EFFECT OF MAGNETIC FLUX DENSITY ON *ESCHERICHIA COLI, SALMONELLA* SPECIES AND CHEMICAL PROPERTIES OF RIVER NJORO WATER

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EGERTON UNIVERSITY

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DECLARATION AND RECOMMENDATION

DECLARATION

I declare that this thesis is my original work and has not been submitted wholly or in part to any institution for award of any degree to the best of my knowledge.

Thirika Anne Mwende

SM13/3117/11

Signature……Date……

RECOMMENDATION

We confirm that this research thesis has been done under our supervision and has our approval to be presented for examination as per the Egerton University regulations.

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DEDICATION

To my beloved parents Mr. and Mrs. Albert Thirika for making my dream a reality

ABSTRACT

The disinfection of water is of prime importance both for the animals and humans, since the presence of microorganisms in water may cause diseases including hepatitis, cholera, typhoid fever and dysentery. Reagents, techniques based on the use of a broad range of chemical agents are commonly used to disinfect water. Among them are chlorine, fluorine, ozone and ions of heavy metals. However, in the case where some organic substances remain in water, chlorine and ozone reacts with them forming carcinogenic substances. Moreover, many reagents are toxic and necessitate safety precautions in their transit and dosage. Some of them when applied result in variations in the composition and properties of water and adversely affect animal and plant life. For this reason, reagent techniques cannot fully meet modern requirements on the disinfection of water. The reagent less techniques are of great interest and may include physical methods based on the use of various physical fields for disinfection. Among them are ultraviolet rays, constant electric fields and magnetic fields. An important advantage of these physical techniques is their ability to act directly on microorganisms, leaving the properties and composition of water virtually intact. In this study experimental results of the disinfection process of water by means of magnetic fields are presented. Water samples were collected from River Njoro. Microbial count (*Salmonella* species *and E. coli*) for the samples were obtained using Membrane Filtration techniques. Spectrophotometry procedure was carried out to determine the concentration of Ca^{2+} and Mg^{2+} ions in the water. The pH value of the water was also taken using a pH meter. The samples were then exposed to different magnetic flux densities (2mT, 6mT and 10mT) at time intervals of 6 hours and 18 hours for each magnetic flux. Membrane filtration, Spectrophotometry and pH measurements were done before and after magnetic treatment of the samples. The data obtained was photographed and presented in tables and bar graphs. It was then subjected to One-way ANOVA, regression and correlation analyses. The means were separated using the Least Significant Difference. The maximum disinfection efficiency was 82.2% for *E. coli* and 77% for *Salmonella* species. It was also found that treatment with magnetic field did not alter the concentration of Ca^{2+} and Mg^{2+} ions as well as the pH values of River Njoro water. Magnetic treatment can possibly be used as a cost-effective method of disinfecting water for domestic consumption to reduce likely incidences of waterborne diseases.

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LIST OF ABBREVIATIONS AND ACRONYMS

AAS	Atomic Absorption Spectrophotometry
ANOVA	Analysis of Variance
APHA	American Public Health Association
CFU	Colony Forming Units
ELF-EMF	Extremely Low Frequency Electromagnetic Field
DBPs	Disinfection By-Products
GPS	Global Positioning System
LSD	Least Significant Difference
MFD	Magnetic Flux Density
MFT	Membrane Filtration Technique
SPSS	Statistical Package for Social Sciences
THM	Tri Halo Methane
UNDP	United Nations Development Programme
USEPA	United States Environmental Protection Agency
WHO	World Health Organization
WRMA	Water Resource Management Authority

CHAPTER ONE

INTRODUCTION

1.1 Background Information

Safe drinking water has been a concern for mankind all over the world for centuries. According to the United Nations Development programme (UNDP), 780 million people lack access to clean drinking water that is approximately one in nine people in any population sample (UNDP, 2010). Moreover, World Health Organization (WHO) reported that over 41% of Kenyans do not have access to clean water and rely directly on rivers as the main source of water (WHO, 2011). Many rivers are reportedly prone to high bacterial contamination due to heightened ecological activities and may therefore be unsuitable for human consumption when untreated (WWAP, 2006).

Study has indicated that the microbial quality of the River Njoro water sources is poor and unacceptable for human consumption due to consistent increase in total and fecal coliforms and also due to pathogenic loading downstream (Kiruki *et al*., 2011). This water is contaminated with pathogens which lead to widespread acute and chronic illnesses which are major cause of death and misery. A recent report holds that about 3.41 million people die each year from water, sanitation and hygiene-related causes (WHO, 2012). The water and sanitation crisis claims more lives through disease than any war claims through guns (WHO, 2011).

One of the key cornerstones of public health and community health is access to safe water however water purification is an expensive process and the government has not provided majority of the population with treated water despite the requirement of the millennium development goals. Moreover, most common water purification methods have been shown to have serious negative impact to health and the environment. Chlorination for instance which is one of the most widely used disinfectants (Vesilind *et al*., 1990) produce chemical compounds known as disinfection by-products (DBPs). Of these, tri halo methanes (THMs) and halo acetic acids (HAAs) found in the highest concentrations in chlorine treated drinking water are linked to high risks of cancer (USEPA, 2003).

There has been growing concern over the health effects of boiling water as a purification method. This is because boiling tends to concentrates harmful inorganic contaminants, hence reducing water potability (Rubenowitz-Lundin and Hiscock, 2005). Again in developing countries, the major source of energy for cooking and boiling water is firewood which is expensive and environmentally unfriendly.

It has been realized that the advantages of using water exposed to magnetic field as compared to normal water are enormous. Magnetic waste water treatment is a process that has been introduced to chemical industries to remove heavy metals and organic pollutants from waste water (Tsouris, 2001; Tomska and Wolny, 2007). Stimulation of both animal and plant growth with magnetic field as a way to increase the quality and quantity of yield has caught the interest of many scientists worldwide (Chastokotenko, 1984; Magnetic technologies, 2004). For instance, it has been established that magnetic treatment of irrigation water can improve primary productivity of water resulting to an increased crop production, a decrease in plant disease rate and an improved taste of agricultural products (Lin and Yotvat, 1990; Duarte *et al*., 1997). It is not clearly known how this is achieved but it could be attributed to the ability of such water to retain and highly dissolve the essential minerals (Johan *et al*., 2004) a position that this study sought to verify.

In latest years, several studies have been performed to verify direct effects exerted by extremely low-frequency (ELF) electromagnetic fields (EMFs) on cell functions. In particular, it has been demonstrated that ELF-EMF can negatively (Kronenberg *et al*., 1985; Lin and Yotvat, 1990; Balcavage *et al*., 1996; Molouk and Amna, 2010) or positively (Smothers *et al.*, 2001; Szkatula *et al*., 2002) affect functional parameters (cell growth and viability) of a cell. The effects of magnetic fields on cells are thus not fully understood since some of the results have been inconsistent and have in other cases contradicted each other. The aim of this study was thus to evaluate the effect of magnetic flux density on the microbiological and chemical properties of water. This study was also aimed at finding the optimum magnetic condition that would eliminate bacterial content in River Njoro water.

1.2 Statement of the Problem

Kenya is a water scarce country and the challenge of availability of clean water has been known to exist for many years. Water purification is an expensive process and the government has not provided majority of the population with treated water. Moreover, reagent techniques based on the use of a broad range of chemical agents are commonly used to disinfect water. Many reagents are toxic and necessitate safety precautions in their transit and dosage. Some of the reagents when applied result in variations in the composition and properties of water and adversely affect animal and plant life. For these reasons, reagent techniques cannot fully meet modern requirements of the millennium development goals in the disinfection of water. This study therefore intended to evaluate the capacity of an applied magnetic flux to disinfect water by directly acting on microorganisms and leaving the chemical composition of water virtually intact.

1.3 Objectives

1.3.1 General objective

To determine the effect of different magnetic flux densities on *Escherichia coli*, *Salmonella species* and chemical properties of River Njoro water

1.3.2 Specific objectives

1. To determine the effect of magnetic flux density on the population of *Escherichia coli* in River Njoro water.
2. To determine the effect of magnetic flux density on the population of *Salmonella species* in River Njoro water.
3. To determine the effect of magnetic flux on the concentration of Ca^{2+}, Mg^{2+} ions and pH of River Njoro water.

1.4 Hypotheses of the study

1 Magnetic flux density does not affect the population of *Escherichia coli* in River Njoro water.

2 Magnetic flux density does not affect the population of *Salmonella species* in River Njoro water.

3 Magnetic flux does not affect the concentration of Ca^{2+}, Mg^{2+} ions and pH of River Njoro water.

1.5 Justification

The challenge of availability of clean water for domestic use has been known to exist for centuries. The WHO estimates that about 2 million lives can be saved per year if there was an adequate and sustainable supply of safe drinking water. This study elucidated an efficient method for water purification that is not harmful to human health and environment. The magnetic flux method is also very convenient in terms of ease in application and can therefore be used locally at household level. As such, the findings of this study carry potential for a definitive and conclusive remedy to one of mankind's oldest problem which is lack of safe and healthy drinking water.

CHAPTER TWO

LITERATURE REVIEW

2.1 Water and Health

Water is vital to life and so the type and quality of the water taken is important. Drinking water can contribute to variable fractions of the total daily requirement of essential minerals which include magnesium, calcium, sodium, potassium, and phosphorus, and their necessary daily requirement is over 100 mg of each (WHO, 1996). Food is the principal source of calcium and magnesium, people consuming refined food may suffer deficiency of these essential minerals, and thus even the relatively low intake of essential elements through drinking water may play an essential role in human health. It has been reported that elements that are present as free ions in water are more readily absorbed from water than from food bound to other substances (Sunderman *et al*., 1989)**.**

Water pollution is regarded as one of the most critical environmental problems with the major categories of pollutants being microbiological, chemical and radioactive elements (WHO, 2004). Throughout history, polluted water is often the main human exposure pathway to infectious pathogens that lead to waterborne disease outbreaks with acute and long-term health effects ranging from diarrhoea, cholera to death (Conroy *et al*., 2001). Emerging contaminants, which have not historically been considered pollutants, have now raised significant concerns to public health professionals, environmental engineers and scientists as these contaminants damage major organs and functional system (Brewer, 2010; Stoev, 2010). As analytical methods continue to improve, recent studies have revealed that emerging or re-emerging microbiological pathogens or chemicals may be present in natural or treated water bodies (WHO, 2004; Kiruki *et al*., 2006; Muia *et al*., 2006).

Chemical, i.e., reagent, techniques based on the use of a broad range of chemical agents are commonly used to disinfect water. Among them are chlorine and chlorine-containing compounds (chlorine gas chlorinated lime, sodium hypocloride, and active chlorine, produced in special electrolyzers made of table salt, chloramines, and others) as well as ozone, fluorine, iodine, ions of heavy metals (including silver), and a number of other substances (WHO, 2012). Chlorination

has gained the most acceptance among the reagent techniques to date. The main reasons for this are the high reliability of the bactericidal effects, the possibility of rather simple monitoring of the residual chlorine, the simplicity of the corresponding equipment, and others. However, in the case where some organic substances remain in water, chlorine reacts with them forming carcinogenic substances. Among the latter are chloramines that induce the cancer (USEPA, 2003). Because of this, drinking water should be carefully purified from organic compounds prior to chlorination.

A similar drawback is also inherent in disinfection processes based on ozone treatment. So-called ozonides are formed on interaction of ozone with some organic compounds. These ozonides also belong to the category of carcinogenic substances. Moreover, many reagents are toxic and necessitate safety precautions in their transit and dosage. Some of them when applied result in variations in the composition and properties of water and adversely affect animal and plant life. For this reason, reagent techniques cannot fully meet modern requirements on the disinfection of water.

Helping people gain access to safe drinking water is one of the most important health-related infrastructure programs in the world hence in the new millennium, there is a growing concern for the need to balance the risks of the need to disinfect the water to reduce the threat of disease from microorganisms against the potential health risks from disinfection byproducts that are formed as a result of adding a disinfectant. The reagent less techniques are therefore of great interest. These physical methods are based on the use of various physical fields for disinfection. Among them are ultraviolet rays, electric discharges in water, cavitation, ultrasound, and so on. An important advantage of physical techniques is their ability to act directly on microorganisms, leaving the properties and composition of water virtually intact. This study therefore intended to address this dilemma by establishing a water purification strategy that circumvents the health risks associated with the classical purification procedures.

2.2 Magnetic flux and Water Properties

It is well-known that water is formed of clusters and the more stable cluster numbers are called magic numbers. The sequence of magic numbers carries essential information about the electronic and ionic structure of the cluster and consequently the water properties (Joshi and Kamat, 1966). If it is possible to modify the magic numbers, many new alternative uses of water

can be found, because the importance of the magic numbers are related to intrinsic properties of water (Kronenberg, 1985). Studies have found that various aspects of water structure changes when exposed to a magnetic field (Coey and Cass, 2000; Amiri and Dadkhan, 2006; Smothers *et al*., 2001; Tai *et al*., 2008). Magnetic field causes physical-chemical changes of natural water parameters, resulting in improvement of filtration and an increase in dissolving properties of water (Moon and Chung, 2000; Smirnov, 2003). An increase of water viscosity under the influence of magnetic field has been explained on the basis of stronger hydrogen bonds (Maheshwari and Grewal, 2009). Magnetic technology is a treatment process that can enhance the separation of suspended particles from the sewage (Johan *et al*. 2004).

