

**EFFECTS OF STOCKING DENSITY ON GROWTH, WATER QUALITY
AND ECONOMIC PERFORMANCE OF MONOSEX NILE TILAPIA
(*Oreochromis niloticus*) REARED IN AQUAPONICS SYSTEM**

BY

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Declaration by the candidate

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DEDICATION

I dedicate this work to God Almighty for it is through Him that I have come so far. I also dedicate this thesis to my dear wife Sylvia, my daughter Shmneh and Son, Israel lastly my beloved parents Duncan and Emily Sabwa.

ABSTRACT

Aquaponic systems integrate the culture of both plants and fish in recirculating aquaculture units. The nutrients derived from the fish are utilized by the plants for their growth in the system. The current study determined the effect of fish stocking density on growth performance, water quality and economics in a Nile tilapia (*Oreochromis niloticus*) - Lettuce (*Lactuca sativa*) aquaponic system. The fish whose initial average weight was 17.9 ± 0.07 g were stocked at densities of 150, 300 and 450 fingerlings m^{-3} in the three treatments; D1, D2 and D3 respectively and raised for 56 days. The treatments were replicated five times. Each treatment was subjected to 16 lettuce plants m^{-2} as the planting density. With respect to increasing stocking density the water quality parameters ranged from dissolved oxygen: 4-7 mg L^{-1} ; pH = 6.3 to 7.3; alkalinity: 64-90 mg L^{-1} ; TAN: 0.32 to 0.57 mg L^{-1} ; NO_3 : 0.13 to 0.36 mg L^{-1} and NO_2 : 0.020 to 0.046 mg L^{-1} . There were significant ($p < 0.05$) interactions between stocking density and the water quality parameters (dissolved oxygen, pH, Total Ammonium Nitrate (TAN), ammonia and NO_2). The growth performance of fish in terms of final weight, Food Conversion Ratio (FCR) and percent survival decreased with increasing stocking density. The final fish weight was 42.6 ± 3.1 g, 32.0 ± 3.8 g and 25.2 ± 4.2 g; the FCR was 1.45 ± 0.13 , 1.66 ± 0.1 and 1.86 ± 0.07 , and % survival was $93.85 \pm 2.1\%$, $89.73 \pm 2.35\%$ and $82.05 \pm 2.9\%$ for stocking densities 150, 300 and 450 fish m^{-3} respectively. The SGR decreased with increasing stocking density giving mean values of 1.5630, 1.1613 and 0.6371 for stocking densities 150, 300 and 450 fish m^{-3} respectively. Lettuce yield increased with increasing stocking density giving final biomass of 166.4 ± 9.8 g, 276.8 ± 23.2 g and 304.6 ± 23.2 g for stocking densities 150, 300 and 450 fish m^{-3} respectively and the lettuce heights were 22.6 ± 1.2 cm, 26.9 ± 0.9 cm and 24.9 ± 0.9 cm for stocking densities 150, 300 and 450 fish m^{-3} respectively. Basing on the enterprise budgets, all the treatments posted positive returns to the risk ($i=18\%$) and were viable investments. The break-even prices were Kshs 196.7, 137.9 and 114.2 kg^{-1} for stocking densities 150, 300 and 450 fish m^{-3} respectively. The break-even prices for variable costs were able to cover the cost of fish and lettuce in the local market as they were below the selling price the farm was able to receive in the market. All the other stocking densities were profitable except at 150 fish m^{-3} and profitability increased with increased stocking density. Stocking density of 300 fish m^{-3} had the best-combined performance in terms of growth of fish and lettuce, water quality and profitability, hence recommended for small-scale aquaponic farmers.

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LIST OF ABBREVIATIONS, ACRONYMS AND SYMBOLS

AOAC	Association of Official Analytical Chemists
AU	African Union
BEP	Break-even price
CCI	Chlorophyll Content Index
CP	Crude protein
CRBD	Completely randomized block design
CC	Carrying capacity
DOM	Dissolved organic matter
FAO	Food Agricultural Organization
FAOSTAT	Food Agricultural Organization Statistics
FCR	Food conversion ratio
GoK	Government of Kenya
IAMS	Integrated Aqua-vegeculture system
IRR	Internal rate of return
NFT	Nutrient film technique
NPV	Net Present Value
POM	Particulate organic matter
RAS	Re-circulating Aquaculture System
SD	Standard deviation
SDGs	Development Goal
SEM	Standard error of means
SGR	Specific growth rate
SSA	Sub-Saharan Africa
STI	Science, Technology and Innovation
STISA	Science, Technology and Innovation Strategy for Africa

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CHAPTER ONE

INTRODUCTION

1.1 Background of the study

Over the last five decades, there has been increased human population growth in the world (FAOSTAT, 2018; Tripathi *et al.*, 2019; Busch and Lacy, 2021), thereby reducing food supply per capita in most countries. Declining land for food production has also contributed to a decline in food security over the past five decades globally, regionally and locally (Cafiero *et al.*, 2018; Chen *et al.*, 2019; Cottrell *et al.*, 2019; Byson *et al.*, 2021). The estimated global food demand from the 8.6 billion people by the year 2030 may exceed 10,094 million metric tons annually while the production may be far much less than this. The production of fish from both capture and aquaculture is projected at 181 million metric tons against the consumption of 151 million metric tons annually by the year 2030 (FAOSTAT, 2018; Chan *et al.*, 2019). Food insecurity is dire in African countries especially in Sub-Saharan Africa (SSA) due to over-reliance on traditional sources as well as the dominance of agrarian and subsistence forms of agricultural production (Nsiah and Fayissa, 2019; Ssozi *et al.*, 2019). Consequently, better food production practices are key remedies for food insecurity in many countries in SSA (Hall *et al.*, 2017; Rademaker and Jochemsen, 2019; Issahaku and Abdulai, 2020).

Aquaculture as a food production practice has been growing rapidly, often exceeding 20% in most countries of SSA during the last five decades (Smith, 2019; Tran *et al.*, 2019; Gephart *et al.*, 2020), assisting in combating food insecurity in areas it has been adopted (Soliman and Yacout, 2016; Asiedu *et al.*, 2017; Kara *et al.*, 2018). FAO

estimates that by the year 2025, more than 50% of fish production in Sub Saharan Africa will be generated from aquaculture (FAOSTAT, 2016). In any case, large-scale aquaculture operations are constrained by the unavailability of land and water resources as well as by environmental concerns (Adeleke *et al.*, 2020). As aquaculture intensifies, production units become more efficient as profit margin increases, endangering land, water and genetic resources due to pollution-related factors (Juju *et al.*, 2020).

As the human population increases, competition for freshwater sources also increases especially from the domestic, industrial and agricultural users. There has also been a large push to have sustainable farming practices that would avail enough food for people without causing environmental degradation or pollution. Studies show that small-scale farming ventures contribute significantly to household income and nutrition, thereby reducing poverty (Hampwaye *et al.*, 2009; Chisonum *et al.*, 2020; Kapembwa *et al.*, 2020; Mhlanga, 2020; Rob *et al.*, 2021). The increasing demand for novel food production has created more demand for fresh water for irrigation. Aquaculture effluents can easily be used for irrigation purposes, where the plants will utilize the available nutrients (Menegaki *et al.*, 2007). To be able to feed the world's ever-growing population, there will be a need to come up with highly productive and sustainable food production systems in urban areas (Nelson, 2007; Fanzo *et al.*, 2020).

In recent deliberations, the African Union (AU) identified advancement in Science, Technology and Innovation (STI) under the theme of Science, Technology and Innovation Strategy for Africa (STISA) 2024, (Agenda 2063) to spearhead the development of aquaculture in SSA (African Union, 2018). There has been a widespread suggestion that aquaculture production techniques that give low yields

should be abandoned and those that upscale or increase fish production employing Science, Technology and Innovation be encouraged (Mavhunga, 2017; Saidi and Douglas, 2018). Several reports have recommended solutions that integrate aquaculture and other production technologies to help in the development of the sector (Ampadu-Ameyaw *et al.*, 2016; Kraemer-Mbula *et al.*, 2018; Nissen *et al.*, 2021).

Aquaponics is an integrated sustainable food production technology (Asciuto *et al.*, 2019; Lennard and Goddek, 2019; Abusin *et al.*, 2020) linking hydroponic production with Recirculating Aquaculture System (RAS) (Endut *et al.*, 2016; König *et al.*, 2018; Colt *et al.*, 2021) to increase fish and vegetable production (Filep *et al.*, 2016). In this system, nutrients from the fish growing unit are used by the plants in the hydroponics component (Goddek and Körner, 2019). The system helps in solving water scarcity problems when the same water is recycled within the same system. It also solves the issue of treatment of wastewater generated from the fish culture (Goddek *et al.*, 2019; Kledal *et al.*, 2019 b). In the process, the plants remove nutrients hence improving the effluents and further enhancing fish production (Goddek and Keesman, 2020; Osti *et al.*, 2020)

The use of aquaponics system has gained attention and popularity in the world as a bio-integrated model for a sustainable food production system (Savidov *et al.*, 2005; Kloas *et al.*, 2015; König *et al.*, 2016). In Kenya, aquaponic is still a new technology at the experimental level but one that has great potential in agri-food systems (Dijkgraaf *et al.*, 2019). Different fish species such as Tilapia (*Oreochromis niloticus*), African catfish (*Clarias gariepinus*), rainbow trout (*Oncorhynchus mykiss*), common carp (*Cyprinus carpio*), barramundi (*Lates calcarifer*) among many others

have been reared in the system (Adler *et al.*, 2000; Rakocy *et al.*, 2004; Filep *et al.*, 2016; Petrea *et al.*, 2016). Different species of vegetables such as iceberg, butterhead, romaine and leaf varieties of lettuce (*Lactuca sativa*), tomatoes (*Solanum lycopersicum*), and basil (*Ocimum basilicum*) also have been successfully grown in the aquaponics system (Pantanella *et al.*, 2010; Sace and Fitzsimmons, 2013; Salam *et al.*, 2014; Filep *et al.*, 2016). The overall growth performances reported in these experiments were dependent on the fish production system and management used.

In the aquaponic system, nutrients (mainly nitrogenous compounds and phosphates), derived from excretions and decomposition of uneaten foods are absorbed by the plants which were grown in the hydroponic component thus harmful by-products of fish production become ameliorative input for plant production (Fang *et al.*, 2017; Setiadi *et al.*, 2018; Kledal *et al.*, 2019a). In traditional aquaculture, water is used once and the resultant effluents are discharged directly into the environment, leading to less efficient water use and production of wastes that pollute waterways adjacent to aquaculture farms (Boyd, 2017; Chatvijitkul *et al.*, 2017; Zhang *et al.*, 2019). However, in an aquaponic system, nutrients from the fish growing unit are used by the plants in the hydroponics component (Goddek and Körner, 2019). Nutrient removal by plants improves the quality of effluent and may also enhance fish production (Nuwansi *et al.*, 2019). Adoption of aquaponic systems amongst farmers in Kenya and beyond is still at an infancy stage with people trying to understand how the system works, which fish and plant species to use and the optimal stocking density of fish and plants (Dijkgraaf *et al.*, 2019).

Stocking density is one of the most critical factors that can influence fish performance in the aquaponic system. It affects fish growth, feed utilization, survival, behaviour,

health, water quality, and gross fish yield (Oké and Goosen, 2019; Maucieri *et al.*, 2019). Running an aquaculture unit close to its carrying capacity (CC) allows one to efficiently use space, reduce variation in the amount of feed input, and eventually maximizing on production (Boxman, *et al.*, 2016). Stocking fish at lower density results in inefficient utilization of space and low yields, whereas stocking fish at densities above the CC impairs the growth performance of fish due to the accumulation of metabolic wastes such as faeces, impairment of fish social interaction, and deterioration of water quality (Birolo *et al.*, 2020; Maucieri *et al.*, 2020).

According to Irwin *et al.* (1999), there exists both positive and negative relationship between the stocking density and fish growth, which is usually species-specific. When fish are stocked at higher stocking densities, their growth decreases and their size variation increases due to the intraspecific competition (Lambert and Dutil, 2001; Capelle *et al.*, 2020). It is therefore important to determine the optimal stocking density for any aquaponic system to optimize both fish and vegetable performance without compromising the water quality and the economic returns associated with the system. Enhanced fish and crop production will likely improve the economic production of the system. The profitability of the system relies on the fish yield obtained from the aquaculture unit and vegetables raised in the hydroponics (Engle, 2015). Moreover, the profitability of the system is related to the amount of fish obtained from the system which is in turn related to the stocking density.

The current study, therefore, investigated the effects of varying stocking density on the growth performance, water quality and economic benefits of Nile tilapia (*O. niloticus*) and lettuce (*L. sativa*) grown in an aquaponics system in Kenya.

1.2 Statement of the problem

The ultimate aim of any fish production system is to increase and/or maintain a high level of overall fish growth performance, survival and good water quality. These requirements ultimately translate to high yield, improved economic benefits and maintenance of ideal environmental conditions within the fish culture system. The open aquaculture systems that have been practised in Kenya, which rely on open ponds, cages or integrated systems have often resulted in low fish growth performance, low economic benefits and sometimes deterioration of water quality that affect the environment whenever the effluent is discharged from the fish culture units (Minoo *et al.*, 2006). To solve the problem of low production, there have been suggestions for increasing fish stocking density. However, increasing the fish stocking densities without changing the culture unit size would probably result in an increased critical standing crop which cannot be supported by the culture system. Consequently, many aquaculture operations result in the deterioration of water quality and impaired fish growth performance with low fish yields. A system that entails recirculating water systems and aquaculture production has been proposed to address these challenges and aquaponics fits in this context very well.

The aquaponic system is a new technology in Kenya (Dijkgraaf *et al.*, 2019), with little documentation on its operation and therefore outcome in terms of fish growth performance, survival and good water quality remain speculative. Similarly, there are neither developed and specified protocols and specifications nor standard operating procedures for aquaponic practices in Kenya. Being a very expensive fish culture system, optimal operation of aquaponics should rely on increasing and/or maintaining a high level of overall fish growth performance, survival and good water quality. The

profitability of recirculating systems is also of major concern since it is expensive to construct and operate the system while returns from the fish component may not be enough to make it sustainable. This raises concerns about the economic benefits achieved from the aquaponic systems in Kenya. These will ultimately translate to high yield, improved economic benefits and maintenance of ideal environmental conditions within the fish culture system. In aquaponics, this can be achieved through proper fish management practice by the fish farmer. To solve the problem of low production, there have been suggestions for increasing stocking density. Increased stocking density without changing the culture unit size will result in an increased critical standing crop that cannot be supported by the culture operation (König *et al.*, 2018). Such an operation will result in the deterioration of water quality and impaired fish growth performance. An aquaponic system, which entails recirculating water systems and aquaculture production, has been proposed to address these challenges. However being a relatively new technology, there is currently no study on several aspects of the culture system and therefore its outcome in terms of fish production remains only at the experimental level (van Gorcum *et al.*, 2019). A knowledge gap on the performance of aquaponics under different stocking densities may hinder the adoption of aquaponics and the generally retarded development of aquaponic culture in Kenya.

1.3 Justification

Agenda 2063 recognizes Science, Technology, and Innovation (STI) as the main driving engine that will assist in achieving the development goals in Africa. This window, therefore, provides an opportunity for investment in STI to increase fish production and sustainable use of water resources. One of the pillars of Kenya's vision 2030 is to achieve food for all (Mwenzwa and Misati, 2014), and meeting the Sustainable Development Goals (SDG) goal number 2. To realize the aforementioned objective, efforts have been geared towards improving fish production in Kenya from the small-scale fish farmers who currently constitute over 95% of fish farmers in Kenya yet produce less than 5% of the total national aquaculture fish production. The remaining 5% of the fish farmers are large-scale and control over 95% of the country's fish production. To achieve high production potential, basic information on aquaculture technology and management practice that enhance production is urgently needed.

Aquaponics systems have certain theoretical and practical advantages including the effluents or wastes derived from the first biological system serving as key important nutrients in the second biological system. The integration of both fish and plants results in a polyculture-type of a system that increases (agro-diversity) and yields multiple products. The system also helps in the removal of the toxic substances present in the effluents, which will just be discharged into the environment in conventional aquaculture systems and can act as a means of controlling soil-borne plant diseases. These nutrients once absorbed, the plant can reduce dependency on fertilizers by nearly 77%.

Some merits associated with aquaponic systems include; the use of plants as a secondary product which end up reducing the pollution load (waste concentration) through nutrient uptake and elimination of soil-borne diseases in the hydroponic system. The system also addresses the nutrient deficiency in arid and semi-arid or water-deficient regions since aquaponics is mostly a soilless culture system and not dependent on fertilization and irrigation. The hydroponic component in the aquaponic system provides the much-needed biofiltration hence reducing the usual effects of feeds on water quality. The costs for individually monitoring water quality parameters and doing the needed analysis is drastically reduced; plants grown in the hydroponic component grow without soil, hence preventing soil-borne plant diseases which eventually results in higher yields (Rakocy *et al.*, 2012).

Although studies have been carried out in recirculating systems in aquaculture (van Gorcum *et al.*, 2019), there has been limited research on the growth performance data of fish species like *O. niloticus* in an aquaponic system in Sub-Saharan Africa (SSA). This study will, therefore, add the much-needed information on the effects of stocking density on the performance of fish, water quality, nutrient balance and the economic benefits in an aquaponics system. The results will be useful in informing policymakers and government agencies on improved fish and vegetable production in the country.

1.4 Objectives of the study

1.4.1 Main objective

The main objective of this study was to evaluate the effects of stocking density on growth performance, water quality and economic viability of mono-sex Nile tilapia (*O. niloticus*) and lettuce (*L. sativa*) in an aquaponics system.

1.4.2 Specific Objectives

The specific objectives of the study were:

1. To determine the effects of stocking density on the growth performance of *Oreochromis niloticus* in an aquaponics system.
2. To determine the effects of stocking density on the yield of lettuce (*Lactuca sativa*) in the aquaponics system integrated with *O. niloticus*.
3. To determine the effects of stocking density on water quality, nutrient removal efficiency in the aquaponics system integrating *O. niloticus* and *L. sativa*.
4. To determine the effects of stocking density of *O. niloticus* and *L. sativa* on the economic viability of the aquaponics system.

1.5 Hypotheses

This study was guided by the following null hypotheses:

- H₀₁:** Stocking density does not influence the growth performance of *O. niloticus* in an aquaponics system
- H₀₂:** Stocking density does not influence the *Lactuca sativa* growth performance in the aquaponics system integrated with *O. niloticus*.
- H₀₃:** Stocking density does not influence water quality, nutrient removal efficiency in the aquaponics system integrating *O. niloticus* and *L. sativa*.
- H₀₄:** Stocking density has no influence on the economic benefits of Nile tilapia (*O. niloticus*) and lettuce (*L. sativa*) in the aquaponics system.

CHAPTER TWO

LITERATURE REVIEW

2.1 Status of global aquaculture

The United Nation's Food and Agriculture Organization (FAO, 2018) projects that to be able to feed the world population by 2050, agricultural output coming from the fisheries sector must increase somehow by over 60%. Achieving this is an enormous challenge especially taking into account the increased cases of hunger, malnutrition and poverty. There is therefore a great need to accelerate and diversify aquaculture operations and production to address the demand for food fish created by the ever-increasing human population globally by the year 2030 (Munguti *et al.*, 2021).

Global aquaculture production which includes aquatic plants in 2016 was 110.2 million tonnes was estimated at \$ 243.5 billion. This production included 80.0 million tonnes of food fish and 30.1 million tonnes of aquatic plants valued at \$ 231.6 billion and \$11.7 billion respectively. The non-food fisheries products were 37, 900 tonnes valued at \$ 214.6 million (FAO, 2018). The demand for fish and seafood has gradually increased due to the ever-growing global population and aquaculture has greatly contributed to meeting these demands with over half of all fish and seafood now being farmed (Tocher *et al.*, 2019). Aquaculture provides most of the fish and its products to the market currently, and thus it helps reduce pressure on the capture fisheries. Its products are relatively higher and thus it contributes to almost half the percentage of the fish products in the market, hence an important protein source globally (FAO, 2010). It is also a platform for provision of

employment to many people and boost of the economy through export of its product. However it's posing various challenges to the environment which have to be addressed immediately including, waste water released by aquaculture to the reservoirs which may lead to eutrophication and hypoxia and lastly the use of fish meal and bait fish (Simeonidou *et al.*, 2013; Duarte *et al.*, 2015). There have been several factors contributing to success in aquaculture; such include; improved water management, economic management, better feeding strategies, fish feeds, improved management of health and agriculture integration (Obiero *et al.*, 2019)

The increase in aquaculture ventures globally has contributed to food security, decrease in malnutrition cases and job creation among many others (FAOSTAT, 2018). However, as aquaculture practices intensify, environmental pollution is still an imminent problem. Traditional recirculation aquaculture systems release effluents into the environment causing pollution and water wastage (Chen *et al.*, 2019). According to Aubin *et al* (2019), to adequately address these challenges and at the same time be able to meet the demand for fish globally, technological advancement in aquaculture is a prerequisite especially in Sub-Saharan Africa.

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meal and bait fish (Simeonidou *et al.*, 2013; Duarte *et al.*, 2015). Aquaculture involves captive rearing and production of aquatic organisms, plants, shellfish and finfish under a controlled condition (FAO, 2010). Aquaculture production methods have been developed by many countries to increase the amount of fish to meet the market demand. Four major systems adopted are; open water system, pond culture, raceway culture and recirculating aquaculture system (FAO, 2010).

Aquaculture is a rapidly growing industry whose products are considered to be a solution for the current world's economic problem, majorly with the massive growth in population (FAO, 2020). Aquaculture is depended for creation of vast job opportunities aside from providing good quality and accessible food. Aquaculture activities have intensified with improved technology and thus more fish in the market. Recent technologies in aquaculture include, cage culture, recirculating systems, aquaponics and many others (Jena *et al.*, 2017).

However, there is a growing concern that the growth of aquaculture has negative implications to the environments near them. The negative impacts of aquaculture which include; destruction of the environment (mangrove forest clearing) to construct aquaculture facilities, pollution of the water meant for human and livestock consumption, eutrophication and nitrification of effluents receiving ecosystems, the issue of escapes and the ecological implications of aquaculture to the natural environment (Martinez-Porchas and Cardova, 2012).

Pollution of the environment by aquaculture is a major concern since it has health implication to people and livestock around the facilities. Therefore it's not ethically correct to release aquaculture wastes (majorly uneaten food, faecal material excreted by fish and other antibiotics used) in water bodies that is used by people. The

concerns regarding release of waste in water include; over enrichment of the water bodies leading to excess algal bloom which is dangerous to aquatic organisms and fish, and antibiotics used may have health implications to people using the water. An alternative method of fish production should be established that will be environmental friendly by minimizing or reducing the release of wastes (Beveridge *et al.*, 1997; Fernandes *et al.*, 2001)

The issue of releasing wastes to the environment by aquaculture systems is a major problem to environment. These nitrogenous wastes get into the systems through feeds given to fish, and the uneaten and the faecal matter produced through the gills and urine leads to accumulation of toxic ammonia in decomposition (Cao *et al.*, 2007; Amirkolaie *et al.*, 2011; Turcios *et al.*, 2014).

2.2 Background and Operation of Aquaponic systems

There is a divergence between the prediction of global food demand and the actual production of food where a large deficit currently exist (Conijn *et al.*, 2018; Gouel and Guimbard, 2019). Many countries in the world have committed themselves to end all forms of food and nutrition insecurity through the Sustainable Development Goals (SDG) (Nyström *et al.*, 2019) by focusing on all forms of food production. Together, finfish and shellfish contribute approximately seventeen percent of animal protein, and seven percent of all proteins, and are important for feeding the billions of people in most developing countries (Deppermann *et al.*, 2019; Obiero *et al.*, 2019). Fish gives high-quality protein with balanced essential amino acids, minerals like zinc and iron, vitamins, docosahexaenoic and eicosapentaenoic omega-3 fatty acids, which are mostly in highly bioavailable forms (Lynch *et al.*, 2020) which renders fish a super

food. An increased and sustainable fish production system should thus bridge the food supply and demand through innovative solutions (Chowdhury *et al.*, 2017; Bentham *et al.*, 2020). During fish production, the conventional monoculture systems for production limit higher production and has less prospective to accommodate an assortment of multi-trophic solutions for aquaculture (van de Vis *et al.*, 2020), which can be achieved through the cultivation of fish and plant.

The cultivation of both fish and plants is old, in historical terms and dates back to 2000 years ago in tropical Asia, India, and China (Jones, 2016; Mathias and Anthony, 2020). Aquaponics (derived from the first four letters of the word “*Aquaculture*” and the last six letters of “*hydroponics*”) (Diver and Rinehart, 2000; Kledal *et al.*, 2019) combines the traditional hydroponic plant husbandry and the recirculating aquaculture system (RAS) in a symbiotic integration (Rakocy, 2012; Palm *et al.*, 2018). Hydroponics involves planting vegetables in a soilless medium while aquaculture involves the culture of aquatic organisms (plants and animals like fish, shell fish) with the aim of making profit. There are different criteria of practising this system, but the common factor in them is that plants obtain moisture and nutrients from fish waste and not soil. Hydroponic is faced by different challenges which include; inadequate nutrient solutions reaching the plants and the initial capital is relatively high. Its advantages include; possibility of growing more than two times in a limited area, there is faster growth of plants hence the vegetables are more palatable and have a good texture, plants do not have to compete for moisture and nutrients and it does not require digging and weeding and above all, soil based characteristics (FAO, 2010). Integrating aquaculture and hydroponics in aquaponics system provides an efficient mineral nutrient recycling, reduce water used by recirculation, reducing

environmental pollution and degradation; however the cost effectiveness and capability of aquaponics to contribute globally to sustainable food production is still a challenge (Rakocy, 2012; Palm *et al.*, 2018).

The potential of Aquaponics can be seen via several lenses, which includes how it contributes to community transformation, implementation of relevant policies and specific programs within a given region. Though aquaponics has the potential to produce food for the world, it is a young science whose development and understanding of its full operation is still being looked at by many players in aquaculture and related fields. There exist several scientific and engineering challenges in aquaponics, but basing on the associated opportunities then this technology is something to look at and invest in (Veludo *et al.*, 2012; Goddek *et al.*, 2015; Shafeena, 2016).

Recirculating aquaculture systems (RAS) were initially developed as a technology for intensive aquaculture, used mainly in areas where water was scarce: these systems made it possible to recycle up to 90–99% of the water with the help of various installed components. The recycling and treatment of water allow the farmer to have greater control on water quality issues and effluent management which assures him or her to have an enabling optimal condition when culturing fish (Turcios and Papenbrock, 2014; Palm *et al.*, 2018). In contrast, RAS systems have several limitations like high capital and operational investments, difficulties in treating fish diseases and general management of the whole system (Schneider *et al.*, 2006). Moreover, as water continually flows, the pumps, degassing units and other important components are installed using a lot of electricity. The elevated electricity costs and high water reuse sometimes can make the whole project uneconomical (Shepherd and

Bromage, 1988). RAS systems are also not very simple to understand and operate; they require technical and experienced supervision and management to ensure that the system works as planned (Lekang, 2007). As time goes by several developments have been effected to address some of these issues thereby simplifying their management (Muir, 1982 and Rosenthal, 1993).

When the RAS system is combined with hydroponics, the nutrient-rich effluent from fish tanks fertilizes the plants grown in the hydroponic component. The roots of the planted crop and rhizobacteria (root-associated bacteria) help to partially remove the nutrients present in the water thereby improving the water quality which is essential for fish production. These nutrients generated from the fish component of the aquaponic system can build up to toxic levels causing fish mortality. So the inclusion of the plants in the system makes sure that most of these nutrients are absorbed by the plants reducing the toxicity. The plants, therefore, act as biofilters stripping off nitrates, nitrites, ammonia and phosphorus before the water is recycled back into the fish rearing tanks.

As aquaculture intensifies, environmental pollution remains an enormous challenge as a result of the generation of substantial amounts of effluents directly from the fish farms (Read and Fernandes, 2003). These effluents are composed of nutrients, various organic and inorganic substances like phosphorus, ammonium, dissolved organic substances (Lekang, 2007). The receiving aquatic ecosystems are hence polluted and this affects the aquatic life. If for example, we assume that the feed conversion ratio (FCR) is between 1 and 3, one, therefore, needs 1 to 3 kilograms of fish feed to generate a kilogram of fish (Houle *et al.*, 2011). Close to thirty-six percent of the fish feed eaten is excreted as organic waste (Bergheim *et al.*, 2009). Close to 65 to 75% of

this feed is unused and remain as waste in water in the form of nitrogen and phosphorus (Read and Fernandes, 2003, Bergheim *et al.*, 2009). Depending on the fish species and culture technique used, close to 80–88% of carbon, 85% of phosphorus and 52–95% of nitrogen are lost to the environment. This happens through wastage of fish feeds, fish excretion, faecal production and respiration. Remediation of aquaculture effluents is crucial due to water scarcity in several areas and the impact the effluents could cause to the environment as a result of nutrient loading (Shepherd *et al.*, 1988; Blancheton, 2000; Read and Fernandes, 2003; Chen *et al.*, 2006). Most RAS systems separate the faecal matter as fast as possible hence reducing the nutrient load which in turn enhances the nitrification process and reduces root clogging and aeration (Azim *et al.*, 2008).

The use of an aquaponics system is a remedy to the above problem since it's a balanced ecosystem whereby the uneaten feeds and the faecal matter excreted by fish are not released to the environment, instead they are acted upon by the biofilters and they are used by the plants as nutrients for growth. These faecal matters when released to the nearby environment leads to excess enrichment of the system which is harmful to the aquatic organism.

