Eutrophication in Ugandan Crater Lakes. A Case Study of Six Crater Lakes Located in Kabarole District Western Uganda.

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by

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Abstract

Volcanic crater lakes in Western Uganda are a significant natural resource to different societal segments. To the local people, the crater lakes provide a source of livelihood, to the Ugandan government the lakes are a boost to the tourism industry, to researchers, the crater lakes play an important role in studies of comparative limnology and, on a global scale, the Western Uganda volcanic crater lakes are heritage sites to be preserved. Despite their numerous values, the crater lakes continue to experience eutrophication challenges largely due to ecosystem disturbance from human activities around the crater lakes themselves and in their entire catchments, but also due to some natural causes.

The rationale for this study was to evaluate eutrophication in the crater lakes through water quality assessment of six freshwater crater lakes. The evaluation is based on the common trophic state variables; chlorophyll-a (Chl-a), total nitrogen (N), total phosphorus (P), and Secchi depth (SD). Other related physicochemical variables were measured both on site and through laboratory experiments. Eutrophication is associated with phytoplankton growth, especially blue-green algae, and as a result, cyanobacterial compositions were also examined.

Trophic state evaluation of the crater lakes was based on the trophic level index (TLI) approach, using the New Zealand TLI system as a base example. The use of the New Zealand TLI system was primarily aimed at testing whether TLI systems developed from other regions can be used to monitor Ugandan crater lakes and, secondly to gain an insight of the systematic similarities and differences between Ugandan crater lakes and New Zealand lakes.

Linear relationships between log-transformed chlorophyll-a and other TLI variables suggested that in the Ugandan crater lakes, total nitrogen and Secchi depth are good proxies for chlorophyll-a whereas total phosphorus is less, suggesting that crater lakes may be nitrogen limited. When the variables were converted into TLI sub-indices, using the New Zealand system, linear regressions of TLc (Chl-a) against TLs (Secchi depth) showed the same relationship between chlorophyll-a and Secchi depth in the crater lakes as in New Zealand model. However, TLn consistently under-predicted TLc with the New Zealand model suggesting that Ugandan crater lakes produce chlorophyll-a for less nitrogen. TLc and TLp (TP) showed a weak relationship. Generally, the New Zealand TLI system characterises

Ugandan crater lakes from a mesotrophic to hypertrophic state, but, the results are not accurate due to lack of similarity among calculated parametric sub-indices. The inaccuracy is attributed to differences in the systematic functioning between Ugandan crater lakes and New Zealand lakes noted above such as predominant nitrogen limitation, high chlorophyll-a synthesis, and a low Chl-a: TN ratio in the crater lakes.

To address the mismatch in the parametric sub-indices, the New Zealand TLI model has been used to formulate a TLI system that is specific to Ugandan crater lakes which increased the similarity in computed TLI sub-indices. The new TLI system improved on the accuracy trophic state estimations and showed that crater lakes range from mesotrophic to hypertrophic state.

Crater lakes with a trophic state above mesotrophic are characterised by extensive catchment modification through agriculture and human settlement. Modified catchments generate nutrients from agricultural activities, such as fertiliser use and farm waste, in addition to household and institutional faecal treatment systems, such as unlined pit latrines and septic tanks. All nutrients generated have the potential to be transported into crater lakes through various pathways. To address the challenge of eutrophication in Ugandan crater lakes, a suitable monitoring programme, such as trophic level index system, should be adopted as the foundation for effective crater lake management. This will also provide a baseline to develop effective nutrient control strategies for craters lakes catchments.

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Chl-a	Chlorophyll-a
DO	Dissolved oxygen
GPS	Geographical Positioning System
NWSC	National Water and Sewerage Corporation
NED	N-1-Naphthylethylenediamine
SD	Secchi Depth
TN	Total nitrogen
TP	Total Phosphorus
TLI	Trophic Level Index
TLx	Trophic Level sub-index
TLp	Trophic Level Index for total phosphorus
TLn	Trophic Level index for total nitrogen
TLs	Trophic Level index for Secchi depth
TLc	Trophic Level index for chlorophyll-a
TSI	Trophic State Index

Chapter 1 **: INTRODUCTION**

1.1 General Background

Eutrophication is generally used to refer to processes and symptoms that respond to the fertilisation of the aquatic systems (Schindler et al., 2008). The most common characteristic of eutrophication is the development of algal biomass, which reduces water clarity. Algae are an essential component of aquatic ecosystems, being active sources of organic matter that support food webs (Pearl, Fulton, Moisander, & Dyble, 2001). Algal growth in freshwater is often controlled by the supply of nutrients (nitrates and phosphates) and light (Cunha, do Carmo Calijuri, & Lamparelli, 2013). Nutrient supplies can be natural sources, such as weathering of catchment rocks (mostly phosphorus (P)) and atmospheric nitrogen (N) fixation by cyanobacteria (Kumar & Krishna, 2013). Nutrients are also a frequent consequence of anthropogenic activities (Ansari, Gill, Lanza, & Rast, 2011), such as agriculture, urbanisation and industrialisation (Downing et al., 1999). Continuous accumulation of nutrients in a waterbody accelerates the primary production of algae and other aquatic plants, but it can also lead to undesirable aspects of eutrophication (Pearl et al., 2001). Eutrophication is marked by increased algal growth, which leads to water quality decline through discolouration, foul smells, and tastes of the affected water, depletion of dissolved oxygen, and alteration of the aquatic food web (Pearl et al., 2001). In addition, when the algal composition becomes dominated by cyanobacteria (blue-green algae), it can produce cyanotoxins that have adverse health effects on humans and animals that come into contact with, or consume, contaminated water (Carmichael, 2001). Studies from various countries have shown that natural and anthropogenic eutrophication contributes significantly to cyanobacterial blooms and are increasingly adding stress to freshwater supplies (Ansari et al., 2011).

To facilitate lake understanding and management, lakes are often classified based on their trophic state, using a trophic level index (TLI) (Ansari et al., 2011). Such a classification most often takes into account water quality variables, such as total phosphorus (TP), total nitrogen (TN), chlorophyll-a (Chl-a), and Secchi depth (SD). The above variables can be used define the trophic condition of a lake and also to monitor water quality changes in a lake over time. Different scholars have taken different approaches to establishing a TLI of waterbodies for different regions. Most of the established index models comprise a continuum of trophic states, such as ultra-oligotrophic, oligotrophic, mesotrophic, eutrophic, supertrophic, and

hypertrophic (Arkadi, 2010; Burns, Rutherford, & Clayton, 1999; Carlson, 1977; Chapra & Dobson, 1981; Cunha et al., 2013). Trophic level index systems, however, tend to be region specific because of systematic differences that exist in waterbodies across different regions. For instance, studies show that tropical lakes tend to be nitrogen limited whereas temperate lakes are mostly phosphorus limited (Huszar, Caraco, Roland, & Cole, 2006). The nutrient enrichment determines the extent of phytoplankton growth in a waterbody. Phytoplankton, estimated by measuring Chl-a concentration (Schindler et al., 2008), is often the principle variable used to define the trophic state. The concept of a limiting nutrient is important in the management of eutrophication, and can be determined by examining the relationship between Chl-a, and each individual nutrient.

There are many freshwater lakes in tropical Africa and the continent is renowned for its large number of tropical lakes of strikingly different sizes and diversity (Talling, 1986). The lakes may be categorised based on their formation processes, size, and location, but most of them owe their formation to the development of, and tectonic activities related to, the Afro-Arabian Rift System (Mills, 2009). Since the beginning of limnological investigations in Africa, around the early 1930s, investigations have centred on the African Great Lakes with limited attention paid to the small freshwater lakes such as volcanic crater lakes (Kizito, Nauwerck, Chapman, & Koste, 1993). Volcanic crater lakes are of particular significance and are numerous and wide spread in the eastern (Uganda, Kenya, Sudan, Tanzania) and western (Cameroon) parts of Africa (Efitre, 2006; Melack, 1976; Mills, 2009). The East African Rift Valley has the largest aggregation of lakes in the tropics (Bootsma & Hecky, 2003) and the majority of them are crater lakes. The crater lakes span a salinity gradient from dilute to hypersaline and display an array of morphological, geological, chemical, physical, and biological characteristics (Mills, 2009). The crater lakes are of great social and economic value to the local people through recreation, provision of water and fish supplies. Despite the various recognised values of crater lakes, Bootsma and Hecky (2003) argued that for the past 40 years, the research interest in African freshwater lakes centred on the need to improve and manage fisheries, and little attention was given to problems of eutrophication or nutrient influx into the lakes, both of which led to unprecedented continuous deterioration of productive water resources.

Uganda has a total area of 241,000 km^2 and 16% (38,560 km^2) of the land surface is covered by freshwater resources (Okello, Portmann, Erhard, Gademann, & Kurmayer, 2010). Lakes account for a significant amount of the available freshwater resources. There are about 89 small volcanic crater lakes in the western part of Uganda along the foot hills of Rwenzori Mountain

(Efitre, 2006; Melack, 1976, 1978). The crater lakes span strong variations in productivity, mixing regimes, morphometry, and salinity (Rumes, Eggermont, & Verschuren, 2011). They are grouped into four main geographic clusters: Fort Portal, Kasenda, Katwe–Kikorongo, and Bunyaruguru clusters (Mills, 2009). They range from latitude 0° 42' N to 0° 19' S and also vary in elevation from 1520 m to 895 m above sea level (Melack, 1978). Two outstanding characteristics are known for the crater lakes in Western Uganda. They present significant limnological variety and there is a natural division into groups based on gradients of either one or two factors, salinity gradient across all lakes and total phosphorus gradient across the dilute lakes (Melack, 1978; Mills, 2009). The dilute lakes are located in Fort Portal and Kasenda (northern cluster) and Bunyaruguru (southern cluster) on the shoulder of the Western Rift Valley whereas the saline lakes lie on the rift valley floor and tend to be hydrologically closed (Rumes et al., 2011). The priority lakes for this study are the freshwater crater lakes close to Fort Portal in Kabarole district Western Uganda. Fort Portal is the principle administrative and commercial town in Kabarole district. Fort Portal is designated to become a tourism city in accordance with the government of Uganda's vision 2040 (Adiyia, Vanneste, Van Rompaey, & Ahebwa, 2014), and crater lakes are significant to the development of tourist infrastructure in this part of the country. The target crater lakes in this study lie within a 30 km radius of Fort Portal and five lakes already have tourism infrastructure such as hotels, lodges, and camp sites linked to them.

Documents show that water quality studies on the large East African tropical lakes such as Lakes Victoria, Turkana, George, Edward, and Kyoga were started in the early 1930s (Mills, 2009) while the limnological investigations on small crater lakes began in the 1970s, with the pioneer studies being short-term and sometimes involving only a single sample from study lakes (Mills, 2009). The first limnologic study on the Ugandan crater lakes was conducted by Beable (1974) who studied the meromictic stratification patterns, and this was followed by Melack (1976), whose survey examined the morphometry, hydrology, and chemical nature of the crater lakes without including biological investigations (Kizito et al., 1993).

Observations from over 10 years of work on Western Uganda crater lakes suggest a steady decline in water quality through eutrophication as result of deforestation around the crater rims by the surrounding communities to meet the increasing demand for wood fuel, building material, and agricultural land expansion (Efitre, 2006). Investigations on the impact of human activities on crater lakes in Western Uganda are still scarce (Rumes et al., 2011). Deforestation facilitates sediment transport and increases nutrient loads entering the lakes, which in turn leads

to phytoplankton growth and eventual eutrophication. One of the marked responses to eutrophication and phytoplankton growth is reduced water clarity due to increased algal biomass and turbidity. Efitre (2006) observed a reduction in water transparency and increased Chl-a concentrations in some of the deforested crater lakes in Western Uganda. Lake Saaka, also part of this study, is reported to be hypertrophic with murky green waters showing high phytoplankton density dominated by the *Microcystis* species (Okello & Kurmayer, 2011). Increased human settlement patterns and agricultural expansion across the catchments of Lake Saaka fuelled routine burning of vegetation resulting into elevated phosphorus loading and accelerating eutrophication processes (Kizito et al., 1993; Rumes et al., 2011). Based on aquatic productivity investigations, some scholars have attributed certain changes in the crater lakes' ecosystem function to eutrophication. Chapman and Chapman (2003) and Efitre (2006) all suggest that the clearing of forests in the crater lake catchments in Western Uganda and the decline in water quality is closely linked to ecosystem alterations, such as reduction in fish productivity and fish depletion.

In the existing literature, documented evidence was not found to suggest that any systematic monitoring of monitoring of eutrophication had been conducted on the high altitude crater lakes in Western Uganda. Furthermore, the available information from studies on the limnological assessment of crater lakes does not allow spatial-temporal analysis of the eutrophication challenges among different crater lakes. Melack (1976) noted that few measurements on the primary productivity of the crater lakes were available to conduct comparative studies on the biological productivity and that situation remains true today. Some published short-term water quality studies on lakes, such as Lake Saaka, show discrepancies in data due to unreliable and less than accurate laboratory analytical techniques and irregular sampling (Mills, 2009). To generate a reliable and independent data set, this study has assessed the trophic state variables and catchment descriptions for the study crater lakes, and evaluated the application of techniques for lake monitoring developed in New Zealand to determine whether monitoring and management strategies can be developed for the Ugandan crater lakes. This research envisaged that monitoring will not only help to manage and prevent eutrophication, but also avoid costly remedial actions on degraded lakes. Six volcanic crater lakes: Saaka, Kyaninga, Nyinambuga, Nyabikere, Mwamba, and Katanda, are used as examples in this study and all are considered small (Efitre, 2006). Lake Saaka is the largest with an area of 0.64 km^2 while Lake Kyaninga is the smallest with an area of 0.32 km^2 and the lakes exhibit variations in altitude, size, and depth (Table 5). Short-term surveys have indicated that the target crater lakes and

many others in the area are undergoing water quality decline manifested by increasing algal blooms. The phytoplankton community in Lake Kyaninga is already dominated by cyanobacteria, but is also comprised of chlorophyta (green algae) and diatoms (Cocquyt, Plisnier, Gelorini, Rumes, & Verschuren, 2010). Since the 1970s, Lake Saaka has been undergoing cultural eutrophication, based on measurements of SD, conductivity, and dissolved oxygen of its surface water (Mills, 2009). It is considered hypertrophic with cyanobacteria accounting for more than 50% of the phytoplankton biovolume (Okello & Kurmayer, 2011).

In addition to nutrient loading, other factors that influence algal blooms include thermal stratification of deep and stagnant water bodies, light penetration, and temperature regimes (Færøy & Barton, 2004). For example, Lake Kyaninga is permanently stratified below 100m, above this depth the lake has a varied mixing frequency between 8-12 m depth daily, 39-47m at least once year and 47-100 m once over several years or decades (Cocquyt et al., 2010). Deeper lakes have the capacity to store P loads in the sediments that can be released depending on factors like pH, dissolved oxygen, mixing, and climate change. There is a greater possibility of sediment nutrient release in shallow lakes than in deep permanently stratified lakes, and furthermore cyanobacteria species such as *Microcystis* have been found to dominate shallow eutrophic lakes that potentially recycle the nutrients (Okello & Kurmayer, 2011). Tropical lake mixing is driven by wind, humidity, and rainfall but not seasonal temperature changes as observed in temperate lakes (Mills, 2009). Deeper volcanic crater lakes in Western Uganda with steep sided walls are sheltered from winds and are permanently stratified with less productivity (Mills, 2009). The paradigm appears to be consistent with the old laws of limnology which suggest that shallow lakes are more productive than deep ones (Kilham & Kilham, 1990), and to this end, this study of different lakes allowed the extremes of size and mixing regimes to be addressed.

1.2 Eutrophication

Eutrophication and the consequent development of algal blooms is a worldwide water quality challenge, threatening freshwater usage (Qin, Liu, & Havens, 2007). Eutrophication involves the influx of excess nutrients into waterbodies which causes the disproportionate growth of algae forming blooms (Rast & Lee, 1978). Algal blooms induce biological, chemical, and physical disturbances in aquatic systems, causing diurnal pH changes, oxygen depletion, temperature changes, odours, fish kills, reduced transparency, and degraded aesthetic values (Leng, 2009). The most noxious phytoplankton group is cyanobacteria, which can produce toxins that pose a major risk to human and ecosystem health and are also often associated with eutrophication (Carmichael, 2001; Galvez-Cloutier & Sanchez, 2007). More than 50 bloom forming cyanobacterial species have been identified and the majority have the capacity to produce toxins that can be either dissolved in lake water (extracellular) or remain in cyanobacteria cells (intracellular) (Ansari et al., 2011). During water treatment, the free extracellular cyanotoxins are difficult to remove and this poses health risks to human such as dermototoxins, hepatotoxins, and fatal neurotoxins (Carvalho et al., 2013). Cyanotoxins can also kill fish and shellfish leading to food web alterations in an aquatic environment (Pearl et al., 2001). The decline in water quality and the reduction of aquatic organism populations thus have social, ecological, and economic consequences. These factors make eutrophication a critical concern to be monitored and addressed in all freshwater environments. The understanding of the relationship between nutrient sources, transfer pathways, and consequences in waterbodies is essential in dealing with the eutrophication problems (Figure 1).

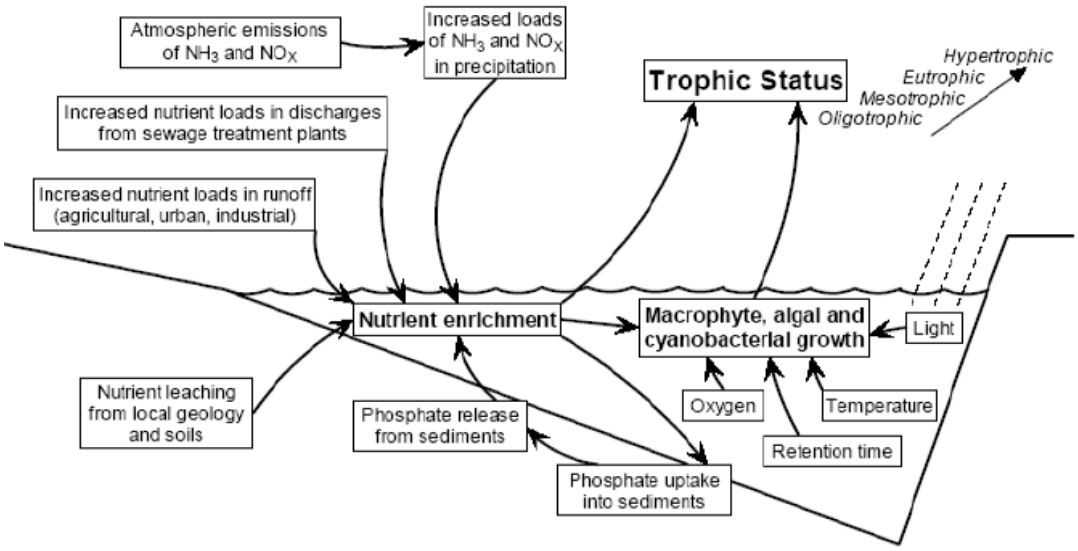


Figure 1: Nutrient cycle in a water systems indicating the causes and consequences of eutrophication (Van Ginkel, 2011)

1.2.1 What causes eutrophication?

Eutrophication can result from both anthropogenic and natural causes. Cultural eutrophication is the term given to anthropogenic activities that generate nutrients that are later released into waterbodies (Leng, 2009). Catchment activities such as deforestation, application of inorganic fertilisers on irrigated farmland, discharge of untreated waste effluents, and grey water from rural and urban settlements are some of the known anthropogenic nutrient sources. For example, Lake Victoria is considered eutrophic due to increased human pressures and urbanisation (Muyodi, Bugenyi, & Hecky, 2010) in the surrounding towns and cities located in the riparian countries Uganda, Kenya, and Tanzania.

Eutrophication by natural causes includes nutrient release from weathering of rocks, from soil, air, and other catchment processes, such as organic decomposition of plants. A common example is the nitrogen fixing bacteria that convert atmospheric nitrogen gas into bioavailable forms (Kumar & Krishna, 2013). In aquatic environments some blue-green algae are able fix atmospheric nitrogen gas and use it to support growth (Ansari et al., 2011). High natural phosphorus has been reported to exist in different parts of Africa, mainly in volcanic areas, and this also influences the nutrient budgets of volcanic lakes (Melack, 1978; Mills, 2009). Natural eutrophication also occurs as lakes age and begin to accumulate sufficient nutrients for algal blooms to occur. Van Ginkel (2011) argues that for eutrophication to take place, high nutrient accumulation must be accompanied by prolonged periods of stagnation of the water

body, suitable temperature and good sunlight regimes. McDowell, Biggs, Sharpley, and Nguyen (2004) believe that aquatic producers are sensitive to even minor inputs of nutrients, such as phosphorus, which implies algal blooms can progressively increase with a gradual increase in nutrient concentration. Pearl et al. (2001) observed that for some lake systems, algal blooms can form under low nutrient loading conditions because they have the capacity to accumulate passively through buoyancy compensation, for buoyant species, under stable water column conditions, or through active mechanisms, such as swimming migrations and high growth rates where growth and division are fast. Species such as *Microcystis* vertically migrate through the water column and take up phosphorus at the sediment water interface and then rise to the water column to form blooms (Conley et al., 2009).

