}
Comamonas testosteroni	4 (15.3)	7 (17.9)	14 (21.5)	0.524^{ns}	3 (17.6)	11 (22.9)	14(21.5)	0.206^{ns}
Escherichia coli	3 (11.5)	3 (7.6)	8 (12.3)	1.278^{ns}	4 (23.5)	5 (10.4)	9(13.8)	1.810^{ns}
Shigella dynteriae	2 (7.6)	3 (7.6)	5 (7.6)	0.771^{ns}	6 (35.2)	6 (12.5)	12 (18.4)	4.333^{*}
Staphylococcus aureus	0 (0.00)	1 (2.5)	0 (0.0)	0.833^{ns}	1 (5.8)	1 (2.0)	2 (3.0)	0.608^{ns}
Enterococcus faecalis	5 (19.2)	4 (10.2)	16 (24.6)	1.274^{ns}	2 (11.7)	6(12.5)	8 (12.3)	0.006^{ns}
Salmonella typhi	2 (7.6)	2 (5.1)	6 (9.2)	0.577^{ns}	1 (5.8)	4 (8.3)	5(7.6)	0.106

Legend: n is the number of fish samples

Numbers in parentheses are the percentages of detected bacteria in the different production system

Statistically significant at *P < 0.05, **P < 0.01, ***P < 0.001; ns = not significant; ‾ no statistics were computed because of the absence of bacteria species in analysed samples

(iv) Bacteria Pathogen Isolated from Pond Water Samples

Water samples collected from each of the 10 sites (n per site = 13) in this study had at least nine or more of all the 19 different species of bacteria (Table 9). In ranking order, the three most prevalent bacteria pathogens in Arusha pond water were *Pseudomonas aeruginosa* (35.3%), *Edwardsiella tarda* (32.3%), and *Enterococcus faecalis* (26.1%), all of which were mostly found in Nambala and Maweni. None of the pond water sampled in Arusha had *Burkholderia cepacia* (Table 9).

On the other hand, the three most occurring bacteria in Morogoro pond water were *Edwardsiella tarda* (35.3%), mostly found in Kihonda, Langali, Mkindo and Tangeni; followed by *Aeromonas sobria* (24.6%) and *Klebsiella* spp (23.0%), both of which were mostly found in Kihonda and Mikese. Interestingly, *Enterococcus faecalis* (1.5%) occurrence was scanty in Morogoro pond water. The other main human bacteria pathogens found in pond water sampled in Morogoro were *Pseudomonas aeruginosa* (20.0%), especially in Langali and Mikese sites; and *Shigella dynteriae* (13.8%) in Tangeni site (Table 9). Overall, the least number of fish bacteria found in pond water sampled in Arusha and Morogoro occurred in Samalia and Mkindo.

Table 9: Bacteria pathogens in fish pond samples

Bacteria species	Arusha (n for each location = 13)						χ^2	Morogoro (n for each location = 13)						χ^2
	Kikwe	Nambala	Maweni	Manyata	Samalia	Total		Kihonda	Langali	Mikese	Mkindo	Tangeni	Total	
Aeromonas sobria	3 (23.0)	1 (7.6)	7 (53.8)	3 (23.0)	1 (7.6)	15 (23.0)	10.400*	4 (30.7)	2 (15.3)	5 (38.4)	2 (15.3)	3 (23.0)	16 (24.6)	2.819ns
Edwardsiella tarda	3 (23.0)	8 (61.5)	6 (46.1)	4 (30.7)	0 (0.00)	21 (32.3)	12.944*	5 (38.4)	4 (30.7)	3 (23.0)	6 (46.1)	5 (38.4)	23 (35.3)	1.749ns
Flavobacterium spp.	0 (0.00)	0 (0.00)	3 (23.0)	4 (30.7)	1 (7.6)	8 (12.3)	9.408*	2 (15.3)	1 (7.6)	1 (7.6)	0 (0.00)	1 (7.6)	5 (7.6)	2.167ns
Streptococcus spp.	1 (7.6)	0 (0.00)	1 (7.6)	0 (0.00)	0 (0.00)	2 (3.0)	3.095ns	1 (7.6)	0 (0.00)	0 (0.00)	4 (30.7)	2 (15.3)	7 (10.7)	8.966ns
Plesiomonas shigelloides	2 (15.3)	3 (23.0)	0 (0.00)	0 (0.00)	0 (0.00)	5 (7.6)	9.915*	2 (15.3)	3 (23.0)	2 (15.3)	1 (7.6)	5 (38.4)	13 (20.0)	4.423ns
Chryseobacterium indoligenes	1 (7.6)	0 (0.00)	1 (7.6)	3 (23.0)	0 (0.00)	5 (7.6)	6.50ns	2 (15.3)	1 (7.6)	5 (38.4)	0 (0.00)	0 (0.00)	8 (12.3)	12.259*
Pseudomonas fluorescens	4 (30.7)	2 (15.3)	0 (0.00)	0 (0.00)	5 (38.4)	11 (16.9)	11.380*	3 (23.0)	0 (0.00)	0 (0.00)	0 (0.00)	3 (23.0)	6 (9.2)	9.915*
Pseudomonas aeruginosa	3 (23.0)	9 (69.2)	4 (30.7)	6 (46.1)	1 (7.6)	23 (35.3)	12.516*	3 (23.0)	6 (46.1)	4 (30.7)	0 (0.00)	0 (0.00)	13 (20.0)	13.077*
Vibrio cholerae	5 (38.4)	2 (15.3)	6 (46.1)	3 (23.0)	0 (0.00)	16 (24.6)	9.452ns	0 (0.00)	4 (30.7)	0 (0.00)	0 (0.00)	1 (7.6)	5 (7.6)	13.000*
Proteus spp.	0 (0.00)	0 (0.00)	4 (30.7)	2 (15.3)	0 (0.00)	6 (9.2)	11.751*	6 (46.1)	0 (0.00)	3 (23.0)	0 (0.00)	0 (0.00)	9 (13.8)	18.571*
Klebsiella spp.	3 (23.0)	0 (0.00)	0 (0.00)	5 (38.4)	6 (46.1)	14 (21.5)	14.020**	6 (46.1)	2 (15.3)	4 (30.7)	2 (15.3)	1 (7.6)	15 (23.0)	6.933ns
Serratia marcescens	3 (23.0)	1 (7.6)	7 (53.8)	3 (23.0)	0 (0.00)	14 (21.5)	13.109*	1 (7.6)	3 (23.0)	6 (46.1)	1 (7.6)	2 (15.3)	13 (20.0)	11.289*
Burkholderia cepacia	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	-	1 (7.6)	1 (7.6)	1 (7.6)	0 (0.00)	1 (7.6)	4 (6.1)	1.066ns
Comamonas testosteroni	4 (30.7)	0 (0.00)	2 (15.3)	1 (7.6)	0 (0.00)	7 (10.7)	8.966ns	1 (7.6)	0 (0.00)	2 (15.3)	4 (30.7)	5 (38.4)	12 (18.4)	8.789ns
Escherichia coli	0 (0.00)	1 (7.6)	4 (30.7)	0 (0.00)	1 (7.6)	6 (9.2)	13.768**	2 (15.3)	2 (15.3)	0 (0.00)	1 (7.6)	1 (7.6)	6 (9.2)	2.571ns
Shigella dynteriae	0 (0.00)	0 (0.00)	1 (7.6)	1 (7.6)	1 (7.6)	3 (4.6)	2.097ns	2 (15.3)	0 (0.00)	2 (15.3)	1 (7.6)	4 (30.7)	9 (13.8)	5.675ns
Staphylococcus aureus	1 (7.6)	0 (0.00)	1 (7.6)	1 (7.6)	0 (0.00)	3 (4.6)	2.097ns	2 (15.3)	4 (30.7)	0 (0.00)	0 (0.00)	0 (0.00)	6 (9.2)	11.751*
Enterococcus faecalis	2 (15.3)	4 (30.7)	6 (46.1)	1 (7.6)	4 (30.7)	17 (26.1)	6.054ns	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	1 (7.6)	1 (1.5)	4.063ns
Salmonella typhi	1 (7.6)	2 (15.3)	1 (7.6)	0 (0.00)	3 (23.0)	7 (10.7)	4.163ns	1 (7.6)	0 (0.00)	0 (0.00)	2 (15.3)	0 (0.00)	3 (4.6)	5.591ns

Legend: n is the number of water samples.

Numbers in parentheses are the percentages of detected bacteria in fish ponds samples.

Statistically significant at *$P < 0.05$, **$P < 0.01$; ns = not significant; ⁻ no statistics were computed because of the absence of bacteria species in analysed samples

(v)   Bacteria Pathogens in Fish Feed Samples

Of the 19 bacteria identified in this study, only five species, namely, *Aeromonas sobria, Klebsiella* spp, *Serratia marcescens, Burkholderia cepacia, Comamonas testosterone* and *Staphylococcus aureus* did not occur in any of the fish feed samples collected from all the 130 sites across Arusha and Morogoro (Table 10). *Escherichia coli* (9.2%) was the most occurring bacteria species in fish feeds collected in Arusha, especially in Kikwe and Samalia sites. In Morogoro, on the other hand, the most occurring bacteria species in fish feeds were *Salmonella typhi* (15.3%) especially in Kihonda, Langali and Tangeni as well as *Vibrio cholera* (12.3%) especially in sites within Mikese (Table 10).

The occurrence of pathogens based on the type of feeding methods used by the farmers in both regions was also investigated. Fish farmers in both Arusha and Morogoro fed their fish using three main feed types: Commercial, on-farm made, and food scraps (Table 11). None of the 19 bacteria species identified in this study occurred in any of the commercial feeds sampled from the 130 study sites across Arusha and Morogoro (Table 11). Overall, only 6 bacteria species, namely, *Aeromonas sobria, Klebsiella* spp, *Serratia marcescens, Burkholderia cepacia, Comamonas testosterone,* and *Shigella dynteriae* did not occur under any of the feeding methods utilized by farmers in all the study locations.

In Arusha, the most occurring human bacterial pathogen in the feeds was *Vibrio cholera* (9.2%), especially in the on-farm made feed samples, followed by *Escherichia coli* (5.7%), mostly found in the food scrap samples. In Morogoro, the most prevalent pathogen in the feeds was *Vibrio cholerae* (9.2%), *Escherichia coli* (9.2%) and *Salmonella typhi* (9.2%), followed by *Plesiomonas shigelloides* (7.6%), all of which were mostly present in the food scraps (Table 10).

The results from the Principal Component Analysis indicated a correlation between bacteria occurrence and feeding types (Fig. 10). A strong correlation was observed between on-farm made feed type (independent variable) with the occurrence of several bacteria fish pathogens. Correlation between the occurrence of the fish pathogen in feed samples and the sites where the samples were collected was also observed.

Table 10: Bacteria pathogens in fish feed samples

| Bacteria | Bacteria Occurrence, n (%) | | | | | | | | | | | | | |
| | Arusha (n for each location = 13) | | | | | | χ^2 | Morogoro (n for each location = 13) | | | | | | χ^2 |
	Kikwe	Nambala	Maweni	Manyata	Samalia	Total		Kihonda	Langali	Mikese	Mkindo	Tangeni	Total	
Aeromonas sobria	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	**0 (0)**	-	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	**0 (0)**	-
Edwardsiella tarda	0 (0)	0 (0)	0 (0)	1 (7.6)	0 (0)	**1 (1.5)**	4.063^{ns}	0 (0)	2 (15.3)	1 (7.6)	2 (15.3)	1 (7.6)	**6 (9.2)**	2.571^{ns}
Flavobacterium spp.	0 (0)	0 (0)	1 (7.6)	0 (0)	0 (0)	**1 (1.5)**	4.063^{ns}	0 (0)	0 (0)	0 (0)	1 (7.6)	0 (0	**1 (1.5)**	4.063^{ns}
Streptococcus spp.	0 (0)	0 (0)	0 (0)	2 (15.3)	1(7.6)	**3 (4.6)**	5.591^{ns}	0 (0)	0 (0)	0 (0)	0 (0)	1 (7.6)	**1 (1.5)**	4.063^{ns}
Plesiomonas shigelloides	1 (7.6)	0 (0)	1 (7.6)	0 (0)	0 (0)	**2 (3.0)**	3.095^{ns}	0 (0)	2 (15.3)	1 (7.6)	2 (15.3)	1 (7.6)	**6 (9.2)**	2.571^{ns}
Chryseobacterium indoligenes	0 (0)	0 (0)	0 (0)	1 (7.6)	1 (7.6)	**2 (3.0)**	3.095^{ns}	0 (0)	1 (7.6)	0 (0)	1(7.6)	0 (0)	**2 (3.0)**	3.095^{ns}
Pseudomonas fluorescens	1 (7.6)	0 (0)	0 (0)	0 (0)	0 (0)	**1 (1.5)**	4.063^{ns}	1 (7.6)	0 (0)	0 (0)	0 (0)	1 (7.6)	**2 (3.0)**	3.095^{ns}
Pseudomonas aeruginosa	2 (15.3)	1 (7.6)	1 (7.6)	0 (0)	0 (0)	**4 (6.1)**	3.730^{ns}	0 (0)	0 (0)	0 (0)	1 (7.6)	0 (0)	**1 (1.5)**	4.063^{ns}
Vibrio cholerae	2 (15.3)	1 (7.6)	1 (7.6)	0 (0)	0 (0)	**4 (6.1)**	3.730^{ns}	2 (15.3)	1 (7.6)	3 (23.0)	1 (7.6)	1 (7.6)	**8 (12.3)**	2.281^{ns}
Proteus spp.	1 (7.6)	1 (7.6)	1 (7.6)	0 (0)	0 (0)	**3 (4.6)**	2.097^{ns}	1 (7.6)	0 (0)	0 (0)	0 (0)	0 (0)	**1 (1.5)**	4.063^{ns}
Klebsiella spp.	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	**0 (0)**	-	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	**0 (0)**	-
Serratia marcescens	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	**0 (0)**	-	1 (7.6)	0 (0)	0 (0)	0 (0)	1(1.5)	**0 (0)**	4.063^{ns}
Burkholderia cepacia	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	**0 (0)**	-	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	**0 (0)**	-
Comamonas testosteroni	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	**0 (0)**	-	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	**0 (0)**	-
Escherichia coli	2 (15.3)	0 (0)	1 (7.6)	1 (7.6)	2 (15.3)	**6 (9.2)**	2.571^{ns}	2 (15.3)	2 (15.3)	1 (7.6)	1 (7.6)	0 (0)	**6 (9.2)**	2.571^{ns}
Shigella dynteriae	0 (0)	0 (0)	0 (0)	1 (7.6)	1 (7.6)	**2 (3.0)**	3.095^{ns}	0 (0)	0 (0)	0 (0)	0 (0)	1 (7.6)	**1(1.5)**	8.095^{ns}
Staphylococcus aureus	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	**0 (0)**	-	1 (7.6)	0 (0)	0 (0)	1 (7.6)	0 (0)	**2 (3.0)**	3.095^{ns}
Enterococcus faecalis	1 (7.6)	1 (7.6)	0 (0)	1 (7.6)	0 (0)	**3 (4.6)**	2.097^{ns}	0 (0)	0 (0)	0 (0)	0 (0)	3 (23.0)	**3 (4.6)**	12.581*
Salmonella typhi	0 (0)	0 (0)	0 (0)	0 (0)	1 (7.6)	**1 (1.5)**	4.063^{ns}	3 (23.0)	3 (23.0)	0 (0)	1 (7.6)	3 (15.3)	**10 (15.3)**	4.727^{ns}

Legend: n is the number of feed samples

Numbers in parentheses are the percentages of detected bacteria in fish ponds samples

Statistically significant at *$P < 0.05$; ns = not significant; $^-$ no statistics were computed because of the absence of bacteria species in analysed samples.

Table 11: Occurrence of bacteria pathogens in different feeding types

Bacteria species	Bacteria occurrence									
	Arusha				χ^2	Morogoro				χ^2
	Commercial (n = 11)	On-farm made (n = 45)	Food scraps (n = 9)	Total		Commercial (n = 8)	On-farm made (n = 46)	Food scraps (n = 11)	Total	
Aeromonas sobria	0 (0)	0 (0)	0 (0)	**0 (0)**	-	0 (0)	0 (0)	0 (0.00)	**0 (0.00)**	**-**
Edwardsiella tarda	0 (0)	1 (2.2)	0 (0)	**1(1.5)**	0.207ns	0 (0)	6 (13.0)	0 (0)	**6 (9.2)**	**1.653ns**
Flavobacterium spp.	0 (0)	1 (2.2)	0 (0)	**1 (1.5)**	0.207ns	0 (0)	1 (2.1)	0 (0)	**1 (1.5)**	**0.254ns**
Streptococcus spp.	0 (0)	2 (4.4)	1 (11.1)	**3 (4.6)**	21.252***	0 (0)	1 (2.1)	0(0)	**(1.5)**	**0.254ns**
Plesiomonas shigelloides	0 (0)	2 (4.4)	0 (0)	**2 (3.0)**	0.420ns	0 (0)	6 (13.0)	0 (0)	**6 (9.2)**	**1.653ns**
Chryseobacterium indoligenes	0 (0)	2 (4.4)	0 (0)	**2 (3.0)**	0.420ns	0 (0)	1 (2.1)	1 (9.0)	**2 (3.0)**	**2.307ns**
Pseudomonas fluorescens	0 (0)	1 (2.2)	0 (0)	**1 (1.5)**	0.207ns	0 (0)	1 (2.1)	1 (9.0)	**2 (3.0)**	**2.307ns**
Pseudomonas aeruginosa	0 (0)	4 (8.8)	0 (0)	**4 (6.1)**	0.868ns	0 (0)	1 (2.1)	0 (0)	**1 (1.5)**	**0.254ns**
Vibrio cholerae	0 (0)	3 (6.6)	1 (11.1)	**4 (6.1)**	0.355ns	0 (0)	7 (15.2)	1 (12.5)	**8 (12.3)**	**1.924ns**
Proteus spp.	0 (0)	0 (0)	3 (33.3)	**3 (4.6)**	0.641ns	0 (0)	1 (2.1)	0 (0)	**1 (1.5)**	**0.254ns**
Klebsiella spp.	0 (0)	0 (0)	0 (0)	**0 (0)**	-	0 (0)	0 (0)	0 (0)	**0 (0)**	**-**
Serratia marcescens	0 (0)	0 (0)	0 (0)	**0 (0)**	-	0 (0)	1 (2.1)	0 (0)	**1 (1.5)**	**0.254ns**
Burkholderia cepacia	0 (0)	0 (0)	0 (0)	**0 (0)**	-	0 (0)	0 (0)	0 (0)	**0 (0)**	**-**
Comamonas testosteroni	0 (0)	0 (0)	0 (0)	**0(0)**	-	0 (0)	0 (0)	0 (0)	**0 (0)**	**-**
Escherichia coli	0 (0)	6 (13.3)	0 (0)	**6 (9.2)**	1.347ns	0 (0)	4 (8.6)	2 (18.1)	**6 (9.2)**	**0.454ns**
Shigella dynteriae	0 (0)	2 (4.4)	0 (0)	**2 (3.0)**	0.420ns	0 (0)	1 (2.1)	0 (0)	**1 (1.5)**	**0.516ns**
Staphylococcus aureus	0 (0)	0 (0)	0 (0)	**0 (0)**	-	0 (0)	0 (0)	0 (0)	**0 (0)**	**-**
Enterococcus faecalis	0 (0)	3 (6.6)	0 (0)	**3 (4.6)**	0.641ns	0 (0)	3 (6.5)	0 (0)	**3 (4.6)**	**0.786ns**
Salmonella typhi	0 (0)	1 (2.2)	0 (0)	**1 (1.5)**	0.207ns	0 (0)	9 (19.5)	1 (12.5)	**10 (15.3)**	**2.068ns**

Legend: n is the number of feed samples.

