STUDIES ON THE EFFECTS OF *HIPPOPOTAMUS AMPHIBIOUS* VECTORED SUBSIDIES ON THE ECOLOGY OF AQUATIC ECOSYSTEM

BY

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A DISSERTATION SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF SCIENCE IN BIOCHEMISTRY OF SOKOINE UNIVERSITY OF AGRICULTURE MOROGORO, TANZANIA

2015
A study was conducted along the Great Ruaha River (GRR) at Ruaha National Park (RNP) for a period of two months (November to December 2013) with the main aim of assessing the effects of Hippopotamus amphibious vectored subsidies on the chemistry of river ecosystems and the ecology of aquatic resident within these systems. The study was done by measuring river water nutrients, physical water chemistry parameters, and stable isotope analysis of aquatic consumers. Study sites were identified in two basic treatment types: river pools containing hippos and those showing no evidence of hippo occupancy. Response parameters that were measured included total dissolved nitrogen (TDN), total dissolved phosphorus (TDP), dissolved organic carbon (DOC), nitrate (NO₃), and ammonia (NH₃). Total dissolved nitrogen, total dissolved phosphorus, and nitrate were significantly elevated in hippo pools relative to non hippo pools (P<0.01). Ammonia, and dissolved organic carbon were not statistically significant between hippo and non hippo pools but their absolute levels were slightly higher in hippo pools. Physical parameters of the GRR were also assessed including measures of dissolved oxygen (DO), pH, conductivity, temperature, and chromophoric dissolved organic matter (CDOM). Dissolved oxygen, pH, and conductivity were significantly higher in the hippo pools compared to non hippo pools (P<0.001). Temperature and CDOM did not vary with hippo density (P>0.05). Stable isotopes of carbon (δ¹³C) and nitrogen (δ¹⁵N) were also assessed during the study to examine whether this technique could help us identify use of hippo delivered nutrients by river consumers such as fish. Carbon and nitrogen stable isotopes were not significantly different for the fish sampled in hippo and non hippo pools. This lack of difference may derive from saturation of C4 plant derived δ¹³C in the GRR ecosystem. This study provides for the first time more insight into the influence of hippo vectored organic material in aquatic ecosystem dynamics in the GRR.
DECLARATION

I, JAMES MPEMBA, do hereby declare to the Senate of Sokoine University of Agriculture that this dissertation is my own original work done within the period of registration and that it has neither been submitted nor being concurrently submitted in any other institution.

James Mpemba (MSc Student)  

The above declaration is confirmed

Prof. B.M.Mutayoba (Supervisor)
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DEDICATION

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<tr>
<td>GPS</td>
<td>Global Positioning System</td>
</tr>
<tr>
<td>ml</td>
<td>Millilitre</td>
</tr>
<tr>
<td>TDN</td>
<td>Total dissolved nitrogen</td>
</tr>
<tr>
<td>G</td>
<td>Gramm(s)</td>
</tr>
<tr>
<td>μgl⁻¹</td>
<td>Micrograms per litre</td>
</tr>
<tr>
<td>mgl⁻¹</td>
<td>Milligrams per litre</td>
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<tr>
<td>μScm⁻¹</td>
<td>Micro Siemens per centimetre</td>
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<td>mm</td>
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<td>%</td>
<td>Percent</td>
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<td>m²</td>
<td>Meter square</td>
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<tr>
<td>Km</td>
<td>Kilometre</td>
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<tr>
<td>MSc</td>
<td>Master of Science</td>
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<tr>
<td>SUA</td>
<td>Sokoine University of Agriculture</td>
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<tr>
<td>TANAPA</td>
<td>Tanzania National Parks</td>
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<tr>
<td>WMA</td>
<td>Wildlife Management Area</td>
</tr>
<tr>
<td>CDOM</td>
<td>Chromophoric Dissolved Organic Matter</td>
</tr>
<tr>
<td>TAWIRI</td>
<td>Tanzania Wildlife Research Institute</td>
</tr>
<tr>
<td>°C</td>
<td>Degree centigrade</td>
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<tr>
<td>DO</td>
<td>Dissolved oxygen</td>
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<tr>
<td>SIA</td>
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<tr>
<td>TDP</td>
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<td>POC</td>
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<td>PO₄-P</td>
<td>Orthophosphate Phosphorus</td>
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<tr>
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<td>Temperature</td>
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<td>EC</td>
<td>Electrical Conductivity</td>
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<td>that is</td>
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CHAPTER ONE

1.0 INTRODUCTION

The hippopotamus (*Hippopotamus amphibious*) is a large, amphibious mammal and common resident in lakes and rivers bordered by grassland throughout sub-Saharan Africa (Grey and Harper, 2002). Hippos are typically herbivorous; selective nocturnal grazers and due to their large body size, they require large areas of grass often exceeding 5 hectares to satisfy their feeding status (Eltringham, 1999). Adults are capable of ingesting approximately 40 kg of short sward grasses throughout nocturnal forays of the catchment (Eltringham, 1999). Hippos are very close croppers and will graze on stands of grass until they are of lawn-like appearance. This is accomplished with the horny edges of the lips where grasses are plucked with an upward movement of the head. Where hippo populations are high such as in the Luangwa valley (Zambia), the density may exceed 42 hippos per km stretch of the river (Tembo, 1987), here hippos require large tracts of land to meet their daily food requirement (Owen-Smith, 1988). Grass biomass and grazing capacity are therefore, considered as one of the major environmental factors limiting hippo population size and density.

Historically, the common hippopotamus occupied an extensive range in Africa and was considered abundant but to date, though the species still occurs widely throughout the continent, its dispersion is patchy and uneven.
There are apparent regional differences in population size and distribution of common hippos in Africa. East African countries (including Uganda, Kenya, Tanzania, Mozambique, and Zambia) form the conservation stronghold for this species and are where the largest numbers of Common Hippos occur. Chomba (2012) estimated the total population of Africa at about 157 000 animals of which 7 000 occurred in West Africa, 70 000 in East Africa and 80 000 in Southern Africa. Although common hippos are found in many West African nations, overall population sizes tend to be much smaller. Of the 36 countries where the common hippopotamus is known to occur, 20 have confirmed declining populations, seven have populations of unknown status, nine have stable populations and three (Algeria, Egypt and Mauritania) have experienced extinctions.
(Lewison and Oliver, 2008). The largest declines have occurred in the Democratic Republic of Congo, a country once thought to have the largest populations (Lewison and Oliver, 2008). Information from The World Conservation Union (IUCN) surveys suggests that, there is a shocking sharp decline in common hippos to countries whose number was thought to be higher (Lewison and Oliver, 2008). These declines have been attributed to two anthropogenic activities: habitat loss as wetlands are converted or impacted by agricultural development and unregulated hunting for meat and ivory.

Kendall (2011) reported that, hippopotamus population in some African countries have fallen sharply to as much as 95%. Furthermore, climate change is predicted to bring major droughts in parts of Africa, reducing available habitat and increasing human–hippopotamus competition for water (Lewison, 2007). Considering the likelihood of additional stress on hippopotamus populations from water shortages, conservation of the species will depend on active management of human–hippopotamus conflict (Eltringham, 1999). In Tanzania, common hippos are widely dispersed, they are found in most National Parks and reserves, with a substantial number in Ruaha National Park (RNP), and although not present anywhere in large numbers, the total probably amounts to several thousands.

Categorized as vulnerable on the IUCN Red List, hippos are under considerable pressure, the primary threats being illegal and unregulated hunting for meat and ivory and habitat loss (Lewison and Oliver, 2008). Illegal or unregulated hunting of Common Hippos has been found to be particularly high in areas of civil unrest (IUCN Red List of Threatened Species). Hippos rely heavily on freshwater bodies, making them vulnerable to drought, agricultural, and rerouting of natural water flows.
Hippos play a key role in the Great Ruaha River (GRR) ecosystems as they shape vegetation structure on land with its nightly grazing habit. Much of the hippo terrestrially derived organic matter may fertilize aquatic ecosystems as they defecate in them during daytime. They may provide a constant subsidy to the system in form of nutrient elimination via egestion and excretion from grass consumed in surrounding terrestrial ecosystem and defecated into the river. These materials may then be regulated by the hydrological pattern into which they are deposited.

The natural habitat for hippo in the GRR continues to get smaller and smaller as humans continue to take over land and water in the wetland that hippo used to settle for agricultural activities (Lewison and Oliver, 2008). In dry season, hippos are forced to move upstream the GRR in search for pools for them to respite as most of the hippo pools dry up. They require water deep enough to cover them, in pools that are close to pasture for grazing. Hippo avoid fast-moving waters, whilst gently-sloping, firm-bottomed beds are preferred, where hippo herds can rest half-submerged and calves can nurse without swimming. Submerging for hippo in water is necessary to prevent their thin, hairless skin from overheating and dehydration from the hot environment of the Great Ruaha during the dry season.
Plate 1: Hippo herd in one of the pools in the Great Ruaha River.

(Source: James Mpemba, 2013)

In RNP, nothing is more important to the common hippos than plentiful water for their constancy. When the GRR and the wetland are healthy, common hippos will thrive and flourish. The future of the Great Ruaha ecosystem and its ecological integrity is in the hands of the people who live and depend on its land and water. Maintaining a healthy and stable ecosystem in the GRR is vital to the livelihood and well being of the common hippos as they play the major role in shaping, protecting and stabilizing the entire ecosystems. Failure to conserve, protect, and restore this valuable mega-mammal, may be putting the whole ecosystem at risk.