Instead of releasing this waste to the environment, it can otherwise be used in agriculture as a source of nutrient to the plants since it contains all the mineral components necessary for plant growth. Therefore, establishing a system that leads to mass production of fish that is environmental friendly is necessary to reuse this particular waste.

Recirculating Aquaculture System (RAS) involves stocking fish at a high stocking density. This leads to high waste production and hence mechanical and biological

filters are needed to keep the water clean for consumption by the fish (Kopsa, 2015). RAS is a new aquaculture technology that is being adopted in many countries to increase the volume of fish produced, to reduce the volume of water used, to reduce fishing activities which have led to the reduction and extinction of some indigenous species and to reduce the chance of fish escape to the wild and vice versa (Rackocy, 2006).

In the past three decades there has been a lot of different aquaponic system designs, different plants and fish species incorporated. Several experimental works have also been documented as people test the operations, designs and management of these systems (Guedar *et al.*, 2011). The results from these experiments and works have reported efficient use of water and land resources, reduction in the risks associated with crop failure, additional food sources, income generation diversification and low operational costs as compared to aquaculture stand-alone ventures. Although it seems that recirculating aquaponic systems show an ingenious diversity with efficient ability to produce multiple products, manage the water quality parameters effectively and ensuring there is no pollution to the environment, however, success and their popularity is still an issue in potential regions of adoption. Low public acceptance of the products harvested from the aquaponic system and undesirable results has led been the main contributor towards the low adoption rates in many parts of the world. The main problem associated with the latter conclusions arises from the disparity between productive units of the system and the un-optimized intensity of the fish and plant species that can be put in the system. There is also very little data on the concentration limits of different nutrient elements at which toxicity or deficiency occurs. As a result of extensive research, the technical knowledge and skills on

management, design, installation and choice of fish and plants to be incorporated has considerably improved and also initiated the implementation of socio-economic studies in the regions where this aquaponic system is adopted. Recent advances by researchers (Houle *et al.*, 2011) have turned aquaponics into a working model for sustainable food production has the potential to supply quality foodstuff for people globally. This has brought hope to folks from different regions especially in the arid, semi-arid and urban centres.

2.2.1 Different types of aquaponic system types and designs

Aquaponics system designs are very much similar to that of recirculating aquaculture systems, the exception being an additional hydroponic component that assists in purifying the effluents and removing the fine and dissolved solids from the system (Forchino *et al.*, 2018). Basic aquaponics systems are composed of four major units (Figure 1), through which the water is circulated: The first component includes the fish production unit: this is the tank array where fish are reared. The fish tanks are designed to allow a bigger percentage of the wastes inform as sludge and uneaten food to be removed and directed to the mechanical filter (Kwon and Kim, 2020). The second unit includes a waste processor: This is a solids filter used to remove from the water all suspended solids that are mainly composed of fish excretions and small portions (about <5%) of uneaten feed (Pfeiffer *et al.*, 2008). The third unit includes a biofilter unit: which is essential for the oxidation of toxic ammonia that is secreted by the fish to less toxic nitrate, thus allowing water recycling without replacement continuously (Van Rijn, 1996). The fourth includes a hydroponic unit: this is where the plants are grown. The plants absorb essential nutrients such as nitrogen and phosphorus with the help of their root system. This removes most of these nutrients in

the water before it is recycled back to the fish tank (Rakocy, 2012). The positioning of the sump may vary depending on how the hydroponic compartment is elevated (ranging from 0.2 m to around 1 m) (Rakocy, 2007; 2012). This helps to reduce additional pumping needs by allowing water to enter the sump by gravity (Rakocy, 2007; Timmons and Ebeling, 2010).

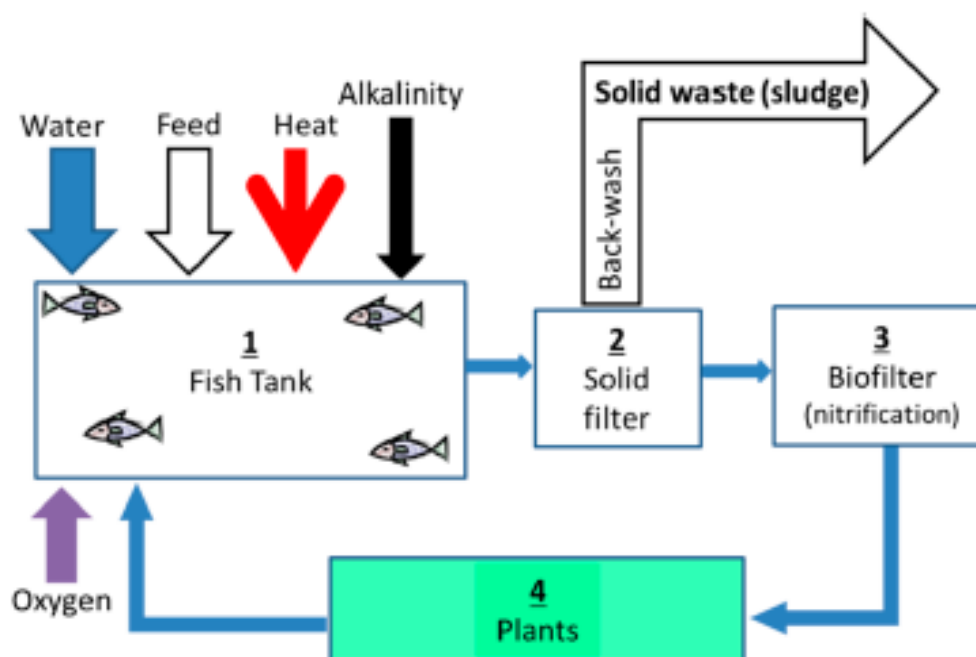


Figure 1: Schematic Aquaponic systems (Yogev *et al.*, 2016)

Modern aquaponics is divided into three main systems, nutrient film technology, deep water or floating raft system, media-based system (Rakocy, 2006). To successfully practice aquaponics for better results, the stocking density of both the fish and vegetables are crucial to ensure a safe ecosystem in which the three main organisms (fish, vegetables, and bacteria) in aquaponics can thrive. The stocking density of tilapia/any fish grown in the system should be optimized to maintain the water quality suitable for the growth of the organisms.

Different types of aquaponic systems exist (Table 1), and they are categorized

according to the growth-bed designs, hydroponics, and method of coupling (coupled or decoupled) (Kloas *et al.*, 2015). The first category includes the floating aquaponic raft system which is widely used to grow various leafy vegetables such as lettuce, spinach, coriander, amaranth among many others (Lennard and Leonard, 2006; Rakocy *et al.*, 2006; König *et al.*, 2018). The second category is the nutrient film technique (NFT) which is used widely to grow several crops such as garlic, tomatoes, cucumber, strawberry and beans (Edaroyati *et al.*, 2017). The last category is composed of a bed that has media. Different materials are used as media, such includes tuff, sand, gravel, expanded clay pellets, ballast, perlite and peat moss (Diver, 2006; Lennard and Leonard, 2006; Rakocy *et al.*, 2006; Love *et al.*, 2014).

Table 1: Examples of aquaponic systems in the literature

Type of aquaponic system	Combinations	References
Floating raft system	Tilapia + basil	Rakocy <i>et al.</i> , 2003
	<i>Oreochromis niloticus</i> + lettuce	Palm <i>et al.</i> (2014)
Nutrient film technique (NFT)	Grass Carp, (<i>Ctenopharyngodon idella</i>) + lettuce (<i>Lactuca sativa</i> L.), dill (<i>Anethum graveolens</i> L.), rocket (<i>Eruca sativa</i>), coriander (<i>Coriandrum sativum</i> L.), and parsley (<i>Petroselinum crispum</i>)	Lennard and wards (2019)
	Pangas (<i>Pangasius hypophthalmus</i>) + marigold (<i>Tagetes erecta</i>)	Mohapatra <i>et al.</i> (2020)
	Climbing perch, <i>A. testudineus</i> + <i>B. alba</i>	Anantharaja <i>et al.</i> (2017)
Media-based bed	Nile tilapia + Common carp + Cucumbers + Tomato + Lettuce	Knaus <i>et al.</i> , 2017
	Nile tilapia + spinach	Rono <i>et al.</i> (2018)
	Nile tilapia + Sweet Wormwood + Pumpkin, + Amaranth	Gichana <i>et al.</i> , 2019

2.2.2 Performance of the aquaponic system

Aquaponic systems demonstrate a high production efficiency when contrasted with traditional aquaculture systems especially in the use of water nutrients, yield per unit area and the limited release of polluted wastes into the environment (Endut *et al.*, 2016). To achieve this, the recirculating system raises huge amounts of fish in relatively small volumes of water, which is treated through sedimentation and biofiltration (Palm *et al.*, 2018) which increase the concentration of non-toxic nutrients as well as the organic matter, that is directed into a secondary tank with economically valuable crops (Kloas *et al.*, 2015; Goddek *et al.*, 2019). The systems can maintain water quality at ranges suitable for fish culture at 20-30% water exchange in the fish holding tank in the system (Shete *et al.*, 2013).

In the hydroponic unit, the available plant and nitrogen-fixing bacteria aid in the breakdown of the by-products, from ammonia to nitrates absorbed by the plants before recycling back the water into the fish rearing unit (Goddek *et al.*, 2016). Planted vegetable act as filters by utilizing the nutrients present hence purifying the water before recycling it back into the fish rearing tank (Pasch *et al.*, 2021). Love *et al.* (2015 b) indicated that when water is recycled daily this ensures that the plants utilize the nutrients derived from the breakdown of both feed and excretion wastes improving the water quality of the system. However, large variations exist which are associated with the origin of the aquaponics system, technological innovations, and levels of investment (Suhl *et al.*, 2016; Karimanzira *et al.*, 2017).

In several studies, edible plants like spinach, coriander, kales among many others, were used to reduce the waste products from recirculating aquaculture systems. However, in the mid-1970s those farmers operating recirculating aquaculture systems introduced the idea of practising aquaponics (Love *et al.*, 2015a). Watten and Busch (1984) described aquaponics as a recirculating system that integrates both plants and fish. During the infancy stages of the development of aquaponics, the first known closed-loop aquaponics system (called an Integrated Aqua-Vegeticulture system, IAVS) that used tilapia culture effluent into sand or gravel-planted tomato beds was designed (Diem *et al.*, 2017). Since then, there have been innovations in aquaponics technology transforming it into a viable enterprise and one that can produce food for the masses (Pantanella, 2008). Over the last thirty years as the integration of aquaculture into the hydroponic system has advanced, there have been several successful aquaponic system designs, working protocols, diverse plants, and aquatic animal species cultured to which we can refer to (Rakocy *et al.*, 2006; Love *et al.*, 2015 a; Diem *et al.*, 2017). Today, aquaponics is primarily practised in greenhouses or outdoor locations where the climate is favourable. Such an environment utilizes both hydroponic and aquaculture procedures of culture and their associated equipment. The recirculating system allows water to flow into the hydroponic beds where the plants are anchored thereby removing the extra ammonia and nitrites, the water from these beds is then recycled back to the fish tanks. The incorporation of recirculating aquaculture units with vegetable production has become a successful model for many players including environmental scientists and private entrepreneurs (Kledal and Thorarinsdottir, 2018).

The merits of promoting aquaponics research and subsequent production of

vegetables and fish give an alternative to the present monoculture initiatives. This system efficiently uses a water resource by allowing very little new water exchange hence maintaining water quality and quantity that is ideal for the growth of both fish and vegetables (Goddek *et al.*, 2015). The nutrients available in the wastewater from the fish production unit can be effectively utilized for plant growth rather than depending on physical fertilizer application (Pantanella, 2018); Aquaponics can be practised in semi-arid and arid areas as well as urban centres where space is limited (Blidariu and Grozea, 2011). The adoption of aquaponics for smallholder farmers is also hailed as the best way to enhance food production among these groups of people who suffer most forms of food insecurity (Goddek *et al.*, 2019).

The dissolved organic matter and fine solids accumulation are well managed when a recommended design ratio is employed. The biofilter depending on its design and components is intended to reduce the levels of ammonia and nitrate hence treating the culture wastewater. The water then flows through the hydroponic bed where some dissolved nutrients are taken up by the plants. Effluents from the aquaculture component have both dissolved and particulate organic matter (DOM and POM respectively) which mainly come from the feeds given to the fish.

These feeds are digested and metabolized while some are not eaten at all remaining as waste in the water in either dissolved form or suspended or settled solids (Joyce *et al.*, 2019). Nitrogen present in the system is an essential element composing of both proteins and nucleic acids. Proteins in the feeds, which represent up to 70% of fish production cost are the ultimate source of nitrogen. Only 35% of this nitrogen is harvested via fish biomass whereas over 65% is excreted in the form of ammonia or completely not consumed by the fish. The microbial community (fungi, bacteria,

viruses, protists and archaea) play important roles in the processes of mineralization and nitrification hence improving the overall productivity of the system (Joyce *et al.*, 2019).

2.3 Fish species reared in aquaponics systems

Fish species cultured in traditional aquaculture systems are also adaptable in aquaponics systems. Aquaponic systems can be established in freshwater, marine, and brackish water environments. Most cultured fish species can tolerate crowding a phenomenon that is good for aquaponic systems (Lennard and Goddek, 2019). As a result, different varieties of fish can be cultured successfully in aquaponic systems (Bich *et al.*, 2020). Many species of freshwater fish, which can tolerate crowding, do quite well in aquaponic systems (Oliveira *et al.*, 2020). Among the warm and cold fishes, tilapia, trout, perch, Arctic char, and bass are well adapted to the recirculating aquaculture system (RAS) (Diver, 2006). There is a growing number of studies that have cultured various fish species in aquaponics including, Nile tilapia (*Oreochromis niloticus*) (Babatunde *et al.*, 2019; Angkha *et al.*, 2020; Hussein *et al.*, 2020), African catfish *Clarias gariepinus* (Knaus *et al.*, 2020), Arctic char (*Salvelinus alpinus*), koi carps (*Cyprinus carpio*) (Paudel, 2020), pikeperch (*Stizostedion lucioperca*), Atlantic salmon (*Salmo salar*), Asian sea bass barramundi (*Lates calcarifer*), sturgeon (order Acipenseriformes), rainbow trout (*Oncorhynchus mykiss*), European eel (*Anguilla anguilla*), and largemouth bass (*Micropterus salmoides*) (Fronte *et al.*, 2019; Birolo *et al.*, 2020; Fischer *et al.*, 2021; Zarantoniello *et al.*, 2021).

Despite the diversity of fish species cultured in aquaponics, under both small-scale and large-scale (commercial) aquaponics, most of the data available for fish

performance in the aquaponics are based on tilapia production (Endut *et al.*, 2016; Lennard and Ward, 2019). In general aquaculture, Tilapia is one of the most widely cultured fish in the world representing more than 75% of world fish production (FAO, 2009), and this has been growing exponentially in recent years.

Palm *et al.* (2014) reports that Nile tilapia can be cultured successfully with lettuce (*Lactuca sativa*) and cucumber (*Cucumis sativus*) giving an average harvestable weight of up to 500 g in weight within less than a year. Knaus and Palm (2017) also reported better growth results when Nile tilapias were grown together with parsley (*Petroselinum crispum*) and basil (*Ocimum basilicum*). Some studies have reported a significant growth in several fish species, like channel catfish, rainbow trout, common carp, koi carp, goldfish, Asian sea bass (barramundi) and Murray cod, in trial aquaponics, but many commercial aquaponic units as described in the literature have used different strains of tilapia successfully (Shete *et al.*, 2013; Palm *et al.*, 2014; Kloas *et al.*, 2015; Rakocy *et al.*, 2016; Makori *et al.*, 2017; Fatima *et al.*, 2018; Yildiz *et al.*, 2019). It has been observed that the success of the aquaponics system relies on the overall performance of the fish in terms of weight gain, food conversion, yield, survival, biomass and on several aspects of management applied in the aquaponics (Maucieri *et al.*, 2019; Stoyanova *et al.*, 2019).

2.4 Plant growth in aquaponics systems

The production of vegetables through the integration of fish and plant production has been widely demonstrated by various researchers (Goto *et al.*, 1996; Rakocy, 2012; and Espinosa *et al.*, 2016; Knaus and Palm, 2017). Based on a meta-analysis of the vegetable production system, the most commonly grown crops among many commercial producers include; peppers (*Capsicum annuum*, 48%), basil (*O. basilicum*, 81%), non-basil herbs (73%), kale (*Brassica oleracea*, 56%), bok choy (*Brassica rapa* subspecies *chinensis*, 51%), tomatoes (*S. lycopersicum*), and Lettuce (*L. sativa*) each at 68%, chard (*Beta vulgaris* subspecies *cicla*, 55%), and cucumbers (*C. sativus*, 45%) (Rakocy *et al.*, 2004 a).

Other crops grown include two varieties of garnish (both Scallion and parsley (Pinho *et al.*, 2018), lettuce (Goto *et al.*, 1996; Jaeger *et al.*, 2019), tomatoes (Savidov, 2004; Karimanzira *et al.*, 2017), basil (Knaus and Palm, 2017), strawberries (Villarroel *et al.*, 2011), cucumber (Tyson *et al.*, 2008) and other herbs (Espinosa *et al.*, 2016). Most of these plants deposit huge amounts of nitrogen to their leaves and this can be manipulated by plant density and nitrogen availability (Seawright, 1998).

Including plants in aquaponic systems improves its performance in terms of yields, nutrient removal and profitability (Engle, 2015). The growth of such plants is rapid and different cropping systems can be used, such as staggered, batch and intercropping (Rakocy *et al.*, 2016). Comparatively, the biomass conversion ratio for crops is superior to that of fish; that is as much as 9 kg of plants, for example, lettuce can be grown using fish manure after feeding the fish with 1 kg of fish feed (Love *et al.*, 2015 a). There is some variation in crop properties based on the different types of aquaponics systems. Fruiting plants such as tomatoes are grown widely in media-

based aquaponic systems (Danner *et al.*, 2019; Eck *et al.*, 2019; Magwaza *et al.*, 2020), while herbs such as basil and thyme are grown widely in both raft and media-based systems (Þórarinsdóttir *et al.*, 2015; Mishra *et al.*, 2020). Leafy plants are grown mostly in any of the three aquaponic systems types, i.e., raft, media-based and nutrient film technique (NFT) (Schmautz *et al.*, 2016).

The number of crops grown in the system in relation to the number of fish reared must be balanced and optimized to increase the output and optimally use the available nutrients (Lennard & Leonard, 2006; Graber & Junge, 2009; Knaus & Palm, 2017; Baßman *et al.*, 2020). Vegetables grown can be harvested in three strategies; staggered cropping, batch cropping and intercropping. Batch cropping and intercropping are good for slow-growing plants like onions while staggered are good for continuous vegetable production (Rackocy, 2006). Plant densities ranging from 16-44 plants m⁻² have been used for 21 to 28 days, mainly on floating aquaponic raft systems. These densities can produce various yields ranging from 1.4 to 6.5 kg m⁻² per crop (Dedium *et al.*, 2012). When the crop is spaced widely (recommended range of 10-30 heads of lettuce per m² (Licamele, 2009) it receives more sunlight, which in turn, improves the colour and nutrient content of the leaves. The content of nutrients is also dependent on growing conditions, such as temperature (Premuzic *et al.*, 2004), irrigation, cultivation methods and type of crop grown (Nozzi *et al.*, 2018).

The selection of plant species to be used in the use of the aquaponics system can be linked to the stocking density of fish and the subsequent nutrient concentration of effluents from aquaculture. In general, lettuce, herbs, and green vegetables (chives, spinach, basil, and watercress) have low to moderate nutritional needs and are well adapted to the aquaponics system conditions (Diver 2006). Other plants like

cucumbers, tomatoes and bell peppers have a higher nutritional demand and do very well in a highly stocked and nutrient stable aquaponics system (Diver, 2006).

Growing crops using soilless media as it is done in hydroponics is now largely considered by many due to its productivity or performance and its associated benefits (Lennard and Ward, 2019). As we move towards the commercialization of aquaponics in many countries, necessary specifications and quality standards must be maintained to ensure quality vegetable production. Productivity in this system is often likened to that of the hydroponic system in terms of nutrient utilization and general performance of the plants grown (Graber & Junge, 2009; Sjøberg, 2016). The amount of nutrients produced in a fish culture system is often affected by several factors key among which may be the amount of feed given to the fish which relates directly to the density of fish stocked in the system (Rakocy, 2007). Vegetables in aquaponics require a source of energy which can be natural (sunlight) or artificial (electricity) to aid in photosynthesis. The Source of light is essential to ensure maximum diffusion of mineral nutrients. 16 essential mineral nutrients are required by vegetables in aquaponics to ensure a high growth rate, high chlorophyll level and faster maturity. The essential nutrients required are classified into two: micronutrients and macronutrients. Micronutrients are needed by vegetables in relatively low amounts while their counterparts-macronutrients- are needed in relatively large amounts.

Rearing fish below the Carrying Capacity (CC) means that the amount of feed introduced is much lower than when fish are raised at a very high stocking density beyond this capacity resulting in a reduction in the overall fish and vegetable yields due to low nutrient availability. Whereas higher fish feed administration may result in the production of high quantities that may affect the plant growth. The content of

nutrients is also dependent on growing conditions, such as temperature (Premuzic *et al.*, 2004), irrigation, cultivation methods and type of crop grown (Nozzi *et al.*, 2018). The ideal temperature ranges of 15-24°C would be for temperate climate vegetables, but temperatures above 26° C can result in bolting and bitter leaves (FAO, 2014).

The plants are grown in the system tap dissolved oxygen (DO) via its root systems and the requirement for optimal growth and root respiration range between 3-7 mg L⁻¹ (Rakocy *et al.*, 2006; Lennard and Goddek, 2019; Maucieri *et al.*, 2019). Levels lower than 3 mg L⁻¹ interfere with water absorption, lowering nutrient uptake, and hence causing loss of cell tissue from roots resulting in an overall reduction of plant growth. Plant roots and microflora can survive with DO levels that are below 3 mg L⁻¹, whereas most fish require higher levels, mostly above 5 mg L⁻¹. Maintaining such levels of dissolved oxygen in the water used is a prerequisite to optimal plant and fish growth, especially if the water has a high organic load (Yildiz *et al.*, 2017; Maucieri *et al.*, 2019).

Plants grown in the aquaponic system require macronutrients and micronutrients. Three of the essential macronutrients carbon (C), oxygen (O) and hydrogen (H) are supplied by both water (H₂O) and carbon dioxide gas (CO₂). Other macronutrients nutrients, which the plants can absorb are; calcium (Ca), nitrogen (N), phosphorus (P), potassium (K), magnesium (Mg), and sulfur (S). The micronutrients required by the plant include boron (B), zinc (Zn), copper (Cu), chlorine (Cl), iron (Fe), manganese (Mn), and molybdenum (Mo). Iron which is the most crucial can be supplemented by adding chelated iron at the rate of 2mg/litre or by adding aquaponics safe organic fertilizer like seaweed tea or compost (FAO, 2018; Rackocy *et al.*, 2006). Vegetables lacking irons are visibly seen by their leaves turning yellow. Magnesium deficiency in

aquaponics can be supplemented by adding dolomite which is a base and it also works to adjust (increasing) pH. Potassium deficiency is dealt with by the addition of dried and ground banana peels in the grow-bed, potassium chloride or potassium hydroxide and lastly calcium deficiency is dealt with by adding either agricultural limestone (CaCO_3), hydrated limestone or calcium chloride (CaCl_2) (Pattillo, 2014). The presence of these nutrients ensure optimal plant growth, but a higher concentration of some of these nutrients can make others unavailable and also toxic (Turkmen and Guner, 2010; Nozzi *et al.*, 2018). It has been observed for example that excess potassium interferes with the uptake of magnesium or calcium and their inclusion in excess may also interfere with the mentioned macro-nutrients (Nozzi *et al.*, 2018). Ammonium (NH_4^+) derived from the fish component as a result of fish excretory products which build up to toxic levels in the water if not removed from the system. This is made possible by the actions of the nitrifying autotrophic bacteria and nitro-bacteria (Delaide, 2017 and Goddek, 2017). The nitroso-bacteria convert the ammonia present in the water to nitrite before the nitro-bacteria transforms it into nitrate (Goddek *et al.*, 2015).

2.5 Effects of stocking density on the performance in aquaponic systems

Several studies have also demonstrated the effect of stocking density on the welfare of cultured fish (Ellis *et al.*, 2002; Turnbull *et al.*, 2005; Liu *et al.*, 2019). In conventional aquaculture, stocking density affects fish growth either negatively or positively depending on the fish species reared (Rahman, 2015). Culturing fish on small-scale operations has often had a lot of setbacks due to inadequate information on the ideal stocking density of fish to be used (Osofero *et al.*, 2009). In aquaculture, stocking density denotes the intensity at which fish are initially stocked into a system. However, it is generally used to refer to the density of fish at any point in time. Stocking density is a major factor that affects fish growth under cultured conditions (de Oliveira *et al.*, 2012; Andrade *et al.*, 2015; El-Saidy *et al.*, 2015; Millán-Cubillo *et al.*, 2016; Chowdhury *et al.*, 2020). High stocking density results in stress leading to high energy demand. Studies have demonstrated the effect of stocking density on cultured fish welfare (North *et al.*, 2006; Turnbull *et al.*, 2008). These studies have reported that as stocking density increases the various aspects that define the fish's welfare are also affected. Such include stress-related factors, swimming, access to food and many others. The relationship between welfare and stocking density of fish can be influenced by various variables like food availability and accessibility and also water quality-related issues (Ellis *et al.*, 2002; Calabrese *et al.*, 2017).

When fish farmers intensify their operations by increasing the stocking densities of the cultured fish species, the problem of land shortage which is common in many places is away addressed (Khattab *et al.*, 2004; de Oliveira *et al.*, 2012; Policar *et al.*, 2013). Although the relationship between the stocking density and fish survival is not very consistent and sometimes even controversial (El-Sayed, 2002; Trenzado *et al.*,

2018), practically, the densities at which farmers stock their fish are based on experience and institution. It is also important to note that stocking density has a direct impact on the potential feed loss and can limit the access to feeds by the fish especially due to the crowding effect (Boujard *et al.*, 2002; Schmittou, 2006). As fish stocking density increases growth reduces, water quality is compromised unless there is very good biofiltration and recycling system, feed access also decreases and this limits the growth performance of the fish (Schmittou, 2006).

Currently, some farmers have adopted intensive tank-culture systems under greenhouse conditions, using recirculation systems where fish can be raised at extremely high stocking densities. The latter often leads to the production of higher yields as compared to the open pond systems (Timmons *et al.*, 2002; Soto-Zarazúa *et al.*, 2011).

Stocking density, therefore, affects the growth of the fish under culture and it is important to choose the optimal density for better fish (Makori *et al.*, 2017). Different stocking densities have been considered for different fish species in aquaponic systems (Table 2).

Some researchers have evaluated the effects of fish density on growth and food utilization (Jørgensen and Jobling, 1993; Lambert and Dutil, 2001) and also survival (Fatima *et al.*, 2018). Several studies have also demonstrated the effect of stocking density on the welfare of cultured fish (Ellis *et al.*, 2002; Turnbull *et al.*, 2005; Baldwin, 2011; Liu *et al.*, 2019) with a few being linked to aquaponics systems directly. In conventional aquaculture, stocking density seems to affect fish growth either negatively or positively depending on the fish species reared (Rahman, 2015). Stocking densities of 106 to 177 fish m⁻³ (even higher densities of up to 500 fish m⁻³)

have been used for tilapia, 300 to 600 fish m⁻³ for goldfish, and 140 to 280 fish m⁻³ for koi carp (Rahmatullah, *et al.*, 2010; Shete *et al.*, 2013; Hussain *et al.*, 2014).

Table 2: Stocking densities of different fish species in aquaponic systems

Fish + plant species combinations	Optimal fish stocking density	Growth period	References
European Carp+ catalogna chicory, lettuce + Swiss Chard	2.5 kg m ⁻³	20 weeks	Maucieri <i>et al.</i> , 2020
Gift tilapia+ morning glory, <i>Ipomoea reptans</i> , and taro, <i>Colocasia esculenta</i>	106 fish/m ³	15 weeks	Rahmatullah <i>et al.</i> , 2010
Rainbow trout+ Lettuce	3.81 kg m ⁻³	16.7 weeks	Birolo <i>et al.</i> , 2020
Goldfish (<i>Carassius auratus</i>) + spinach (<i>Spinacea oleracea</i>)	500 fish m ⁻³	8.6 weeks	Shete <i>et al.</i> , 2013b
Koi carp + gotukola (<i>Centella asiatica</i>)	2.1 kg m ⁻³	8.6 weeks	Nuwansi <i>et al.</i> , 2021
Tilapia + Indian spinach (<i>Basella alba</i>)	167 fish m ⁻³	8 weeks	Rayhan <i>et al.</i> , 2018
Koi Carp + spinach (<i>Beta vulgaris var. bengalensis</i>).	1.4 kg m ⁻³	8.6 weeks	Hussain <i>et al.</i> , 2014

Studies researching the effects of stocking density on the performance of fish in aquaponics have used tilapia more than any other species of fish (Jørgensen *et al.*, 1993; Lambert and Dutil 2001) and survival (Chakraborty and Mirza 2007; Szkudlarek and Zakes' 2007). Based on extensive studies on tilapia, most of the stocking densities for raising tilapia are affected by the age of the fish at stocking (Endut *et al.*, 2009; Pantanella, *et al.*, 2010; Sace and Fitzsimmons, 2013; Salam, *et al.*, 2014; Filep, *et al.*, 2016). Extremely higher fish stocking densities can be used in aquaponic systems than the recommended densities, such as 30 larvae m⁻² at depths

ranging from 1.8 m to -2.4 m in depth for intensive rearing of climbing perch (*Anabas testudineus*) in earthen ponds in the Mekong Delta, Vietnam (Diem *et al.*, 2017). When raising fingerlings, the stocking densities that have been used range from 1,000 fingerlings m⁻³ to 10,000 fingerlings m⁻³ grown up to table size (250-400g) in ultra-intensive systems with a lot of mechanical aerations. It is worth noting that the growth performance of fish is often influenced by the feeding regime and frequency used, fish interactions and the overall size of the rearing units used (Wahab *et al.*, 1995; Wang *et al.*, 2009). Ntanzi *et al.* (2014) indicated that there was an inverse relationship between the stocking density and growth performance of tilapia. The same relationship has also been reported in several aquaponics trials done by Knaus and Palm (2017), Silva *et al.*(2017) and Rayhan *et al.*(2018).

