EFFICACY OF *ADANSONIA DIGITATA* L. SEEDS AS A NATURAL SOURCE FOR WATER PURIFICATION

BY

Ocholi Paul EDOGBANYA

B.Sc. BIOLOGY (ABU) 2011

M.Sc/SCI/42689/2012 – 2013

A THESIS SUBMITTED TO THE SCHOOL OF POSTGRADUATE STUDIES

AHMADU BELLO UNIVERSITY, ZARIA

IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF THE DEGREE M.Sc. BIOLOGY

DEPARTMENT OF BIOLOGICAL SCIENCES

FACULTY OF SCIENCE

AHMADU BELLO UNIVERSITY, ZARIA

NIGERIA.

JUNE, 2015

DECLARATION

I declare that the work in this thesis entitled "Efficacy of *Adansonia digitata* L. seeds as a natural source for water purification" has been carried out by me in the Department of Biological Sciences Ahmadu Bello University, Zaria. The information derived from the literature has been duly acknowledge in the text and in the list of references provided. No part of this thesis was previously presented for another degree or diploma at this or any other institution.

Ocholi Paul EDOGBANYA _____ _____

Signature Date

CERTIFICATION

This thesis entitled "Efficacy of *Adansonia digitata* L. seeds as a natural source for water purification" by Ocholi Paul EDOGBANYA, meets the regulations governing the award of the degree of M.Sc. Biology of the Ahmadu Bello University, and is approved for its contribution to knowledge and literary presentation.

Dr. M.A.Adelanwa
Chairman, Supervisory Committee Signature Date
Department of Biological Sciences,
Faculty of Science,
Ahmadu Bello University, Zaria.

Prof. D.S.Abolude
Member, Supervisory Committee Signature Date
Department of Biological Sciences,
Faculty of Science,
Ahmadu Bello University, Zaria.

Prof. A.K.Adamu
Head of Department Signature Date
Department of Biological Sciences,
Faculty of Science,
Ahmadu Bello University, Zaria.

Prof. A.Z. Hassan
Dean, School of Postgraduate Studies Signature Date
Ahmadu Bello University, Zaria.

ACKNOWLEDGEMENTS

My profound appreciation goes to my supervisors, Dr. M.A.Adelanwa and Prof. D.S.Abolude, for taking out time despite their tight schedules to supervise me and make relevant input in this work, to ensure that it comes up to standard. I am equally grateful to the Staff of the Department of Biological Sciences, especiallyMallam A. Magaji in the hydrobiology laboratory for his assistance during thesoxhlet extraction and Dr. Mrs. B. Suleiman for all her wise counsel. The Staff of Department of Water Resources and Environmental Engineering , especially Mr. P.C. Alika, Mr. J. Ameh and Mallam Y. Yakubu for allowing me use their laboratory and all the assistance offered me during thebiocoagulation experiments. The Staff of Department of Pharmaceutics and Pharmaceutical Microbiology, most especially Pastor Dangana Ezekiel, for putting me through in the disinfection experiments. The Staff of Department of Chemistry, especially Dr. O.J. Ocholi and Mr. Oche in the M.Sc. laboratory, for their assistance during the adsorption experiments.

I want to say a big thank you to my parents Mr. and Mrs. E.A.Edogbanya, for their support financially,socially, morally, spiritually and otherwise, I LOVE YOU. I also want to appreciate my fellow soldiers (my brothers), Akoji, Reuben, Benjamin, Ephraim, and Michael for believing so much in me.

Special thanks goes to my Father in Zaria, late Elder S.A. Ocholi. It pains my heart that you left us during the course of this study. I wish you were here to see the end of it all. Thank you for the immeasurable love you lavished on me. To my mummy in Zaria, Mrs. M. Ocholi, thank you for everything, the lord is your strength. I want to also appretiate all my other relatives in Zaria: Especially Dr. Amana, his wife Aisha and their children (Unyo and Ugane), thank you for providing me with a home during my course of study. To the rest of you all, Ene and his wife Lizzy, Odike (espirit de corps) and his wife Faith, Okpass, Ebus, thank you for your love and understanding.

My heart felt gratitude also goes to my friends and colleagues. To my sweetheart, Promise Ayuba(Masoyina), for standing with me all through, it was as though we did the masters together, I LOVE YOU. And to Marvellous (My friend and brother), Ugo, Enenwa, Kamba and all others that I cannot mention, thank you for sticking to me all through.

Finally to my Spiritual family, The God-life Assemble, and my spiritual father, Pastor Chingtok I. Ishaku, thank you for the love and spiritual training to take the nations.

DEDICATION

This work is dedicated to my friend and companion, THE HOLY SPIRIT, Who gave me divine inspiration, grace, and speed, to finish this work. I LOVE YOU.