As hippos control the entire ecosystem in the GRR by moving nutrients between aquatic and terrestrial environment, its disappearance could affect the whole wildlife biodiversity as hippo dung provides nutrients to flora and fauna in the river.
The presence of substantial number of hippo herds in Tanzanian watersheds is likely having major impacts on watershed ecology; however there is a paucity of information on the nature, magnitude, dynamics, or mechanisms of these effects (Naiman and Rogers, 1997; Jacobs et al., 2007). We still lack useful information on how these amphibious mega-mammals connect the aquatic and terrestrial ecosystems they traverse, how they move materials across these boundaries, what the ecological and biogeochemical significance is of these transfers, or what other emergent effects that their presence may be having on watershed ecosystem ecology. Because of their amphibious life, any changes in river hydrology will affect their population sizes, movement, and biochemical effects they have on the whole ecosystems.

1.1 Problem Statement and Justification of the Study

The hydrology of RNP watersheds are greatly in flux as a result of disturbance and climatological change in recent decades. The health of this river and water resources govern important properties of wildlife ecology. In particular the ecology of the resident common hippopotamus is likely to be greatly affected by change to these river systems. Furthermore, the effects of hydrology on hippos are likely to cause numerous cascading effects on the river systems that hippos themselves influence. The focus of this research was to address these issues.

The variability of the river system requires that we make creative efforts to understand how physical forcing affects the chemistry, utilization, and attenuation of subsidy delivery, as well as the nascent ecological effects of the subsidies. Hippos are certain to have many spatially and temporally variable effects on the regional ecology of these systems. Hence, I challenged myself to make integrative descriptions of these dynamic effects and to identify the major causes of this variation. Results from this work are
envisaged to make an important contribution to the understanding of the ecological importance of the largest faunal components of ecosystems. The Hippopotamid lineage has been greatly reduced in diversity and distribution in the past several million years. Uncovering the effects of extant hippos would provide a first source of information on what ecosystem level functions went globally or regionally extinct as results of the reduction and simplification of this ecologically unique lineage. It is anticipated that results from this research might fundamentally improve the understanding of the key mechanisms and dynamic processes that control the ecology of watersheds, provide quantitative insight into the contemporary and historical controls that strong interactors have on watershed ecosystems, and create an important stimulus for future ground-breaking advancements in watershed science.

Results from this whole-system investigation of hydrological patterns, ecosystem connections, material fluxes, and ecological relationships will likely make an impact on our understanding of watershed health, hippo ecology, and watershed-landscape dynamics in Tanzania and across an entire continent. There is much opportunity for results from this research to positively impact the environment of Tanzania and the societies that depend on Great Ruaha River. Some speculation has already been generated about the possible provision of key ecosystem services by hippos e.g. providing nutrient subsidies that enhance local fisheries (Mosepele et al., 2009; Schrank, 2009), but this study provides the first scientific examination of these putative connections. Results from this work will likely greatly improve our understanding of the possible impacts that climate change and future water use patterns may have on hippos and the ecosystem-wide processes that hippo control. Most studies have concluded that climate change and human population growth continue to reduce already low river water flow rates (Ritchie et al. 2008). If these projections are accurate, we will need data to understand and respond to the figurative and
literal “downstream” ecosystem impacts of this change. My hope is that this data will improve our understanding of the ecology of this river, better comprehend the factors that impact these rivers, and more clearly see how this change in rivers may cause cascading effects on whole watershed ecosystems. Results from this work will also contribute towards the development of a new understanding of how a single strong interactor can determine whole ecosystem processes. All indications suggested that hippos may prove to be one of the most important biological forces in watershed ecology yet studied.

1.2 Objectives of the Study

1.2.1 General Objective

The general objective of the study was to determine how hippos directly and indirectly affect the ecology of aquatic ecosystem by vectoring nutrients and consuming materials.

1.2.2 Specific Objectives

Specifically the study intended to;

i. Examine the relationship between hippo density and nutrients measured in the Great Ruaha River waters.

ii. Assess how hippo density affects other important attributes of water chemistry of the Great Ruaha River.

iii. Examine the effect of hippo density on stable isotopes of carbon and nitrogen of consumers in the Great Ruaha River waters.
CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Biological information of *Hippopotamus amphibious*

*Hippopotamus amphibious* is a large river-living mammal in tropical Africa, and one of only two extant species in the family Hippopotamidae. Hippo is classified in the phylum Chordata, subphylum Vertebrata, class Mammalia, order Artiodactyla, and family Hippopotamidae. They are found exclusively in rivers throughout the savannah of Africa, particularly in the sub-Saharan Africa.

The common hippopotamus has a barrel-shaped body, smooth hairless skin and short stout legs, an adaptation to move swiftly on water and land, whilst their short stout legs provide powerful actuation through water, and their webbed feet allow them to navigate on shallow river bottoms. The mouth is wide, and the incisors and lower canines are large ivory tusks that grow throughout their life.

![Plate 2: A barrel-shaped body of *Hippopotamus amphibious* and its front view.](image)

*Source: James Mpemba, 2013*
Common hippos are among the largest mammals with males achieving body weights greater than 2 500 kg (Parker, I.S.C. personal communication, 2005). The tail is abbreviated and flattened with a sparse fringe of bristles at the tip. Location of eyes, ears, and nostrils high on their head allows them to remain mostly submerged while still being able to breathe and stay aware of their surroundings. Hippo closes its nostrils and folds its ears when it submerges into the water to prevent water from entering them.

The skin has a unique structure in that, they lack scent and sweat glands, and instead they have mucous glands which secrete a thick oily layer of red pigmented fluid. The secretion is sometimes referred to as "blood sweat", but is neither blood nor sweat. This fluid is a combination of hipposudoric acid and norhipposudoric acid which create a sunscreen effect by absorbing ultraviolet rays from the sun and prevent the growth of disease causing bacteria. The secretion is initially colourless and turns red-orange within minutes, eventually becoming brown (Coughlin and Fish, 2009). The colour of the skin is greyish black with a pink tinge; the skin around the eyes and ears is pinkish-yellow and the gape of the mouth is flesh-coloured (Arman and Field, 1973). The alimentary canal of the hippo is able to break down the tough cellulose which makes up a large part of its diet. The stomach consists of four chambers which function like those of ruminants with micro-organisms fermenting and producing enzymes which break down cellulose (Arman and Field, 1973). Hippos do not ‘chew the cud’ and are known as ‘pseudo ruminants’.

Lewison and Oliver (2008) described the reproductive strategy of the hippo as one well adapted to the semi-arid environments of Africa. They are polygynous, meaning that one bull mates with several females in the social group. Although breeding is not strictly seasonal, conception usually occurs during the dry season, between February and August, and births usually occur during the rainy season, between October and April.
(de Magalhaes and Costa, 2009). When resources become limiting, hippo maintain stable populations by delayed sexual maturity and fecundity and hence adjust to the carrying capacity of the environment. Hippo populations are capable of rapid increase when resources become abundant. Therefore, this finding has important implications for hippo management in Ruaha National Park.

Hippo population dynamics is sensitive to annual rainfall variability, as during the dry season the population tend to be small in number because of lack of forage, heat stress, and increased vulnerability to diseases (Chomba et al., 2013). Conversely, in response to higher than average rainfall, common hippo populations exhibit dramatic population expansion. Information from The World Conservation Union (IUCN) surveys suggests that, there is a shocking sharp decline in common hippos to countries whose number was known to be higher (Lewison and Oliver, 2008). These declines have been attributed to two anthropogenic activities: habitat loss as wetlands are converted or impacted by agricultural development and unregulated hunting for meat and ivory.

Tanzania is one among countries hosting a great number of hippos which are estimated to be 20 000-30 000 and are found in National Parks (NP) and Game reserves (GR). This population is distributed in the following areas, Ruaha NP, Lake Manyara NP, Arusha NP, Serengeti NP, Mikumi NP, Rubondo NP, Tarangire NP, Burigi GR, Maswa GR, Biharamulo GR, Selous GR, Moyowozi GR, and Ngorongoro Conservation Area. Ruaha NP is one of the parks in the country hosting large number and provides ideal environment for hippopotamuses to flourish. The large number of hippos in Tanzania is attributed to strict protection and legal enforcement provided by the government. Poaching has continued to be the major challenge facing national parks in Tanzania. In particular, the recent poaching wave involving killing elephants in almost all national
parks in the country. Sport hunting and poaching of hippos in Tanzania is rare and is less commonly reported. In addressing this problem of escalated poaching, TANAPA has invariably increased and diversified anti-poaching strategies over the years, including increasing budgetary allocations, increasing the number of rangers and strengthening intelligence gathering and prosecution activities. Provision of innovative intervention approaches to rangers in these areas has also been stepped up.

2.2 Hippo habitats

The daily Hippo movements between water and the grazing range result in trampling and erosion to river banks. When an exit point from a river becomes unusable due to erosion, hippo will begin another nearby and, over a number of years; this process can reshape rivers. With sufficient grazing available, hippos tend to remain close to rivers. However, drought, arid conditions or competition with humans may cause hippo to seek resources some distance from their daily living space. Grazing pressure tends to decrease with distance from water (Lewison and Carter, 2004). The significance of the relationship between daily living space and grazing range is that simple comparisons of hippo densities or attempts to assess carrying capacity are invalid without an understanding of the constraints affecting the daily living space of each population.