Studies have shown that use of irrigation water that has been exposed to a magnetic field recorded higher crop yields. For instance, it has been established that magnetic treatment of irrigation water can improve primary productivity of water (Lin and Yotvat, 1990; Duarte *et al*., 1997). Stimulation of plant growth and development with magnetic field as a way to increase the quality and quantity of yield has caught the interest of many scientists worldwide (Chastokotenko, 1984). Some studies have shown that there is an increase in growth and yield parameters of tomatoes, strawberry and broad beans by the application of magnetic fields (Danilov *et al*., 1994; Podleoeny *et al*., 2004; Esitken and Turan, 2004). It is not clearly known how this is achieved but it could be attributed to the ability of such water to retain the essential minerals, a position that this study sought to verify.

2.3 Bio effects of magnetic field

The exposure of living tissue to various types of electric and magnetic fields is a commonly encountered event (Blum and Roller, 1982). Since AC and DC magnetic fields are most likely to penetrate the body, they are the components of electromagnetic fields that are usually studied in relation to the effects on living systems (Balcavage *et al*., 1996). It has been realized that magnetic gradients induce forces which affect biological materials (Smirnov, 2003). This has been attributed to biological materials having small magnetic susceptibility regardless of whether they are diamagnetic or paramagnetic (Magnetic Technologies, 2004).

The phenomenon of effective penetration of the atmospheric oxygen into the solutions treated with a magnetic field is advantageous from one point of view. Most microorganisms breaking down the organic compounds are aerobic by nature (Abel, 2002). Thus, in the magnetically

treated water, with increased oxygen concentration, their growth is more intense and so, is the degradation of organic matter responsible and essential for bacterial metabolism (Molouk and Amna, 2012). This in turn leads to decrease in length of bacterial cell, decrease in cellular thickening, and disappearance of most elements from the cytoplasm and eventually inability of the cells to divide (Mohamed *et al*., 1997). Additionally, water forms 80% of bacterial cells, so when it's physical and chemical properties are changed by magnetic force, the growth of bacterial cells are inhibited as their composition changes. This is in accordance with (Strasak *et al*., 2002).

Microorganisms are single-celled organisms surrounded by a phospholipid membrane. The Purpose of the membrane is two-fold. First, it physically contains the cell's organelles and the other cellular machinery (proteins) needed for survival. Second, it maintains separation between the intracellular and extracellular salt solutions in which the cell exists (Macfaddin, 2000). This separation of the ions across the bacterial cell wall and the maintenance of the impermeable phospholipid membrane are essential for cell life (Neill, 2004). The bacteria cell membrane contains protein channels that transport different ions across the membrane to control both electrical and chemical potential that exists across. When microorganisms are subjected to a strong magnetic flux density, the ability of the protein channels to maintain the electrical and chemical potential across the cell's membrane is greatly affected. In brief, the membrane is drastically torn apart and the microorganism is destroyed (He *et al*., 2009).

Electromagnetism increases the competency of *E.coli* hence plasmids may become more antibiotic resistant when treated to electromagnetism (Neill, 2004). Static magnetic field causes inhibition of cell division in *E.coli* and inability to form colonies on solid media (EL-Sayed *et al*., 2006). Some other studies found that magnetic fields could be a general stress factor in bacteria (Molouk and Amna, 2010). The general stress response to a magnetic field is found in all bacteria, plant and animal cells and is remarkably conserved across species. In a study on the mutagenicity of magnetic fields exposure also reported that strong static magnetic fields can cause mutations in *S. typhimurium* and *E.coli* (Mohamed *et al*., 1997). There is great inhibition in the growth and concentration of *Pseudomonas aeruginosa* bacteria when exposed to magnetic field using wireless magneto-elastic, which facilities the sterilization process specially in canned

food (Pengfei *et al*., 2007). Exposing *Serratia marcescens* to magnetic flux leads to the inhibition of its growth (Piatti *et al*., 2002).

The question of how microbiological and chemical parameters of water are affected by magnetic field is not completely answered. This study was therefore intended to throw more light on as to whether magnetic flux density affects these parameters.

2.4 Origin of magnetic flux density

Magnetic field is defined as the area around a magnet where the magnets pull and repulsion effect is felt (Chaby and Sherwood, 1995). The field is stronger near the poles of the magnet and weaker further away from the poles The existence of magnetic field is shown diagrammatically by use of imaginary lines called magnetic field lines. A group of field lines around a magnet constitutes magnetic flux. To understand the meaning of magnetic flux (Φ) and magnetic flux density (B) think first about an ordinary bar magnet. Around the magnet there is a magnetic field and this gives a 'flow of magnetic energy' around the magnet. It is this flow of energy that we call magnetic flux (Φ). Magnetic flux flows from the north pole of a magnet round to its south pole as shown by the arrows on the lines in figure 1. Looking at the diagram it is seen that there is as much flux flowing 'from the north pole' as there is 'flowing into the south pole'. Magnetic flux is given the symbol Φ and is measured in units called Webers (Blum and Roller, 1982).

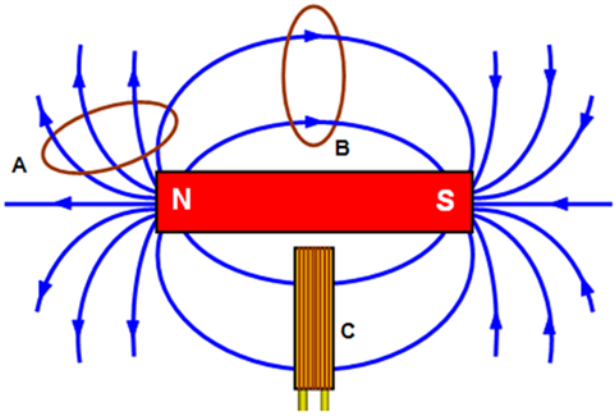

Figure 1: Magnetic field lines due to a bar magnet

However, the amount of magnetic flux flowing through a given area will change from one point to another around the magnet. This can be understood by thinking about a loop of wire placed in the field at two different points (A and B). It is seen that in position B there are a smaller number

of magnetic field lines passing through the loop than there is when it is in position A. The amount of flux passing through a unit area at right angles to the magnetic field lines is the flux density (B) at that point. Flux density is measured in Tesla (T) where 1 T = 1 Wbm-2. Magnetic flux density (B) is therefore defined as the force acting per unit current per unit length on a wire placed at right angles to the magnetic field. (Duffin, 1990).

2.5 Electromagnetism

Electromagnetism relates to the magnetic field generated around a conductor when current is passed through it. Every orbiting electron in motion around its nucleus generates a magnetic field. When electrons are forced to leave their parent atom by voltage and flow towards the positive polarity, they are all moving in the same direction and each electron's magnetic field will add to the next. The accumulation of the electron fields will create a magnetic field around the conductor (White, 1987; Chaby and Sherwood, 1995).

2.5.1 Direct current (DC) versus Alternating current (AC) electromagnetism

It is known that a magnetic field results when a current flows through any piece of conductor and wire. If many coils are wound in the same direction, an electromagnet is formed (Duffin, 1990). A Direct current (DC) voltage produces a fixed current in one direction and therefore generates a magnetic field in a coil with fixed polarity, as determined by the right hand rule (Chaby and Sherwood, 1995). For a coil carrying direct current we find the polarity as shown in Figure 2

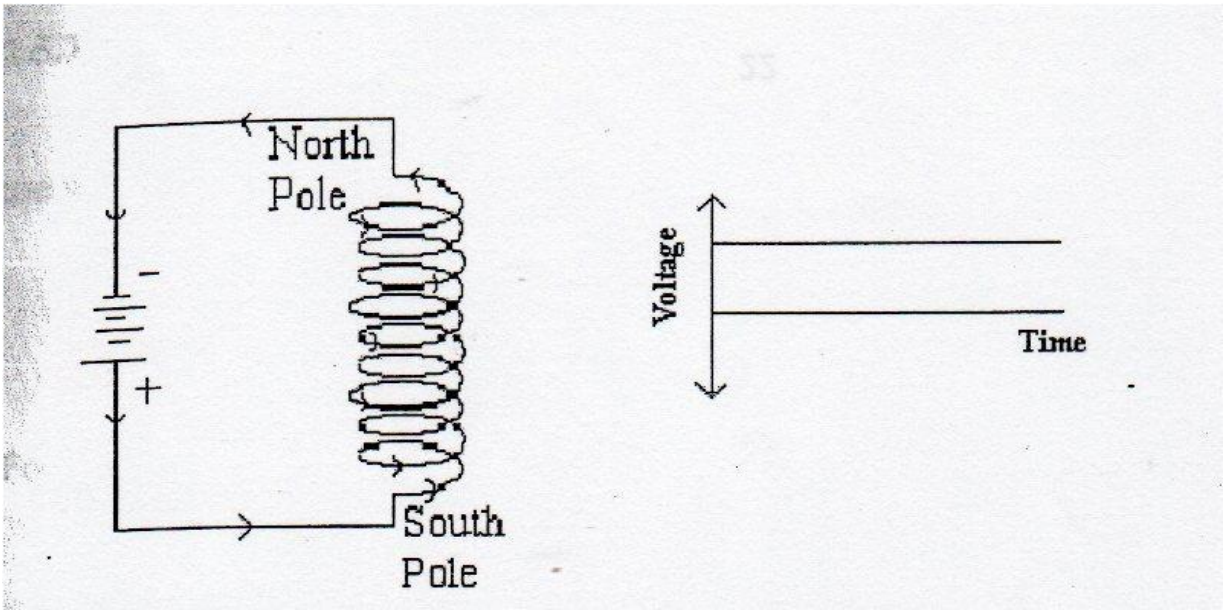

Figure 2: Field due to direct current

Alternating current (AC) is continually varying and as the polarity of the magnetic field is dependent on the direction of the current flow (left-hand rule) the magnetic field will also be alternating in polarity (Figure 3).

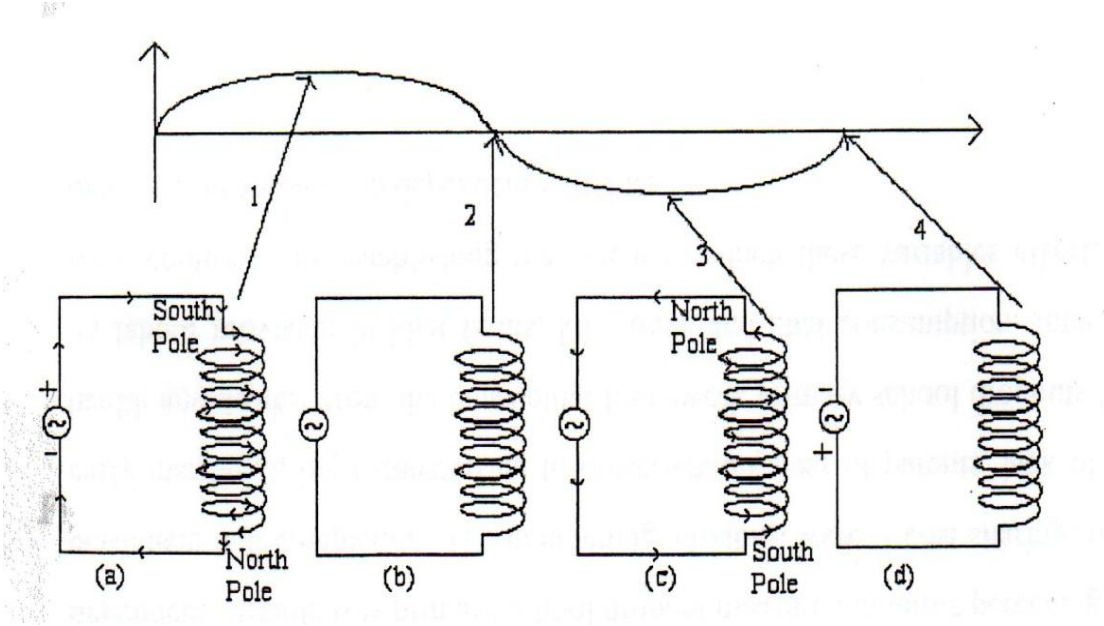

Figure 3: Changing field due to alternating current

2.6 Magnetic field flux

Magnetic field flux (B) is used in determining the strength of a given magnetic field and has been derived from Biot Savart law as shown in the equations 1(a) and (b) (Chabay and Sherwood, 1995).

$$B = \frac{\mu_o}{4\pi}\frac{qvXa}{a^2} \quad \text{..... 1(a)}$$

for a single moving particle

$$B = \frac{\mu_o}{4\pi}\int\frac{IdlXa}{a^2} \quad \text{..... 1(b)}$$

for current distribution

2.6.1 Magnetic field flux on axis of a circular coil

The field due to circular coil with two elements at opposite ends of a diameter of the coil shown in the Figure 4 can be derived by applying Biot-Savart law.