Okello and Kurmayer (2011) reported natural eutrophication in three Ugandan freshwater lakes: George, Edward, and Mburo. These lakes exhibited high concentrations of total phosphorus (>100 µg/l) and chlorophyll-a (>70 µg/l), depicting hypertrophic conditions. The authors suggested that this was due to all the lakes being shallow, accumulated overtime nutrients from several rivers running from the Rwenzori Mountain and the lakes being a habitat of abundant wildlife such as hippopotami whose excreta is believed to contribute large amounts of nutrients (Okello & Kurmayer, 2011). The location of the lakes in the national parks makes them vulnerable to excreta from wildlife which too can be regarded as natural eutrophication. Accumulation and release of nutrients in lake sediments is an important aspect of gradual eutrophication by both natural and cultural mechanisms. This is because nutrients trapped in bottom sediments can be released back into the water under reducing conditions and the phenomenon is particularly common with sediment-bound phosphorus because it tends to be released from phosphorus rich sediments when they go anoxic (Waters, 2016). Increased sediment oxygen demand through eutrophication can release more phosphorus and this leads to a positive feedback for algal blooms. In contrast development of hypoxia in the bottom waters can lead to the reduction of nitrates through denitrification and anaerobic oxidation of ammonia at the oxygenated/deoxygenated water interface (Conley et al., 2009). The above nutrient dynamics also regulate nutrient bioavailability in the surface water layers in the event of lake mixing (Lewis Jr, 2002). The observed increase in phytoplankton assemblage biomass in Lake Victoria since the 1980s is assumed to be a result of increased sediment-bound recycling of nutrients, whose accumulation was result of intensified lake catchment disturbances during the 20th century (Hecky & Bugenyi, 1992; Mills, 2009). Søndergaard,

Jensen, and Jeppesen (2003) argue that nutrient load from sediments can play an essential role in phosphorus dynamics, mostly for shallow lakes that are phosphorus limited.

When the duration of external nutrient loading is short, lake recovery from eutrophic to oligotrophic takes a short period (McDowell et al., 2004), but, longer periods of external nutrient loading excessively enriches sediments with excess nutrients that continue to be recycled in the lake system even after reducing external loads. A large resident load within a lake prevents, or delays, water quality improvements even after remedial actions are inferred to reduce external nutrient load (Søndergaard et al., 2003). Large and deep lake systems that form anoxic hypolimnion with reducing conditions and have less amounts of metals to form complexes with phosphorus are more vulnerable to elevated P loading from the hypolimnetic water layers (Kilham & Kilham, 1990).

In addition to nutrients and light, phytoplankton growth is further influenced by other factors such as light intensity, water temperature, and pH (Ansari et al., 2011). In this regard phytoplankton, and more especially blue-green algae, grow efficiently and abundantly with increased sunlight and temperature (Qin et al., 2007). Pronounced seasonal algal maxima are observed during dry seasons (in the tropics) and summer seasons (in temperate regions), when light intensity and temperature are high and there is low flow and high water residence time (Ansari et al., 2011). Incidentally these seasons can also be marked by increase in catchment activities, such as intensive irrigation on fertilised farmland.

Seasonal variability influences phytoplankton growth in freshwater bodies. Warmer seasons are linked to an increase in phytoplankton growth (Kalff & Knoechel, 1978) and colder seasons may well mean there is a reduction in plankton biomass, but, compared to temperate lakes, the lakes in the tropical region are potentially vulnerable to algal blooms throughout the year due to high air temperatures (Poste, Hecky, & Guildford, 2013). Conditions that enhance nutrient regeneration and increased algal productivity year around in the tropical lake systems were referred to as the "Endless summer" by Kilham and Kilham (1990).

Uganda has a complex patchwork of climatic regimes (Mills, Ryves, Anderson, Bryant, & Tyler, 2014) that are generally tropical (NEMA, 2016), modified locally by the presence of large inland waterbodies, topography, and maritime influences (Mills, 2009), with two wet seasons between March and mid-May (long rains), and October to December (short rains) (Endfield, Ryves, Mills, & Berrang-Ford, 2009; Mills, 2009; Mills et al., 2014). The remaining

intermediate months are dry with minimum precipitation (Okello & Kurmayer, 2011). These seasonal changes regulate phytoplankton growth in crater lakes. The light availability and high air temperature during the dry season generally facilitates an increase in phytoplankton biomass and the changes in rainfall regimes, run-off, and erosion have an influence on nutrient loading from the catchment into the lakes (Mills, 2009). Similarly, climatic influences on algal productivity have been documented in studies conducted on large tropical lakes. Akiyama, Kajumulo, and Olsen (1977) observed very high densities of *Anabaena* which accumulated on the surface of Lake Victoria during the dry period from November to January when the lake water was calm (Okello & Kurmayer, 2011). Regarding seasonality of phytoplankton growth in temperate regions, annual data shows increased phytoplankton growth in spring, summer, and autumn and reduced growth intensity during winter (Fruh, Stewart, Lee, & Rohlich, 1966). Unlike in temperate regions, in Africa and the rest of the tropical belt, the seasonality of phytoplankton is believed to be mainly regulated by either hydrologic (water input-output) or hydrographic changes (water column structure and circulation) (Talling, 1986).

1.2.2 Controlling eutrophication

Due to severe the consequences of eutrophication in freshwater ecosystems, there is need to build sustainable eutrophication control programmes. (Conley et al., 2009). Nitrogen and phosphorus are the critical nutrients that support phytoplankton growth (Ansari et al., 2011). The management and control of eutrophication and algal growth can be achieved primarily by reducing the amount of nutrients entering the waterbody, but there is also a need to understand the interaction between the physical, chemical, and biological factors (Pearl et al., 2001) that drive the complex relationships between algal blooms, nutrients, and the environment. For instance, nutrients may originate from point and nonpoint sources and may require different management strategies to mitigate eutrophication. Whereas the control of point source pollution can be achieved relatively easily, diffuse nutrient sources are difficult to both measure and regulate (Carpenter, Ludwig, & Brock, 1999). Because of this development of appropriate and specific management strategies, in accordance with the source of nutrients, is essential for the effective eutrophication management (Conley et al., 2009). Natural eutrophication presents another challenge when it comes to nutrient management. Studies in nutrient stoichiometry for temperate lakes indicate that there is more nitrogen concentrations than phosphorus (Kumar & Krishna, 2013) due to various natural nitrogen sources such as NH_4, organic matter

decomposition and atmospheric nitrogen deposition. Hence studies show that in many temperate waterbodies phytoplankton growth is largely limited by phosphorus rather than nitrogen (Kim, Hwang, Shin, An, & Yoon, 2007). In such conditions of elevated natural nitrogen supplies, reducing anthropogenic nitrogen influx may not achieve improved water quality hence attention must be focused on managing phosphorus.

Cultural eutrophication is markedly wide spread and the urgency to reduce anthropogenic nutrient inputs into freshwater systems has been extensively recognised (Conley et al., 2009). Because of the possibility of costly interventions, management strategies should aim for cost-effectiveness. For low-cost interventions the concept of limiting nutrients can be applied in eutrophication management. Huszar et al. (2006) argued that the algae-nutrient relationship in an aquatic environment generated an understanding of the limiting nutrient, which can be a valuable management tool. For instance, in the 1970s, Schindler conducted long term experiments, and found that phosphorus was the primary limiting nutrient in Lake 227 and, the reductions in phosphorus loading and removal resulted into positive improvements of lake water quality (Conley et al., 2009). Nutrient bioassay experiments are used to determine the nutrient limiting eutrophication and phytoplankton growth in lake. Lakes are typically considered to be phosphorus limited when the atomic TN:TP ratio is >15, and nitrogen limited when the atomic TN:TP ratio is <7 (Mills, 2009). The ratios between 7 and 15 indicate potential co-limitation by both TN and TP. Despite the experimental evidence, Schindler et al. (2008) asserted that eutrophication management cannot be achieved through nitrogen control, because nitrogen limitation is a symptom of over-fertilisation with phosphorus. The observation of short-term nitrogen limitation in bioassays thus may not be a reliable indicator of the potential to reverse eutrophication by controlling nitrogen. Mesocosm experiments and improved mechanistic understanding of eutrophication control show that nitrogen is prime factor for eutrophication in many lakes and estuaries in the temperate region, due to increased redistribution of reactive nitrogen in the biosphere through agriculture and urbanisation (Howarth & Marino, 2006). Given the above scholarly arguments, and depending on the sources of nitrogen and phosphorus in the lake's catchment, identifying the limiting nutrient will focus the management interventions in a cost effective way and achieve the desired water quality. It may be difficult to separate anthropogenic nutrient impacts from natural ones (Larson, Collier, & Buktenica, 2007), but there is a growing body of evidence that eutrophication and aquatic primary production are much enhanced by anthropogenic nutrient pollution. With the availability of adequate resources, it is imperative that where possible dual-

nutrient management strategies be implemented when controlling eutrophication (Conley et al., 2009) in order to sustain water quality improvements.

1.2.3 Trophic state classification

Freshwater lakes can be classified based on their trophic status (Carlson, 1977; Mills, 2009), mixing (Lewis Jr, 1983), and basin type, or by origin (Wetzel, 2001). For water quality purposes, the trophic classification is the most relevant and also the most broadly accepted method (Lewis Jr, 1983). This is primarily because it evaluates chlorophyll-a (as a descriptor of algal biomass), which is the root cause of great public concern in many aquatic systems due its severe consequences. (Howarth & Marino, 2006). Despite a number of trophic state schemes that have been proposed by researchers, the Trophic State Index (TSI) system, that uses a continuum of trophic states to classify lakes, offers a great potential to standardise the concept and has been widely adopted (Kalff & Knoechel, 1978). Trophic state classification depends on the degree of organic and inorganic nutrient enrichment in the lake (Ansari et al., 2011), the depth of light penetration, and algal biomass. The above parameters can be measured in terms of total phosphorus, total nitrogen, Secchi depth and chlorophyll-a. The four parameters indicate the life supporting capacity of a lake and can be used to classify lakes using trophic states in ascending order, from oligotrophic, mesotrophic, eutrophic, to hypertrophic. The classification can be conducted based on the measurements of each individual parameter and/or by averaging the sub-indices of the parameters into a single numerical TLI. The numerical TLI method is a simple water quality assessment approach (Lewis Jr, 1983) because it considers only a few parameters and these can be easily measured.

Before the TLI system was initially introduced, limnologists used to classify lakes using only two trophic states: Oligotrophic lakes, which were believed to be "under fed", and eutrophic lakes, which were "well fed" (Fruh et al., 1966). The trophic assessment was multi-parameter in context based on nutrient flux, organism numbers (plants and animals), rates of production, and physicochemical properties, such as morphology (Fruh et al., 1966). A multi-parameter index, however, was found to be limited in its application due to the number of parameters to be measured, biogeographical differences between lakes, and also because assumptions had been made, without clear evidence, that there were linear relationships between some of the indices (Carlson, 1977). In addition, Fruh et al. (1966) cast doubt on the effectiveness of the

multi-parameter index due to the diverse nature of the aquatic ecosystems and the limited resources available for assessing them at the time. Classifying lake systems based on organism numbers may depend on the taxonomic competence of the investigator and can be ecologically obscuring because it ignores the entire ecosystem functioning of the waterbody (Kalff & Knoechel, 1978).

During the 1970s classification was refined to include a mesotrophic state because this was believed to be a transition from an oligotrophic to eutrophic state (Rast & Lee, 1978). Even with an additional mesotrophic state, the nomenclatural categories assigned were still small in number, lacked clear delineation between them, caused information loss when lakes were lumped together, and the system was also insensitive to minor trophic changes (Carlson, 1977). Furthermore Shapiro and Maloney (1979) noticed that the few categories were typological, overlapping without standard limits. Although there was perceived improvement through the addition of more parameters, the classification still required the examination of several diverse criteria such as nutrient concentrations, shape of oxygen curve, and composition of flora and fauna (Carlson, 1977; Fruh et al., 1966), and these assessment methods required a lot of resources and time because of the dynamic nature of lakes ecosystem. Rast and Lee (1978) envisaged the need for a numerical trophic state index that would more appropriately assign trophic conditions based on simple and practical parameters that are statistically easy to measure.

Carlson (1977) established the initial numerical trophic classification of lakes based on the relationship between three parameters; chlorophyll-a, Secchi depth and phosphorus, within a scale of transparency variations. The relationship is based on the general understanding that increase in algal biomass reduces water clarity, and that algal growth is directly related to nutrients and, in the North American waters under study, the particular nutrient of concern was phosphorus. Thus an increase in phosphorus concentration generally led to an increase in algal biomass, which reduced water clarity. Carlson first established the relationship between transparency (Secchi disc) and algal biomass (chlorophyll-a), across a range of transparency lakes. He used Secchi depth values to construct the first Trophic State Index equation, TSI_{SD}, which defined oligotrophic, mesotrophic, and eutrophic in terms of water clarity (Galvez-Cloutier & Sanchez, 2007). By using regression equations of transparency against total phosphorus and chlorophyll-a, Carlson further developed two other indexes, $TSI_{Chl\text{-}a}$ and $TSI_{TP,}$ that yielded similar numerical values, on average, for all the lakes in the data set. Thus an integrated index, the $TSI_{Average}$, could be derived from the three variables (Galvez-Cloutier &

Sanchez, 2007). Carlson's TSI scale ranged from 0 (ultra-oligotrophic) to 100 (hypertrophic) (Table 1), and the index was constructed so that an increase in TSI by 10 units correlated with halving of the Secchi depth and doubling of the phosphorus concentration (Mills, 2009) and algal biomass. There were however, limits on the utility of such indices: the indices derived from individual variables can only allow the testing of hypothesis of ecosystem functioning in terms of a related variable (Kalff & Knoechel, 1978), and the primary dependence on Secchi depth may create inaccurate results in situations of very turbid, coloured and very clear waters (Carlson, 1977; Mills, 2009). Carlson's kind of classification does not show the combined effect of different water quality variables on the lakes trophic state. Cunha et al. (2013) further observed that Carlson's TSI was only applicate to temperate region lakes which studies have found to be mostly phosphorus limited. Application of such a trophic classification to warm water or tropical lakes, is not helpful because tropical lakes have specific sensitivities and systematic differences not shared with the temperate lakes. In particular many tropical lakes tend to be nitrogen limited, have high nitrogen losses (through denitrification) at high temperatures (Huszar et al., 2006), high rainfall leading to increased nonpoint pollution, high evaporation rates, and increased primary production throughout the year (Cunha et al., 2013).

Table 1: Carlson's trophic state classification for temperate lakes (Galvez-Cloutier & Sanchez, 2007)

TSI	Classification	Description
<30	Oligotrophic	Clear water, dissolved oxygen throughout the year in the hypolimnion
30-40	Oligotrophic	Deep lakes still exhibit classical oligotrophy but some shallower lakes will become more anoxic in the hypolimnion during summer
40-50	Mesotrophic	Water moderately clear, but increasing probability of anoxia in the hypolimnion during summer
50-60	Eutrophic	Lower boundary of classical eutrophic; decreased transparency, anoxic hypolimnion during summer, microphyte problems evident and warm water fisheries only
60-70	Eutrophic	Dominance of blue-green algae, algal scum probable, extensive macrophyte problems
70-80	Hypertrophic	Heavy algal blooms possible throughout summer, dense macrophyte beds, but extent limited by light penetration,
>80	Hyperutrophic	Algal scum, summer fish kills, few macrophytes, dominance of rough fish

In addition to chlorophyll-a, Secchi depth and total phosphorus, another vital water quality parameter, total nitrogen, has been added to the trophic level index variables to cater for nitrogen limited lakes and also expand the use of trophic level index concept to lake systems from different regions. The multivariate index can describe water quality changes over a period of time (Kumar & Krishna, 2013), and help to determine the limiting nutrient of phytoplankton

growth in different lakes or regions. In addition, more intermediate trophic categories such as ultra-oligotrophic, microtrophic, supertrophic, and hypertrophic have been added to the traditional continuum of three classes that were established by Carlson (1977) and other pioneer scholars, to enhance discrimination of the index and increase sensitivity towards small changes in the trophic status of lake systems. Burns et al. (1999) developed a multidimensional trophic classification for New Zealand lakes. The New Zealand classification system was based on five trophic index variables which included chlorophyll-a, total phosphorus, total nitrogen, Secchi depth for shallow non-stratified lakes. The fifth variable of Hypolimnetic Volumetric Oxygen Demand (HVOD) was added to cater for stratified deep lakes. The TLI for New Zealand lakes considered total nitrogen because many New Zealand lakes are nitrogen limited. Unlike the previous TLI classifications schemes developed from summer data, the New Zealand classification system was designed to use de-seasonalised annual average measurements of trophic level index variables (Burns et al., 1999).

Primarily trophic state determination of lakes is meant to monitor the current state of water quality, but lake classification is also intended to enhance the communication water related discourse between technical people and the public (Carlson, 1977). To this end an established numeric trophic state classification system such TLI can simply convey information about lake water quality in such away to be understood by different stakeholders with interests in the sustainable management and use of water resource (Arkadi, 2010). Better understanding of the lake's trophic status can also trigger freshwater management actions such as nutrient management programmes within the catchments of degraded lakes (Galvez-Cloutier & Sanchez, 2007).

1.2.4 Development of a trophic level index system

Development of a trophic level index is based on the empirical relationships between water quality parameters such as chlorophyll-a, total phosphorus, total nitrogen and Secchi depth (Cunha et al., 2013). The principle of TLI is to generate a composite numerical value which is a sum of the main water quality parameters under consideration. Several numerical TLI systems have been proposed by researchers, but the ideal TLI system should be composed of simple and practically measurable parameters whose values are determined easily using less sophisticated statistical and analytical methods (Rast & Lee, 1978). Carlson (1977) noted that an ideal TLI should be able to express the diverse aspects of trophic state from multi-parameter indices and display the simplicity found when using a single parameter index. It should be

noted that a TLI system from one region may not be a applicable another region, but rather can be adopted by either adding more parameters or performing distinct calibrations on adapted models and regressions with local data (Cunha et al., 2013). For instance, Cunha et al. (2013) adapted Carlson's model of temperate lakes to develop the trophic state index of tropical and subtropical lakes, which he described as the arithmetic mean of TSI_{TP} (trophic state index with respect to TP) and $TSI_{CHL\text{-}a}$ (trophic state index with respect to chlorophyll-a).

Trophic level indices that are estimated by averaging the trophic state sub-indices of any chosen TLI variables need to be weighted such that each sub-index yields a similar value (Kumar & Krishna, 2013). The general correspondence among the trophic level values and individual sub-indices is established in the form of a set of logarithmic function that connects the trophic levels with water quality parameters (Arkadi, 2010). Taking a general expression; $\mathbf{TL_i = a_i + b_i \log (Par_i)}$ where i varies from 1 to 4, Par_i, represents SD, TN, TP and Chl, and a and b are coefficients, the two coefficients need to be set to ensure that sub-indices converge for a given water quality. Aggregate water quality TLI can then be calculated as the arithmetic mean of the of the four trophic sub-indices whose trophic level values range from *2* (regarded as oligotrophic) to *7* (regarded as hypertrophic) (Arkadi, 2010). Using the above description, the aggregate TLI of a lake can be calculated as the arithmetic mean of the four sub-indices using Eqn 1.1.

$$\mathrm{TLI} = 1/4(\mathrm{TLc} + \mathrm{TLs} + \mathrm{TLp} + \mathrm{TLn}) \qquad \text{Eqn 1.1}$$

where TLc represents the trophic state index with respect to chlorophyll-a, TLs for trophic state index with respect to Secchi depth, TLp for trophic state index with respect to total phosphorus and TLn for trophic state index with respect to total nitrogen.

In applying the above approach, Burns et al. (1999) developed a TLI system for New Zealand lakes using linear regression models between the Chl-a and TP, SD, TN. The resulting empirical equations (Eqn 1.2 to 1.5) were used to calculate the trophic level sub-indices (TL_X) of each parameter in each particular study lake.

$$TLc = 2.22 + 2.54 \log (Chl - a) \quad \text{Eqn 1.2}$$

$$TLs = 5.10 + 2.60 * \log (1/SD - 1/40) \quad \text{Eqn 1.3}$$

$$TLp = 0.218 + 2.92\log(TP) \quad \text{Eqn 1.4}$$

$$TLn = -3.61 + 3.01\log(TN) \quad \text{Eqn 1.5}$$

The equations normalise the annual average values, and coefficients of constants are set so that, for an average New Zealand lake, the TLx values of the four parameters were similar. The TLI of a particular lake and year was finally calculated using the expression Eqn 1.1.