ABSTRACT

Scientific studies aimed at evaluating theefficacy of *Adansonia digitata* L. seeds as a natural source for water purification, was determined in some laboratories in Ahmadu Bello University, Zaria, from 23rd july, 2014 to 7th November, 2015. Dried fruits of *A. digitata* were collected from the Department of Biological Sciences, Ahmadu Bello University, Zaria.The seeds were excised, washed with sterile distilled water, sundried, powdered, defatted using n-hexane and stored in the refrigerator for further studies.Model polluted water was prepared in the laboratory while surface water was obtained from the Kubanni Reservoir, Ahmadu Bello University, Zaria. Experiments were carried out in triplicates with powdered defatted seeds at doses of 0mg/l (control), 50mg/l, 100mg/l, 150mg/l and 200mg/l as biocoagulant, disinfectant and biosorbent. Jar test experiments were carried out for the biocoagulation studies; disinfection experiments for disinfection studies; and batch experiments for the adsorption studies. The experimental design was Cross Randomized Design (CRD) and One way Analysis of Variance (ANOVA) was used to determine significant difference among means of the various parameters measured. The New Duncan Multiple Range Test (NDMRT) was used in separating means with different significant values. Regression analysis was used for the kinetic studies. The confidence level was taken at 95% ($p<0.05$). Results revealed that an optimum dose of 150mg/l was able to reduce turbidity of model water significantly ($p < 0.05$) from 20.06NTU to 6.57NTU, but there was no significant reduction ($p >0.05$) in the turbidity of surface water. As a disinfectant, a dose of 200 mg/l was able to significantly reduce ($p < 0.05$) Total Coliform Count of the model water and surface

water from 1.65×10^4 cfu/ml and 4.27×10^2 cfu/ml to 5.00×10^2 cfu/ml and 6.67×10^1 cfu/ml respectively. As biosorbent a dose of 200mg/l was able to reduce the initial concentration of Cd^{2+} and Pb^{2+} by 72.98% and 97.49% respectively in model water; and by 93.52% and 100% respectively in surface water. The adsorption of Cd^{2+} and Pb^{2+} onto the biosorbent followed the 2^{nd} order Kinetic model with R^2 value of 0.993 and 0.998 respectively. The 2nd order constants (k_2) obtained for Cd^{2+} and Pb^{2+} were 2.29×10^{-5} and 5×10^{-6} respectively.*Adansonia digitata* seeds possess biocoagulant, disinfectant and biosorptive potentials, but it is not significantly effective for highly turbid and contaminated water. The seeds may possibly be used to complement other conventional chemicals in the purification of water.

TABLE OF CONTENTS

Title page i
Declaration ii
Certification iii
Acknowledgements iv
Dedication v
Abstract vi
Table of Contents viii
List of Figures xii
List of Tables xiii
List of Plates xiv
List of appendices xv
Abbreviations xvii

CHAPTER ONE
1.0 INTRODUCTION 1
1.1Preamble 1
1.2 Statement of Research Problem 4
1.3 Justification 4
1.4 Aim 5
1.5 Objectives 5
1.6 Null Hypotheses 5

CHAPTER TWO
2.0 LITERATURE REVIEW 6
2.1 Conventional Methods of Water Treatment 6
2.1.1 Coagulation (flocculation and sedimentation) 7
2.1.2 Filtration 8
2.1.3 Ion exchange 8
2.1.4 Adsorption 8
2.1.5 Disinfection 8

2.2 Plant Seeds as Biocoagulants and Disinfectants 9
2.2.1 Mechanism of action of seeds as biocoagulants and disinfectants 10
2.2.2 Advantages of the use of seeds as biocoagulants and disinfectants 11
2.2.3 Disadvantages of the use of seeds as biocoagulants and disinfectants 12
2.3 Plant Seeds as Biosorbents 12
2.3.1 Mechanism of action of seeds as biosorbents 14
2.3.2 Advantage of the use of seeds as biosorbents 14
2.3.3 Disadvantages of the use of seeds as biosorbents 15
2.4 Kinetic Models for Biosorption 16
2.4.1 The pseudo-first-order kinetic Model 16
2.4.2 The pseudo-second-order kinetic Model 17
2.5 *Adansonia digitata*L. 18
2.5.1 Classification 18
2.5.2 General description 18
2.5.3 Distribution 20
2.5.4 Uses 20
2.5.4.1 *Food uses* 20
2.5.4.2 Domestic non-food uses, other than medicinal 22
2.5.4.3 *Medicinal uses* 23

CHAPTER THREE
3.0 MATERIALS AND METHODS 24
3.1 Collection, Identification and Preparation of Samples 24
3.1.1 Collection of water samples 24
3.1.2 Collection, identification and preparation of plant material 25
3.1.3.1 *Digestion* 25
3.1.3.2 *Distillation* 26
3.1.3.3*Titration* 26
3.3 Biocoagulation Studies 27
3.3.1 Preparation of model turbid water 27
3.3.2 Extraction of active component from *Adansonia digitata* seeds 27
3.3.3 Jar tests experiment 28
3.3.4 Measurements of physicochemical parameters 28

3.3.4.1 *Turbidity* 28
3.4.4.2 *Electronic conductivity (EC) and total dissolved solid (TDS)* 28
3.4.4.3 *pH* 28
3.4.4.4 *Total suspended solids (TSS)* 28
3.4.4.5 Total alkalinity 30
3.4.4.6 *Hardness* 30
3.4.4.7 Chemical oxygen demand (COD) 31
3.5 Disinfection Studies 32
3.5.1 Preparation of *Escherichia coli* culture 32
3.5.2 Disinfection experiment 33
3.6 Adsorption Studies 33
3.6.1 Preparation of stock solutions of heavy metals (Lead and Cadmium) 33
3.6.2 Batch adsorption experiments 34
3.6.3 Kinetic studies 35
3.7 Statistical Analysis 36