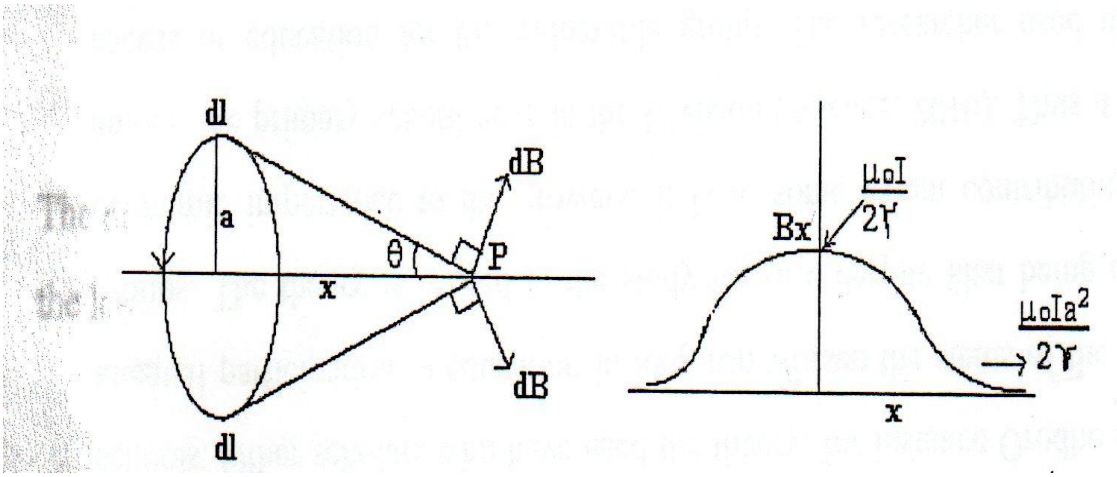

Figure 4: Magnetic field produced by a circular coil

The magnitude of the field due to the element will be given by

$$dB = \frac{\mu_o}{4\pi} \cdot \frac{idl}{\sqrt{x^2+r^2}} \quad \ldots\ldots 2$$

The components of dB to the axis, resolved perpendicularly will cancel, while those resolved along the X-axis will add up. This applies to every such pair of elements. The total field at P is simply the sum of resolved parts along x-axis. This is shown to give along the coil axis a magnetic magnitude given by equation

$$B_x = \frac{\mu_o I \pi r^2}{2r\pi^2 (r^2+x^2)^{3/2}} \quad \ldots\ldots 3$$

Where B_y and B_z are equal to zero;

When x=0 at the centre of the coil axis

$$B = \frac{\mu_o I}{2\pi r^2} \quad \ldots\ldots 4$$

2.6.2 Helmholtz coils

There are numerous ways of producing uniform magnetic fields but the methods commonly employed in the research laboratory are few in number. One of this is shown in Figure 5. In this case, two circular coils of the same radii are arranged such that the distance from one coil to the other is the radius of the coil. This is known as Helmholtz coils. A highly uniform field will be produced at the axial point midway between the coils if the distance apart is properly adjusted. When current is allowed to flow towards the same directions in both coils, the magnetic field strength is of bigger magnitude.

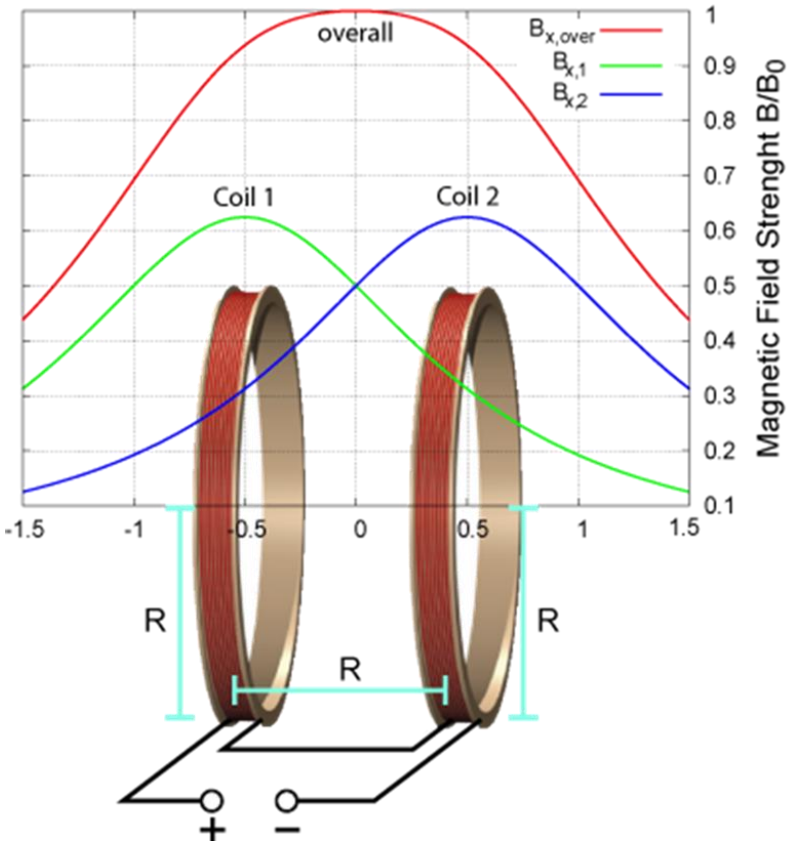

Figure 5: Magnetic field lines around Helmholtz coils

Considering equation 3, for magnetic field flux on the axis of a circular coil, then by replacing

$x = r^{1/2}$ gives

$$B_x = \frac{\mu_o I r^2}{2\left\{r^2+\left(\frac{r}{2}\right)^2\right\}^{3/2}} = \frac{\mu_o I r^2}{2\left(\frac{5}{4}r^2\right)^{3/2}} = \left(\frac{4}{5}\right)^{3/2}\frac{\mu_o I r^2}{2r^3} = 0.717\,\frac{\mu_o I}{2r} \quad \text{.....5}$$

If N is the number of turns in one of the two identical coils then

$$B_x = 0.717\,\frac{\mu_o IN}{2r} \quad \text{.....6}$$

Since the 2 coils contribute equal amount of the field at the centre, then

$$B = 2\times 0.717\,\frac{\mu_o IN}{2r} = 1.43\frac{\mu_o IN}{2r} \quad \text{..... 7}$$

Where;

r is the radius of the coils and I is the current flowing through the coils.

Since resultant field strengths thus produced depends a great deal upon the nature of the coil itself, the magnitude of B between the poles is usually measured by experimental methods.

This study investigated the ability of the magnetic flux density to inhibit the growth of *E. coli* and salmonella species in river Njoro water without altering the essential chemical composition of the water.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Study Area

River Njoro (Figure 6) is located within the Kenyan Rift Valley in Nakuru County and is approximately 60 kilometers (km) in length (Jenkins *et al* 2004). It is a high altitude stream with its source at the eastern segment of Mau Hills around 2700 m a. s. l and terminates in Lake Nakuru. Its geographical location is S00° 34.588´; E035° 54.684´ (Mathooko, 2001). It is the major source of water for domestic purposes (drinking, washing, cooking and bathing), industrial and agricultural use for the surrounding community including Nakuru town, Njoro town, Kenya Agricultural and Livestock Research Organisation (KALRO) and Njoro Canning factory.

3.2 Sample Collection

Water samples were collected at Njoro Bridge, along River Njoro, (Figure 6. Sampling was done weekly for five months using sterilized water sample bottles (250ml). Magnetic treatment of the samples was done in the Physics Laboratory of Egerton University. Microbiological parameters and spectrophotometry were evaluated from Water Resource Management Authority (WRMA) laboratories in Nakuru.

3.3 Sample Analysis

Analysis was done within 0-24 hours after sampling to avoid changes of the bacteria count due to growth or die off (Neill, 2004). Analysis involved the use of membrane filtration (MF) procedures to estimate the initial number of *E. coli* and *salmonella* species present in the water samples. An Atomic Absorption Spectrophotometer (Buck Scientific 210VGP) was used to determine the initial concentrations of the calcium and magnesium ions in the water sample according to Gul and Safdar (2009). The initial pH value of the sample was taken using a pH meter (model H198129).

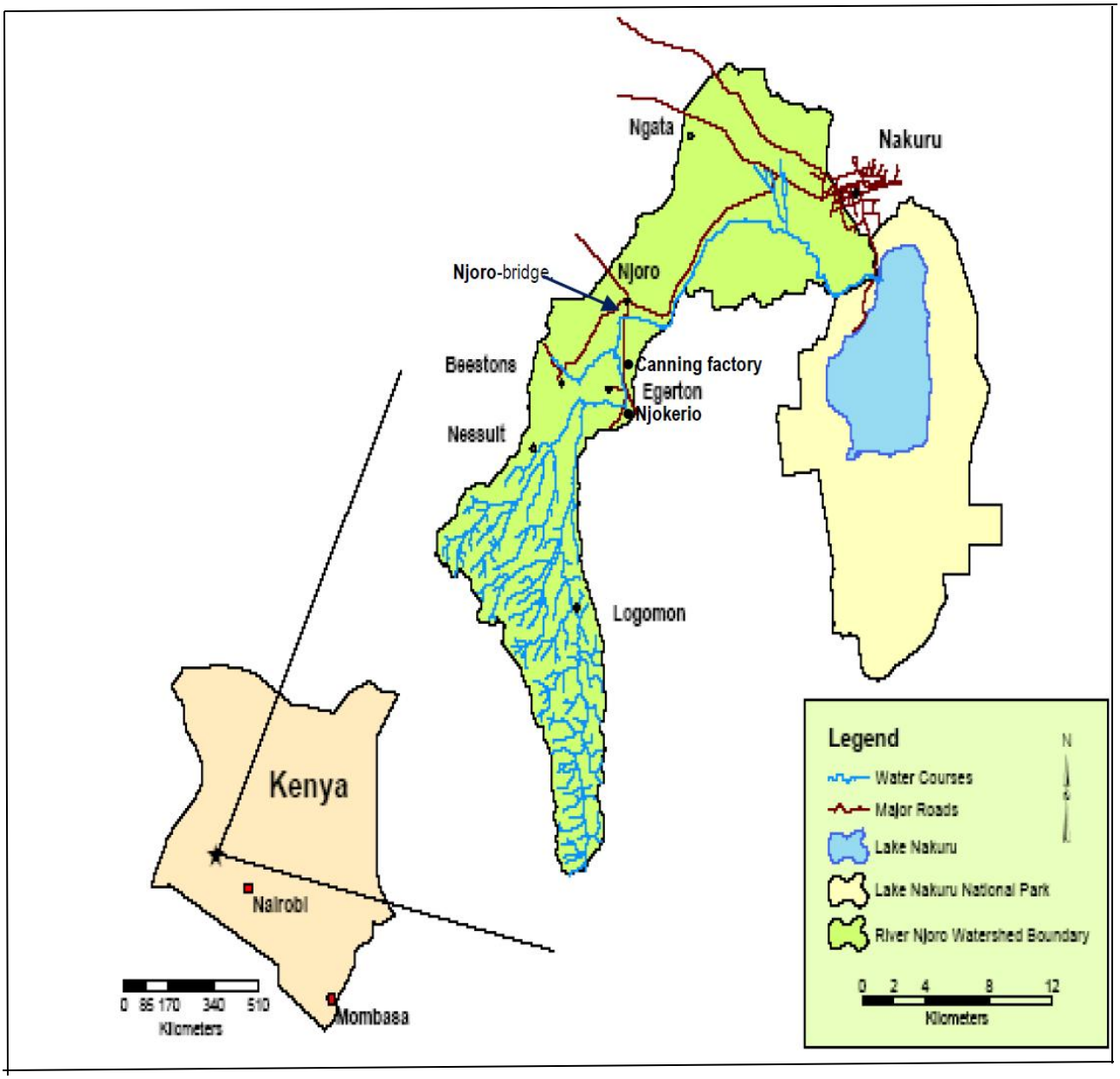

Figure 6: Map of River Njoro from source in Mau Forest to Lake Nakuru.

3.3.1 Membrane filtration technique

Membrane filtration was done according to American Public Health Association (APHA) Standard Methods (2008). Appropriate dilutions of the water sample were done using distilled water. 100ml of the sample or its dilution was aseptically filtered through a membrane filter (47mm diameter, 0.45μm pore size) on a filtration unit. The filter was then taken off using a pair of sterilized forceps and placed on the surface of the corresponding culture media. Colony forming units (CFU) per 100 ml of the sample or its dilution were calculated as described in the United States Department of Agriculture, Food Safety and Inspection Services manual (USDA-FSIS, 2008).

a) Isolation of *E. coli*

For *E. coli* count, filters were placed on Chromocult agar (merck) plates and incubated at 37^0C for 18-24 hours. Typical colonies appearing dark blue were counted as *E. coli* and expressed as CFU's /100ml.

b) Isolation of *Salmonella* species

For *Salmonella* species, the filters were placed on *Salmonella Shigella* agar and incubated at 37^0C for 24 hours. Black colonies on the plate indicated the presence of *salmonella* species and their numbers were also expressed as CFU's /100ml.