Because the New Zealand trophic level index system includes total nitrogen (due to nitrogen limitation in many of New Zealand lakes), it is a suitable TLI system to be tested among the tropical lakes where wide spread nitrogen limitation is common. The New Zealand TLI system has been applied to data from this study to identify the systematic similarities and/or differences between Ugandan crater lakes and New Zealand lakes.

Burns et al. (1999) proposed a monitoring and classification system for New Zealand lakes (Table 2) comprising seven trophic levels and class boundaries of the four key water quality variables; TP, TN, SD, and Chl-a, that define the trophic status for several New Zealand lakes.

Table 2: The trophic states and trophic level class boundaries of key variables that define different lakes in New Zealand (Burns et al., 1999)

Lake type	**Trophic level**	**Chl-a (mg/m^3)**	**Secchi depth (SD) (m)**	**TP (mg/m^3)**	**TN (mg/m^3)**
Ultra-Microtrophic	0.0 to 1.0	0.13 - 0.33	31 - 24	0.84 - 1.8	16 - 34
Microtrophic	1.0 to 2.0	0.33 - 0.82	24 - 15	1.8 - 4.1	34 - 73
Oligotrophic	2.0 to 3.0	0.82 - 2.0	15 - 7.8	4.1 - 9.0	73 -157
Mesotrophic	3.0 to 4.0	2.0 - 5.0	7.8 - 3.6	9.0 - 20	157 - 337
Eutrophic	4.0 to 5.0	5.0 - 12	3.6 - 1.6	20 - 43	337 - 725
Supertrophic	5.0 to 6.0	12 - 31.0	1.6 - 0.7	43 - 96	725 -1558
Hypertrophic	6.0 to 7.0	>31	<0.7	>96	>1558

1.2.5 Limitations of the trophic level index (TLI) system

Despite wide usage for trophic state estimation, the TLI approach has some limitations. In situations where different species of algae occupy different lake layers (Fruh et al., 1966), a common characteristic of stratified lakes, chlorophyll-a analysis of the surface water may not represent the standing crop of phytoplankton biomass in the water column. In some instances, the pigment composition in some phytoplankton groups is not constant hence biasing the chlorophyll estimations. Furthermore, different phytoplankton groups may interfere with Chl-a absorbance and, the chlorophyll content of algal groups vary with age (Fruh et al., 1966). Computing the arithmetic mean to a single TLI value remains a challenge due to the lack of similarity among the values of the sub-indices (Cunha et al., 2013).

Despite the same trophic state variables, their relationship varies with different lake conditions. For instance, the relationship between Chl-a and SD in shallow and turbid lakes with suspended sediments and naturally coloured geothermal lakes is different from that of deep and clear lakes. In coloured and turbid lakes, SD is not related to the amount of algal biomass. Turbid and coloured lakes are likely to show highly eutrophic conditions with respect to SD measurements without having undesirable characteristics of eutrophic conditions such as low dissolved oxygen, fish kills, toxicity and the noxious algal blooms.

1.2.6 Trophic state monitoring in Uganda and other tropical regions

Water quality decline in a Ugandan lake is difficult to assess, because the government does not have a consolidated national management tool (such as TLI) to determine and periodically monitor the state of water quality in lakes. Based on the available literature, none of the East African countries has developed a monitoring plan to assess the extent of eutrophication based on nutrients and chlorophyll-a or any other water quality variables. Mills (2009) argued that classification for tropical Africa lakes has not been explored and it is partly hindered by lack of water quality data to support development of a reliable trophic classification system. Some scholars have estimated the trophic states of Ugandan lakes by making comparisons with water quality from other regions, while others have chosen to directly apply TLI systems developed from other regions to estimate the current state of the Ugandan lakes. For instance Okello and Kurmayer (2011) classified high altitude volcanic crater Lakes Saaka, Mburo, George, and Edward as being hypertrophic by setting threshold values for the hypertrophic state of total phosphorus concentration at >100 µg/l and chlorophyll-a concentration at >70 µg/l . Similarly,

Cocquyt et al. (2010) estimated the trophic state of Lake Kyaninga based on the Swedish lakes classification system by Forsberg and Ryding (1980), and North American lakes TLI by Carlson (1977). Cunha et al. (2013) argued that a clear trophic state index system of tropical African lakes is needed for data grouping or organisation and water resource management decision making. The use of trophic classification systems meant for lakes in other regions risks overestimation of the enrichment conditions due to differences in land use shifts and climatological attributes.

In comparison to the findings of previous scholars with respect to trophic classification for tropical and subtropical lakes, Cunha et al. (2013) proposed a trophic classification for waterbodies in the tropical South America using monitoring datasets of lakes and reservoirs from Sao Paulo and South-eastern Brazil. The classification included six trophic categories including; ultra-oligotrophic, oligotrophic, mesotrophic, eutrophic, supertrophic, and hypertrophic. The chlorophyll-a and total phosphorus results were compared to the existing trophic state indices for temperate and tropical lakes systems (Table 3).

Table 3: Comparison of trophic state boundaries for total phosphorus and chlorophyll-a for tropical lake systems and temperate lake systems proposed by different researchers (Cunha et al., 2013)

Trophic state category	**Total phosphorus (µg/l)**			
	North American lakes	**OECD lakes**	**Tropical and subtropical lakes (Brazil)**	**Tropical and Subtropical (Sao Paulo-Brazil)**
	Carlson (1977), Carlson and Simpson (1996)	**(Vollenweider & Kerekes, 1982)***	**Salas and Martino (1991)****	**Cunha et al., 2013****
Ultra-oligotrophic	-	≤2.5	-	≤15.9
Oligotrophic	≤12.0	2.6–8.0	≤21.3	16.0–23.8
Mesotrophic	12.1–24.0	8.1–25.0	21.4–39.6	23.9–36.7
Eutrophic	24.1–96.0	25.1–80.0	39.6–118.7	36.8–63.7
Supereutrophic	-	-	-	63.8–77.6
Hypereutrophic	≥96.1	≥80.1	≥118.8	
Trophic State Category	**Chlorophyll-a (µg/l)**			
	Carlson(1977), Carlson and Simpson (1996)	**Vollenweider and Kerekes (1982)***	**Salas and Martino (1991)****	**Cunha et al., 2013****
Ultra-oligotrophic	-	≤0.7	–	≤2.0
Oligotrophic	≤2.6	0.8–2.1	≤3.6	2.1–3.9
Mesotrophic	2.7–6.4	2.2–6.3	3.7–6.7	4.0–10.0
Eutrophic	6.5–56.0	6.4–19.2	6.8–17.4	10.1–20.2
Supereutrophic	-	-	-	20.3–27.1
Hypereutrophic	≥56.1	≥19.3	≥17.5	≥27.2

***Annual arithmetic means, **Annual geometric means**

The class boundaries of total phosphorus indices of temperate lakes are more restrictive compared to the tropical lakes (Table 3), and this can be attributed to high rain fall intensity that increases nutrient loading in the tropical lakes (Cunha et al., 2013). The restrictive chlorophyll-a values in temperate lakes can also be explained by seasonal monitoring, mainly conducted during summer and spring seasons, corresponding with higher primary production during that time of the year. The short summer-spring period may limit the optimum growth of phytoplankton. Meanwhile, tropical lake systems have long periods of light availability and the potential for high temperatures, hence they are likely to be productive whole year increasing

the productivity of phytoplankton biomass (Cunha et al., 2013). Aquatic systems whose surface water temperatures periodically exceed 20°C and are stratified, have increased phytoplankton growth (Pearl et al., 2001) a condition typical of tropical lake systems. Cunha et al. (2013) reported that when Salas and Martino's (1991) trophic classification for warm water lakes was verified against water quality data from some African lakes, the results were quite close indicating the classification is likely do better on African lakes. There is, however, no reliable information where the South American classification systems have been applied to African lakes. Both temperate and tropical classification systems have been interchangeably used by different researchers based on the objectives of the studies. For instance, Okello et al. (2010) classified 12 Ugandan freshwater lakes based on a classification system proposed by Vollenweider and Kerekes (1982) for lakes in OECD countries, by using the following class boundaries;

Mesotrophic: TP (10–35 µg L^{-1}), chlorophyll-a (3–8 µg L^{-1}), Secchi depth (3–1.5 m)
Eutrophic: TP (35–100 µg L^{-1}), chlorophyll-a (8–25 µg L^{-1}), Secchi depth (1.5–0.7 m)
Hypertrophic: TP (100_350 µg L^{-1}), chlorophyll-a (≥25µg L^{-1}), Secchi depth (<0.7 m)

Table 4: Trophic level indices for SD, TP, Chl-a and the trophic states of 12 freshwater lakes from Uganda based on Vollenweider and Kerekes (1982) method of classification proposed for temperate lakes in OECD countries (Okello et al., 2010).

Lake	**SD (m)**	**TP(µg L-1)**	**Chl-a (µg L-1)**	**Trophic state (OECD countries)**	**New Zealand TLI**	
					NZ TLI	**Category**
Swamp	0.1	138	0	-	6.9	Hypertrophic
Nyabikere crater lake	1.1	380	10.5	Eutrophic	5.8	Supertrophic
Nkuruba crater lake	3.1	76.5	8	**Eutrophic**	**4.7**	**Eutrophic**
Lake George	0.2	164.5	103	**Hypertrophic**	**7.0**	**Hypertrophic**
Lake Edward	0.4	123.5	33.5	**Hypertrophic**	**6.2**	**Hypertrophic**
Nkugute crater lake	0	40	5	Mesotrophic	4.4	Eutrophic
Lake Mburo	0.3	143	38.5	**Hypertrophic**	**6.3**	**Hypertrophic**
Lake Nabugabo	1.0	27	9.5	Mesotrophic	4.7	Eutrophic
Lake Victoria (Bunjako)	1.2	54	9	**Eutrophic**	**4.9**	**Eutrophic**
Lake Victoria(Murchison)	0.8	113.5	18	Eutrophic	5.6	Supertrophic
Lake Victoria (Napoleon)	1.1	72.5	10.5	Eutrophic	5.2	Supertrophic
Pond (Jinja)	0.2	217.5	27.5	**Hypertrophic**	**6.6**	**Hypertrophic**

Lakes Nkuruba, Nkugute, and Nyabikere are among the crater lakes in Western Uganda. Lake Nkugute was listed as mesotrophic whereas Lakes Nkuruba and Nyabikere were both eutrophic

(Table 4).The classification gives a general indication of water quality in the Ugandan lakes based on three parameters, but the exclusion of total nitrogen from the parameters considered casts doubt on the accuracy of the trophic estimation due reported dominancy of nitrogen limitation among tropical lakes. A classification system that considers total nitrogen alongside Chl-a, TP, and SD would be more appropriate for tropical lakes.

To illustrate the discrepancies in trophic predictions between in TLI models from different regions, the New Zealand trophic classification system has been applied to SD, TP and Chl-a data by (Okello et al., 2010) (Table 4) The regression equations from Burns et al. (1999) have been used to calculate the trophic level sub-indices TLc, TLp and TLs and the values averaged into TLI. Some of the trophic states agree with the OECD results while other differ. Where the trophic states do not agree, the estimates by the New Zealand system appear to be of a higher trophic status than the estimates by the OECD system. It not possible to conclude about the actual or near trophic estimation given that both systems are designed for different temperate lakes, and secondly, neither accounts for total nitrogen. Generally the findings demonstrate that different TLI systems are likely to give different trophic predictions, and that TLI systems are geographically specific.

1.3 Problem statement

The Lakes Saaka, Kyaninga, Nyabikere, Nyinambuga, Mwamba, and Katanda are among the 80 freshwater crater lakes located in Western Uganda (Efitre, 2006; Mills, 2009). The crater lakes have undergone water quality decline due to eutrophication and the increase of phytoplankton biomass. Phytoplankton growth reduces water transparency, depletes dissolved oxygen and produces toxins (Kumar & Krishna, 2013), all of which adversely affect the sustainability of freshwater use and disrupt ecosystem functions. The crater lakes are essential sources of domestic water and fish protein to a surrounding large community (Campbell, Hecky, Dixon, & Chapman, 2006; Poste et al., 2013). Socioeconomically, the lakes provide employment and revenue to the communities where the infrastructure developments such as camp sites and hotels built on the crater rims support tourism businesses.

Eutrophication and increased phytoplankton growth significantly risks the loss of social, economic and cultural values provided by the lakes. For instance, bloom forming cyanobacteria makes the water unsuitable for most of the recreational and consumptive uses by reducing water clarity giving it the undesirable murky green colour. Where phytoplankton standing crop is dominated by toxic cyanobacteria species, it poses a potential risk to humans and animals (Færøy & Barton, 2004). Cyanobacteria are the dominant phytoplankton species in most eutrophic lakes in Ugandan (Okello & Kurmayer, 2011) and most cyanobacteria species are capable of producing toxins. The genus *Microsystis* has been found in eutrophic crater lakes and further research showed that *Microcystis* proliferation may be favoured by shallow and eutrophic conditions (Okello & Kurmayer, 2011), and it is just these conditions that characterise crater lakes, such as Lake Saaka.

The exposure to cyanobacterial toxins risks harm to human wildlife and animals from getting hepatotoxins, neurotoxins, and dermatotoxins (Okello et al., 2010). Lake Saaka is hypertrophic and has high *Microcystis* levels (Poste et al., 2013). Although Lake Kyaninga has low aquatic productivity (oligotrophic/mesotrophic), cyanobacteria still dominate its phytoplankton community (Cocquyt et al., 2010) and threaten the opportunities provided by the crater lake. For instance, the spectacular Kyaninga Lodge was recently established on the rims of Lake Kyaninga. Visitors to the hotel and the resident community enjoy the amenity and recreational values of the lake by swimming and kayaking. The lake is also a source of potable water for the lodge and the community. In terms of ecosystem health and functioning, Efitre (2006)

noticed that eutrophication adversely affected fish production and growth, with stunting of fish and reduction in fish production in 20 crater lakes located in Western Uganda. The above crater lake values and functions risk being lost if nothing is done to address the declining water quality. The first stage of in managing eutrophication is to develop a coherent and representative scheme for crater lake water quality monitoring which can used to plan for appropriate intervention depending on the state of each particular lake.

1.4 Research aim and objectives

Aim of the study

The study aim is to establish the trophic status of crater lakes located in Kabarole district Western Uganda, and to determine whether methods developed for lake monitoring in New Zealand, specifically the Trophic Level Indicator approach (Burns et al., 1999), can be applied in Ugandan crater lake systems.

Specific objectives

The objectives of the study are to assess eutrophication in crater lakes in Western Uganda, identify some possible causes, and develop a trophic level index that may be suited to a long term monitoring programme. This has been accomplished by determining:

1. Water quality in terms of *in situ* measures of pH, dissolved oxygen, temperature, conductivity, and clarity in the target crater lakes
2. Concentration of chlorophyll-a
3. Amount of nitrogen and phosphorus in the lakes
4. The potential sources of nitrogen and phosphorus around and within crater lakes
5. The potentially toxic algae in the crater lakes
6. Application of the Trophic Level Index scoring system

The above objectives have allowed the identification and attribution of water quality issues and the development of monitoring and management strategies for the crater lakes to support sustainable utilisation.

Chapter 2 : **MATERIALS AND METHODS**

2.1 Study site

The study lakes were randomly selected from the northern cluster of (Fort Portal and Kasenda) crater lakes located in Kabarole district. Northern cluster crater lakes lie on the moist shoulder of the Western Rift Valley, and are typically considered to be dilute lakes (Mills, 2009), compared to crater lakes on the floor of the rift valley. The area around Kabarole district is characterised by rolling hills, and most of the lakes in this area occupy crater kettles with steep sided walls that were formed as a result of volcanic activities dating back to approximately 11,000 years ago (Efitre, 2006). The surface area of most of the crater lakes is less than 1 km^2 (Table 5) and have depth variations ranging from 0.25 m to >180 m (Chapman, Chapman, Crisman, & Nordlie, 1998; Kizito et al., 1993; Melack, 1978). The lakes in Fort Portal lie at a higher altitude (1520 m asl) than those in Kasenda area (1220-1400 m asl) (Mills, 2009). The climate and vegetation of the Fort Portal and Kasenda areas are similar (Melack, 1978). Both receive a bimodal rainfall pattern with two seasons: March-May and September-November (Chapman et al., 1998), with mean annual rainfalls of 1702 mm (1990-2005), mean daily minimum temperature of 14.9°C, and the mean daily maximum temperature of 20.2°C (Efitre, Chapman, & Murie, 2009). There is a pronounced natural vegetation gradient with altitude, and forest reserves such as Kibale forest in the Kasenda area have medium altitude, moist, semi-deciduous forest (Taylor, Marchant, & Robertshaw, 1999). In the non-protected areas, comprising the crater lake catchments, the natural vegetation has been intensively modified thorough land use (Efitre et al., 2009). Widespread deforestation has left crater lakes catchments vulnerable to run-off fostering sediment and nutrient transportation into the waterbodies. The details of the study lakes have been described by GPS locations, size, depth, and altitudes (Table 5).

Table 5: The GPS locations, altitude, size and depth of the study lakes. Data are derived from (Efitre, 2006; Mills, 2009; Rumes et al., 2011)

Lake	**GPS location**		**Altitude (m)**	**Size (km^2)**	**Depth (m)**
	Latitude	**Longitude**			
Kyaninga	0°42'8.11"	30°17'46.6"	1567	0.32	152
Katanda	0°29'0.67"	30°15'42.6"	1340	0.37	146
Nyinambuga	0°30'48.1"	30°19'17.4"	1408	0.40	130
Mwamba	0°27'55.6"	30°16'24.6"	1308	0.45	203
Nyabikere	0°30'5.83"	30°19'32.0"	1393	0.44	56.7
Saaka	0°41'19.2"	30°14'34.4"	1568	0.64	7.8

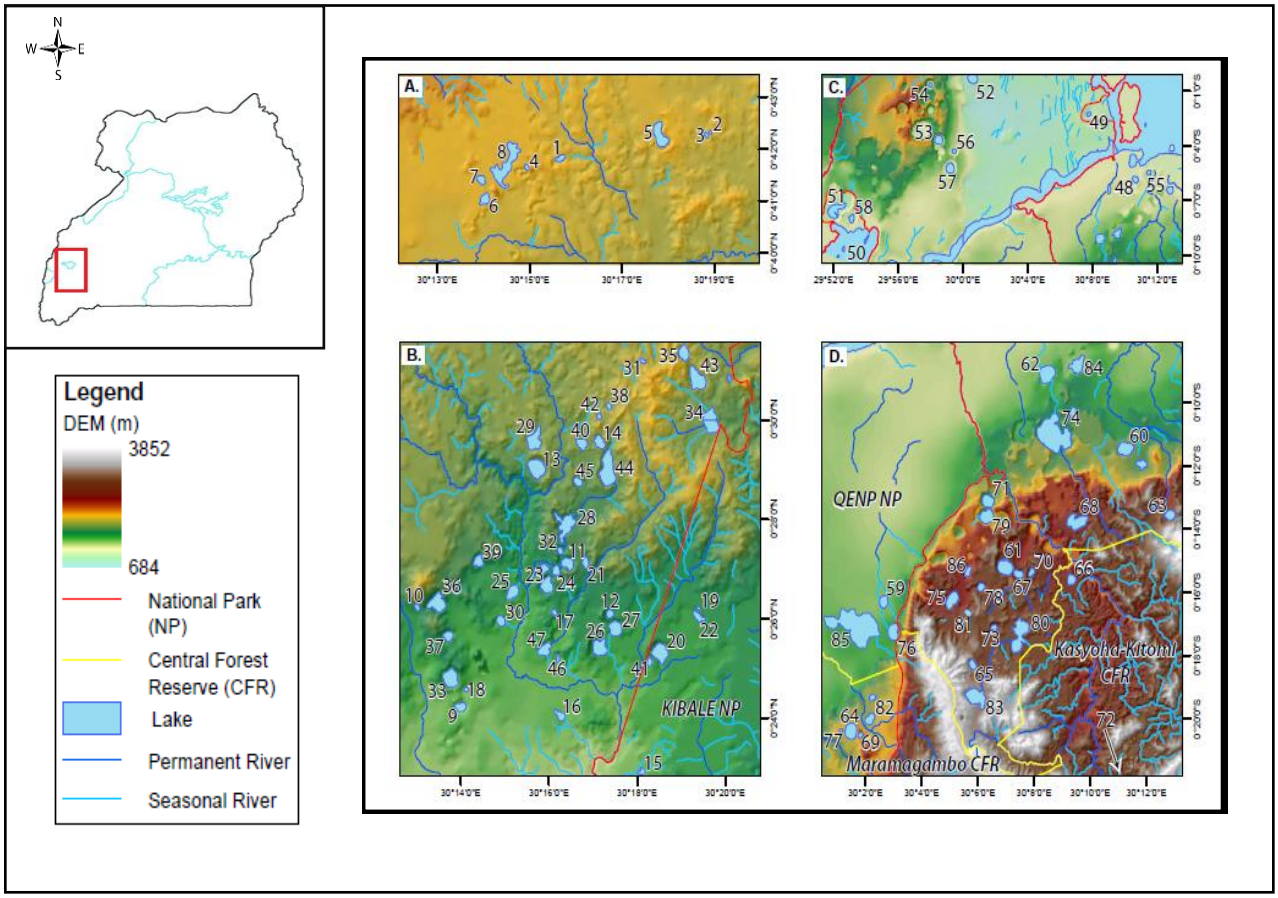


Figure 2: Elevation models showing the four main clusters of crater lakes. The lakes are numbered 1-86 across the clusters. (A) Fort-Portal, (B) Kasenda, (C) Katwe-Kikorongo, and (D) Bunyaruguru. Study lakes include 5, 8,13,28,43, and 44 (Mills, 2009)

The six study crater lakes lie within a 30km radius of Fort Portal. The Kasenda cluster has a higher concentration of craters, and more defined inflows and outflows, than does the Fort Portal area (Figure 2). I assume that the selected crater lakes represent the geological, hydrological and limnological characteristics of the rest of the lakes because they are all formed as result of volcanic activities, geographically associated with the Western Rift Valley, experience same climatic conditions, and their catchments soils are composed of volcanic ash and alluvial deposits (Mills, 2009), and densely populated.