CHAPTER FOUR

4.0 RESULTS 37
4.1 Confirmation and Protein Content of *Adansonia digitata* Seed 37
4.2 Biocoagulation Studies and its Efficacy on Physicochemical Parameters on Surface and Model Water 37
4.3 Disinfection Studies. 52
4.4 Adsorption Studies 52
4.4.1 Effect of pH on biosorption efficiency 52
4.4.2 Effect of initial concentration of Cd^{2+} and Pb^{2+} on biosorption efficiency 52
4.4.3 Effect of contact time on biosorption efficiency 54
4.4.4 Effect of dosage of biosorbent on biosorption efficiency 54
4.5 Kinetic Studies 60
4.5.1 Adsorption of Cd^{2+} 60
4.5.2 Adsorption of Pd^{2+} 60

CHAPTER FIVE

5.0 DISCUSSION..... 62

5.1 The Protein Content of *Adansonia digitata* seeds and its Effect as Biocoagulant..... 62

5.2 *Adansonia digitata* Seeds as Disinfectant..... 63

5.3 *Adansonia digitata* Seeds as Biosorbent..... 64

5.4 Kinetic Studies..... 66

CHAPTER SIX

6.0SUMMARY, CONCLUSIONS AND RECOMMENDATIONS..... 67

6.1 Summary..... 67

6.2 Conclusions..... 68

6.3 Recommendations and Further Studies..... 68

REFERENCES..... 70

APPENDICES..... 79

LIST OF FIGURES

Figure 2.1: Schematic diagram of conventional methods of water purification 6

Figure 2.2: Schematic diagram showing mechanism of coagulation 7

LIST OF TABLES

Table 4.1: Plant verification and protein content 38

Table 4.2: Effect of biocoagulant on different parameters of Model turbid water 47

Table 4.3: Effect of biocoagulant on different parameters of surface water. 48

Table 4.4: Effect of biocoagulant on Total Coliform Counts in model and Surfacewater 53

Table 4.5: Effect of pH on the Adsorption of Cd^{2+} and Pb^{2+} from model water onto *Adansonia digitata* seed biosorbent 55

Table 4.6: Effect of initial concentration of ions on the adsorption of Cd^{2+} and Pb^{2+} from model water onto *Adansonia digitata* seeds 56

Table 4.7: Effect of dosage of biosorbent on the removal of Cd^{2+} and $Pb^{2+}$from model water onto *Adansonia digitata* seed biosorbent 57

Table 4.8: Effect of contact time on the adsorption of Cd^{2+} and Pb^{2+} from model water onto *Adansonia digitata* seed biosorbent 58

Table 4.9: Effect of dosage of biosorbent on the adsorption of Cd^{2+} and Pb^{2+} from surface water onto Adansonia digitata seed biosorbent 59

Table 4.10: Pseudo 1^{st} and 2^{nd} order constants for the Adsorption of Cd^{2+} and Pb^{2+} onto *Adansonia digitata* seed biosorbent 61

LIST OF PLATES

Plate I. *Adansonia digitata*tree 39

Plate II: Dried fruits of *Adansonia digitata* 40

Plate III : Fruits of *Adansonia digitata* broke open 41

Plate IV: Seeds of *Adansonia digitata* 42

Plate V: Powered seeds of *Adansonia digitata*..... 43

Plate VI: Cake residue of defatted Powdered *Adansonia digitata* seeds 44

Plate VII: Model water before treatment with *Adansonia digitata* seed biocoagulant 49

Plate VIII: Model water after treatment with *Adansonia digitata* seed biocoagulant 50

Plate IX: Formation of flocs during jar experiment after about 5 minutes of stirringmodel water with a concentration of 150mg/l of Adansonia digitata seed biocoagulant..... 51

LIST OF APPENDICES

Appendix I: Figure showing effect of different doses of *Adansonia digitata*seed biocoagulant on turbidity of water 79

Appendix II: Figure showing the effect of different doses of *Adansonia digitata*seed biocoagulant on TSS of water 80

Appendix III: Figure showing the effect of different doses of *Adansonia digitata*seed biocoagulant on COD of water..... 81

Appendix IV: Figure showing the effect of different doses of *Adansonia digitata*seed biocoagulant on hardness water 82

Appendix V: Figure showing the effect of different doses of *Adansonia digitata*seed biocoagulant on alkalinity of water 83

Appendix VI: Figure showing the Effect of different doses of *Adansonia digitata*seed biocoagulant on TDS of water..... 84

Appendix VII: Figure showing the effect of different doses of *Adansonia digitata* biocoagulant on EC of water 85

Appendix VIII: Figure showing the effect of different doses of *Adansonia digitata*seed biocoagulant on pH of water 86

Appendix IX: Figure showing the effect of different doses of *Adansonia digitata*seed biocoagulant on Total Coliform Count of water 87

Appendix X: Figure showing the effect of pH on the biosorption efficiency of *Adansonia digitata* seed biosorbent on Cd^{2+} and Pb^{2+} in model water ... 88

Appendix XI: Figure showing the effect of initial concentration of solution on the biosorption efficiency of *Adansonia digitata* seed biosorbent on Cd^{2+} and Pb^{2+} in model water 89

Appendix XII: Figure showing the effect of initial concentration of metal ions on the biosorption efficiency of *Adansonia digitata* seed biosorbent on Cd^{2+} and Pb^{2+} in model water 90

Appendix XIII: Figure showing the effect of dosage of *Adansonia digitata* seed biosorbent on biosorption efficiency of Cd^{2+} and Pb^{2+} in model water... 91

Appendix XIV: Figure showing the effect of dosage of *Adansonia digitata* seed biosorbent on biosorption efficiency of Cd^{2+} and Pb^{2+} in surface water. 92