The relationship between the survival of the fish reared and the stocking density used is not very consistent and sometimes somehow controversial (El-Sayed, 2002). Practically, the stocking densities used by many fish farmers and researchers are based on their experience and intuition. Makori *et al.*(2017) and Yildiz *et al.* (2017) reported that stocking density affects the water quality, which indirectly affects the growth and survival of fish.

2.6 Water quality attributes and nutrient removal in aquaponics

2.6.1 Water quality attributes in aquaponic systems

Fish depend entirely on water to perform all their biological activities. Aquaculture ecosystems are made up of physical, chemical and biological factors that interact collectively and individually to influence culture performance (Schmittou, 2006). The fish excrete ammonia and urine into the water column. The ammonia level also increases due to the accumulation of uneaten food. During protein digestion, ammonia quickly builds up and accumulates in the water adversely affecting the growth of the fish.

Many studies have shown that combination plants and bacteria can efficiently remove or convert ammonia from fish water and thereby maintain healthy living conditions for both the fish and plants (Nichols and Savidov, 2011; Tyson *et al.*, 2011). The key water quality variables related to the culture of *Oreochromis niloticus* and other fish species are temperature, dissolved oxygen (DO) and hydrogen-ion concentration (pH). However, other parameters such as ammonia, nitrates, phosphates, alkalinity and hardness also have significant impacts within these ecosystems (Abolude, 2007).

Temperature is one of the most important environmental variables and a major metabolic modifier in fishes because fish assume approximately the same temperature as their surroundings. It affects their activity, behaviour, feeding, growth, survival, reproduction and efficiency of food conversion (Dupree and Hunner, 1994; Handeland *et al.*, 2008). Temperature impacts tilapia culture in two major ways: firstly the temperature of the water where the fish are located and secondly, the temperature stratification of the water column in which the fish lives (Schmittou,

2006). The ideal temperature for tilapia ranges from 23 to 30°C with the optimal being 28°C.

Dissolved oxygen is the most important parameter, requiring continuous monitoring in tilapia culture systems. Low levels of dissolved oxygen are critical to *Oreochromis niloticus* culture and are responsible for massive fish kills; either directly or indirectly, as compared to all other problems combined (Schmittou, 2006). *Oreochromis niloticus* grows well at DO levels greater than 3 mg/l while lettuce will grow satisfactorily at a DO level of at least 4 ppm. Oxygen naturally enters and dissolves into the water via diffusion at the air-water interface and oxygen-releasing photosynthesis. Diffusion is relatively insignificant unless there is considerable wind action. Low dissolved oxygen is associated with increased ammonia, increase in free carbon dioxide, decreased pH, increased nitrite, increased fish metabolism, increased water temperature, abundant gill parasites and many other factors, which when combined can significantly reduce fish production performances (Schmittou, 2006).

The effect of pH on the chemical, biological and physical properties of water systems make its study very crucial to the lives of the fish and plants grown in the aquaponic system. There is, therefore, a need to regularly monitor the pH levels in both the hydroponic and fish components of the aquaponic system. The ideal pH levels for the culture of tilapia ranges from 6.5 to 8.5. Plants especially lettuce can do very well in pH beginning from 6 up to 8 (Ross, 2000).

Ammonia is the third important respiratory gas after oxygen and carbon dioxide in fishes (Zhang *et al.*, 2011). It is the principal nitrogenous product of fish metabolism that originates from the deamination of amino acids. Culture water is prone to ammonia toxicity if build-up from fish wastes and uneaten feed are not put into check.

Studies have shown that long-term ammonia exposure can hinder fish growth (Hegazi and Hasanein, 2010), cause gill hyperplasia (Benli *et al.*, 2008), liver tissue deterioration (Shingles *et al.*, 2001; Zhu *et al.*, 2020; Mangang and Pandey, 2021) and fish mortality.

Most bony fishes are very sensitive to ammonia toxicity; when subjected to chronic ammonia stress, the fish antioxidant defence system will be damaged (Caruso *et al.*, 2011) thus reducing the body's ability to clear free radicals (Romano and Zeng, 2007). Low levels of ammonia nitrogen have been reported to impact negatively on fish health and growth rate of the fish cultured (Remen *et al.*, 2008) while Chen *et al.*, (2011) reported that the immune response of tilapia (*O. niloticus*) is restrained with exposure to ammonia toxicity.

Although the study by (Chen *et al.*, 2011) probed into the effect of acute ammonia toxicity on the fish immune system; little is known about the long-term exposure of ammonia on fish immune responses (Lemarié *et al.*, 2004). Earlier studies on fish physiological responses to acute and chronic stressors relating to welfare have been reported (Caruso *et al.*, 2008). While acute stress can have different effects on fish; severe stress can have lethal consequences (Maricchiolo *et al.*, 2008).

Water hardness which can be categorized into general hardness and carbonate hardness or alkalinity which is a measure of buffering capacity (FAO, 2014; Sallneva, 2016) is also a very important parameter to look at in aquaponic systems. Hardness can be soft or very hard hardness which compromises the organisms in aquaculture and it's also measured on ppm.

2.6.2 Nutrient removal in aquaponics

These microorganisms are responsible for the breakdown of the uneaten food forming nitrates which are later absorbed by the plants. They additionally assume a few significant roles in recycling aquaculture system (RAS) like influencing nutrients fluxes, diseases, water quality and can be utilized as immediate nourishment for target species (Blancheton *et al.*, 2013). This eventually allows for the removal of undesirable nutrients from the water making it possible for water reuse. The bacteria in the biofilter is the foundation of plant productivity in an aquaponic system for it converts the toxic ammonia present in the water as a result of fish excrete and uneaten feed deposits to nitrate via a process called nitrification. Nitrification is one of the most efficient processes that take place in recirculating aquaculture that has a biofiltration component (Gutierrez-Wing *et al.*, 2012).

The biofilter present in the RAS system is comprised of nitrifying bacteria that live on the system's submerged surfaces naturally and it begins to develop in the presence of ammonia in the culture water. The Nitrosomonas bacteria oxidize the toxic ammonia into nitrite (NO_2^-), whereas Nitrobacter bacteria oxidize nitrite into nitrate (NO_3^-), which is relatively safe for fish (Keuter *et al.*, 2011; Li *et al.*, 2021). The nitrate is then utilized by plants as nutrients. Plants absorb nitrate as well as ionized and unionized ammonia. The resultant nitrates are essential nutrients that the plants require for growth (Marschner, 2003); therefore proper management of the aquaponic system facilitates the thriving of beneficial nitrifying bacteria hence improving the sustainability of the aquaponics system. High ammonia levels usually prevent uptake of nutrients thereby changing the ionic capacity of the water. Plants can absorb excess nitrate from the fish component hence natural filtration by plants takes place in the

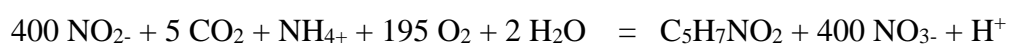
aquaponics system. Nitrite is then oxidized by *Nitrobacter* species to nitrate (NO_3^-) which is absorbed by the growing lettuce plants (Broadly *et al.*, 2003; Velichkova *et al.*, 2019). A mechanical filter removes particulate matter from the fish wastes before being processed by the biological filter. Removal of large solids prior reduces the population of heterotrophic bacteria (Supajaruwong *et al.*, 2021), hence causing minimal competition with nitrifying bacteria thereby converting ammonia to nitrate (Timmons *et al.*, 2002). The nitrification process releases hydrogen ions (H^+) which in turns increases the water pH (Timmons *et al.*, 2002). The nitrifying bacteria determine the status of the aquaponic system. Their absence will make the system toxic resulting in mass fish mortality. A temperature range between 15-30°C, pH ranges between 6.5 to 8.5 and DO levels above 3 mg L⁻¹ provides an ideal environment for the bacteria to reproduce in the aquaponics system (Gichana *et al.*, 2019; Joyce *et al.*, 2019).

Nitrification of 1 mole of ammonia consumes 2 moles of dissolved oxygen (O_2) and yields 1 mole of nitrate, one mole of water (H_2O), and 2 moles of hydrogen ions (H^+) (Bernstein, 2011). The overall reaction of nitrification can be summarized as follows (Haug and McCarty, 1972):

Nitrosomonas



Nitrobacter



This nitrogen transformation removes ammonia from the water column. Nitrate is not harmful to fish except for extremely elevated levels (96-h LC50 > 1000mg/L NO_3^- –

N), and is a primary source of nitrogen for plants in an aquaponic system. Plants especially the leafy ones take in nitrate and ammonium for their growth.

Plant nutrient uptake is an electrically neutral process. Uptake of NH_4^+ may depress the uptake of the essential cations like K^+ , Ca^{2+} and Mg^{2+} . The optimum nitrate to ammonium ratio for vegetables grown in hydroponics is 75:25. The water quality in the aquaponic system should be ideal to support the growth and development of the plants, fish and also bacteria present in the system sustainably. There is inadequate information on the relationship between water quality on nitrifying bacteria and linking this to the conditions present in aquaponic systems (Tyson *et al.*, 2004).

The amount of feed given corresponds to the amount of waste released and nutrients supplied. The feeding ratio used has a direct influence on the number of plants that can be grown (Rakocy, 2007). When fish are fed daily, a continuous supply of nutrients for plant growth is produced. This ensures the leafy plants grow optimally preventing any nutrient build-up by taking up nutrients for growth and in return cleans up the effluents from the fish growing unit. Each square meter of hydroponic growing area in a tilapia-lettuce aquaponic system removes about 0.83 g of total N and 0.17 g of total P per day hence reducing their discharge into the environment (Akter *et al.*, 2018). The nutrient load increases in proportion to the stocking density and feeding rate used.

Approximately 25% of the nutrients and carbon that aquaculture units get from the aquafeeds administered is assimilated as fish biomass (Yogev *et al.*, 2016; Paudel *et al.*, 2020). Yogev *et al.* (2016), reports that about 35% of what is remaining is excreted as total ammonia nitrogen (TAN). The latter is oxidized and made potentially available to plants, whereas the remaining 40% is discharged from the

system as sludge (Yogev *et al.*, 2017). The continuous removal and treatment of organic matter from the aquaponic system help to minimize pollution and associated costs. Solid fish waste can be used as fertilizers in field crops, can be composted or discharged into rivers and streams with or without treatment (Summerfelt *et al.*, 1999). When this sludge is wasted nearly 50% of the available input nutrients that would be used for plant biomass production is lost. Goddek *et al.* (2016) suggested different ways of recycling these nutrients, but more information is still needed if aquaponic systems will be one day largely adopted by farmers.

Endut *et al.*(2009) generally reports removal of Biological oxygen demand (BOD), Total suspended solids (TSS), Total ammonium nitrate (TAN), nitrite-nitrogen, total phosphorus and nitrate-nitrogen by 47-65 %, 67-83 %, 64-78 %, 68-89 %, 43-53 % and 42-65 % respectively when the flow rate range between 0.8- 4 L min⁻¹. Endut *et al* (2016) also reported a reduction of nutrient effluent TAN concentration in a water spinach-based aquaponics by 88.7% from 0.85 to 0.09 mg L⁻¹. The same authors reported a reduction of TAN concentration in a mustard green aquaponics system by 78.2 % to 0.18 mg L⁻¹. In an aquaponic setting involving green mustard and spinach, the concentration of NO₂-N that was originally ranging between 0.02-0.17 mg L⁻¹ was reduced by 92.5 and 86.67 %.

Schmittou (2006) observes that aquaculture ecosystems are made out of physical, chemical and biological components which interact independently and collectively to impact culture performance. The physico-chemical component includes water quality variables such as DO, Temperature, TSS, TDS, ammonia, nitrite, nitrate, alkalinity just to mention a few, whereas the biological component comprises of the fish, plants and bacteria. According to Sidoruk and Cymes (2018), the fish give off ammonia

through gills and urine, including any undigested feeds and fish egesta and excreta that alter the water quality considerably. Fish growth is adversely affected when fish wastes are continuously released in the water causing ammonia to build up in the fish tank (Boyd *et al.*, 2012). The combination of plants and bacteria efficiently removes or converts ammonia from fish water, thereby maintaining healthy living conditions for both the fish and plants (Hu *et al.*, 2015; Rakocy *et al.*, 2016 and Yildiz *et al.*, 2017).

The key water quality variables related to *O. niloticus* and other cultured fish species are temperature (range from 24°C to 26°C), dissolved oxygen (4.86-10.53 mg L⁻¹) and hydrogen-ion concentration (pH) (range from 6.1 to 8.3). However, other parameters such as ammonia (ranging from 0.003 to 0.25 mg L⁻¹), nitrates (ranging from 10-50.7 mg L⁻¹), phosphates, alkalinity and hardness also have significant impacts within aquaculture ecosystems (Shoko *et al.*, 2014; Makori *et al.*, 2017). The organic solids in the form of faecal matter and uneaten food must be removed in the aquaponics system to avoid failure of the system. While other fish species like goldfish and Koi carp can tolerate relatively lower pH ranges (5-9), Tempero *et al.* (2002) suggest that the pH should always be kept close to neutral.

Luo *et al.* (2015) indicate that a basic pH increases ammonia toxicity and acidic pH increased nitrite toxicity in water. When the pH levels are low and high they reduce the nitrification processes. The stocking density of fish has a direct effect on water quality due to the production of carbon dioxide, consumption of oxygen and other metabolites that cause nitrification of the culture water (Yildiz *et al.*, 2017). Water quality is impacted by the feeding rates chosen, water flow and exchange conditions in the system and lastly plant density used in the hydroponics compartment (Makori *et*

al., 2017; Maucieri *et al.*, 2019). These authors indicate the need to monitor changes in water quality parameters in the aquaponic system regularly.

The respiration of the fish, bacteria and plant roots increases the concentration of carbon dioxide concentrations in water. This in turn reacts with water forming weak carbonic water which also lowers the pH of water. The free CO₂ discharged during respiration reacts with water creating carbonic acid, hence lowering the pH of the water. Carbon dioxide rarely causes harmful effects on fish, but higher concentrations prevent the supply of oxygen by lowering the pH of the blood at the gills (Alatorre-Jacome *et al.*, 2012). High carbon dioxide levels do not cause harmful effects when there is an efficient oxygen supply in the water.

Davidson *et al.* (2013) report a deterioration of water quality if partial water exchange and continuous flow rate (of not less than 0.8 L min⁻¹) is minimized. Timmons *et al.* (2002) also reported that as stocking density increases the water quality also deteriorates, hence there is a need to discharge the effluent and replace range it with fresh water at a 5 to 10 % range of the recirculating water per day (Timmons *et al.*, 2002). The level of dissolved oxygen, for example, goes down and this, in turn, affects feed intake and hence fish growth directly. The deterioration of water quality may be a result of the weight of the fish, the volume of the tank and the organic matter that accumulate from the faecal matter of the fish and vegetable residues (Maucieri *et al.*, 2019).

2.7 Economic performance of aquaponics systems

Aquaponics is capital and knowledge-intensive food production technology. Adequate capital and operational funds are a prerequisite when establishing and running aquaponic systems. In developing the aquaponics system, certain important economic points must be considered to make this system profitable. These include; First: the overall capital needed to establish the required infrastructure and buy the equipment needed like pumps, aerators, water test kits among many others (investment cost); Secondly, the annual operational costs; and thirdly, realistic estimates of the prices of fish and crops you intend to grow (Engle, 2015).

The amount needed may vary with the level of intensification or the size of the system used (Engle, 2015). The items to be considered when doing the cost estimates of this system would include greenhouse structure and materials, the tanks, PVC pipes and other accessories, submersible pumps and filtration systems (Rieger *et al.*, 2015; Johnson, 2016).

Economic analyses have been reported for both small research-scale and commercial-scale warm water fish species culture (Jenkins *et al.* 1996; Bailey *et al.* 1997). With realistic estimates based on the amount (kg) of fish to be produced, and the volume and type of crops that can be grown, a clear understanding of the kind of risks that could occur is of paramount importance. Larger aquaponic systems (producing 20,160 heads of lettuce and 1428 kg of tilapia weekly) can have an internal rate of return (IRR) of about 21.7 % whereas a smaller system (5040 heads of lettuce and 357 kg of tilapia) will have its IRR at 9 % (Rupasinghe and Kennedy, 2010).

The IRR, therefore, increases with the increase of both the plants and fish raised in the

system (Bailey *et al.*, 1997). The economic return of any aquaponic system depends on the price of both the plants grown and the fish raised and the IRR can range from 0 to 57 % (Rupasinghe and Kennedy, 2010). Baker (2010) reports a break-even price of a lettuce and tilapia aquaponics system as \$ 3.30 kg⁻¹ and \$ 11.01 kg⁻¹, respectively, indicating a positive economic return.

Goddek *et al.*, (2015) reported that the management of cycling of nutrients especially nitrogen and phosphorus and maintaining recommended pH levels can make aquaponics a viable project. Profitability can be very difficult in some places due to climatic issues (like drought, storms, intense heat among many others), high land prices and strict legislation that do not favour viable aquaponic systems. Other issues that affect the profitability of aquaponics are low prices of vegetables grown, low prices of fish, high inputs of energy and high labour costs. It has been noted that aquaponic systems established in urban settings do very well unlike those in rural and semi-urban settings due to the availability of a ready market (Stadler *et al.*, 2017). Fang *et al.* (2017) further suggest that when aeration is done moderately, the energy cost to produce 1 kg of fish reduce by nearly 44 %. Several authors have reported profitability as marginal (Goodman, 2011; Tokunaga, 2015), net gain only on vegetables but not fish (Love *et al.*, 2015a).

Goddek *et al.* (2015) reported scarcity of data on profitability in large farms because most private companies practising aquaponics are confidential. Dadgupta and Bryant (2017) reported profitability only when big fish and vegetables are sold. Engle (2015) and Rakocy (2012) report profitability only when correct vegetables are grown. English (2015) recommends that an aquaponic system that has lettuce and basil as the main crops makes the whole venture economically viable. Bailey and Ferrarezi *et al.*

(2017) report that prices set by the market forces determine the cost of the vegetable grown in the aquaponic system.

In a survey done among 68 aquaponic growers in Europe, 15.2% reported profitability whereas 71.4 % did not report any income but concluded that this venture can be profitable. The unprofitability of this system was linked to energy costs mainly because of the temperate environment (Villarroel *et al.* 2016). Under such conditions water is heated, air cooling and circulation done, there is also supplemental lighting that increases the variable cost of production. English (2015) agrees with the latter statement on energy costs but recommend that when doing a site survey one should not choose extremely cold regions.

Despite all these outcomes, several challenges are associated with aquaponic systems that include power outages, diseases or parasites which can become very difficult to manage or control (Goddek *et al.*, 2015) and hence can adversely affect the anticipated economic returns. Prevention or control of pests that attack the crops can have detrimental effects on the fish. Most pesticides are very toxic to fish and cannot be used to control pests attacking the plants (Rakocy, 2012; Saraf *et al.*, 2014; Goddek *et al.*, 2015).

Engle (2015) on the other hand recommends an underestimation of the anticipated yields of both plants and fish and a slight overestimation of the costs when developing an aquaponics business plan to buffer any adverse effects brought about by unforeseen circumstances. Costing for labour is crucial because of the deliveries, supervision, repairs and continuous monitoring of the aquaponic system. The overall cost of the system can be measured per every m² and this is directly influenced by how complex the system has designed (Goddek *et al.*, 2015). Tokunaga *et al.* (2015)

gave an estimate of labour costs and pegged it at 46 % of the total operating costs. This will also take at least 40 % of total annual costs. Engle (2015) states that it is not profitable to run an aquaponic system basing on the fish portion only because the cost of fish reared may be less than the market price. Many reports link the profitability of the aquaponic system to the inclusion of the vegetable portion (Rakocy *et al.*, 2012; Engle, 2015; Tokunaga *et al.*, 2015).

Several reports indicate that the fish alone cannot assure profitability of an aquaponics system (Bailey *et al.* 1997; Goodman, 2011; Engle, 2015), but crops like basil and lettuce planted in aquaponic systems can make profitability possible. Love *et al.* (2015a) in a survey on aquaponics established a relationship between the sales of non-food items from aquaponics practising farms and their profitability. In their study, 31% of the respondents interviewed reported profitability receiving between \$1000 and \$ 4,999 within the first 12 months. In the same study, 55 % of the respondents predicted profitability in the next 12 to 36 months.

Savidov (2004) identified lower fish survival rates, nutrient deficiencies at the beginning of the establishment, determination on which crop to grow and not to grow, root rot challenges and lastly how to control water levels to avoid flooding and water wastage as the major problems beginners will face. Savidov (2004) also expressed survey-respondents fears on issues to do with bacterial counts in the water and possibilities of the crops especially the vegetables being affected. He concluded that it will be important for aquaponic farmers not to ignore such concerns because it will affect the demand for aquaponic products. To earn some profit and recover nearly all the expenses including capital expenses fish and plants must be reared continuously. In that, as the farmer waits for the fish to reach market size he or she can do several

planting and harvesting of the crop he or she has chosen. One culture factor that may lead to the recuperation of the operational costs and high capital utilized earning a profit is raising the fish near to their maximum production capacity (Rakocy *et al.*, 2006). Building and equipping a commercial-sized aquaponics greenhouse can cost Kshs 10,000 to Kshs 30,000, contingent upon the aquaponic system design and choice of the different components of the system.

Aquaponics is considered one of the most effective food production systems in terms of the number of products produced per unit volume of water. It takes approximately 500 litres of water to produce Kshs 100 of the product (fish and lettuce), whereas producing cattle takes more than 100 times as much water to produce a Kshs100 of the product (Rakocy *et al.*, 2004b). Goodman (2011) carried out a study of small and medium-scale aquaponic systems using 2,800 litres and two-14,200 litres aquaponic systems to investigate the profitability. It was found that 75% of aquaponics systems researched and evaluated could not break even by selling only fish and vegetable. However, as one creatively diversifies the business model, profits begin to stream in.

Given the overall inadequacy of the economic data and the inconstancy of the profitability matrices utilized in the existing literature, it is not possible to make a clear conclusion on the aquaponics system at this point.

2.8 Application/adoption of aquaponics in sub-Saharan Africa

As an efficient food production system aquaponics has gained attention worldwide (Love *et al.*, 2014, 2015), including in most SSA countries. However, several factors hinder the adoption and development of aquaponics for sustainable food production. Disposal of wastewater is a major concern in aquaculture, Recirculating aquaculture systems are therefore seen as means of reducing the huge volumes of discharged wastewater. Even though the volume of discharge is reduced in RAS, the pollution load (in terms of organic matter and dissolved nutrients per unit of discharge is higher. This may pose a danger to the environment, and as far as RAS is concerned, an additional expense of treating the water may be incurred (Li *et al.*, 2019; Maigual-Enriquez *et al.*, 2019). The introduction of a hydroponic component to recirculating systems is intended to reduce the discharge of aquaculture effluents into the environment, and hence extending water use as well as its conservation.

The ultimate aim of any fish production system is to increase and/or maintain high levels of overall fish growth performance, survival, and good water quality. These will ultimately translate to high yield, improved economic benefits, and maintenance of ideal environmental conditions within the fish culture system. The open aquaculture systems that are commonly practised in many sub-Saharan African countries, including Kenya, often result in low fish growth performance, low economic benefits, and sometimes deterioration of water quality in recipient water bodies (Minoo *et al.*, 2016).

To solve the problem of low production, there have been suggestions for increasing stocking density. Increased stocking density without changing the culture unit size will result in an increased critical standing crop that cannot be supported by the

culture operation (Opiyo *et al.*, 2014). This will result in the deterioration of water quality and impaired fish growth performance. An aquaponic system, which entails recirculating water within the production unit, has been proposed to address these challenges. However being a new technology in many SSA countries, studies on several aspects of the culture system are limited, and not many species of fish are grown. Many studies are also carried only at the experimental level (van Gorcum *et al.*, 2019). A knowledge gap on the appropriate fish stocking density to achieve optimal outputs has hindered the adoption of aquaponics and generally retrogressed development of aquaculture in many SSA countries.

Aquaponics is capital and knowledge-intensive food production technology. Adequate capital and operational funds are a prerequisite when establishing and running aquaponic systems. Even though most SSA countries have placed a lot of investment on food production, and specifically on the growth of Aquaculture, the requisite investment in research on new methods of food production, including aquaponics, is very limited. Thus, there is scarce information and data about many fish species that are needed to maximize production in aquaponics operations. Limited investment in research also extends to limited investment in new methods of food production by farmers. Several studies have deemed profitability in aquaponic systems to be marginal (Goodman 2011; Tokunaga *et al.* 2015), or being a net loss on the fish but a net gain on when vegetables are incorporated (Love *et al.* 2015). This tends to discourage farmers but with the right choice of valuable crops, profitability is assured.

2.9 Knowledge gaps in aquaponics research and practice

Several gaps exist regarding aquaponic research and practice. Below are some of them:

- i) Determining the best/optimal fish stocking density that farmers, researchers and other interested parties can use in closed-loop aquaponic systems for the best growth performance of fish.
- ii) There is a need to know how different densities of fish would affect the growth of plants in aquaponic systems. How do these densities affect the chlorophyll level, weight of the harvested leafy vegetables? The latter being very important since it affects consumer's taste and preferences.
- iii) Since the nutrient derived from the fish component drives the aquaponics systems, establishing an optimal fish density for maximum nutrient production is a prerequisite.
- iv) There is limited data on the profitability of aquaponic systems in regions where it is practised. A survey done in the USA reveals that only 33 % of recirculating systems were profitable, whereas many others still hoped to see positive returns as they continue farming. Several things can be done to increase the profitability of the system, one being increasing the stocking density of the fish while maintaining a constant plant density (Cammies *et al.*, 2021).
- v) More research on fish waste solubilization to transform all added nutrients into plant biomass is needed.

2.10 Conclusions

Although some studies have been carried out in recirculating systems in aquaculture, there has been limited research on the growth performance data of fish species like *O. niloticus* and *C. gariepinus* in an aquaponic system in Sub-Saharan Africa (SSA). Certainly, less information is available for other fish species, despite Africa having one of the most diverse fishes in the world. For aquaponics to grow as a sector of sustainable food production there is a need for investment in the technology and knowledge base of the farmers. Specifically, there is a need to come with cost-effective technologies that can lower the initial cost of setting up the system and its operational costs. Several designs can be adopted or modified using locally available materials, and associated challenges can always have sustainable remedies. Aquaponic systems can easily be powered by solar energy and this will reduce electricity bills and risks especially during blackouts and power surges.

Available data show that aquaponics in many SSA countries is still done at small-scale levels, and this is done also by few people with the most reported studies being experimental. Nevertheless, aquaponics has the potential to address food security and nutritional challenges in many SSA countries. Policies aimed at overall food production and food security need to add more weight to the growth of new technologies for food production under the blue economy mantra. Incentives and knowledge on the concept of aquaponics need to be disseminated among starting practitioners. Moreover, creating the right conditions that better support aquaponics entrepreneurs are needed to attract investors and practitioners.

To foster the adoption and growth of aquaponics in SSA countries, this study recommends the following:

1. Agricultural policies in many SSA countries do not include aquaponics. There is a need for verifiable aquaponics studies to influence the government policy-making process. This will eventually help in funding, credit, and extension support, targeting both new and experienced aquaponic system entrepreneurs (Mchunu *et al.*, 2018).
2. There is a need for investment in research to study all components of an aquaponic system, particularly relating to the various tropical fish and plant species.
3. There is also a great need to establish pest and disease management protocols that are accommodative and non-toxic to fish reared in different aquaponic systems regardless of the design and type used.
4. There is a need to adopt optimal stocking densities to ensure high productivity in both the fish and plant components of the aquaponic system
5. Lastly, the issue of profitability needs to be addressed by focusing, not only on the economic profitability of the system but also on the environmental benefits too (Greenfeld *et al.*, 2019). Furthermore, profitability can be achieved when the right crops are incorporated. The growing of these crops can be staggered or different varieties included

CHAPTER THREE

MATERIALS AND METHODS

3.1 Study Area

This study was carried out at the University of Eldoret in the Aquaponic research unit ($0^{\circ}32' 51.3972''$ N, $35^{\circ}12'16' 11.2044''$ E) at an altitude of 2,140 m above sea level (Figure 2). University of Eldoret area receives a mean annual rainfall of 1124 mm with temperatures ranging between 17°C and 26°C (Chebet *et al.*, 2017). The experiment was set up at the greenhouse, housed by the Department of Fisheries and Aquatic Sciences and located at the Fish Farm.

This experiment was done from February 2017 to April 2017.