2.2 Measurement of physicochemical parameters

For each of the study lakes, pH, dissolved oxygen, temperature, conductivity, and Secchi depth were measured on site.

2.2.1 Dissolved oxygen, pH, temperature and conductivity

Dissolved oxygen, pH, temperature, and conductivity were measured using a portable HACH HQ40D digital multi-meter. The pH and conductivity probes were calibrated with standard solutions prior to taking measurements. A one point verification of the dissolved oxygen probe at 100% saturation was carried out in humid air.

Measurements were taken by immersing the probes into the lake and noting readings from the meter until stability was reached. Each parameter was measured three times and averaged.

2.2.2 Secchi depth

Secchi depth measures the clarity of a waterbody. During fieldwork, water clarity was measured using two instruments; a Secchi disc when clarity exceeded 1 m, or a SHMAK clarity tube, when it was less than 1 m.

Secchi disc measurements were taken by lowering the disc into the waterbody and noting the depth when the disc just disappeared from sight. The disc was then lowered a little further and then raised until it reappeared and the depth recorded at this point. The SD was calculated as the average depth at disappearance and reappearance (Kilroy & Biggs, 2002). The measurement procedure was repeated three times and the values were averaged.

For lakes with clarity less than 1 m, the traditional Secchi disc gave an imprecise measurement, and therefore a water clarity tube was used to measure water clarity. Steel and Neuhauser (2002) developed the following protocol which was used for lakes with clarity less than 1 m. A clarity tube, a 1 m length of Perspex tubing with a clear Perspex base, was filled with water, a matt black disc inserted inside the tube and a stopper placed on the tube. The matt black disc was mounted on a magnet and could be moved inside the tube using an external matching magnet. While someone was looking horizontally through the water that was inside the tube, the black disc was moved away until it just disappeared from sight and the length noted on the

scale. The disc was moved further away and then back again until it just reappeared, and a second reading was taken. The two readings were averaged. The procedure was carried out three times and the values averaged to calculate the SD of the lake.

2.3 Water sampling

Surface samples were collected at a depth of 0.1-0.25 m. Two sampling points were chosen from each lake, one site inshore and the other offshore. The offshore samples were collected approximately 20 m from the inshore point, using a canoe or boat. Water samples of approximately 3 L were collected from each sampling point using a clean sampling container.

For chlorophyll-a analysis, at each lake, a precise volume of water was drawn from the collected sample using a graduated syringe and filtered through a glass fibre filter fitted in the filter holder of a plastic cartridge (Burns et al., 1999). The filtering process was repeated until it became impractical to filter more water through, and the total water volume filtered was noted down. The filter was removed from the cartridge and inserted in 15 ml centrifuge tube and chilled in a cool box. The filtration process was repeated three times and the sample centrifuge tubes were later transferred to National Water and Sewerage Corporation (NWSC) water quality laboratory in Kampala for chlorophyll-a analysis. The remaining water was returned to the laboratory in a chilled box for preparation of TN and TP samples. 50 ml centrifuge tubes were used to transfer the water samples for nutrient analysis to the laboratory. The centrifuge tubes were rinsed three times with sampled water before being used to prevent any contamination. A volume of 0.1 ml of concentrated H_2SO4 was added to 50 ml of the water samples for preservation and to prevent phosphorous from sticking to the plastic centrifuge tubes (Clasceri, Greenberg, & Eaton, 1998). The samples were kept chilled in a cool box and later transferred to a field laboratory for refrigeration. The laboratory analysis of total nitrogen and total phosphorus was carried out in New Zealand. During field work, the water samples were kept refrigerated in a Ugandan laboratory before being transferred to the water quality laboratory at Lincoln University in New Zealand for analysis.

2.4 Chlorophyll-a analysis

Laboratory handling of chlorophyll-a samples proceeded with the freezing the chlorophyll filter papers at -20^0C for one hour to burst phytoplankton cells.10 ml of 96% ethanol were added to chlorophyll centrifuge tubes and the samples were again kept dark and at 4^0C for 24 hours to allow chlorophyll extraction. After 24 hours, the samples were brought to room temperature

and placed in a warm water bath (60^0C) for approximately two minutes to fully extract any remaining chlorophyll. The chlorophyll extract was filtered and the extract was analysed by spectrophotometry, where absorbance was read at both 664 nm and 750 nm (Clasceri et al., 1998)

To detect low levels of chlorophyll-a in some of the water samples, a Turner Aqua Fluor fluorometer was used. This instrument measures chlorophyll by fluorescence and can give more accurate values for low levels of chlorophyll-a. In the laboratory analysis of chlorophyll-a, the fluorometer was calibrated using a solution of chlorophyll in 96% ethanol of known concentration. Both the spectrophotometer and the fluorometer were zeroed using 96% ethanol.

2.5 TN and TP analysis

A HACH spectrophotometer, in conjunction with appropriate reagents, was used to determine the concentration of total phosphorus and total nitrogen in the water samples. An alkaline persulfate digestion method was used to oxidise all forms of the two nutrients to nitrate and phosphate.

2.5.1 Total phosphorus analysis

The persulfate digestion of total phosphorus was carried out by mixing 18 ml of the sample with 3.75 ml solution of potassium persulfate and sodium hydroxide (Rice, Baird, Eaton, & Clesceri, 2012). The sample was digested in an autoclave at 120^0C for one hour and then allowed to cool to room temperature where, after neutralisation of the digest using sodium hydroxide, total phosphorus analysis was completed using the ascorbic reduction method. The principle for ascorbic acid reduction method is that ammonium molybdate and antimony potassium tartrate react with orthophosphate to form phosphomolybdic acid, which is reduced to molybdenum blue by ascorbic acid (Eaton, Clesceri, & Greenburg, 1995; Mackereth, Heron, & Talling, 1979) .

Procedure

A volume of 10 ml of the digested water sample was pipetted into two centrifuge tubes. One centrifuge tube was labelled blank and the other labelled sample. A drop of phenolphthalein indicator was added to each centrifuge tube. If pink colour developed, it was discharged dropwise with 5N sulphuric acid.

A combined sample reagent and combined blank reagent were prepared by mixing the following four reagents in the following proportions: 25 ml of 5N sulphuric acid, 2.5 ml of antimony potassium tartrate, and 7.5 ml of ammonium molybdate and 15 ml of ascorbic acid. The combined blank reagent was made with all the reagents, except for antimony potassium tartrate and ascorbic acid.

A volume of 1.6 ml of combined blank reagent was added to the sample blanks. 1.6 ml of combined sample reagent was added to the samples and this was done at one minute intervals. After 10 minutes, the absorbance was read in spectrophotometer at a wavelength of 880nm. Before taking the sample absorbance, the spectrophotometer was zeroed with deionised water.

Calibration of total phosphorus method

The concentration of total phosphorus of the water samples was determined by converting spectrophotometric absorbance into concentration using calibration curves. Potassium hydrogen phosphate was used to prepare a standard calibration curve for total phosphorus (Appendix B, Table B5). To estimate the recovery of organic phosphorus in the persulfate digestion, two standard curves were prepared using two organic phosphate solutions, glucose 6 phosphate (simple phosphate) and Inositol (stronger phosphate) (Appendix B, Table B4). Results showed positive linear response to organic P concentration and an 80-90% recovery with for glucose 6 phosphate and 60-70% recovery for inositol (Figure 3). Due to differences in the recovery of the two organic P compounds, the inorganic phosphorus calibration was applied to all the samples.

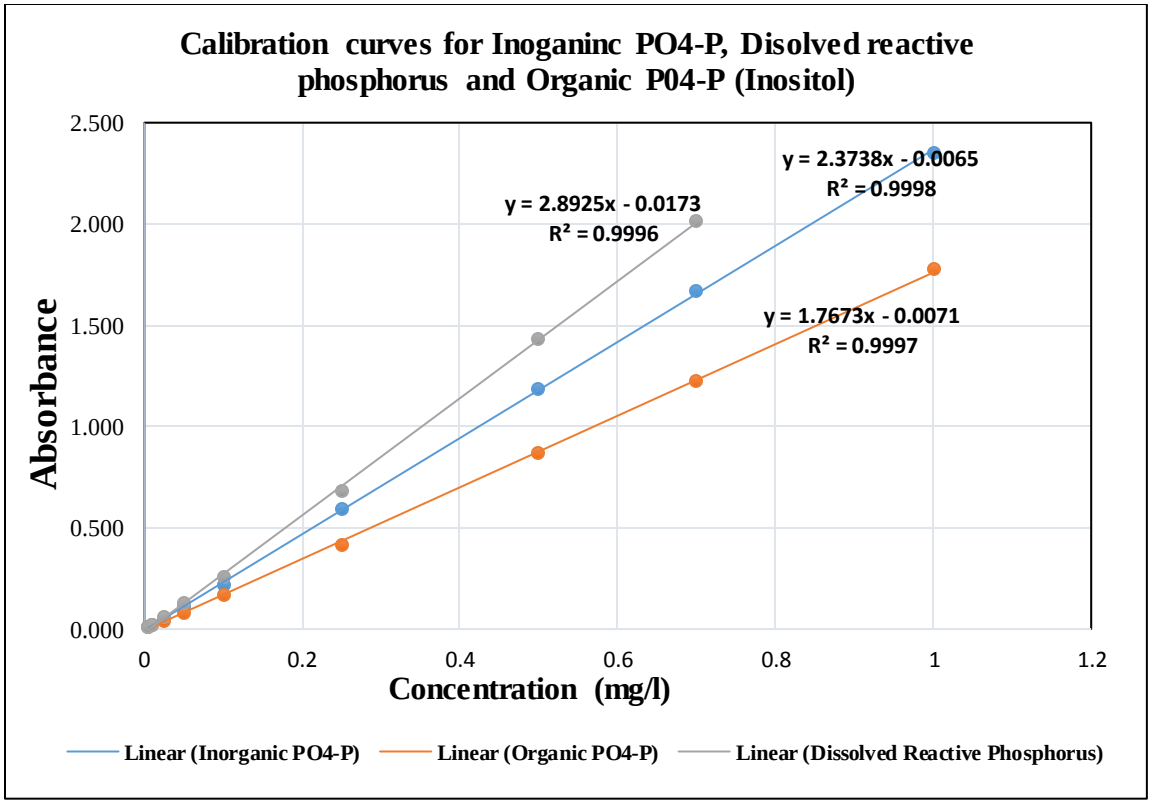


Figure 3: Calibration curves for total inorganic PO_4-P, dissolved reactive phosphorus and organic PO4-P (Inositol). Curves were fitted using least square regression in excel.

2.5.2 Total nitrogen analysis

The persulfate digestion of total nitrogen was carried out by mixing 18 ml of the sample with 3.75 ml solution of potassium persulfate and sodium hydroxide (Rice et al., 2012). The sample was digested in a hot block at 150^{0}C for one hour. The preferred method of autoclave digestion could not be used because of contamination issues associated with the autoclave available. After one hour, the digest was allowed to cool to room temperature, the digest neutralised, and total nitrogen was analysed as nitrate using the cadmium reduction method. The principle for the cadmium reduction method is that nitrate is reduced to nitrite in the presence of cadmium. Nitrite is subsequently diazotised with sulphanilamide and coupled with N-1-Naphthylethylenediamine dihydrochloride (NED) to form a coloured azo dye (Mackereth et al., 1979)

Procedure

The suitable pH for nitrate analysis is between 6-9 (Rice et al., 2012). After the persulfate digestion, the water samples pH were more acidic (below pH of 2). For this reason, the sample pH were adjusted towards neutral (6.5-7.5) using 3N sodium hydroxide. The pH was measured using a pH electrode.

A sample volume of 10 ml was auto pipetted into a 15 ml centrifuge tube. Then 3 ml of ammonium chloride was added followed by 1 ml of borax solution and 0.5 – 0.6 g of spongy cadmium. The centrifuge tubes were tightened and samples shaken using a wheel for 20 minutes. 1.4 ml of sample was then auto pipetted into new centrifuge tube and 200 µl of sulphanilamide added then mixed thoroughly. After 4-6 minutes, 200 µl of NED was added and mixed thoroughly, followed by 8.2 ml of deionised water to make the solution up to 10 ml. Sample blanks were prepared by adding 1 ml of the original digested sample to a centrifuge tube and adding 9 ml of deionised water to make the same concentration as the derivative sample. After 10 minutes, both the sample and sample blank absorbance were read in a spectrophotometer at a wave length of 543 nm. Before taking sample absorbance, the spectrophotometer was zeroed with deionised water.

Calibration of total nitrogen method

In principle, total nitrogen concentration in the water samples was estimated by converting sample absorbance into concentration using a standard calibration curves.

The standard calibration curve for total nitrogen was prepared by running standards of potassium nitrate through persulfate digestion (Appendix A), and to estimate the capacity of the method to recover organic nitrogen the samples, a curve for ethylenediaminetetra-acetic acid (EDTA) standards was prepared (Figure 4). The EDTA curve showed a linear response and 68-78% recovery of organic nitrogen. Because of the good recovery shown by EDTA, the organic calibration curve was used to determine total nitrogen concentration in the samples.

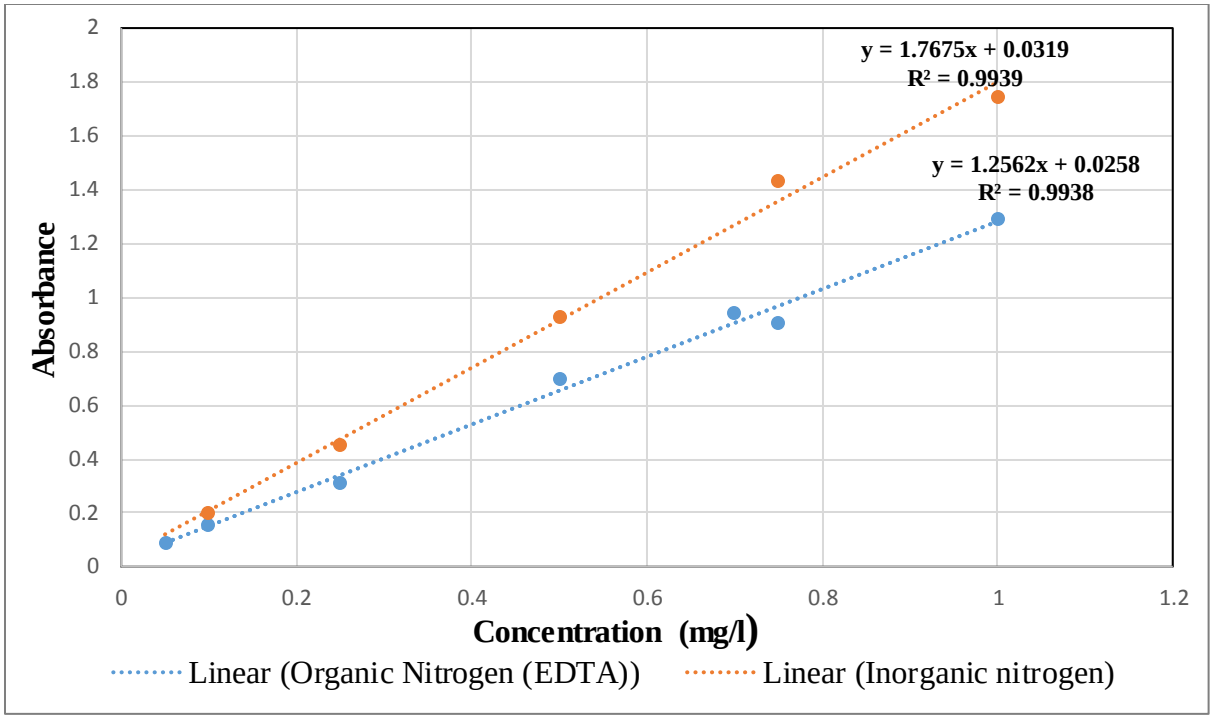


Figure 4: Calibration curves for inorganic nitrogen and organic nitrogen (EDTA). Curves were fitted by least square regression using excel.

2.6 Identification of potential sources of nitrogen and phosphorus in the catchments

The crater lakes in Western Uganda were formed by volcanic activities, have steep sided walls and are located at a high altitude (Mills, 2009). They are believed to have been formed when steep sided river valleys were blocked by flowing volcanic lava (Chapman et al., 1998). The lake catchments are predominantly occupied by rural communities and intensively used for agriculture because of fertile alluvial volcanic soils. A fieldwork catchment assessment was conducted to establish the influence human activities and natural factors may have on the water quality in the crater lakes. Generally the catchment assessment involved description of the existing anthropogenic activities that can potentially release nutrients into the lakes. Natural factors assessed included surface lake tributaries crossing the catchments and discharging into the crater lakes, catchment sizes, topography, riparian vegetation and their capacity to buffer crater lakes. Where surface tributaries were identified, water samples were collected and the nutrients then analysed. The findings were then used to establish the relationships between the catchment characteristics and the current water quality of the lakes.

2.7 Potentially toxic algae

Because of the inability to analyse the toxicity of the algae in the field, a qualitative procedure, based on international experience, was used to identify the presence of potentially toxic algae in the water samples.

Sampling toxic algae

Water samples of 500 ml were collected from the study lakes to investigate the presence of potentially harmful cyanobacteria. 5 ml of Lugol's iodine solution was added to preserve the water samples and encourage sedimentation (Clasceri et al., 1998). The preserved sample bottles were kept in the dark by wrapping them in aluminium foil, then chilled in a mobile cool box and transferred to NWSC laboratory for microscopic examination.

Laboratory examination of toxic algae

Qualitative methods were applied in the laboratory examination of cyanobacteria. The samples were left to settle overnight, and three quarters of the water was decanted off to leave the concentrated cyanobacterial cells at the bottom of the sample bottle. A plastic dropper was then used to extract a water sample from the concentrated sample to a glass slide and observed under a microscope at a magnification of 400x. Cyanobacteria cells were qualitatively identified to genus level, identification was based on *Cyanobacterial monitoring and cyanotoxin analysis* (Meriluoto & Codd, 2005), and morphological keys (cell forms) from the *International guidance manual for the management of toxic cyanobacteria* (Newcombe, 2009). An abundance of cells in the water samples was classified as dominant, abundant, occasional, or rare. If samples did not have any cells in the 500 ml samples, field sampling was repeated and the sample volume was doubled to 1 L. After settling at 4^0C for 48 hours, 800 ml of the liquid was decanted off and the remaining sample left to settle for another 24 hours, where more 150 ml of the sample was decanted of leaving only 50 ml. After subsequent decanting, the remaining volume was examined under the microscope.

Figure 5: laboratory setting for microscopic examination of cyanobacteria at NWSC central laboratory in Kampala, Uganda (Author's photo)

2.8 Data analysis and management

Both qualitative and quantitative data were produced in this study. Quantitative data were a result of field observations and the laboratory analysis of both the nutrients and chlorophyll-a. The data were entered into a Microsoft Excel spreadsheet and summarised. The results were presented using tables and in graphical form to enable easy visualisation. Linear graph correlations were used to show the relationships between water parameters.