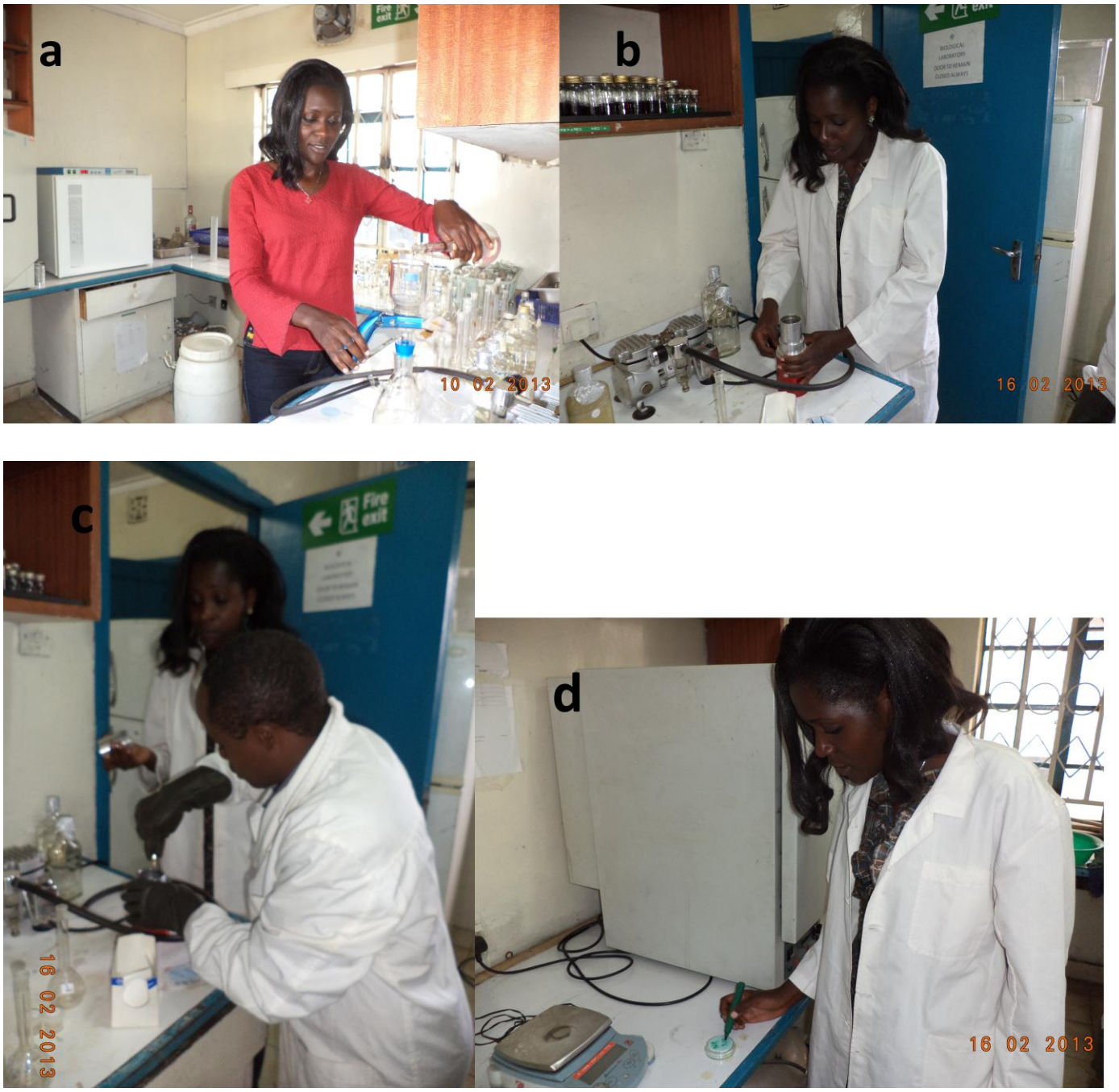

Plate 1: (a, b, c, d) Membrane filtration Analysis at WRMA laboratory Nakuru by Anne Thirika.

3.3.2 Determination of concentration of Ca^{2+}, Mg^{2+} and pH value

Calmagite calorimetric method was used to measure the concentration of Calcium and Magnesium ions in the water. For calcium test, the water samples were mixed with calcium indicator solution and placed in a portable data logging spectrophotometer model (HACH/2010). Readings were taken as mg/L Ca as $CaCO_3$. For magnesium test, the water samples were mixed with magnesium indicator solution and placed in a portable data logging spectrophotometer model (HACH/2010). Readings were taken in mg/L Mg as $CaCO_3$. The stored programme number for calcium was 220 and for magnesium was 225. A pH meter model H198129 was used to measure the pH value of the water.

3.3.3 Water samples magnetic B-field application

The water samples were subjected to three different magnetic flux densities (2mT, 6mT and 10mT) treatments in addition to control which was not subjected to a magnetic field. For each magnetic field the samples were exposed for 2 time periods (6hours and 18 hours). The magnetic B field was produced by Helmholtz coils which were connected to a power supply producing a direct current. An ammeter was also connected in series with the power supply to measure the amount of current passing through the Helmholtz coils. Magnetic flux densities were varied using different currents as well as increasing the number of coils. A teslameter was used to measure the amount of the magnetic B-field produced.

Plate 2: Experimental setup for water samples magnetic B-field application

3.4 Data Analysis

All experiments were replicated three times and the statistical significance of each difference observed among the mean values was determined by standard error analysis. All mean data were statistically analysed with a general linear model procedure of statistical analysis system. The Statistical analysis was done using the Statistical Package for Social Sciences (SPSS) software (Version 17.5). Pearson correlation analysis was carried out to determine the relationship between magnetic field intensity and bacterial count, mineral composition and pH. One-way ANOVA with 95% confidence level was done to compare the effect of the magnetic field intensities with the bacterial count, mineral concentration and pH value. The Least Significant Difference (LSD) was also done to compare the effect of the different magnetic field intensities with each of the three parameters measured.

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 Microbiological parameter

This study investigated the biological effect of magnetic fields, as a component of the non-ionizing radiations, on a unicellular system. Pathogenic microorganism, especially *Escherichia coli*, was chosen to be an experimental model for many reasons; it is widely distributed in the environment such as soil, water and air. *E. coli* is a member of the normal intestinal flora of humans. It causes several diseases such as urinary tract infection, wound infection, traveler's diarrhea, sepsis and meningitis. *S.*typhi was also chosen as it is the main cause of typhoid fever.
The bacterial cultures were exposed to different MFD (2 mT, 6 mT and 10 mT) at 25^{o}C. The duration of exposure was 6 hours and 18 hours which were within the range of the bacterial logarithmic growth phase.

4.1.1 Effect of magnetic flux density on *E. coli*

Different trends on the densities of *E. coli* at different levels of treatment were noted and results obtained indicated in Table 1 and Figures 7,8, 9 and10. Presence of *E. coli* was characterized by development of blue colonies on the surface of the Chromocult culture medium. Plates 3 and 4 (a, b, c, d) show *E. coli* density without B-field exposure (control), under a B-field exposure of 2 mT, of 6 mT and of 10 mT for 6, and 18 hours respectively. It was observed that the control recorded the highest number of CFUs for both 6 and 18hours. Increasing the magnitude of magnetic field led to a significant decrease in the numbers of *E. coli* with the least number of CFUs given by the highest magnetic flux density.

MFD was able to decrease *E. coli* concentration in River Njoro water significantly and hence proved to be a possible small scale cost-effective method of disinfecting water for domestic consumption and reducing incidences of waterborne diseases. The duration of time needed for complete eradication of *E. coli* through magnetic flux application was probably due to the strength of the MFD. More time of exposure to radiation for disinfection was required for low magnetic fields.

This is similar to (Haile *et al*., 2013) who studied the effect of pulsed magnetic field intensity on bactericidal property in sterilization fresh watermelon juice and showed that the overall bactericidal effect was strengthened as the magnetic field intensity increased.

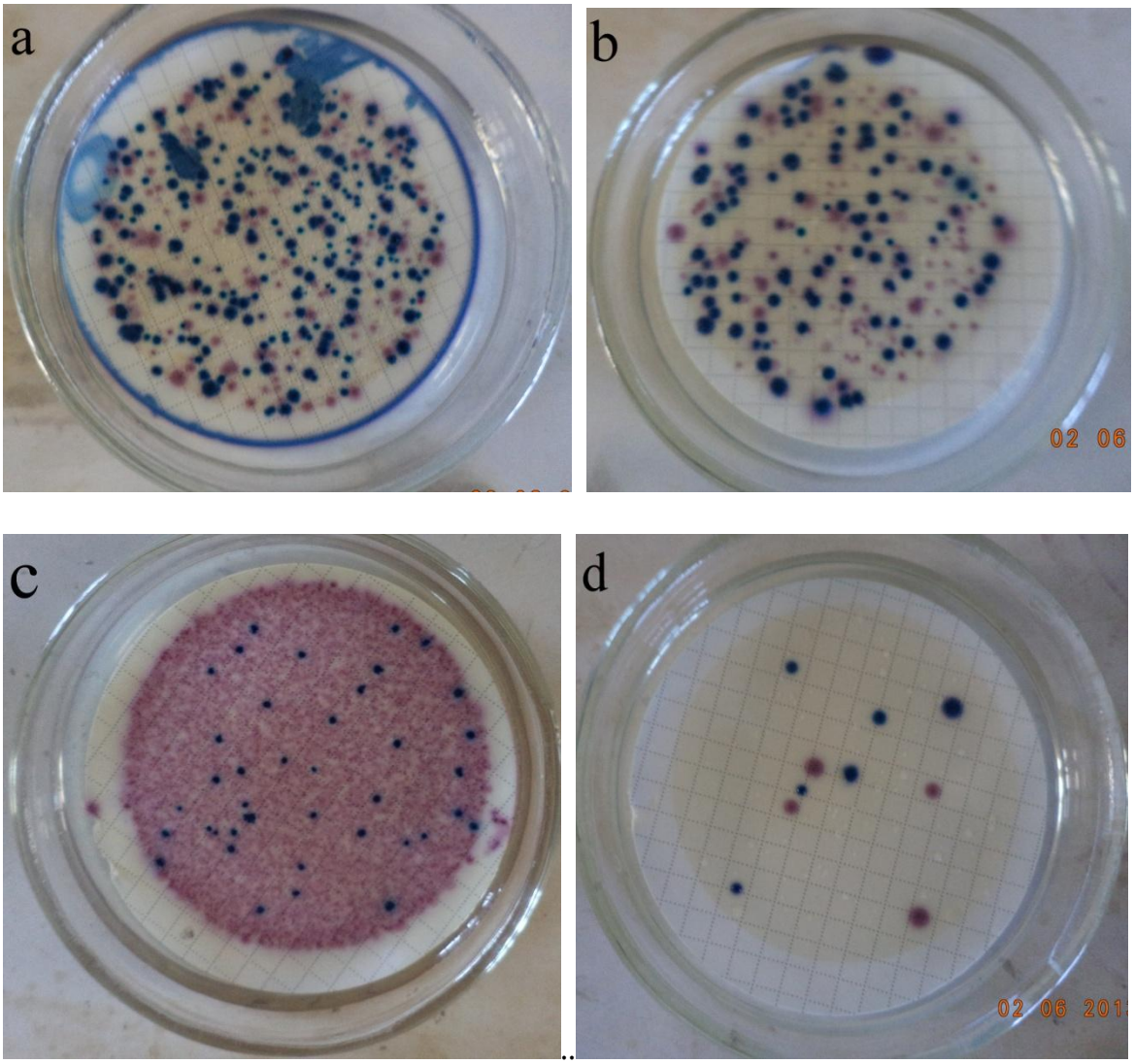

Plate 3: (a, b, c, d) *E. coli* density (blue colonies) after 6 hours of exposure: a -without magnetic field; b – magnetic field of 2mT; c – magnetic field of 6 mT, d- magnetic field of 10 mT.