Numbers in parentheses are the percentages of detected bacteria from different feeding types

Statistically significant at *P < 0.05, **P < 0.01, ***P < 0.001; ns = not significant; - no statistics were computed because of the absence of bacteria species in analysed samples.
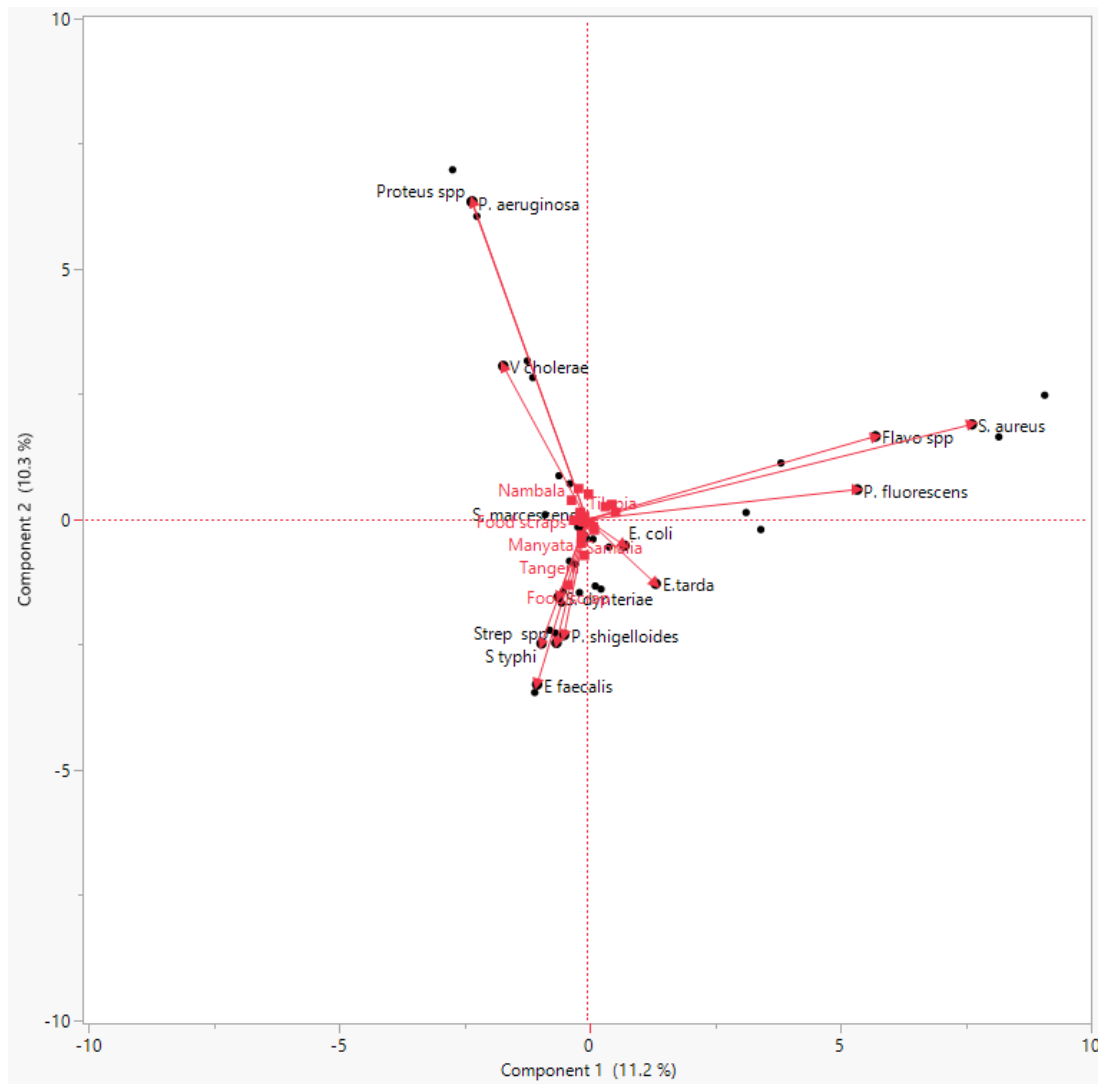
Figure 10: **Principal component analysis (PCA). Showing the relationship between bacteria pathogen occurrence and independent variables (Feed types and sites) that emanated insignificant effects (p<0.05)**

4.1.4 Bioaccumulation and Distribution Pattern of Heavy Metals in Aquaculture Systems

(i)   Heavy Metals in Sediments

Chromium (Cr) was the most abundant heavy metal found in the sediment samples and was especially higher in Morogoro region (Table 12). In Morogoro, the heavy metals concentrations (dry weight basis) in the sediments were measured as 6.10 – 9.28 mg/kg for Cr; 0.80 – 3.82 mg/kg for Pb; 2.54 – 3.60 mg/kg for Cd; 1.65 – 2.69 mg/kg for Hg; and 1.47 – 1.95 mg/kg for As. In Arusha, the heavy metals concentrations (dry weight basis) in the sediments were measured as 2.68–5.56 mg/kg for Cr; 0.93 – 3.54 mg/kg for Pb; 0.75 – 1.34 mg/kg for Hg; 0.64 – 1.32 mg/kg for Cd; and 0.68 – 1.29 mg/kg for As. Higher amounts of Cr were found in sediment samples from Mkindo (9.28 mg/kg) located in Morogoro while the least was found in Maweni (2.68 mg/kg) located in Arusha.

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Overall, sediment samples collected in Kihonda had the highest Cd (3.60 mg/kg), Hg (2.69 mg/kg), and Pb (3.82 mg/kg) concentrations (Table 12). Among Arusha sites studied, sediment samples from ponds in Samalia had the highest Pb and Cd concentrations of 3.54 and 1.32 mg/kg, respectively; while those from Nambala had the highest As, Hg and Cr concentrations of 1.29, 1.46 and 4.65 mg/kg dry weight basis, respectively.

Table 12: Heavy metals concentration (mg/kg, dry weight) in sediment samples from various locations in Arusha and Morogoro regions of Tanzania

Location	n	Arsenic	Lead	Mercury	Cadmium	Chromium
Morogoro						
Kihonda	13	1.66 ± 0.46^a	3.82 ± 2.79^a	2.69 ± 1.68^a	3.60 ± 1.02^a	8.30 ± 4.05^a
Langali	13	1.47 ± 0.67^a	1.43 ± 0.89^b	1.71 ± 0.92^a	2.54 ± 0.90^b	6.10 ± 3.37^a
Mikese	13	1.86 ± 0.74^a	2.83 ± 2.87^{ab}	1.84 ± 0.83^a	2.76 ± 0.79^{ab}	7.01 ± 4.04^a
Mkindo	13	1.95 ± 1.28^a	0.80 ± 0.53^b	1.95 ± 0.97^a	2.88 ± 1.05^{ab}	9.28 ± 3.76^a
Tangeni	13	1.70 ± 0.93^a	2.43 ± 1.08^{ab}	1.65 ± 0.68^a	2.77 ± 0.92^{ab}	7.64 ± 4.98^a
Mean, μ		**1.73 ± 0.82**	**2.26 ± 1.63**	**1.97 ± 1.01**	**2.91 ± 0.94**	**7.67 ± 4.04**
Arusha						
Kikwe	13	0.70 ± 0.50^b	0.93 ± 0.62^b	1.34 ± 0.80^a	0.64 ± 0.41^{ab}	5.40 ± 1.46^a
Nambala	13	1.29 ± 0.41^a	2.08 ± 1.45^{ab}	1.46 ± 0.92^a	1.10 ± 0.63^a	4.65 ± 0.95^a
Maweni	13	0.68 ± 0.37^b	0.94 ± 0.61^b	0.75 ± 0.50^a	1.19 ± 0.59^a	2.68 ± 0.82^b
Manyata	13	0.82 ± 0.28^{ab}	1.59 ± 1.02^b	1.05 ± 0.63^a	0.90 ± 0.52^{ab}	5.56 ± 1.98^a
Samalia	13	1.04 ± 0.26^{ab}	3.54 ± 2.19^a	0.98 ± 0.56^a	1.32 ± 0.34^a	4.36 ± 1.32^a
Mean, μ		**0.91 ± 0.36**	**1.82 ± 1.18**	**1.12 ± 0.68**	**1.03 ± 0.49**	**4.53 ± 1.31**
Probable effect concentration (PEC)[*]		**33.0**	**128.0**	**1.06**	**4.98**	**111.0**

Values are mean concentrations ± standard deviation expressed in mg/kg dry weight.

Values within a column with different letters are significantly different at *P*<0.05 level (Duncan's multiple range test).

n is the number of samples.

[*]Consensus-based sediment quality values for freshwater ecosystems called the probable effect concentration (PEC) guidelines proposed by MacDonald *et al.* (2000)

(ii)   Heavy Metal in Fish Feed

In Morogoro, all fish feed showed undetectable levels of As and Hg, while levels ranged from 4.01 to 8.39 mg/kg for Cr, from 0.40 to 2.94 mg/kg for Cd, and from 4.57 to 5.64 mg/kg for Pb (Table

13). On the other hand, all fish feed samples collected from Arusha sites had concentrations (dry weight basis) ranging from 2.49 to 4.75 mg/kg for As and 0.41 to 0.62 mg/kg for Hg. The comparison among sites revealed that Cr was the most abundant heavy metal in the fish feed samples tested in this study, particularly from Kihonda in Morogoro.

Table 13:   Heavy metals concentration (mg/kg, dry weight) in fish feed samples from various locations in Arusha and Morogoro regions of Tanzania

Location	n	Arsenic	Lead	Mercury	Cadmium	Chromium
Morogoro						
Kihonda	13	ND	4.57 ± 2.34^a	ND	2.82 ± 0.83^a	8.39 ± 5.30^a
Langali	13	ND	5.64 ± 2.59^a	ND	2.94 ± 0.57^a	5.77 ± 3.84^{ab}
Mikese	13	ND	5.03 ± 1.77^a	ND	2.77 ± 0.78^a	7.20 ± 3.41^{ab}
Mkindo	13	ND	4.79 ± 2.19^a	ND	2.74 ± 0.78^a	5.23 ± 1.74^{ab}
Tangeni	13	ND	4.59 ± 2.47^a	ND	0.40 ± 0.19^a	4.01 ± 1.42^b
Mean, μ		**ND**	**4.92 ± 2.27**	**ND**	**2.33 ± 0.36**	**6.13 ± 3.14**
Arusha						
Kikwe	13	3.66 ± 1.89^{ab}	0.86 ± 0.79^b	0.62 ± 0.31^b	3.37 ± 0.63^{ab}	5.11 ± 2.05^a
Nambala	13	3.12 ± 1.03^b	1.12 ± 0.46^{ab}	0.41 ± 0.08^b	1.78 ± 0.61^a	4.67 ± 1.67^a
Maweni	13	2.49 ± 1.15^b	1.66 ± 0.48^a	0.46 ± 0.17^a	1.31 ± 0.81^{ab}	3.02 ± 0.63^b
Manyata	13	4.75 ± 0.94^a	0.89 ± 0.65^{ab}	0.53 ± 0.14^{ab}	1.07 ± 0.36^b	5.31 ± 1.12^a
Samalia	13	3.76 ± 1.56^a	0.97 ± 0.45^b	0.46 ± 0.18^{ab}	1.22 ± 0.85^{ab}	2.87 ± 0.64^b
Mean, μ		**3.55 ± 1.31**	**1.10 ± 0.57**	**0.50 ± 0.18**	**1.75 ± 0.65**	**4.20 ± 1.22**

Values are mean concentrations ± standard deviation expressed in mg/kg dry weight.
Values within a column with different letters are significantly different at $P<0.05$ level (Duncan's multiple range test).
n is the number of samples.
ND – Not detected.

(iii)   Heavy Muscles Metals in Fish

In Morogoro region, the range of heavy metals (dry weight basis) concentrations in fish muscles were 0.43 – 0.80, 1.04 – 3.44, 0.47 – 0.84, 1.99 – 3.97 and 4.61 – 9.50 mg/kg for As, Pb, Hg, Cd, and Cr, respectively. The concentrations in Arusha region were 1.02 – 1.49, 0.58 – 0.94, 0.35 – 0.95, 1.38 – 3.55, and 2.53 – 5.57 mg/kg for As, Pb, Hg, Cd and Cr, respectively (Table 14). The highest Pb and Hg concentrations were found in Tangeni (3.44 mg/kg) and Manyata (0.95 mg/kg) in Arusha, respectively. On average, fish muscle samples from Mikese in Arusha had the highest content (dry weight basis) of Cd (3.97 mg/kg) and Cr (9.50 mg/kg).

Table 14: Heavy metals concentration (mg/kg dry weight) in fish muscle samples from various locations in Arusha and Morogoro region of Tanzania, compared with the recommended daily dietary allowances

Location	n	Arsenic	Lead	Mercury	Cadmium	Chromium
Morogoro						
Kihonda	13	0.65 ± 0.18^{abc}	1.68 ± 0.35^{b}	0.63 ± 0.12^{ab}	3.19 ± 1.10^{ab}	7.81 ± 2.25^{ab}
Langali	13	0.80 ± 0.24^{a}	1.04 ± 0.77^{b}	0.84 ± 0.12^{a}	1.99 ± 0.50^{c}	7.19 ± 3.11^{b}
Mikese	13	0.46 ± 0.12^{bc}	1.69 ± 0.65^{b}	0.62 ± 0.34^{ab}	3.97 ± 0.56^{a}	9.50 ± 0.98^{a}
Mkindo	13	0.67 ± 0.19^{ab}	1.46 ± 0.49^{b}	0.70 ± 0.18^{ab}	2.40 ± 0.81^{bc}	4.61 ± 0.98^{c}
Tangeni	13	0.43 ± 0.26^{c}	3.44 ± 0.93^{a}	0.47 ± 0.27^{b}	3.25 ± 0.43^{a}	8.50 ± 1.22^{ab}
Mean, µ		**0.60 ± 0.20**	**1.86 ± 0.64**	**0.65 ± 0.21**	**3.55 ± 0.68**	**7.52 ± 1.70**
Arusha						
Kikwe	13	0.76 ± 0.15^{c}	0.76 ± 0.16^{b}	0.35 ± 0.13^{c}	3.55 ± 0.28^{a}	4.31 ± 0.61^{bc}
Nambala	13	1.49 ± 0.30^{a}	0.77 ± 0.18^{b}	0.40 ± 0.08^{ab}	1.38 ± 0.44^{ab}	4.76 ± 0.89^{ab}
Maweni	13	1.22 ± 0.14^{ab}	0.79 ± 0.19^{b}	0.80 ± 0.04^{a}	2.75 ± 0.67^{ab}	5.57 ± 0.83^{a}
Manyata	13	1.02 ± 0.38^{bc}	0.58 ± 0.21^{ab}	0.95 ± 0.04^{a}	2.72 ± 0.81^{ab}	3.77 ± 0.73^{c}
Samalia	13	1.24 ± 0.40^{ab}	0.94 ± 0.15^{a}	0.50 ± 0.51^{ab}	2.38 ± 0.86^{ab}	2.53 ± 0.65^{d}
Mean, µ		**0.95 ± 0.27**	**0.80 ± 0.18**	**0.60 ± 0.16**	**2.56 ± 0.61**	**4.19 ± 0.74**
Recommended Daily Dietary Allowance (mg/day/ person)		0.13*	0.21*	0.03**	0.06*	0.20*

Values are mean concentrations ± standard deviation expressed in mg/kg dry weight.
Values within a column with different letters are significantly different at *P*<0.05 level (Duncan's multiple range test).
n is the number of samples.
*Recommended daily dietary allowances.
**Established recommended daily dietary allowance.