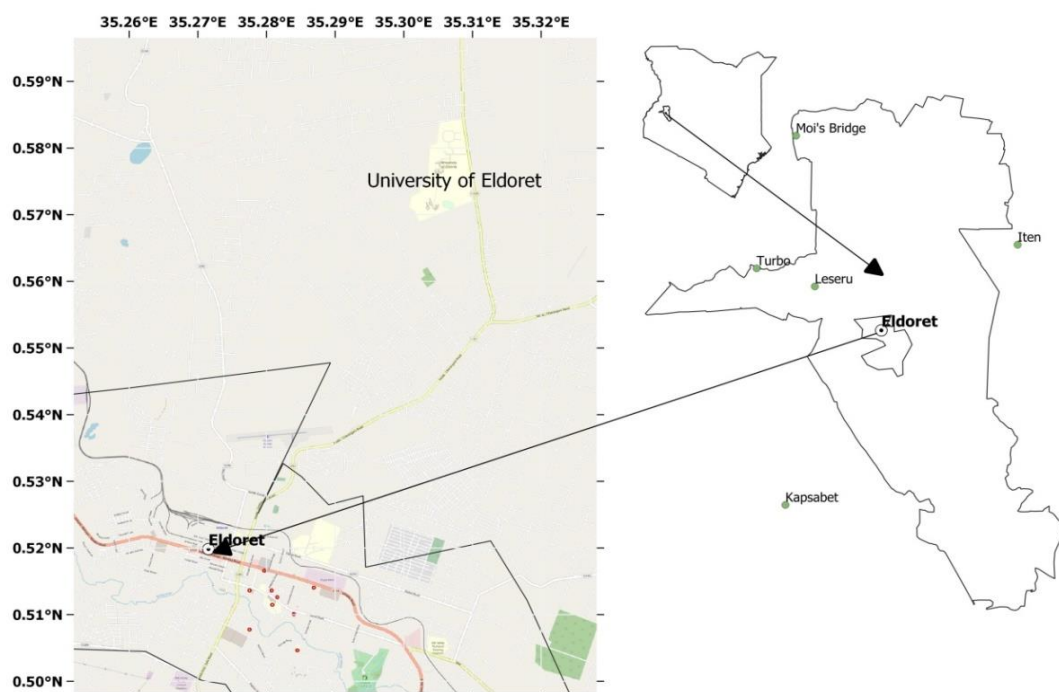


Figure 2: Location of the University of Eldoret in Eldoret town, Kenya

3.2 Source of Nile tilapia brood stock

Nile tilapia *O. niloticus* brood stock was harvested from Lake Victoria. The fish were conditioned in two 50 m² concrete tanks in a nearby fish farm for 3 days. On the fourth day, the fish were put in a 200 L open tank. The tank was filled with water to 75 % capacity. 0.05 % of sodium chloride (common salt) was added to the water to reduce the stress issues associated with live fish transportation. The brooders were carefully put in the water in the tank and transported to Eldoret using an open tank system (Fish were carried in a 200 L open tank using a pickup) via road. On arrival at the University of Eldoret fish farm (Plate 1), the fish were acclimatized to the environment by allowing the ponds water to mix slowly with the tank water as the fish swim out into the ponds.



Plate 1: University of Eldoret fish farm (Ngugi *et al.*, 2007)

After 4 days the feeding commenced where the fish were fed twice a day at 1000 hours and 1600 hours using a feeding rate of 2 % body weight of the fish. The fish were fed on a formulated diet containing 35 % crude protein (CP) twice a day.

3.2.1 Fry production

The brood fish were stocked in two earthen breeding ponds (10 m × 10 m) at a ratio of one male: one female, and after 14 to 21 days of stocking, the females were robbed of all eggs in their buccal cavity and flashed into plastic bowls and immediately transferred to the hatching jars for artificial incubation. The eggs were stocked at a rate of 1,000 eggs L⁻¹. The incubation system was composed of two improvised three-litre MacDonald Zug jar with temperatures controlled by an Eheim Jager Aquarium Thermostat Heater (model number: 3619090 of 300 Watts) which was set at 26°C

while the water flow was set at 2 L min^{-1} . The inflow rate was set by simply adjusting the gate valve to allow water to fill a 2 L calibrated funnel in 60 seconds. The water available was municipal tap water. The municipal water was de-chlorinated by allowing it to settle in a 2000 L lidless reservoir tank for 48 hours. After 48 hours the water was released into the experimental tanks via automated valves.

3.2.2 Sex reversal and nursing of the fry

The incubation of the eggs in the hatching jars lasted for 72 hours. 95% of the eggs hatched. The design of the jars allowed the larvae to follow the flow of water and they were collected in receiving 40 Litre rectangular basins. The larvae were later siphoned into four 200 L rearing tanks ready for the sex reversal process. The tanks were all fitted with thermostat heaters set at 26°C and after 48 hours all the fry had absorbed their yolk sacs. After this, the larvae were feed on a 17α methyl-testosterone impregnated diet for three weeks for sex reversal. The diet (crude protein level of 42 % were prepared within the University using locally available feed ingredients as shown in (Table 5). 6 g of the hormone was mixed with 500 ml of pure ethanol (95 %), to form a stock solution which was put in a vial before refrigeration. 60 mg of this solution mixed with a kilogram of feed. The impregnated feed was put under a shade to allow the alcohol in it to escape. The feed was stored and used for feeding the fry. The fries were fed 6 times daily (0800, 1000, 1200, 1400, 1600 and 1800 hours) at a feeding rate of 10 % body weight. The feeding of the fry lasted for 21 days. All the sex-reversed fry were stocked in two 1 m^2 hapa nets and fed using a hormone-free a rich protein commercial powder of CP 42 % until they averaged 17 gram each. The fingerlings were then transferred to the aquaponic experimental tanks.

3.3 Feed Preparation

The fingerlings were fed with feed formulated at the fish farm containing 35 % Crude protein (CP) and 7 % crude lipid. The feed ingredients included *Omena*, *R. argentea* fish meal, wheat bran, cottonseed cake and rice polish. The solar-dried *R. argentea* were bought from the Mbita market in Siaya County. The exact ingredient proportion of the diet is shown in Table 3, 4 and 5 for fish brooders, tilapia sex reversal feed and the fingerling feed respectively. All ingredients were ground individually into a fine powder using an electrical grinding mill, measured in the respective proportions then mixed and subjected to proximate analysis.

The proximate analysis was determined at the University of Eldoret Fisheries Laboratory following the standard analytical procedure recommended by AOAC (Williams, 1984). Equal proportions of sunflower oil and cod liver oil (1:1) were added as a lipid source in the test diets.

Table 3: Ingredient formulation and proximate composition for the brooders feed

Ingredient	% Inclusion
Wheat bran	35
Freshwater shrimp (<i>Ochong'a</i>)	25
Cottonseed meal	15
Soybean meal	14
Salt (NaCl)	1
Vitamin premix	1
Cassava leaf meal	3
Sunflower oil	2
Blood meal (batch)	4
TOTAL	100
Proximate composition (%)	
Dry matter	90.4
Ash	8.22
Crude protein	35.07
Digestible crude protein	28
Lipid	9
Fibre	6.5
Total n-3	0.8

Total n-6	1.4
Available P%	1.06

Table 4: Ingredient formulation and proximate composition for the sex reversal diet

Ingredient	% Inclusion
Wheat bran	20
Freshwater shrimp (<i>Ochong'a</i>)	36
Cottonseed meal	20
fish meal	22
Salt (NaCl)	1
Vitamin premix	1
TOTAL	100
Proximate composition (%)	
DM%	91.938
Ash%	13.477
CP%	42.00
Dig CP%	35.529
Lipid%	9.991
Fibre%	6.552
Available P%	1.9775

Table 5: Ingredient formulation and proximate composition of the diet used for feeding the experimental fish during the study

Ingredient	% Inclusion
Wheat bran	38
Fresh water shrimps (<i>Ochong'a</i>)	25
Cottonseed meal	17
Soybean meal	10
Salt (NaCl)	1
Vitamin premix	1
Cassava leaf meal	3
Blood meal (batch)	5
TOTAL	100
Proximate composition (%)	
Dry matter	90.27
Ash	8.29
Crude protein	35.03
Digestible crude protein	27.60
Lipid	7.13
Fibre	6.90
LOA (18:2n-6)	0.76
LNA (18:3n-3)	0.05
EPA (20:5n-3)	0.40
DHA (22:6n-3)	0.17
Total n-3	0.62
Total n-6	0.77
Total phospholipid	3.11

3.4 Preparation of the aquaponic system

The aquaponic system consisted of 0.1 m³ recirculating aquaculture and 0.1 m³ hydroponic units, 0.05 m³ improvised columnar filter system (for mechanical filtration), and 0.02 m³ sumps (Figure 3).

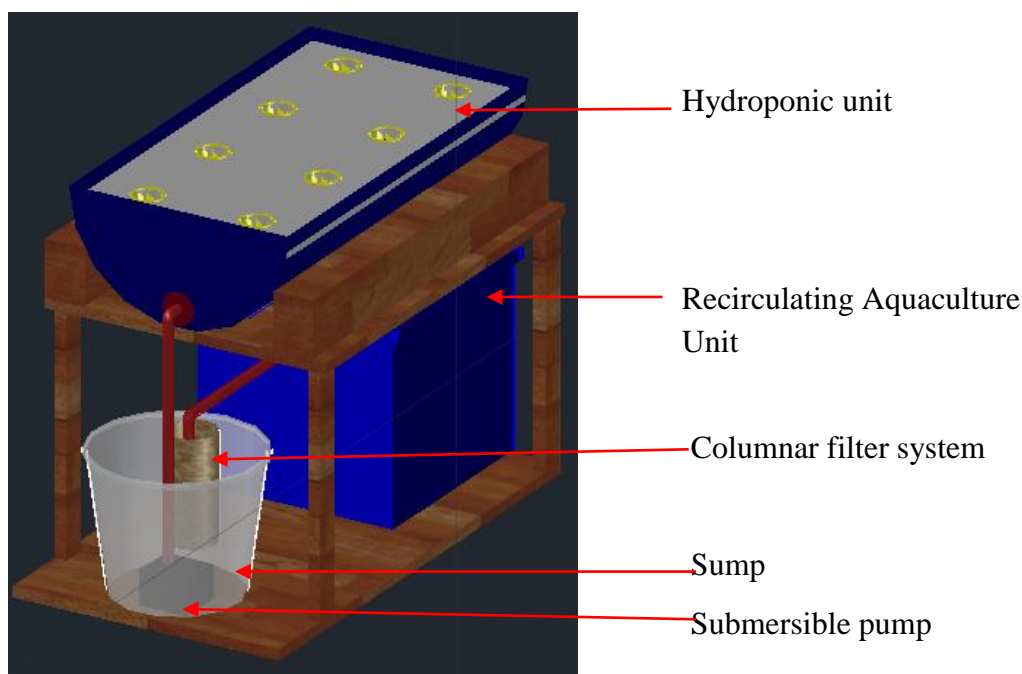


Figure 3: Different sections of the aquaponic system used in this experiment

The system had a 2 m³ reservoir that had de-chlorinated municipal tap water. The flow rate into each system was set at 1.5 L min⁻¹. Fifteen (15) Styrofoam boards of dimensions 1 m x 0.5 m x 0.03 m (length, width and thickness) were used. The boards were placed to float in the hydroponic unit for anchoring the plants. Each board had 8 evenly drilled holes (spacing of 6 inches apart) that were 4 cm in diameter and 1-inch depth. Each of the drilled holes had a plastic plant pot filled with 5-10 mm ballast up to the brim for supporting the plants (lettuce used in the experiment). The pots had 6-9 open strips (Plate 2) to allow plant roots to sprout out and reach nutrients in the hydroponic unit (Figure 4).



Plate 2: Photographs of aquaponic net pots at the research site (Source, author, 2017)

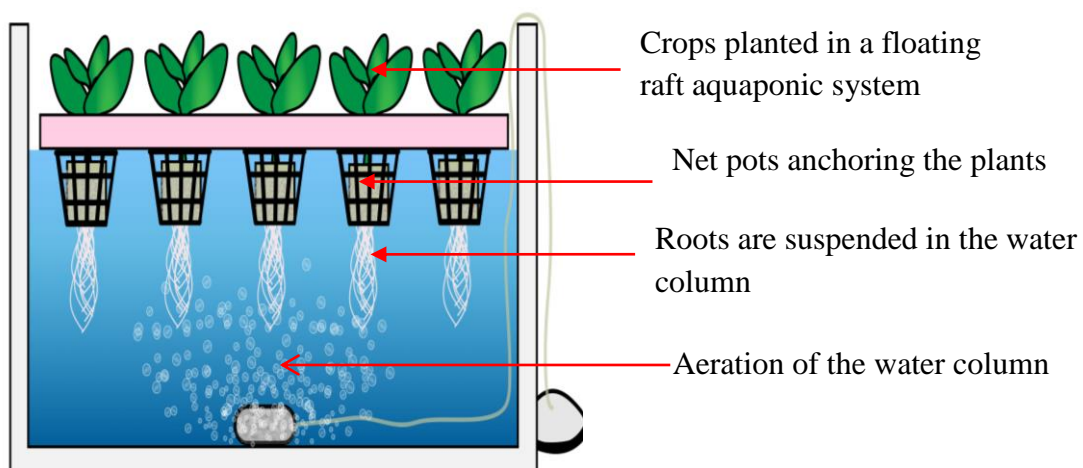


Figure 4: Plants anchored in a net pot in a floating aquaponic system (Source: <http://leafliftsystems.com/2018/06/18/introduction-to-hydroponic-system-types/>)

3.5 Acquisition of lettuce

The lettuce (*L. Sativa*) seeds used in this experiment were sourced from Simlaw Seed Company in Eldoret, Ref. 3405. The seeds were then placed on plastic trays (whose dimensions were 0.5 m by 0.3 m by 0.01 m) placed in a greenhouse. Each tray had a 5 mm thick layered cotton wool covering the entire tray area. The seeds were evenly broadcasted on wet cotton wool and covered using a transparent lid and left to germinate. Every evening the lid was removed and seeds watered by wetting the cotton wool. This was necessary to ensure that there was enough moisture in the wool that is a prerequisite for germination. After 7 days, 99 % of the seeds germinated. The germinated plants were then immediately transplanted into nursery hydroponic system units where they were allowed to grow and fully develop roots for a period of 7 more days. All the healthy plants with well-developed roots were then uprooted and replanted in the hydroponic unit, anchored in the plant pots (Plate 3). After 56 days the lettuces were fully grown and leaves were ready for harvest (Plate 4).



Plate 3: (a) Floating raft for plants, (b) Lettuce seedlings in plant pots (round containers) anchored on floating Styrofoam (Source, author, 2017)

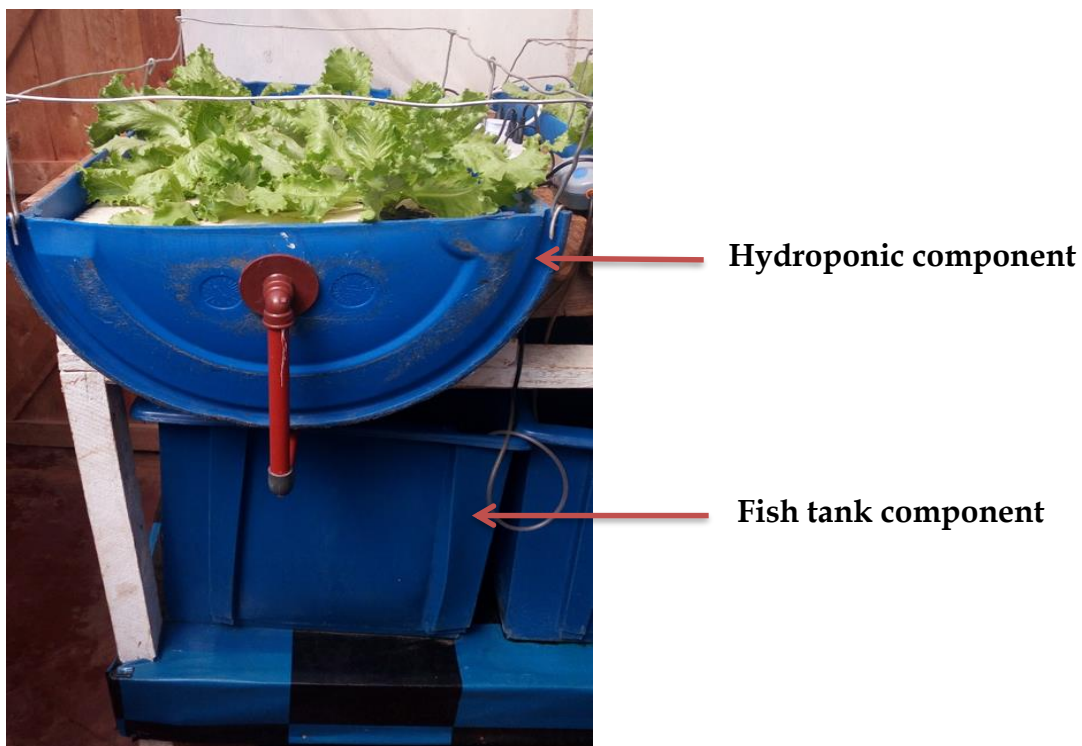


Plate 4: A complete aquaponic system with fully grown lettuce plants (Source, author, 2017)

3.6 Experimental Design

The experimental setup included fifteen plastic rectangular tanks (100 L capacity; dimensions 0.5 m by 0.5 m by 0.6 m length, width, and depth respectively) that were used in a completely randomized block design (CRBD) (Figure 5). There were three aquaponic treatments each stocked with monosex Nile tilapia fingerlings of an average size of 17.9 ± 1.7 g (35 days old). The fish were stocked at densities of 150 fingerlings m^{-3} , 300 fingerlings m^{-3} , and 450 fingerlings m^{-3} for treatments D1, D2, and D3 respectively. These three treatments were replicated five times. Each treatment was being subjected to a 16 lettuce m^{-2} as the planting density (each 0.5 m^2 Styrofoam board had 8 planting holes). The fish were fed to satiation three times a day at 1000 hours; 1200 hours and 1600 hours respectively using a 35 % crude protein (CP) formulated diet. The average amount of feed given was 3.1, 5.9 and 6.5 g/tank/day for densities of 150 fingerlings m^{-3} , 300 fingerlings m^{-3} , and 450 fingerlings m^{-3} respectively.

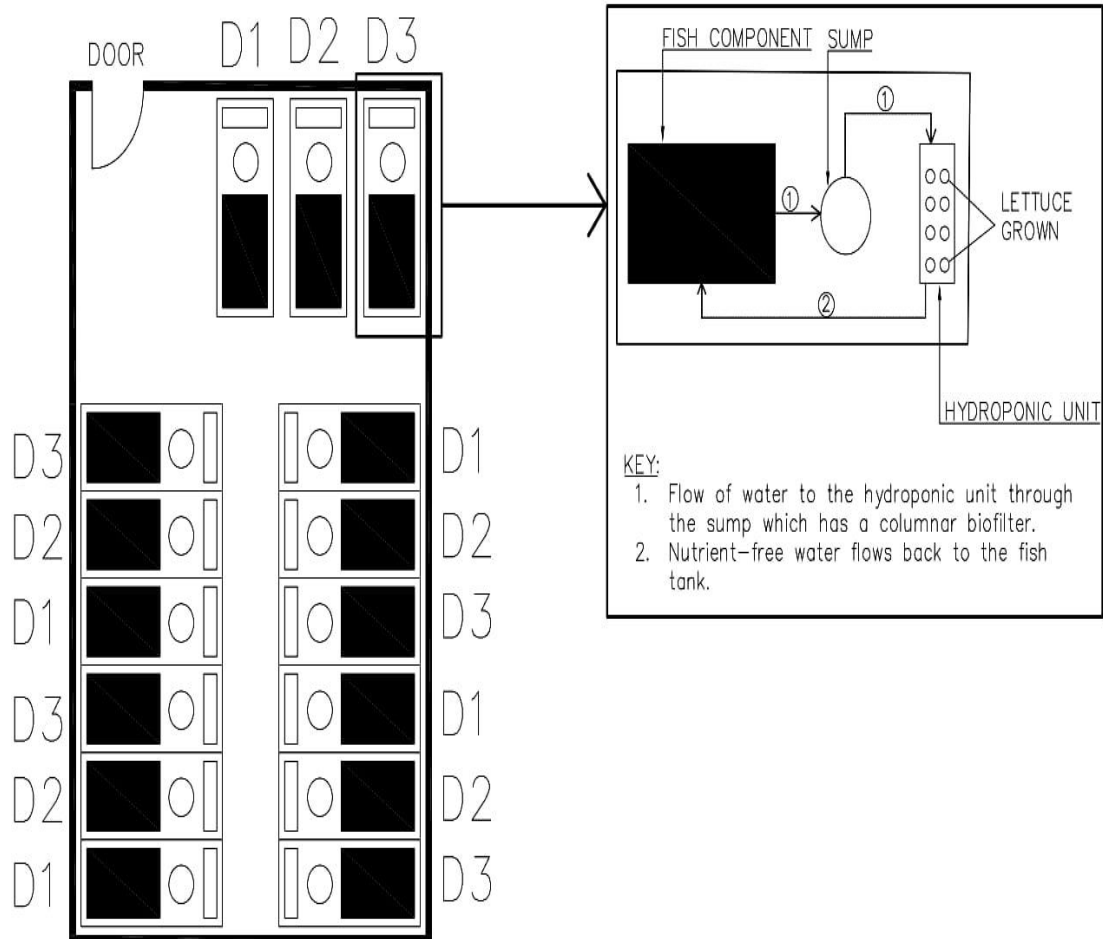


Figure 5: Experimental setup for the aquaponic system. D1, D2 and D3 are stocking densities for *O. niloticus* fingerlings at 150, 300 and 450 fish m⁻³, respectively

3.7 Water quality analyses

Data on dissolved oxygen (DO) concentration, pH, water hardness, and temperature were checked daily in the recirculating fish rearing tank and the hydroponic tank using a YSI 540 DO meter and an EcoSense pH 10 A Pen Tester. Once every week the water samples were collected from the two units for analysis of TAN, nitrates, nitrite, potassium, and soluble reactive phosphorus (SRP) following the methodologies described by the manufacturer using an optical photometer YSI 9500 (YSI Incorporated, Yellow Springs, OH, USA) ($\pm 1\%$ precision)(YSI, I. 2014). The amount of water entering and that leaving the system before and after treatment was maintained at a flow rate of 1.5 L min^{-1} with a help of pre-set automatic gate valves. The water lost through transpiration, evaporation, and periodic flushing was replaced weekly. The water was sampled at the outflow and inflow points of the aquaponic system.

Water quality parameters analyzed included: Dissolved Oxygen (DO, mg/L), pH, Hardness (mg L^{-1}), Nitrates (mg L^{-1}), Nitrites (mg L^{-1}), PO_4 (mg L^{-1}), Potassium (mg L^{-1}) and TAN (mg L^{-1}). All water quality parameters were presented as means \pm SD. Water quality parameters were determined in the recirculating aquaculture system (RAS) and hydroponic. The nutrient removal rate was calculated as:

$$\frac{\text{Nutrient concentration in hydroponic unit} - \text{Nutrient concentration in RAS}}{\text{Nutrient concentration in RAS}} \times 100 .$$

3.8 Fish Sampling and Analysis

A random sample of 10, 10 and 20 fingerlings per tank was collected for weight and length measurements weekly for eight weeks (56 days) for stocking densities 150, 300 and 450 fingerlings m⁻³ respectively. The difference in the number of fish sampled was based on at least 30 % of fish in the tank. This represented 33-66 % of the fish stocked. Each week individual fingerlings were weighed to the nearest 0.01 g with a weighing balance (WJEUIP, Model WA50002Y, W & J Instrument Co. LTD, China) and their lengths were measured using a pair of callipers to the nearest 0.1 mm.

The weekly data on measurements of weight and the daily amount of feed provided were used for calculation the Specific Growth Rate (SGR), Food Conversion Ratio (FCR).

Changes in fish weight were determined using the formula: Final weight – Initial weight while SGR (% BW/D) was determined as;

$$\text{SGR}(\% \text{BW/Day}) = \frac{\text{Ln}W_2 - \text{Ln}W_1}{t} \times 100 ;$$

Where,

W1 = initial weight

W2 = final weight

t = time in days.

The FCR was calculated weekly as the ratio between total feed fed (g) and weight gain (g) for that period. Food Conversion Ratio (FCR) was calculated as:

$$\text{FCR} = \frac{\text{Total feed fed}}{\text{Weight gain}}$$

3.9 Analysis of plant parameters

With the aid of a chlorophyll concentration meter, AtLeaf Plus (resolution ± 0.1 , Chlorophyll Content Index (CCI) unit and repeatability $\pm 1\%$) absorbance of the wavelengths was recorded weekly for each treatment and replicates. The CCI value (comprising of both chlorophyll *a* and *b*) which is proportional to the amount of chlorophyll in the plant leaf was stored in the memory of the meter and later own downloaded into a computer using a dedicated software interface and a data cable. After every three days, the height of individual plants was taken. The height was measured from the Styrofoam surface to the top of the main plant stem. The number of leaves for each plant was also counted, including the tips of newly emerging plants. At the end of the experiment, the plants in the floating boards were carefully removed, washed slightly and blotted using a soft paper towel to remove all the surface moisture attached to the plant. The individual plants were then weighed immediately using an electronic balance (readability 0.01 mg, model VI-200) and the data recorded. The data on the growth performance of the lettuce were also recorded i.e. the height of the plant, number of leaves, length of the roots, CCI and final dry weight of the harvested plant (Plate 5).



Plate 5: A section of the experiment with fully grown lettuce (Source, author, 2017)

3.10 Economic analysis data

The cost of installing the 15 aquaponic units as presented in the purchase receipts was recorded in an excel sheet. The total weight of the fish and lettuce and the survival data were used to calculate the total biomass. The market price for fish and lettuce was set at Kshs 400 and 300 respectively. All the expenses incurred including the cost of inputs, labour, repairs, bills were documented. The revenues derived from the sale of both fish and lettuce were also documented and used for the economic analysis of the aquaponic system.

During economic analysis, the costs of the aquaponic unit were calculated to get the overall capital outlay for the aquaponic unit. Fish and plant yields were computed based on the overall fish and plant biomass at harvest and the total revenue earned determined by multiplying the biomass and the market price at the time of the study. An enterprise budget was utilized to decide on income, expenses and returns of the aquaponic system under different stocking densities. The profitability of the venture was examined utilizing the net returns above variable costs. The break-even price for the enterprise was calculated using the formula.

$$\text{Breakeven price} = \frac{\text{Fixed cost per unit}}{1 - (\text{Variable cost per unit} / \text{Selling Price per unit})}$$

A sensitivity analysis was used to reenact the net returns due to variation in alternative market prices and yield variability on breakeven prices.

3.11 Data Analysis

All the data on fish weight and length, plant weight, height, number of leaves as well as water quality parameters (temperature, dissolved oxygen, nitrates, ammonia, nitrites and phosphates) were entered in MS Excel according to the treatment to facilitate processing for statistical analyses. Growth was determined using changes in mean weight and specific growth rate (SGR).

The relationship between body weight and SGR against time was demonstrated by regression analysis of the form $\text{Weight/SGR} = \beta_0 + \text{Weight} \cdot \text{Time}$; where β_0 is a constant. A test for the common slope was used to compare coefficients in regression equations.

Differences in the FCR were determined using One Way ANOVA. The survival rate was calculated based on the number of fish alive as a percentage of the total stocked fish after every 7 days. Differences in the survival among treatments were analyzed using survival analysis trends.

Bivariate relationships between individual water quality parameters were analyzed using Persons Correlation. Meanwhile, the interrelationships among the water quality parameters were analyzed using Factor analysis.

Generalized linear mixed-effects models (GLEMs) was used to test the effect of stocking density on water quality variables pH, water hardness, nitrite (NO_2^-) and nitrates (NO_3^{2-}), SRP, potassium (K) and TAN (NH_4^+) with the `lme` function in the *nlme* package in R (Pinheiro *et al.* 2016, R Core Team 2019). Similar to water quality variables, GLEMs were used to test the effect of stocking density on fish growth using length, weight, SGR and FCR as response variables. GLEM was used instead

of Generalized Additive Models (GAMs) after residuals demonstrated largely linear responses to fish stocking density. For each response variable, the GLEM models included stocking density (150, 300 and 450 fingerlings m^{-2} ; categorical variable) and time (week 1- week 7) as fixed effects, and tank as a random effect to test whether the position of the tanks affected water quality and fish growth. Time was and its interaction with stocking density (stocking density \times time) as fixed factors. The model setup included the following equation; Response~Stocking density * Time, random = ~1| Tank, where the response variables included water quality and growth parameters. A separate model for each variable was run with the 8 observations (7 weekly measurements, including day 1).

An initial GLEM ‘full’ model was fitted that included fingerlings stocking density and time as fixed effects, and ‘tank’ as a random effect with Poisson distribution and a log link function (Bolker *et al.* 2009). The distribution of all response variables and their residuals were inspected for normality using q-q plots and histograms. To identify the most parsimonious model including only significant predictor variables for water quality and fish growth, a step-wise ANOVA approach based on the Akaike Information Criterion (AIC) was used to achieve an optimal model that explained most variation without the random effect and interaction, with the lowest AIC among non-significant models indicating the best model (Burnham & Anderson, 2004). For each model, a marginal R^2 (R^2_{m} , variance explained by fixed factors) was computed and conditional R^2 (R^2_{c} , variance explained by the entire model, i.e. by fixed and random factors) coefficients with the ‘r.squared GLMM’ function in the *MUMIN* package (R Core Team, 2019). All analyses were conducted with an alpha of 0.05.