Appendix XV: Figure showing the pseudo 1^{st} order kinetic model for the adsorption of Cd^{2+} onto *Adansonia digitata* seed..... 93

Appendix XVI: Figure showing the pseudo 2^{nd} order kinetic model for the adsorption of Cd^{2+} onto *Adansonia digitata* seed biosorbent 94

Appendix XVII: Figure showing the pseudo 1^{st} order kinetic model for the adsorption of Pb^{2+} onto *Adansonia digitata* seed biosorbent 95

Appendix XVIII: Figure showing the pseudo 2^{nd} order kinetic model for the adsorption of Pb^{2+} onto *Adansonia digitata* seed biosorbent 96

ABBREVIATIONS

ANOVAAnalysis of Variance

CODChemical Oxygen Demand

CRD Cross Randomized design

DBPs Disinfection By-products

NDMRTNew Duncan Multiple Range Test

ECElectrical Conductivity

NOM Natural Organic Matter

TDS Total Dissolved Solids

TSSTotal Suspended Solids

GPS Global Positioning System

CHAPTER ONE

1.0 INTRODUCTION

1.1 PREAMBLE

Water is a basic resource, it is also the most essential liquid substance, and the sustenance of life is highly dependent on its availability(FAO, 2014). The importance of water cannot be overemphasized; as it is used for domestic, agricultural, industrial, economic and commercial purposes.

WBCSD (2005) reported that over 70% of the surface of theearth is covered by water, but less than 3% of the world's water is fresh andconsumable, while the rest is seawater and undrinkable. Of this 3% over 2.5% is frozen, locked up in Antarctica, the Arctic and glaciers, and not available to man. Thus humanity rely on 0.5% for all of man's and ecosystem's fresh water needs.This 0.5% is thewater found in lakes, rivers, reservoirsand those underground sources that areshallow enough to be tapped at anaffordable cost. Only this amount isrenewed regularly by rain and snowfall,and therefore available on a sustainablebasis (Colwell and Sasson, 1996). An old adage captures the dangerous paradox of water this way: "Water, water, everywhere, but not a drop to drink." It's everywhere, yet most of it is inaccessible. Even the little amount available to man has drastically reduced over the years due to pollution.

The environment has a way of multiplying and giving back what man gives to it, so man has become a snare to himself, by constantly discharging harmful substances into his

environment (Edogbanya*et al.*, 2013b). The problem of water pollution has become an issue drawing global attention. Every day, 2 million tons of sewage, industrial and agricultural waste is discharged into the world's water (UN WWAP, 2003). The United Nations estimates that the amount of wastewater that results in pollution, produced annually is about 1.5 million m^3, this is six times more water than exists in all the rivers of the world(UN WWAP, 2003).

Polluted water causes serious health implications worldwide. It is estimated that 3,900 children die each day due to dirty water or poor hygiene (WHO, 2004). 1.8 million people die every year from diarrhoeal diseases (including cholera) – the equivalent of 15 killer tsunamis each year or 12 Boeing 747 crashes every day(WBCSD, 2005). Food and Agricultural Organisation (FAO) of the United Nations (U.N) revealed that in African countries, particularly Nigeria, water related diseases had been interfering with basic human development (FAO, 2007).

Heavy metals are known to be one of the most fatal pollutants of water. This is because they are very poisonous even in very minute quantities in water (Igwe and Abia, 2006; Mudgal*et al*., 2010; Edogbanya*et al*., 2013b). They find their way into surrounding water bodies majorly as a result of adverse activities carried out by man such as: discharge of untreated industrial wastes into water bodies; improper use of agro-allied chemicals such as pesticides, herbicides and fertilizers; mining activities etc. (Celik and Demirbas, 2005; Mudgal*et al*.,2010; Banerjee,*et* al., 2012; Edogbanya*et al*., 2013b). The health hazards of heavy metals are enormous: Lead (Pb) is known to have adverse effects especially in children, causing anemia, cancers, reproductive disorders and hormone disorders;

Cadmium (Cd) affects internal organs like the Kidneys and liver; Mercury (Hg) causes a Hunter-Russell syndrome, Minamata disease and gastrointestinal disorders; Arsenic (As) leads to various dermal effects like: hyperkeratosis, hyperpigmentation and hypopigmentation; Copper (Cu) causes gastrointestinal disorders; Chromium (Cr) is known to be mutagenic, carcinogenic and teratogenic (Asmatullah and Shakoori, 1998; Chen and Dixon, 1998; Igwe and Abia, 2006; Mudgal*et al.,* 2010).

Drinking water treatment involves a number of unit processes. Commonly used chemicals for the various treatment units are synthetic organic and inorganic substances (such as alum, chlorine, acrylamide and activated carbon), usually these chemicals are expensive and are not readily available especially in the rural areas (Ghebremichael, 2004). Apart from these they also constitute a number of health problems - for instance the use of alum have been reported to cause Alzheimer's disease(Crapper *et al*.,1973; Miller *et al.*,1984; Ghebremichael*et al.*,2005); while some synthetic organic polymers, such as acrylamide, have strong neurotoxic and carcinogenic effect (Choubey*et al.,* 2012); Chlorine being a strong oxidizing agent reacts with Natural Organic Matter (NOM) to form Disinfection By-Products (DBPs), and these DBPs have been associated with increased risk for cancer and other health related issues (Morris *et al.,* 1992;Cantor *et al.,* 1998; Villanueva *et al.,* 2006; Richardson *et al.*, 2007). They have also been reported to be non-eco-friendly as they tend to affect non-target organisms (Ahluwalia *et al.,* 1998) and are usually non-biodegradable (Pritchard *et al.,* 2009).