(iv)   Estimated Daily Intake (EDI)

The results of estimated daily intake are presented in Fig. 11. In Morogoro region, the EDI for As, Pb, Hg, Cd, and Cr was estimated to be 0.013, 0.041, 0.014, 0.065 and 0.0166 mg/person/day dry weight, respectively; which represents 4.43, 13.69, 4.79, 21.77 and 55.32% of the total heavy metals consumption, respectively. In Arusha region, the EDI for As, Pb, Hg, Cd, and Cr was estimated to be 0.025, 0.017, 0.013, 0.057 and 0.093 mg/person/day dry weight, respectively; which represents 12.38, 8.30, 6.48, 27.61 and 45.24% of heavy metals total consumption, respectively.
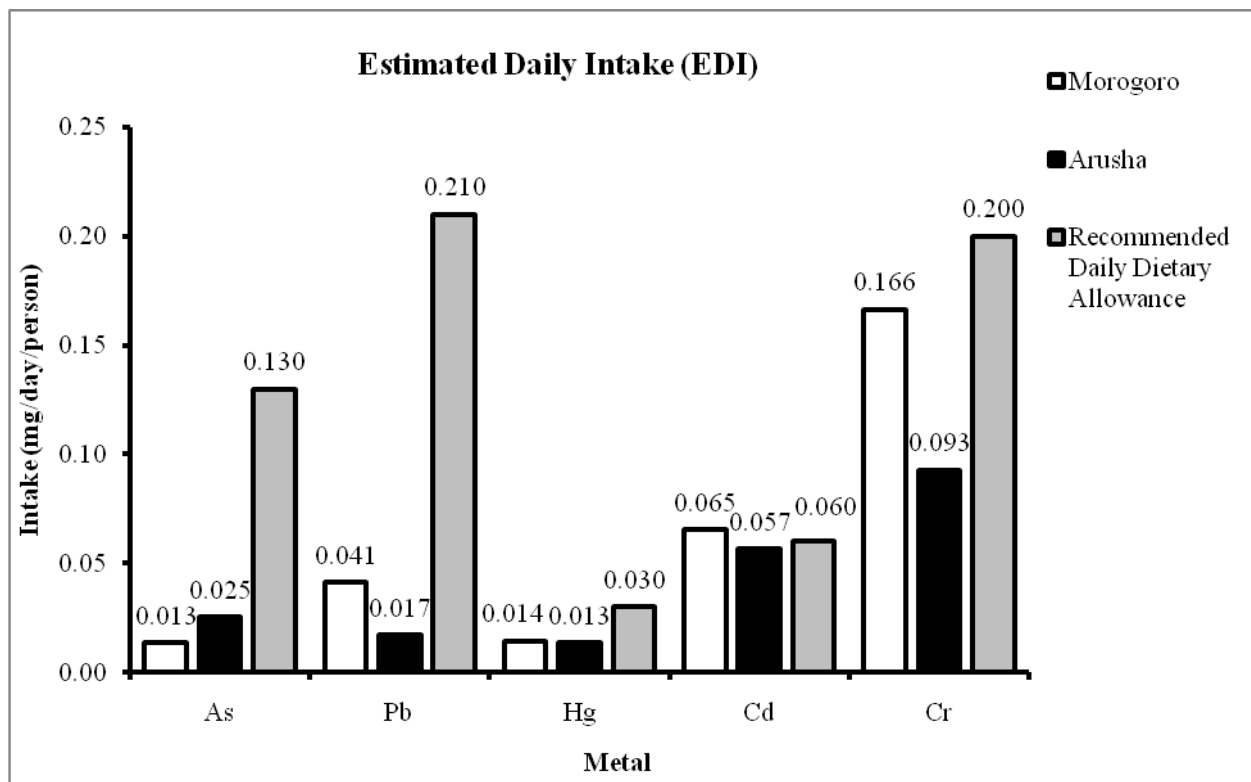
Figure 11: Comparison of the estimated daily intake (EDI) of heavy metals from fish muscle samples as well as the recommended daily dietary allowance

(v)   Target Hazard Quotient (THQ) and Hazard Index (HI)

Non-carcinogenic effects of the heavy metals were assessed using target hazard quotient (THQ) and Hazard Index (HI). In this study, we found that the THQ values of all heavy metals were <1 in both locations except for Cd in Morogoro region (Fig. 12a). The accepted value for THQ is less than 1 (WHO, 1976). The highest THQ was in Morogoro region (1.091 for Cd); while the lowest was in Arusha region (0.001 for Cr) which is considerably below the acceptable limit. Equation 3 was used to obtain the HI for both regions studied. The HI results were found to be greater than 1, that is, Morogoro and Arusha regions had HI values of 3.6676 and 2.6266, respectively, for all heavy metals of all studied locations (Fig. 12b), suggesting there is a non-carcinogenic health risk related to exposure and ingestion of these five heavy metals collectively through ingestion of fish from these locations. In both regions, the total HI was found to be dominated by Cd contribution, that is, 30 and 36% in Arusha and Morogoro regions, respectively, followed by Hg, that is, 13 and 17%, respectively (Fig. 12b).

(vi)   Estimated Carcinogenic Risk

The CR values below 10^{-6} are generally considered as negligible while above 10^{-4} are unacceptable. The estimated CR values of As, Pb, Cd, and Cr due to exposure from fish consumption from

locations within the studied regions are shown in Fig. 12c. In this study, CR values were between 10^{-4} to 10^{-8}, indicating no cancer risk due to exposure of current concentrations of As, Pb, Cd, and Cr in the fish samples studied from both regions.
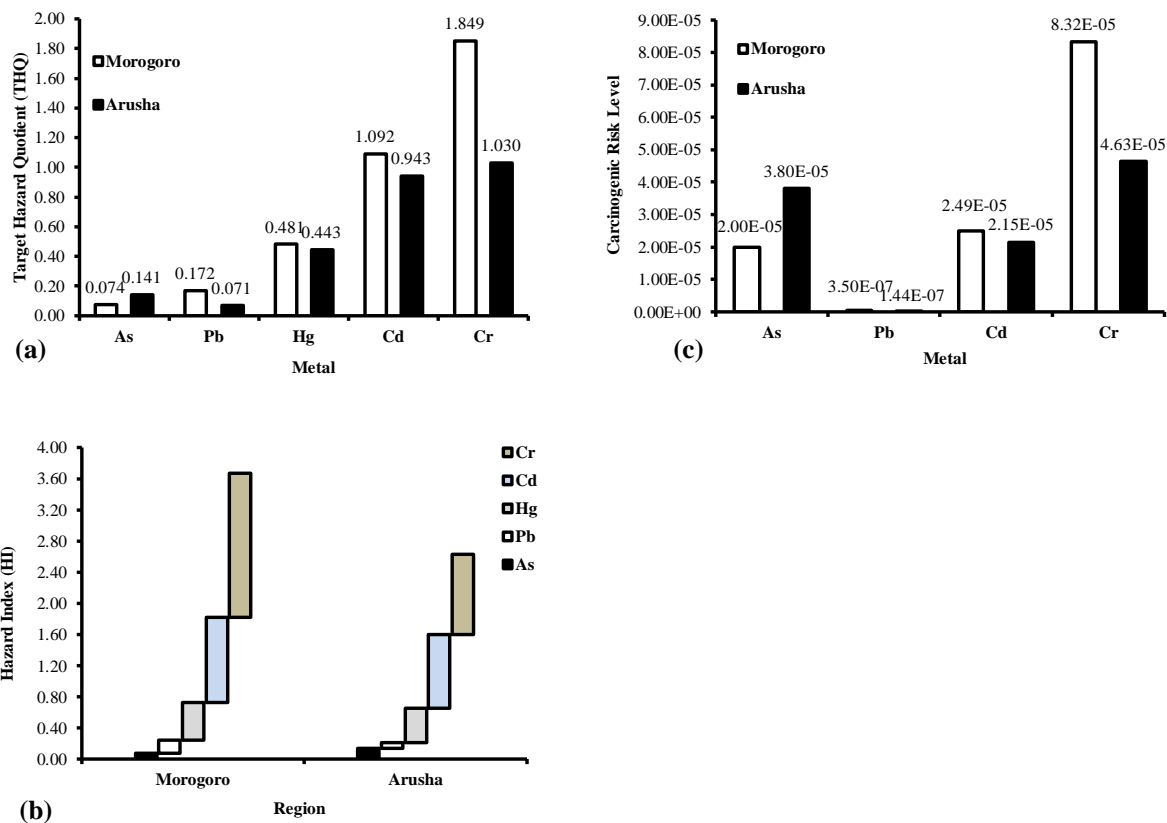
Figure 12: Hazard risk estimation: (a) Target Hazard Quotient (THQ); (b) Hazard Index (HI) (c) estimated carcinogenic risk (CR)

4.1.5 Design of a Context-specific Climate-smart Sustainable Fish Pond

(i) Construction of the Climate-smart Fish Pond

When selecting the materials for the system, affordability and availability were the two critical factors considered. The climate-smart pond was contructed in an open field in an east-west direction to ensure the solar component received plenty of sunlight to power the pump as well as allow for direct rainfall into the ponds. Each of the two climate-smart ponds had dimensions of 2 m length $\times$ 2 m width $\times$ 1.5 m depth, capable of holding at least 4000 liters of water. The pond sat above ground, and the 1m depth was adequate to prevent any external runoff from entering the pond. The base of the ponds sloped at about 3% toward the outlet drain valves. The major components of the first section that had no filter were iron frame support structure, polyethylene membrane (pond liner), and drain valves, while the second section with a filter included solar-powered pump and filteration

system built from locally available materials such as cotton, charcoal, gravel, and sand in a recycled food-grade 55-gallon barrel. The pump helped with pushing used water through the filter layers to be cleaned and recycled back into the pond. This also allowed re-oxygenation of the water through the turbulence created.

All pipes were made of smooth plastic with the standard Hazen-Williams C-value of 150. Only six (6) ninety-degree (90°) pipe elbow fittings were used to channel the water through the filter and back to the pond. The total length of all the PVC pipes in the system was approximately 10 feet. Any losses due to friction and fittings in the PVC pipes were assumed to be negligible, hence, no adjustments were made in flow calculations to account for losses. The pump was capable of pushing water through the filter at a constant rate of 530 liters/hr at 3 feet of head. The ponds were filled with water collected from a small local river with the assumption that it was free of any harmful chemicals.

The construction of the climate-smart fish ponds was commissioned in December 2019 and completed in late January 2020 prior to the start of the experiment in February 2020. One laborer with expertise and experience in welding and plumbing worked on this project. The pond design is affordable, can be built in rural areas of Tanzania using locally available materials, uses little space and can be easily scaled up when expansion is desired.

(ii)   Physicochemical Analysis of Pond Waters

The prototype climate-smart ponds were designed with 1.5 m depth above ground to ensure they are resilient to any external sources of pollution, especially from surface runoffs and flooding. On a biweekly basis, the water in the pond with filter was filtered and recycled back to the pond while for the pond without a filter, half of its water was drained and replaced with fresh supplies from the river. Table 15 shows values (mean ± standard deviation) of quality parameters measured on water sampled from the control pond and the newly designed system with and without filter. In this study, the mean daytime pond water temperature between February and August for the control pond, climate-smart pond with and that without filter were 27.2 ± 0.43 °C, 24.7 ± 0.50 °C and 27.4 ± 0.42 °C, respectively.

Over the same period, the mean pH of pond water sampled from the control pond, climate-smart pond with and that without filter were 9.7 ± 1.94, 6.6 ± 0.28 and 7.4 ± 0.36, respectively (Table 15). The mean nitrate levels in the pond water sampled from the control pond, climate-smart pond with and that without filter were 14.7 ± 3.23 mg/L, 1.6 ± 0.34 mg/L and 3.8 ± 0.78 mg/L, respectively (Table 15). The mean ammonia levels in water samples from the control pond,

climate-smart pond with and that without filter were 1.6 ± 0.71 mg/L, 0.2 ± 0.11 mg/L and 0.5 ± 0.16 mg/L, respectively.

The mean dissolved oxygen (DO) reported in water samples from the control pond, climate-smart pond with and that without filter were 4.3 ± 1.75 mg/L, 7.6 ± 0.77 mg/L and 5.8 ± 1.54 mg/L, respectively (Table 15). In this study, the BOD levels in water samples from the control pond, climate-smart pond with and that without filter were 29.6 ± 4.94 mg/L, 4.3 ± 0.46 mg/L and 12.0 ± 1.26 mg/L, respectively (Table 15).

Carbonates and bicarbonates are the most critical contributors to water alkalinity (quantity of base present in water). In this study, the total alkalinity for water samples from the control pond, climate-smart pond with and that without filter were 57.4 ± 10.36 mg/L $CaCO_3$, 23.2 ± 1.89 mg/L $CaCO_3$ and 45.8 ± 2.45 mg/L $CaCO_3$, respectively.

In this study, the hardness for water samples from the control pond, climate-smart pond with and that without filter were 264 ± 16.92 mg/L $CaCO_3$, 105 ± 3.09 mg/L $CaCO_3$ and 114 ± 6.99 mg/L $CaCO_3$, respectively. Lastly, turbidity (measure of water clarity or discoloration) levels <35 NTU are considered ideal for tilapia production. In this study, the mean turbidity value for water sample from the control pond was >35 NTU, and could be due to dissolved soil particles and floating sediment materials. However, the mean turbidity values for water samples from the climate-smart pond with and that without filter were 3.9 ± 0.58 NTU and 14.3 ± 3.45 NTU, respectively, suggesting they are ideal for fish health (Table 15).

(iii)   Fish Measurements

Fish reared during the six-month period of this study grew to a mean length of 13.1 ± 1.42 cm for control pond (mean weight of 482 ± 78.95 g), 20.2 ± 1.78 cm for climate-smart pond with filter (mean weight of 520 ± 87.30 g), and 18.4 ± 1.54 cm for the climate-smart pond without filter (mean weight of 502 ± 81.90 g) (Table 15). Low-cost, home-made balanced diet formulated using insect-protein and locally available ingredients from plant sources was regularly given to the stock in the climate-smart ponds.

Table 15: Mean physicochemical water parameters in fish ponds in three ponds

	Parameters	Conventional concrete pond (control)	Climate-smart pond with filter	Climate-smart pond without filter
Water quality parameters	Temperature (°C)	27.2 ± 0.43	27.4 ± 0.50	27.4 ± 0.42
	pH	9.7 ± 1.94	6.6 ± 0.28	7.4 ± 0.36
	Nitrate (mg/L)	14.7 ± 3.23	1.6 ± 0.34	3.8 ± 0.78
	Ammonia (mg/L)	1.6 ± 0.71	0.2 ± 0.11	0.5 ± 0.16
	DO (mg/L)	4.3 ± 1.75	7.6 ± 0.77	5.8 ± 1.54
	BOD (mg/L)	29.6 ± 4.94	4.3 ± 0.46	12.0 ± 1.26
	Alkalinity (mg/L $CaCO_3$)	57.4 ± 10.36	23.2 ± 1.89	45.8 ± 2.45
	Hardness (mg/L $CaCO_3$)	264 ± 16.92	105 ± 3.09	114 ± 6.99
	Turbidity (NTU)	38.0 ± 5.08	3.9 ± 0.58	14.3 ± 3.45
Fish measurement	Fish length (cm)	13.1 ± 1.42	20.2 ± 1.78	18.4 ± 1.54
	Fish weight (g)	482 ± 78.95	520 ± 87.30	502 ± 81.90

Values are mean ± standard deviation; *n* is the number of ponds; °C = degrees Celcius; DO = Dissolved Oxygen BOD = Biological Oxygen Demand; mg/L = concentration expressed in milligrams per liter; NTU = Nephelometric Turbidity Units, a turbidimeter (nephelometer) measurement of light intensity as a beam of light passes through a water sample at 90 degrees; cm = centimeter; g = grams

(iv) Technology Needs Assessment to Solve Climate-change Problems

Table 16 summarises the potential of this project to solve food insecurity in rural communities and address gender inequity in aquaculture sector in Tanzania.

Table 16: Technology needs assessment

Problem	Satisfied?	Insights
Food insecurity	Yes	The system is designed for pond fish farming with better freshwater management to maintain aquacultural productivity, support food security and nutrition. The climate-smart pond design also make biosecurity measures possible and provides increased monitoring for water quality and disease outbreaks to minimize production losses, especially for socio-economically vulnerable communities in Tanzania.
Climate change vulnerability	Yes	The design is an environmentally-friendly fish production system. It has the capacity to enhance resilience to climate change and related disasters. For example, the design is raised by 1.5m above ground to prevent influx of flood water that may introduce pollutants, diseases, parasites and harmful algal blooms during rainy seasons. The system also filters and recycles water which is critical during drought when freshwater supplies are scarce.
Gender inequity	Yes	The system is simple to manage; rural women farmers would find it easy to operate.

4.1.6 Feed Formulation

This is the first time Google Sheet Program has been used to formulate fish feed diet (Table 18). The experimental BSFL diet delivered crude protein and crude lipid content that met nutrition requirement of fish based on age and weight. The ratios of spirulina:moringa:BSFL meal in the BSFL diet was 1:1.8:24 as a complete replacement for fishmeal (Table 18). This ratio provided adequate crude protein, crude lipid and essential amino acid requirements of *Oreochromis niloticus* (Table 3). Rice bran and corn bran had relatively high levels of carbohydrates (Table 17), and were primarily included for their protein sparing function (Table 18). Basil powder added aroma and palatability while wheat gluten was primarily used as a binder, but it also contributed crude protein to the diets (Table 18). Based on USDA National Nutrient Database for Standard Reference, the other highest contributors of crude protein were moringa, BSFL meal, sunflower seedcake, and spirulina. Crude lipid mainly came from BSFL meal and rice bran (Table 17).

Proximate chemical analysis validated Google Sheets Program as a useful tool in determining nutritional profile of fish feed formulations. Calculated crude protein and crude lipids in the experimental BSFL diet were 35.34 and 15.92%, respectively; while their values determined through chemical analysis were 34.19 and 19.78%, respectively. These values were relatively higher than those reported for the commercial feed, which were 22.65 and 10.97%, respectively (Table 19). The Google Sheets Software output also indicated that the experimental BSFL diet had high crude fiber value (15.2g/100 g feed), which was in contrast to the analytical value reported (5.88 g/100 g feed). A similar large difference was also observed between the computed and analyzed crude ash values for BSFL diet (Table 19). These differences indicate the importance of having all nutritional data populated in the Nutrasheets[TM] especially for those ingredients such as BSFL that aren't found in the USDA National Nutrient Database for Standard Reference. There was no significant (P<0.05) difference in proximate compositions of crude fiber, crude ash and DM between both the experimental BSFL diet and commercial diet (Table 19). In this study, significantly (P<0.05) higher content of NFE was found in the commercial feed (53.65 ± 0.93 g/100 g feed) compared to the experimental BSFL diet (38.83 ± 0.74 g/100 g feed). On the other hand, from Google Sheet Program output, concentrations of all essential amino acids in the experimental diet were consistent with the Essential Amino Acid (EAA) requirements for *Oreochromis niloticus* (Table 19).