The New Zealand trophic level index system (Burns et al., 1999) has been applied to calculate the trophic level indices of the TLI variables and extra graphical correlations for the sub-indices created.

Qualitative data came from microscopic observation of cyanobacteria and crater lakes catchment assessments. This data included catchment descriptions and classification of cyanobacteria species to genera using cell structural forms.

Chapter 3 **: RESULTS**

This chapter summarises data generated from field measurements, laboratory experiments, and qualitative descriptions. The first subsection shows a result summary of the physicochemical measurements. The second subsection summarises laboratory experimental results of nutrients and toxic algae. The last subsection summarises qualitative catchment assessment of the potential nutrient sources, dynamics of nutrient transport, and results of nutrient analysis from the tributaries of the crater lakes.

3.1 Physicochemical parameters

Table 6: Summary of physicochemical parameters of study lakes (June, July 2016).

Lake	DO (mg/l)	DO (%) Saturation	pH	Cond (µS/cm)	Temp (^{0}C)	SD (m)
Kyaninga (10:15 a.m.)	7.8	111.6	8.19	435.3	24.6	3.00
Nyinambuga (10:00 a.m.)	7.4	105.5	8.69	353.0	25.3	0.62
Nyabikere (3:25 p.m.)	6.7	94.7	8.67	267.7	24.6	0.26
Katanda (11:00 a.m.)	7.0	102.3	8.90	426.7	27.1	0.67
Mwamba (3:40 p.m.)	6.9	99.2	8.60	418.3	26.3	0.56
Saaka (9:50 a.m.)	12.8	179.0	7.52	649.3	22.7	0.22
Saaka (2:40 p.m.)	14.7	215.6	7.86	613.0	25.4	
Saaka (3:00 p.m.)	16.2	237.9	7.86	647.7	25.4	

The measurements were undertaken during June and July 2016, the dry season in Western Uganda; it did not rain during the course of this fieldwork, but some sampling mornings were overcast. The measurements for each lake were taken once between the hours of 10 a.m. to 4 p.m. In addition, the diurnal changes of the parameters were captured by two further measurements of Lake Saaka (9:50 a.m., and 2:40 p.m.). Dissolved oxygen ranges from 6.7 mg/l (94.7%) to 16.2 mg/l (237.9%) (Table 6). The results from Lake Saaka show that there was an increase in dissolved oxygen from morning to the afternoon. The pH values range from 7.52 to 8.9, with all lakes showing slightly alkaline pH. The results of Lake Saaka suggest that there is an afternoon rise in pH. The conductivity ranged from 267.7 µS/cm in Lake Nyabikere to 649.3 µS/cm in Lake Saaka. The results from the additional measurements at Lake Saaka indicate that there are no diurnal changes in conductivity. Temperatures varied from 22.7 ^{0}C to 27.1 ^{0}C. Secchi depth was greatest at Lake Kyaninga, at 3.0 m, compared to Lake Saaka which was the least transparent with a Secchi depth of 0.22 m.

3.2 Chlorophyll-a

Lake Kyaninga had the lowest chlorophyll-a concentration of 4.6 ug/l, while Lakes Nyabikere and Saaka had the highest at 164 µg/l and 172 µg/l respectively. The other lakes fell between these extremes (Table 7).

Table 7: Chlorophyll-a concentrations in the six surface water samples of the study lakes (June - July 2016). End value is the mean of inshore and offshore samples

Lake	**Chlorophyll-a (µg/l)**
Kyaninga	4.6
Katanda	12
Nyinambuga	24
Mwamba	48
Saaka	172
Nyabikere	164

3.3 Total nitrogen and total phosphorus

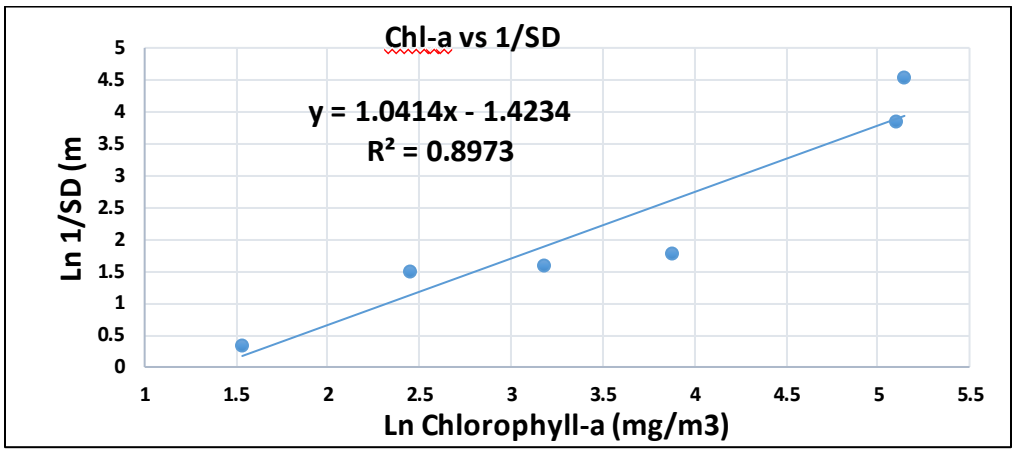
Nutrient concentrations varied among the study lakes. Concentrations of total nitrogen were always higher than those of total phosphorus. Lake Kyaninga showed the lowest TP and TN concentrations of 15 µg/l and 70 µg/l respectively. Lake Nyabikere had the highest TN concentration of 1140 µg/l, while Lake Saaka had the highest TP concentration of 125 µg/l, but both lakes displayed extremely high nutrient concentrations. The TN:TP ratios for lakes Nyinambuga, Mwamba, and Nyabikere were the highest, with similar values between 12 and 13.6 (Table 8). Lakes Kyaninga and Katanda had the lowest TN:TP ratios of 4.7 and 4.5 respectively. Lake Saaka shows an intermediate nutrient ratio, almost twice those of Lakes Kyaninga and Katanda.

Table 8: Total phosphorus and total nitrogen concentrations, and TN: TP ratios in the six study lakes. End value is the mean of inshore and offshore samples. TN: TP is expressed by mass.

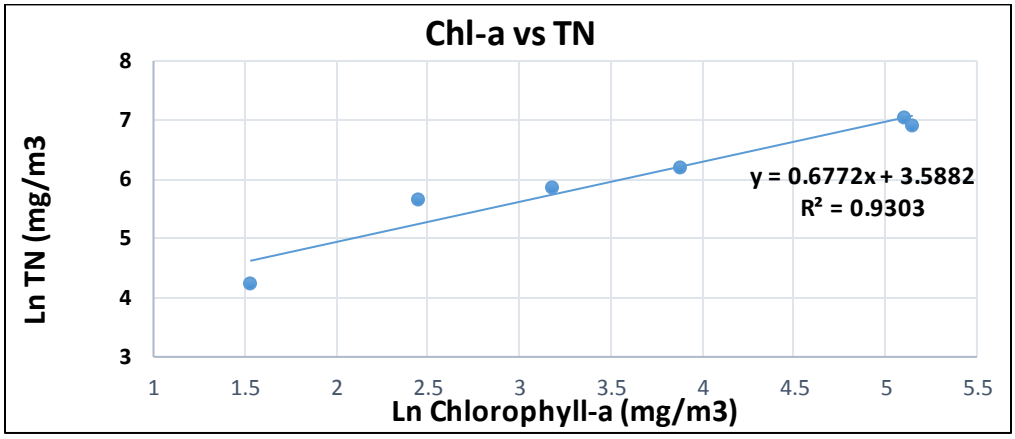
Lake	**TN (µg/l)**	**TP (µg/l)**	**TN:TP**
Kyaninga	70	15	4.7
Katanda	290	64	4.5
Nyinambuga	350	29	12.1
Mwamba	490	36	13.6
Saaka	1020	125	8.2
Nyabikere	1140	87	13.1

3.4 Relationship between trophic level index variables

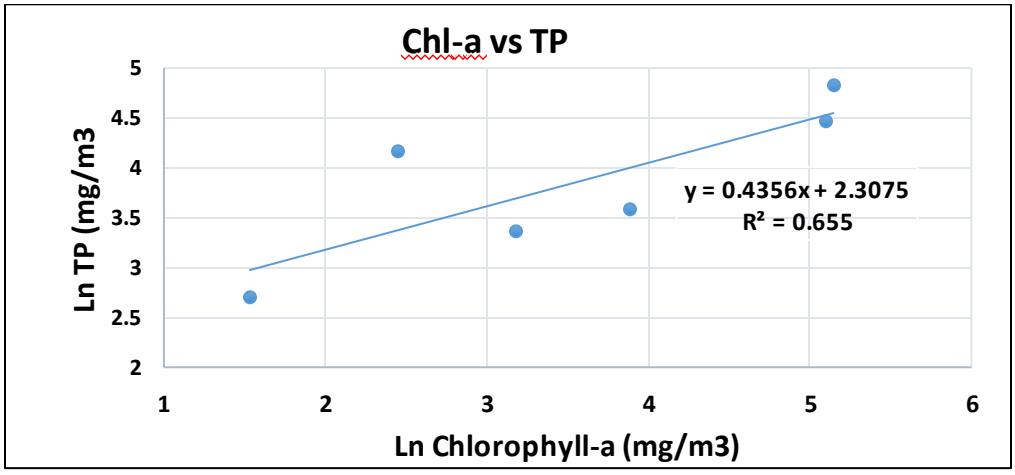
Total nitrogen, total phosphorus, chlorophyll-a and Secchi depth are among the commonly accepted variables for estimating the trophic state of freshwater lakes (Burns et al., 1999). Relationships have been created among the selected variables to calculate the trophic states of freshwater lakes, based on the assumption that there are causative links between this group of variables. By applying a similar approach to the results of this study, I explored relationships between chlorophyll-a and the other variables. The variables were all log-transformed to make them normally distributed.



(a) Chlorophyll-a and Secchi depth



(b) Chlorophyll-a and total nitrogen



(c) Chlorophyll-a and total phosphorus

Figure 6: Scatterplots and linear regressions of log-transformed chlorophyll-a versus; **(a)** Log 1/SD, **(b)** Log TN **(c)** Log TP

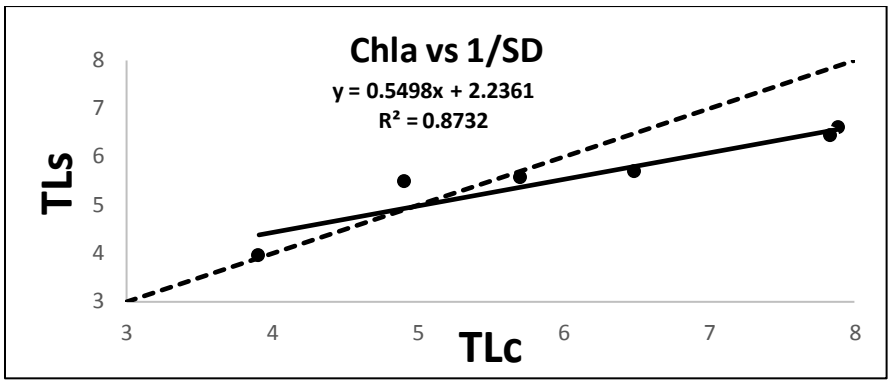
The relationships show positive correlations between chlorophyll-a and 1/SD, TP, and TN. The correlation coefficient R^2 suggests the strength of the relationships for the correlation (Figure 6). In this regard chlorophyll-a has a strong relationship with TN (R^2=0.9303) and 1/SD (R^2=0.8973), and a lesser relationship with TP (R^2=0.655).

Trophic level Indices

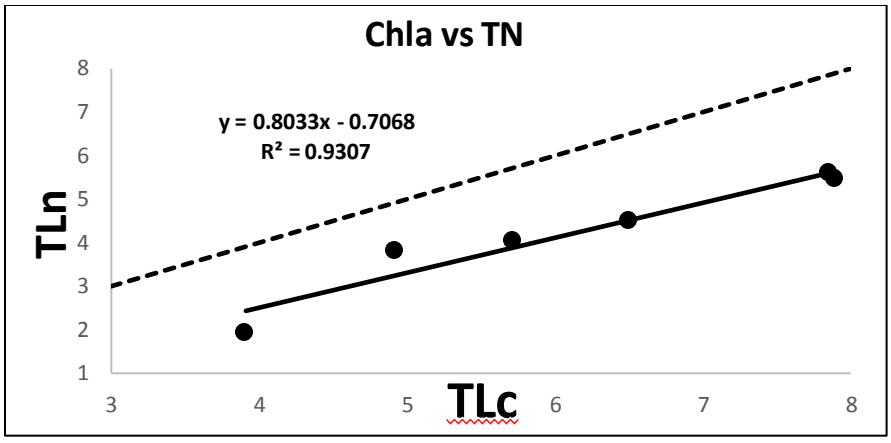
To determine how well the Ugandan crater lakes fitted the trophic categories developed in other locations, such as New Zealand, the inter-relationships between the TLI variables were further assessed by calculating individual trophic level sub-indices based on the New Zealand TLI system (Table 9). Taking an average of TLc, TLn, TLp and TLs, and applying the New Zealand TLI scoring system, the results characterise Lake Kyaninga as mesotrophic, Lakes Katanda and Nyinambuga as eutrophic, Lake Mwamba as supertrophic, and Lakes Saaka and Nyabikere as hypertrophic (Table 9). Sub-indices, however, frequently varied considerably from the TLI value, and this was further investigated using a regression analysis of relationships between TLc and other sub-indices (Figure 7).

Table 9: Trophic level sub-indices of chlorophyll-a, Secchi depth, total nitrogen and total phosphorus, and the trophic categories of the study lakes based on New Zealand TLI classification system.

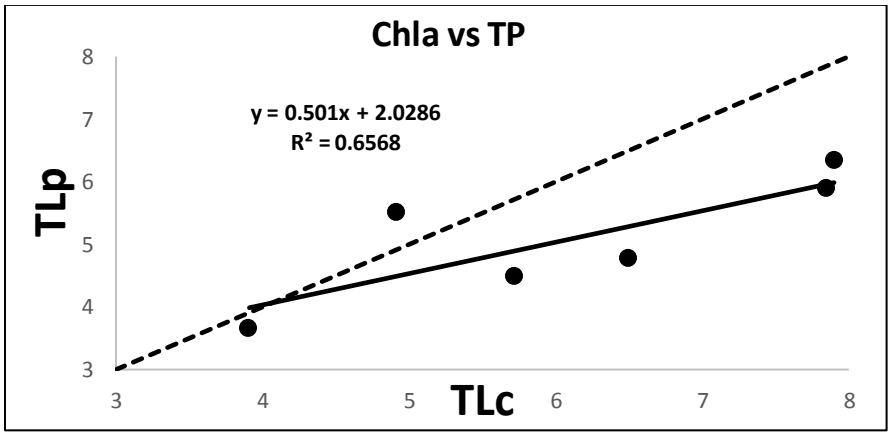
Lake	TLc	TLs	TLn	TLp	TLI	Standard Error	Category (overall)
Kyaninga	3.91	3.94	1.94	3.65	3.36	0.48	Mesotrophic
Katanda	4.92	5.48	3.80	5.49	4.92	0.40	Eutrophic
Nyinambuga	5.72	5.56	4.05	4.49	4.95	0.41	Eutrophic
Mwamba	6.50	5.66	4.49	4.76	5.35	0.46	Supertrophic
Nyabikere	7.90	6.59	5.45	6.34	6.57	0.51	Hypertrophic
Saaka	7.85	6.42	5.59	5.88	6.44	0.50	Hypertrophic



(a) TLc vs TLs



(b) TLc vs TLn



(c) TLc vs TLp

Figure 7: Scatter plots and linear regressions between the trophic level index of chlorophyll-a (TLc) and the trophic level indices of; **(a)** Secchi depth -TLs **(b)** total nitrogen-TLn and **(c)** total phosphorus-TLp.

Each scatter plot (Figure 7) shows two lines. The thick line shows the regression relationship between the trophic level indices among the crater lakes, while the broken line is a 1:1 linear fit indicating where the points "should" fall on the New Zealand TLI system. The TLc vs TLs curves (Figure 7a) show close similarities between Ugandan crater lakes and the New Zealand lake systems, implying Chl-a vs 1/SD relationship in the Ugandan crater lakes is consistent with the New Zealand trophic level index model. The linear relationship for TLc vs TLn among the crater lakes (Figure 7b) is significant, but it is different from that of New Zealand lakes and suggests that with less total nitrogen, Ugandan crater lakes produce more chlorophyll-a than New Zealand lake systems. Total nitrogen is a good proxy of chlorophyll-a in Ugandan crater lakes, but the relationship developed for the variables in New Zealand is not applicable. The TLc vs TLp curves (Figure 7c) shows two thirds of study crater lake produce more chlorophyll-a with less phosphorus than New Zealand systems but the correlation between the Chl-a and TP is less significant than that for nitrogen.

3.5 Identification of potential sources of nitrogen and phosphorus in the catchments

3.5.1 Agricultural farmland

Most of the crater lakes are located in rural areas and their catchments are occupied by communal settlements. Some crater lakes have tourism infrastructure, such as hotels and camp sites, large public institutions, such as schools, were seen in some lake catchments. The most significant observations in the wider part of the lake catchments, however, were the agricultural farms and communal settlements which exert pressure on the lakes riparian and catchment vegetation. For some of the crater lakes such as Lakes Saaka, Mwamba, and Nyabikere, the catchment riparian vegetation has been cleared up to the shoreline exposing the lakes to contaminants, sediment run-off, and leaching from neighbouring farms (Figure 8).

Figure 8: Deforested shoreline of Lake Mwamba in Kabarole district Uganda (Author's photo)

3.5.2 Waste management

Waste treatment systems were found at institutions such as hotels, lodges, and schools located in the study lakes' catchments. The establishment of some of the infrastructure close to the lakes is underpinned by commercial and recreational values provided by the waterbodies. Most of the institutions use conventional underground septic tanks with seepage overflows for the disposal of grey water and sewage. Some of the systems are located very close to the lake shores, which is increasing the potential for nutrients to leach into lakes. Particular cases were observed at Kyaninga Lodge close to Lake Kyaninga, Ndali Lodge close to Lake Nyinambuga, the Holy Cross Training institution and the Mountains of the Moon University both close to Lake Saaka, Nyabikere Safari Park and also the CVK camp site close to Lake Nyabikere. Some waste treatment systems have existed for more than 20 years and have therefore been leaching nutrients into the lakes for a long time.

Figure 9: A septic tank serving a Holly Cross Institution located right above the shore of Lake Saaka (Author's photo)

The crater lakes catchments were found to be widely inhabited by populations of rural communities without plans for human waste management. Every household in the catchment used unlined pit latrines for faecal waste disposal. Some latrines were found located close to the lakes or the lake tributaries. Lakes such as Saaka and Nyabikere had large catchment sizes with dense communal settlements.

3.5.3 Lake catchment size and riparian vegetation

The crater lakes catchments land forms have either gentle low land relief or steep slopes with varying catchment sizes. Lake Kyaninga has a relatively small catchment compared to Lake Saaka and the two catchments are morphometrically different. Lake Kyaninga is located on a hill top and has steep carter walls. In addition, Lake Kyaninga has a thick riparian forested vegetation around its steep slopes touching the lake shore. Lake Saaka is located in the middle of a gentle plain and the landscape from wide catchment slopes towards the lake or has a gently sloping peripheral regions (Melack, 1976). The riparian and catchment vegetation surrounding the lake has greatly diminished due to the expansion of agricultural farmland. With the exception of Lake Kyaninga, most of the crater lakes catchments in this study have lost vegetation due to agricultural and settlement activities.