Since the use of conventional methods for water treatment in developing countries is unsustainable, there is a need to consider alternative technologies of water treatment using naturally occurring materials (Pritchard *et al.,* 2009). One of the areas that holds great

prospect is the plant kingdom especially the use of plant seeds(e.g. *Moringaoleifera*seeds) in the treatment of water (Dalen *et al.,* 2009; Subramaniu*met al.,* 2011; Edogbanya*et al*., 2013a).

Adansonia digitata – Baobab (commonly known as "Kuka" in northern Nigeria) is a deciduous tree belonging to the Malvacaea family and is indigenous to arid central Africa (Yazzie *et al*., 1994; Bremer *et al.*, 2003). It is widely distributed and can be found in most of Sub-Sahara Africa's semi-arid and sub-humid regions as well as in western Madagascar (Diop*et al.*, 2005). Different parts of the tree are used as foods and medicines including the back fibres, no part of the tree is a waste (Igboeli*et al.,* 1997; Gebauer*et al*., 2002; Sidibe and Williams, 2002; De Caluwé*et al.,* 2010). This research attempts to evaluate the efficacy of seeds of *Adansonia digitata* as biocoagulant, disinfectant and biosorbent in water purification.

1.2 Statement of Research Problem

Water pollution has become a global phenomenon and a thing of concern across the nations of the world. It is estimated that about 748 million people, continue to rely on unimproved drinking water sources, of whom almost a quarter (173 million people) still rely on direct use of surface water (WHOand UNICEF, 2014).

Worldwide, infectious diseases such as waterborne diseases are the number one killer of children under five years old and more people die from unsafe water annually than from all forms of violence, including war (WHO, 2004).

The use of chemicals in the treatment of water is expensive, and is not readily available especially in the rural areas (Balandrin*et al.,* 1985; Muyibi and Alfugara, 2002).

Chemicals used in the treatment of water have been reported to be hazardous to man and his environment (Ghebremichael*et al.*,2005; Pritchard *et al.*, 2009).

1.3 Justification

Several plant seeds have been reported to contain coagulant proteins, which is responsible for their: biocoagulative, disinfectant, and biosorptive properties that enhances the purification of water (Muyibi and Alfugara*et al*., 2003; Amagloh and Benang*et al*.,2009 Oboh*et al*., 2008; Dalen *et al.*, 2009; Yongabi*et al*., 2011; Subramanium*et al.*, 2011).

The use of alternative technologies (such as the use of plants) in the purification of water has not been fully explored, and the findings of this work would give vital information that would contribute towards the solving of the problem of water pollution (especially in the rural areas).

1.4 Aim

The aim of this work is to study the efficacy of *Adansonia digitata*seeds as a natural sourcefor water purification.

1.5 Objectives

i. To determine the protein content of the seeds of *Adansonia digitata.*

ii. To evaluate the efficacy of the seeds of *Adansonia digitata* as a biocoagulant.

iii. To evaluate the efficacy of the seeds of *Adansonia digitata* as a disinfectant.

iv. To evaluate the efficacy of the seeds of *Adansonia digitata* as a biosorbent.

v. To determine the rates of adsorption of heavy metals by *Adansonia digitata* seeds.

1.6 Null Hypotheses

i. The seeds of *Adansonia digitata* do not contain significant amount of protein.

ii. The seeds of *Adansonia digitata* have no significant biocoagulative effect in water.

iii. The seeds of *Adansonia digitata* have no significant disinfectant effect in water.

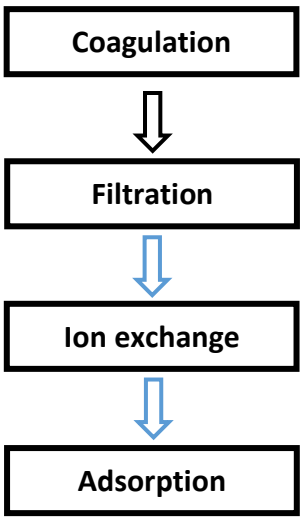
iv. The seeds of *Adansonia digitata* have no significant biosorptive effect in water.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Conventional Methods of Water Treatment

The treatment of water for drinking involves a number of combined processes based on the quality of the water source. Water is treated so as to reduce turbidity and the amount of microbial load present in it (Bodlund, 2013). Water suppliers use a variety of treatment processes to remove contaminants from drinking water(USEPA, 2004). These individual processes may be arranged in a "treatment train" (a series of processes applied in sequence). The most common processes include coagulation (flocculation and sedimentation), filtration, and disinfection used for surface water. Other treatment trains also include ion exchange and adsorption method as shown in figure 2.1(USEPA, 2004).