The plant final biomass, shoot length, root length and total chlorophyll were presented as means \pm SD and their treatment differences analyzed using One Way ANOVA. Median values were determined on the data collected for the number of leaves counted every week. Differences in the number of leaves in the stocking density treatment were analyzed using the Kruskal Wallis test. Boxplot with whiskers for the three stocking density with plant weight, shoot length, root length and number of leaves, including weekly plant height per stocking density were generated using R statistical software. Multiple linear regression of the number of leaves versus plant weight to compare slopes at different stocking density was conducted using Statgraphic Centurion XVI software and finally bar graphs generated with Statistica 8.

During sensitivity analysis, The Excel Package, What-if Analysis, 1 way and 2-Way Table was utilized to investigate the variability of single and combined variables respectively affecting the yield variability or break-even prices accordingly. Cash flow projections were based on the aquaponic stocking densities and were computed using Net Present Value analysis and Internal Rate of return as detailed elsewhere (Bailey *et al.*, 1997).

CHAPTER FOUR

RESULTS

4.1 Effects of stocking density on fish growth performance

Fish growth data at different stocking densities in the aquaponics during the 8 weeks are shown in Figure 6. Fingerlings stocked at 150 fish m⁻³ grew from 17.9±1.7 g to 42.6±3.1 g, while those stocked at 300 fish m⁻³ grew from 18.2±2.2 g to 32.0±3.8 g, while fish stocked at 450 fish m⁻³ grew from 18.2±1.9 g to a weight of 25.2±4.2 g. The growth data were subjected to weight-time regression and the following relationships were obtained: The equations; (i), (ii) and (iii) show how weight changes with time.

- i) 150 fish m⁻³: Weight = 3.2767*Time + 15.345 (R² = 0.8931)
- ii) 300 fish m⁻³: Weight = 1.9645*Time + 16.279 (R² = 0.8307)
- iii) 450 fish m⁻³: Weight = 1.0175*Time + 17.794 (R² = 0.6262)

Where; R² is the coefficient of determination which shows that the models are reliable to explain the relationships in the data. There was a significant effect of stocking density on fish growth (Figure 6 and 7, Table 6). Fish reared at 150 fish m⁻³ at a significantly greater rate ($p < 0.05$) than did the growth at 300 fish m⁻³ and 450 fish m⁻³. At the end of the experimental period (56 days), fish stocked at 150 fish m⁻³ weighed approximately 33 % heavier than fish stocked at 300 fish m⁻³ and 70 % heavier than fish stocked at 450 fish m⁻³. Meanwhile fish stocked at 300 fish m⁻³ weighed approximately 27 % heavier than fish stocked at 450 fish m⁻³.

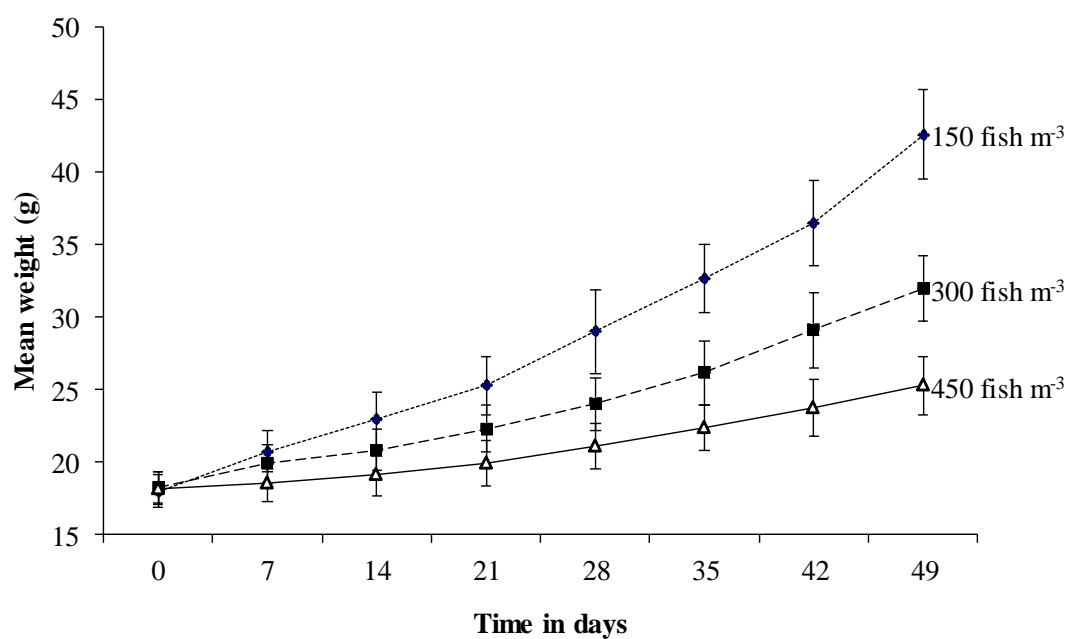


Figure 6: Growth of *O. niloticus* fingerlings in terms of weight under different stocking densities in the aquaponic system. Dotted line = 150 fish m⁻³, dash-line = 300 fish m⁻³ and solid line = 450 fish m⁻³

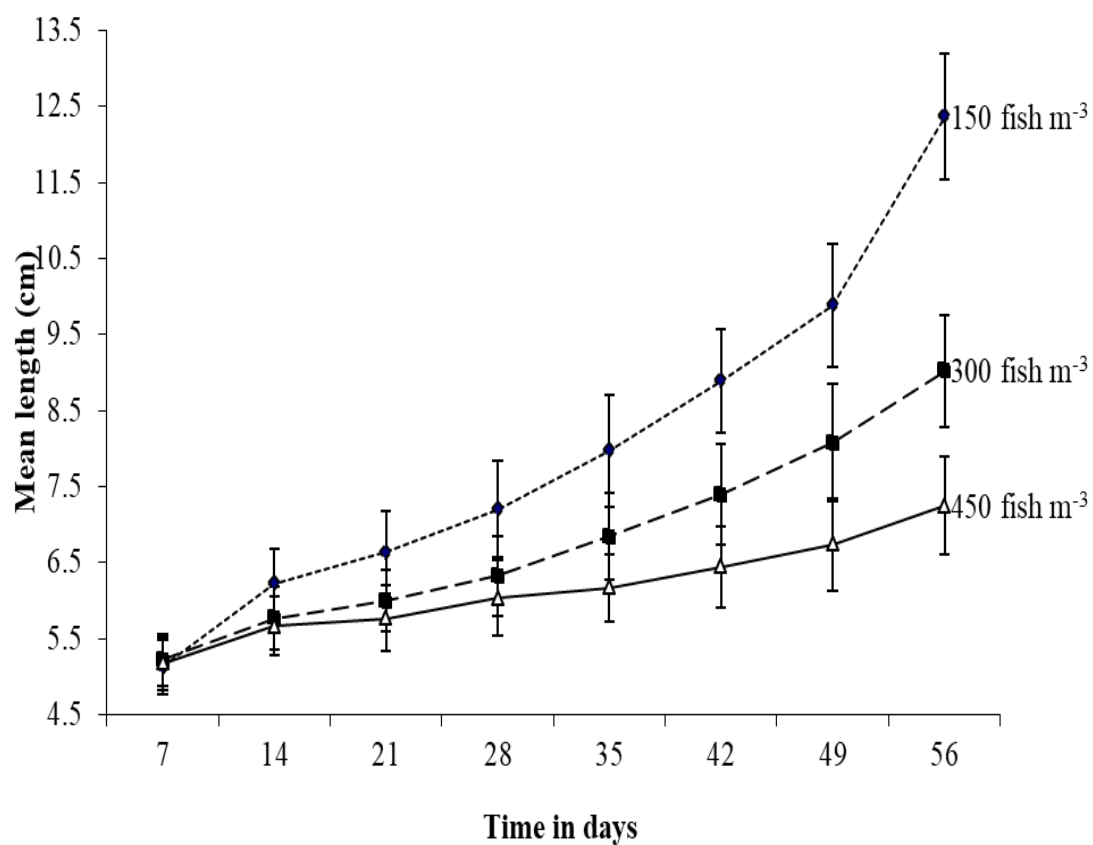


Figure 7: Growth of *O. niloticus* fingerlings in terms of length under different stocking densities in the aquaponic system. Dotted line = 150 fish m⁻³, dash-line = 300 fish m⁻³ and solid line = 450 fish m⁻³

Table 6: Results of generalized mixed-effects models for fish length and weight over the experimental period

	Fish length	Fish weight	SGR	FCR
Fixed effects	β (SE) t-value	β (SE) t-value	β (SE) t-value	β (SE) t-value
Intercept	4.51 (0.14) 32.5***	15.84 (0.52) 31.1***	2.74 (0.15) 18.6***	1.55 (0.02) 94.5***
Stocking density	0.22 (0.03) 6.6***	0.79 (0.12) 6.5***	-0.60 (0.07) 6.6***	0.14(0.01) 18.6***
Time in weeks	1.22 (0.02) 69.2***	4.24(0.06) 66.9***	-0.03 (0.004) -6.1***	-0.01(0.001)-20.9***
Stocking density X Time	-0.33 (0.01) -40.0***	-1.12 (0.03) -39.3 ***	0.005 (0.002) 2.4*	0.002 (0.0002) 9.1***
ANOVA for fixed effects	F-value	F-value	F-value	F-value
Intercept	3456.2***	3103.2***	1671.2***	346903.5***
Stocking density	2361.9***	2136.4***	218.2***	3517.8***
Time in weeks	1735.0***	6986.8***	106.2***	1094.5***
Stocking density X Time	1598.3***	1464.5***	5.5*	82.9***
Random effect				
Tank (intercept) SD	0.26	0.97	<0.001	<0.001
Residual SD	0.65	2.31	0.28	0.03
$R^2_{GLMM(m)}$	0.84	0.83	0.73	0.97
$R^2_{GLMM(c)}$	0.87	0.86	0.73	0.97

Note: The ‘full’ model included stocking density (150, 300 and 450 fingerlings m⁻³), time in weeks and a stocking density × time interaction as fixed effects and tank as a random effect. The marginal R² (GLMM[m]; fixed effects only) and the conditional R² (GLMM[c]; fixed and random effects) represent the proportion variance explained by each model. SE = standard error; SD = standard deviation; *p < 0.05, **p < 0.01, ***p < 0.001.

To facilitate direct comparison between fish growth at different stocking densities, SGR was calculated between each weight collection of fish in different treatments (Figure 8). Generally, SGR decreased with the growth of fingerlings over time, with the greatest decrease recorded in fingerlings stocked at 150 fish m⁻³. Based on the analysis of the rate of change, fish stocked at 150 fish m⁻³ grew faster than fish stocked at 450 fish m⁻³ and fish stocked at 300 fish m⁻³. Data were subjected to linear regression analysis and the following relationships were obtained: The equations; (i), (ii) and (iii) show how the specific growth rate (SGR) changes with time. The relationship is negative and the slope is very steep.

- i) 150 fish m⁻³: $SGR = 2.4008 - 0.0299 * Time$ ($R^2 = 0.9882$)
- ii) 300 fish m⁻³: $SGR = 1.7984 - 0.0227 * Time$ ($R^2 = 0.9882$)
- iii) 450 fish m⁻³: $SGR = 1.1828 - 0.0194 * Time$ ($R^2 = 0.9882$)

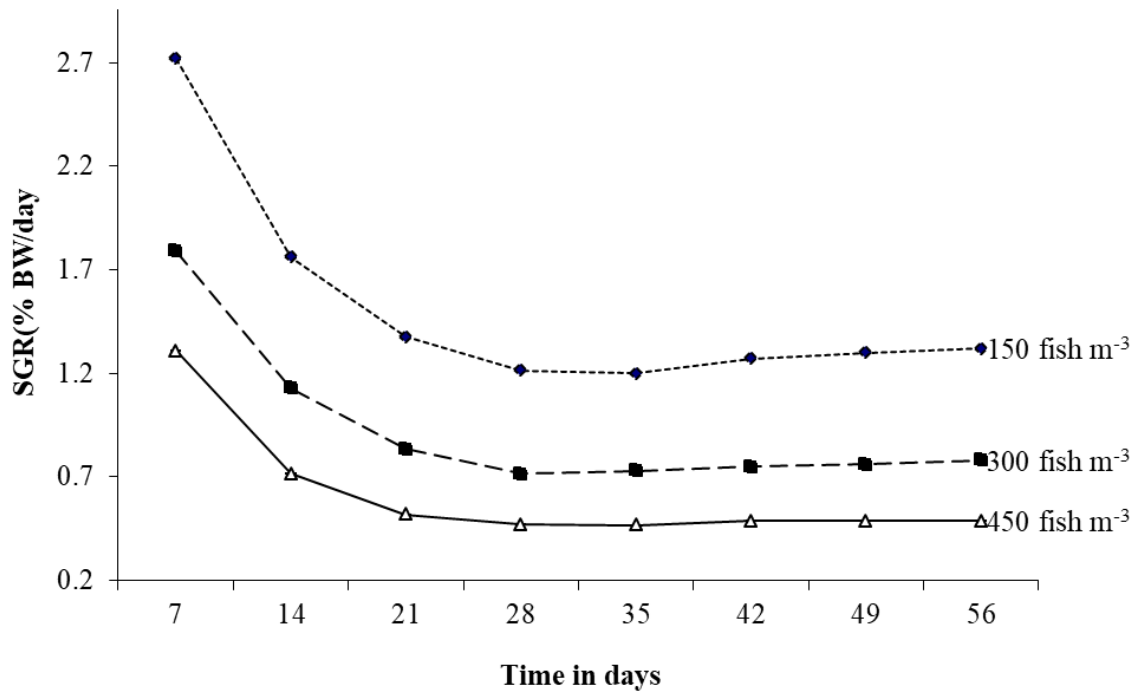


Figure 8: The SGR of *O. niloticus* fingerlings reared under different stocking densities.

Dotted line = 150 fish m⁻³, dash-line = 300 fish m⁻³ and solid line = 450 fish m⁻³

There were differences in the survival of fingerlings across treatments over the experimental period (Table 7). At the end of the study period (56 days), survival was significantly highest ($H = 13.445$, $df = 2$, $P = 0.0002$) at stocking density 150 fish m^{-3} ($93.85 \pm 2.11 \%$), followed by fish stocked at density 300 fish m^{-3} ($89.73 \pm 2.35 \%$), which was similar to fish stocked at 450 fish m^{-3} ($82.05 \pm 2.9 \%$).

Table 7: Percent (%) Survival of *O. niloticus* for 56 days reared under different stocking densities

Days	150 fish m^{-3}	300 fish m^{-3}	450 fish m^{-3}
0	100	100	100
7	98.7	97	95.4
14	96.9	94	90.1
28	93.2	88	79
35	91.7	85.7	74.8
42	90.5	84.7	73
49	89.9	84.2	72.1
56	89.9	84.2	72

On the contrary, the highest stocking density of fish had the highest FCR (Figure 9). The FCR was significantly higher at a stocking density of 450 fish m⁻³ followed by fish stocked at density 300 fish m⁻³ and least in fish stocked at 150 fish m⁻³. By the end of the 56 days, the FCR was 1.45± 0.13, 1.66± 0.1 and 1.86± 0.07 for 150, 300 and 450 fish m⁻³ respectively. The lowest calculated FCR of 1.45± 0.13 was the best but generally, all the treatments had FCRs below 2. This range is ideal for aquaponic systems and general aquaculture.

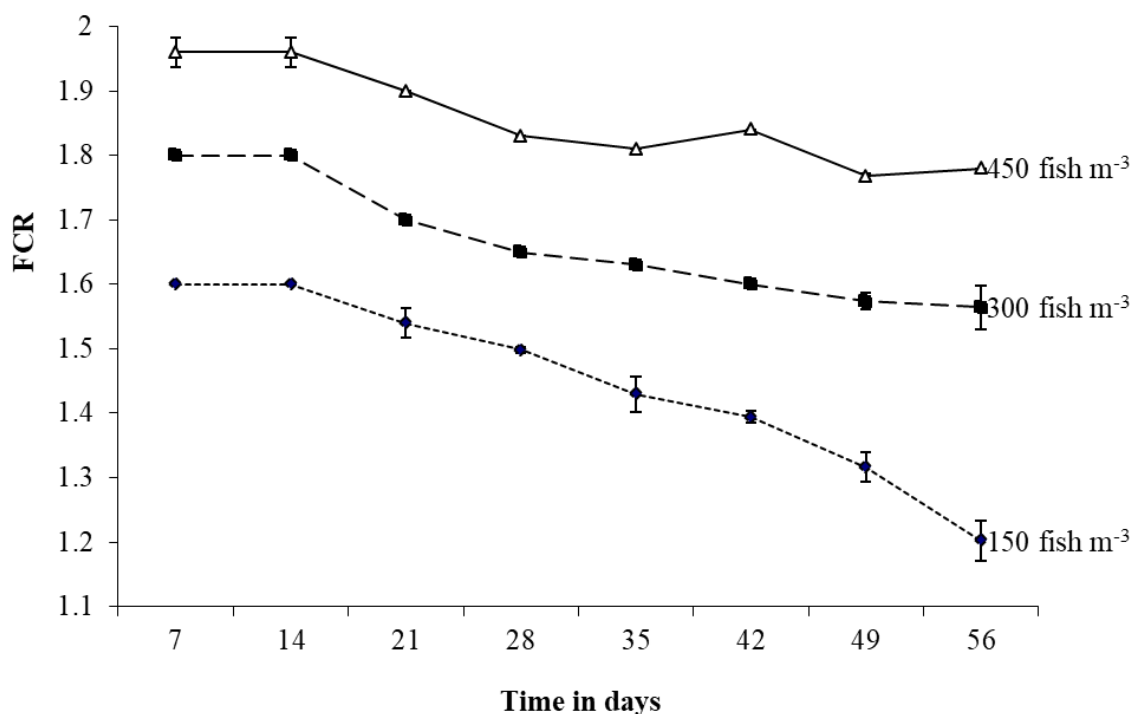


Figure 9: The FCR of *O. niloticus* fingerlings reared under different stocking densities.

Dotted line = 150 fish m⁻³, dash-line = 300 fish m⁻³ and solid line = 450 fish m⁻³

4.2 Effects of fish stocking density on *Lactuca sativa* growth performance

The growth parameters of *L. sativa* in terms of the biomass of the lettuce, their shoot length, root length and the number of leaves as a function of the stocking density of fish (Table 8). Production in terms of total biomass ranged from the lowest value of 166.4 ± 9.8 g in treatment stocked at 150 fish m^{-3} , to the highest 304.6 ± 23.2 g in treatment stocked at 450 fish m^{-3} . Generally, the production in terms of total production, yield and mean per plant of *L. sativa* increased with increasing stocking density. Production of *L. sativa* increased with the increasing stocking density of the fish. A similar trend as those of production was observed for yield kg^{-1} and mean weight per plant with respect to the stocking density of fish. The total chlorophyll concentration in the lettuce leaves also increased with increased stocking density as indicated in the table below.

Table 8: Biomass and growth parameters of *L. sativa* based on the stocking density of *O. niloticus* in aquaponics (data are presented as means \pm SEM)

Growth parameters (Average)	Stocking density (fish m^{-3})		
	150	300	450
Biomass (g)	166.4 ± 9.8^a	276.8 ± 23.2^b	304.6 ± 23.2^c
Shoot length (cm)	22.6 ± 1.2^a	26.9 ± 0.9^c	24.9 ± 0.9^b
Root length (cm)	27.2 ± 1.9^a	39.6 ± 1.7^c	33.9 ± 1.7^b
Number of leaves (\pm median)	20 ± 2.1	21 ± 1.7	22 ± 1.9
Total chlorophyll ($\mu\text{g L}^{-1}$)	0.019 ± 0.002^a	0.056 ± 0.007^c	0.032 ± 0.004^c

Values in the same row with different superscripts are significantly different ($p < .05$).

Lower case letters indicate significant differences among the treatments

Box plot was used to explain the relationship between the fish stocking density and the different plant yield measurements as shown in figures 10 (a-d) below. Plant weight showed very little variation at the stocking density of 150 fish m^{-3} but varied as the density increased from 300 fish m^{-3} to and 450 fish m^{-3} . The plant weight was between 100 (g) and 700 (g) figure 10 a. The Shoot length (15-37.5 cm), root length (20-60 cm) and the number of leaves (15-28) did not vary much when the three stocking densities were compared, Figure 10 (b,c and d) respectively.

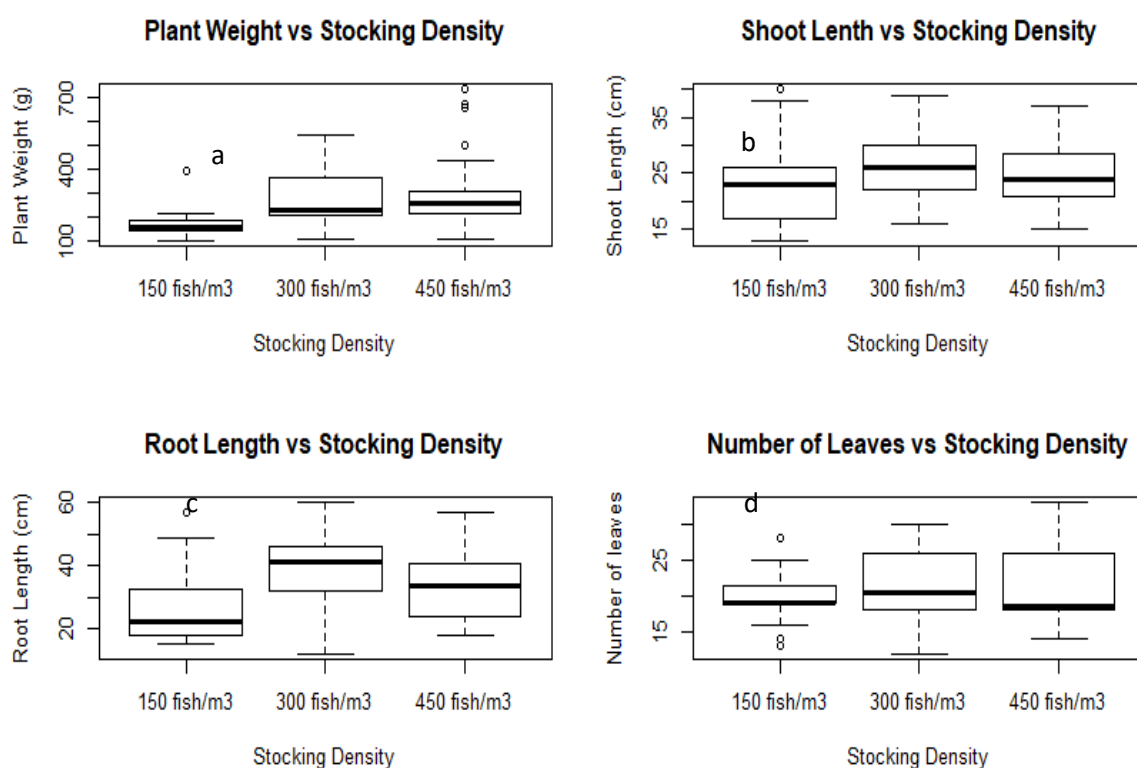


Figure 10: (a-d); Box plot Fish Stocking density vs. (a) plant weight, (b) shoot length, (c) root length and (d) number of leaves

Box plot on weekly variation in plant height at the three stocking densities was as shown in figure 11 (a-c). At stocking density 150 fish m^{-3} (figure 11 a), significant variation was observed between week 3 and week 4 where the whiskers, first and the third quartile were

very different comparing the two weeks. This was also observed between week 5 and week 6 through the final plant height in the whole duration did not exceed 130 mm. The Stocking density of 300 fish m^{-3} (figure 11 b) also demonstrated significant growth and variation in plant height from week 3 when comparing whiskers, first and the third quartile. This was observed between week 3 and 4, week 4 and 5 and between week 5 and 6. The final plant height exceeded 160 mm. Finally, at stocking density 450 fish m^{-3} (figure 11 c), variation in growth by comparing whiskers, first and third quartile started from week 2. Plant height at this stocking density was also generally longer than the other two stocking densities from the second week through to the last week. The first quartile of plant height at week 6 was above 200 mm and some of the final heights were above 220 mm. Comparing the whiskers, first and the third quartile at the last week, plant height at stocking density 450 fish m^{-3} was longer than the other two stocking densities.

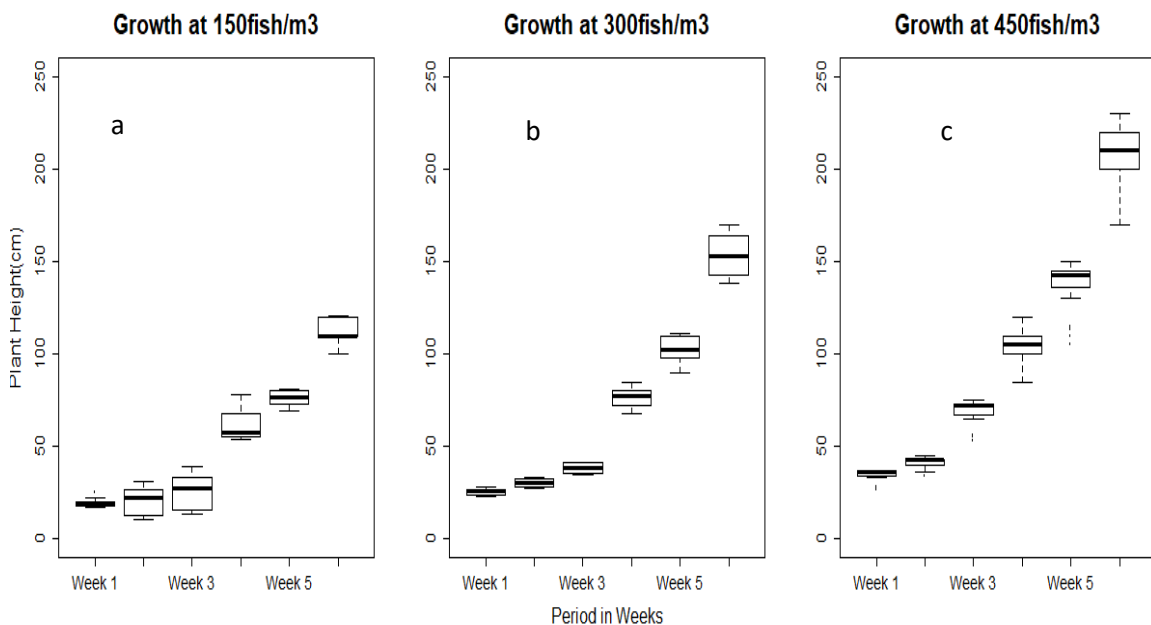


Figure 11: (a-c) Weekly variation of lettuce's height (cm) at (a) 150 fish m^{-3} (b) 300 fish m^{-3} and (c) 450 fish m^{-3}

The final mean plant weight per stocking density was significantly different ($p < 0.05$) at 150 fish m^{-3} (mean weight 166.4 ± 9.8 g) as compared to the other two stocking densities. At 300 fish m^{-3} , the mean weight was 276.8 ± 23.2 g while at 450 fish m^{-3} the mean weight was 304.6 ± 23.2 g (Figure 12). This means that at higher stocking densities the mean weight of harvested lettuce also increased.

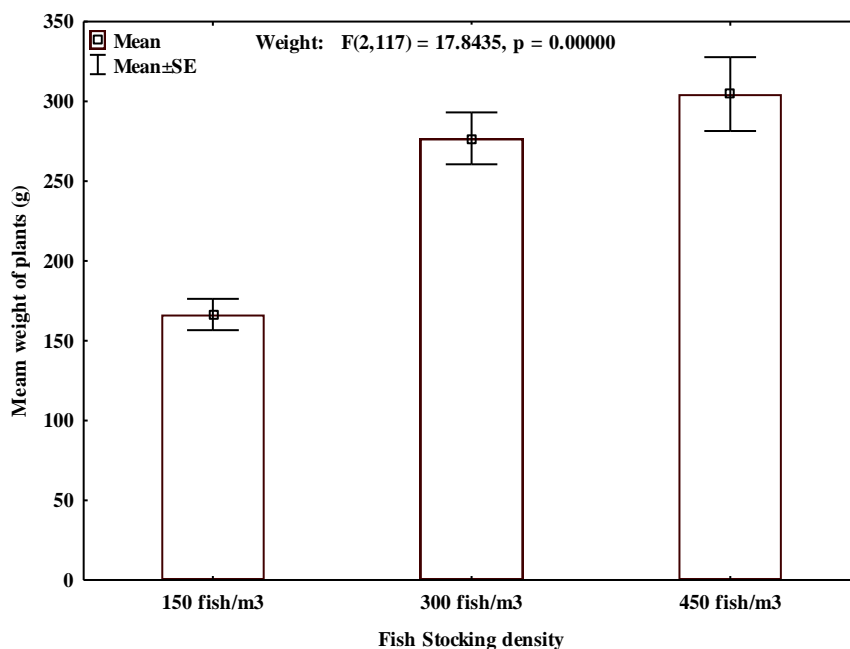


Figure 12: Fish stocking density vs. plant weight under different stocking densities

Multiple linear regression compared slopes for the number of leaves at the three stocking densities with plant weight (Figure 13), and r^2 and adjusted r^2 of 63.2 and 61.6 % respectively. This relationship signified that the number of leaves increased with an increase in plant weight. Comparing stocking density 150 fish m^{-3} and 300 fish m^{-3} , the slopes were not significantly different, instead, they were almost parallel (12.3 and 12.5 respectively) only that at stocking density 300 fish m^{-3} , the plants were heavier at the

same number of leaves as compared to stoking density 150 fish m⁻³, meaning that the leaves were heavier (though the same number) at stoking density 300 fish m⁻³ than 150 fish m⁻³. However, stocking density 450 fish m⁻³ had a significant slope (slope = 19.6 and $p = 0.001$) which was also steeper than the other two stocking densities, thus inferring more leaves and heavier plants than the previous two stocking densities hence better yield number of leaves and plant weight).