In terms of growth performance, there was significant (P<0.05) increase in length and weight of fish fed on experimental BSFL diet compared to those fed on commercial diet (Table 19). The length and weight of fish fed on BSFL diet were 1.26× and 1.69× greater than those fed on commercial diet, respectively (Table 19). The average daily weight gain for the fish fed on BSFL diet was

significantly (P<0.05) higher (1.76× greater) than for those fed on commercial feed. Feed intake between the two diets differed significantly (P<0.05) throughout and was 1.39× higher with the experimental BSFL diet relative to the commercial feed (Table 20). Protein efficiency ratio (PER) was significantly (P<0.05) higher when fish were fed the experimental BSFL diet even though their protein intake was relatively lower (Table 20). Fish fed with the commercial feed had comparatively better feed conversion ratio (FCR) (17.9 ± 0.16) compared to those fed with experimental BSFL diet (14.4 ± 0.09). However, the specific growth rate (SGR) followed the same trend as mean weight gain, average daily weight gain and protein intake. From our results, the SGR in fish fed with experimental BSFL diet was 1.2× better during the 90 days of feeding regimen than those fed with commercial diet (Table 20).

Table 17: Composition of essential amino acids (EAA), crude protein (CP) and crude lipid of the main ingredients (as is)[1]

EAA	Rice bran[2]	Corn bran[2]	Fresh basil[2]	Sunflower seedcake[2]	Moringa leaf powder[2]	Spirulina seaweed[2]	Black soldier fly larvae meal[3]
Tryptophan	0.11		0.04	0.74	0.05	0.93	0.58
Threonine	0.56		0.10	1.96	0.09	2.97	1.54
Isoleucine	0.57		0.10	2.40	0.08	3.21	1.73
Leucine	1.02		0.19	3.50	0.21	4.95	2.80
Lysine	0.65		0.11	1.98	0.13	3.03	2.26
Methionine	0.31		0.04	1.04	0.22	1.15	0.76
Phenylalanine	0.64		0.13	2.47	0.12	2.78	1.63
Valine	0.88		0.13	2.78	0.11	3.51	2.48
Arginine	1.06		0.12	5.07	0.13	4.15	2.00
Histidine	0.36		0.05	1.33	0.07	1.09	1.24
Carbohydrate	49.69	85.64	2.65	35.83	50.0	23.90	
Crude Protein	13.35	8.36	3.15	48.06	30.0	57.47	42.1
Crude Lipids	20.85	0.92	0.64	1.61	0	7.72	26.0
% Moisture	6.13	4.71	92.06	7.47	8.00	4.68	4.00

[1]Values reported as is. Google Sheets Program uses individual moisture content of each ingredient in computing best practical output based on desired evaporation/absorption rate.
[2]USDA National Nutrient Database for Standard Reference.
[3]EAA profile of black soldier fly meal reared in vegetable waste (Spranghers *et al.,* 2017)

Table 18: Formulation and composition of experimental Black Soldier Fly diet and control food (g/100 g diet)

Dry ingredients	Proportion (%)		Function
	BSFL Diet[1]	Control diet (commercial feed)	
Rice bran	11.0	25.0	Provide high amounts of carbohydrates which are relatively inexpensive sources of energy that may spare protein (which is more expensive) from being used as an energy source.
Corn bran	10.8	21.0	
Basil powder	1.2	0	Source of micronutrients. Adds aroma and increases palatability. Has antimicrobial properties.
Sunflower seedcake	14.0	12.0	Source of lipids. Allow for good water stability.
Moringa leaf powder	4.0	0	Rich in proteins, vitamins, carotenoids, ascorbic acid and minerals e.g. iron.
Spirulina seaweed	2.2	0	Cyanobacteria, a natural pigment. Also source of high levels of crude protein and essential amino acids.
Black soldier fly larvae (BSFL) meal	52.8	0	Excellent source of crude protein and lipids.
Vital wheat gluten	4.0	4.0	Binder material to provide stability to the pellet.Excellent sources of protein.
Soybean meal	0	18.0	Source of protein and metabolizable energy.
Groundnut cake	0	20.0	Source of protein, carbohydrate, crude fibre and minerals.
Total (%)	100	100	
Ratio[2]	1:1.8:24	0	

[1]Quantities weighed as is. Google Sheets Program uses individual moisture content of each ingredient in computing best practical output based on evaporation/absorption desired.
[2]Ratio of spirulina:*Moringa oleifera* leaves:black soldier fly larva (BSFL) meal for fish meal replacement.

Table 19: Calculated and proximate nutritional as well as amino acid composition (g/100 g of diet) of diets used in the study

Parameters	BSFL Diet	BSFL Diet	Control diet (commercial feed)	Nile tilapia requirement[1]
	Google Sheets Values[2]	Proximate Analysis Values[3]		
Crude Protein(g/100g)	35.34	34.19 ± 0.37^a	22.65 ± 0.71^b	
Crude Lipids (g/100g)	15.92	19.78 ± 0.06^a	10.97 ± 0.14^b	
Crude Fiber (g/100g)	15.2	5.88 ± 0.10^a	5.20 ± 0.08^a	
Carbohydrate (g/100g)	21.9	—	—	
NFE	42.82	38.83 ± 0.74^a	53.65 ± 0.93^b	
DM (%)	90.0	82.3 ± 0.15^a	90.7 ± 0.26^{ab}	
Gross Energy (kJ/g)[4]	15.56	21.1 ± 0.02^a	18.92 ± 0.28^b	
Ash (%)	5.92	10.20 ± 0.24^a	12.73 ± 0.47^a	
P/E ratio[5]	13.7	12.58 ± 0.36^a	18.92 ± 0.28^b	
Cost ($/kg)	0.35	1.55		
EAA (g/100g, DM)[6]				
Tryptophan	0.4	—	—	0.28–0.3
Threonine	1.1	—	—	1.05–1.1
Isoleucine	1.2	—	—	0.87–1.0
Leucine	1.9	—	—	0.95–1.9
Lysine	1.4	—	—	1.43–1.6
Methionine	1.0	—	—	0.75–1.0
Phenylalanine	1.2	—	—	1.05–1.6
Valine	1.6	—	—	0.78–1.5
Arginine	1.7	—	—	1.18–1.2
Histidine	0.8	—	—	0.48–1.0

[1]NRC (1993, 2011).
[2]Google Sheets Software output values.
[3]Proximate analysis values (n =3).
[4]Calculated using Crude Protein = 23.9 kJ/g, Crude Lipids = 39.8 kJ/g, NFE = 17.6 kJ/g (Schulz *et al.*, 2005).
[5]P/E = Protein to energy ratio in mg protein per kJ gross energy.
[6]Google Sheets Software calculated values using complete protein and protein digestibility factor of 0.91.
— Not determined.

Table 20: Growth performance and feed utilization of *Oreochromis niloticus* fed with experimental Black Soldier Fly diet compared to commercial feed for 90 days

Parameters		Control diet (commercial feed)	Experimental diet BSFL feed
Average Fish length (cm)	Initial	3.4 ± 0.97	3.4 ± 0.97
	Final	8.5 ± 1.12^a	10.9 ± 1.54^b
Average Fish Weight (g)	Initial	4.8 ± 0.95	4.8 ± 0.95
	Final	111 ± 17.20^a	188 ± 20.11^b
Mean weight gain (g)		106 ± 16.58^a	183 ± 19.40^b
Average daily weight gain (g)		1.26 ± 0.04^a	2.17 ± 0.02^b
Feed intake (g)		1896 ± 9.22^a	2642 ± 10.04^b
Protein intake (g)		83.7 ± 4.88^a	77.3 ± 5.26^b
Protein efficiency ratio (PER)		1.26 ± 0.15^a	2.36 ± 0.13^b
Specific Growth Rate (SGR%)[1]		3.49	4.08
Feed conversion ratio (FCR)		17.9 ± 0.16^a	14.4 ± 0.09^b

[1]t used in calculation is 90 days.

Figures in each row with different superscript are significantly different ($P < 0.05$) from each other.

4.2 Discussion

4.2.1 Farmer's Knowledge on Aquaculture Management Practices

Aquaculture is essential because fish demand is increasing. Fish is a vital source of animal protein in the human diet. Commercial fish farming supplements capture fisheries. In Tanzania, fish farming plays a great role in food security and livelihood to many households.

In Arusha and Morogoro regions, the most common holding structures for aquaculture production were found to be earthen ponds and concrete tanks. These culture methods have become more intensive for producing higher yields (Akinwole *et al*., 2014) to meet the demand level for fish. The choice of culture facility could have been influenced by the cost of fish pond establishment and availability of space or awareness of available innovations.

Fish farming in Tanzania is practised by smallholder producers using various production systems. Intensive monoculture system, where only one fish species is raised, was the most predominant aquaculture method used by farmers in both regions. As previously noted by Adeogun *et al*. (2007) and Akinwole *et al*. (2014), this culture system enables the farmer to make the feed that will meet the requirement of a specific fish species. As other authors previously reported (Brummet *et al*., 200), tilapia was the most cultured fish species in both zones followed by catfish. Tilapia is a traditional and favourite dish in Africa. In Tanzania, for example, it is consumed as an affordable source of protein in poor rural communities as well as in affluent urban centres. Therefore, the markets for tilapia are diverse (Norman-Lopez *et al*., 2008) and tilapia product prices are increasingly becoming favourable for traders.

However, commercial fish farmers face many production challenges. One of the biggest constraints in aquaculture production in Tanzania is the high cost of commercial nutritious feed. Fish farmers

require high quality feeds to produce high-quality fish that would attract commensurate prices high enough to ameliorate whatever constraints they may face. In fact, fish feed and feeding are reportedly responsible for over 70% of operating cost in fish production (Edet *et al*., 2018). This ultimately cuts into the farmers' profits. This findings revealed that most of the fish farmers in Arusha and Morogoro regions preferred to formulate their own fish feed using locally available raw ingredients such as vegetable proteins and cereal grains to reduce feeding cost. Another related study previously reported that fish producers have to cope with high production costs associated with fish feeds (Gabriel *et al*., 2007). The good fish feed should provide proper nutrition so that the fish can feed efficiently and grow to their full potential.

Despite the fact that the locally formulated fish feed can significantly reduce fish farming cost (Gabriel *et al*., 2007), fish farmers must understand the nutritional requirements of their fish stock at every stage while developing the fish feed. Fish feed formulations and feed preparation require knowledge of the nutritional requirements for various fish species and the skill in feed manufacturing. Nutrients essential to fish are similar to those required by most other animals. Fish nutrition, however, is an inexact science (Aizam *et al*., 2018). Fish feed blends or formulas must deliver balanced nutrients and should consider fish age and specific nutritional requirement. Though the farmers we surveyed are able to prepare their own fish feeds from locally available ingredients, the majority lacked the basic knowledge and technology for proper feed formulation for their tilapia and catfish stocks. Arguably, these farmers reported a high percentage of fish loss and low fish weight gain than expected. A fish feed with low nutritional values and poor texture can decrease fish appetite; poor feeding will, in turn, increase their susceptibility to diseases, morbidity, and mortality (Elfitasari & Albert, 2017). In this study, we observed that the farmers never analyzed the locally grown raw ingredients they used in making their fish feeds prior to use or the finished blends for nutrient content. It is concerning how little importance was placed on fish nutritional needs and diet.

Pond feeding can also be done using manure (livestock waste), which is an ecologically appropriate method for raising fish. Manure in the ponds provides energy for the fish as well as nutrients and organic matter for autotrophic and heterotrophic production. Even though the use of manure in aquaculture reportedly reduces feed costs and enrich the ponds with additional food sources such as planktons (Kang'ombe *et al*., 2006), this system require fish farmers to adhere to good aquaculture practices to ensure safety of the consumers, especially against pathogenic microorganisms (Kamaruddin *et al*., 2015).

Related to that, fish farmers using manure should never use feed containing banned compounds considered harmful to humans. Similarly, farmers utilizing wastewater to rear fish must consider the potential risk of contaminants and other industrial chemical residues that might compromise the safety of the consumers (Uddin *et al*., 2018; Ali *et al*., 2016). Undoubtedly, fish grown under environmentally friendly practices and good aquaculture practices is a responsible way for farmers to avoid banned substances in their facilities. In this study, all the fish farmers surveyed reported not to use any of the internationally banned compounds in their facilities.

Water quality plays an important role in the production of fish, especially in intensive aquaculture systems (Boyd, 2017). In fact, suitable water quality parameters are pre-requisite for the healthy production of sufficient fish and fish food. The productivity of a water body depends on the physical, chemical, and other intrinsic factors. In Arusha and Morogoro regions, boreholes and tap water are the two major sources of water used by the fish farmers.

Pond sizes the farmers use in both regions are depicted in Table 1. Pond water capacity depends on pond depth and size. Fluctuation in water depth would result from evaporation, rain and water seepage. Inadequate water depth is one of the most important factors for fish mortality (Baleta *et al*., 2019). Lower water depth would not provide the fish with sufficient space for movement and feeding. In this study, the fish farmers in both regions reported water scarcity during the dry season and flooding during the rainy season. For those using boreholes, water scarcity resulted from low water level often experienced during prolonged drought. Some farmers (15%) reported using rivers as their main source of water for their ponds. There are multiple challenges fish farmers in both region face regardless of the water source. Two issues that were common were high water temperature and flooding. The farmers complained about high water temperature during dry hot seasons or drought. Various strains of tilapia and catfish differ with respect to their tolerance to water temperature in terms of feeding, growth, and spawning. However, overall, the ideal water temperature for good health and growth should be between 20-30 °C. It's been reported that when pond water warms up, the metabolic rates of tilapia and catfish also rise, leading to, in some cases, death (Qiang *et al*., 2019).

Flooding during the rainy seasons was the other huge concern for the fish farmers. Flooding results in losses when pond structures are compromised and fish wash away. Flooding also increases fish susceptibility to diseases, infections, and contamination, potentially from the compounds from industrial and agricultural runoff (Reid *et al*., 2019; Rutkayova *et al*., 2018). Even though some fish farmers acknowledged that the integrity of their pond structures was a challenge during rainy

seasons, no statistical correlation between pond structure and diseases occurrence was observed. This could have been due to the fact that the data were collected during the dry season.

In general, monitoring water quality parameters require technical knowledge and appropriate equipment. Majority of the farmers surveyed did not apply proper management of water quality due to lack of basic aquaculture management knowledge and technique. It was observed that most of the fish farmers rely on visual checks to monitor the quality of their pond water. Over 70% of the farmers did not treat the water in their ponds before adding the fingerlings. The water of poor quality can cause diseases and infections as other authors previously reported (Mishra *et al.,* 2018). It was also found that the farmers discharged the wastewater from their ponds into the environment untreated. It's well understood that a load of pollutants in wastewater such as suspended solids, nitrates, phosphates, trace elements and microorganisms can lead to pollution of natural water bodies (Amirkolaie, 2008; Cao *et al*., 2007).

Good practice in the management of pond water is, therefore, necessary to avoid or reduce the negative impacts of aquaculture effluents on the environment. Proper wastewater management and practices by the fish farmers can help protect the future of natural water resources. Overall, control of aquaculture health is critical for fish farmers to realize maximum productivity. In this regard, farmers must invest in disease diagnostic strategies to prevent outbreaks that often lead to significant stock losses. Unfortunately, it was found that fish farmers had poor disease diagnostic capacity for preventing and controlling potential infectious diseases of their fish in an aquaculture environment. From the observations, a limited number of fish farmers had proper aquaculture training. The majority had poor record-keeping practices and lacked health management plans. Aquaculture disease diagnoses require special knowledge and technique without which proper diagnosis can be complicated and challenging for the uneducated.

In this respect, good farm management, proper disease diagnosis and prevention based on globally accepted principles are some applicable strategies for ensuring sustainable aquaculture which can be recommended for fish farmers locally. It is therefore important for the farmers to train and qualify for global Good Aquaculture Practices (GAP) certification as per the global GAP aquaculture standard of 2013. None of the fish farmers surveyed had this important certification. No wonder many fish farms in developing countries with poor farm management methods and untrained personnel experience the high occurrence of disease outbreaks (Opiyo *et al*., 2018).

In aquaculture, disease infections come in many forms and can occur at any stage of growth but the highest mortalities are in fingerlings. The most important bacterial infection is bacillary necrosis of *Pangasius* (BNP) followed by motile aeromonad septicaemia (MAS) (Phu *et al*., 2016). Both

diseases are common during the beginning of wet rainy seasons. In this study, very few farmers reported cases of disease outbreaks in general.

Antibacterial drugs added to feeds are the most common treatment for BNP and MAS. However, there were some farmers who preferred dissolving the antimicrobial powder into a solution before adding into the pond water. Oxytetracycline was the most commonly used antibacterial drug by fish farmers surveyed in both regions. Farmers need to have proper knowledge of antimicrobial drug use because abuse and misuse can cause widespread resistance to several commonly used drugs. (Chuah *et al*., 2016; Ye *et al*., 2013; Romero *et al*., 2012). Studies have shown antibiotic resistance by some pathogenic bacteria to streptomycin, chloramphenicol and enrofloxacin (Liu *et al*., 2017).

External parasitic infections predispose fish to bacterial infections (Huston & Cain, 2018) leading to reduced growth and thus poor weight gain. Protozoan parasites are especially problematic and can be severe during the wet rainy seasons. The most common parasiticides used by fish farmers to treat their ponds were mebendazole, copper sulfate and trichlorfon added to the water. Based on this survey, it was observed that many fish farmers lacked the technical training required to diagnose aquatic diseases and make informed treatment choices in case of an outbreak. Most of them relied on past experiences. It's therefore easy to see how drugs and chemicals can be misused.

A pond that has high-quality clean water is important in producing healthy fish. Disinfectants can be used throughout the production cycle for the purpose of improving the quality of the water and disinfecting the farm as well as personal protective equipment (PPE). In this study, it was found that fish farmers used various chemical agents for disinfection primarily on PPEs and treating pond water. Chlorine and iodine solutions were the most commonly used disinfectants in Morogoro and Arusha, respectively. Chlorine, when used, must be neutralized to avoid killing of fish. Additionally, organic matter in water can react with chlorine and calcium hypochlorite leading to unintended toxicity (Macedo *et al*., 2019). Iodine and iodine-containing compounds reported by the farmers in both regions can be toxic and must be adequately rinsed off when used to disinfect PPEs (Postigo & Bozo, 2019). Sadly, none of the farmers surveyed was aware of the food safety hazards and environmental threat associated with the use of these chemical agents.