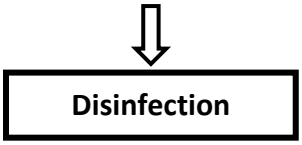
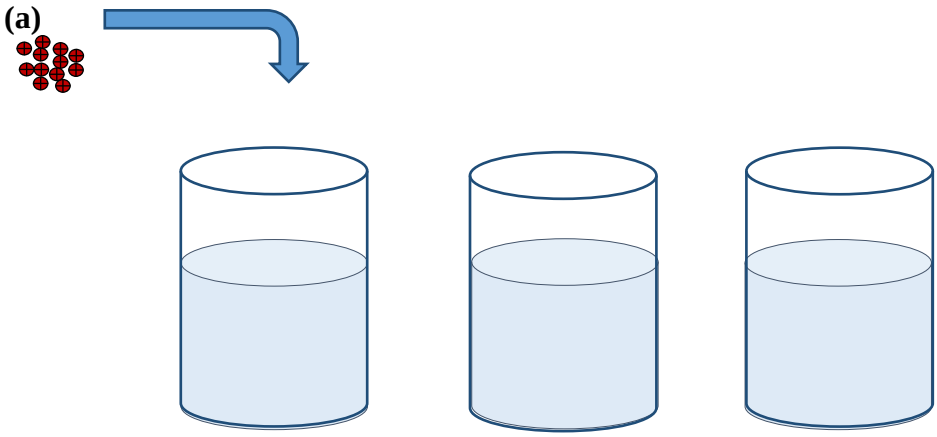
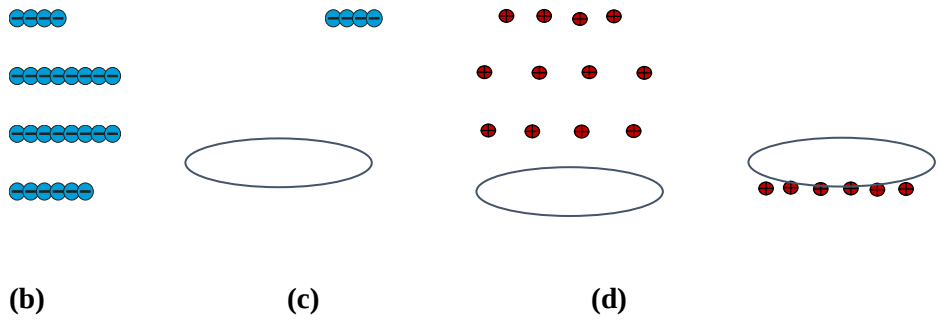


Figure 2.1: Schematic diagram of conventional methods of water purification. (Blue arrows indicate links that may be absent in some system units)

2.1.1 Coagulation (flocculation and sedimentation)

Coagulation is a process that involves the addition of coagulant (e.g alum) in water, which causes particles to aggregate into larger particles that can settle (USEPA, 2004). Turbidity is caused as a result of suspended particles and natural organic matter (NOM) present in the water. These particles are negatively charged and are therefore repelling each other, making it impossible for them to aggregate and settle (USEPA, 2004). The particles are carriers of unwanted contaminants and pathogenic organisms. In order to decrease the turbidity of water some positively charged chemicals (usually Alum and iron salts or synthetic organic polymers) are added(USEPA, 2004). This will destabilize the particles by neutralization of the negative charges. Flocculation is the agglomeration of these particles into large size particles known as flocs, which will settle by gravity (sedimentation). Turbidity of water is most commonly measured by turbidimeter and expressed in Nephelometric Turbidity Units (NTU) (Bodlund, 2013; USEPA, 2004). Figure 2.2 shows the process of coagulation and flocculation





(a) Coagulant (b) Suspended negatively charged suspended particles causing turbidity
(c) Flocs formation (d) Sludge formation (sedimentation)

Figure2.2 :Schematic diagram showing mechanism of coagulation

2.1.2 Filtration

Filtration clarifies water and enhances the effectiveness of disinfection. Many water treatment facilities use filtration to remove all particles from the water. Those particles include clays and silts, natural organic matter, precipitates from other treatment processes in the facility, iron and manganese, and microorganisms(USEPA, 2004).

2.1.3 Ion exchange

USEPA (2004) reported that ion exchange is a processes used for removing inorganic contaminants if they cannot be removed adequately bysedimentation or filtration. It is a process that can be used in the treatment of hard water. It can also be used in the removal of arsenic, chromium, excess fluoride, nitrates, radium, and uranium etc.

2.1.4 Adsorption

Adsorption is a process that involves the addition of an adsorbent (e.g activated carbon) in water for the removal of organic contaminants, unwanted colouring, and taste-and-odour-causing compounds. This compounds stick to the surface of granular or powder activated carbon and are thus removed from the drinking water (USEPA, 2004).

2.1.5 Disinfection

Water is usually disinfected before it is released into the distribution system to ensure that potentially dangerous microbes are killed (USEPA, 2004). Chlorine, chloramines, or chlorine dioxide are mostly used because they are very effective disinfectants (USEPA, 2004), not only at the treatment plant but also in the pipes that distribute water to our homes and businesses, since they have residual effects (USEPA,2004). Ozone is also a powerful disinfectant, and ultraviolet radiation is an effective disinfectant and treatment for relatively clean source waters, but neither of these are effective in controlling biological contaminants in the distribution pipes, and they are expensive methods (USEPA, 2004).