Multiple linear regression equations were:

- i) At 150 fish m⁻³ the model reduces to Plant Weight (g) = -76.8394 + 12.3634*Leaves
- ii) At 300 fish m⁻³, the model reduces to Plant Weight (g) = 13.9526 + 12.5458*Leaves
- iii) At 450 fish m⁻³, the model reduces to Plant Weight (g) = -132.859 + 19.6837*Leaves

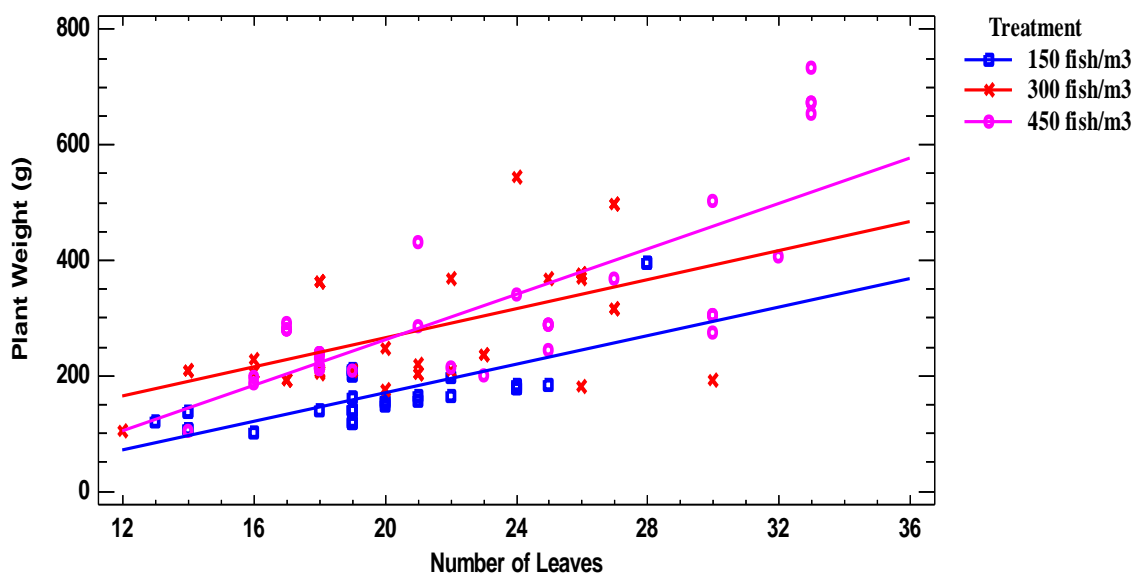


Figure 13: Number of leaves vs the plant weight under different stocking densities

4.3 Effects of stocking density on water quality

Stocking density had a significant effect on alkalinity, nitrite, nitrate, SRP, potassium (K) and ammonium in the RAS and hydroponic components (Table 9 and 10). The GLMM model was performed for the RAS component only because this had a direct effect on fish condition and growth. Stocking of fish affected most water quality variables giving significant responses. The RAS component recorded higher levels than the hydroponic unit for almost all variables, indicating the ameliorating effects afforded by the lettuce. Nutrients were particularly lower suggesting uptake for plant growth.

The water quality parameters in the aquaponics system stocked with *O. niloticus* at different stocking densities are provided in Table 9. The water quality parameters were DO: 4-7 mg L⁻¹; pH = 6.3 to 7.3; alkalinity: 64-90 mg L⁻¹; TAN: 0.32 mg L⁻¹ to 0.57 mg L⁻¹; NO₃: 0.13 to 0.36 mg L⁻¹ and NO₂: 0.020 to 0.046 mg L⁻¹. There were significant ($p < 0.05$) effects of stocking density on dissolved oxygen, pH, TAN, ammonia and NO₂. The DO ranged from 4 to 7 mg L⁻¹ and generally decreased with increasing stocking density. The pH values across the three treatments ranged from 6.89 to 7.6 for the RAS component and 6.78 to 7.44 for the hydroponic component. The concentration of TAN ranged from 0.32 to 0.57 mg L⁻¹ and generally increased at increasing stocking density. The concentration of NO₃ ranged from 0.91 to 1.3 mg L⁻¹ and increased with increasing stocking density. The concentration of NO₂ significantly ($p < 0.05$) increased with increasing density. Descriptive statistics of water quality parameters were used as covariates in the determination of stocking density on the growth performance of *O. niloticus* in the aquaponic system.

Table 9: Water quality parameters (means \pm SD) in aquaponics stocked with *O. niloticus* at different stocking densities

Parameters	Compartment	150 fish m ⁻³	300 fish m ⁻³	450 fish m ⁻³
Temperature (°C)	RAS	23.64 \pm 0.65	23.23 \pm 0.17	23.95 \pm 0.66
	Hydroponic	24.43 \pm 1.38	23.09 \pm 0.15	24.13 \pm 0.69
Dissolved oxygen (mg L ⁻¹)	RAS	5.35 \pm 1.36 ^c	4.86 \pm 0.56 ^b	3.83 \pm 0.41 ^a
	Hydroponic	5.79 \pm 1.13 ^c	5.12 \pm 0.88 ^b	4.35 \pm 0.1 ^a
pH	RAS	7.60 \pm 0.02 ^c	7.33 \pm 0.05 ^b	6.89 \pm 0.04 ^a
	Hydroponic	7.44 \pm 0.02 ^c	7.20 \pm 0.01 ^b	6.78 \pm 0.03 ^a
Hardness (mg L ⁻¹)	RAS	185.0 \pm 2.9 ^b	168.8 \pm 4.8 ^a	184.7 \pm 5.6 ^b
	Hydroponic	120.0 \pm 4.8	120.0 \pm 5.2	120.0 \pm 10.2
Nitrates (mg L ⁻¹)	RAS	1.11 \pm 0.041 ^a	1.16 \pm 0.012 ^b	1.34 \pm 0.022 ^b
	Hydroponic	0.91 \pm 0.03 ^a	0.98 \pm 0.011 ^b	1.30 \pm 0.022 ^c
Nitrites (mg L ⁻¹)	RAS	0.01 \pm 0.001 ^a	0.05 \pm 0.02 ^b	0.08 \pm 0.06 ^c
	Hydroponic	0.01 \pm 0.001 ^a	0.04 \pm 0.002 ^b	0.08 \pm 0.005 ^c
SRP (mg L ⁻¹)	RAS	2.40 \pm 0.007 ^a	2.34 \pm 0.031 ^b	2.47 \pm 0.012 ^c
	Hydroponic	2.31 \pm 0.088 ^c	2.09 \pm 0.03 ^a	2.19 \pm 0.007 ^b
Potassium (mg L ⁻¹)	RAS	5.24 \pm 0.03 ^b	5.54 \pm 0.06 ^c	4.78 \pm 0.37 ^a
	Hydroponic	5.04 \pm 0.03 ^b	5.34 \pm 0.04 ^c	4.62 \pm 0.05 ^a
TAN (mg L ⁻¹)	RAS	0.03 \pm 0.004 ^b	0.014 \pm 0.00 ^a	0.029 \pm 0.004 ^b
	Hydroponic	0.02 \pm 0.002 ^b	0.01 \pm 0.001 ^a	0.027 \pm 0.005 ^c

RAS = Recirculating Aquaculture System

Values in the same row with different superscripts are significantly different ($p < .05$).

Lower case letters indicate significant differences among the treatments

Table 10: Results of generalized mixed-effects models for water quality variables

	Hardness	Nitrate	Nitrite	SRP	K	Ammonium
Fixed effects	β (SE) t-value	β (SE) t-value	β (SE) t-value	β (SE) t-value	β (SE) t-value	β (SE) t-value
Intercept	269.50 (23.25) 11.6***	-3.38 (1.61) -2.11*	-0.06 (0.10) - 0.53	2.98 (0.35) 8.39***	5.07 (2.18) 2.32*	0.04 (0.03) 1.17
Stocking density	1.88 (10.76) 0.17	1.03 (0.74) 1.39	0.80 (0.05) 1.61	-0.40 (0.16) -2.46*	-0.18 (1.01) -0.17	0.003 (0.01) 0.21
Time in weeks	-20.86 (4.59) -4.55***	2.20(0.32) 6.94***	0.03 (0.02) 1.26	0.09 (0.07) 1.30	0.44 (0.43) 1.03	-0.001(0.006) - 0.11
Stocking density X Time	-0.12 (2.13) -0.06	-0.68 (0.14) -4.64***	-0.01 (0.01) -0.89	-0.08 (0.03) -- 2.36*	-0.23 (0.20) -1.14	-0.002 (0.003) - 0.56
ANOVA for fixed effects	F-value	F-value	F-value	F-value	F-value	F-value
Intercept	2008.2***	81.0***	62.7***	971.9***	156.4***	20.4***
Stocking density	0.11	38.1***	3.3	103.5***	7.02**	0.42
Time in weeks	147.5***	48.9***	1.3	5.4*	0.004	2.77
Stocking density X Time	0.003	21.5***	0.8	5.6*	1.29	0.32
Random effect						
Tank (intercept) SD	0.002	<0.001	<0.001	<0.001	<0.001	0.004
Residual SD	43.70	3.02	0.20	0.67	4.11	0.06
$R^2_{GLMM(m)}$	0.55	0.47	0.04	0.48	0.06	0.03
$R^2_{GLMM(c)}$	0.55	0.47	0.04	0.48	0.06	0.03

Note: The ‘full’ model included stocking density (150, 300 and 450 fingerlings m⁻³), time in weeks and a stocking density × time interaction as fixed effects and tank as a random effect. The marginal R² (GLMM[m]; fixed effects only) and the conditional R² (GLMM[c]; fixed and random effects) represent the proportion variance explained by each model. SE = standard error; SD = standard deviation; *p < 0.05, **p < 0.01, ***p < 0.001

The PCA plot for the distribution in water quality parameters in the hydroponics and RAS with respect to stocking density is shown in Figure 14 while the factor loading of the correlation matrix between the water quality variables and stocking density are shown in Table 11 representing 47.46% (Approximately 50%). Based on the factor analysis diagram (Figure 14) and the factor loading table (Table 11), the TAN, nitrate, NO_3 and hardness were largely influenced by the stocking density of fish.

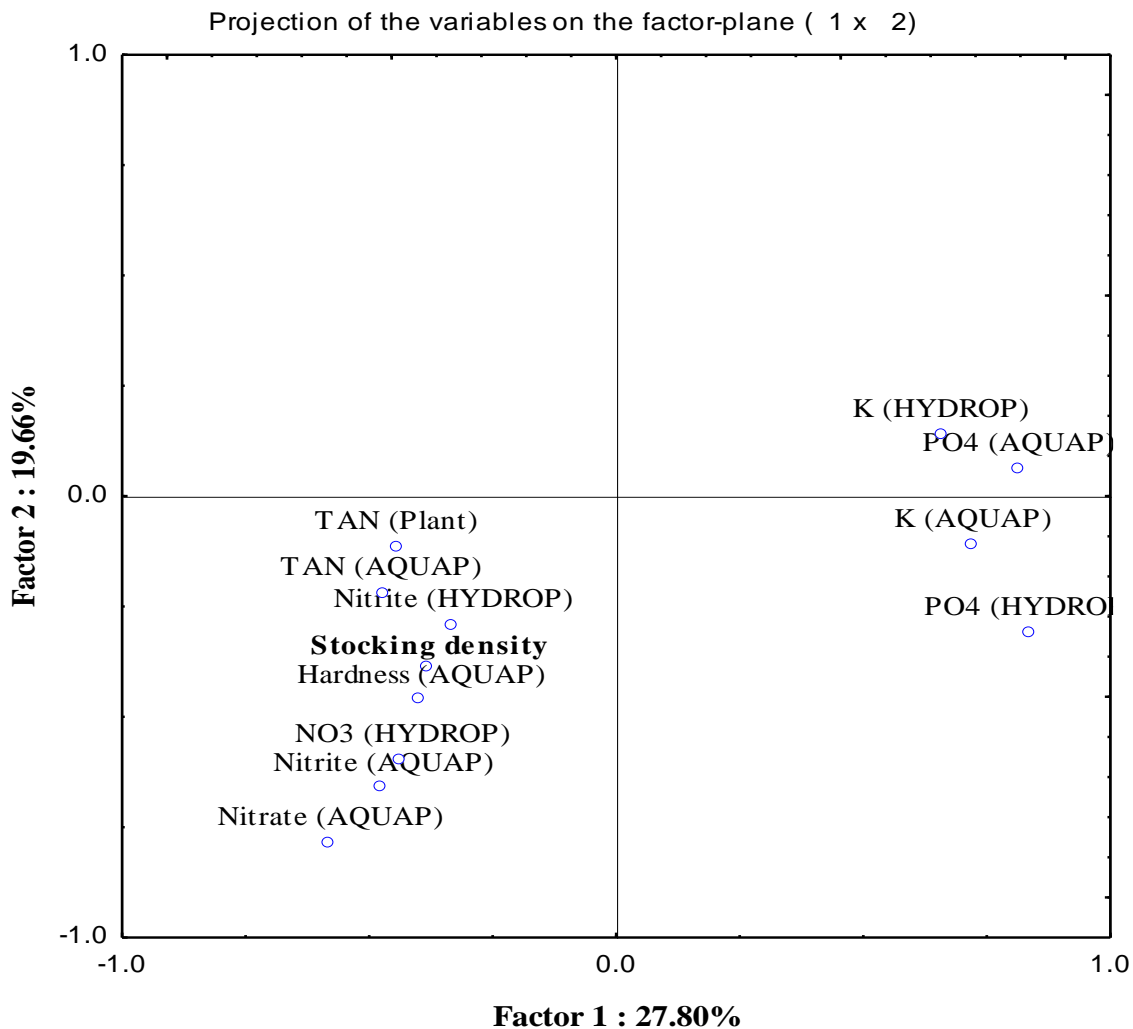


Figure 14: Results of Factor analysis of the stocking density on water quality variable

Table 11: Factor loading table of the variables, based on correlations

Variable	Factor 1	Factor 2
Nitrate (AQUAP)	-0.185	0.781
Stocking density	-0.292	-0.437
Nitrite (AQUAP)	-0.477	-0.558
Hardness (AQUAP)	-0.401	-0.459
NO ₃ (HYDROP)	0.039	0.945
Nitrite (HYDROP)	0.336	-0.288
K (HYDROP)	0.658	0.143
K(AQUAP)	0.715	-0.067
TAN (Plant)	-0.444	-0.112
TAN (AQUAP)	-0.475	-0.116

The nutrient removal rates of the hydroponics are shown in Table 12. The highest percentage removal efficiency occurred for hardness, nitrite and TAN. Meanwhile stocking density significantly affected the removal efficiency of nitrate, nitrite, PO₄ and TAN. The highest nitrate, nitrite and TAN removal efficiency occurred at density 150 followed by 300 fish m⁻³ while stocking density 300 and 450 fish m⁻³ removed more PO₄ than stocking density 150 fish m⁻³.

Table 12: Nutrient removal efficiency (%) of the hydroponic unit

Water quality parameters	150 fish m ⁻³	300 fish m ⁻³	450 fish m ⁻³
Alkalinity	35.1 ± 4.2 ^a	28.9 ± 4.3 ^a	35.0 ± 7.5 ^a
Nitrates	18.0 ± 2.2 ^b	15.5 ± 3.3 ^b	3.0 ± 3.4 ^a
Nitrites	50.0 ± 7.2 ^b	35.1 ± 9.6 ^b	2.4 ± 0.4 ^a
PO ₄	3.9 ± 0.3 ^a	12.0 ± 3.4 ^b	12.8 ± 3.4 ^b
Potassium	4.0 ± 1.3 ^a	3.7 ± 1.1 ^a	3.5 ± 0.4 ^a
TAN	21.9 ± 3.4 ^c	14.3 ± 3.1 ^b	6.9 ± 1.1 ^a

Values in the same row with different superscripts are significantly different (p<.05).

Lower case letters indicate significant differences among the treatments

4.4 Economic performance of the aquaponic system

To construct an aquaponics system, the total capital calculated was approximately KES 62,450 (\$ 624.5) for each aquaponic system based on the different stocking densities as used in the study (Table 13). The main units of aquaponics were the reservoir tanks, the RAS composed of fish tanks and the hydroponic unit composed of a hydroponic unit (0.07 m³) and a sump. The RAS unit consisted of approximately 90 % of the costs with the remaining 10% accounted for by the hydroponic unit. The cost was reduced by designing the water to flow through gravity from the reservoir tank.

Table 13: Items required, quantity, unit cost, and total cost for the *O. niloticus* and *Lactuca sativa* production components of the aquaponic unit

Items	Unit quantity	Price/Unit(KES)cost	Price/unit (\$)	Total cost	Cost (\$)
RAS component					
Aeration tubes	3	100	1	300	3
Air pump	3	1000	10	3,000	30
Airstones	5	250	2.5	1250	12.5
Bench platform	1	24,000	240	8,000	80
Biofilter system	5	1000	10	5,000	50
Plastic tanks	5	4,000	40	20,000	200
Piping	Assorted			2,000	20
Reservoir tanks	1	18000	18	6,000	60
Submersible pump	5	1500	15	7,500	75
Sump/waste pipe	5	300	3	1,500	15
Styrofoam	1	1000	10	1,000	10
Tangit	500 ml	600	6	200	2
Subtotal				55,750	557.5
Hydroponic units					
Hydroponic tank	1	3500	35	3,500	35
Pipes and fittings	Assorted			3,000	30
Plant pots	120	5	0.05	200	2
Subtotal				6,700	67
Total aquaponic				62,450	624.5

NB: Exchange rate: 1\$=100 KES, (2017)

The total yields of *O. niloticus* and *Lactuca sativa* in each treatment unit are shown in Table 14. Fish yield increased with increasing stocking density, ranging from 6 kg per tank at stocking density 150 fish m⁻³ to 10.33 kg per tank at stocking density 450 fish m⁻³. *Lactuca sativa* yields ranged from 1.66 kg at stocking density 150 fish m⁻³, to the highest value of 3.04 kg at a stocking density of 450 fish m⁻³.

Table 14: Yields of *O. niloticus* and *Lactuca sativa* at different stocking densities after 56 days

	Stocking density (fish m ⁻³)		
	150	300	450
Total fish yield (kg m ⁻³)	6.00 ± 1.42 ^a	8.56 ± 1.43 ^b	10.33 ± 2.55 ^c
<i>Lactuca sativa</i> yield (kg m ⁻³)	1.66 ± 0.98 ^a	2.76 ± 0.23 ^b	3.04 ± 0.23 ^c

Values in the same row with different superscripts are significantly different (p<.05).

Lower case letters indicate significant differences among the treatments

Based on the yields, and other operating expenses, the enterprise budget of the aquaponics system consisting of *O. niloticus* and lettuce at different stocking densities was determined (Table 15). All the treatments posted positive returns and were viable investments. The break-even prices for variable costs were able to cover the cost of fish and lettuce in the local market as they were below the sale price of Ksh 400/kg and Kshs 300/kg respectively that the aquaponic systems were able to get in the market.

Table 15: Overall enterprise budgets for *O. niloticus* and *Lactuca sativa* in a model aquaponic at different stocking densities for 56 days

	Fish stocking density		
	150 fish m ⁻³	300 fish m ⁻³	450 fish m ⁻³
Survival (%)	93.3	90	90.2
Mean fish yield (kg m ⁻³)	6	8.56	10.33
Mean <i>Lactuca sativa</i> yield (kg m ⁻³)	1.66	2.76	3.04
Total revenue from fish (Kshs)	4200	5992	7231
Revenue from <i>Lactuca sativa</i>	498	828	912
Gross revenue	4698	6820	8143
Variable costs			
Cost of feeds	132.54	186.9	208.392
Field labour	1500	1500	1500
Cost of maintenance of equipment	350	350	350
Water	650	650	650
Electricity	800	800	800
Miscellaneous	600	600	600
Sub-total variable costs	4032.5	4086.9	4108.4
Interest on operating cost	725.857	735.642	739.51056
Total variable cost (TVC)	4758.4	4822.5	4847.9
Fixed costs			
Bench platform	800	800	800
Amortization	200	200	200
Interest on the fixed cost	180	180	180
Total fixed cost	1180	1180	1180
Total cost (TC)	5938.4	6002.5	6027.9
Net returns above TVC	-60.4	1997.5	3295.1
Net returns above TC	-1240.4	817.5	2115.1
Break-even price	196.7	137.9	114.2

Assumption: The market price of *O. niloticus* was Kshs 400 kg⁻¹ and Lettuce Kshs 300 kg⁻¹.

The cash flow projections were made over a 5-year cycle within a year for each of the aquaponic units. There was initial cash to buy capital items during the establishment of the research unit. In the preceding years, there were operating expenses and revenues generated from sales. The net present value (NPV) and internal rate of return (IRR) were determined for each of the model units (Table 16). A discount rate of 18% was used to ascertain the NPV. It was observed that sales of fish and *Lactuca sativa* at a stocking density of 150 fish m⁻³ were not profitable. However, all the other stocking densities were profitable except at 300 fish m⁻³ increased with increased stocking density. Investors must decide their necessities for adequate returns while choosing their aquaponic facility size. On the off chance that the IRR is excessively low, at that point other speculation openings must be found.

Table 16: Net present value (NPV) and internal rate of return (IRR) for the model aquaponics at different stocking densities

Years	150 fish m ⁻³		300 fish m ⁻³		450 fish m ⁻³	
	NPV	IRR	NPV	IRR	NPV	IRR
1	-12,061.5	4.2	-1103.5	16.2	-12,061.5	16.2
2	-8,545.2	5.7	-8,143.2	16.7	5,545.21	16.7
3	-7,610.6	7.2	-4,610.62	17.2	5,610.62	17.2
4	-5,697.9	8.4	-1,654.1	17.8	3,697.95	17.8
5	-1,026.2	10.7	26.4	16.7	1,026.15	16.7

Result of the sensitivity analysis showing how the variations in the cost of production affect the profitability of the enterprise under different stocking density scenarios is shown in Table 17. For the range of profitability given, the cost of production above Kshs 6000 m⁻³ was not profitable regardless of the stocking density. Meanwhile, at a low stocking density below 150 fish m⁻³, production cost above Kshs 4400 m⁻³ resulted in negative returns and should not be countenanced. At stocking densities extending between 240 to 500 fish m⁻³ seem to yield profit throughout most of the range of production prices, and the size of the potential profits is considerable.

Table 17: Sensitivity analysis of the profitability of the aquaponics to alternative variation in the cost of production

Market Price (Kshs)	Stocking density (fish m ⁻³)							
	122.88	153.6	192	240	300	360	432	518.4
400	1802.5	2153.1	2591.4	3139.2	3824.0	4508.8	5330.6	6316.7
1200	3402.5	3753.1	4191.4	4739.2	5424.0	6108.8	6930.6	7916.7
2800	4202.5	4553.1	4991.4	5539.2	6224.0	6908.8	7730.6	8716.7
3600	3402.5	3753.1	4191.4	4739.2	5424.0	6108.8	6930.6	7916.7
4400	1802.5	2153.1	2591.4	3139.2	3824.0	4508.8	5330.6	6316.7
5200	-597.5	-246.9	191.4	739.2	1424.0	2108.8	2930.6	3916.7
6000	-3797.5	-3446.9	-3008.6	-2460.8	-1776	-1091.2	-269.4	716.7

At a stocking density of 300 fish m⁻³, the total cost involved is Kshs 2,800 and this gives an operating profit of 6224.0 and this is our current model. By increasing the market price from Kshs 400 to 4400 all the stocking densities give a positive net

return or operating profit. However when the market price increases beyond the current model of Kshs 2800 profitability remains positive but lower values are recorded. Profitability remains positive up to a cost price of Kshs 5,200 and turns negative past the Kshs 5,200 point for all the stocking densities ranging from 122.88 to 518.4 fish m⁻³. At higher densities i.e. past 432 fish m⁻³ profitability or positive net returns can be achieved. As the density reduces from 300 to 192 a positive net return is assured up to the cost price of Kshs 5, 200 whereas the densities of below 192 only produce a positive net return up to Kshs 4,400.

CHAPTER FIVE

DISCUSSION

5.1 Effects of stocking density on fish growth performance

Although fish performance in aquaponics has not been extensively studied in Kenya, the system is comparable to other culture units in terms of fish performance (Endut *et al.*, 2016) where important factors such as stocking density should be determined as a function of the growth of the fish cultured. During the study, the growth of *O. niloticus* in aquaponics was evaluated based on fish stocked at an initial average weight of 17.9 ± 1.7 g for 8 weeks. Generally, the fish weight increased with decreasing stocking density.

There were significant interactions between stocking density and time (weeks) in all parameters of growth examined; length, weight, SGR and FCR. Fish stocked at 150 fish m^{-3} had the highest mean weight and final length. Based on the analysis of change, this fish also demonstrated faster growth as compared to fish stocked at densities 300 and 450 fish m^{-3} . Rayhan *et al.* (2018) obtained comparable outcomes, where stocking density was related to the average weight gain and length in tilapia. The present results, therefore, suggest that the weight of fish decreased inversely with increasing stocking density and the patterns appeared consistent throughout the experiment period which concurs with those established by Ferdous *et al.* (2014) and Rayhan *et al.* (2018).

Generally, the fish weight increased with decreasing stocking density. An increase in fish weight by approximately 250% during a similar rearing period has been obtained in studies done in tanks (Siddiqui and Al-Harbi, 1999; Yoo and Lee, 2016) and

recirculating aquaculture systems (Wang *et al.*, 2019). However, higher values than what the present study describes have been reported in experiments done in commercial aquaponics (Endut *et al.*, 2009). Although fish performance in aquaponics has not been extensively studied, the system is comparable to other culture units in terms of growth performance (Endut *et al.*, 2016) where important factors such as stocking density should be determined as a function of the growth of the fish cultured. The current study demonstrated that the stocking density chosen influences the growth of the fish. It is evident from the literature that fish raised in aquaponic systems can achieve a growth performance similar to those raised in conventional aquaculture and recirculating aquaculture systems (Baßmann *et al.*, 2017; Maucieri *et al.* 2019). This, therefore, suggests that the current growth performance may have been lower than what is reported in other experiments and also in commercial aquaponics, perhaps because of the better water recirculation used in those reported studies.

The current study showed that the SGR increased with decreasing stocking density of fish. Steeper slopes were seen in the regression equations and the effect of stocking density gave a negative relationship. Palm *et al.* (2014) working also in the aquaponic system reported a specific growth rate of 0.71% per day in Nile tilapia whose initial weight was 174 g and were initially stocked at 5.6 kg m⁻³. Greenfeld *et al.* (2018) additionally reported that the SGR of Koi Carp (*Cyprinus carpio*) whose initial weight of 4.24 g raised in an aquaponic system decreased with increasing density of fish from 1.4 kg m⁻³ to 2.1 kg m⁻³ to 2.8 kg m⁻³. Maucieri *et al.* (2019) also observed a substantial reduction of SGR with increasing stocking density in juveniles of tilapia reared in aquaponic systems. This shows that as stocking density increases feed

utilization efficiency reduces which also affects the somatic growth. Higher stocking densities above the tank's carrying capacity causes water quality deterioration which increases stress in fish.

The FCR increased with increasing stocking density and this could probably be due to the utilization efficiency of the food given and the amount of feed given in each treatment a phenomenon also reported by AL-Harbi and Siddiqui, (2000). Several studies report a negative effect as stocking density increases on the feed conversion ratio of fish in both the aquaponic and conventional aquaculture systems (Tran *et al.*, 2019; Maucieri *et al.*, 2019). However, other authors still working in aquaponic systems did not report any significant changes in FCR being affected by the changes in the stocking density (Hayat *et al.*, 2018 and Maucieri *et al.*, 2019).

The survival rate of the fish decreased with increasing stocking density. This may probably be due to the crowded condition created as a result of the higher densities and the resulting competition for space and food, even though the fish were fed to satiation. Similar results of higher densities creating crowding conditions were also reported by Gibtan *et al.* (2008) and Person-Le Ruyet *et al.* (2008).

5.2 Effects of stocking density of fish on the growth of *Lactuca sativa*

Plant growth can be used to evaluate the suitability or efficiency of a hydroponic sub-system (Diem *et al.*, 2017). The production in terms of total production, yield and mean per plant of *Lactuca sativa* increased with increasing stocking density a phenomenon also reported by Estrada-Perez *et al.* (2018). Generally, the average plant (lettuce) production in terms of total biomass increased with increasing stocking density. The lettuce shoot and root length and the number of leaves counted also increased with increasing stocking density of fish in the aquaponic system a phenomenon similarly reported by Estrada-Perez *et al.* (2018).

The observed increase in plant production at high density may be related to high nutrient released from the recirculating units and was available to the plants mainly as NO_3 . Although a high stocking fish density was observed to result in lower growth rates of lettuce due to the promotion of denitrification (Endut *et al.*, 2010). It was also noted that the lettuce grown in the 300 fish m^{-3} treatment were generally heavier than those in the 150 fish m^{-3} treatment even though the number of leaves was nearly the same. The heaviness in the leaves was probably because the leaves were broader possibly due to the high concentration of nutrients absorbed from the systems with high stocking density. The slope in the 450 fish m^{-3} treatment was much steeper than the other two densities hence yielding more leaves and having a higher plant weight.