2.3 Historical background of the Great Ruaha River

The Great Ruaha River (GRR) rises in Tanzania’s Kipengere mountains, flows through the Usangu plains (a very important region for irrigated agriculture, mostly rice), and along the eastern boundary of Ruaha National Park - now larger than the Serengeti after the 2008 extension with an area today of 200 226 square km. This area is also home to a rich variety of wildlife, including: greater and lesser kudus, giraffes, zebras, elephants, hippos, African wild dogs, cheetahs, leopards, lions and over 400 bird species. They all
depend on the water from the Ruaha, as do the six million people who live near the river and rely on it for drinking water, fish, water for their crops and livelihoods. Great Ruaha is about 475 km (300 mi.) long, its tributary basin has a catchment area of 68,000 km² and the mean annual discharge is 140 m³/s (Mtahiko et al., 2006). The GRR supplies 22% of the total flow of the Rufiji catchment system. Farming is the main source of income for around 90% of the basin’s population, who grow crops like rice, maize, beans, vegetables, groundnuts, fruits, millet and potatoes.

The park is part of the Rungwa-Kizigo-Muhesi ecosystem, which includes Rungwa Game Reserve, Kizigo and Muhesi Game Reserves, the Mbomipa Wildlife Management Area, and several other protected areas (RNP, 2014). The rivers’ headwaters are in the Kipengere Mountains. From there the river descends to the Usangu plains, an important region for irrigated agriculture and livestock in Tanzania (RNP, 2014). The Great Ruaha is the main tributary of the Rufiji basin - an important basin for Tanzania, contributing to about 50 percent of the country’s electricity supply, via the Mtera and Kidatu dams. It is also important for food security, with rice and other crop production in the Usangu plains. The river eventually reaches the Mtera Dam and then flows south to the Kidatu Dam. The river continues southwards and flows across the Selous Game Reserve before reaching the Rufiji River. The major rivers contributing to the GRR are Lukosi, Yovi, Kitete, Sanje, Little Ruaha, Kisigo, Mbarali, Kimani and Chimala whereas the small ones include Umrobo, Mkoji, Lunwa, Mlomboji, Ipatagwa, Mambi and Mswiswi rivers.

The GRR, which is the life blood of the Ruaha National Park, is under threat. It ceased to flow for the first time in living memory during the dry season of 1993 and this drying-up has continued every year since then, with the period of non-flow increasing to several months. In the years, coincident with the continuing drying up of the GRR various
programmes of so-called “improvement” of smallholder irrigated rice schemes were undertaken in the Usangu catchment.

As water flow in the Great Ruaha River (GRR) has declined in recent years and large sections of the river dry out during the dry season due to the limited effectiveness of the current government policy in managing farmers’ degradation of the wetland, the hippopotamus population sizes and their spatial distribution in the park has continued to decline in recent years (Kendall, 2011).

The drying of the river has a major impact on biodiversity, wildlife and people’s lives and their livelihoods. The dry season of 2003 was the most desperate; scores of hippo were forced to mass together in muddy pools and many died (Banga, P. Personal communication, 2015). To date the distribution of some of the mammals whose livelihoods depend on GRR has changed markedly as a result of their search for water, leading them into conflicts with the villagers around the Park.

2.4 Hippo Ecosystem Roles

Because of their massive size, hippos play an important role in their ecosystems. They are ecosystem engineers as they create and change the land in and around their habitat. They do this by moving a lot of soil around with their huge body when moving in and out of their habitat. Daily activities both in and out of water create habitat and shelter for smaller organisms. Hippos gut content after being egested is partially fermented and thus an ideal substrate for further microbial activity. Many terrestrial insects such as members of the Scarabaeidae (e.g. Dung beetles) are known to utilise dung either directly for food or as a nutritional source for their developing offspring.
Hippos in the GRR graze on grasses that surround river bank, prompting the retention of the grasslands for other organism which end up stabilizing the whole ecosystem. Generally, hippos play a significant role in their ecosystems, as they keep grasses short for other animals.

2.5 Cross-ecosystem linkages created by the common hippopotamus

Hippopotamus is a semi-aquatic, inhabiting rivers and lakes where territorial bulls preside over a stretch of river and groups of 5 to 30 females and youngs. Hippos terrestrial foraging is far reaching, they often travel a few hundred meters where they spend approximately five hours a night intensely grazing. While hippopotamuses rest near each other in the water, grazing is a solitary activity and they are not territorial on land. In their natural habitat, water helps to cool them from the heat of the hot day. It also helps them to conserve energy for future use as they use a great deal of it when moving around on land. That is part of why they feed so heavily when they are on land. Their natural environment is at risk due to being killed for sport, meat, and ivory of their teeth as well as loss of habitat.

The Common hippos control important linkages between aquatic and terrestrial ecosystems and these effects in turn are influenced by river hydrology. Hippo dung subsidies and grazing may directly affect key components of river and terrestrial ecosystems.
Plate 3: Hippo terrestrial-aquatic linkage

Source: Concept proposal of Douglas McCauley, 2013

Nutrients are essential chemical elements that aquatic organisms need to survive and reproduce (McCartney, 2010). Important nutrients that influence aquatic life include Carbon, nitrogen, phosphorus, and suspended particles. Knowledge of these element cycles is essential for our understanding of ecosystem biogeochemistry. In aquatic systems, nitrogen and phosphorus are the two nutrients that most commonly limit maximum biomass of algae and aquatic plants (UNEP and GEMS, 2006). Since the availability of these elements is often less than biological demand, environmental sources can regulate or limit the productivity of organisms in aquatic ecosystems (UNEP and GEMS, 2006). Therefore, insufficient or imbalance in the nutrient ratio of the aquatic ecosystem affects its structure, stability, and functions. The amount of oxygen, carbon, nitrogen, suspended particles, and phosphorus in the GRR are of special interest because of the regulatory role that these elements have in aquatic ecosystems. The anticipation was that the activities of hippos by fertilizing the Great Ruaha River (GRR) could be the source of these nutrients.
CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Study area description

3.1.1 History and site description of Ruaha National Park (RNP)

The present study was conducted in Ruaha National Park (RNP), Tanzania during the months of November and December 2013. These months formed the late dry season commencing from October to the short rains period in the area which occurs from December. Principally, the study was done in the Great Ruaha River (GRR) which flanks the border of the Ruaha National Park (Figure 2, 3). The GRR ecosystem was selected as a study area due to hosting a large number of hippos and large diversity of ecosystem variability.

Figure 2: Map of Ruaha National Park

RNP is located in the middle of Tanzania about 130 kilometres from Iringa Municipal of Iringa Region and in the Southern Tourist Circuit (Latitudes 7° and 8° S and Longitudes 34° and 35° E) of Tanzania. Currently RNP is the largest park in Tanzania and one of the largest national parks in Africa covering an area of about 200 226 square kilometres. The park was gazetted in 1910 as Saba Game Reserve by the Germans and thereafter named as Rungwa Game Reserve in 1946 by the British. In 1964 the southern portion of the reserve was gazetted as Ruaha National Park and in 1974, a small section of south eastern part of the GRR was incorporated into the park. The name “Ruaha” originates from the Hehe word “Ruvaha”, which means “river”. The Park is currently part of Rungwa-Kisigo–Muhezi ecosystem, which covers more than 45 000 km². Recently, in 2008, Usangu Game Reserve and other important wetlands in Usangu basin were annexed into the park, making it the largest park in Tanzania and East Africa. The park harbours one of the largest hippo populations in the country.

3.1.2 Climate and hydrology

RNP has a bimodal pattern of rain forest; usually the long low rainfall intensity season begins November to February, while the short season with high rainfall intensity is between March and April. The park experiences its dry season between June and October. RNP unique ecosystem is shaped by a wide range of climatic types, from dry, hot, rift valley zones and riverine forests along the Great Ruaha River, to sweeping savannas and cold mountain forests. The annual mean rainfall ranges between 500 mm – 800 mm with the average annual temperature of about 28°C.
3.1.3 Vegetation structure, Animal distribution, diversity, and physical features of Ruaha National Park

The park is characterized by semi-arid type of vegetation, baobab trees, Acacia and other species. There are over 1,650 plant species that have been identified. The park is the transitional point of two vegetation zones, the Zambezian (characterized by Miombo vegetation) and Sudanian (characterized by Acacia vegetation).

Located at the convergence zone of northern and southern species, Ruaha is an extremely diverse ecological area that is home to several endemics, including the Ruaha Red-Billed Hornbill and endangered species such as the African Wild Dog. The park is also the southern limit of the ranges of Grant's gazelle and Striped Hyena. The park harbours over 400 species of birds and it is the only park in Africa where Roan and Sable Antelope and Greater and Lesser Kudu can be found together.
RNP has the highest concentration of elephants than any national park in East Africa and harbours one of the largest hippo populations in the country. It is also home to a wide diversity of mammals like Kudu, Sable and Roan antelopes, lions, leopards, cheetah, giraffes, zebras, elands, impala, bat eared foxes and Jackals (RNP, 2014). The park also harbours a number of reptiles and amphibians such as crocodiles, poisonous and non-poisonous snakes, monitor lizards, agama lizards and frogs.