2.2 Plant Seeds as Biocoagulants and Disinfectants

A number of plant seedshave been reported to have potentials to serve as biocoagulants and disinfectants in the purification of water (Edogbanya*et al.*,2013a). Women in rural areas of Sudan (Jahn and Dirar, 1979), Tanzania (Marobhe*et al.*,2007) and India (Saif*et al.,* 2012) treat their water with Moringa seed powder prior to use as drinking water. Amagloh and Benang (2009) reported that a concentration of *Moringaoleifera*powder of 12.0 g/1000 ml loading dose as coagulant gives similar effect on turbidity compared with alum of loading doses of 10.0 g/1000 ml and 12.0 g/1000 ml. Pritchard *et al.* (2010) reported that *Moringaoleifera* removed 84% turbidity of water

initially at 146 NTU. Attempts have been made to characterize and purify the active biocoagulative protein of *Moringaoleifera* and considerable success have been achieved (Ghebremichael, 2005). Apart from the turbidity removal properties, *Moringaoleifera*has been reported to have antimicrobial properties in water (Amagloh and Benang,2009;Choubey*et al*, 2012; Pritchard *et al*, 2011;Yongabi *et al.*,2011).Diaz *et al.*(1999) reported that *Prosopisjulifloras*eed extract was able to produce a final water whose turbidity was close to the required standard of 5NTU with both high (100–200 NTU) and low (30–40 NTU) initial turbidities.Choubey*et al.* (2012) reported that turbidity reduced up to 95.89% for highly turbid water which is almost as same as the reduction capacity of alum. *Cicerarientinum*was also found to possess antimicrobial properties (Choubey*et al.*, 2012). Subramaniu*met al.* (2011) reported that seed extract of *Phaseolus vulgaris and Zea mays* could reduce turbidity level from 250 NTU to 100 and 55NTU respectively, after 24hrs standing period. Marobhe (2007) reported that fine (0.200mm<Grain size<0.212mm)and the large grain size(0.425mm<Grain size <0.850mm) of *Parkinsoniaaculeata,Vignaunguiculata, Voandzeiasubterranea*seed powder, were found to reduce turbidity of water from 205NTU to 19 and 20NTU, 23 and 26NTU, and 21 and 25NTU respectively; the seeds' extracts were also found to possess antimicrobial properties in water. Saefudin*et al*.(2012) reported that at a concentration of 0.1% over 98.5% turbidity removal was achieved using ***Viciafaba* seed extract.** Dalziel (1973) reported the coagulative and disinfectant property of *Parkiabiglobossa*.Yongabi*et al.* (2011) reported that the seed extract of *Perseaamericana, Garcinia kola, Carica papaya* were able to reduce the turbidity of water gotten from three different streams in Cameroon from 27, 33, and 117 NTU to 13.2, 14.11 and 42.6 NTU; 7.11, 11.3 and 20 NTU and 9.9,

9.4 and 11.9 NTU respectively. The seed extract of these plants were also reported to have antimicrobial properties (Yongabi*et al.,* 2011). Yongabi*et al*.(2012) reported that seed extracts of *Hibiscus sabdariffa* and *Jartrophacurcas* had both biocoagulative and disinfectant properties.

2.2.1 Mechanism of action of seeds as biocoagulants and disinfectants

The chemical compositions of the active coagulating agent of seeds like *Moringaoleifera* have been debated. Several researchers have described the active component from a water extract as cationic and proteinaceous (Ndabigengesere*et al*.,1995; Muyibi and Alfugara,2003). However a few have argued that the active component from a salt extract responsible for coagulation is an organic polyelectrolyte that is neither protein nor polysaccharide (Okuda *et al.*,2001). Researchers have suggested the coagulation mechanism for the water extracted active agent is adsorption and charge neutralization. The antimicrobial (disinfectant) property of seeds have continued to be researched. Findings support recombinant proteins both removing microorganisms by coagulation as well as acting directly as growth inhibitors of the microorganisms (Doerr, 2005).

2.2.2 Advantages of the use ofbiocoagulants and disinfectants from plant seeds

Compared to the commonly used chemicals for coagulation and disinfection, seeds have a number of advantages (*Ghebremichael, 2004)*:

- It is of low cost.
- Produces biodegradable sludge.
- Produces lower sludge volume.

- Does not significantly affect the pH of the water.
- Has little or no health implications.

2.2.3 Disadvantages of the use of seeds as biocoagulants and disinfectants

The use of seeds as biocoagulantsand disinfectants also has some disadvantages(Ndabigengesere*et al*., 1995; Okuda *et al.*, 2001; *Ghebremichael, 2004*).

- Most of the coagulation and antimicrobial studies of seed extracts are based on laboratory scale experiments and household level applications.
- The major limitation in using crude seed extracts for large-scale water treatment application is the release of organic matter and nutrients (nitrate and phosphate) to the water and unfortunately many researches do not give account of the dissolved organic matter left behind after seeds are used. The organic and nutrient released from the seed can be avoided either by purifying the coagulant component or by removing the released substances from the water.

2.3 Plant Seeds as Biosorbents

Biosorption is simply a kind of adsorption involving adsorbates which are biological in origin – commonly called biosorbents (Gavrilescu, 2004). Several plant seeds have been reported to have the potential to serve as biosorbents(Edogbanya*et al.*,2013b).

Kumari*et al.* (2006) reported that seeds of *Moringaoleifera* made recoveries of up to 60.21% for As (III) and 85.60% for As (V); Sharma *et al.*(2007) reported 76.59% for Cd (II), 68.85% for Cr(III) and 60.52% for Ni (II); Mancy*et al.*,(2013) reported 88% for Cr(VI) and 69% for Fe(III) , Meneghel*et al.*(2013) reported a maximum adsorbance(q_m) of