The main growth-limiting nutrient is usually nitrogen which is absorbed by the plants as either nitrate or ammonia and this is influenced by the level of CO_2 in the water (Eck *et al.*, 2019). The higher growth performance of plants is normally seen when nitrogen is supplied as a combination of both ammonium and nitrate (Maucieri *et al.*,

2019). Lettuce can absorb ammonia and promote oxidation with the help of root borne bacteria (Widyastuti (1998) as cited by Portalia *et al.*, 2019). The effect of the absorbed nutrients on the lettuce crops was evident since the plants recorded different chlorophyll levels as stocking density increased.

5.3 Effects of stocking density on water quality

Except for a few cases in this experiment, water quality variables across treatments were within the favourable range required for fish culture even under high stocking density but were significantly influenced by stocking density. A similar observation was made by Rahmatullah *et al.* (2010), who in their study directly linked stocking density of fish to changes in water quality. The dissolved oxygen (DO) levels recorded in this experiment were within the recommended range of 3-6 mg L for the culture of Nile tilapia as suggested by many authors (Xu *et al.*, 2006; Tran-Duy *et al.*, 2008; Abdel-Tawwab *et al.*, 2014; Hassaan *et al.*, 2014; Tran-Ngoc *et al.*, 2016).

The dissolved oxygen range reported in the current experiment is in line with suggestions made by Hillary and Boyd (1997), Xu *et al.* (2006), and Rahmatullah *et al.* (2010) without taking account of stocking density. The decrease in DO levels as stocking densities increased (Jørgensen *et al.*, 1993) could probably result in the mortalities observed in the current experiment. The mortality in fish increases when they are crowded and lack adequate food resulting in cannibalism. The water quality is also deteriorating as stocking density increases a condition that will stress the fish and result in fish kills (Yang *et al.*, 2020). The low DO levels could also be associated with reduced water circulation; hence there is a need to allow the water to circulate as aeration also goes on. It is also associated with greater total respiration and metabolic

demand which increases with increasing stocking density (Teichert-Coddington and Green, 1993).

Low concentrations of dissolved oxygen in the aquaponics system may probably be due to the fast decomposition of the fish metabolites and feed materials and present in the culture system (Yildiz *et al.*, 2017). The solubility of DO decreases with the increase in temperature (Rakocy, 2007; Yildiz *et al.*, 2017) but generally, the different stocking densities did not influence this. A similar DO range of 3-5 mg L⁻¹ was also reported in experiments done by Yildiz *et al.* (2017) and Maucieri *et al.* (2019). In their studies, these authors suggest that anything below this range lowers nutrient uptake hence resulting in a reduction in plant and fish growth. The fish, plants and bacteria in the aquaponic system require adequate amounts of DO for maximum health and growth (Rakocy, 2007). Oxygen is also an important requirement in the process of nitrification.

The pH and temperature were within the recommended ranges ideal for aquaponic systems as also reported by (Rakocy *et al.*, 2006 and Maucieri *et al.*, 2019) even though they kept varying across the different stocking densities of fish in both the RAS and hydroponic units. The pH decreased with increasing stocking density of fish in the system, an observation also reported by several authors (Goddek *et al.*, 2015; Kloas *et al.*, 2015; Yildiz and Bekcan (2017). Yildiz and Bekcan (2017) for example recommended that for the success of the nitrification process in the aquaponic unit pH should be kept around 7. pH below 6 can disrupt the nitrification process causing the aquaponic system to fail (Goddek *et al.*, 2015; Yildiz *et al.*, 2017). Regardless of the stocking density used the pH levels were within the recommended levels for both fish and plant culture. The process of nitrification is more efficient at pH levels higher

than 7 and ceases at pH lower than 6. Low pH is toxic to fish and it also affects the solubility of nutrients hence affecting the plant growth and yields negatively (Rakocy, 2007).

There were significant interactions between stocking density and time (weeks) in water hardness, nitrite, nitrate and SRP, implying that as fish grew, they added more nutrients into the system and their uptake by the plants (lettuce) could not keep up with the loading rates. This phenomenon was also reported by Graber and Junge (2009) and Rakocy *et al.* (2016). Interestingly, TAN did not respond to stocking density, as expected, and this could be attributed to several factors. Firstly, ammonia is very volatile and highly labile, suggesting that it was likely the N species that was most up taken by the plants. Similar observations on plant uptake of ammonium in the aquaponic systems were also made by Gichana *et al.* (2018). Secondly, it was likely transformed into other species of N (nitrite and nitrate) by biogeochemical processes mediated by changes in pH and dissolved oxygen concentrations (Goddek *et al.*, 2015; Delaide, 2017 and Goddek, 2017).

The concentration of nitrite and nitrate in this study increased with increasing stocking density a phenomenon also directly related to the amount of feed administered. Rahmatullah *et al.* (2010) report a linear increment of TAN and nitrate with increasing stocking density and feed input. The removal of TAN, nitrite, and nitrate from the water as indicated by the lower values in the hydroponic unit as compared to the RAS unit was probably due to plant uptake (Zarantoniello *et al.*, 2021). Microorganisms present in the system assimilate $\text{NO}_3\text{-N}$ in the water or the root of the plants grown with the help of biofilms (Azam and Ifzal, 2006). These nutrients are important for the growth of lettuce.

5.4 Effects of stocking density on economic performance

Recirculating aquaculture systems provide the economic benefit of fish harvest, plant harvest as well as difficult to quantify low water consumption (Rakocy *et al.*, 2016; Engle, 2015). Although there is high fish production that enhances the highest net profit, it likewise includes economic investment as electricity is consumed by the submersible pumps for water flow. The merit of the aquaponic system cuts down this cost to some level by production of the second crop of vegetables which improves the system's profit margin (Tokunaga *et al.*, 2015). According to Quagraine *et al.*, 2017 the initial investment is normally higher because of the cost of installation. The inclusion of a plant like lettuce which can be harvested at least thrice during the overall production period thereby increasing the revenue of the aquaponic system

In the current study, the total production of yields of Nile tilapia and *L. sativa* in each treatment increased with increasing stocking density. This is consistent with work done by Yoo *et al.* (2016) who linked yields of plants being affected by the stocking density of fish. Based on the yields, and other operating expenses, the enterprise budget of the aquaponics system consisting of *O. niloticus* and lettuce at different stocking densities all the treatments except that of stocking density of 150 fish m⁻³ posted positive returns to the risk and were viable investments.

This is in agreement with Shoko *et al.* (2016), who in their work reported positive net returns in all the stocking densities used. This, therefore, implied that stocking densities above 300 fish m⁻³, if chosen, should yield a profit. Hence profitability increased with increased stocking density, a phenomenon also observed and reported by Rahman *et al.* (2006) and Shoko *et al.* (2016). Stocking densities 300 and 450 fish

m^{-3} had significant positive effects on production, suggesting that higher densities may be used to obtain higher biomass and to improve the profitability of the system. This agrees with Rahman *et al.* (2006) who linked density to more yields and profitability.

The break-even prices in the current experiment decreased with increasing stocking density. The break-even prices for variable costs were able to cover the cost of fish in the local market as they were below the sale price provided in the market. Goodman (2011), Tokunaga *et al.* (2015), Engle (2015) and Shoko *et al.* (2016) commend that the break-even prices are important financial aspects to be considered in any enterprise because they indicate the profitability of the operation in the short and long term, provided the commodity price at the market is higher than the break-even prices obtained.

The sensitivity analysis shows how the variations in the cost of production affect the profitability of the enterprise under different stocking density scenarios. It denotes that enterprise viability is very sensitive to sale price changes received for the harvested product, highlighting the largest impact on annual returns (Di Trapani *et al.*, 2014). Basing on the current study, the range of profitability given, the costs of production above Kshs 6000 m^{-3} were not profitable regardless of the stocking density used. Meanwhile, at a low stocking density below 150 fish m^{-3} , production cost above Kshs 4400 m^{-3} resulted in negative returns and should not be permitted. At stocking densities extending between 240 to 500 fish m^{-3} seem to yield profit throughout most of the range of production prices, and the size of the potential profits is considerable. A similar phenomenon was also reported by Sogbesan and Ibiyo (2003) and Ngugi *et al.* (2012). The best model would therefore be at a market price of Kshs 2800 which

gives a positive operating profit or net return across the stocking the different stocking densities. This model's market price value also gives the highest net returns especially from stocking densities above 240 to 518.4 fish m⁻³.

CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

The conclusions of this study are as follows:

i) **Effects of stocking density on growth performance of *O. niloticus***

The growth performance of Nile tilapia fingerlings in terms of weight and length and growth parameters FCR and SGR was significantly influenced by the stocking density. The mean weight and length and the SGR decreased with increasing stocking density whereas the FCR increased with increased stocking density. Generally, the fish performed better at lower i.e. 150 fish m⁻³ than at higher stocking densities of 300 and 450 fish m⁻³ respectively. This, however, doesn't rule out the fact that higher stocking densities can still be used especially when good water quality parameters are maintained.

ii) **Effects of stocking density on growth performance of *L. sativa***

Generally, the production in terms of total production, yield and mean per plant of *L. sativa* increased with increasing stocking density. A similar trend as those of production was observed for yield per kg and mean weight per plant with respect to stocking density. The amount of feed given has a direct effect on the amount of nutrients the system can generate. The higher the stocking density the higher the volume of nutrients the aquaponic system generates for the plants. Stocking densities of 300 and 450 fish m⁻³ had the highest growth performance of lettuce. It was evident

that stocking densities of fish positively affects the growth performance, the total yield and the chlorophyll level in lettuce grown in a small scale aquaponics system.

iii) Effects of stocking density water quality change and nutrient removal efficiency

Stocking density had a significant effect on the water quality of aquaponic systems such that the lower the stocking density the better the water quality in terms of pH, DO, TAN, nitrite, nitrate and SRP. Regardless of the stocking density used, the water quality parameters measured were within the recommended concentrations.

pH was around 7, and this will be instrumental in the nitrification process where a drop in pH will result in disrupting the process. DO levels were within the recommended levels. Having adequate quantities of DO in the system assists in ensuring both the (plant) lettuce and the fish grows healthily and fast.

Nutrient removal witnessed in this experiment is a result of nutrient uptake by the plants grown in the system. The higher values of the nutrient removal recorded were probably related to the growth of lettuce. Growth of lettuce increased with increasing stocking density hence more nutrients removed in 450 fish m⁻³ treatment than 300 and 150 m⁻³ respectively.

iv) Effects of stocking density on economic benefits of the aquaponics system

Generally, the total yield of fish and lettuce increased with increasing stocking density. The enterprise budgets for aquaponic farms with different stocking densities for fish and lettuce production indicated all the treatments posted positive returns to

the risk and were viable investments.

The cash flow projections were made over 5 years for each of the units with the net present value (NPV) and internal rate of return (IRR) were calculated for each of the model farms. A discount rate of 18 % was used to calculate the NPV. The sales of fish and *L. sativa* at a stocking density of 150 fish m⁻³ were not profitable and hence not worthwhile.

The break-even prices for variable costs were able to cover the cost of fish in the local market as they were below the sale price of Ksh 10 per case that the farm was able to receive in the market. At a discount rate of 18%, it was observed that sales of fish and *L. sativa* at a stocking density of 150 fish m⁻³ were not profitable. All the other stocking densities were profitable except at 300 fish m⁻³ and 450 fish m⁻³. The optimal density that gave the highest profits based on the different economic analysis was the treatment that was stocked with 300 fish m⁻³.

6.2 Recommendations

1. Farmers can adopt higher stocking densities of fish of 300-400 fish m⁻³. But even higher densities can be used than what is recommended in this study as long as they invest in aeration and filtration systems.
2. Farmers to adopt higher fish stocking densities in their aquaponic systems to ensure enough nutrients are available for the plants to utilize.
3. Higher stocking densities in the fish component affects the water quality, hence there is needed to ensure that optimal levels are maintained for the growth of both lettuce and Nile tilapia.
4. Investors must determine their requirements for acceptable returns when choosing the optimal densities for their farm size. If the IRR is too low then other investment opportunities must be found. The farmers can also increase the stocking density of fish and lettuce to maximize profits without compromising the water quality of the aquaponic system.

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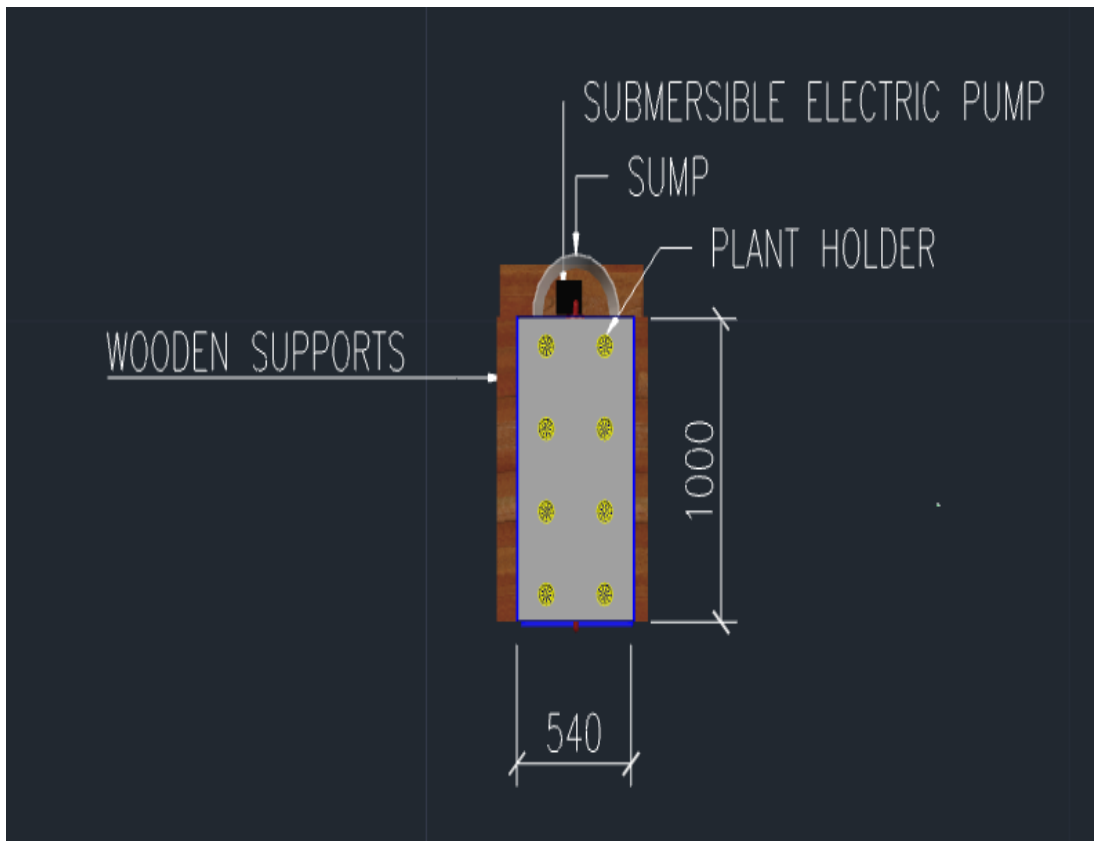
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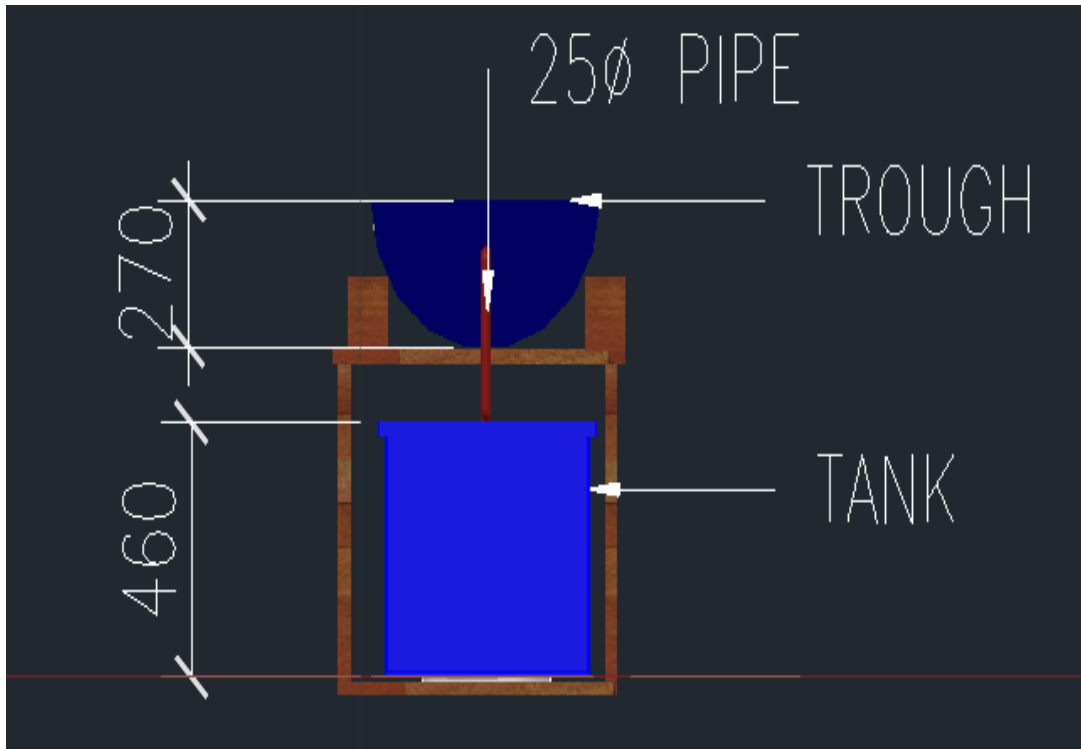
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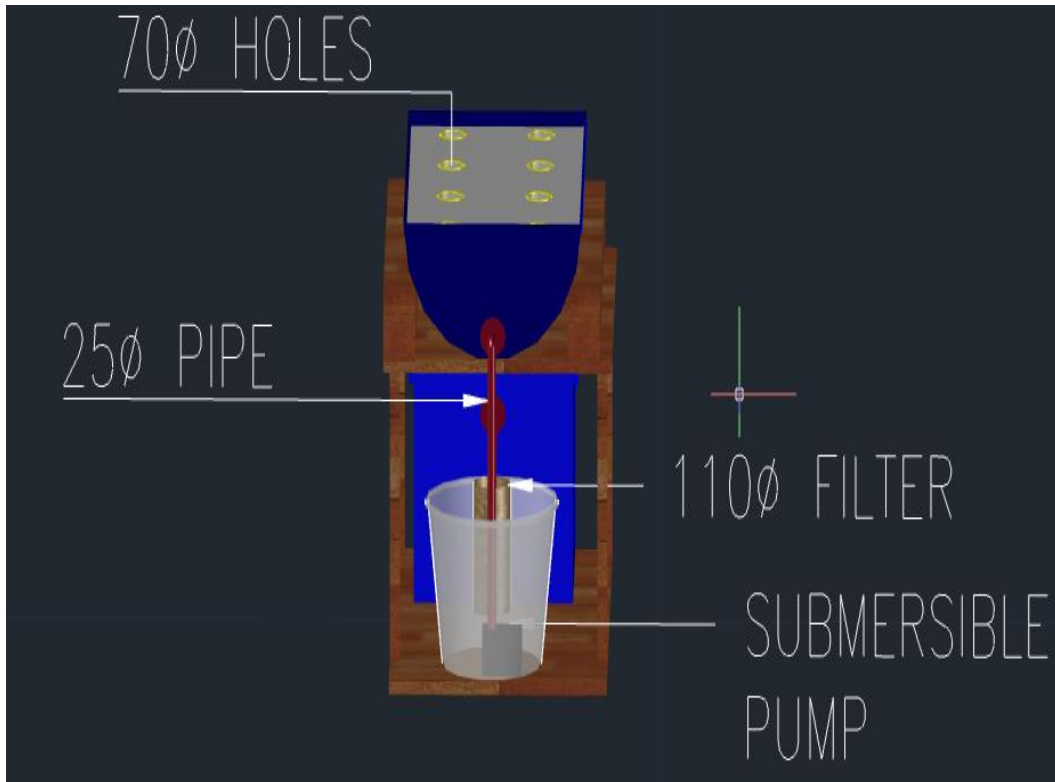
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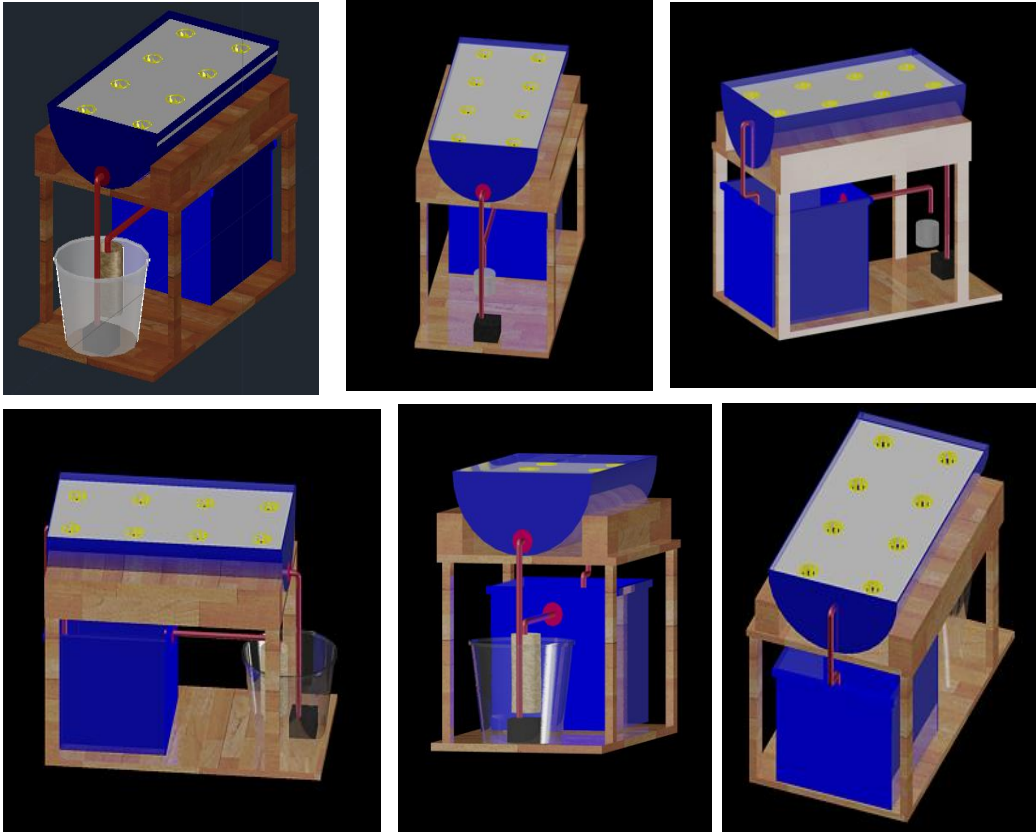
APPENDICES

Appendix I: Aerial view of the setup with width and length specifications



Appendix II: Cross-sectional view of the setup showing height specifications

Appendix III: Setup showing key plumbing components

Appendix IV: Different views of the aquaponic setup used

Appendix V: Proximate procedures used

Crude protein

A simple method as proposed by Chow *et al.* (1980) was used. 1 g of the sample being tested was weighed and placed in a Kjeldahl flask. The following were added 10 g potassium sulphate, 0.7 g mercuric oxide and 20 ml concentrated sulphuric acid. The flask was fixed with the digester and heated till the solution became colourless. After 30 minutes of additional heating, the solution was set aside to cool by gradually adding 0.09L of distilled water followed by 0.025 L. The solution was stirred evenly for 3 minutes. 80 ml of 40% sodium hydroxide solution was then added, as the flask remained tilted until two layers form. The flask was immediately connected to the distillation unit, which was heated. 0.05 L of the distillate that has ammonia was collected. This solution was titrated with standard chlorhydric acid.

Calculations

$$[\text{Nitrogen in the sample (\%)} = 100\left[\frac{A*B}{C}0.014\right]$$

$$\text{Crude protein (\%)} = \text{nitrogen in sample} \times 6.25$$

Where:

A= The standard Chlorhydric acid;

B= The Normality of the standard acid

C= the weight of the sample

Crude lipids

Extraction flasks are removed from the kiln and cooled in a dryer and weighed. 5 g of dry sample of the ingredients are placed in the extraction thimble which is then connected to the extraction unit. The flask that had 66% of petroleum ether of the total volume was then connected to the extractor. The extraction took approximately 2 hours. The ether was evaporated by distillation and the flasks cooled. The defatted sample was used in determining crude fibre.

Calculations

$$\text{Crude lipid content (\%)} = 100 \frac{B - A}{C}$$

Where: A= Weight of the empty flask; B= Weight of the flask with fat; C= the weight of the sample

Crude fibre

3 g of defatted fat is weighed and placed in a flask and 200 ml boiling sulphuric acid solution is added. The contents are boiled for 30 minutes. By maintaining the volume of distilled water constant and swirling the flask periodically to remove particles adhering to the sides. Buchner funnel Lined with filter paper and pre-heated with boiling water. At the same time, at the end of the boiling period, the flask was removed, and the contents filtered by suction. The filter paper is then washed with boiling water and the residue transferred to the flask. The residue was then washed using HCL solution, boiling water and petroleum ether. The crucible is then placed crucible in a kiln set at 105°C for 12 hours then cool in a dryer. The crucible was

weighed with the residue. It was then placed in a furnace at 550° C for 3 hours and weighed again

Calculations

$$\text{Crude fibre content (\%)} = 100 \frac{A - B}{C}$$

Calculations

$$\text{Crude fibre content (\%)} = 100 \frac{A - B}{C}$$

Where: A= Weight of crucible and residue; B= Weight of the crucible with ash; C= the weight of the sample

For ash

5 g of the dry sample is placed in a crucible previously calcined and brought to constant weight. It is then placed in a furnace and heat at 550°C for 12 hours; left to cool and transferred to a dryer then weighed.