3.2. Study design
This study constituted two major components. Field work and laboratory activities well designed to assess the three specific objectives of this study namely to assess the relationship between hippo density and nutrients measured in the Great Ruaha River waters, to assess how hippo density affects other important attributes of water chemistry of the Great Ruaha River and lastly to examine the effect of hippo density on stable isotopes of carbon and nitrogen of consumers in the Great Ruaha River waters.

Field study sites selection
The field work was done for a period of 6 weeks. The first three days were used for study site selection and this involved both vehicle and foot surveys along the GRR to identify ideal pools with and without hippos. Thirteen pools containing hippos (hippo pools) and twelve pools without hippo (non-hippo pools or controls) were selected and marked using a Global Positioning System (GPS). The pools selected are shown in Figure 4.
Figure 4: Map of study site – location of sampling points in the Great Ruaha River
During identification of study sites, only accessible hippo pools and non hippo pools were selected as some of the sites were not easily accessible. Within the sampling frame, the pool named (1) in the figure above was named as H (Hippo pool), the second pool (2) was named as N (Non hippo pool), which served as a control site. The naming of subsequent pools depicted in Figure 4 followed the same trend and the hippo pool and its control pool were close enough to allow making comparison during the study.

3.2.1. Examining the relationship between hippo density and nutrients measured in the Great Ruaha River waters.

To better understand the relationship between hippo density and nutrients measured in the GRR waters, one of the 250 ml water sample replicates for each sample collected in the field were transported chilled to the laboratory at Sokoine University of Agriculture (SUA), in the Department of Soil Science for nutrient analysis. The remaining replicates per sample were sent to University of California, Berkeley Centre for Stable Isotope Biogeochemistry, USA for further analysis. At SUA, the samples were analysed for Ammonium and nitrate for estimation of Total Dissolved Nitrogen (TDN). Samples were further analysed for total dissolved phosphorus (TDP) using the methods elaborated under.

The concentration of Ammonium (NH$_3$N) and nitrate (NO$_3$N) in water samples analysed at SUA was found to be very low and this was also confirmed at Berkeley by reanalysing the samples for similar parameters. Total dissolved nitrogen was measured directly at the University of California, Berkeley Centre for Stable Isotope Biogeochemistry (CSIB), USA. In addition the Centre also analysed total dissolved phosphorus, and particulate organic carbon.
3.2.2 Assessing how hippo density affects the water physical parameters of the Great Ruaha River.

To directly measure the relationship between hippo density and water physical parameters measured in the GRR, chemical and physical water properties in hippo pools and control pools lacking hippos were assessed. In the field, each study pool was visited three times a week and at each visit, pool water samples were collected using a one litre plastic container (see plate 4 below) which was tightly fastened on a long metal stick. At each sampling, the container was dipped inside the pool water at a depth of about 30 cm and removed when the container was full of water.

Plate 4: Plastic container for water sampling
Source: own field work photo

At each sampling exercise, two water samples were taken directly from the pool centre, one 10m downstream and another 10 m upstream. The water sampler and the collection bottles were rinsed three times with pool water before repeating the water collection procedure. Within 1 min of sampling, water was immediately analysed for pH, Conductivity, Dissolved Oxygen (DO), and temperature using HQ40d Portable Multi-
Parameter Meter (HACH brand) as shown in Plate 5 below. The data obtained from these duplicate samples (upstream and downstream samples) was averaged and recorded.

Plate 5: Biogeochemical analysis of sampled pool water using a Handheld Multi-Parameter Meter (HACH brand) and (insert) a portable Handheld Multi-Parameter Meter set up

Source: Own field work photo

In addition, at one sampling site (H21), a dissolved oxygen sensor (HOBO Dissolved Oxygen Logger U26-001) was mounted for 6 hours to assess and compare the variation in oxygen concentration from the top to the bottom of the pool water surface. A DO sensor was firstly installed partially on the water surface at hippo pool sampling point (H21) and data were recorded. On the same sampling point (H21), DO sensor was deeply installed to the bottom of the pool and data were recorded. This procedure allowed comparison of DO
content in the same hippo pool. The DO sensor was also allowed to record data at the bottom of the hippo pools for 6 hours.

Following initial field analysis, sampled water was transferred into six 250 ml plastic containers and stored chilled in coolers containing ice cold water awaiting further analyses. A make shift field laboratory was setup at the camp site every evening after field work and was used to analyse for Chromophoric Dissolved Organic Matter (CDOM) in 3 ml of water sample taken from one of the six 250 ml chilled stored respective water samples using a Fluorometer (Turner designs, AquaFluor) see plate 6 below.

Plate 6: A make shift field laboratory setup for sample processing

3.2.3 Examining the relationship between hippo density and stable isotopes of carbon and nitrogen in the Great Ruaha River waters.

To identify the origin of nutrients being used by members of river pool and associated riparian food webs, naturally-occurring C and N stable isotopes were measured at the University of California, Berkeley Centre for Stable Isotope Biogeochemistry, USA lab in the samples collected from watershed sites. The target samples for these studies were blood and tissue samples harvested from fish found both in hippo and non-hippo pools. A
fishing net was used to catch different types of fish found in both hippo and non hippo pool, and after their species identification, fish length was recorded using a tape measure (see Plate 7a-d below).

Plate 7: (a) Representative fish netted (b) Golden dart” (above) and “spotted dart” (below). (c) Rufiji Tilapia and (d) Red tailed dart obtained from study pools.

Names applied are morphotype markers.

Fish blood and tissue samples were carefully collected using tweezers, 2 ml syringes, and stored in sampling plastic vials, then clearly labelled. The sampled blood and tissues were preserved into the vials and packed in the whirl pak (Plastic bags for sample storage) bags and stored in ice-cold cool boxes. In the evening make shift lab, fish tissue and blood samples were dried at ~60°C using a Food dehydrator (NESCO®/American Harvest®) (Plate 8). The dried blood and tissue samples were well wrapped with aluminium foil and preserved dry for stable isotope analysis which was conducted at CSIB, USA. Time for
collecting stable isotope samples at the sampling site ranged but generally lasted about 45 minutes depending on the number of samples taken.

Plate 8: Food Dehydrator ((NESCO®/American Harvest®) used for drying fish blood and tissue samples for stable isotope analysis.

Source: Field work

3.3 Laboratory analysis

3.3.1. Determination of Ammonia (NH₃-N) using distillation titrimetric method

Determination of (NH₃-N) was performed in the Department of Soil Science, SUA using a distillation titrimetric procedures as outlined in the standard methods for analysis of wastewater (APHA, 1992). For this work, 250 ml of the water samples were titrated with 0.02 N H₂SO₄ using Boric Acid as an indicator. Ammonia free distilled water was used as blank. The concentration of ammonia nitrogen was calculated using the following formula:

\[ \text{mg/L Ammonia N} = \frac{(A - B) \times 280}{mL \text{ sample}} \]

Where:

A = volume of sulphuric acid titrated for sample (mL)

B = volume of sulphuric acid titrated for blank (mL)
3.3.2. Determination of Nitrate (NO$_3$-N) using UV Spectrophotometric method

The Total Nitrogen (or Nitrate) in pool water samples were analysed using the UV spectrophotometric method in the Department of Soil Science, SUA using the standard procedure for wastewater analysis (APHA, 1992). Pool water samples to be analysed were first filtered using Whatman filter paper no 1 to remove all suspended particles. To 50 mL of clear filtered samples, 1.0 mL HCl were added and mixed thoroughly. The standards in the range of 0-7 mg NO$_3$-N/L were used for calibration. The absorbance of test samples and calibration standards was read against distilled deionised water at 220 nm using a spectrophotometer (Bio Mate UV-Vis Thermo Scientific) while 275 nm was used to determine interference due to dissolved organic matter.

3.3.3. Determination of phosphorus, ortho phosphate (o-PO$_4$-P) using Ascorbic Acid Spectrophotometric method

Infrared spectrophotometric method for determination of (o-PO$_4$-P) was performed in the Department of Soil Science, SUA using the standard procedure for wastewater analysis (APHA, 1992). Essentially, the water samples were first filtered using Whatman Number 1. About 50 ml of filtrate were transferred to an Erlenmeyer flask and 8 mL of combined reagents (5 N sulphuric acid solution, Potassium antimonyl tartrate solution, Ammonium molybdate solution, and containing Ascorbic Acid) was added and mixed thoroughly and left to stand for 20 minutes to allow colour formation. The concentration of PO$_4$-P was measured at 880 nm using a spectrophotometer machine with infrared phototube, (Bio Mate UV-Vis Thermo Scientific).
3.3.4. Determination of total dissolved nitrogen (TDN) using combustion method.

Frozen water samples were shipped to the University of California, Davis Analytical Laboratory, USA, for analysis of total dissolved nitrogen. Essentially, all particulates in water samples for TDN were homogenised into fine particles. About 50 mL of water samples were injected using an autosampler into a high temperature (850ºC) combustion reactor with an oxidative catalyst, converting all forms of nitrogen to nitric oxide (NO). The NO was quantitated with a chemiluminescent detector (ANTEK 8060 Equimolar Detector Nitrogen liquid chromatography HPLC-CLND).