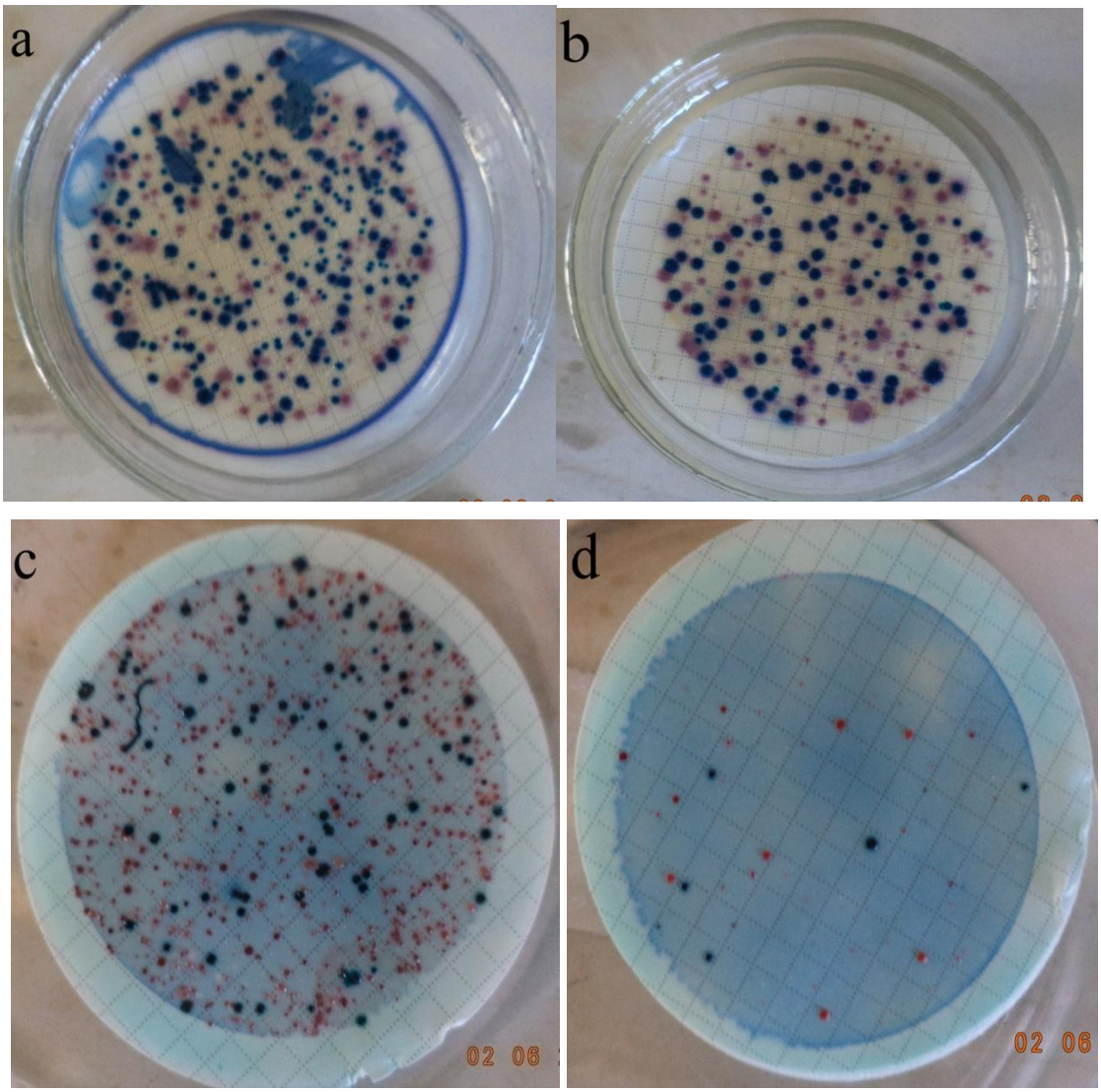

Plate 4: (a, b, c, d) *E. coli* density (blue colonies) after 18 hours of exposure: a -without magnetic field; b – magnetic field of 2 mT; c - magnetic field of 6 mT, d- magnetic field of 10 mT.

Results in Table 1 showed that, the highest means of *E. coli* were recorded with the control. The *E. coli* numbers decreased proportionally by the increase in the magnetic intensity. The least magnetic intensity of 2mT at 6hrs gave a mean number of 337.5 CFU/100ml but after 18 hrs the number increased to 542.5 CFU/100ml. The magnetic intensity of 6mT had more inhibitory effect on *E. coli* at both 6hrs and 18 hrs of magnetic treatment, where bacteria counts were 225 CFU/100ml and 237.5 CFU/100ml respectively. This study showed that the highest magnetic

intensity of 10mT gave the least mean numbers of *E. coli* as 122.5 CFU/100ml and 125 CFU/100ml after 6hrs and 18hrs of exposure respectively.

The acceptable level of *E. coli* in any drinking water is 0 – 100 CFU /100 ml (WHO, 2006). In this study, the highest MFD gave a disinfection level of 122.5 CFU/ 100ml of water which is very close to the WHO maximum acceptable contamination level of *E. coli* in drinking water. This level could be reached by increasing the strength of MFD slightly. Looking at the trend of the results obtained, as the MFD is increased the number of CFUs are decreased significantly.

Table 1: Effect of different magnetic flux densities on *E. coli* numbers (CFU/100ml) after 6hrs and 18 hrs of exposure (average ± standard deviation)

Time of exposure	MFD	Mean ± Std. Dev
6 Hrs	control	687.5 ± 172.3
	2 mT	337.5 ± 95.35
	6mT	237.5 ± 102.1
	10 mT	122.5 ± 42.72
18 Hrs	control	680.0 ± 176.3
	2 mT	542.5 ± 153.3
	6mT	225.0 ± 68.56
	10 mT	125.0 ± 51.96

Figures 7 and 8 show a strong inverse relationship between magnetic flux density and mean numbers of *E. coli* bacteria. It is clear that with increasing strength of magnetic field, the mean numbers of *E. coli* decreased significantly. The application of electromagnetic pulses evidently causes a lethal effect on *E. coli* cells (Mei *et al*., 2007).

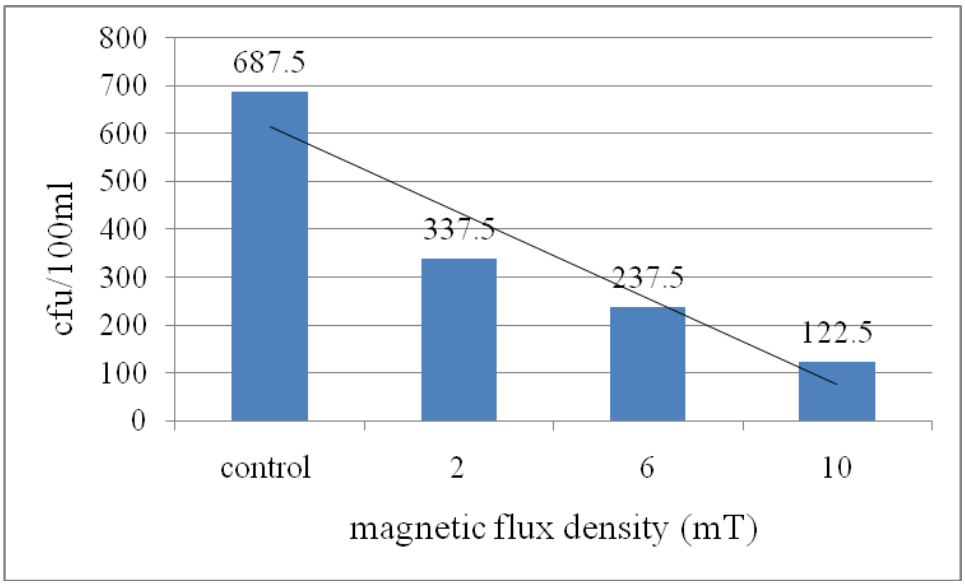

Figure 7: Mean densities of *E. coli* (cfu/100ml) due to different strengths of MFD after 6 hours of exposure.

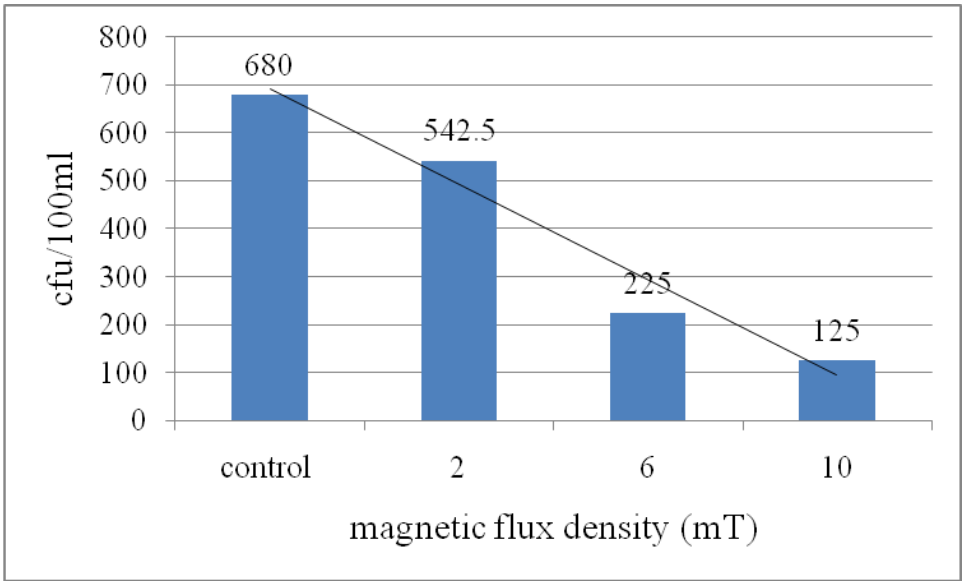

Figure 8: Mean densities of *E.coli* (cfu/100ml) due to different strengths of MFD after 18 hours of exposure

This study indicates that treatment of water with constant magnetic fields always gives rise to a disinfection effect. The magnitude of this effect depends on the strength of magnetic flux density used as well as the time of exposure. Fields of 2mT, 6mT and 10mT caused percentage decrease of 50.9, 65.5 and 82.2 respectively in the number of *E.coli* as compared to the control after 6 hours of exposure (figure 9) and a percentage decrease of 20.2, 66.9 and 81.6 respectively after 18 hours of exposure (figure 10).

The maximum disinfection level for *E.coli* attained in this study was 82.2% with increasing strength of the magnetic flux and period of exposure. This could possibly imply that with a slight increase in both the MFD and time of exposure, the disinfection level could get to at least 99%. The main damaging role of the magnetic fields might be on the cellular membrane that strongly affects, not only the cellular physiological functions, but also the cell-to-cell communications of the bacteria (Molouk *et al.*, 2010).

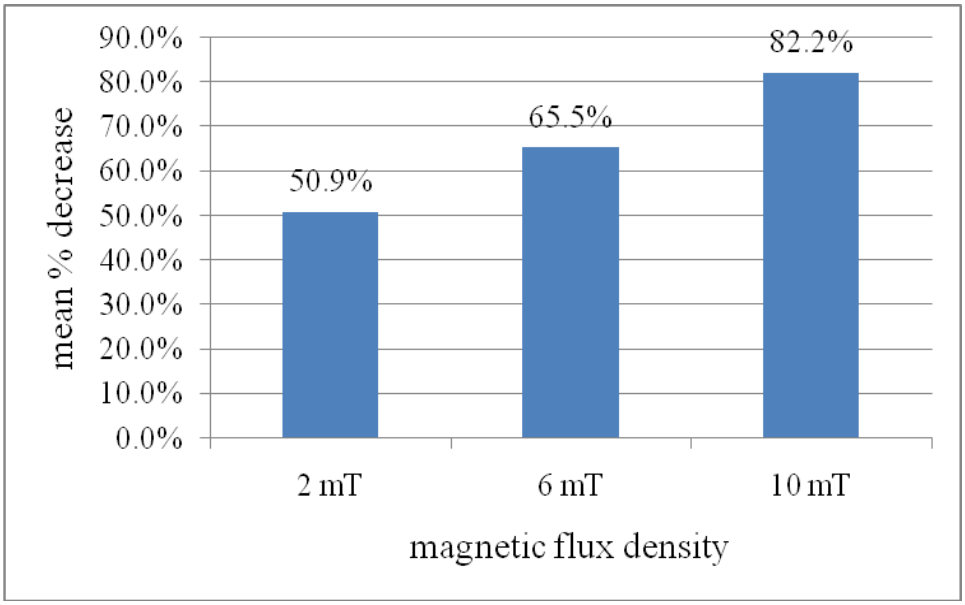

Figure 9: Percentage decrease in the mean number of *E.coli* in water after 6 hours of exposure to different intensities of MFD (2 mT, 6 mT and 10 mT).

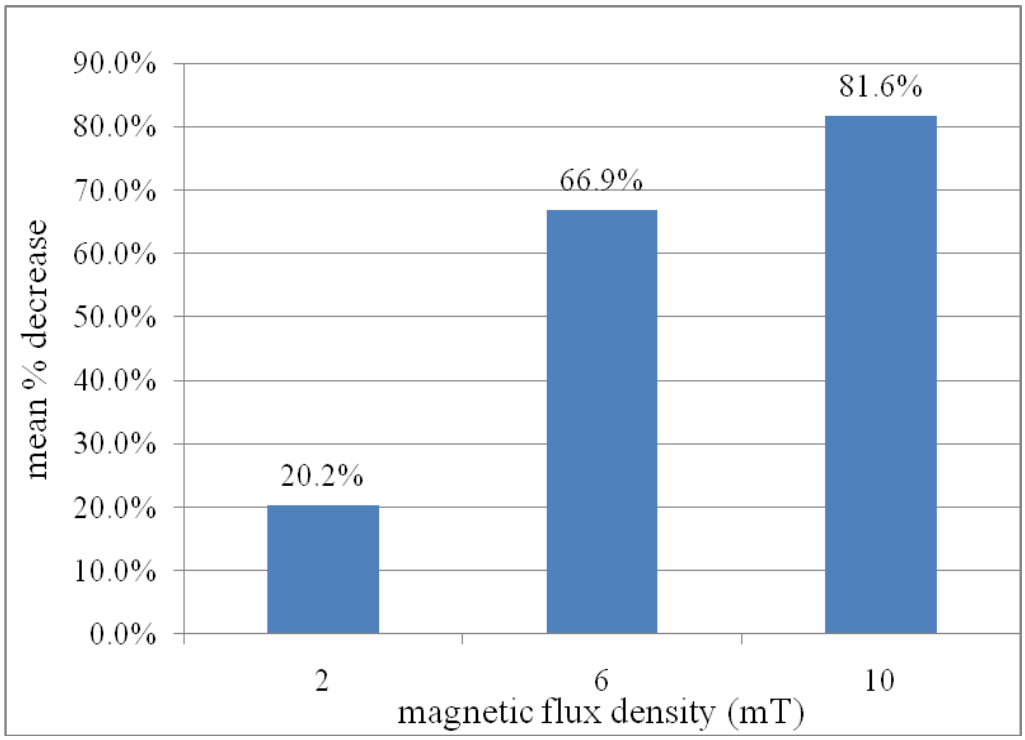

Figure 10: Percentage decrease in the mean number of *E. coli* in water after 18 hours of exposure to different intensities of MFD (2mT, 6mT and 10mT).