4.2.2 Prevalence of Fish Parasites in Nile Tilapia (*Oreochromis niloticus*) and African Catfish (*Clarias gariepinus*) and Physicochemical Characteristics of Pond Water

Aquaculture productivity is dependent on a wide range of factors. Successful management of aquaculture systems requires an understanding of water quality parameters, which is determined by abiotic factors such as temperature, pH, nitrate, ammonia, dissolved oxygen (DO), biological oxygen

demand (BOD), alkalinity, turbidity, and hardness, among others (Bhatnagar & Devi, 2013). These water quality parameters can have profound effects on pond productivity, fish health and oxygen availability.

The ideal water temperature considered optimum for feeding, spawning, good health, and growth of Nile Tilapia and African catfish is about 25 to 27 °C (Kausar & Salim, 2006). The mean temperature previously reported in 13 fish ponds located in urban and rural areas of Morogoro region was 26 ± 3.1 °C (Mdegela *et al.,* 2011). Ngugi *et al.* (2007) gave a range of between 20 and 35 °C as ideal for tilapia culture. These previous studies are consistent with our current findings.

Different types of fish tolerate different pH levels. The ideal pH for tilapia culture ranges between 6.0 to 9.0 (DeWalle *et al.,* 2011). Fish and other aquatic vertebrates have an average blood pH of 7.4. Therefore, ponds with pH levels close to fish blood pH would be ideal, the majority of which were found in Morogoro region. The pH below 5.0 or above 10 may stress fish and cause heavy mortality (Ekubo & Abowei, 2011). The pH values higher than 10.0 were reported in ponds within Samalia and Maweni in Arusha region. From our results, some fish farmers in Arusha and Morogoro must begin monitoring the pH by recording weekly readings to provide an excellent indication of any developing problem. Previously, Mdegela *et al.* (2011) reported mean pH from 13 fish ponds located in urban and rural areas of Morogoro as 6.8 ± 0.8, which was within the recommended standard range for Nile tilapia and African catfish production.

Naturally, water is saturated with dissolved oxygen (DO) in equilibrium with air but fluctuates considerably depending on the prevailing temperature of the water (Eze & Ogbaran, 2010; Meck, 2000). Decreased DO in pond water (especially during the night when photosynthesis stops and fish releases CO_2 through respiration) may lead to poor feeding of fish, starvation, reduced growth, and fish mortality (Bhatnagar & Garg, 2000). The DO level > 5 mg/L is essential for good pond productivity (Bhatnagar & Singh, 2010; Bhatnagar *et al.,* 2004). Though sensitivity to low levels of DO is species-specific, most fish species are distressed when DO falls to 2 – 4 mg/L, leading to detrimental effects on growth and feed utilization, while mortality usually occurs at concentrations less than 2 mg/L (Bhatnagar *et al.,* 2004). In this study, the DO levels were measured in pond water samples collected during the day. From the results, 80% of the sites studied in Morogoro region had mean DO levels ranging from 5.3 ± 2.3 mg/L to 9.1 ± 4.1 mg/L, which are excellent for pond life. On the other hand, average DO levels in ponds studied in Arusha ranged from 3.6 ± 0.8 mg/L to 4.3 ± 1.8 mg/L, and would most likely be lower during the night (Boyd, 2010). This is particularly concerning in constrained environments considering a concentration of >4 mg/L DO is recommended for optimum pond life (Ntengwe & Edema, 2008). Reports indicate that African

catfish can tolerate 20 to 30 mg/L CO_2 in pond water if DO concentration is above 5 mg/L (Wanja *et al.,* 2020). Since aquatic plants use CO_2 for photosynthesis, water quality guidelines established by Bhatnagar and Devi (2013) provide a framework that small-scale fish farmers in Arusha and Morogoro may use to control amounts of aquatic weeds and phytoplankton in their ponds to manage DO levels. One must note, however, that minor fluctuations in the DO content in pond water are a natural occurrence and that fish have developed adaptive mechanisms to cope with these changes.

The Biological Oxygen Demand (BOD), which is a measure of biodegradable organic matter in the ponds, has an inverse relationship with DO. High BOD levels in pond water might be harmful for aquatic life (Mukherjee & Dutta, 2016) and can arise from unconsumed feed, fish waste, as well as surface runoff and soil erosion caused by rainfall (Odokuma & Okpokwasili, 1996). The recommended BOD level in pond water is 20 mg/L (Boyd, 2003). In this study, BOD values for Arusha were consistently lower than for Morogoro sites, suggesting a geographic influence on water sources and organic matter composition. The highest BOD was found in Kihonda in Morogoro region, suggesting higher organic matter in these ponds than the rest. Such a load of organic matter upon degradation can mineralize to release sufficient nutrients for phytoplankton and other aquatic plants to thrive.

Water alkalinity and hardnes levels were measured because they can have profound effects on pond productivity. Generally, a total alkalinity of > 20 mg/L $CaCO_3$ is necessary for good pond productivity. Lower alkalinity reduces buffering capacity of water (Eze & Ogbaran, 2010). From the results, alkalinity values measured in both regions ranged between 20 – 80 mg/L and varied from site to site. This results concur with Mdegela *et al.* (2011) report who found mean alkalinity values in fish ponds located in Morogoro region as 78.7 ± 34.1 mg/L. In concrete ponds, lime may leach out into the water and increase the alkalinity of the pond. Thus, alkalinity is also related to the amount of dissolved calcium magnesium in the water and tends to be higher in harder water (Eze & Ogbaran, 2010). However, bacterial actions, which release acidic compounds, naturally neutralize the basic components into the water and helps decrease alkalinity.

Hardness level in the range of 100 to 250 mg/L is ideal for aquaculture; a value of 250 mg/L hardness matches the calcium concentration of fish blood (Wanja *et al*., 2020). In this study, the only sites that had water hardness levels lower than 100 mg/L were Manyata and Samalia, both in Arusha region. Fish farmers in these locations should consider adjusting hardness using agricultural limestone or use another suitable source of water for maximum productivity. All sites in Morogoro region had water hardness in the range of $\geq 189 \leq 300$ mg/L, and it would be reasonably safe to assume that these hardness levels reflect sufficient calcium concentrations for fish. Previously,

Mdegela *et al.* (2011) reported average water hardness of 76.6 ± 24.1 mg/L from 13 fish ponds located in Morogoro urban and rural areas, suggesting possible differences in characteristics of soil and bedrock where ponds surveyed in this study were located.

Nitrogen is usually present in fish ponds as ammonia or nitrate. Ideally, the ammonia concentration in pond water should be zero. The minimum acceptable ammonia level suitable for pond fishery is < 0.2 mg/L (Bhatnagar & Singh, 2010). According to the Bureau of Fisheries and Aquatic Resources (BFAR), ammonia levels of between $0.02 – 0.05$ mg/L are optimum for tilapia growth (Makori, 2017). The results are in contrast to these values. In this study, the ammonia levels in Arusha and Morogoro regions ranged from 1.0 ± 0.7 mg/L to 1.9 ± 1.0 mg/L and 1.0 ± 0.8 mg/L to 1.6 ± 1.0 mg/L. Fortunately, the alkalinity levels in all sites studied were ≤ 80 mg/L; this is desirable since the less toxic ionized form of ammonia (ammonium, NH_4^+) is more prevalent in low alkaline waters (Wurts & Durborow, 1992). Previously, Mdegela *et al.* (2011) reported ammonia levels of 1.0 ppm in fish ponds located in Morogoro urban and rural areas, which concurs with this findings in Kihonda, Mkindo and Tangeni sites. Reports indicate that ammonia concentration of >0.6 mg/L in pond water can cause mortality in fish (Wanja *et al.,* 2020). Sadly, all ponds surveyed lacked aeration, yet several farmers failed to replace pond water regularly throughout the production cycle. For such farmers, the addition of quicklime is potentially a cost-effective way to manage ammonia levels within their fish ponds.

The favorable range of nitrate in the aquaculture pond is 0.1 to 4.0 ppm (Santhosh & Singh, 2007). The presence of nitrate in ponds could be from the fish feed and surface water runoff. Excessively higher nitrate concentrations in pond water are indicative of pollution (Eze & Ogbaran, 2010). Mdegela *et al.* (2011) reported extremely low nitrate concentrations (0.20 ppm) in water samples from multiple ponds within Morogoro region, which included Mkindo where it was found levels in the range of 3.1 ± 0.7 mg/L. Though nitrate is very important to aquatic plants, its concentrations in the pond water should be controlled to avoid eutrophication. This can be effectively achieved through routine water changes and utilization by plant algae (Meck, 2000).

Turbidity measurements were taken to determine water clarity or discoloration levels. Reports indicate turbidity levels between 30 to 35 NTU are favorable for tilapia growth and the lower the better for freshwater fish (Ojwala, 2018). In this study, turbidity values varied considerably from site to site, but were all less than 35 NTU, therefore are ideal for fish health. Though turbidity is usually only an aesthetic problem (such as muddy water), high turbidity can hinder sunlight penetration into the pond, affecting pond life. Zooplankton blooms, surface runoffs, disturbance of sediments by fish, and using rivers as a water source can all affect pond water discoloration.

Overall, substandard fish environment and poor pond management were observed in multiple fish ponds surveyed in the current study, especially in farms with earthen ponds which often provide a favorable environment for benthic microinvertebrates to proliferate (Kirjuśina & Vismanis, 2007). A majority of the farmers in the rural areas had no access to extension services of fish farming. However, the lack of disease occurrence in several sites studied despite poor management of some ponds may be ascribed to the fact that fish, especially Tilapia do not frequently succumb to disease endemics and have remarkable mechanisms for recovery from infections (Kirjuśina & Vismanis, 2007).

Monogeneans are flatworms (Platyhelminthes), are host- and site-specific, ectoparasitic and have special posteriorly positioned organs for attachment onto their host's skin or gills (Iyaji *et al.,* 2009). Monogenean trematodes such as *Dactylogyrus* sp and *Gyrodactylus* sp can proliferate rapidly especially in ponds under high stocking densities as their life cycle require only one host, thus poses a great threat to fish cultures (Mansell *et al.,* 2005). In this study, *Dactylogyrus* sp and *Gyrodactylus* sp were recorded from the gills of Nile tilapia and African catfish samples from both Arusha and Morogoro regions, and their prevalence varied by region. Multiple reports exist on the prevalence of monogenean infection in East Africa. For instance, in Kenya, *Dactylogyrus* sp was reported in farmed tilapia at a prevalence of 48.1% in Nyeri county (Mavuti *et al.,* 2017) and 3.5% in Kiambu county (Maina, 2017), which affected the health and quality of fish. Thus, fish farmers in Tanzania need to be informed that if left uncontrolled, monogenean infestation could lead to serious high morbidity and mortality and thus economic losses.

In this study, *Acanthocephalus* sp had the highest prevalence among all parasites recovered, which concurred with studies conducted by Ashmawy *et al*. (2018). The *Acanthocephalus* sp. are mostly endoparasitic with at least one intermediate host in their life cycle. The *Acanthocephalus* sp was recovered from the gastrointestinal tract of both Nile tilapia and African catfish and found a significant ($P < 0.05$) difference in the prevalence of this parasite between all sites studied in both regions. Comparatively, Mavuti *et al.* (2017) isolated *Acanthocephalus* sp from tilapia reared in earthen ponds at a lower prevalence of 0.8% in Tetu, Nyeri County, Kenya. Florio *et al.* (2009) also found prevalence of *Acanthosentis* sp in Kenya (7.1%) and Uganda (13%) in all the fisheries systems (wild and caged). Additionally, Chacha and Lamtane (2014) reported the prevalence of *Acanthocephalus* sp as high as 47.9% in unconfined environments such as Lakes Uba and Ruwe in Tanzania. The disparity between this results and these other reports can be ascribed to the differences in feeding habits or the availability of intermediate hosts such as crustaceans. Earthen ponds with overgrown vegetation are the most at risk of encouraging *Acanthocephalus* sp infestation because intermediate hosts such as amphipods, isopods, copepods, or ostracods thrive in such environments

(Eyo *et al*., 2013). In the survey, some farmers admitted sourcing fingerlings from fellow farmers whose quality is unknown. This could lead to the transmission of parasites such as *Acanthocephalus* sp between farms. Furthermore, since most farmers produced fish on a subsistence level, several admitted to practicing partial harvesting and left some fish to continue inbreeding in the ponds.

Digenean trematodes such as *Diplostomum* metacercariae require multiple hosts (e.g. snails, fish, and piscivorous birds) to propagate and can be found externally or internally in the fish organs (Thon *et al.,* 2017). *Diplostomum* sp In this study, *Diplostomum* sp was recovered from the vitreous humour of the eyes of fish and found that it infested significantly high numbers of African catfish in Arusha (72.7%) than in Morogoro region (15.7%). Significant ($P < 0.05$) difference in the overall prevalence of *Diplostomum* sp was found in both Nile tilapia and African catfish in Arusha and Morogoro regions. A similar influence of ecological zone on parasite infestation was also reported in Kenya where the prevalence of *Diplostomum* sp in farmed Nile tilapia was found as 54.3% in Tana River (Mathenge, 2010) and 1.9% in Nyeri county (Mavuti *et al.,* 2017). Earthen ponds are good environments for digenean helminthes because snails, crustacean copepods and leeches can be vectors/intermediate hosts. Eliminating intermediate hosts through good aquaculture management can substantially minimize its prevalence in aquaculture systems.

In this study, the larvae of *Contracaecum* sp were recovered from the abdominal cavity of Nile tilapia and African catfish samples from both Arusha and Morogoro regions. In both regions, the occurrence of *Contracaecum* sp was higher in African catfish than Nile tilapia. This may be related to diet and water sources. Both fish species feed on all intermediate hosts including detritus, benthic invertebrates like arthropods, mollusks, mud, and other small fish, potentially accumulating the parasite larvae (Mavuti *et al*., 2017; Thon *et al.,* 2017). This presents a huge challenge to farmers using earthen ponds because chance proximity of infected mollusks has implications on *Contracaecum* sp. prevalence. Therefore, the aquaculture sector must focus on the control of nematodes to safeguard public health and prevent production losses.

Protozoans in fish are highly pathogenic. Ciliophora, especially trichodinids, is characterized by cilia for locomotion, round shape when seen dorsally, and a ring with hook-like denticles (Mavuti *et al*., 2017). In this study, *Trichodina* sp was recovered from the skin of Nile tilapia and African catfish samples. Compared to this results, Mavuti *et al*. (2017) reported *Trichodina* sp occurred in both farmed Nile tilapia and African catfish at a much lower overall prevalence of 1.4% in Nyeri County, Kenya. All infested fish were from earthen ponds, which are highly prone to siltation and vegetation cover that often support parasitism. Overstocking and poor pond management can exacerbate protozoan infections in fish. Other reports from the East Africa region showed a high

prevalence of ciliates in various aquaculture systems (Florio *et al.,* 2009, Akoll *et al.,* 2012). Therefore, for profitable and sustainable aquaculture in Tanzania, awareness of fish health and good farm management practices should be encouraged.

Leeches are differentiated from monogenean parasites by the presence of body segmentation. Leeches suck blood from the soft tissues and exposed organs, which can potentially kill the fish (Mavuti *et al.*, 2017). In this study, leeches were observed in gills, nostrils, and the anus of both Nile tilapia and African catfish samples. The use of rivers as a source of water could be a possibility of leeches being introduced into farmed fish reared exclusively in concrete ponds. Comparatively, Mavuti *et al.* (2017) reported leeches in the gills of farmed Nile tilapia and African catfish with an overall prevalence of 2.7% in Nyeri Central sub-county, Kenya. Additionally, Iyaji and Eyo (2008) reported leeches on the mouths of silver catfish with an overall prevalence of 19%.

4.2.3 Occurrence of Fish Bacteria Pathogens Isolated from Farmed Nile Tilapia (*Oreochromis niloticus*) and African Catfish (*Clarias gariepinus*)

In this present study, 19 species of bacteria were isolated and identified from Nile tilapia and African catfish farms in Arusha and Morogoro. In the study area, farmers were practicing small scale fish farming; Nile tilapia was the most commonly farmed fish in both regions, thus more bacteria species were identified in the tilapia farms. *Aeromonas sobria* and *Edwardsiella tarda* were the two most prevalent fish bacteria found, both in Nile tilapia and African catfish farms across Arusha and Morogoro. Other researchers have also reported similar findings in farmed fish elsewhere (Wamala *et al.,* 2018; Walakira *et al.,* 2014; Joh *et al.,* 2013; Newaj-Fyzul *et al.,* 2008; Ribeiro *et al.,* 2010), indicating the ubiquitous nature of these pathogens in the aquatic environment. *Aeromonas sobria* and *Edwardsiella tarda* are common pathogens known for causing heavy mortalities in farmed and wild fish (Mohanty & Sahoo, 2007; Hemstreet 2010; Nielsen *et al.,* 2001; Walakira *et al.,* 2014).

Vibrio cholera was the most common human bacterial pathogen isolated from both Nile tilapia and African catfish farms in Arusha and Morogoro followed by *Pseudomonas aeruginosa*, *Enterococcus faecalis*, *Escherichia coli*, *Salmonella typhi*, *Shigella dynteriae* and *Staphylococcus aureus*, across both regions, suggesting possible contamination with human wastes, especially faecal contamination from sewage and surface runoff as well as animal waste used as feed.

The absence of some bacteria in African catfish farms in one region versus the other, namely, *Flavobacterium* spp, *Streptococcus* spp, *Chryseobacterium indoligenes*, *Proteus* spp, *Shigella dynteriae* and *Staphylococcus aureus* was surprising since both regions had similar fish species, production systems (earthen and concrete ponds) and feed sources (commercial, on-farm made and

feed scraps). Plausible reasons for this observation could be differences in the source of pond water, personnel hygiene, ponds management, and human and animal activities surrounding the locations sampled in this study.