3.3.5. Determination of total dissolved carbon (DOC) in water samples

To determine total dissolved carbon in water samples, 50 ml of water sample was injected by means of integrated autosampler into the high temperature combustion reactor with an oxidative platinum catalyst in an oxygen rich atmosphere. The concentration of carbon dioxide generated was measured with a non-dispersive infrared (NDIR) gas analyser (GFC7000 series Ultra Trace Gas Analyser, Teledyne Technologies Company). In this reactor, at the temperature of 850ºC all organic and inorganic carbon was oxidized into the gaseous carbon dioxide (CO₂). Platinum catalyst that was present in the reactor catalyzed the oxidation of all organic and inorganic carbon to completion. The carbon dioxide was measured at 4.2 um by infrared (IR) detection.

3.3.6. Determination of stable isotope

The dried blood and tissue samples were analysed at Centre for Stable Isotope Biogeochemistry (CSIB), University of California, Berkeley, USA. Carbon (¹³C) and Nitrogen (¹⁵N) in solid materials were analyzed by continuous flow (CF) dual isotope
analysis method using a CHNOS Elemental Analyzer interfaced to an IsoPrime100 mass spectrometer (Leco CHNS 932, Isomass Scientific Inc). The long-term external precision for C and N isotope analyses was ± 0.10 ‰ and ± 0.15 ‰, respectively.

3.4 Statistical analysis
The data obtained were compiled, coded and analysed using SAS (Statistical Analysis System) program (Version 8.3) for Window®. Results from experimental samples were expressed as means ± standard error. Data were assessed by t-test and tests for differences between the means were done using one tailed t-test. The statistical significance between hippo abundance and all nutrient/chemical response values was analysed. The GPS data were processed using Arc view system and GIS image produced as map.
CHAPTER FOUR

4.0 RESULTS

4.1. Assessing the relationship between hippo density and nutrients measured in the Great Ruaha River waters.

4.1.1. Assessment of Ammonia (NH₃-N)

To allow the better determination of nitrogen ammonia (NH₃-N) in the river waters collected during the study period, all water samples were analysed for NH₃-N and results were compared (Table 1).

<table>
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<th>Parameter</th>
<th>Regression coefficient (R²)</th>
<th>Slopes</th>
<th>Y-Intercept</th>
<th>p-value</th>
<th>F</th>
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</thead>
<tbody>
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<td>DOC(mg/L)</td>
<td>0.3768</td>
<td>1.134</td>
<td>22.813</td>
<td>&lt;0.35</td>
<td>15.11</td>
</tr>
<tr>
<td>TDN(mg/L)</td>
<td>0.2991</td>
<td>0.046</td>
<td>1.5357</td>
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<tr>
<td>TDP(µg/L)</td>
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<td>3.715</td>
<td>124.83</td>
<td>0.02</td>
<td>6.52</td>
</tr>
<tr>
<td>NO₃(mg/L)</td>
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<td>0.001</td>
<td>0.0206</td>
<td>&lt;0.01</td>
<td>12.87</td>
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<tr>
<td>NH₃(mg/L)</td>
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<td>0.001</td>
<td>0.2181</td>
<td>0.89</td>
<td>0.02</td>
</tr>
</tbody>
</table>

Nutrient concentrations in the GRR water samples varied with hippo density along the longitudinal axis in the river. The results (Figure 5) indicated an insignificant relationship between NH₃-N concentrations and hippo density (R²=0.0011). The concentrations of NH₃-N did not vary with hippo density at different sites of the river during the November to December month.
Figure 5: Mean Concentration of Nitrogen, Ammonia (NH$_3$-N) in relation to hippo density

4.1.2. Determination of Nitrate (NO$_3$-N)

The results of nitrate concentrations assessed in two months (November to December) between hippo and non hippo pools in the GRR are presented in Figure 6. Nitrate concentrations varied with number of hippo in the study sites. It was evident that nitrate (NO$_3$-N) exhibited a positive relationship with hippo density ($R^2 = 0.345$, $P < 0.01$) (Table 1) where NO$_3$-N tended to increase as hippo density increased. The estimated number of hippos in each sampling pool varied from each other. The concentrations of NO$_3$-N within the hippo pools in the GRR were generally high and increased sharply with increasing number of hippos along the river.
4.1.3. Assessment of dissolved organic carbon (DOC)

In order to assess whether hippo abundance along the GRR was associated with an increase in nutrients in river waters, an attempt was done to assess the amount of total dissolved organic carbon (DOC) between hippo and non-hippo pools. Results of DOC are presented in Figure 7. The particulate organic carbon ranged from 5.31 mg/L at non-hippo pool sampling point (N6) to 157.89 mg/L at hippo pool sampling point (H28). The mean value for DOC in hippo pools was 52.74 mg/L (n=13) and that of non-hippo pools was 17.52 mg/L (n=12). There was no statistical significant difference in DOC between hippo and non-hippo pools (P<0.35) (Table 1). Despite this lack of statistical significant relationship it is interesting to observe that the highest DOC values tended to occur in the pools with the highest mean number of hippos.
4.1.4. Assessment of total dissolved nitrogen (TDN)

The mean values for total dissolved nitrogen in hippo pools (n=13) and non hippo pools (n=12) were 2.83 mg/L and 1.30 mg/L, respectively. During the entire period of study, total dissolved nitrogen (TDN) values ranged from 0.54 mg/L at non hippo pool sampling point (N6) to 6.61 mg/L at hippo pool sampling point (H28). The next highest value was 6.25 mg/L at hippo pool sampling pool (H21). Results from this study revealed that, the amount of total dissolved nitrogen was dependent on hippo density. Hippo pools with higher number of hippos namely H21, H25, H27, H28, H36, and H37 had remarkable amount of TDN compared to pools with less number of hippos (had almost six times higher) compared to pools with less number of hippos (N15, N17, N18, N23, N25, N27, and N28). Statistical analysis showed that, total mean dissolved nitrogen in hippo pools was significantly higher when compared to non hippo pools values (P < 0.01) see Table 1.
4.1.5. Assessment of total dissolved phosphorus (TDP)

Generally, TDP concentrations in pool water ranged from 55.10 ug/L to 559.83 ug/L in hippo pools and 22.44 ug/L to 154.33 ug/L in non hippo pools. There was a positive relationship with hippo density as the increased hippo density led to significant increase in TDP in respective pools (P<0.02) (Table 1).

The measured total dissolved phosphorus concentrations in hippo and non hippo pools are depicted in Figure 9. The mean total phosphorus in non-hippo pools was 54.18 µg/L. Like other measured variables, the total pool phosphorus concentrations also tended to increase with increased number of hippos and this increase was 280.35 µg/L. The simultaneous increase in total dissolved phosphorus concentrations with hippo density were highly significant (R² =0.2068, p˂0.01715) (Table 1).
4.2 Assessing how hippo density affects the water physical parameters of the Great Ruaha River.

The large hippo herds were located at few pools which still had enough water to accommodate a substantial numbers. Most of them were concentrated in those areas which still had habitat suitable to their survival.

To clearly assess the contribution of common hippo on water quality parameters, the hippo pools were modelled as rectangle to calculate water volume in relation to the number of hippo residing in respective pools and their ongoing activities. The estimate number of hippos at each sampling site ranged from 0 to 10 (Appendix 1). The method of hippo survey employed at each sampling site was by river bank counts. For example, at hippo pool sampling point (H25), the estimates for length, width, and depth were 85 cm, 14 cm, and 96.67 cm which gave water estimate of total 115037.3 cm$^3$ and the pool had an estimate number of hippo of nine (9).
The control group pools (n=12) those having apparently clear water and as they were monitored every time of sampling, they were confirmed to harbour a very low hippo density. All hippo containing pools (n=13), had water which was very turbid and contained treading down sediment and a permanent surface of faecal matter. Hippo pool, water colour ranged from black to brown and there was no river water flow during the study period. Decomposing organic matter obtained from the bottom of the hippo pools were completely black (Plate 9).

Plate 9: Decomposing plant material found at the bottom of the hippo pool (H 17)

4.2.1 Assessment of water physical parameters in the GRR

During the field studies conducted during the period of Nov-Dec 2013, the water physical parameters (pH, dissolved oxygen concentration, conductivity, chromophoric dissolved organic matter (CDOM), and temperature) in the GRR were assessed during the dry (November month) and at the onset of the wet season (December month).
4.2.2 pH assessment before the onset of rain (November)

Water pH in samples collected during the November month ranged from $7.52 \pm 0.09$ in hippo pools to $8.24 \pm 0.13$ in non hippo pools (Table 2) and these differences were statistically significant ($P<0.001$) see Table 2. Generally hippo pools contained water with remarkably lower pH compared to the control non hippo pools.