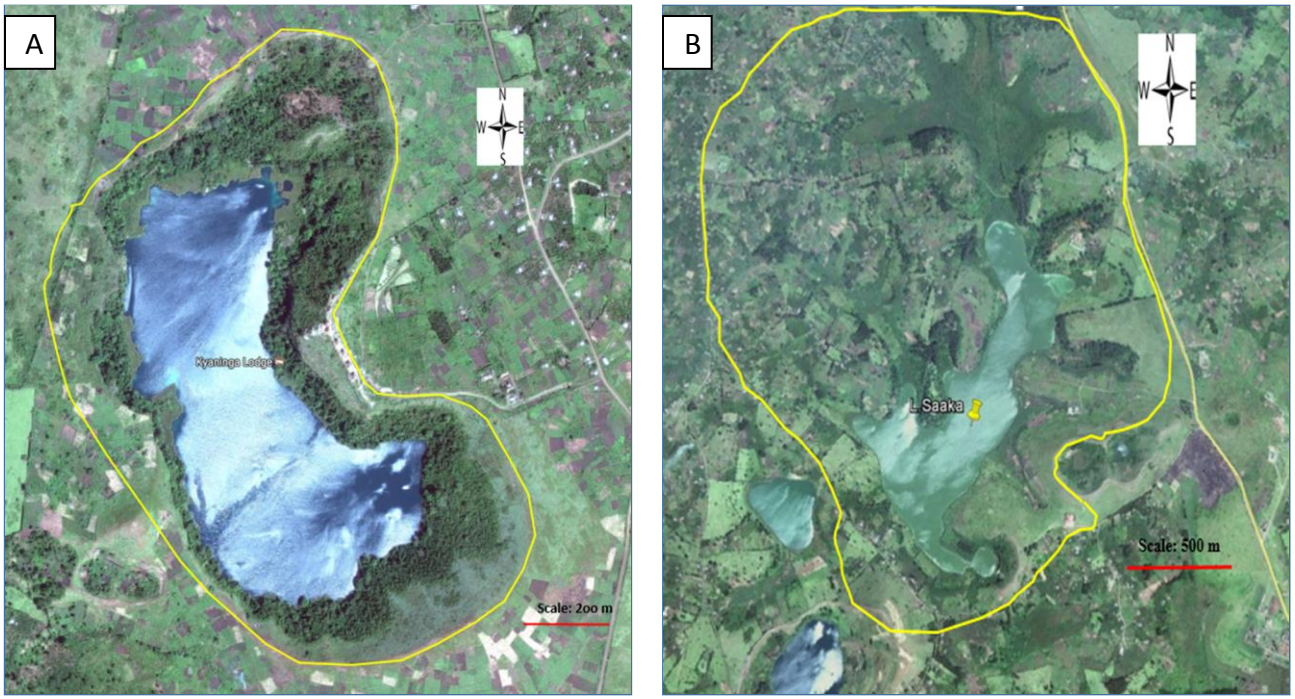


Figure 10: A google map showing the catchments and riparian zones for Lakes Kyaninga (A) and Saaka (B) (Google maps)

Of the six study lakes, only three have overland tributaries: Nyinambuga, Mwamba, and Nyabikere. Lakes Saaka, Kyaninga, and Katanda appear to be groundwater fed. Tributaries flow across agricultural farmlands and through communal settlements before discharging into the crater lakes. The water from tributaries is used for domestic purposes, canal irrigation and stock watering by the local communities. In addition to natural nutrient concentrations, human activities on the streams may influence the quality of water discharged into the crater lakes. To characterise nutrient concentrations in the streams, water samples were taken from each tributary and analysed for total phosphorus and total nitrogen concentrations (Table 10). A total of seven streams were identified and are named in reference to the lake into which they discharge the water.

Table 10: Total phosphorus and total nitrogen concentrations in the tributaries, and the TN:TP ratios expressed by mass.

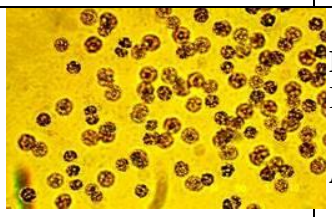
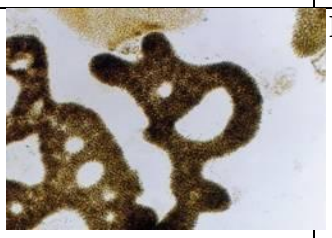
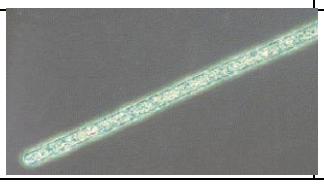
Stream	**TN (µg/l)**	**TP (µg/l)**	**TN:TP**
Nyinambuga Stream	1238.14	733	1.7
Mwamba Stream	992.59	300	3.3
Nyabikere Stream 1	148.49	868	0.2
Nyabikere Stream 2	438.93	398	1.1
Nyabikere Stream 3	443.32	336	1.3
Nyabikere Stream 4	574.05	359	1.6
Nyabikere Stream 5	523.49	366	1.4

Lake Nyabikere receives five surface tributaries from its catchment. All the inflows show significantly high nutrient concentrations. Nyinambuga stream shows the highest nutrient concentrations (Table 10). The rest of the streams have considerable amounts of nutrients. In every case TP concentrations were very high, always exceeding in-lake concentrations.

3.6 Cyanobacteria

Microscopic examinations of potentially toxic algae were conducted at 400x magnification using an inverted microscope (Appendix E). Algae were identified to genus by internal structures and morphological forms (Table 11). Most of the study lake samples had more than one type of potentially toxic algae. *Microcystis* and *Anabaena* species were common in the lake water samples. *Anabaena* was dominant or abundant in five of the six lakes, while *Microcystis* was in three. No potentially toxic cyanobacteria were observed in the water samples from Lake Kyaninga.

Table 11: The distribution of potentially toxic cyanobacteria in the five Ugandan crater lakes, based on optical microscopy at 400x. The photos used to illustrate microscopic appearance have been adopted from (Meriluoto & Codd, 2005) and (Newcombe, 2009).

Cyanobacteria genera	Internal cell structure	Distribution
Microcystis sp		**Dominant** in Lake Saaka and Nyabikere **Abundant** in Lake Nyinambuga
Microcystis sp		**Rare** in Lake Saaka
Cylindrospermopsis sp		**Abundant** in Lake Saaka **Occasional** in Lake Mwamba
Coiled Anabaena sp		**Dominant** in Lake Nyinambuga **Abundant** in Lake Mwamba **Occasional** in Lakes Saaka, Nyabikere, and Katanda
Anabaena sp		**Dominant** in Lake Mwamba **Abundant** in Lakes Saaka and Katanda
Planktothrix sp		**Abundant** in Lake Nyinambuga

Chapter 4 **DISCUSSION**

4.1 Introduction

The motivation behind my study was to assess the trophic states of crater lakes in Western Uganda using the traditionally accepted trophic state water quality variables. In addition, I set out to determine how well existing trophic indices, developed in other geographic regions, fit this set of lakes, by applying the New Zealand TLI system as an example (Burns et al., 1999). Preference was given to the New Zealand TLI system mainly because it includes total nitrogen in the trophic assessment, which many studies identify as the most common limiting nutrient for eutrophication and phytoplankton growth in tropical water ecosystems. Many New Zealand lakes are nitrogen limited, which makes them more closely related to tropical crater lakes of Western Uganda.

The evaluation was guided by the principles of TLI application, such as linear relationships between the water quality variables and the TLI sub-indices. The assessment was conducted in a series of stages; first, by testing the suitability of the New Zealand TLI scheme on Ugandan crater lakes where it was assumed that Ugandan crater lake systems behave similarly to New Zealand lakes. Second, the basic principles of the New Zealand TLI system were applied to confirm any systematic similarities and/or differences between the New Zealand lakes and Ugandan crater craters. Last, where systematic differences were found, efforts were made to assemble water quality data from previous studies on the crater lakes and to use that data to reformulate the New Zealand model in order to generate a new TLI scheme that specifically fits the Ugandan crater lakes. The new system could also be applied to estimate the trophic states of other freshwater bodies in Uganda.

4.2 The application of TLI system to Ugandan crater lakes

Direct application of the New Zealand TLI scheme developed by Burns and colleagues in 1999 classifies Lake Kyaninga as mesotrophic, Lakes Nyinambuga and Katanda as eutrophic, Lake Mwamba as supertrophic, and Lakes Saaka and Nyabikere as hypertrophic (Table 9) The TLI scheme provided a good separation of lakes according to trophic condition, but in evaluating the usefulness of the scheme on Ugandan lakes it is also useful to examine the correlations between the sub-indices and their associate variables, because these showed patterns dissimilar to New Zealand lakes.

4.3 Linear relationships among TLI variables

The linear relationships between log-transformed Chl-a and TP, TN, SD provided some useful suggestions concerning the correlations among the water quality parameters, which can be used to infer eutrophication processes and phytoplankton in the crater lakes. The linear regressions showed that SD and TN are good proxies for Chl-a, with the Pearson correlation coefficient of Chl-a vs SD and Chl-a vs TN being r^2=0.90 and r^2=0.93 respectively. TP showed a weak predictive relationship with Chl-a (r^2=0.66). That there is a stronger relationship between TN and Chl-a than that between TP and Chl-a suggests that nitrogen may be the nutrient limiting chlorophyll-a accumulation in Western Ugandan crater lakes, although further research would be required to confirm this. The limiting nutrient is determined by the relative availability of ambient nitrogen and phosphorus (Downing et al., 1999) and differential rates of nutrient loading, retention, and removal determine the ratio of these two key plant nutrients in lakes (Verburg et al., 2013). Crater lakes receive excess phosphorus flux from chemical weathering of their volcanic catchments rocks, a phenomenon that is believed to be particularly prominent at high temperatures (Lewis Jr, 1996) and the inflows to the crater lakes consistently contain high concentration of TP and low TN:TP ratios. High P loading may also be related to intensive clearing of catchment vegetation, which raises sediment transport and can increase the loading of particulate associated phosphorus into the lakes (Waters, 2016). The general observation is that in the tropics nitrogen limiting progressively becomes more severe with increasing catchment disturbance (Downing et al., 1999). The data I collected also conforms to this expectation.

Furthermore, it also useful to mention that, based on the good relationship between SD and Chl-a, phytoplankton appears to play a primary role in attenuating light in the crater lakes, which means that this low-cost and, technically simple technique could be used to estimate phytoplankton biomass in the crater lakes.

4.4 The Principles of TLI application

An assessment of the suitability of the New Zealand TLI system on Ugandan crater lakes was based on the relationships among the trophic level sub-indices of the variables. To be able to apply the New Zealand TLI system to any lakes outside New Zealand, the target lakes are required to satisfy the condition of similarity among calculated TLx values (TLp, TLn, TLs TLc) (Burns et al., 1999), as TLx values of each target lake should ideally lie within the same

trophic category. However, the TLx values derived in this study (Table 9) show a mismatch in all the study lakes which suggests systematic differences between New Zealand and the Western Ugandan crater lakes. In addition, there is a marked bias in the TLx values with respect to total nitrogen (TLn) which apparently are lower than the TLx values for TP, SD, and Chl-a. When the linear regression among TLx variables in the crater lakes is compared with the line fitted to New Zealand systems (Figure 7), more insight into the systematic similarities and differences between New Zealand lakes and Western Ugandan crater lakes is obtained. Based on TLx regressions, the relationship between Chl-a and SD in Ugandan crater lakes is compatible with the New Zealand model, implying similarity in the way that phytoplankton affects light penetration. However, the relationship between Chl-a and TN is strikingly different, such that for the same amount of nitrogen, Ugandan crater lakes appear to develop more chlorophyll-a than New Zealand lakes. This observation relates closely with the findings of Lewis Jr (1996) who noted that for a given nutrient concentration, the primary productivity in tropical lakes is twice as high as it would be for temperate lakes.

The observed systematic differences between the sets of lake systems suggests that the New Zealand TLI system may be unsuitable for assessing eutrophication of the crater lakes in Uganda, suggesting the need for development of a new TLI scheme that is appropriate and reliable, accounting for the systematic functioning of Ugandan crater lakes. The desired TLI system can easily be achieved by reformatting the New Zealand TLI model using water quality data from Ugandan crater lakes to account for specific interactions among water quality variables and how they influence eutrophication.

4.5 Developing a TLI system for crater lakes

To solve the mismatch observed in the New Zealand system, a new TLI system is proposed using data from the Ugandan crater lakes. To increase the applicability of the systems, I have accumulated a more comprehensive set of water quality data from Ugandan crater lakes, largely based on merging my data with a study conducted by Mills (2009) on the limnology, palaeolimnology, and palaeoenvironmental history of the crater lakes in Western Uganda. These data were used to reformulate the New Zealand TLI model based on the New Zealand and internationally recognised relationships between the TLI variables. The Mills (2009) dataset is comprised of measurements and analyses of surface water samples from 42 crater lakes during July-August 2006 and January-February 2007 (both dry seasons). In the interest of formatting the New Zealand TLI model, only crater lakes with data on the four TLI variables

(TP, TN, Chl-a, and SD) are considered. Thirty-six crater lakes had complete information on the required TLI variables (Table 12).

Table 12 : TP, TN, Chl-a and SD measurements from 36 crater lakes in Western Uganda from this study (bold entries) and that of (Mills, 2009).

Lake	SD(m)	TP (µg/l)	TN (µg/l)	Chl-a (µg/l)	Log(1/SD - 1/40)	LogTP	LogTN	Log (Chl-a)	TN:TP
Bugwagi	1.35	25	296	6.74	-0.15	1.39	2.47	0.83	12.0
Kako	2.93	5	86	1.2	-0.50	0.66	1.94	0.08	18.6
Kamunzuka	7.70	2	66	0.7	-0.98	0.23	1.82	-0.16	39.4
Kamweru	0.43	32	656	13.9	0.36	1.51	2.82	1.14	20.4
Kanyanmukali	0.94	188	444	7.2	0.02	2.27	2.65	0.85	2.4
Karolero	1.49	20	374	5.4	-0.19	1.31	2.57	0.73	18.2
Kasenda	1.75	18	662	3.0	-0.26	1.27	2.82	0.47	35.8
Katinda	0.35	19	768	25.4	0.45	1.29	2.89	1.41	39.7
Kifuruka	1.15	35	497	8.6	-0.07	1.55	2.70	0.94	14.1
Kigezi	1.35	16	283	6.6	-0.15	1.21	2.45	0.82	17.3
Kikorongo	1.60	1372	11914	4.0	-0.22	3.14	4.08	0.60	8.7
Kitagata	0.05	43	1821	25.5	1.30	1.63	3.26	1.41	42.6
Kyaninga	6.60	5	104	0.8	-0.90	0.73	2.02	-0.09	19.5
Kyasanduka2	1.30	211	893	203.0	-0.13	2.32	2.95	2.31	4.2
Kyasanduka3	0.18	211	893	203.0	0.76	2.32	2.95	2.31	4.2
Kyerbwato2	0.64	11	228	1.7	0.19	1.05	2.36	0.24	20.1
Kyogo	2.90	273	343	2.0	-0.50	2.44	2.54	0.31	1.3
Lugembe	0.78	21	529	5.7	0.10	1.33	2.72	0.76	24.7
Lyantonde	2.80	5	258	2.3	-0.48	0.74	2.41	0.36	47.3
Mahuhura	6.60	50	86	1.2	-0.90	1.70	1.94	0.07	1.7
Mirambi	1.00	18	633	11.1	-0.01	1.26	2.80	1.04	35.2
Muijongo	2.70	7	331	2.0	-0.46	0.82	2.52	0.31	50.3
Murabyo	2.03	10	322	0.0	-0.33	0.99	2.51	#NUM!	32.9
Murusi	1.40	5	309	1.3	-0.16	0.71	2.49	0.12	60.7
Njarayabana	0.75	360	498	3.8	0.12	2.56	2.70	0.57	1.4
Nkuruba2	1.49	25	659	5.9	-0.19	1.40	2.82	0.77	26.3
Nyamogusingiri	0.50	30	1536	14.8	0.30	1.47	3.19	1.17	51.9
Nyamirima	4.40	4	245	2.4	-0.69	0.61	2.39	0.37	60.3
Nyamogusani	1.20	152	285	3.5	-0.09	2.18	2.46	0.54	1.9
Nyamswiga	1.80	33	391	4.0	-0.28	1.52	2.59	0.60	11.7
Nyierya	1.35	16	470	14.0	-0.15	1.21	2.67	1.15	29.2
Nyinambulita	4.28	26	166	1.8	-0.68	1.41	2.22	0.25	6.4
Nyungu	0.47	119	1498	57.3	0.33	2.08	3.18	1.76	12.6
Saaka2	0.60	37	894	32.5	0.22	1.56	2.95	1.51	24.5
Wandakara2	0.31	26	752	9.1	0.51	1.41	2.88	0.96	29.0
Wankenzi	0.36	24	835	11.4	0.44	1.39	2.92	1.05	34.1
Kyaninga	**3.00**	**15**	**70**	**4.6**	**-0.51**	**1.18**	**1.85**	**0.6656**	**3.9**
Katanda	**0.67**	**64**	**290**	**11.5**	**0.17**	**1.81**	**2.46**	**1.0611**	**4.9**
Nyinambuga	**0.62**	**29**	**350**	**23.9**	**0.20**	**1.46**	**2.54**	**1.3788**	**5.7**
Mwamba	**0.56**	**36**	**490**	**48.4**	**0.25**	**1.56**	**2.69**	**1.6849**	**6.5**
Saaka	**0.22**	**125**	**1020**	**171.6**	**0.66**	**2.10**	**3.01**	**2.2345**	**7.9**
Nyabikere	**0.26**	**87**	**1140**	**164.19**	**0.58**	**1.94**	**3.06**	**2.2153**	**7.8**

Linear regressions of log-transformed values of Chl-a vs those of TP, TN and SD revealed correlations that were statistically significant based on the *p* value < *0.05*. (Table 13)

Table 13: Results of least square linear regressions between log-transformed values of Chl-a and TN,TP and SD.

Relationship	**R^2**	***p* value**
Log (Chl-a) vs Log TP	0.34	0.005
Log (Chl-a) vs Log TN	0.57	5.2×10^{-5}
Log (Chl-a) vs Log (1/SD – I/40)	0.57	1.1×10^{-6}

To reassess the trophic states and algal productivity in the crater lakes, a set of TLx equations were generated using scatter plots and linear regression models of TLc vs Log TP, Log TN and Log (1/SD-1/40). Chl-a is used as the primary descriptor of eutrophication. TLc values of 3, 4 and 6 are assigned to corresponding Chl-a concentrations of 2, 5 and 30 µg/l (Vant, 1987), which are internationally recognised thresholds between oligotrophic, mesotrophic, and eutrophic trophic categorisations. These numbers and TLc values were selected by Burns et al. (1999), while developing the New Zealand TLI system, to ensure a linear relationship between the desired trophic number and log-transformed chlorophyll-a concentration (Figure 11). The resulting linear regression equation (Eqn 4.1) can then be used to calculate the TLc of the crater lakes.

TLc = 2.2248 + 2.5537Log(Chl-a)……..Eqn 4.1

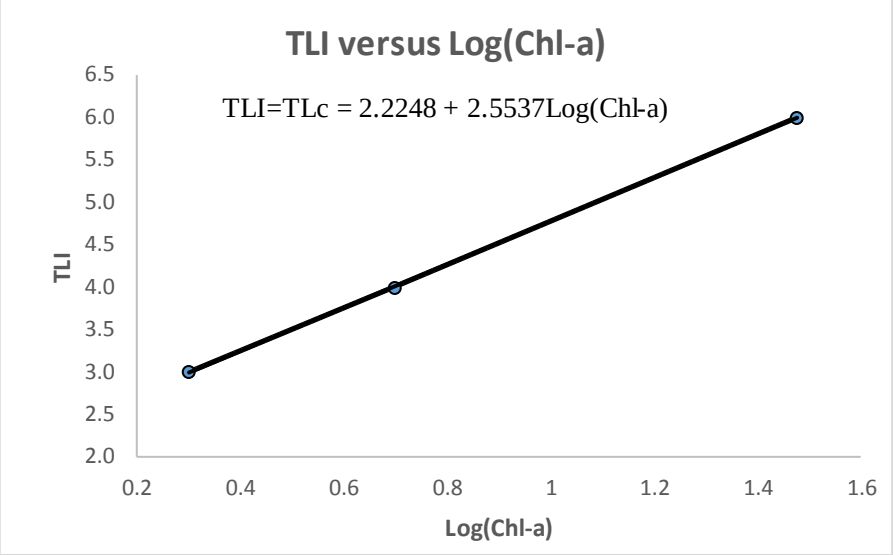
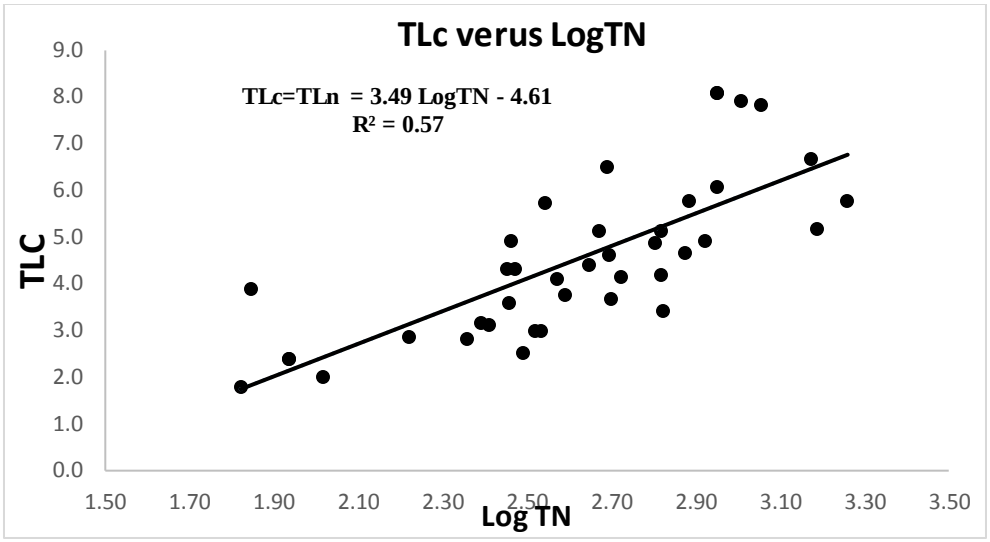
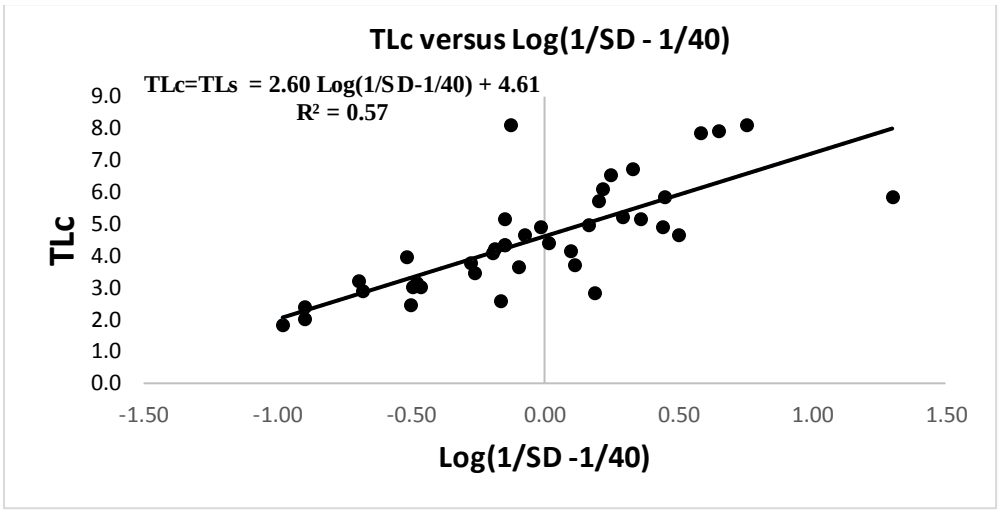


Figure 11: Chlorophyll-a values plotted against the proposed general TLI scale for New Zealand (Burns et al., 1999).