Figure 11 shows the relationship between different magnetic field densities (2mT, 6mT and 10mT), different times of exposure (6hours and 18hours) and mean percentage decrease in the density of *E. coli*. It is clear from the figure 11 that when *E. coli* bacteria are exposed to magnetic field, there is bactericidal effect which depends on the strength of the field and the time of exposure. The optimum decrease in the mean number of *E. coli* occurred after 6 hours of exposure to MFD of 10mT which recorded a percentage decrease of 82.2%.

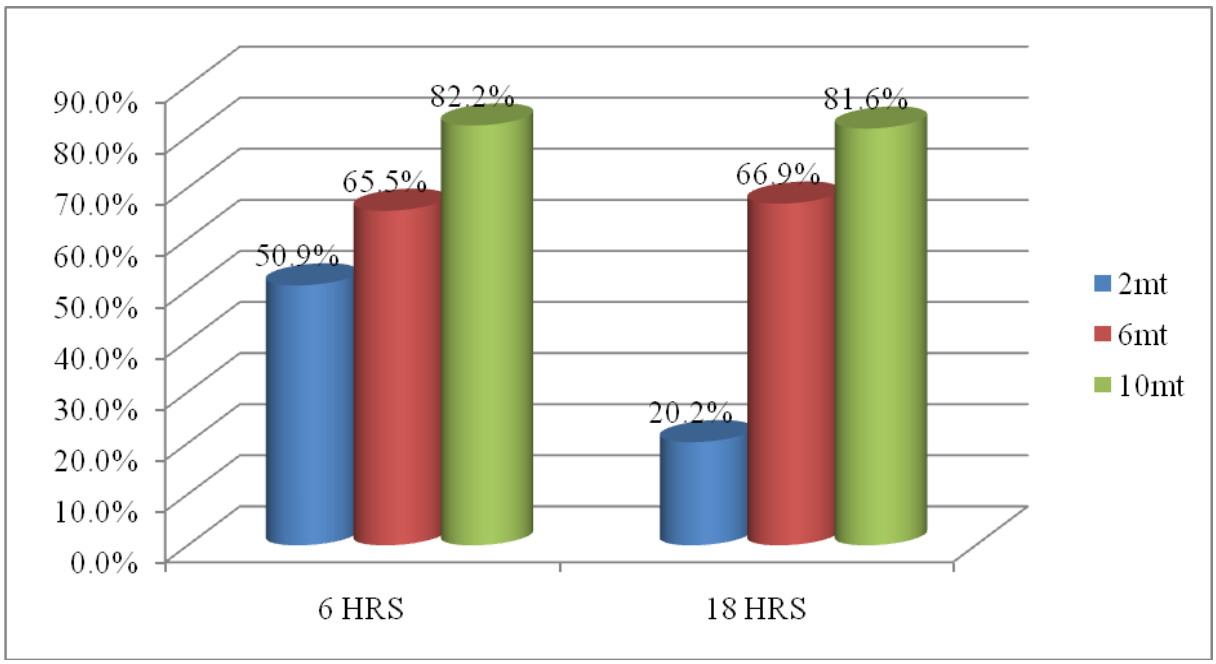

Figure 11: Percentage decrease in the number *E.coli* (cfu/100ml) in river Njoro water after 6 hours and 18 hours of exposure to different intensities of magnetic flux density

An inhibitory effect of EMF on the growth of *E. coli* may be due to the interaction between electric charges induced by EMF and that of the cytoplasm membrane resulting in partial abolishment of electric potential of the cytoplasm membrane with a subsequent decrease in the macromolecular biosynthesis. Also EMF may cause damage of bacterial DNA and inhibition of its replication. The cellular membrane of the microorganism could also have been affected by the external magnetic field, resulting to a disturbance in their metabolic activity and, consequently, a change in their cell division (Mohammed *et al*., 2011).

Correlation analysis demonstrated that MFD was negatively significantly correlated with mean number of CFU ($r = -.999$, $p<0.05$) and ($r = -976$, $p<0.05$) for 6hours and 18hours of exposure respectively as shown in tables 2 and 3.

Table 2: Correlation between MFD (mT) and mean number of *E. coli* (cfu/100ml) after 6 hours of exposure

		MFD	MEAN
MFD	Pearson Correlation	1	-.999(*)
	Sig. (2-tailed)	.	.026
	N	3	3
MEAN	Pearson Correlation	-.999(*)	1
	Sig. (2-tailed)	.026	.
	N	3	3

* Correlation is significant at the 0.05 level (2-tailed).

Table 3: Correlation between MFD (mT) and mean number of *E. coli* (cfu/100ml) after 18 hours of exposure

		MFD	MEAN
MFD	Pearson Correlation	1	-.976(*)
	Sig. (2-tailed)	.	.024
	N	4	4
MEAN	Pearson Correlation	-.976(*)	1
	Sig. (2-tailed)	.024	.
	N	4	4

* Correlation is significant at the 0.05 level (2-tailed).

4.1.2 Effect of MFD on *Salmonella* Species.

Black colonies on the surface of the SS agar showed presence of *Salmonella* species. Plates 5 and 6 (a, b, c, d) show densities of *salmonella* species without B-field exposure (control), under a B-field exposure of 2 mT, of 6 mT and of 10 mT for 6, and 18 hours respectively. The results in plate 5 show that there is a significant decrease in the number of CFU's with increasing magnetic field after 6 hours of exposure.

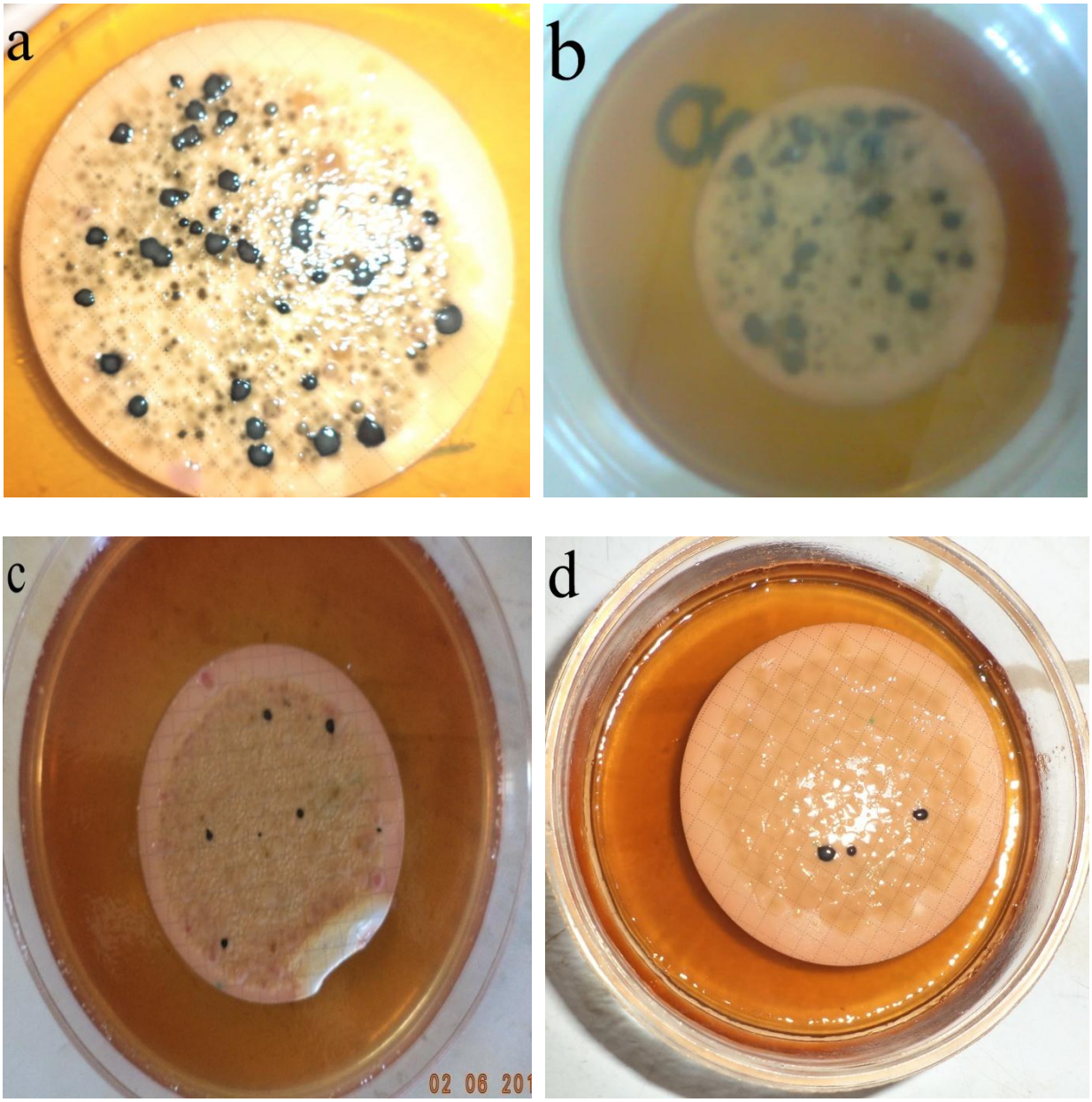

Plate 5: (a, b, c, d) *Salmonella* species densities (black colonies) after 6 hours of exposure: a -without magnetic field; b – magnetic field of 2 mT; c – magnetic field of 6 mT, d- magnetic field of 10 mT

Increasing the time of exposure to 18hours led to a slightly different trend in the number of colonies formed by the different strengths of the magnetic fields used.

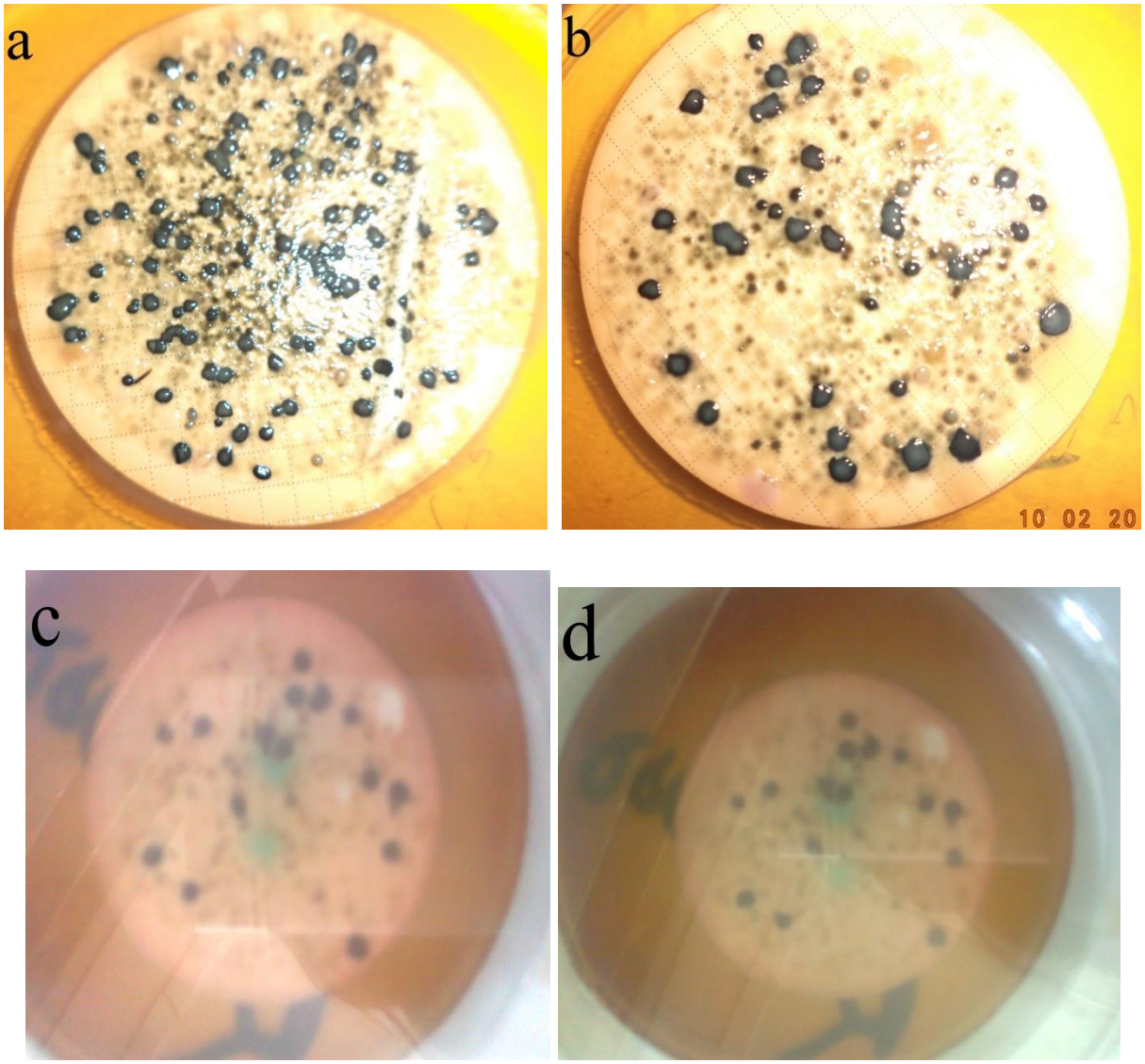

Plate 6: (a, b, c, d); *S*almonella Species densities (black colonies) after 18 hours of exposure: a -without magnetic field; b – magnetic field of 2 mT; c – magnetic field of 6 mT, d- magnetic field of 10 mT

From the present data it can be easily deduced that MFD has bactericidal effects on the density of *Salmonella* species in water after 6 hours of exposure and with increasing strength of the field. The highest mean number of *Salmonella* species was recorded with the control. Magnetic flux of 2 mT gave a mean number of 16.25 cfu/100ml, 6mT decreased the number further to 9.75 cfu/100ml and 10mT recorded the least mean number of 4.25 cfu/100ml. Increasing the time of exposure to 18 hours caused slight decrease in the mean number of the *Salmonella* species as compared to the control. Magnetic fields of 2mT, 6mT and 10mT decreased the mean number of

salmonella species to 14.5, 11.75 and 13.75 cfu/100ml respectively. This is shown in table 4, figure 12 and figure 13.