Table 2: Mean ± SEM values of the physicochemical water parameters in the dry November month

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Hippo pools</th>
<th>Non Hippo pools</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>DO mg/l</td>
<td>$2.599 \pm 0.984$</td>
<td>$7.892 \pm 0.924$</td>
<td>0.0003</td>
</tr>
<tr>
<td>CDOM mg/l</td>
<td>$279.281 \pm 18.567$</td>
<td>$262.695 \pm 22.424$</td>
<td>0.4936</td>
</tr>
<tr>
<td>Temp °C</td>
<td>$29.035 \pm 1.035$</td>
<td>$29.658 \pm 1.036$</td>
<td>0.4875</td>
</tr>
<tr>
<td>pH</td>
<td>$7.517 \pm 0.086$</td>
<td>$8.237 \pm 0.129$</td>
<td>0.0002</td>
</tr>
</tbody>
</table>

4.2.3 Temperature assessment in the dry season

The temperature values of the water during the dry season varied widely throughout the day (Table. 2). Daytime mean water temperature of the GRR varied from a minimum of $29.04$ °C in the hippo pools to a maximum of $29.66$ °C in the non hippo pools. By dawn, the surface water temperature was marked low and it was high in the mid-afternoon. There was a considerable spatial variation in water temperature from pool to pool depending on time, shadowing by vegetation and depth. Statistical analysis showed that, there was no significant difference in temperature between the hippo pools and non hippo pools ($P>0.05$) during the dry season (Table 2).
4.2.4 Assessment of dissolved oxygen in the GRR

Dissolved oxygen content in hippo and non hippo pools assessed during the dry season are presented in Table 2 and Figure 10. The mean ± sem value of dissolved oxygen in the hippo and non hippo pools were 2.60 ± 0.98 mg/L and 7.89 ± 0.92 mg/L (Table 2), respectively and these differences were statistically significant (P < 0.01). The minimum DO values were observed at hippo pool sampling sites (H28, H27, and H21) in the dry season. The maximum DO values were observed at non hippo pool sampling sites (N17, N30, and N28).

![Figure 10: Variability (mean± sem) of dissolved oxygen between hippo pools and non hippo pools in the GRR](image)

To assess the DO variability from the surface to the bottom of the pool, such DO values were assessed at hippo pool sampling point (H21) using dissolved oxygen sensor. This pool was located at the Msembe Bridge characterised by very black water with a lot of hippo dung floating and bubbling. The length, width, and depth of the pool were 70 cm, 30 cm, and 260 cm, respectively and the estimated hippo scale (hippo amount) was 9.
Results showed that, there was a remarkable minimum amount of dissolved oxygen (0.011 mg/L) at the bottom of this pool. The mean values of dissolved oxygen near the surface of the hippo pool was 4.443 mg/L and these values were statistically higher (P<0.001) compared to the DO values which were recorded at the bottom of the pool.

4.2.5 Assessment of water conductivity in the GRR

Water conductivity in hippo and non hippo pools showed an inverse relationship when compared to DO values above (Figure 11). Water conductivity was significantly higher (P<0.01) in hippo pool (-18.173 ± 7.839 μS/cm) when compared to values recorded in non hippo pool (-69.377 ± 7.720 μS/cm).

![Figure 11: Variability (mean ± sem of conductivity between hippo and non hippo pools in the GRR](image)

4.2.6 Assessment of chromophoric dissolved organic matter (CDOM) in hippo and non hippo pools in the GRR

The mean CDOM content in hippo and non hippo pools during the dry season (November) are shown in Table 2. The mean value of CDOM content in hippo pools and non hippo pools were 279.281 mg/L and 262.695 mg/L, respectively). No significant
differences in CDOM content were observed between hippo pools and non hippo pools during the dry season (P>0.05).

4.3 Water chemistry conducted during the onset of wet season (December)

4.3.1. Pool temperatures between hippo and non hippo pools

The temperature assessment conducted between hippo pools and non hippo pools were done for five consecutive days and results are presented in Figure 12. There was no significant changes in daily water temperatures in the hippo and non hippo pools from day one to day five of monitoring and neither significant temperatures were observed between the two (hippo and non-hippo pool) groups. Lowest temperatures were recorded on Day one and Day four had a maximum amount.

![Temperature Chart](image)

**Figure 12:** Variability mean ± sem of temperature between hippo and non hippo pools in the GRR after the onset of rains

4.3.2. Dissolved oxygen assessment

DO was also assessed for five days following the onset of rains. Dramatic changes in the DO concentrations were observed and Figure 13 shows these changes in one hippo pool
number H16 and Figure 13 shows the variability in mean ± sem dissolved oxygen between hippo and non hippo pools after the onset of rains in GRR. During monitoring, the DO concentration was marked high in day one to day four but dramatically dropped on day five as the river waters started to decrease in volume.

![Graph showing dissolved oxygen fluctuations](image)

**Figure 13: Oxygen fluctuations within a hippo pool (H16) during the wet season**

With the exception of day four, the mean temporal DO values between the hippo and non hippo pools followed a similar trend and the values were not significantly different between the two groups.
Figure 14: Variability in mean ± sem dissolved oxygen between hippo and non hippo pools during the onset of rains in GRR

4.3.3. Chromophoric dissolved organic matter (CDOM)

After the onset of rains in early December month, there was heavy dewfall in RNP and resulting in rapid increase in water levels in both temporally and permanent water pools. CDOM contents of the GRR waters were compared between hippo pools and non hippo pools for five days following the onset of rains and results are presented in (Figure 15). There was no significant changes in CDOM content in the hippo and non hippo pools from day one to day five of monitoring and neither significant CDOM content were observed between the two (hippo and non-hippo pool) groups. However, the mean values tended to increase on the fourth and fifth day of monitoring. The mean values between hippo pools on the fourth and fifth day of monitoring were 254.5 mg/L and 212.4 mg/L, respectively while corresponding values for non hippo pools were 326.4 mg/L and 217.1 mg/L.
Figure 15: Variability in mean ± sem of chromophoric dissolved organic matter between hippo and non hippo pools during the onset of rains in GRR

4.4 Assessment of stable isotopes of carbon and nitrogen between hippo and non hippo dwellers in the Great Ruaha River waters.

4.4.1 Assessment of carbon stable isotope ($\delta^{13}$C)

Stable isotope of carbon ($\delta^{13}$C) was analysed in the present study as a way of tracing the origin of nutrients consumed by aquatic organisms in study pools. The mean $\delta^{13}$C recorded from Tilapia in hippo pools and non hippo pools were almost similar and were -18.65 ‰ and -18.32 ‰, respectively. (Table 3). The same trend was observed in $\delta^{13}$C values measured in the Red tailed dart, obtained from hippo (-19.99) and non hippo (-20.27) pools.
Table 3: Mean ± sem of carbon isotope data from Tilapia and Red tailed dart

<table>
<thead>
<tr>
<th>Species</th>
<th>d 13C</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Hippo pool</td>
<td>Non hippo pool</td>
</tr>
<tr>
<td>Tilapia</td>
<td>-18.65± 0.32</td>
<td>-18.32± 0.39</td>
</tr>
<tr>
<td>Red tailed dart</td>
<td>-19.99± 0.62</td>
<td>-20.27± 0.65</td>
</tr>
</tbody>
</table>

4.4.2 Assessment of nitrogen stable isotope (δ¹⁵N)

Nitrogen stable isotope (δ¹⁵N) was also assessed from fish blood and tissue samples obtained from the study sites in the GRR to assess the origin of nutrients used by river consumers. Tilapia and red tailed dart were sampled, analysed and results are presented in Table 5. The mean values of δ¹⁵N in tilapia obtained from hippo pool was (9.94 ± 0.27) and non hippo pools (9.66 ± 0.41). In red tailed dart, data were taken and measured for δ¹⁵N, and the mean value observed in hippo pool was 11.99 ± 0.21 whereby in non hippo pool the value was 13.01± 0.81. The values in tilapia and red tailed dart were not statistically different (Table 4).

Table 4: Mean ± sem of nitrogen isotope data from Tilapia and Red tailed dart

<table>
<thead>
<tr>
<th>Species</th>
<th>d 15N</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Hippo pool</td>
<td>Non hippo pool</td>
</tr>
<tr>
<td>Tilapia</td>
<td>9.94± 0.27</td>
<td>9.66± 0.41</td>
</tr>
<tr>
<td>Red tailed dart</td>
<td>11.99± 0.21</td>
<td>13.01± 0.81</td>
</tr>
</tbody>
</table>
CHAPTER FIVE

5.0 DISCUSSION

The present study which was done for a period of two months (November to December 2013 in the Great Ruaha River, was mainly aimed at assessing the effect of *Hippopotamus amphibious* vectored subsidies on the chemistry of river ecology and the ecology of aquatic organisms resident within these ecosystems. The effects of hippo vectored subsidies in the GRR were assessed by measuring water nutrients, water parameters, and stable isotope analysis of aquatic consumers in samples collected from two different sampling points, those containing hippos and those with no hippos. There have been several previous studies which have investigated the effect of hippos on terrestrial and aquatic ecosystems (Ajibade, 2008; Kendall, 2011; Wolanski and Gereta, 1999), but none of these used similar study design used in the present investigation to address the interactions of several factors which occurs in pools habited by hippopotamus and which play roles in nutrient recycling. The GRR ecosystem was selected as a suitable site for conducting these types of studies in Tanzania due to the huge population of hippos which habit this river and large diversity of ecosystem variability along the entire river.

The present investigation covered a period of 2 months (November to December, 2013) and was aimed at assessing the role of common hippos in aquatic ecosystem in the GRR and was done by combining the use of GPS information to map several sites on and along the river which could help provide more insight on the effect of hippopotamus vectored nutrients to the GRR waters. In addition to the identification of the effect of hippos on the river waters in RNP all locations were GPS identified.
5.1. Assessing the relationship between hippo density and nutrients measured in the Great Ruaha River waters.