In this present study, *Aeromonas sobria* was the most prevalent bacterial species found in fish samples in both Arusha and Morogoro. The results of the present study are higher than those previously reported in farmed tilapia in Southern highland and Northern Tanzania (24.6%) (Mzula *et al.,* 2019) and Egypt (25%) (El Deen *et al.,* 2014). Farmers in Southern highland and Northern Tanzania previously reported outbreaks of aeromonads diseases in the hot season (Mzula *et al.,* 2019). Aeromonads diseases outbreak has been shown to occur mainly in the summer (Ibrahem *et al.,* 2008), and the most reported in Tanzania occurred in 2009 on wild tilapia at the Mtera hydroelectric power dam (*Oreochromis niloticus*) (Shayo *et al.,* 2012). Aeromonads have also been reported to cause a high mortality rate in farmed fish elsewhere (Sreedharan *et al.,* 2012; Hemstreet, 2010; Nielsen *et al.,* 2001; Chen & Lu, 1991). Another species of this genera, in particular *Aeromonas hydrophila*, the causative agent of aeromonad septicemia of fish, has previously been detected and isolated in apparently healthy fish in Nigeria (Omeje & Chukwu, 2014) suggesting farmers need to improve their pond management system to minimize the risk of *Aeromonas* outbreak.

Edwardsiella tarda, a highly pathogenic fish bacteria (Joh *et al.,* 2013) was the second most prevalent fish bacteria identified in the fish samples in both Arusha and Morogoro study sites. The occurrence of *Edwardsiella tarda* pathogens has previously been reported in farmed Nile tilapia and African catfish in Morogoro (Emil, 2017) and wild fish in Uganda (Wamala *et al.,* 2018). *Edwardsiella* infection in farmed fish can occur through the introduction of contaminated faecal matter, feed or water into the ponds. Some of the other isolated bacteria in this study have been reported to have caused diseases to humans including *Plesiomomas shigelloides* (Chen *et al.,* 2013), *Chryseobacterium* spp (Calderón *et al.,* 2011) and *Comamonas testosteroni* (Bayhan *et al.,* 2013).

In this study, the prevalence of human bacteria pathogens in fish samples collected in both regions was also high, especially for *Pseudomonas aeruginosa*, *Vibro cholera, Enterococcus faecalis* and *Escherichia coli.* Lack of biosecurity measures and poor pond management practices during the entire production process may be ascribed to a possible source of contamination within the sites sampled in this study. Interestingly, *Staphylococcus aureus*, another human bacteria pathogen, did not occur on any of the fish samples collected from the 65 farms/locations studied within Arusha, despite the bacteria being present in 2 of the 54 Nile tilapia farms sampled. The observed occurrence

of this pathogen at the farm level could have been from the type of pond used since it was isolated from one concrete pond used by the farmers.

Farmers in both regions mostly used concrete ponds for fish production activities. About 95% of all the 19 identified bacteria in this study occurred in concrete ponds. The trend of pathogen occurrence in either earthen or concrete pond type showed that *Aeromonas sobria* and *Edwardsiella tarda* were still the most prevalent among the fish bacteria, followed by *Serratia marcescens* and *Plesiomonas shigelloides* which have both been associated with pathogenicity in fish (Baya *et al.,* 1992; Cruz *et al.,* 1986).

Indeed, pond structure and location have a significant impact on fish exposure to pathogens, especially from human activities. Poorly constructed ponds made the exchange of water difficult. Of the human bacteria pathogens identified from the ponds, *Vibrio cholera, Pseudomonas aeruginosa,* and *Enterococcus faecalis* were the most prevalent in both regions. This knowledge is particularly essential in understanding disease etiologies to ensure a proper response to clinical diseases fish farmers might encounter.

Equally important is the water quality in which the fish live. Known pathogens, including *Aeromonas sobria* and *Edwardsiella tarda* were frequently isolated and identified in pond water from multiple locations in Arusha and Morogoro. This could be ascribed to the intensive fish farming practices used by these farmers, low water quality, and high organic matter characteristics of aquaculture systems especially earthen ponds which had a comparatively higher presence of these two bacteria in this study.

Mathew *et al.* (2014) and Emil (2017) reported that water scarcity in some districts of Morogoro led to the deterioration of the quality of pond water, subjecting farmed fish to opportunistic infection such as *Edwardsiella tarda* and those of the genus *Pseudomonas* (Park *et al.,* 2012)*. Edwardsiella tarda* infections can be worsened by several environmental factors including temperature (Park *et al.,* 2012). At the time of sample collection in this study, Arusha and Morogoro pond water temperatures ranged between 21 to 27 ºC and 24.3 to 28 °C, respectively. These were within range for farming tropical fish (Bhatnagar & Devi, 2013) but also adequate for the growth of most pathogenic bacteria often reported in fish (Noga, 2010).

Overall, the observed high prevalence and diversity of bacteria in pond water samples in both regions could be due to direct contamination of pond waters by bacteria from surrounding soils, especially in earthen ponds, human activities around the farms as well as from various feed sources and personnel pond management methods. Emil (2017) previously reported that several fish farmers

in Morogoro were using streams as a source of their water without first disinfecting it, while pests, vehicles, and people traffic into the ponds were not controlled.

The presence of human bacteria pathogens such as *Pseudomonas aeruginosa* and *Enterococcus faecalis* isolated from pond water in every location studied in Arusha should be concerning due to the life-threatening complications they can cause to humans. The only pathogen that was not found in pond water in any location studied in Arusha was *Burkholderia cepacia*, suggesting that its presence in fish samples from this region must have been contamination from earthen ponds found only within Maweni and Manyata, since it was never identified in any fish feed samples analyzed. In general, sampled sites in Arusha and Morogoro had at least nine or more of the 19 bacteria species identified in this study, indicating this high diversity could be from a variety of sources including high stocking densities characteristic of pond aquaculture as well as contamination from humans, surrounding soils, runoff water, and feed, etc.

Some farmers admitted to using animal manure to fertilize their ponds. Animal waste in fish feeding, though nutritionally and cost beneficial in intensive aquaculture (Elsaidy *et al.,* 2015), has been shown to introduce pathogenic bacteria into aquaculture environment (Venglovsky *et al.,* 2016). For example, coliforms such as *Escherichia coli* are not the normal flora in fish and are mostly associated with faecal contamination. The type of manure used to fertilize fish ponds varied between farms and depended on the animal species reared by the farmer.

Of course, fish farmers in Tanzania reportedly use other feeding methods, especially on-farm made and food scraps to supplement feeds, because of unavailability and high cost of commercial feeds (Kaliba *et al.,* 2006, Mathew *et al.,* 2014). As such, proper hygiene is essential in handling locally sourced feeds to minimize the negative impacts on fish safety and consumer health. Indeed, the absence of bacteria in all the commercial feed samples collected from fish farmers in both regions suggests important Good Aquaculture Management Practices (GAMP) were followed during their manufacture.

However, a variety of pathogenic bacteria were isolated and identified from on-farm made and food scrap feed sources sampled in this study and could be attributed to personnel hygiene and cross-contamination during handling, particularly the relatively high occurrence of human bacterial pathogens such as *Vibrio cholera* and *Escherichia coli.*

Additionally, the absence of *Aeromonas sobria* in the feed samples collected in all 130 locations and feeds used in this study suggest that the observed occurrence of this virulent species in the fish, pond water and pond type could be attributed to other sources such as soil and surface runoff, among

others. There were no biosecurity principles practised in any of the farms visited in this study exacerbating the risk of transmission of the identified pathogens from pond to pond, especially given that fishing nets were commonly shared between ponds.

Finally, this current study provides knowledge regarding prevalent etiological agents in fish and fish farms within the Arusha and Morogoro regions of Tanzania. Some of the isolated bacterial pathogens in this study have been reported to have caused serious disease elsewhere, including in farm-cultured eels in Korea (Joh *et al.,* 2013) and tilapia in Trinidad (Newaj-Fyzul *et al.,* 2008). When pathogens are known and properly profiled, control of disease in a susceptible population can, therefore, be correctly managed.

4.2.4 Bioaccumulation and Distribution Pattern of Heavy Metals in Aquaculture Systems

Fish farmers in Arusha and Morogoro regions use earthen or concrete ponds for their aquaculture production. In this study, we report for the first time the heavy metal contamination of sediments in ponds used for Nile tilapia and African catfish production in Tanzania. Different levels of As, Pb, Hg, Cd, and Cr were seen amongst the villages studied. Most notably, the results showed pond sediment samples from Morogoro region had significantly higher Cr concentrations (6.10 – 9.28 mg/kg) compared to those in Arusha (2.65 – 5.56 mg/kg). As previously revealed by other authors (Mdegela *et al*., 2009), the observed variation could be caused by the differences in anthropogenic activities surrounding the sites studied. For instance, Kihonda village in Morogoro region had the highest concentrations of Cr indicating their proximity to urban settlements and industrial activities likely contributed to the observed values.

Despite WHO providing maximum tolerable soil concentrations for heavy metals, none exists for sediments. As such, a sediment quality control regulation in aquatic environments varies considerably between countries (Australian New Zealand Food Authority [ANZFA], 2000). Unfortunately, Tanzania does not have its sediment quality guideline for heavy metals. However, since MacDonald *et al*. (2000), had proposed consensus-based sediment quality guidelines for freshwater ecosystems called the probable effect concentration (PEC) which reflects causal rather than correlative effects, we compared our heavy metal concentrations in sediment samples with the PEC guideline. The PEC values for As, Pb, Hg, Cd and Cr are 33.0, 128.0, 1.06, 4.98, and 111.0 mg/kg dry weight (MacDonald *et al*., 2000). In this study, it was report for the first time that the concentrations of the other heavy metals in the fish pond sediments sampled in Arusha and Morogoro were lower than the PEC values, except for Hg. Some authors reported that heavy metal concentrations in fish pond sediments in Bangladesh decreased in the sequence of Cr>Pb>Cd (Das

et al., 2017), which is similar to mean results from Arusha region. The results also indicate that fish pond sediments in both regions contain heavy metals at levels that potentially could cause long-term contamination of fish. One source of heavy metals in the sediment is the unconsumed pellet feed that often settle at the bottom.

WHO's maximum tolerable soil concentrations for Pb, Hg and Cd are 84.0, 7.0 and 4.0 mg/kg, respectively (WHO, 2011). By comparison, sediments from river basins, lakes, water treatment dams and coastal beaches in Tanzania have shown heavy metal concentrations higher than the established limits (Kishe & Machiwa, 2003; Embedded and Ubiquitous Computing [EUC], 2006; Nziku, 2013). This might be ascribed to greater surface area per unit of mass of the fine particles in the sediments, clay minerals, and significantly larger volumes of water, which increases the adsorption capacity and solubility of the heavy metals (Kishe & Machiwa, 2003).

In this study, fish feed samples from all locations in the Morogoro region had undetectable levels of both As and Hg (below instrumental detection limits). Another study conducted in Missouri, USA, found that some commercial fish feed used in aquaculture and natural ponds contained As 1.81, Pb 9.16, Hg 0.07, Cd 2.37, and Cr 1.42 mg/kg diet, dry weight (wt.), which were within the acceptable limit. However, the results show Cr concentrations in fish feed samples collected from all sites studied were consistently higher than other metals studied, similar to previously reported results from commercial feeds used in farmed shark catfish (*Pangasius hypophthalmus*) in Bangladesh (Das *et al*., 2017). To the best of my knowledge, no information is available regarding heavy metal concentrations on commercial fish feeds currently sold to fish farmers in Tanzania. The results indicate that fish farmers from all sites studied except Maweni and Samalia used commercial fish feeds with higher than the maximum Cr consumption level permitted in feeds (5.0 mg/kg) by the European Union (Javed & Usmani, 2016). This increases the likelihood for Cr to bioaccumulate faster in fish tissue, posing a health risk to consumers. This can likely happen in Tanzania since some commercial fish feed used by fish farmers in Bangladesh were revealed recently to contain higher levels of heavy metals that exceeded WHO's limits for food safety (Tacon *et al*., 2009). Therefore, the type and source of the raw ingredients used in the feed formulations influence feed quality. Crops, especially those grown in polluted soils or irrigation water, urban and peri-urban areas, are proficient in accruing high heavy metals and can contaminate fish feeds when used as input in the formulations (Kumar *et al*., 2017).

Fish is a better material for detecting heavy metals contaminating aquaculture ecosystems because fish can accumulate these compounds directly from water and through the feeds (Van *et al*., 2003). This is critical because muscles contribute to the greatest mass of the flesh that humans consume.

Concentrations of heavy metals, As, Pb, Hg, Cd, and Cr in muscle tissues of Nile tilapia and African catfish, two species highly consumed in Tanzania, are summarized in Table 3 and compared with the recommended daily dietary allowances (ANZFA, 2000). According to these data, in Morogoro region, the ranking order of the mean concentration of the heavy metals in fish muscles was Cr>Cd>Pb>Hg>As; while in Arusha, the trend was Cr>Cd>As>Pb>Hg (Table 3).

In this study, the highest concentration of Cr in fish muscle was found in Mikese in Morogoro (9.50 ± 0.98 mg/kg dry weight), equating to 0.210 mg of Cr per day considering the daily fish consumption rate for a 60 kg Tanzanian adult is 22.1 g on average (Ullah *et al.*, 2017). Thus, the level is similar to the recommended daily dietary allowances of 0.20 mg per day per person and doesn't constitute a significant threat to fish consumers (RDA, 1989). Samalia in Arusha had the least Cr concentrations (2.53 ± 0.65 mg/kg dry weight), equating to 0.056 mg of Cr per day. The FAO and WHO permissible limit of Cr in freshwater fish is 0.05 µg/g (Al-Busaidi *et al.*, 2011). The levels of Cr in fish muscle from this study were similar to those reported in Bangladesh in freshwater fishes from Dhaleshwari River (6.92 – 12.23 mg/kg dry weight), Buriganga River (5.27 – 7.38 mg/kg dry weight), and ponds in Noakhali districts (3.21 mg/kg). Long-term chronic exposure to Cr may cause respiratory, gastrointestinal, liver, kidney and cardiovascular problems as well as disruption of disrupt cellular integrity and functions (Ahmed *et al.*, 2009; Ahmed *et al.*, 2010; WHO, 2011).

The Cd has no known biochemical benefits to humans. Long-term dietary exposure to Cd from marine fish and other mammals may cause kidney dysfunction, liver tumor, reproductive problems, prostate and ovarian cancers, as well as endocrine disruption and oxidative damage, pulmonary dysfunction, and kidney problems (Underwater Electric Potential [UEP], 1997; Duruibe *et al.*, 2007; Javed & Usmani, 2016; Renieri *et al.*, 2017). The highest Cd content (3.97 ± 0.56 mg/kg) in fish muscles was found in Mikese, Morogoro, equating to 0.088 mg Cd per day from fish for a 60 kg Tanzanian adult consuming 22.11 g fish per day on average (Ullah *et al.,* 2017), which is slightly higher than the recommended daily dietary allowances of 0.06 mg per day per person (Redundancy Analysis [RDA], 1989), increasing the risk to human health. The lowest concentration was 1.38 ± 0.44 mg/kg in Nambala, Arusha, equating to 0.030 mg Cd per day for a Tanzanian adult. The EU maximum tolerable limit for Cd in fish is 0.05 mg/kg (Young, 2005). The contamination of Cd was possibly due to industrial discharges and mining activities in the regions. The amount of Cd measured in other freshwater fish species in Bangladesh was also found to be above the EU maximum tolerable limit, especially from Dhaleshwari River, and was attributed to urbanization effects (WHO, 2011). However, compared to this results, a previous study in Morogoro's Mindu Dam found much lower Cd levels between 0.003 – 0.090 mg/kg wet weight in hairy river prawn

(*Macrobrachium rude*), African sharptooth catfish (*Clarias gariepinus*), and Wami tilapia (*Oreochromis urolepis*) and attributed its accumulation in tissues to anthropogenic sources and traffic emission (Mdegela *et al.*, 2009).

Consumption of food contaminated with Pb causes many adverse health effects including disruption of the cognitive development in children, mental retardation, and even death (EUC, 2006; Javed & Usmani, 2016). In this study, the maximum Pb level observed in the fish muscles was 3.44 ± 0.93 mg/kg in Tangeni, Morogoro, equating to 0.076 mg of Pb per day for a Tanzanian average fish consumer; while the minimum was 0.58 ± 0.21 mg/kg in Manyata, Arusha, equating to 0.013 mg Pb per day (Table 3). These levels were significantly lower than the recommended daily dietary allowances of 0.21 mg per day per person (RDA, 1989).

In other fishes consumed in Tanzania, the mean Pb content reported from Nile perch (*Lates niloticus*) muscles sampled in Mwanza, Kagera and Mara regions on the shores of Lake Victoria were 0.36 ± 0.15, 0.41 ± 0.12 and 0.28 ± 0.11 mg/kg, dry weight basis, respectively (Hassanien & Shahawy, 2011). These equate to between 0.006 – 0.009 mg of Pb per day per person for a Tanzanian average fish consumer. Additionally, the levels of Pb in Nile perch sampled from five major fish processing factories at the shores of Lake Victoria in Mwanza and Musoma reportedly ranged from < 0.01 – 0.08 µg/g ww (Hassanien & Shahawy, 2011). Levels of Pb in the muscles of African sharptooth catfish (*Clarias gariepinus*), Lake Rukwa tilapia (*Oreochromis rukwaensis*) and Singida tilapia (*Oreochromis esculentus*) from Tanzania's Lake Rukwa were found to range between 0.01 to 1.9 µg/g, 0.12 to 0.88 µg/g and 0.02 to 1.4 µg/g, respectively, and were below WHO permissible limits (Wu *et al.*, 2013). However, high concentrations of Pb were detected in muscles from wild and farmed milkfish (*Chanos chanos*) and wild mullet (*Mugil cephalus*) from Tanzania mainland (Mtwara), Zanzibar islands (Pemba and Unguja) and the Indian Ocean, which exceeded maximum levels set by FAO/WHO of 0.3 mg/kg ww (Mwakalapa *et al.*, 2019).