Table 4: Effect of different strengths of MFD on *Salmonella* species numbers (CFU/100ml) after 6hrs and 18 hrs of exposure (average ± standard deviation)

Time of exposure	MFD	Mean± Std. Dev
6 Hrs	control	18.50 ± 6.19
	2 mT	16.25 ± 6.70
	6mT	9.750 ± 4.65
	10 mT	4.250 ± 2.99
18 Hrs	control	18.25 ± 6.13
	2 mT	14.50 ± 4.93
	6mT	11.75 ± 2.94
	10 mT	13.75 ± 1.73

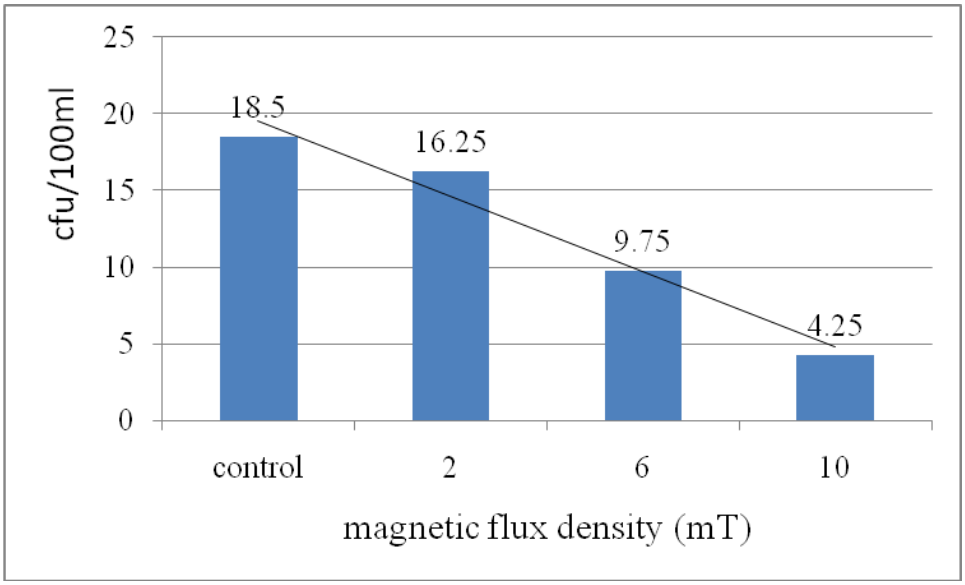

Figure 12: Mean densities of *Salmonella* species (cfu/100ml) due to different strengths of MFD after 6 hours of exposure

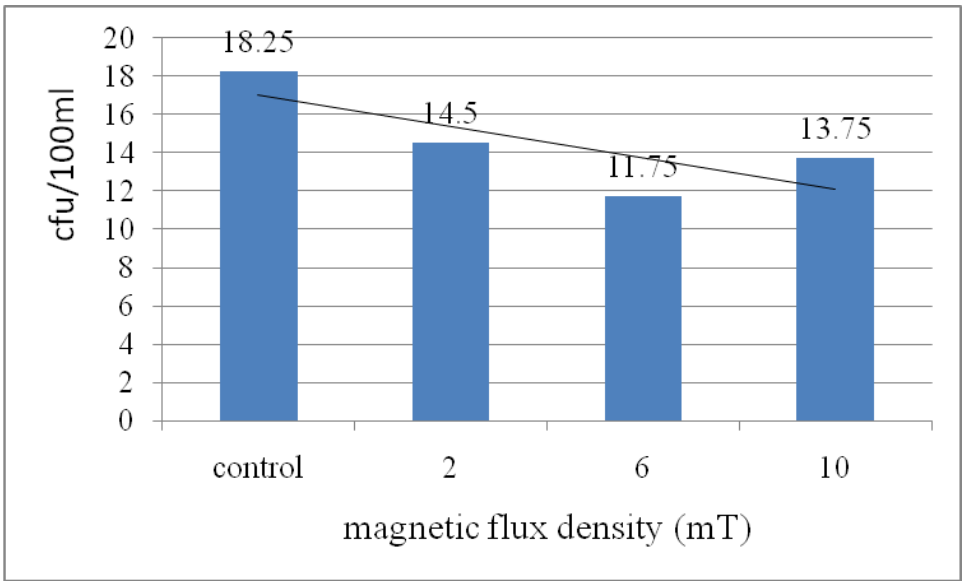

Figure 13: Mean densities of *Salmonella* species (cfu/100ml) due to different strengths of MFD after 18 hours of exposure

Figure 14 and figure 15 shows the percentage decrease of *Salmonella* species as a function of the strength of the magnetic field after 6 and 18 hours of exposure respectively. It is clear from Figure 14 that for the exposure period of 6 hours, an increase in the strength of the magnetic field caused a remarkable decrease in the mean percentage of the *Salmonella* species as compared to the control. However, at the exposure period of 18 hours, there was a very slight decrease in the mean percentage of the *Salmonella* species with a stimulation case at a magnetic flux density of 10mT. This is similar with Mohammed *et al* (2011) who reported that exposing *S. tyhi* to a 20 G magnetic field increased their cell division and cell number. This is shown in Figure 15.

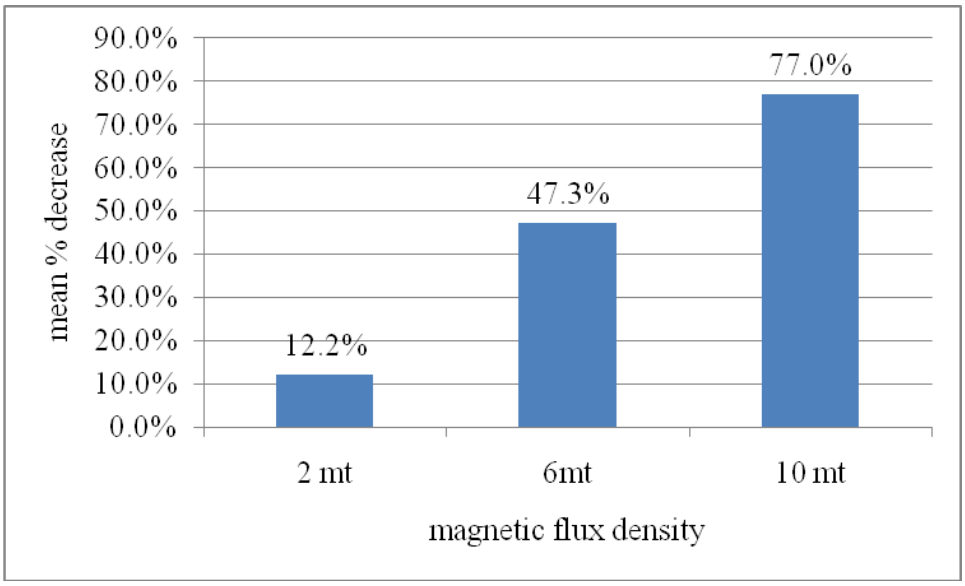

Figure 14: Percentage decrease in the mean number of *Salmonella* species in water after 6 hours of exposure to different intensities of MFD (2mT, 6mT and 10mT).

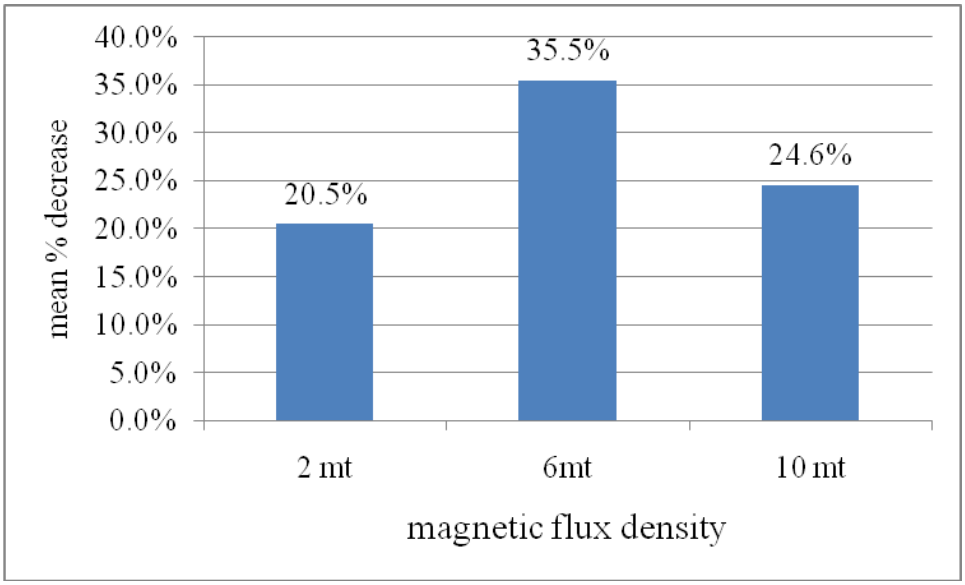

Figure 15: Percentage decrease in the mean number of *Salmonella* species in water after 18 hours of exposure to different intensities of MFD (2mT, 6mT and 10mT).

The results in Figure 16 indicate considerable changes in the percentage mean density of *Salmonella* species for the two exposure periods 6 hours and 18 hours. For the exposure period of 6 hours, maximum percentage decrease (77%) occurred at a magnetic field strength of 10mT.

Increasing the exposure period to 18 hours caused a different trend in the density of the *Salmonella* species. It is possible that MFD has a short period inhibition of *Salmonella* Species These results are in agreement with (Jaffe, 1983) who reported that the electromagnetic field was used either to inhibit or to stimulate the growth of the microorganism under appropriate conditions. For this case the exposure period of 6 hours shows an inhibition case while that of 18 hours shows a stimulation case. This could be as a result of the external magnetic field strength being very large relative to the bio magnetic field of the cells which causes a disturbance in their metabolic function. Del-Re *et al*. (2010).

The fact that MFD could inhibit *Salmonella* Species growth is quite promising as a potential technique for disinfecting water contaminated with *Salmonella* Species. Moreover, the relevance of the findings is strongly supported by the fact that *Salmonella* species are quite heterogeneous and the said genera is an assembly of over 2600 serotypes. Further the infective dose of *Salmonella typhi* the most pathogenic species is 10^7 cells/ml (USEPA, 2011).

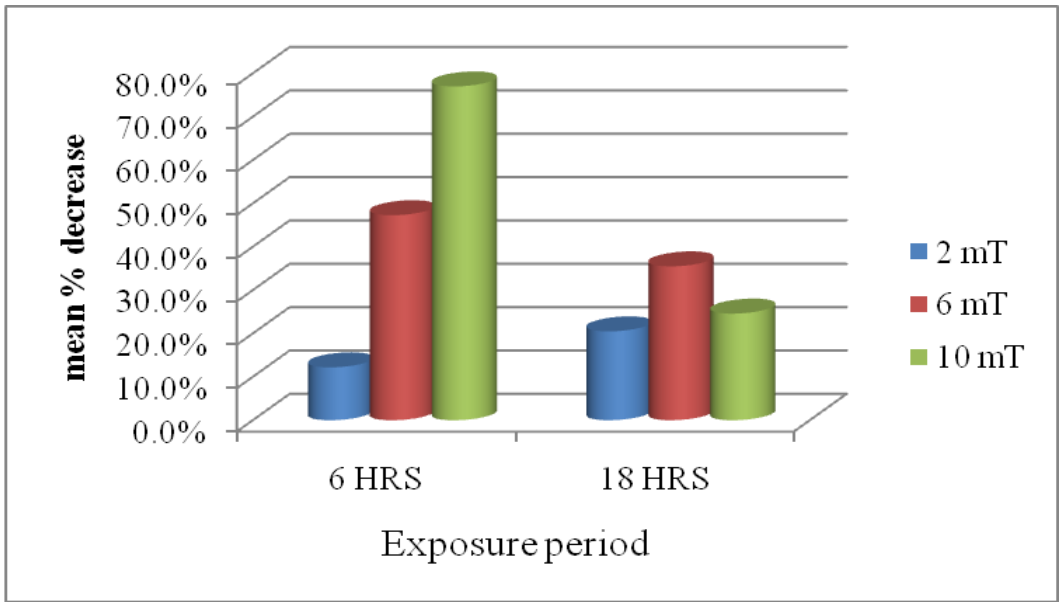

Figure 16: Percentage decrease in the number *Salmonella* species (cfu/100ml) in river Njoro water after 6 hours and 18 hours of exposure to different intensities magnetic flux density.