5.1.1 Assessment of ammonia

Results of the current study have clearly shown that *Hippopotamus amphibius* in the GRR have no effect on the concentration of ammonia as lower concentrations were measured in both pools (hippo and non hippo pools). Statistically, there was no significant difference in ammonia concentrations between hippo and similar control pools (P >0.05). These results conform to the study of Ajibade *et al.* (2008) who reported no significant correlation in ammonia between the dry and wet season when ammonia was assessed in hippo pools. Ammonia is one of several forms of nitrogen that exist in aquatic environment. It undergoes reversible oxidation and reduction processes as it travels through the environment. Nitrogenous compounds decompose and oxidize to form ammonia. Ammonia, along with other inorganic nutrients in the right proportion facilitates plant growth. In excess, ammonia is likely to be toxic to aquatic organisms and NH$_3$ is the most toxic form. The toxicity of ammonia to fish and aquatic organisms is remarkable, even in very low concentrations. When levels reach 0.06 mg/L, fish can suffer gill damage. Ammonia levels greater than approximately 0.1 mg/L are often indicative of polluted waters.

The danger ammonia poses for fish and other aquatic organisms depends on the water’s temperature and pH, along with the dissolved oxygen and carbon dioxide levels. Francis-Floyd *et al.* (2012) reported that of all the water quality parameters that affect fish, ammonia is the most important after oxygen, especially in most aquatic systems. Ammonia causes stress and damages gills and other tissues, even in small amounts. Fish exposed to low levels of ammonia over time are more susceptible to bacterial infections, have poor growth, and will not tolerate routine handling as well as they otherwise would.
Ammonia is a killer particularly when present in higher concentrations, and many unexplained loss in aquatic abundance of organisms likely been caused by ammonia (Camargo and Alonso, 2006).

5.1.2. Determination of nitrate

Results of the current study have clearly shown that *Hippopotamus amphibius* in the GRR have an increasing effect in nitrate concentrations as higher concentrations were measured in pools with high number of hippos. These findings are in agreement with previous studies done by Subalusky *et al.* (2014) who found that daily hippopotamus loading into the river by excretion and egestion produces 3499 kg Carbon, 492 kg Nitrogen and 48 kg Phosphorus. Nitrate concentrations were expected to be high in hippo pools as hippos are thought to be the driver of aquatic ecosystem by vectoring nutrients from terrestrial ecosystems.

In the present study, it was observed that, there was a strong statistical significant relationship between hippo density and nitrate in all the study sites under investigation (hippo and similar control pools) during the whole study period. Hippo pools were predominantly higher in nitrate compared to non hippo pools during the study. Nitrates are a form of nitrogen, which is found in several different forms in terrestrial and aquatic ecosystems. Nitrates (NO₃⁻) are essential nutrients for growth of both plants and other aquatic organisms e.g. fish. These results concur with the study conducted by Camargo and Alonso (2006) who reported nitrates in river waters stimulate or enhance the development, maintenance and proliferation of primary producers, resulting in eutrophication of aquatics. Nitrogen in the form of nitrate is an important element for algae growth which is an important component of an aquatic ecosystem and a vital
component for the web ecosystems. Together with phosphorus, nitrates in aquatic ecosystem can accelerate eutrophication, and hence causing dramatic increases in aquatic plant growth and changes in the types of plants and animals that live in the water system. This observation was also reported by Durand et al. (2011) who suggested that the role of nitrate in freshwater ecosystems is as one of the key nutrients, along with carbon, phosphorus (P) and silica (Si), required to support primary production by higher plants and algae. Nitrogen also plays a role in determining the food web structure and relative productivity of any water body through microbial, algal and plant uptake of nitrate in the form of both inorganic nitrogen species and dissolved organic nitrogen (DON). Any change in the rate of supply of nitrate to a water body, will lead to changes in the productivity of the water body and its microbial metabolism, with associated secondary effects in terms of microbial, plant and animal community species composition and relative abundance, and the structure and balance of the aquatic food web.

Excess nitrates can create conditions that give rise to hypoxia (low levels of dissolved oxygen) and can become toxic to warm-blooded animals at higher concentrations (10 mg/L or higher) under certain conditions (APHA, 1992). Due to existence of high concentration of nitrate in the study sites with high number of hippos, there is a high possibility that aquatic organisms in these sites are severely affected. Such a pattern was anecdotally reflected in our preliminary surveys of vertebrate diversity and abundance across our sample sites with varying degrees of nitrate and hippos. The only way to minimize the possibility of nitrate overload is to ensure there is a continuous flow of the GRR.
Interestingly, results from this study showed that hippos build important ecological connections between terrestrial and aquatic ecosystem adding essential nutrients to the river consumers. This study also provides a proof that hippos are able to bring together terrestrial-aquatic nutrients and energy into other component of river system ending up nourishing aquatic organisms. Further studies are invited to further extend the study and track the significance of hippos in relation to nutrient availability in the GRR.

5.1.3. Assessment of dissolved organic carbon (DOC)

Dissolved organic carbon differed slightly between different sampling points under investigation (hippo and similar control pools) during the study period. Hippo pools were predominantly higher in dissolved organic carbon compared to non hippo pools during the entire period of study. The sites that displayed high dissolved organic carbon values are, hippo pool sampling point (H28) and hippo pool sampling point (H27) were characterized by hosting high densities of hippos. Subalusky et al. (2014) reported that, hippopotamus inputs to rivers (excretion and egestion) provide evidence that hippopotamus are important resource vectors in sub-Saharan African rivers, even when compared to other sources of carbon and nutrients. Studies indicate that DOC content in rivers is indicative of the accumulation of organic matter vectored from terrestrial ecosystem (Sachse, 2005). The high DOC values observed at H27 and H28, and the slight difference observed during the dry season between hippo and control pools are most likely the result of decaying hippo derived organic material and low water levels in the GRR. Similar findings have previously been reported in other areas (Ajibade, 2008 and McCartney, 2010 ). The influx of dissolved organic carbon in aquatic ecosystems is essential for food supplement, supporting growth of microorganisms and plays an important role in the global carbon cycle through the microbial loop. Moreover DOC is a key indicator of organic loadings in rivers and other aquatic systems, as well as supporting terrestrial
processes (Kirchman et al., 1991). Results from this study indicate that dissolved organic carbon concentrations in the study pools varied considerably depending on the presence or absence of hippos. Moran and Zepp (1997) reported that dissolved organic carbon stimulate the growth and activity of microorganisms in aquatic environment.

This study demonstrated the effect of common hippos in aquatic organisms in the GRR. Aquatic dwellers in the GRR obtain their nutrients and flourish due to the presence of common hippopotamus. Hippos contribute to the release of dissolved and particulate organic carbon and are important components in the carbon cycle and serve as a primary food sources for aquatic food webs. Several lines of evidence from these results show that, DOC alters aquatic ecosystem chemistries by contributing to acidification in low-alkalinity, weakly buffered, freshwater systems. Furthermore, DOC forms complexes with trace metals, creating water-soluble complexes which can be transported and taken up by organisms. Thus, reductions in DOC from aquatic ecosystem can have important consequences on watershed productivity. Finally, organic carbon, as well as other dissolved and particulate matter, can affect light penetration in aquatic ecosystems, which is important for the ecosystem's phototrophs that need light to subsist.

5.1.4. Assessment of total dissolved nitrogen (TDN)

The current findings have demonstrated significant effect of Hippopotamus amphibious to the river dwellers in the GRR. The typical high concentrations of total dissolved nitrogen which were observed in hippo pools are a clear indication that hippos are the driver of aquatic community in the GRR.
Total dissolved nitrogen showed a significant increase in hippo pools when compared with non hippo pools. It was evident that, in all the study sites with high number of hippos, total dissolved nitrogen was markedly high compared to the control pools. The sites high in total nitrogen were dispersed depending on the aggregation of hippos in the study sites suggesting that differences in hippo dung loading rates have played a role in creating these observed differences in TDN. The sites high in total dissolved nitrogen were hippo pool sampling points (H21) and (H28). Benstead et al. (2010) reported that excretion of nitrogen (N) and phosphorus (P) in aquatic ecosystems has a direct and potential important role for aquatic consumers in nutrient cycling.

Nitrogen is very essential for the survival of both aquatic and terrestrial organisms. Nitrogen enters the ecosystem in several chemical forms and also occurs in other dissolved or particulate forms, such as tissues of living and dead organisms. Kelly (2008) reported that, nitrogen enrichment in aquatic ecosystems results into increased plant biomass and primary productivity which in turn leads to increased species abundance. The primary role of TDN in freshwater ecosystems is as one of the key nutrients, along with carbon and phosphorus, required to fuel up the productivity of water bodies by enriching species abundance.

5.1.5. Assessment of total dissolved phosphorus (TDP)

Results of the current study have evidently shown that total dissolved phosphorus observed in the water samples collected was significantly varying between hippo and non hippo pools. Elevated total dissolved phosphorus values were evident on pools with high number of hippos. A number of sites ranked high in total phosphorus, including the hippo pool sampling point (H21), hippo pool sampling point (H17) and hippo pool sampling point (H28). All of the sampling sites during the dry season were not connected which have
contributed to the build up of nutrients at these sites. Comparison between hippo pools and non hippo pools may explain the richness in some organic matter in the sampling sites as higher concentrations of phosphorus were evident in hippo pools. These findings are in line with those of Gereta and Wolanski (1998) who reported that the Seronera river sediment in suspension was extremely rich in phosphorus.