After calculating the TLc values (Eqn 4.1) from Chl-a concentrations of crater lakes (Table 12), scatter plots and linear regression models are plotted between TLc values and the corresponding log-transformed values of TP, TN and (1/SD – 1/40) (Figure 12). The resulting linear equations (Eqn 4.2 to 4.4) can be used to calculate the trophic level sub-indices of study lakes with respect to total phosphorus (TLp), total nitrogen (TLn) and Secchi depth (TLs) (Table 14)





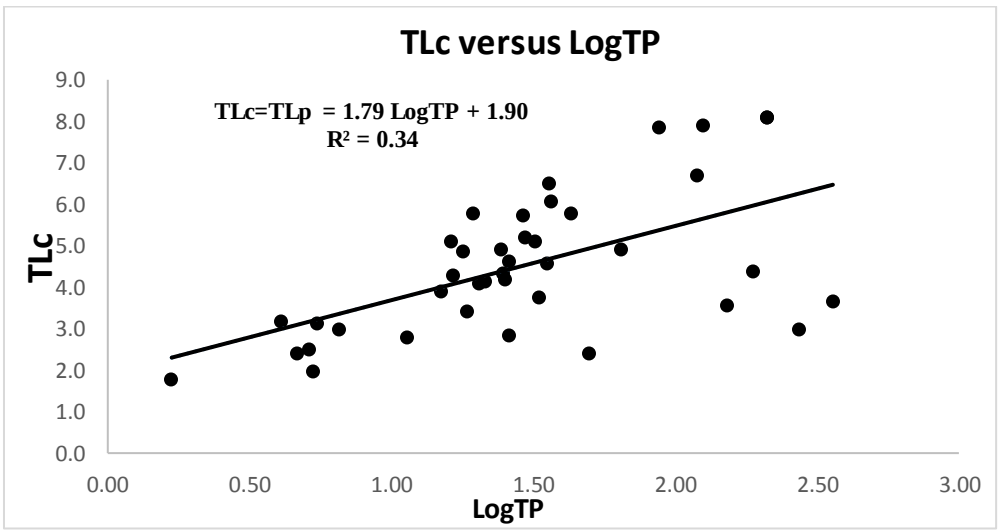


Figure 12: Regression models of the trophic level index with respect to Chl-a (TLc) against the log-transformed values of TP, TN, and (1/SD - 1/40)

The four linear regression equations represent the new TLI system considered suitable for assessing eutrophication and algal productivity in Ugandan crater lakes. It can be used to calculate the average trophic level index of each study lake with respect to Chl-a TP, TN, and SD (Table 14).

$$TLs = 4.61 + 2.60 \log (1/SD - 1/40) \quad \text{Eqn 4.2}$$

$$TLp = 1.90 + 1.79 \log(TP) \quad \text{Eqn 4.3}$$

$$TLn = -4.61 + 3.49 \log(TN) \quad \text{Eqn 4.4}$$

Reassessing the similarity among the TLx values (Table 14) based on the new TLI system showed an improvement in the agreement of calculated sub-indices, and hence of accuracy in trophic state estimation based on multiple variables. Comparing the standard error of calculated sub-indices under the New Zealand TLI system (Table 9) with that of the Ugandan TLI system (Table 14), there is an improvement in the similarity of the sub-indices for Lakes Katanda, Nyinambuga and Mwamba while the rest did not show any improvement. Even so, the new system may not be definitive due to limited confidence in the data from a one-off sampling and the potential non-representativeness of samples. Despite these limitations, the predictions are likely to account for the systematic functioning of crater lakes.

Table 14: Trophic level indices for Chl-a, TN, TP,SD, and avearge TLI value for the study lakes based on reformated equations using water quality data from previous studies on Ugandan crater lakes.

Lake	TLc	TLs	TLn	TLp	TLI	Standard Error	Category (NZ)
Kyaninga	3.9	3.3	1.8	4.0	3.3	0.51	Mesotrophic
Katanda	4.9	5.0	4.0	5.1	4.8	0.25	Eutrophic
Nyinambuga	5.7	5.1	4.3	4.5	4.9	0.32	Eutrophic
Mwamba	6.5	5.3	4.8	4.7	5.3	0.41	Supertrophic
Saaka	7.9	6.3	5.9	5.6	6.4	0.51	Hypertrophic
Nyabikere	7.8	6.1	6.1	5.4	6.3	0.51	Hypertrophic

The new TLI system can also be tested by determining the trophic state of Lake Kyaninga using data from Cocquyt et al. (2010) for three consecutive years: August 2007, August 2008, and

April 2009 (Table 15). Disregarding the missing values of TN in 2007 and TP in 2009 the resulting TLI values for 2007, 2008, and 2009 are 4.0, 3.9, and 2.6 respectively. The results show that, based on the New Zealand trophic categories, Lake Kyaninga was mesotrophic during 2007 and 2008 and shifted to oligotrophic in 2009. It should be noted that in Western Uganda August is dry and April is a rainy season. Considering assessment results from 2016 (Table 14), Lake Kyaninga could be predominantly mesotrophic but can occasionally shift to oligotrophic during the wet season probably due to dilution through precipitation and reduced chlorophyll synthesis due to limited solar insolation. The consistency of the mesotrophic state of Lake Kyaninga in August of 2007, 2008, and 2016 gives confidence about the reliability of the new TLI system for predicting average trophic states of crater lakes. While some seasonality may be evident, there are generally less marked seasonal changes in dissolved nutrients and biomass in tropical lake systems than in temperate ones (Viner, 1977), and the frequent sampling may be less problematic in such conditions.

Cocquyt et al. (2010) reported Lake Kyaninga to be predominantly oligotrophic with brief shifts to mesotrophic after deep mixing. Their findings were based on the Carlson (1977) trophic scheme but this may be considered unsuitable due to non-consideration of TN and, it is suitable for temperate lake systems that are mostly productive during summer and early fall.

Table 15: TP, TN, SD and Chl-a values of Lake Kyaninga and corresponding trophic sub-indices and TLI during 2007, 2008 and 2009.

Lake	Chl-a	SD	TN	TP	TLc	TLs	TLn	TLp	TLI
Kya 2007	5.24	4.7	-	70	4.1	2.7	-	5.2	4.0
Kya 2008	9.27	3.9	270	20	4.7	3.0	3.9	4.2	3.9
Kya 2009	3.49	6.05	70	-	3.6	2.4	1.8	-	2.6

4.6 The non-compatibility of New Zealand TLI system on Ugandan lakes

Systematic differences between the New Zealand and Ugandan crater lakes systems, are illustrated by the relationships between the TLI variables and can be used to explain why the New Zealand TLI system is unsuitable for Ugandan crater lakes. As discussed above, the Chl-a vs TN relationship revealed a lower Chl-a:TN ratio in Ugandan crater lakes compared to New Zealand lakes because the Ugandan lakes produce more Chl-a for less nitrogen than New Zealand lakes. The crater lakes in Uganda are subject to a very different seasonality compared to New Zealand lakes. The Western Ugandan crater lakes straddle the equator from latitude 0°42 N to 0°19 S (Melack, 1978) and because of this they receive high insolation making them

attain relatively high annual water temperatures and small seasonal temperature changes due to long periods of solar irradiation, high air and water temperatures most of the year. These conditions are favourable for increased chlorophyll-a synthesis. In contrast, the New Zealand lakes lie between latitudes 35° S and 48° S (Alla et al., 2016) and experience extreme conditions of winter with reduced periods of solar insolation, and hence lower chlorophyll-a synthesis. Despite many New Zealand lakes being nitrogen limited, similar to what is observed in the crater lakes in Western Uganda, experiments of nitrogen assimilation in cultures of green algae (Bongers, 1956) indicated that at high light intensities, such as the conditions around the equator, nitrogen assimilation by phytoplankton cells increases leading to increased photosynthesis, consequently increased chlorophyll production. New Zealand lakes lie at high altitude receive relatively less light intensity leading to low photosynthesis.

Ugandan crater lakes also display a weak Chl-a vs TP relationship. As discussed above, this is likely to reflect the fact that TP is not a limiting nutrient. Volcanic soils are rich in phosphorus and crater lakes receive a hydrological washout of P loads from their catchments (Talling, 1986). N:P ratios by weight of 13.6:1, the highest seen in this study, are equivalent to 6:1 by moles, also suggests that crater lakes are more likely to be nitrogen limited than phosphorus limited (Ansari et al., 2011; Mills, 2009)

4.7 The variation of trophic states among crater lakes

Based on standard Chl-a criteria, the crater lakes vary from mesotrophic to hypertrophic state, suggesting the mesotrophic state may be lowest trophic level attained among crater lakes. Given the presence of N fixing cyanobacteria in the mesotrophic lakes, this mesotrophic nature may be due to high natural phosphorus fluxes from catchments maintaining a high P concentration in the lakes; a high nitrogen budget enhanced by the composition of N fixing cyanobacteria in the crater lakes; and a tendency of high Chl-a synthesis, prevalent in the equatorial region.

The factors that takes crater lakes above mesotrophic can be attributed to those that regulate external and internal nutrient processes and other biophysical factors that specifically affect the development of phytoplankton. One of the variable features of crater lakes that may contribute to trophic state is the catchment size and land use. Considering hydrological nutrient inputs, the nutrient status of run-off water and surface tributaries is highly variable and differs depending upon the catchment type (Talling, 1986). The highly eutrophic Lakes Saaka and Nyabikere are characterised by large catchments with intensive farming activities mixed with

dense human settlements. Cultural eutrophication around Lake Saaka was first noticed in the early 1970s based on measurements of conductivity, SD, and supersaturation of dissolved oxygen in the surface waters which indicated elevated algal productivity (Crisman, Chapman, Chapman, & Prenger, 2002). Campbell et al. (2006) further noted that over the past half century Lake Saaka has been severely affected by surrounding agriculture leading to significant eutrophication. For example a government prison farm started operations in the mid-1950s along the catchment of Lake Saaka. During the expansion of the prison farm significant deforestation reduced the riparian vegetation, exacerbating sediment run-off and nutrient load into the crater lake. The large catchment sizes around Lakes Saaka and Nyabikere are highly modified due to an increased demand for farmland expansion and human settlements in the fertile alluvial catchment soils. Crater lakes with small catchments and less anthropogenic disturbance appear to have better water quality. An example is Lake Kyaninga which has a small and least disturbed catchment with lowest trophic state, albeit mesotrophic. The nature of catchment morphometry also influences human habitation. Bootsma and Hecky (1993) demonstrated that unlike the steep cliff shorelines that are unattractive for human habitation, the lowland relief of the nearshore catchments of lakes are characterised by human habitations and high population densities. Bootsma and Hecky (1993) further noted that although low relief may result into reduced erosion and run-off potential, these are countered by the close proximity of human settlement and anthropogenic activities on the lake, which are particularly evident in crater lakes with low topography (Figure 8 and Figure 9).

There is a potential association between water quality and crater lakes morphometry. Lake Kyaninga has a steep, forested cliff above the water surface which buffers the lake from excess sediments and nutrients in the run-off. Melack (1978) noted that volcanic crater lakes in Western Uganda are maar type lakes with morphometric features such as water depth, steep cliffs and height of crater rim above the water surface. The morphometric variations influence lake stratification, mixing, and internal nutrient loading processes, which may also regulate nutrient cycling and phytoplankton. Among the study lakes, Lake Saaka is the shallowest (7.8 m) followed by Lake Nyabikere (57 m). These two lakes are open to sweeping wind currents because of the gentle slopping topography of their lateral catchments, and low land relief of the nearshore environment. Winds create turbulence and mixing within the water column and raise bottom nutrients which become bioavailable for supporting further algal growth. Meanwhile Lake Kyaninga, the deepest known maar crater lake in Western Uganda, has a very steep and forested nearshore topography (Cocquyt et al., 2010) sheltering it from wind currents

and hence preventing lake overturn and maintaining a calm steady state. The permanent stratification of Lake Kyaninga below 100 m (Cocquyt et al., 2010) may lead to nutrient loss from the mixed layers due to sinking of organic matter (Bootsma & Hecky, 1993). Melack (1978) suggests that shallow crater lakes such as Lake Saaka are likely to support more productive aquatic systems than deep lakes such Lake Katanda due to regeneration and mixing of nutrients associated with shallow lake systems.

Western Uganda crater lakes with tributaries accumulate nutrients loads via these well-defined inflows, according to whether the flow is constant or seasonal. The hypertrophic state of Lake Nyabikere is partly attributed to excessive nutrient loading by five tributaries that run across its busy catchment. The high nutrient concentrations in tributaries (Table 10) indicate a significant influence from surface tributaries on crater lake water quality, but also show the potential nutrient generation by catchments. Bootsma and Hecky (1993) observed that high nutrient concentrations in surface flow/run-off signify intense catchment disturbance through land clearance and cultivation which subsequently released the nutrients. Closed crater lake systems may only be subject to nutrient loading through run-off during the rainy season, and to an extent through ground water flow. The presence of riparian margins in closed lake systems, such the forests on the slopes of Lake Kyaninga, can buffer excessive sediment and nutrients carried by run-off and prevent or reduce eutrophication processes. Similarly Lake Nkuruba, a closed crater lake in Kasenda area, is reported to be mesotrophic which is partly linked to the surrounding intact forest along its steep slopes (Campbell et al., 2006).

4.8 The potential nutrient sources

Understanding the relative importance of different nutrient sources is fundamental for inferring the impacts of disturbances on a lake (Bootsma & Hecky, 1993). Generally, agriculture farms occupy the largest proportion of crater lake catchments in Western Uganda and consequently substantial amounts of nutrients are generated from agricultural wastes and applied fertilisers. Fertilisers are commonly used on the tea plantations in the Kasenda area (Vonesh, 2001). Beyond the influence from natural nutrient sources, the amount of nutrient flux from crater lake catchments also is depends on the type and intensity of anthropogenic disturbance (Chapman & Chapman, 2003). The common pathways through which external nutrients enter lake ecosystems include groundwater, fluvial or atmospheric inputs (Smith, Tilman, & Nekola, 1999). Despite this, Bootsma and Hecky (2003) noted that for the smaller tropical lakes, nutrient concentrations are largely regulated by hydrologic inputs from the littoral catchments.

Deforestation through slash and burn agriculture, commonly practised by communities to clear agricultural land around the crater lakes, enhances nutrient pulses through all pathways; surface flow, groundwater, and atmospheric deposition. Downing et al. (1999) noted that the practice of burning releases substantial organically bound nutrient stocks into the soil, transferring about 96% of N and more than 40% P into the atmosphere which can later become deposited into regional lake ecosystems. Despite the fact that P is not abundant in a volatile form due to its close association with particulate matter, its increased atmospheric concentration is enhanced by higher atmospheric particulate load generated from burning activities and elevated wind erosion resultant from deforestation and land tillage (Bootsma & Hecky, 1993).

In addition, nutrients are generated from localised sewage treatment systems such as underground septic tanks and unlined pit latrines that are common at institutions and households around crater lake catchments. Because rural areas are not connected by centralised sewerage treatment systems, every household or institution uses a private system for excreta treatment leading to high density of excreta disposal sites. Although no specific study regarding sewage-related nutrient release has been conducted in rural areas, studies around dense communities in Kampala city indicate that substantial amounts of nutrient rich wastewater originating from unlined pit latrines and septic tanks enters aquifers (Nyenje, Foppen, Kulabako, Muwanga, & Uhlenbrook, 2013). Around the lake catchments, nutrient plumes from such contamination join ground water which consequently feeds into tributaries and the crater lakes. As an example of this, an underground septic tank adjacent to Lake Saaka (Figure 9) located at the Catholic institution, has been in operation since the 1970s. The long period of usage is believed to have indirectly contributed nutrients into Lake Saaka partly contributing to its hypertrophic state. Similarly other septic tanks close to Lakes Nyabikere and Kyaninga have the potential to influence lake water quality depending on the period of usage and volumes of excreta generated. The system on Lake Kyaninga was established in the early 2000s. Despite the lake showing a mesotrophic state, continuous use of the septic tank potentially risks contributing to the deterioration of the lake if no sound monitoring and management plan is put in place.

Bootsma and Hecky (1993) listed riverine and tributary inputs as another key nutrient source. Rural communities in the crater lake region depend on surface streams for a numerous uses including domestic water supply, watering stock, and direct discharge of grey water. These uses generally leave a nutrient foot print in the tributaries. For this reason, the severity of

nutrient concentrations observed in the tributaries is a composition from multiple sources that indirectly influence the nutrient budgets.

4.9 The phytoplankton composition in the crater lakes

Although my investigations of phytoplankton composition were restricted to single samples identified only to genus level, observations of the dominance of cyanobacteria agree with other studies from this region. Okello and Kurmayer (2011) identified seven cyanobacteria genera including *Anabaena, Microcystis, Planktolyngbya, Planktothrix, Cylindrospermopsis, Chroococcus* and *Aphanocapsa* in five Ugandan lakes including Lake Saaka, which were dominant and regularly contributed more than 50% of the phytoplankton biovolume in the lakes. In addition, Poste et al. (2013) observed that during periods of optimum growth, the cyanobacteria community composition in Ugandan lakes can make up >80% of the total phytoplankton biovolume. Generally, the algal seasonality in African lakes is influenced by considerable changes in radiant energy incomes, water temperature, and oftentimes the lake water input (Talling, 1986). In this study four cyanobacteria genera were identified, *Microcystis, Planktothrix, Cylindrospermopsis* and *Anabaena,* and all of them have been previously identified in Ugandan lakes. *Microcystis, Planktothrix* and *Anabaena* have all been identified as microcystin producing genera (Chorus & Bartram, 1999; Okello & Kurmayer, 2011). Microcystin toxins are of great concern to public health due to their potential effects they pose to human health, animals, and other aquatic organisms. Regarding ecosystem functioning, *Cylindrospermopsis* and *Anabaena* have the potential to fix atmospheric nitrogen which is believed to be the main supply of nitrogen for phytoplankton growth among the crater lakes.