The Hg is ubiquitous in nature and is present in several forms. Methyl mercury can build up in certain fish at levels greater than those in the surrounding water (Machiwa, 2005). Chronic exposure to Hg may cause toxicity of the gastrointestinal, renal and nervous systems, loss of vision, hearing, mental retardation, and death to humans (Sarasiab *et al.*, 2014). In this study, the minimum and maximum Hg contents were found as 0.95 ± 0.04 mg/kg in Manyata and 0.35 ± 0.13 mg/kg in Kikwe, respectively, both from Arusha region, equating to 0.021 and 0.008 mg of Hg per day, respectively, for a Tanzanian average fish consumer. These levels were lower than the recommended daily dietary allowances of 0.03 mg of Hg per day per person (Tchounwou *et al.*, 2003). The EU maximum tolerable limit for Hg in fish samples is 1.0 mg/kg (Young, 2005). The mean Hg content

(0.60 – 0.65 mg/kg) reported in the fish muscles studied in Morogoro and Arusha regions were lower than the EU maximum tolerable limit for Hg. Previous studies in Tanzania found mean Hg concentrations from Nile perch (*Lates niloticus*) muscles as 0.23 ± 0.15, 0.08 ± 0.04, and 0.09 ± 0.03 µg/g, dry weight basis, for Mwanza, Kagera, and Mara, respectively (Hassanien & Shahawy, 2011). In contrast, Mshana found that concentrations of Hg in the muscles of African sharptooth catfish (*Clarias gariepinus*) and Singida tilapia (*Oreochromis esculentus*) from Tanzania's Lake Rukwa varied between 0.03 to 0.33 µg/g and <0.01 to 0.29 µg/g, respectively, and were above WHO permissible limits of 0.14 µg/g , indicating that the fishes were not safe for human consumption (Al-Busaidi *et al*., 2011).

The chronic build-up of As severely affect different body organs including the CNS, kidneys, gastrointestinal tract, heart, skin, and lungs (Sarasiab *et al*., 2014). In this study, the minimum and maximum As contents were found as 0.43 ± 0.26 mg/kg in Tangeni, Morogoro and 1.24 ± 0.40 mg/kg in Samalia, Arusha, respectively, equating to 0.027 and 0.010 mg of As per day, respectively, for a Tanzanian average fish consumer, and were below both the recommended daily dietary allowances of 0.13 mg of As per day per person and the WHO maximum tolerable daily intake of 0.05 mg/kg body weight per day (RDA, 1989). Comparatively higher levels of As were found in Arusha locations compared to Morogoro. There is no maximum permissible limit of As in fish sample set by the EU. Compared to our results, higher levels of As in different freshwater fish species have been reported in the literature and were in the range of 1.97 – 6.24 mg/kg dry weight in some edible fishes from Bangshi River at Savar in Bangladesh and 0.091 – 0.53 mg/kg wet weight in some commonly consumed fish species from urban rivers around Dhaka city, Bangladesh (Rahman *et al*., 2012; Islam *et al*., 2015). It's been reported that As contamination originates from both natural and anthropogenic processes (Saha *et al*., 2016). Increased urbanization and fast industrial development in Arusha region likely contributed to rapid pollution growth. Studies indicate a high accumulation of As in most fish tissues occurred in polluted areas (Jankong *et al*., 2007). Some studies also show that freshwater fish can convert inorganic Arsenic to organic Arsenic (Šlejkovec *et al*., 2004).

The results revealed that Cr contributed the highest estimated daily intake (EDI) of heavy metals in fish muscles from both Arusha and Morogoro. All the EDI values were significantly lower than their respective recommended daily dietary allowances, confirming that the fish species studied are safe for human consumption. The results agreed with those of Ullah *et al*. (2017) who investigated health risk implications in Bangladesh from dietary intake of heavy metals in cultured fish. In this study, the least EDI was found in Hg and As from Arusha and Morogoro, respectively.

The target hazard quotient (THQ) was used to estimate the potential non-carcinogenic risk of the heavy metals to consumers of Nile tilapia and African catfish in Tanzania. Average heavy metal concentration from each of the regions was used in calculating THQ for the residents of Arusha and Morogoro. In this study, the THQ for each of the metals was less than 1 in all locations, except for Cd in Morogoro region, suggesting that consumers of fish farmed in Arusha and Morogoro would not experience significant health risks if they only ingest individual heavy metal through fish. In Morogoro, the THQ values for the targeted heavy metal followed the descending order of Cr>Cd>Hg>Pb>As; while that of Arusha followed Cr>Cd>Hg>As>Pb. In comparison, Mhina reported that THQ values from Nile perch in Mwanza, Kagera and Mara regions of Lake Victoria followed the descending order of Pb>Cd>Hg (Mhina, 2016). Elsewhere, the THQ values reported in commonly consumed fish in Bangladesh followed the descending order of As>Pb>Hg>Cd>Cr (Ahmed *et al*., 2015). whereas some imported species followed As>Cd>Pb>Cr, indicating the influence of species and geographical location on heavy metal bioaccumulation in fish (Wang *et al*., 2005).

Exposure to two or more heavy metals may result in cumulative and/or interactive effects and is expressed as HI (total THQ) (RDA, 1989). In this study, HI values from fish sampled from Arusha and Morogoro were significantly greater than 1, suggesting exposure to a mixture of the five examined metals through consumption of the fish presents a significant non-carcinogenic risk to human health. It means that the possibility of health risk associated with the non-carcinogenic effect is significantly high for the continuous consumption of the farmed fishes studied. In contrast, Mhina (2016) found that THQ and HI of Hg, Cd and Pb ingestion in Nile perch muscles from Mwanza, Kagera and Mara regions of Lake Victoria was less than 1, suggesting no non-carcinogenic health risk from ingestion of Hg, Cd, and Pb individually and collectively through the Nile perch consumption in these areas (Mhina, 2016). In this study, however, the major risk contributor was Cr with 50.42 and 39.20% of the total HI values, in Morogoro and Arusha, respectively. The HI values greater than 1 was also reported in eight cultured fish consumed in Bangladesh and the major risk contributor was As with 80.66% (Wang *et al*., 2005).

Figure 7c shows the carcinogenic risk (CR) for As, Pb, Cd, and Cr due to the consumption of Nile tilapia and African catfish from the Arusha and Morogoro regions of Tanzania. The CR values lying between 10^{-6} and 10^{-4} are considered acceptable while those above 10^{-4} are unacceptable (WHO, 1976). In this study, the estimated CR values for the five heavy metals were lower than 10^4, indicating a low risk of cancer due to their exposure through fish consumption from the studied regions. The CR values for heavy metals higher than the unacceptable range have been reported in

Bangladesh, indicating a greater risk of cancer in marine fishes consumed in that country (Wang *et al*., 2005).

4.2.5 Design of a Context-specific Climate-smart Sustainable Fish Pond

Many farmers in Tanzania are finding that fish production is profitable. However, the aquaculture sector in the country faces increasing constraints as competition for the available freshwater resources for agricultural and reactional activities intensifies. Therefore, freshwater fish farming in the country need production systems that are more resilient to climate-change impacts such as droughts and floods. The success of pond aquaculture to meet climate-change challenges is largely dependent on the design and management of the fish pond. In Tanzania, conventional, small-sized excavated earthen or concrete ponds are the most commonly practiced form of aquaculture system (MLFD, 2015). These ponds are especially susceptible to flooding and overgrown vegetation which predisposes farmed fish to diseases, bacteria, parasites, heavy metal contamination, and predation. Flooding may also introduce pollutants from sewage and surface runoffs into the ponds, thereby reducing dissolved oxygen levels and destroying the fish as a result of harmful algal bloom during rainy seasons. On the other hand, drought may reduce availability or reliability of freshwater sources such as spring rivers.

Therefore, to reduce vulnerability to long-term climate change impacts, the aquaculture sector needs better freshwater management. A viable approach for Tanzania would be to invest in context-specific climate-smart pond design that: (a) improves efficacy of freshwater usage, (b) reduces watershed pollution caused by aquaculture effluent discharge, (c) has resilience to climate-change variability, (d) increases aquacultural productivity, and (e) combines a high level of biosecurity with low risk of disease and external contamination. In this study, a climate-smart pond was designed and constructed using locally available materials with the capacity to boost resilience and potentially increase fish production with less water and land through improved freshwater utilization. Two special features of the design included a solar-powered water pump and a filtration system that ensured water can be stripped of organic load through a strata of cotton, activated charcoal, gravel and fine sand, then re-oxygenated through turbulence while being recycled back into the pond. Solar energy is abundant, clean and renewable, thus helps reduce the environmental impact of aquaculture by eliminating carbon development. The climate-smart ponds had a polyethylene membrane (pond liner) to hold the water and prevent seeping losses and was constructed on an open field area to allow for both direct sunlight and rainfall. The depth of the climate-smart ponds was 1.5 m above ground to prevent influx of water from floods or surface runoffs during the wet months.

Water quality parameters were measured of the climate-smart pond with the filtration capability and compared them values to those from a climate-smart pond without a filter and a conventional concrete control pond. For the six-month duration of this study, fish stocked in all the 3 ponds studied were fed the same feed formulation; however, the farmer followed his normal aquaculture practices to manage the fish culture in the concrete pond. Water quality parameters have profound effects on pond productivity, fish health and oxygen availability.

Water temperature is one of the most important factors for pond aquaculture (Eze & Ogbaran, 2010). Pond water temperature normally follows that of the prevailing climate, and in a large water body, can vary throughout the fish pond. The pond liner used in this study was black, which could theoritically absorb and retain heat, hence lead to substancial warming up of the water that affect fish metabolic rate and/or evaporation loses. Moreover, high water temperature increases toxicity of ammonia and decreases dissolved oxygen levels (Meck, 2000), and in extreme cases may lead to fish mortality (Eze & Ogbaran, 2010). The ideal water temperature considered optimum for Nile Tilapia production is about 25 to 27 °C (Kausar & Salim, 2006). The mean pond water temperature reported in this study ranged from 27.2 to 27.4 °C, which was ideal for the fish. The mean temperature previously reported in over a dozen fish ponds located in Morogoro region was 26 ± 3.1 °C (Mdegela *et al.,* 2011), which is consistent with our current findings.

In pond aquaculture, abiotic factors such as pH, nitrate, ammonia, dissolved oxygen (DO), biological oxygen demand (BOD), alkalinity, turbidity and hardness should be monitored to ensure proper fish health (Bhatnagar & Devi, 2013). The design of the climate-smart pond with filtration mechanism used in this study made it possible to manage levels of these critical aquaculture water quality parameters. The water samples tested in this study were collected during mid-morning time. Compared to the climate-smart pond without a filter, the one with a filter had on average 2.4 times less nitrate, 2.5 times less ammonia, 1.3 times more dissolved oxygen, 2.8 times less biodegradable organic matter, 2 times less alkalinity, 3.6 times less turbidity and 1.1 times less hardness levels. When compared to the conventional concrete pond, the climate-smart pond with a filter had on average 9.2 times less nitrate, 8 times less ammonia, 1.8 times more dissolved oxygen, 6.9 times less biodegradable organic matter, 2.5 times less alkalinity, 9.7 times less turbidity, and 2.5 times less hardness levels. These results show the importance of frequency of water change, pond cleanliness and good aquaculture practices, thus validating the filtration system as essential for good pond life and overall pond hygiene management.

It was observed that some challenges with the climate-smart pond without the filter. First, the system couldn't be drained completely prior to replacing the water. Second, some unconsumed feed and

fish waste remained at the bottom of the pond, which likely contributed to decreased DO, elevated BOD levels, high nitrate, and increased ammonia compared to the pond with the filtration mechanism. Decreased DO may lead to poor feeding and reduced growth in pond aquaculture (Bhatnagar & Garg, 2000) while high BOD levels in pond water might be harmful for aquatic life (Mukherjee & Dutta, 2016). Mdegela *et al.* (2011) reported ammonia levels of 1.0 ppm in water samples from multiple earthen and concrete fish ponds within Morogoro region, which understandably were relatively higher than results from climate-smart ponds studied in the present research. However, Mdegela *et al.* (2011) reported extremely low nitrate concentrations (0.20 ppm), which is in contrast to the values reported in the conventional concrete pond studied in this experiment conducted in the same region. And third, adding river water every two weeks to the climate-smart pond without filter compromised biosecurity since it had the potential of introducing parasitic and bacterial pathogens to the fish.

Home-made feed formulation comprising of a mixture of plant ingredients and insect-protein was used in this study. The recorded weight and length measurements of fish in each pond structure showed growth performance over the six-month study period. Fish feeding was based on fish weight; the amount of feed given to the fish was adjusted every two weeks in the climate-smart ponds, while the farmer who kept no records of fish measurements, had limited knowledge of proper feeding regimen for optimal pond productivity. On average, fish reared in climate-smart ponds had better weight gain and length compared to those in conventional concrete pond.

For the rural poor in Tanzania, capital is a major constraint to constructing and operating a climate-smart fish pond. The design of climate-smart ponds is economically feasible and appropriate for small-scale house-holds in rural communities in the country. Energy is the most cost-prohibitive item for operating the climate-smart pond filtration system. However, the installation of 12-Watt solar-powered water pump reduces the energy demand of the system, thus energy costs for the filtration system is eliminated. The solar-pump used is both durable and sustainable since sunlight is abundant in Tanzania throughout the year. In terms of solving for gender inequity, the climate-smart pond was designed in a way that's simple to manage for grassroots farmers including women who are seeking to engage in small-scale fish farming with minimal expenses for subsistence and economic sustainability.

4.2.6 Fish Feed Formulation

This is the first study to utilize Google Sheets Program in feed formulation in order to investigate the growth performance and nutritive value of locally made fish feed in Tanzania using a

combination of spirulina, moringa leaf powder and black soldier fly larva (BSFL) meal as complete replacement for fish meal. The BSFL was prepared from unwanted kitchen matter such as food waste, rotting fruits and vegetable remnants since natural populations of *H. illucens* are adapted to decompose decaying organic materials (Spranghers *et al.,* 2017). This study was conducted in Morogoro from , all water quality parameters recorded from the climate-smart pond used in this study were within optimal ranges for Nile tilapia production (Djissou *et al.,* 2017). Except for BSFL meal, all the ingredients used could be sourced from places such as flour mills, supermarket stores and municipal markets, and have been reported as major feed supplement for cultured fish in Tanzania (Mmanda *et al.,* 2020).

Using Google Sheets Program, the experimental BSFL diet was formulated to provide 35 g protein per 100 g of feed, while considering the essential amino acid (EAA) requirement for Nile tilapia. There are large discrepancies between the reported requirement for lysine, tryptophan and methionine in cultured Nile tilapia (National Research Council [NRC], 1993; El-Sayed, 2004). However, our findings indicate that black soldier fly larva meal, *Moringa oleifera* leaf powder and spirulina have the potential to supply the EAA required for Nile tilapia production. Médale and Kaushik (2009) reported that EAA levels in the diet must be adequate to reduce nitrogenized catabolism and poor diet conversion. In thus study, I confirm that Google Sheets Program enables feed formulation that takes into account protein digestibility in order to obtain good growth performances and feed utilization. Furthermore, this results corroborate the study by Zhao *et al.* (2010) who showed that it's possible to predict growth performance of animals fed with food formulated with more precison.

The crude protein content (34.19 ± 0.37 g/100g) for the experimental BSFL diet was also within the range (25 – 35% CP) required for proper growth of Nile tilapia (Abdel-Tawwab *et al.,* 2010) but lower than the CP content required (40% CP) for maximum growth rate (Wang *et al.*, 2005). The BSFL diet was processed into dry pellets with a final moisture content of 10% and was nutritionally balanced, water stable, with the proper size and texture. Based on Google Sheets Program output, the BSFL diet had crude lipid and caloric content of 15.92% and 15.56 kJ/g, respectively. Basil powder was added at a rate of 1.2% of the formula to increase palatability and provide antimicrobial property. Antibacterial herbal ingredients prevent the growth of microorganisms such as bacteria, fungi and protozoa that cause fish disease and mortality (Kingston, 2008).

In contrast, the commercial feed had on average crude protein (22.65 ± 0.71 g/100 g), crude lipids content (10.97 ± 0.14 g/100 g) and ash (12.73 ± 0.47 g/100 g), which were significantly lower than for the experimental BSFL diet. This variation between experimental and control feeds can be

attributed to differences in ingredients and their amounts in the respective feed recipes. The results agree with Mmanda *et al.* (2020) finding that commercial fish feeds sold in Tanzania had crude protein content ranging from 22.0 to 55.0 g/100 g and ash content of 11.0 to 26.0 g/100 g. The high crude fiber content (15.2 g/100 g) in the experimental BSFL diet reported through the Google Sheets Program compared to proximate chemical analysis value (5.88 ± 0.10 g/100 g) could be an artifact related to data entry in which some ingredients nutrition values are available only in fresh basis while others are provided in dry matter basis. This can further be explained by the lack of significant (P<0.05) difference in crude fiber levels obtained after chemical analysis in the control commercial diet compared to the experimental BSFL diet. Lastly, this is the first time fish feeds have been formulated from local feed ingredients having the potential to supply the EAA required for proper fish growth and health using Google Sheets Program.

Other than BSFL meal, the chemical composition of BSFL diet ingredients was populated into the Google Sheets Program from the USDA National Nutrient Database for Standard Reference using Nutrasheets[TM] add-on designed to custom batches and compute nutrition facts and formulation costs. Nutrition values for individual feed ingredients were generally within the range reported in Tanzania by other researchers (Mutayoba *et al.,* 2011; Munguti *et al.,* 2012; Madalla *et al.,* 2013). Optimization of dietary protein is necessary since it directly affects the cost of feed and nitrogen loading in the culture system. The calculated cost of the experimental BSFL diet was $0.35 per kg of feed. Comparatively, the cost of the commercial feed used in this study was high (1.55 $/kg of feed). Generally, the cost of commercial aquafeeds in Tanzania is high (Mmanda *et al.,* 2020). High feed cost often lead to elevated production costs that's increasingly uneconomical to rural small-scale fish farmers given low returns (Mmanda *et al.,* 2020). Therefore, fish diets formulated entirely from local feed ingredients can improve farm productivity and lower production expenses.

Based on observation, experimental BSFL diet was well accepted by the fish since tilapia rapidly and repeatedly swam to consume the feed throughout the duration of study. In terms of growth performance, fish fed with experimental BSFL diet and commercial feed grew to a mean length of 10.9 ± 1.54 cm and 8.5 ± 1.12 cm, respectively, during the 90-day study period; while their mean weight during the same duration was 188 ± 20.11 g and 111 ± 17.20 g, respectively. The good overall growth performances obtained with experimental BSFL diet used in this study confirm the suitability of the formulation and its nutrient composition for Nile tilapia production. From the result, formulating and feeding fish with a diet having crude protein content of 34.19 ± 0.37 g/100 g confirm the work of Médale *et al.* (2013) who reported that fish of lower trophic level such as tilapia and carp should be fed with diet containing 30% protein to ensure optimal production performance.