A study conducted by Hoberg et al. (2002) examined the aquatic food web dynamics and it was observed that high concentrations of phosphorus were measured during the dry season. Highest values of total nitrogen and total phosphorus were found close to the river. A study conducted by Ajibade et al. (2008) on water quality parameters in hippo pools reported that there was a marked increase in phosphate content in the dry season compared to the wet season particularly to those pools with common hippos. The information regarding the role of hippopotamus to aquatic dwellers in the GRR was similar to that reported in other studies (Kilham, 1982). Nutrients availability is the major factor which drive both aquatic and terrestrial ecosystem. High nutrient level is associated with a remarkable increase in primary productivity of aquatic organisms. The high amount of nutrient levels observed in hippo pools were due to leaching and partial decomposition of hippopotamus dung (Kilham, 1982). The very large amounts of total dissolved phosphorus in hippo pools suggests that hippo strongly influence aquatic ecosystems by vectoring large volumes of terrestrial matter into these systems.

5.2. Assessing how hippo density affects the water quality parameters of the Great Ruaha River

The November 2013 field studies coincided with the dry season in Ruaha and there was no water flow in the GRR during this period hence there was significant variabilities in dissolved oxygen (DO) content, electrical conductivity, and pH across various sampling
points between hippo pools and control pools. DO, electrical conductivity, and pH increased linearly with increasing number of hippos while the mean temperature and chromophoric dissolved organic matter (CDOM) values had no statistical significant difference between hippo and non hippo pools. However, there was some observed increase of these parameters in some hippo pools (H16 and H17).

Following the onset of wet season which started in earlier December the water flow in the GRR resumed and all the sampling pools were reconnected with water. The significant higher dissolved oxygen, conductivity, and pH which was observed in pools containing hippo compared with non hippo pools during the dry season, was no longer evident during the wet season. The secret behind low values of dissolved oxygen, conductivity, and pH during the wet season was attributed to the flowing river (water coming in and out) leading into diluting all the material that were brought by hippos. Results from this study are in line with that of Premlata Vikal (2009).

Oxygen dissolved in water as well as pH variability influence aquatic life respiration and their survival. Dissolved oxygen is necessary to many forms of life including fish, invertebrates, bacteria and plants living in aquatic environments. These organisms use oxygen in respiration, similar to organisms on terrestrial ecosystems. Fish and crustaceans obtain oxygen for respiration through their gills, while plant life and phytoplankton require dissolved oxygen for respiration when there is no light for photosynthesis (EPA, 2014). The amount of dissolved oxygen needed varies from creature to creature in the water bodies. Bottom feeders, crabs, catfish and worms need minimal amounts of oxygen (1-6 mg/L), while shallow water fish need higher levels (4-15 mg/L) (Osmond et al., 1995). Microbes such as bacteria and fungi also require dissolved oxygen. These
organisms use DO to decompose organic material at the bottom of a body of water. Microbial decomposition is an important contributor to nutrient recycling.

Dissolved oxygen enters into water from the atmosphere by diffusion and from algae and aquatic plants when they photosynthesize. Dissolved oxygen is removed from water when organisms metabolize or breathe. Because photosynthesis occurs during the day when there is light, DO often increases in the day. At night, DO drops as organisms metabolize. DO can be low in streams if metabolism (breathing) is greater than DO inputs or if water flowing in from upstream is already low. Low DO often occurs when organic matter or nutrient inputs are high, and light inputs are low as were the conditions characterizing high hippo pool sites.

pH is a measure of the amount of acidity/alkalinity in the stream. A low pH observed in the GRR waters during the dry season indicated more acidity. pH revealed a clear trend along the longitudinal axis in the GRR, values were relatively higher in hippo pool sampling points and decreased sharply in the control pools. The reason behind the high pH values observed in hippo pools was attributed to the river flow. When the river stops flowing (dry season), hippo-vectored organic matter concentrate in the pools making the DO and pH go higher. If the pH of water is too high or too low, the aquatic organisms within it will die. pH can also affect the solubility and toxicity of chemicals and heavy metals in the water (USGS, 2013). The majority of aquatic creatures prefer a pH range of 6.5-9.0, though some can live in water with pH levels outside of this range.
As pH levels move away from this range (up or down) it can stress animal systems and reduce hatching and survival rates. The further outside of the optimum pH range a value is, the higher the mortality rates. The more sensitive a species, the more affected it is by changes in pH. In addition to biological effects, extreme pH levels usually increase the solubility of elements and compounds, making toxic chemicals more mobile and increasing the risk of absorption by aquatic life (EPA, 2012).

5.3. Stable isotope comparison of consumer tissue between hippo and non hippo pools

Stable isotopes offer exciting possibilities as natural tracers that reflect the origin and transformations of key biological elements. With respect to the complete dataset on stable isotopes, in pool fish (Tilapia and Red tailed dart) no significant correlations were evident between the hippo pools and the control pools. The fact that there was no statistical differences observed between hippo pools and control pools suggests two possibilities: 1) that indeed there was no difference in the contribution of hippo-derived organic matter between these pools or 2) that alternative sources of organic carbon that looked isotopically similar to hippo dung (e.g. elephant dung) may have washed into non-hippo pools obscuring our ability to detect differences specifically derived from carbon and nitrogen isotopes.

In ecological studies, stable isotope analysis is useful in quantifying the relative importance of allochthonous carbon and nitrogen sources to aquatic ecosystems (Meili, 1992; Jones et al., 1994). According to various studies, Stable isotopes have been extensively used to trace element cycles and their incorporation into different food webs (Middelburg, 2014). However, the results obtained demand more detailed study to deeply investigate how hippo vectored nutrients and energy are used by river consumers.
CHAPTER SIX

6.0. CONCLUSION AND RECOMMENDATIONS

6.1. Conclusion

The findings obtained from different studies undertaken in this dissertation have clearly demonstrated significant hippopotamus activity against the tested parameters in the GRR. Hippo-derived allochthonous materials contributed to several important sources of local chemical differences observed in the GRR. The lower pH and DO values observed in hippo pools during the dry season made the waters to be less suitable for many aquatic organisms. Results of this study revealed that *Hippopotamus amphibious* is having a remarkable influence on chemistry and consequently the ecology of pools they inhabit. Hippo dung is nutritionally rich in total dissolved nitrogen, total dissolved phosphorus, dissolved organic carbon and suspended particles. The site variations in the physicochemical parameters of the GRR water were as a result of the effects of hippos on the rivers and the prevailing weather conditions of the GRR. Hippo vectored subsidies contributed a lot to the water chemistry of the GRR. The hippo pools and its control pools physicochemical parameters of the GRR waters were within the range that can support aquatic life particularly during the wet season. However, the lower DO and pH values in the hippo pools and the extreme concentration of dung in the hippo pools made the waters of the GRR not to be suitable for supporting many aquatic species. Statistical analysis showed that there were significant differences between the hippo and non hippo pools sampling points for the different parameters examined. The presence of hippopotamus in the pools affected some parameters such as CDOM, Conductivity, DO, and pH significantly. The DO and pH contents were strongly significant (P<0.05). The problem of water quality in the hippo pools particularly during the dry season should be addressed
with all weightiness. Watersheds should be protected for the lifelong survival of aquatic organisms and of the GRR at large.

An interesting point for future analysis would be to re-examine the relationships between river nutrient concentrations and the abundance of hippos standardized by these pool-specific volumes of water.

6.2. Recommendations

- Further studies should be conducted to determine the effect of hippopotamus subsidies on the Great Ruaha River waters and the mechanism of action on hippo subsidies transfers from terrestrial to aquatic ecosystems.
- Although *Hippopotamus amphibious* has influence on aquatic life, and since irrigation has been identified as the major pressure on water resources in the GRR, much emphasis should be directed on how to rescue the life of hippos as well as the life of the GRR.
- There should be further studies on the flow, stability and degradation of the Usangu wetland which is the source of waters of the great Ruaha River and the main supporting body of aquatic life in the great Ruaha River.
- Since RNP is one of the regions park hosting a greater number of hippopotamus in the country, there is a need to carry out further studies using other methods to determine the effect of hydrology on hippopotamus life and the river ecosystem.
- Since the GRR is the key component of the Ruaha National Park ecosystem, and as it is the only source of water in the Park, the area deserves detailed investigations on quantity and quality of its water.
• In spite of these results of preliminary effect of nutrients and water chemistry parameters to aquatic animals, further studies, especially long-term studies, are required to check and improve the effect of hippo vectored material (hippo dung) on aquatic dwellers in the GRR.
REFERENCES


Appendix 1: The GRR information on hippopotamus status and river characteristics

<table>
<thead>
<tr>
<th>Study site</th>
<th>Water colour</th>
<th>Method of survey</th>
<th>Estimate number of common hippo</th>
<th>Year surveyed</th>
</tr>
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<tbody>
<tr>
<td>H-15</td>
<td>Black</td>
<td>River bank counts</td>
<td>3</td>
<td>Nov – Dec 2013</td>
</tr>
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