Correlation analysis demonstrated that MFD was negatively significantly correlated with mean number of CFU ($r = -.999$, $p<0.05$) and ($r = -995$, $p<0.05$) for 6hours and 18hours of exposure respectively as shown in tables 5 and 6.

Table 5: Correlation between MFD (mT) and mean number of *Salmonella* species (cfu/100ml) after 6 hours of exposure

		Magnetic Flux Density	Mean number of *Salmonella Species*
Magnetic Flux Density	Pearson Correlation	1	-.999(**)
	Sig. (2-tailed)	.	.001
	N	4	4
Mean number of *Salmonella Species*	Pearson Correlation	-.999(**)	1
	Sig. (2-tailed)	.001	.
	N	4	4

** Correlation is significant at the 0.01 level (2-tailed).

Table 6: Correlation between MFD (mT) and mean number of *Salmonella* species (cfu/100ml) after 18 hours of exposure

		Magnetic Flux Density	Mean number of *Salmonella Species*
Magnetic Flux Density	Pearson Correlation	1	-.995(**)
	Sig. (2-tailed)	.	.005
	N	4	4
Mean number of *Salmonella Species*	Pearson Correlation	-.995(**)	1
	Sig. (2-tailed)	.005	.
	N	4	4

** Correlation is significant at the 0.01 level (2-tailed).

4.2 Chemical properties.

All measurement of the chemical properties was carried out at the same temperature (25°C) with the intention of reducing the errors.

Table 7: Effect of different strengths of MFD on concentration of Ca^{2+}, Mg^{2+} and the pH value of water samples after 6hrs and 18 hrs. of exposure (mean values).

		Ca^{2+} (mg/L)	**Mg^{2+} (mg/L)**	**PH**
6 HRS	control	3.058	4.745	8.225
	2mT	3.062	4.750	8.188
	6mT	3.061	4.752	8.345
	10mT	3.062	4.755	8.385
18 HRS	control	3.058	4.745	8.230
	2mT	3.061	4.758	8.290
	6mT	3.061	4.757	8.335
	10mT	3.065	4.760	8.362

4.2.1 Effect of MFD on concentrations of Ca^{2+} and Mg^{2+} ions

Results were subjected to variance analysis for validation of the increase tendencies of the measured parameters (Table 7 and appendix 5, 6, 7, 8). The results of Ca^{2+} and Mg^{2+} ions concentration at both 6 hours and 18 hours showed an insignificant change with increasing magnetic field density. After a lot of serious laboratory measurements and chemical analysis, it is concluded that; the total dissolved salts remain the same; no magnesium or calcium is removed from the water by magnetic treatment. This in turn implies that MFD is a possible effective way of disinfecting water without altering the essential element composition of the treated water.

Minerals consist approximately 4% of human body mass. The group of major minerals includes magnesium and calcium among others, and their necessary daily requirement is over 100 mg each. Food is the principal source of calcium and magnesium. Nonetheless, people consuming

refined food may suffer deficiency of essential nutrients, and thus even the relatively low intake of essential elements through drinking water may play an essential role in human health. It has been reported that elements that are present as free ions in water are more readily absorbed from water than from food bound to other substances (USEPA, 2014)

This result does not agree with Matasova *et al*. (2005) and Krzemieniewski *et al*. (2004) who observed a clear inverse correlation between metallic elements concentrations and magnetic field intensity.

4.2.2 Effect of MFD on water pH

Data obtained with untreated and treated water did not reveal any significant changes in pH values (Table 7 and appendix 9, 10). The change in pH value was negligible and inconsistent with increasing magnetic density at both 6 hours and 18 hours of treatment. The MFD treatment in this study kept the water pH at the acceptable level for drinking water which again shows that it is a promising technique for disinfecting water.

Other studies have shown similar or different results of pH of water treated with magnetism to this study as outlined below. Changes in the pH of distilled water of up to 0.4 pH units have been reported by Joshi and Kamat (1966). However, Quickenden (2002) found no pH change in double distilled water subjected to a very strong magnetic field of 24 000 Gauss. Tai et al (2008) cited that Ellingsen and Kristiansen showed that their water sample's pH decreased from pH 9.2 to 8.5 after magnetic treatment. (Busche *et al.*, 1985) showed an initial decrease in pH of 0.5pH units from 7.0 to 6.5, followed by a gradual increase throughout the time of the experiment to pH 7.5 – 8.0. Parsons et al (1996) also recorded a decrease of 0.5 pH units after passing water through a Magnetic field.

Yamashita et al. (2003) witnessed, what he considered slow and large pH fluctuations (0.05 -0.1) during the first several hours of magnetically treating distilled water. His results indicated that to accurately evaluate the effects of magnetic fields on water, subtle experimental conditions such as field conditions produced by common lab devices and procedures cannot be ignored. He also states that extending measurements beyond several hours may be essential to observe accurately the effects of magnetizing water. From these experiments, it appears the fluctuations in pH change from experiment to experiment suggest that unforeseen interactions are contributing to

pH change such as impurities picked up from the treatment device or inaccuracies in the pH meter.

CHAPTER FIVE

CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

Based on the findings of this study, the following conclusions can be drawn;

1. The maximum disinfection efficiency was 82.2% for *E. coli* exposed to 10mT magnetic flux density for a period of 6 hours.
2. The maximum disinfection efficiency was 77% for *Salmonella species* exposed to 10mT of magnetic flux density for a period of 6 hours.
3. Treatment with magnetic field did not alter the concentration of Ca^{2+} and Mg^{2+} ions as well as the pH value of River Njoro water.

5.2 Recommendations

Arising from the findings of this study, further research on the following areas concerns has to be done:

1. A study to show how long the inhibitory effects of magnetic field lasts in the treated water after withdrawing the field.

2. A study to show effect of magnetic field treatment on other strains of bacteria.

3. Investigation of effect of different intensities of magnetic field on heavy metals in water

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APPENDICES

APPENDIX 1: Multiple Comparison of *Salmonella* species and Magnetic Flux Density at 6 Hours

Dependent Variable: Number of microorganisms

LSD

		Mean Difference			95% Confidence Interval	
(I) FIELD	(J) FIELD	(I-J)	Std. Error	Sig.	Lower Bound	Upper Bound
Control	2 mT	2.2500	3.77078	.562	-5.9658	10.4658
	6 mT	8.7500(*)	3.77078	.039	.5342	16.9658
	10 mT	14.2500(*)	3.77078	.003	6.0342	22.4658
2 mT	Control	-2.2500	3.77078	.562	-10.4658	5.9658
	6 mT	6.5000	3.77078	.110	-1.7158	14.7158
	10 mT	12.0000(*)	3.77078	.008	3.7842	20.2158
6 mT	Control	-8.7500(*)	3.77078	.039	-16.9658	-.5342
	2 mT	-6.5000	3.77078	.110	-14.7158	1.7158
	10 mT	5.5000	3.77078	.170	-2.7158	13.7158
10 mt	Control	-14.2500(*)	3.77078	.003	-22.4658	-6.0342
	2 mT	-12.0000(*)	3.77078	.008	-20.2158	-3.7842
	6 mT	-5.5000	3.77078	.170	-13.7158	2.7158

* The mean difference is significant at the .05 level

APPENDIX 2: Multiple Comparison of *Salmonella* species and Magnetic Flux Density at 18 Hours

Dependent Variable: Number of microorganisms

LSD

		Mean Difference			95% Confidence Interval	
(I) FIELD	(J) FIELD	(I-J)	Std. Error	Sig.	Lower Bound	Upper Bound
Control	2 mT	3.2500	4.83800	.514	-7.2911	13.7911
	6 mT	8.7500	4.83800	.096	-1.7911	19.2911
	10 mT	6.7500	4.83800	.188	-3.7911	17.2911
2 mT	Control	-3.2500	4.83800	.514	-13.7911	7.2911
	6 mT	5.5000	4.83800	.278	-5.0411	16.0411
	10 mT	3.5000	4.83800	.483	-7.0411	14.0411
6 mT	Control	-8.7500	4.83800	.096	-19.2911	1.7911
	2 mT	-5.5000	4.83800	.278	-16.0411	5.0411
	10 mT	-2.0000	4.83800	.687	-12.5411	8.5411
10 mT	Control	-6.7500	4.83800	.188	-17.2911	3.7911
	2 mT	-3.5000	4.83800	.483	-14.0411	7.0411
	6 mT	2.0000	4.83800	.687	-8.5411	12.5411

APPENDIX 3: Multiple Comparison of *E.coli* and Magnetic Flux Density at 6 Hours

Dependent Variable: Number of microorganisms

LSD

(I) FIELD	(J) FIELD	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
Control	2 mT	20.0000	177.51760	.912	-366.7776	406.7776
	6 mT	150.0000	177.51760	.415	-236.7776	536.7776
	10 mT	195.0000	177.51760	.294	-191.7776	581.7776
2 mT	Control	-20.0000	177.51760	.912	-406.7776	366.7776
	6 mT	130.0000	177.51760	.478	-256.7776	516.7776
	10 mT	175.0000	177.51760	.344	-211.7776	561.7776
6 mT	Control	-150.0000	177.51760	.415	-536.7776	236.7776
	2 mT	-130.0000	177.51760	.478	-516.7776	256.7776
	10 mT	45.0000	177.51760	.804	-341.7776	431.7776
10 mT	Control	-195.0000	177.51760	.294	-581.7776	191.7776
	2 mT	-175.0000	177.51760	.344	-561.7776	211.7776
	6 mT	-45.0000	177.51760	.804	-431.7776	341.7776

APPENDIX 4: Multiple Comparison of *E.coli* and Magnetic Flux Density at 18 Hours

Dependent Variable: Number of microorganisms

LSD

		Mean Difference			95% Confidence Interval	
(I) FIELD	(J) FIELD	(I-J)	Std. Error	Sig.	Lower Bound	Upper Bound
Control	2 mT	27.5000	190.61031	.888	-387.8042	442.8042
	6 mT	160.0000	190.61031	.418	-255.3042	575.3042
	10 mT	220.0000	190.61031	.271	-195.3042	635.3042
2 mT	Control	-27.5000	190.61031	.888	-442.8042	387.8042
	6 mT	132.5000	190.61031	.500	-282.8042	547.8042
	10 mT	192.5000	190.61031	.332	-222.8042	607.8042
6 mT	Control	-160.0000	190.61031	.418	-575.3042	255.3042
	2 mT	-132.5000	190.61031	.500	-547.8042	282.8042
	10 mT	60.0000	190.61031	.758	-355.3042	475.3042
10 mT	Control	-220.0000	190.61031	.271	-635.3042	195.3042
	2 mT	-192.5000	190.61031	.332	-607.8042	222.8042
	6 mT	-60.0000	190.61031	.758	-475.3042	355.3042

APPENDIX 5: Anova for Concentration of Calcium Ions in Water at 6 Hours of Exposure to Magnetic Flux Density

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	0.070669	3	0.023556	0.596959	0.62904	3.490295
Within Groups	0.473525	12	0.03946			
Total	0.544194	15				

APPENDIX 6: Anova for Concentration of Calcium Ions in Water at 18 Hours of Exposure to Magnetic Flux Density

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	0.127925	3	0.042642	1.376833	0.297063	3.490295
Within Groups	0.37165	12	0.030971			
Total	0.499575	15				

APPENDIX 7: Anova for Concentration of Magnesium Ions in Water at 6 Hours of Exposure to Magnetic Flux Density

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	0.083025	3	0.027675	0.207336	0.88936	3.490295
Within Groups	1.60175	12	0.133479			
Total	1.684775	15				

APPENDIX 8: Anova for Concentration of Magnesium Ions in Water at 18 Hours of Exposure to Magnetic Flux Density

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	0.34545	3	0.11515	0.702866	0.56833	3.490295
Within Groups	1.96595	12	0.163829			
Total	2.3114	15				

APPENDIX 9: Anova for pH value of Water at 6 Hours of Exposure to Magnetic flux Density

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	0.106819	3	0.035606	0.315245	0.814135	3.490295
Within Groups	1.355375	12	0.112948			
Total	1.462194	15				

APPENDIX 10: Anova for pH value of Water at 18 Hours of exposure to Magnetic Flux Density

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	0.040219	3	0.013406	0.129365	0.940801	3.490295
Within Groups	1.243575	12	0.103631			
Total	1.283794	15				