Djissou *et al.* (2016) observed similar results when a mixture of leaf maggots, *Azolla filiculoides* and *Dialum guineense* leaf powders were used as complete replacement for fishmeal in the diet of *Oreochromis niloticus*. The observed difference in length and weight gain in this study could be due to the nature of the diets and the digestibility of the two feeds, perhaps driven by processing methods.

High feed intake in fish fed experimental BSFL diet suggest better palatability, which helps improve dietary nutrient intake and increased growth performance. However, plants often contain compounds that can negatively impact appetite, digestion, nutrient absorption and metabolism (Francis *et al.,* 2001). Generally, fish has high digestibility (>90%) of proteins from vegetable origin (NRC, 2011), but anti-nutritional factors such as fibres and tannins (Burel & Médale, 2014) can limit bioavailability of some amino acids (Cai & Burtle, 1996). For example, the leaves of *Moringa oleifera* contain tannins known to hinder protein digestibility (Richter *et al.,* 2003), which could explain the low protein intake reported in fish fed with experimental BSFL diet used in this study. In fact, tannins have been shown to reduce growth performance in fish of the genus Tilapia and *Labeo rohita* (Jackson *et al.,* 1982).

The results suggest that replacement of fishmeal using BSFL meal may be limited to 53% inclusion without further amendments to BSFL meal through the addition of moringa leaf powder and spirulina. In fact, key growth metrics such as fish length, fish weight, protein efficiency ratio and SGR% were all higher in fish fed with experimental BSFL diet (Table 4) indicating the ratio of spirulina:*Moringa oleifera* leaf powder:black soldier fly larva (BSFL) meal for fish meal replacement established in this study suited tilapia production. Improvements in tilapia performance at higher levels of BSFL meal inclusion are possible given that as currently formulated in this study, the experimental BSFL diet containing only 52.8% BSFL meal already provides EAA concentrations that meet *Oreochromis niloticus* requirement.

The Feed conversion ratio [FCR] of fish fed with experimental BSFL diet was significantly (P<0.05) lower (14.4 ± 0.09) than that of fish fed commercial feed (17.9 ± 0.16). The protein intake in fish fed with experimental BSFL diet might account for a portion of the reduced FCR. Lower FCR values have been reported in shrimp reared in green-water systems (ponds or tanks) due to the continuous access to supplemental nutrients (Ye *et al.,* 2011; Bulbul *et al.,* 2013). Thus, our results indicate that it may have been better to overfeed the fish with experimental BSFL diet to increase access to food and thereby avoid artificially reducing growth rates. Other researchers have reported much lower FCR values (<2.0) in Nile tilapia fed with housefly maggot meal diets (Ogunji *et al.,* 2008).

By the end of the growth trial, fish fed with commercial diet had lower SGR and PER despite having high protein intake levels and significantly higher (18.92 ± 0.28 g) dietary protein to energy ratio (P/E ratio). This may suggest that the digestibility of the commercial feed might have been lower than for the experimental BSFL diet, indicating the advantage of using Google Sheets Program in formulating feeds to precisely meet growth requirements of fish. Appropriate protein levels in the diet is essential to ensure adequate growth of fish. The gross energy reported in the commercial feed used in this study was significantly ($P<0.05$) lower than in experimental BSFL diet. Further, the protein content in the commercial feed was found as lower than recommended for different life stages of Nile tilapia (Thongrod, 2007). Ogunji *et al.* (2008) reported the importance of maintaining a proper P/E ratio in the diet, confirming also that energy supply must be adequate so that dietary protein is not metabolized for energy. The P/E ratio of feeds used in this study are optimal for growth of Nile tilapia (Ogunji & Wirth, 2000). Overall, the results of this study indicate that fish fed with experimental BSFL diet with black soldier fly larva, *Moringa oleifera* and spirulina as the main protein sources had the best growth performance than the fish fed with commercial diet made from other plant-protein sources as a complete fish protein replacement.

CHAPTER FIVE

CONCLUSION AND RECOMMENDATIONS

5.1   Conclusion

This study revealed that in both regions, tilapia and catfish are the main fish species raised in an intensive monoculture system either in concrete or earthen ponds utilizing borehole or tap water resources. However, a significant setback for farmers to achieve sustainable development of fish farming is the lack of relevant aquaculture training and/or awareness of available innovations on fish farming. Therefore, based on this study, fish farming management is still underdeveloped considering their poor water quality management approaches, untreated wastewater discharge into the environment, lack of accurate disease diagnostic methods, limited knowledge on feed formulation, preparation and quality, and very importantly, limited understanding of the drugs and chemical agents used for prevention and control of diseases.

Fish farmers in both regions face many constraints and challenges, including diseases and high production costs driven by high feed cost. Diseases are a major concern for the sustainability and profitability of fish farming. However, in both regions, there was a general lack of responsible farming practices including poor record-keeping and non-adherence to recommended dosage of the antibacterial drugs, therapeutic chemicals, and disinfectants. Additional research is recommended in order to illuminate possible development of widespread resistance to commonly used drugs this could cause.

The farmers also mentioned that they faced weather-driven challenges such as occasional high water temperatures and seasonal flooding. These natural factors have understandably been huge production challenges, so I would recommend the farmers to adopt problem planning prevention methods and locally applicable strategies based on globally accepted principles to minimize losses.

This study has revealed the physicochemical characteristics of fish pond water and the prevalence of three endo- and four ecto-parasites, bacteria pathogens as well as heavy metal accumulat on farmed Nile tilapia and African catfish in the northern (Arusha) and eastern (Morogoro) regions of Tanzania. Based on my observation and results, farmers need to be informed on how key water quality parameters, pond management, feed quality, stocking density, pond type, source of water and frequency of water change, among others, affect pond productivity, parasite and bacteria infestation, disease outbreaks, fish quality and human health. As aquaculture matures in Tanzania, efforts should be placed on adherence to sustainable production practices.

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The climate-smart ponds designed in this study addressed a number of social concerns, including providing solution to food insecurity due to climate change vulnerability and gender inequality. It's affordable, environmentally friendly and has the potential to increase food fish production with less labour and land among the rural communities while improving freshwater utilization and minimizing aquaculture effluent discharge into the environment. Solar energy used to power the water pump is renewable, thus, this system is sustainable, making it more resilient to climate-change impacts by filtering and recycling the water. This technology can be adapted to diverse and changing climatic conditions around the country; therefore, it has the capacity to enhance socio-economic growth and food nutrition.

This study give insight into the use of Google Sheets Program in fish feed formulation using plant- and insect-based protein sources as possible alternatives for fishmeal replacement in aquaculture diets. The software enables quick and judicious control of multiple factors during feed formulation including protein digestibility of individual ingredients and recipe cost optimization while taking into consideration EAA requirement for fish growth. Compared to commercial aquafeed, results of this study show an excellent overall growth performance of tilapia fed with experimental BSFL diet which can help fish farmers reduce production cost and increase pond productivity. Also, the findings indicate that BSFL reared on kitchen wastes is a low-cost and environmentally-friendly high quality protein resource for fish production. However, it is important to determine the apparent digestibility coefficient of BSFL meal to correctly and completely compute its EAA contribution in the diet.

5.2   Recommendations

Aquaculture is a specialized industry. The commercial fish farming industry in Tanzania, like any other agricultural practice, is regulated by the government. To ensure fish farmers realize greater economic returns, the Ministry of Agriculture, Livestock and Fisheries should incentivize certification programs and conduct farmer education through regular agricultural extension services to help aquaculture continue to develop into an environmentally and socially responsible food production endeavour. Of course, gaining skills in fish farming techniques and having access to equipment for disease diagnosis will improve farmers' ability to solve some of the challenges they reportedly faced in their ponds. Additionally, the government should promote efforts to develop sustainable aquaculture operations. In this study, it was noticed that the majority of the farmers heavily relied on past experiences when administering drugs had limited knowledge regarding fish diets, and used outdated technologies to manage their ponds. I believe that substituting the traditional

top-down extension system with farmer-participatory knowledge system will greatly maximize farmers' access to quality innovations in aquaculture.

Due to the high prevalence of several parasites in these two most economically important fish species in Tanzania, a proper pond management practices, awareness in fish health and fish disease prevention at the community level is paramount important to minimize parasitic infection and improve aquaculture in Tanzania. The adoption of climate-smart pond structures is recommended as well as improvement to currently utilized earthen and concrete ponds to minimize and/or eliminate entry of floodwater.

Small- and large-scale fish farmers should adopt this technology to improve their aquaculture production efficiency and reduce losses from diseases, stress and often preventable external contamination. This study has shown the possibility of inclusion of maggot meal in the formulation of fish feed and that this meal can be used to supplement fishmeal to about 50% inclusion levels which gave the best growth performance from this study. The economic analysis also justifies the growth performance findings. Based on -these results, the use of maggot to supplement the costly fishmeal to about 50% inclusion levels is recommended to fish farmers and the feed industry.

Finally, strengthening collaborative research on aquaculture between government research institutions and academia should be encouraged for better fish health management in Tanzania. Of course, still more studies are required to explore the magnitude of the microbial infestation and heavy metal distribution in fish under aquaculture farming in other regions of the country and the safety of fish feed formulated since some plants such as moringa has reported to contain antinutritonal factors such as tannin.

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Appendix 1: Research Tool (Questionnaire)

Questionnaire: Investigation of the existence of aquaculture management practice in Tanzania

1: PARTICULARS

If family, the name of the owner of the ponds:	If associative, the name of the association:
The number of dependents :	Name of the president:
The number of beneficiaries:	The number of members of the association:

2. LOCATION

Location of ponds:	**Residence of fish farmers:**
District:	District:
Administrative post :	Administrative post:
Locality:	Locality:
Village:	Village:
Walking distance or time between home and the fish farm:	
Observation:	

3. DESCRIPTION OF FISH FARM AND MANAGEMENT PRACTICES

Aquaculture type ☐ Earth pond ☐ Cages ☐ Other (specify)	Dimension of the ponds/cages (m2):	Year of the start of the activity:	The initial number of juveniles:
Aquaculture practices ☐ Integrated ☐ Non-integrated ☐ Other (specify)			
Management system ☐ Extensive ☐ Semi- intensive Intensive			
Production practice ☐ Monoculture ☐ Poly culture other			
Farmed (s) specie (s): ☐ Tilapia ☐ catfish			

☐ Carp Other			
Feeding: ☐ Commercial feedstocks ☐ Food scraps ☐ Vegetable ☐ Manure ☐ Corn Bran ☐ Sorghum ☐ Millet ☐ Rice ☐ Other (specify)			
How often fish diseases occur ☐ Very often ☐ Really ☐ Never			
Chemicals use ☐ Antibiotics ☐ Pesticides ☐ Other (specify)		**Purposes** ☐ Disease control and prevention ☐ Growth promotor ☐ Water treatment ☐ Other (specify)	**Manufacturer details** ☐ Name ☐ Expiring date ☐ Intended use ☐ Mode of application
People involved in the care of fish: ☐ Family members ☐ Persons hired ☐ Members of the association ☐ Others		**Qualification** ☐ Trained ☐ Untrained with experience ☐ Untrained without experience	
Maintenance status of aquaculture species: ☐ Poor ☐ Good ☐ Acceptable			

Excellent			

4. PRODUCTION

Provision of fingerlings:	Number of harvest : Partial harvest_____ Total harvest_____	Quantity of harvest: Partial harvest_____ Total harvest_____	Approximate size or weight of fish:
Destination of fish produced: □ House hold consumption/association □ Sales for local market □ Export □ Sale of juveniles □ other			

5. TECHNICAL ASSISTANCE

During the opening of ponds/cages: □ Yes □ No	In monitoring ponds/cages: □ Yes □ No	When fish diseases occur □ Yes □ No		
Source of assistance: □ Veterinarian □ Extension officer □ other				
Frequency of assistance: □ Weekly □ Monthly □ Quarterly □ Semiannually □ Annually □ On-call				

6. ENVIRONMENTAL ASPECTS

how often is the water in fish pond/cage changed □ weekly □ monthly □ once before stocking □ never □ other	

How much water is the exchange each time? ☐ 10% ☐ 20% ☐ 50% ☐ 75% ☐ 100%	
Is water entering the ponds treated: ☐ Yes ☐ No	
Is water leaving the ponds treated: ☐ Yes ☐ No	
Does the water leave the ponds/cages used for vegetables/crops irrigation? ☐ Yes ☐ No	
The water in the ponds comes from: ☐ Tap ☐ Well ☐ River ☐ Lake ☐ Source ☐ Subsoil ☐ Estuary ☐ Sea ☐ other	

7. PROBLEMS FACED

☐ No problem ☐ Low technical assistance ☐ Lack of technical assistance ☐ Lack/Inadequate knowledge of aquaculture management practices ☐ Lack of alternatives to disease control	

Appendix 2: Consent Form

NELSON MANDELA AFRICAN INSTITUTION OF SCIENCE AND TECHNOLOGY (NM-AIST)

Office of the Deputy Vice Chancellor
Academic, Research & Innovation

Direct Line: +255 027 2970002
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Tengeru,
P. O. Box 447
Arusha.

PI's contacts; Email: nyametef@nm-aist.ac.tz; Mobile No. 0757204091

For more information you may contact the following;

Chairperson,

The Northern Tanzania Health Research Ethics Committee (KNCHREC),

P. O. Box 447, Arusha, Tanzania.

Informed Consent for the participants of the study on the development of a climate-smart aquaculture framework for nutritious and safer food in Tanzania

Introduction: Aquaculture has great potential to improve food and nutrition security in developing countries where more than 815 million chronically undernourished individuals reside. Fish is packed with protein, vitamins, and minerals with high biological value. It is also an excellent source of omega-3-fatty acids which is important for brain development and cognitive performance of young children and adolescents. In most developing countries, aquaculture is conducted on a small scale for the purpose of addressing family-level subsistence and livelihood needs. The trends indicate that the sector continues to intensify its system and practices. This kind of aquaculture practices is characterized by high stock density in a limited space, creating a stressful condition and increasing fish susceptibility to diseases. Fish farmers heavily use antibiotics, pesticides, disinfectants and parasiticides for disease prevention and treatment. The residue from these chemicals can end up in the human gut upon consumption of contaminated fish. Therefore, this study aimed at investigating types of management practices used by fish farmers in Tanzania and the effectiveness of fish health management practices, identify safety hazards and develop a climate-smart aquaculture framework

Procedure: If fish farmers consent to participate in this study, they will be requested to participate in answering questions from the questionnaire. Fish samples, fish feed samples and water samples

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will be taken for laboratory analysis to identify safety hazards such as heavy metals residue, antibiotics residue, pesticide residue and the presence of pathogens. The results of this study will be disclosed to you and will only be used for the purpose stated in this study.

Compensation: By participating in this study, questionnaires answering will be done free of charge and farmers will be compensated for samples that will be collected for laboratory analysis at the cost of 5000 Tsh per farmer. The results of the finding will be communicated to influential stakeholders so that appropriate measures can be taken. Non-participation will not be punished in any way whatsoever.

Risks and Precautions: sample collection which will be done are non-invasive, thus no pain and risks will be involved for the fish. However, we are requesting fish farmers to be patient during the period of study which is approximated to take about 4months which will sometimes be inconvenient and you will have to postpone your activities. Time to participate in this study will not be compensated, however, we will try to use very minimal time as possible.

Confidentiality: Any records relating to your participation will be strictly confidential. Your names will not be used in any reports from the study. The participation in this study is voluntary and you may withdraw from the study at any time without fear of any reprisals. You are free to ask any questions or any clarification after you have read and understood the consent form explained to you.
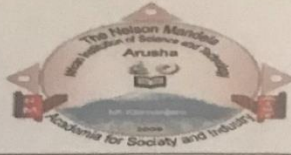
Participant statement

I…………………………………………… have understood the above information explained to me by the researcher and I agree to take part in this study and I can withdraw at any time without giving a reason.

Participants Name:…………………………………..Signature………………… Date…………..

Researcher's Name:…………………………………Signature …………………....Date…………..

Appendix 3: Ethical Clearance Certificate

Kibong'oto Infectious Diseases Hospital- Nelson Mandela African Institution of Science and Technology- Centre for Educational Development in Health, Arusha (KIDH-NM-AIST-CEDHA) -KNCHREC

RESEARCH ETHICAL CLEARANCE CERTIFICATE

Research Proposal No: KNCHREC00025   28th OCTOBER 2019

Study Title: DESIGNING A CONTEXT-SPECIFIC CLIMATE-SMART AQUACULTURE FRAMEWORK FOR SAFE AND NUTRITIOUS FOOD IN TANZANIA

Study Area: Arusha and Morogoro Region

PI Name: FRIDA ALBINUSI NYAMETE

Co-Investigator:

Institutions: NM-AIST School of Life Science and Bio-Engineering (LiSBE) of the Nelson Mandela African Institution of Science and Technology

The Proposal has been approved by KNCHREC on 18TH OCTOBER 2019

1. Subject to this approval you will be required to submit your progress report to the KNCHREC, National Institute for Medical Research, and Ministry of Health Community Development Gender Elderly and Children
2. Publication of your findings is subject to presentation to the KNCREC and NIMR Approval.
3. Copies of final publication should be made available to KNCHREC, National Institute of Medical Research and Ministry of Health Community Development Gender Elderly and Children.

Duration of Study Renewal: Subject to Renewal within ONE YEAR

Span From: 18th OCTOBER 2019 to 17TH OCTOBER 2020.

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Mr. Simon Njeya
Secretary
KNCHREC

Chairperson
KNCHREC

RESEARCH OUTPUTS

(i)   Publications

Frida, N., Musa, C., Titus, M., & Jofrey, R. (2020). Bioaccumulation and distribution pattern of heavy metals in aquaculture systems found in Arusha and Morogoro regions of Tanzania. *International Journal of Environmental Analytical Chemistry*, 2020, 1-19.

Frida, N, Musa, C., Titus, M., & Jofrey, R (2020). Prospects for aquaculture development in Africa in the context of a changing climate: A review. *International Journal of Biosciences,* 17(4), 1-13,

Frida, N., Musa, C., Titus, M., & Jofrey, R. (2020). Prevalence of fish parasites in Nile tilapia (Oreochromis niloticus) and African catfish (Clarias gariepinus) and physicochemical characteristics of pond water in Arusha and Morogoro, Tanzania. *International Journal of Biosciences,* 17(6), 76-91.

(ii)   Poster Presentation