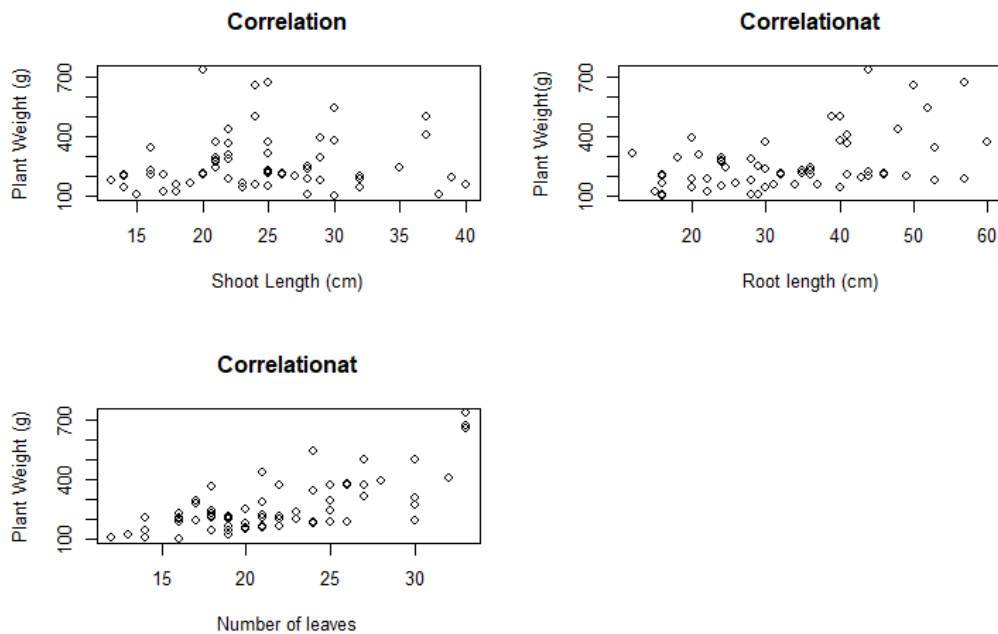
Calculations

$$\text{Ash content (\%)} = 100 \frac{A - B}{C}$$

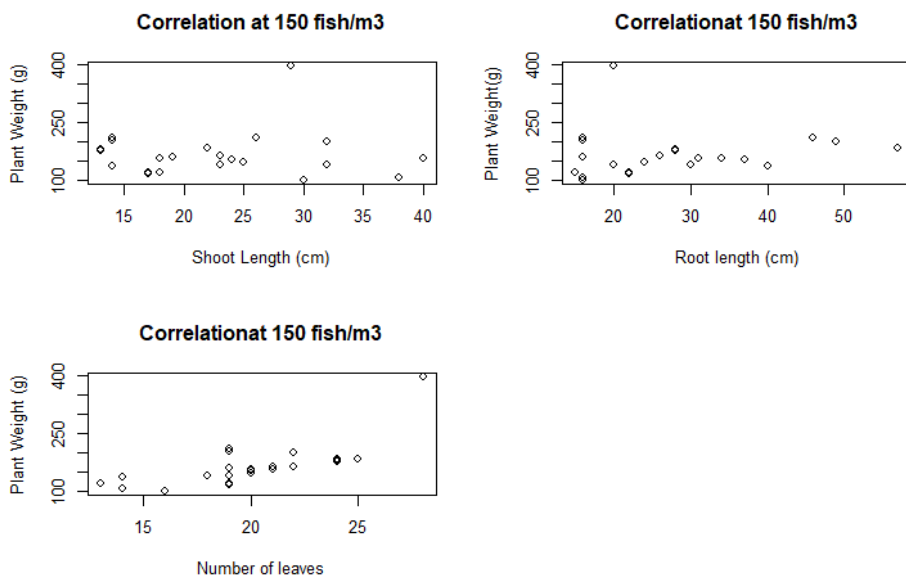
Where A= Weight of the crucible with the sample, B = Weight of crucible with ash and C= Weight of sample

Appendix VI: Additional data analysis

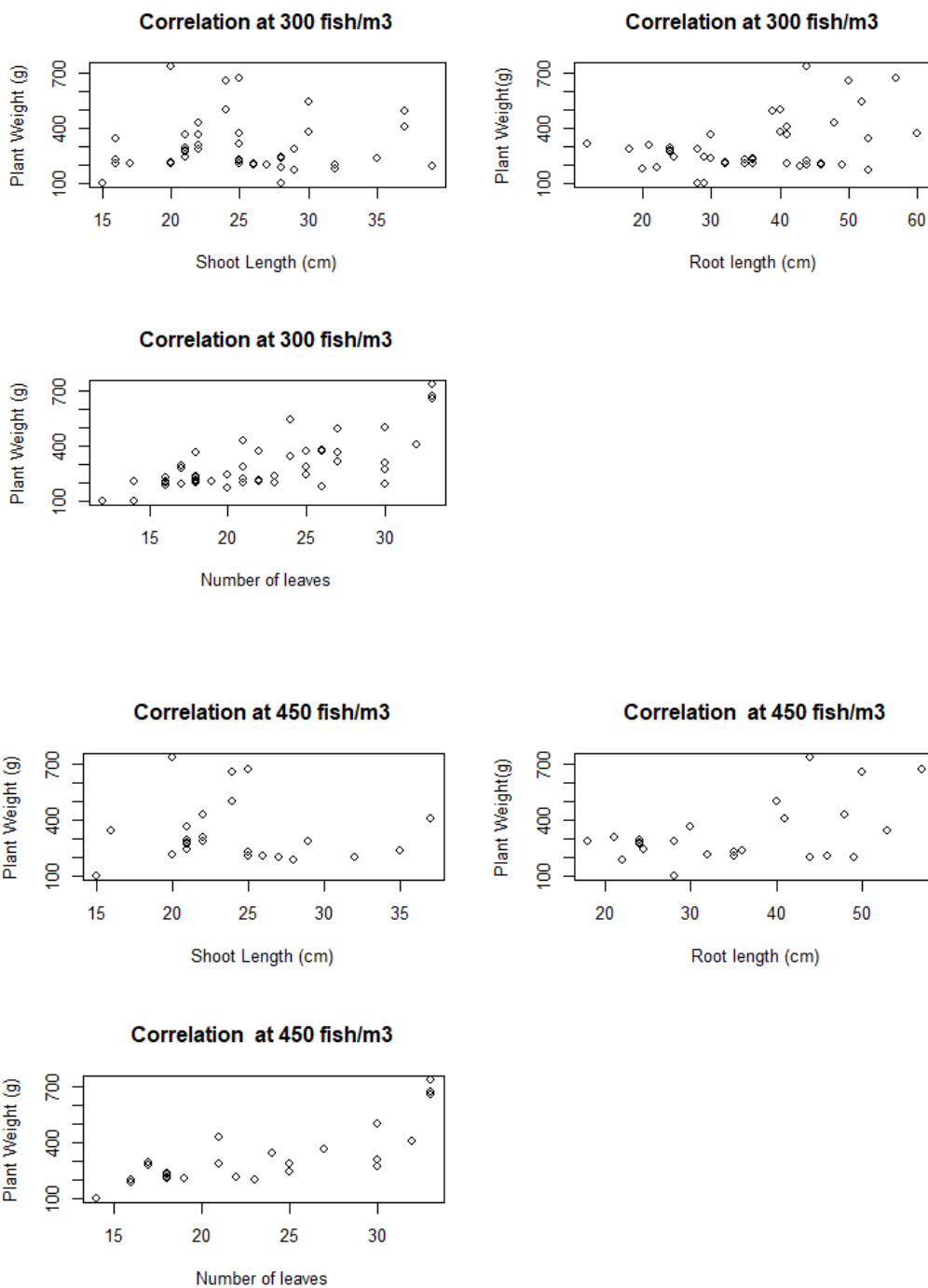
Correlation with pulled data: Plant weight vs Shoot length, Root length and Number of leaves



Correlation at 150 fish/m3



Correlation at 300 fish/m³



Correlation at 450 fish/m³

Appendix VII: Comparison of Regression Lines - Weight versus Leaves by Treatment

Dependent variable: Weight

Independent variable: Leaves

Level codes: Treatment

Number of complete cases: 120

Number of regression lines: 3

Multiple Regression Analysis

		<i>Standard</i>	<i>T</i>	
<i>Parameter</i>	<i>Estimate</i>	<i>Error</i>	<i>Statistic</i>	<i>P-Value</i>
CONSTANT	- 76.8394	68.3884	-1.12357	0.2636
Leaves	12.3634	3.42059	3.6144	0.0004
Treatment=300 fish/m3	90.7921	90.035	1.00841	0.3154
Treatment=450 fish/m3	- 56.0199	83.4945	-0.670941	0.5036
Leaves*Treatment=300 fish/m3	0.18238	4.37921	0.0416467	0.9669
Leaves*Treatment=450 fish/m3	7.32035	4.0058	1.82743	0.0703

Coefficients

<i>Treatment</i>	<i>Intercept</i>	<i>Slope</i>
150 fish/m ³	-76.8394	12.3634
300 fish/m ³	13.9526	12.5458
450 fish/m ³	-132.859	19.6837

Because the P-value in the ANOVA table is less than 0.05, there is a statistically significant relationship between the variables at the 95.0% confidence level.

Analysis of Variance

<i>Source</i>	<i>Sum of Squares</i>	<i>Df</i>	<i>Mean Square</i>	<i>F-Ratio</i>	<i>P-Value</i>
Model	1.15544E6	5	231089.	39.13	0.0000
Residual	673296.	114	5906.1		
Total	1.82874E6	119			

(Corr.)

The R-Squared statistic indicates that the model as fitted explains 63.1825% of the variability in Weight. The adjusted R-Squared statistic, which is more suitable for comparing models with different numbers of independent variables, is 61.5677%. The standard error of the estimate shows the standard deviation of the residuals to be 76.8512. This value can be used to construct prediction limits for new observations by selecting the Forecasts option from the text menu. The mean absolute error (MAE) of 55.343 is the average value of the residuals. The Durbin-Watson (DW) statistic tests the residuals to determine if there is any significant correlation based on the order in

which they occur in your data file. Since the P-value is greater than 0.05, there is no indication of serial autocorrelation in the residuals at the 95.0% confidence level. To test for statistically significant differences between the intercepts and/or slopes, select Conditional Sums of Squares from the list of Tabular Options.

R-Squared = 63.1825 percent

R-Squared (adjusted for d.f.) = 61.5677 percent

Standard Error of Est. = 76.8512

Mean absolute error = 55.343

Durbin-Watson statistic = 2.24366 (P=0.9084)

Lag 1 residual autocorrelation = -0.12737

Combined equation

$$\begin{aligned} \text{Weight} = & -76.8394 + 12.3634 * \text{Leaves} + 90.7921 * (\text{Treatment}=300 \text{ fish/m}^3) - \\ & 56.0199 * (\text{Treatment}=450 \text{ fish/m}^3) + 0.18238 * \text{Leaves} * (\text{Treatment}=300 \text{ fish/m}^3) + \\ & 7.32035 * \text{Leaves} * (\text{Treatment}=450 \text{ fish/m}^3) \end{aligned}$$

At 150 fish/m³ the model reduces to

$$\text{Plant Weight (g)} = -76.8394 + 12.3634 * \text{Leaves}$$

At 300 fish/m³, the model reduces to

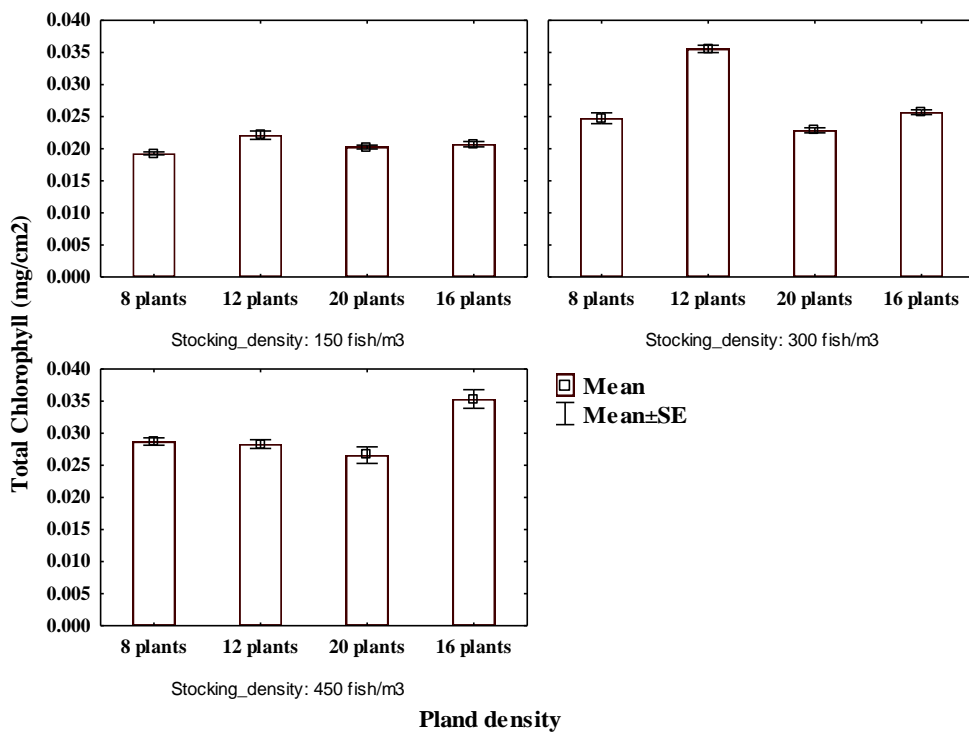
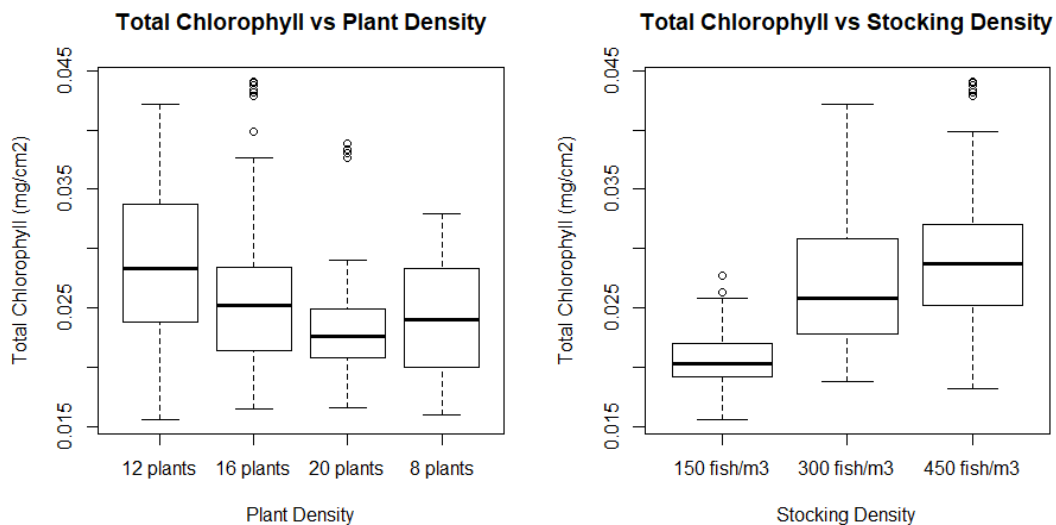
$$\text{Plant Weight (g)} = 13.9526 + 12.5458 * \text{Leaves}$$

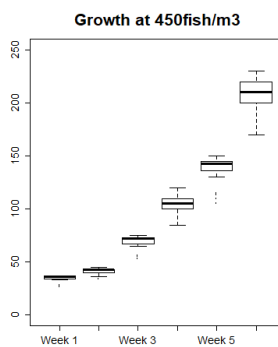
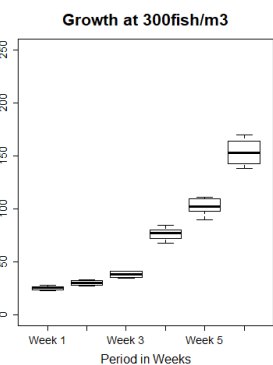
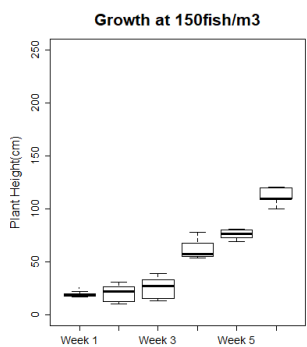
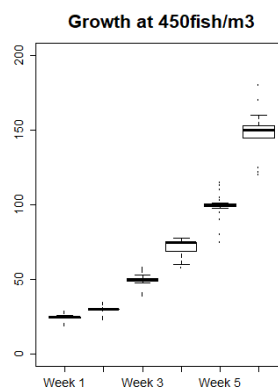
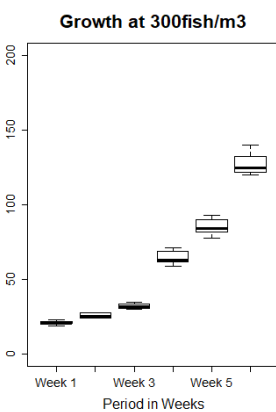
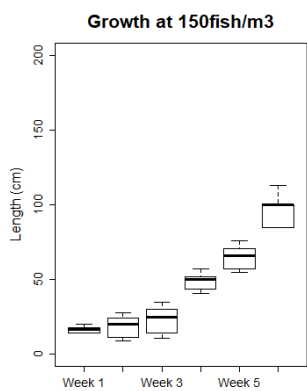
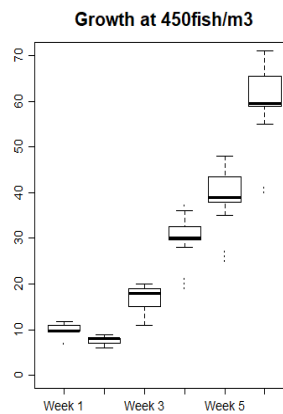
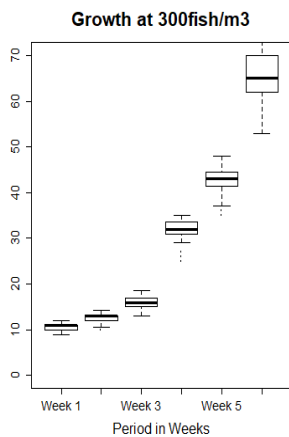
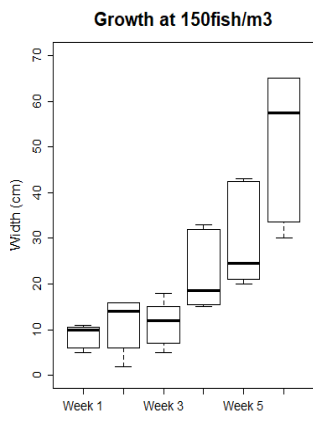
At 450 fish/m³, the model reduces to

$$\text{Plant Weight (g)} = -132.859 + 19.6837 * \text{Leaves}$$

Appendix VIII: Results from the pilot study

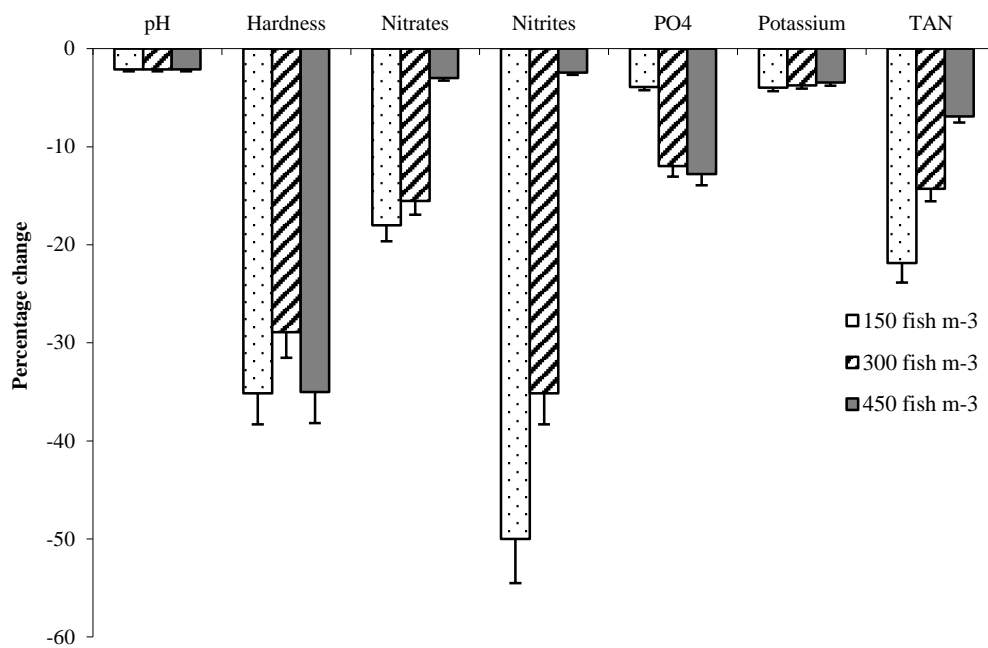
Comparison of the plant density on chlorophyll content of lettuce





Appendix IX: Regression outputs comparing changes in nitrate in the aquaponics system in relation to other studied parameters

	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>
Intercept	0.5233	2.8477	0.1837	0.8543	-5.0668	6.1133
Days	-0.0106	0.0049	-2.1629	0.0309	-0.0202	- 0.0010
Stocking density	-0.0014	0.0002	-5.6183	0.0000	-0.0019	- 0.0009
Fish weight	0.0882	0.0108	8.1507	0.0000	0.0670	0.1095
Chlorophyll	1.5565	3.2668	0.4764	0.6339	-4.8563	7.9692
Height of plant	0.0301	0.0050	6.0259	0.0000	0.0203	0.0399
Length	-0.0119	0.0067	-1.7837	0.0749	-0.0250	0.0012
pH (HYDROP)	-0.1736	0.3819	-0.4547	0.6495	-0.9233	0.5760
Nitrite (AQUAP)	-0.1259	0.3501	-0.3596	0.7193	-0.8132	0.5614
Hardness (AQUAP)	-0.0033	0.0006	-5.3849	0.0000	-0.0045	- 0.0021

Appendix 1: Nutrient removal in the aquaponic system

Appendix 2: Regression statistics

Nitrate in Aquaponic						
<i>Regression Statistics</i>						
Multiple R	0.6415					
R Square	0.4115					
Adjusted R Square	0.4047					
Standard Error	0.5256					
Observations	789					
<i>ANOVA</i>						
	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>	
Regression	9	150.4770	16.719	60.5	9.84218	
		419	6713	129	E-84	
Residual	779	215.2369	0.2762			
		952	991			
Total	788	365.7140				
		371				
	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>
Intercept	0.5233	2.8477	0.1837	0.8543	-5.0668	6.1133
Days	-0.0106	0.0049	-2.1629	0.0309	-0.0202	0.0010
SD	-0.0014	0.0002	-5.6183	0.0000	-0.0019	0.0009
Weight	0.0882	0.0108	8.1507	0.0000	0.0670	0.1095
Chlor	1.5565	3.2668	0.4764	0.6339	-4.8563	7.9692
Height_plant	0.0301	0.0050	6.0259	0.0000	0.0203	0.0399
Length	-0.0119	0.0067	-1.7837	0.0749	-0.0250	0.0012
pH(HYDROP)	-0.1736	0.3819	-0.4547	0.64	-0.9233	0.5760

				95		
Nitrite(mg/l -N)-AQUAP	-0.1259	0.3501	-0.3596	0.71	-0.8132	0.5614
				93		
Hardness(AQUAP)	-0.0033	0.0006	-5.3849	0.00	-0.0045	-
				00		0.0021

Nitrate in hydroponics

Regression Statistics

Multiple R	0.6179
R Square	0.3818
Adjusted R Square	0.3746
Standard Error	0.0604
Observations	789

ANOVA

	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	9	1.752027053	0.19466967	53.4477	1.61462E-75
Residual	779	2.837310493	0.00364225		
Total	788	4.589337546			

	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>
Intercept	-0.8611	0.3270	-2.6337	0.0086	-1.5029	-0.2193
Days	0.0053	0.0006	9.4036	0.0000	0.0042	0.0064
SD	0.0002	0.0000	6.1319	0.0000	0.0001	0.0002
Weight	-0.0067	0.0012	-5.4276	0.0000	-0.0092	-0.0043

Chlor	0.5345	0.3751	1.4250	0.01	-0.2018	1.2707
				55		
Height_plant	-0.0063	0.0006	-	0.00	-0.0074	-
			10.938	00		0.0051
			8			
Length	0.0068	0.0008	8.8633	0.00	0.0053	0.0083
				00		
pH(HYDROP)	0.1242	0.0438	2.8332	0.00	0.0382	0.2103
				47		
Nitrite(mg/l -N)-AQUAP	0.3634	0.0402	9.0400	0.00	0.2845	0.4423
				00		
Hardness(AQUAP)	0.0000	0.0001	0.0119	0.99	-0.0001	0.0001
				05		

**Phosphates in the
aquaponics**

Regression Statistics

Multiple R	0.72317
	6481
R Square	0.52298
	4222
Adjusted R	0.51747
Square	3128
Standard Error	0.19783
	4198
Observations	789

ANOVA

	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	9	33.42689	3.714099	94.89663	6.5562E-
		474	415		119
Residual	779	30.48879	0.039138		
		013	37		
Total	788	63.91568			

487

	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>
Intercept	0.2325	1.0718	0.2169	0.8283	-1.8714
Days	0.0075	0.0018	4.0670	0.0001	0.0039
SD	-0.0007	0.0001	-7.9263	0.0000	-0.0009
Weight	0.0004	0.0041	0.1005	0.9200	-0.0076
Chlor	-5.6846	1.2295	-4.6235	0.0000	-8.0981
Height_plant	-0.0105	0.0019	-5.5790	0.0000	-0.0142
Length	0.0169	0.0025	6.7206	0.0000	0.0120
pH(HYDROP)	0.1998	0.1437	1.3902	0.1649	-0.0823
Nitrite(mg/l -N)-	1.2075	0.1318	9.1633	0.0000	0.9488
AQUAP					
Hardness(AQUAP)	-0.0039	0.0002	-17.1546	0.0000	-0.0044

Phosphate in hydroponics

Regression

Statistics

Multiple R	0.76813
	869
R Square	0.59003
	7047
Adjusted R Square	0.58530
	0633
Standard Error	0.20114
	5178
Observations	789

ANOVA

	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	9	45.3619	5.0402	124.5746	2.3697E-

					144	
Residual	779	31.5179	0.0405			
Total	788	76.8798				
	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>
Intercept	0.7829	1.0897	0.7184	0.4727	-1.3563	2.9220
Days	0.0315	0.0019	16.8717	0.0000	0.0279	0.0352
SD	0.0005	0.0001	5.2789	0.0000	0.0003	0.0007
Weight	-0.0058	0.0041	-1.4103	0.1589	-0.0140	0.0023
Chlor	-5.0911	1.2501	-4.0726	0.0001	-7.5451	-
						2.6372
Height_plant	-0.0191	0.0019	-10.0142	0.0000	-0.0229	-
						0.0154
Length	0.0204	0.0026	7.9930	0.0000	0.0154	0.0254
pH (HYDROP)	0.1350	0.1461	0.9241	0.3557	-0.1518	0.4219
Nitrite (mg/l - N)-AQUAP	0.5355	0.1340	3.9970	0.0001	0.2725	0.7985
Hardness(AQUAP)	-0.0055	0.0002	-23.9151	0.0000	-0.0060	-
						0.0051
Ammonia in aquaponics						
<i>Regression Statistics</i>						
Multiple R	0.2705					
R Square	0.0732					
Adjusted R Square	0.0625					
Standard Error	0.0816					
Observations	789					
ANOVA						
	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>	
Regression	9	0.4093	0.0455	6.8344	1.69916	

					E-09	
Residual	779	5.1833	0.0067			
Total	788	5.5926				
	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>
Intercept	1.4242	0.4419	3.2229	0.0013	0.5567	2.2917
Days	0.0009	0.0008	1.1862	0.2359	-0.0006	0.0024
SD	0.0000	0.0000	-0.6645	0.5066	-0.0001	0.0000
Weight	0.0018	0.0017	1.0772	0.2817	-0.0015	0.0051
Chlor	-2.1369	0.5069	-4.2153	0.0000	-3.1321	-
						1.1418
Height_plant	0.0011	0.0008	1.4783	0.1397	-0.0004	0.0027
Length	-0.0018	0.0010	-1.7389	0.0824	-0.0038	0.0002
pH (HYDROP)	-0.1902	0.0593	-3.2088	0.0014	-0.3065	-
						0.0738
Nitrite(mg/l -N)- AQUAP	0.1167	0.0543	2.1484	0.0320	0.0101	0.2234
Hardness(AQU AP)	0.0002	0.0001	2.2876	0.0224	0.0000	0.0004

Ammonia in hydroponics

Regression Statistics

Multiple R	0.3083
R Square	0.0951
Adjusted R Square	0.0846
Standard Error	0.0576
Observations	789

ANOVA

	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	9	0.2719	0.0302	9.09	3.721E-43
				43	13

Residual	779	2.5881	0.0033			
Total	788	2.8600				
	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>
Intercept	0.9908	0.3123	3.1729	0.0016	0.3778	1.6038
Days	0.0036	0.0005	6.6848	0.0000	0.0025	0.0046
SD	0.0001	0.0000	3.2087	0.0014	0.0000	0.0001
Weight	-0.0012	0.0012	-0.9837	0.3256	-0.0035	0.0012
Chlor	-2.3160	0.3582	-6.4654	0.0000	-3.0192	1.6128
Height_plant	0.0004	0.0005	0.6712	0.5023	-0.0007	0.0014
Length	-0.0013	0.0007	-1.7971	0.0727	-0.0028	0.0001
pH(HYDROP)	-0.1213	0.0419	-2.8963	0.0039	-0.2035	0.0391
Nitrite(mg/l -N)-AQUAP	0.0034	0.0384	0.0893	0.9288	-0.0719	0.0788
Hardness(AQUAP)	-0.0001	0.0001	-1.0085	0.3135	-0.0002	0.0001

Appendix XII: Table: Enterprise budget table used during Sensitivity analysis

	Fish stocking density		
	150	300	450
Mean fish yield (kg/m ³)	6	8.56	10.33
Mean Lactuca sativa yield (kg/m ³)	1.66	2.76	3.04
Total revenue from fish (Kshs)	4200	5992	7231
Revenue from Lactuca sativa	498	828	912
Gross revenue	4698	6820	8143
Variable costs			
Cost of feeds	132.54	186.9	208.392
Field labour	1500	1500	1500
Cost of maintenance of equipment	350	350	350
Water	650	650	650
Electricity	800	800	800
Miscellaneous	600	600	600
Sub-total variable costs	4032.5	4086.9	4108.4
Interest on operating cost	725.857	735.642	739.51056
Total variable cost (TVC)	4758.4	4822.5	4847.9
Fixed costs			
Bench platform	800	800	800
Amortization	200	200	200
Interest on the fixed cost	180	180	180
Total fixed cost	1180	1180	1180
Total cost (TC)	5938.4	6002.5	6027.9
Net returns above TVC	-60.4	1997.5	3295.1
Net returns above TC	-1240.4	817.5	2115.1
Break even	196.7	137.9	114.2
Stocking density	150	300	450
Kg of fish sold	6	8.56	10.33

Price/kg	400	400	400
Costs	2750	2800	2850
Total revenue	2900	4224	5032
Profit	150	1424	2182
	750		
	0.04	0.028533333	0.022955556

Appendix XIII: Correlation coefficients showing the nitrate in the aquaponics system in relation to other studied parameters

Nutrients	Nitrate (AQUA)	Nitrite (AQUA)	NO ₃ (HYDR)	Nitrite (HYDR)	PO ₄ (AQUA)	PO ₄ (HYDR)	K (AQUAP)	K (AQUA)	Ammonia (AQUA)	Ammonia (Plant)
Nitrate (AQUA)	1									
Nitrite (AQUA)	-0.4485	1								
NO ₃ (HYDR)	0.7073	-0.3799	1							
Nitrite (HYDR)	-0.0082	0.3535	-0.2314	1						
PO ₄ (AQUAP)	-0.1147	0.2073	0.0699	0.0231	1					
PO ₄ (HYDR)	-0.1387	0.3264	-0.0529	0.2208	0.8820	1				
K(AQUAP)	-0.1227	0.3316	-0.0030	0.1492	0.6361	0.5745	1			
K (HYDR)	-0.1573	0.1581	0.1797	0.2684	0.4165	0.3541	0.3274	1		
Ammonia (AQUA)	-0.0478	-0.0565	-0.0085	-0.1591	-0.1719	-0.2373	-0.1000	-0.3284	1	
Ammonia (Plant)	0.0108	-0.1966	-0.0720	-0.1274	-0.0708	-0.0678	-0.1565	-0.3077	0.7963	1

Where: AQUA and HYDR represents the fish and the plant component in the experiment respectively.

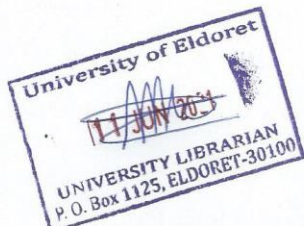
Appendix 3: Similarity report

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