Microcystis and *Anabaena* are dominant and abundant among crater lakes (Table 11) which implies some crater lake conditions may be more favourable for some cyanobacterial species than others. For instance, the genus *Microcystis* is dominant in hypertrophic lakes while *Anabaena* is relatively dominant in eutrophic and mesotrophic lakes. This distribution pattern may also signify a succession among cyanobacteria species along the trophic enrichment gradient. Poste et al. (2013) reveal that high total TP concentrations, lower TN: TP ratios and lower CO_2 availability are some of the conditions that favour an increase in cyanobacteria biomass, in particular *Microcystis,* in Ugandan lakes. Poste et al. (2013) also noted that the above conditions existed in shallow lakes such as Lake Saaka, and the shallow nearshore sites of deeper lakes such as Lake Victoria. Shallow and eutrophic sites provide ideal conditions for

the proliferation of *Microcystis* cyanobacteria biomass (Okello & Kurmayer, 2011), explaining the dominancy of *Microcystis* observed in Lakes Saaka and Nyabikere given their shallow depth and highly nutritious state. It should also be noted that uncontrolled eutrophication among of crater lakes risks intense colonisation of toxic algae across all crater lakes, including the ones that are still mesotrophic such as Lake Kyaninga, which will further deprive the region of the values from these useful natural resources.

Chapter 5 CONCLUSION AND RECOMMENDATIONS

5.1.1 Conclusions

Trophic evaluation of the volcanic crater lakes in Western Uganda can readily be achieved by assessing four commonly accepted TLI variables: TN, TP, Chl-a, and SD. The connection between the individual parameters and their influence on eutrophication processes conform to established relationships between Chl-a and nutrients and water transparency. Despite adhering to the traditional relationships, the trophic assessment in the crater lakes cannot be accurately estimated using TLI systems developed in other countries such as New Zealand because of the underlying systematic ecosystem differences between the lakes from the different regions. The differences can be explored through calculating trophic level sub-indices, and through regression analysis of relationships of TLI variables and trophic sub-indices.

The analysis conducted within this project has shown than volcanic crater lakes in Western Uganda are nitrogen limited; they receive excess P flux from the volcanic catchments leading to high ambient P concentration in the surface waters. The phytoplankton in crater lakes synthesise Chl-a for a smaller amount of nitrogen compared to New Zealand lakes, and the community phytoplankton composition in Ugandan crater lakes includes nitrogen fixing cyanobacterial genus such as *Anabaena* and *Cylindrospermopsis*. Because of high concentrations of P and high Chl-a synthesis, Ugandan crater lakes attain higher trophic levels than New Zealand lakes and cannot be evaluated using the New Zealand TLI system. However, reformulating the New Zealand TLI model with data from Ugandan crater lakes generated a new TLI system that accounted for systematic ecosystem functioning and was able to increase the accuracy of trophic estimation among study crater lakes.

Based on the New Zealand trophic categories, the TLI assessment shows that Ugandan crater lakes even in their pristine state are not oligotrophic and, mesotrophic state is likely to be the lowest trophic status that can be attained due to the aforementioned reasons of high ambient P and high Chl-a concentrations. This may also imply that TLI estimations of previous studies based on systems developed in other countries may have risked either underestimation or overestimation of trophic status in Ugandan crater lakes.

Eutrophication of crater lakes in Western Uganda is highly attributed to the influence of anthropogenic catchment disturbance. The catchments of the crater lakes have been severely modified through agricultural expansion and increased human settlement activities increasing

potential of nutrient loading. Crater lakes such as Lakes Saaka and Nyabikere, with extensive and prolonged catchment disturbance, are highly degraded. The eutrophication process also resulted into the colonisation of cyanobacteria including toxin producing species such as *Microcystis* and *Planktothrix*, which threaten ecological sustainability and freshwater usage. But, crater lakes that are considered to be of good water quality such as Lake Kyaninga are still of great ecological value to multiple stakeholders. It is essential that comprehensive aquatic conservation procedures are adopted to sustain lakes of good water quality and restore the water quality of degraded lakes.

5.1.2 Recommendations

It is important to recognise the limitations of this study and the effect they may have had undermining trophic state estimations. Six crater lakes were sampled of the many crater lakes in Western Uganda with variations in biogeochemical characteristics. The small number of study lakes may obscure the broad limnologic characterisation of water quality among volcanic lakes. Second, this study sampled surface waters, assuming a well-mixed water column where there may be variations in depth and the extent of stratification within the crater lakes. Lastly, one-off sampling may have obscured temporal variations in the TLI variables and how they are influenced by annual climatic changes. To minimise the effect of annual climatic changes, sampling was conducted in the dry season when growth of algal biomass was expected to be maximal within the small seasonal range expected for near-equatorial systems. An expectation based on results from previous studies, for example, previous research on Lake Kyaninga showed small variations in TLI parameters measured throughout the dry seasons of three consecutive years.

Despite these limitations, the results reveal useful findings about the trophic state and ecosystem functioning of the volcanic crater lakes in Western Uganda. These findings will provide a baseline for long-term, detailed studies on eutrophication in freshwater bodies in Uganda. Currently, there is no national strategy for lake management in Uganda, but these simple techniques of trophic state monitoring can provide a foundation for understanding lake management. For this reason, adopting such a trophic level indicator approach nationwide in Uganda would serve as a monitoring tool to help develop sustainable nutrient management strategies in lake catchments.

The trophic level index system formulated in this study for monitoring crater lakes in Western Uganda still requires improvements and experimentation, for example, removing limitations

through collecting a primary dataset and conducting long-term depth integrated sampling of a more representative number of volcanic crater lakes. If a nationwide lake management strategy is developed along the lines suggested it should be possible to preserve the water quality of lakes than have not undergone eutrophication, and crucially improve the degraded lakes for them to remain a valuable resource to all stakeholders.

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Appendices

Appendix A: Total nitrogen calibrations

Table A1: Concentrations and absorbance of nitrate calibration curve

Sample	Standard nitrate Concentration	Spectral Absorbance
		DR6000 5cm cell
B1	0	0.015
B2	0	0.015
B3	0	0.016
B4	0	0.014
B5	0	0.015
2	2	2.974
1	1	1.741
0.75	0.75	1.435
0.5	0.5	0.929
0.25	0.25	0.453
0.1	0.1	0.197

Table A2: Concentrations and absorbance for EDTA calibration curve

Samples		Conc. (mg/l)	Volume of sample (Wt3)	Volume of digested sample (Wt5)	Final Vol pH adjusted sample	Absorbance	Absorbance corrected
						5cm Cell	5cm cell
Blanks	B1	0	21.8012	21.5443	22.2405	0.021	0.021
	B2	0	21.8621	21.6988	22.2852	0.032	0.033
	B3	0	21.8186	21.6386	22.1564	0.029	0.029
	B4	0	21.8551	21.593	22.0726	0.027	0.027
	B5	0	21.8015	21.644	22.0532	0.027	0.027
EDTA Standards	0.05a	0.05	21.9671	21.8176	22.404	0.097	0.099
	0.05b	0.05	21.9821	21.7036	22.1808	0.096	0.097
	0.05c	0.05	21.9326	21.5989	22.1173	0.091	0.092
	0.1	0.1	21.9597	21.9039	22.2463	0.153	0.155
	0.25	0.25	21.736	21.4245	22.0103	0.308	0.312
	0.5	0.5	21.9066	21.6859	22.0963	0.689	0.695
	0.7	0.7	21.7434	21.525	22.1836	0.92	0.939
	0.75	0.75	21.7432	21.4859	22.1451	0.889	0.905
	1	1	21.8612	21.7164	22.2688	1.268	1.292
Digested nitrate standards	0.25	0.25	21.5496	21.4619	22.0119	0.341	0.348
	0.1	0.1	21.7891	21.7231	22.1693	0.149	0.152
Undigested nitrate standards	0.25	0.25				0.459	
	0.1	0.1				0.187	

Appendix B: Total Phosphorus calibrations

Table B3: Concentrations and absorbance for a Dissolved Reactive Phosphorus (DRP) calibration curve

	Sample	Concentration (mg/l)	Absorbance	
			5cm cell	2cm cell
Blanks	B1	0	0.002	0.001
	B2	0	0.002	0.001
	B3	0	0.002	0
	B4	0	0.002	0.001
	B5	0	0.002	0.001
PO4-P Standards	0.005	0.005	0.012	0.005
	0.005	0.005	0.011	0.006
	0.005	0.005	0.011	0.005
	0.01	0.01	0.022	0.01
	0.025	0.025	0.062	0.029
	0.05	0.05	0.128	0.06
	0.1	0.1	0.255	0.12
	0.25	0.25	0.678	0.325
	0.5	0.5	1.432	0.683
	0.7	0.7	2.017	0.961
	1	1	2.891	1.381

Table B4: Concentrations and absorbance for organic phosphorus (Inositol) calibration curve

	Sample	Concentration (mg/l)	Absorbance		1st Absorbance correction		2nd Absorbance correction (minus blank absorbance)	
			5cm cell	2cm cell	5cm cell	2cm cell	5cm cell	2cm cell
Blanks	B1	0	0.024	0.011	0.024	0.011	-0.001	-0.001
	B2	0	0.025	0.012	0.025	0.012	0	0
	B3	0	0.026	0.013	0.026	0.013	0.001	0.001
	B4	0	0.026	0.013	0.026	0.013	0.001	0.001
Organic P Standards (Inositol)	0.005	0.005	0.036	0.017	0.036	0.017	0.011	0.005
	0.01	0.01	0.044	0.021	0.044	0.021	0.019	0.009
	0.025	0.025	0.069	0.033	0.068	0.033	0.043	0.021
	0.05	0.05	0.109	0.051	0.108	0.05	0.083	0.038
	0.1	0.1	0.193	0.091	0.191	0.09	0.166	0.078
	0.25	0.25	0.446	0.211	0.44	0.208	0.415	0.196
	0.5	0.5	0.901	0.427	0.892	0.423	0.867	0.41
	0.7	0.7	1.266	0.6	1.25	0.593	1.225	0.58
	1	1	1.82	0.865	1.798	0.855	1.773	0.843
Organic P (Glucose 6-PO4)	0.1	0.1	0.219	0.104	0.216	0.103	0.191	0.091
	0.5	0.5	1.036	0.492	1.011	0.48	0.986	0.468
PO4-P	0.1	0.1	0.248	0.118	0.245	0.116	0.22	0.104
	0.5	0.5	1.219	0.572	1.203	0.565	1.178	0.553

Table B5: Concentrations and absorbance for potassium hydrogen phosphate (PO4-P) calibration curve

	Sample	Concentration (mg/l)	Absorbance		Absorbance corrected	
			5cm cell	2cm cell	5cm cell	2cm cell
Blanks	B1	0	0.003	0.001	0.003	0.001
	B2	0	0.003	0.001	0.003	0.001
	B3	0	0.002	0	0.002	0
	B4	0	0.003	0.001	0.003	0.001
	B5	0	0.003	0.001	0.003	0.001
PO4-P Standards	0.005	0.005	0.012	0.005	0.012	0.005
	0.01	0.01	0.021	0.01	0.021	0.01
	0.025	0.025	0.052	0.024	0.051	0.024
	0.05	0.05	0.106	0.051	0.104	0.05
	0.1	0.1	0.219	0.103	0.215	0.101
	0.25	0.25	0.603	0.287	0.592	0.282
	0.5	0.5	1.212	0.577	1.189	0.566
	0.7	0.7	1.714	0.81	1.672	0.79
	1	1	2.397	1.143	2.352	1.121
Organic P	0.5	0.5	1.016	0.484	0.999	0.476
	0.7	0.7	1.547	0.737	1.525	0.726

Appendix C: Total nitrogen analysis of water samples

Table C6: Total nitrogen analytical data for crater lakes water samples

Sample	Dilution factor	Absorbance	Absorbance corrected	Corrected Absorbance less blank absorbance	Concentration (mg/l)	Total conc. (mg/l)	Final Average conc. (mg/l)
		5cm cell					5cm
B1	1	0.028	0.028	-0.004	-0.03	-0.03	
B2	1	0.035	0.035	0.003	-0.02	-0.02	
B1	1	0.036	0.037	0.001	-0.02	-0.02	
B2	1	0.035	0.035	-0.001	-0.02	-0.02	
Saaka Inshore	5	0.308	0.315	0.283	0.2	1.01	1.02
Saaka Inshore	5	0.343	0.352	0.32	0.23	1.16	
Saaka Offshore	5	0.333	0.337	0.305	0.22	1.1	
Saaka Offshore	5	0.254	0.26	0.228	0.16	0.79	
Kyaninga Inshore	5	0.066	0.067	0.035	0	0.02	0.07
Kyaninga Inshore	5	0.068	0.069	0.037	0.01	0.03	
Kyaninga Offshore	2	0.138	0.138	0.106	0.06	0.12	
Kyaninga Offshore	2	0.111	0.113	0.081	0.04	0.08	
Katanda inshore	2	0.263	0.262	0.226	0.16	0.32	0.29
Katanda inshore	2	0.246	0.246	0.21	0.14	0.29	
Katanda offshore	2	0.201	0.2	0.164	0.11	0.22	
Katanda offshore	2	0.259	0.258	0.222	0.15	0.31	
Mwamba inshore	5	0.163	0.163	0.127	0.08	0.39	0.48
Mwamba inshore	5	0.208	0.21	0.174	0.12	0.58	
Mwamba offshore	5	0.172	0.174	0.138	0.09	0.44	
Mwamba offshore	5	0.189	0.193	0.157	0.1	0.51	
Nyabikere inshore	5	0.345	0.352	0.327	0.24	1.19	1.14
Nyabikere inshore	5	0.327	0.332	0.307	0.22	1.11	
Nyabikere offshore	5	0.332	0.337	0.312	0.23	1.13	
Nyabikere offshore	5	0.331	0.334	0.309	0.22	1.12	
Nyinambuga inshore	5	0.151	0.152	0.127	0.08	0.39	0.35
Nyinambuga inshore	5	0.134	0.136	0.111	0.07	0.33	
Nyinambuga offshore	5	0.146	0.149	0.124	0.08	0.38	
Nyinambuga offshore	5	0.135	0.134	0.109	0.06	0.32	
0.1 EDTA	1	0.159	0.161	0.129	0.08	0.08	
0.1 EDTA	1	0.16	0.162	0.137	0.09	0.09	
0.5 EDTA	1	0.61	0.62	0.595	0.45	0.45	
0.5 EDTA	1	0.529	0.538	0.506	0.38	0.38	

Table C7: Total nitrogen analytical data for samples from lake tributaries

Stream / tributary sample analysis data							
Sample	**Dilution factor**	**Absorbance**	**Absorbance corrected**	**Corrected Absorbance less blank absorbance**	**Concentration (mg/l)**	**Total conc. (mg/l)**	**Final Average conc. (mg/l)**
B1	1	0.048	0.048	-0.001	-0.02	-0.02	
B2	1	0.044	0.045	-0.004	-0.03	-0.03	
Nyinambuga Stream	5	0.37	0.375	0.326	0.24	1.19	1.23
Nyinambuga Stream	5	0.396	0.398	0.349	0.26	1.28	
Mwamba Stream	2	0.769	0.773	0.724	0.55	1.11	0.99
Mwamba Stream	2	0.626	0.626	0.577	0.44	0.88	
Nyabikere Stream 1	20	0.084	0.085	0.036	0.01	0.12	0.12
Nyabikere Stream 1	20	0.084	0.085	0.036	0.01	0.11	
Nyabikere Stream 2	2	0.324	0.323	0.274	0.2	0.39	0.43
Nyabikere Stream 2	2	0.376	0.376	0.327	0.24	0.48	
Nyabikere Stream 3	5	0.199	0.2	0.151	0.1	0.49	0.44
Nyabikere Stream 3	5	0.177	0.177	0.128	0.08	0.4	
Nyabikere Stream 4	2	0.485	0.483	0.434	0.32	0.65	0.57
Nyabikere Stream 4	2	0.386	0.382	0.333	0.24	0.49	
Nyabikere Stream 5	5	0.215	0.215	0.166	0.11	0.55	0.52
Nyabikere Stream 5	5	0.2	0.202	0.153	0.1	0.5	
0.25 EDTA	1	0.394	0.399	0.35	0.26	0.26	
0.25 EDTA	1	0.396	0.398	0.349	0.26	0.26	
0.5 EDTA	1	0.765	0.769	0.72	0.55	0.55	
0.5 EDTA	1	0.748	0.749	0.7	0.54	0.54	

Appendix D: Total phosphorus analysis of water samples

Table D8: Total phosphorus analytical data for crater lakes water samples

Sample	Dilution factor	Absorbance 5cm cell	Absorbance corrected	Corrected Absorbance less blank absorbance	Total conc. (mg/l)	Final Average conc. (mg/l)
B1	1	0.009	0	0.009	0.002	0
B2	1	0.01	0	0.01	0.003	
B3	1	0.006	0.006	0	0.003	
B4	1	0.006	0.006	0	0.003	
Nyabikere Inshore	1	0.221	0.003	0.216	0.088	0.087
Nyabikere Inshore	1	0.21	0.002	0.207	0.085	
Nyabikere Offshore	1	0.219	0.006	0.215	0.086	
Nyabikere Offshore	1	0.215	0.002	0.212	0.087	
Katanda Inshore	1	0.171	0.004	0.176	0.071	0.064
Katanda Inshore	1	0.26	0.005	0.256	0.104	
Katanda Offshore	1	0.145	0.004	0.143	0.057	
Katanda Offshore	1	0.165	0.002	0.161	0.065	
Mwamba Inshore	1	0.103	0.007	0.102	0.039	0.036
Mwamba Inshore	1	0.113	0.012	0.111	0.04	
Mwamba Offshore	1	0.088	0.01	0.087	0.031	
Mwamba Offshore	1	0.09	0.007	0.089	0.033	
Saaka Inshore	1	0.295	0.29	0.277	0.119	0.125
Saaka Inshore	1	0.305	0.301	0.289	0.124	
Saaka Offshore	1	0.311	0.305	0.292	0.126	
Saaka Offshore	1	0.319	0.313	0.301	0.13	
Kyaninga Inshore	1	0.036	0.035	0.027	0.014	0.015
Kyaninga Inshore	1	0.036	0.035	0.029	0.015	
Kyaninga Offshore	1	0.068	0.067	0.056	0.026	
Kyaninga Offshore	1	0.034	0.033	0.027	0.014	
Nyinambuga Inshore	1	0.066	0.065	0.055	0.026	0.029
Nyinambuga Inshore	1	0.064	0.063	0.053	0.025	
Nyinambuga Offshore	1	0.05	0.049	0.039	0.019	
Nyinambuga Offshore	1	0.07	0.069	0.059	0.028	
0.1 Org-P	1	0.162	0.16	0.154	0.068	
0.5 Org-P	1	0.858	0.845	0.839	0.356	

Table D9: Total phosphorus analysis data for lake tributary water samples

	Dilution factor	Absorbance	Sample Blank absorbance	Absorbance corrected	Absorbance less sample absorbance	Concentration (mg/l)	Total Conc. (mg/l)	Average conc. (mg/m3)
		5cm cell						
B1		0.002	0.000	0.002	0.000	0.00	0.00	
B2		0.002	0.000	0.002	0.000	0.00	0.00	
B3		0.002	0.000	0.002	0.000	0.00	0.00	
Nyinambuga Stream	2	0.934		0.013	0.922	0.899	0.382	0.733
Nyinambuga Stream	2	0.863		0.014	0.852	0.828	0.351	
Mwamba Stream 1	2	0.378	0.007	0.372	0.363	0.16	0.31	0.300
Mwamba Stream 1	2	0.351	0.006	0.344	0.336	0.14	0.29	
Nyabikere Stream 1	5	0.412	0.000	0.407	0.405	0.17	0.87	0.868
Nyabikere Stream 1	5	0.414	0.000	0.408	0.406	0.17	0.87	
Nyabikere Stream 2	2	0.469	0.002	0.463	0.459	0.20	0.39	0.398
Nyabikere Stream 2	2	0.486	0.003	0.479	0.474	0.20	0.40	
Nyabikere Stream 3	2	0.404	0.000	0.395	0.393	0.17	0.34	0.336
Nyabikere Stream 3	2	0.401	0.000	0.392	0.390	0.17	0.33	
Nyabikere Stream 4	2	0.443	0.009	0.433	0.422	0.18	0.36	0.359
Nyabikere Stream 4	2	0.444	0.013	0.432	0.417	0.18	0.36	
Nyabikere Stream 5	5	0.174	0.001	0.171	0.168	0.07	0.37	0.366
Nyabikere Stream 5	5	0.174	0.001	0.170	0.167	0.07	0.37	
0.1 Org-P	1	0.158	0.000	0.154	0.152	0.07	0.07	
0.5 Org-P	1	0.870	0.000	0.853	0.851	0.36	0.36	
0.7 Org-P	1	1.249	0.000	1.216	1.214	0.51	0.51	

Appendix E: Photos of microscopic observation of cyanobacteria in a water sample from Lake Saaka (Authors photo)