7.864 mgg^{-1} for Cd(II) at optimum conditions. Egila*et al.*(2011) reported that seeds of *Carica papaya* had a maximum adsorbance of 4.1 mg/100 cm^3 and 3.63 mg/100 cm^3 for Mn (II) and Pb (II) respectively at 33°C and contact time of 90mins; Hadi*et al.* (2011) reported that seeds had a maximum adsorption capacity (q_m) of 212 mgg^{-1} for Cu (II). Oboh andAluyor (2008)reported that sour sop seeds achieved the removal of 77.6, 68.5, 56.4and 40.6% for Cu(II) , Ni(II), Zn(II) and Pb(II) respectively after a contact time of 120min. Lima *et al.*, (2007) reported that *Araucaria angustifolia*seed wastes, natural and modified(with Congo Red), had a maximum adsorbance (q_m) of 32.2 and 68.6 mgg^{-1} of Cu(II) respectively at optimum conditions. Zamani*et al.*(2013) reported that *Peganumharmala*seedsadsorbed about 95 % of Pb(II), 75 % of Zn(II) and 90 % of Cd(II) from 45 ml of aqueous solution containing 20mgl-^{1} of each cation with 2 g of adsorbent at pH 4.5 after 15 min. Sen (2012) reported that *Jartrophacurcas* seed hull had a maximunadsorbance (q_m) of 11.8906mgg^{-1} for Cd(II). Gupte*et al.*(2012) reported that up to 90 % of Cu(II) was adsorbed by 0.1 g of soaked seeds of *Ocimumbasilicum* within 2.5 hr. Mancy*et al.*(2013) reported that a removal of about 94.6 % and 70% for Cr (VI) and Fe (III) respectively were observed using *Tamarindusindica* seeds as biosorbent, at pH 4 and temperature of 311K. Yoa*et al.*(2010) reported that the adsorption capacity(q_m) of chestnut (*Castaneamollissima*) shell for Cu(II) was determined with the Langmuir model and was found to be 12.56mgg^{-1} at 293K.

Mancy*et al.*(2013) reported that *Azadiracthaindica*seeds had recoveries of up to about 70% and 58% for Cr(IV) and Fe(III) respectively at pH 4 and temperature of 311K; while Stanley *et al.*(2013) reported 57.6 % and 35% for Cr(IV) and Fe(III) at same conditions. Gohulavani*et al.*(2013)reported that *Azadiracthaindica*seed shells had a maximum

maximum adsorption capacity of 66.66 mgg-^{1} for Ni(II). Raj *et al.* (2013) reported that natural *Leucaenaleucocephala* seeds used as biosorbent had a removal capacity of 81.88% and 92.61 %, for As (III) and As (V) respectively, while when modified with polyethylenimine the removal capacity increased to 85% and 99 %, for As (III) and As (V) respectively. Ahalya*et al.*, (2005) reported that dried seed (gram husk) of *Cicerarietinum* showed 99.9% removal of Cr(VI); Kale (2012)reported that the maximum adsorption (q_m) of 5g of sulphonated*Cicerarietinum* biomass was 74% for Pb(II). Omorogie*et al.*(2012) reported that at a temperature of 333 K, the highest amount of metal biosorbed (q_m) for Cd(II) and Hg(II) obtained were 6.30 and 6.15 mgg^{-1} respectively.

Jimoh*et al.*(2013) reported that using *Citrus sinensis* (Sweet orange)seeds as biosorbent maximum removal of Pb(II) and Zn(II) occurred at pH 6 with 98.69% and 99.82% respectively while that of Cd(II) ion was found to be 98.61% at pH 7. Stanley *et al.*(2013) reported that *Strychnospotatorum* shows removal efficiency of 69% of Chromium (VI) and 58% of Iron (III) at acidic pH and 0.1 mg L^{-1} of initial metal ion concentration.

2.3.1 Mechanism of action of seeds as biosorbents

For agricultural products (such as plants seeds), the mode of sorption can be attributed to two main terms; intrinsic adsorption and coulombic interaction (Gang and Weixing, 1998). The coulombic term results from the electrostatic energy of interactions between the adsorbents and adsorbates, which results in affinity of the sorbent for the sorbate. The process continues until equilibrium is established between the amount of sorbate bound to the sorbent and the portion remaining in the solution. The degree of sorbent affinity for the sorbate determines its distribution between the solid phase and liquid phase (Das *et al.*

2008). The charges on substrates as well as softness or hardness of charge on both sides are mostly responsible for the intensity of the interaction. Coulombic interaction can be observed from the adsorption of cationic species versus anionic species on adsorbents (Gang *et al.* 1998). The intrinsic adsorption of the materials is determined by their surface areas, which can be observed by the effect of different sizes of adsorbent on adsorption capacity (Igwe and Abia, 2005).

2.3.2 Advantage of the use of seeds as biosorbents

Biosorption have several advantages over conventional techniques (Igwe and Abia, 2006; Singh *et al.*, 2014). They include:

- Cheap: the cost of the biosorbent is low since they often are made from abundant or waste material.
- High uptake capacity: they have the ability to adsorb high quantity of Heavy metals.
- Metal selective: the metal sorbing performance of different types of biomass can be more or less selective on different metals. This depends on various factors such as type of biomass, mixture in the solution, type of biomass preparation and physicochemical treatment.
- Regenerative: biosorbents can be reused, after the metal is recycled.
- No sludge generation: no secondary problems with sludge occur with biosorption, as is the case with many other techniques, for example, precipitation.
- Metal recovery possible: In case of metals, it can be recovered after being sorbed from the solution.

- Competitive performance: biosorption is capable of a performance comparable to the most similar technique, e.g. ion exchange treatment.

2.3.3 Disadvantages of the use of seeds as biosorbents

In spite of the advantages of biosorption it also have some disadvantages (Ahluwulia and Goyal,1998 ; Das *et al.,* 2008). They include:

- Early saturation i.e. when metal interactive sites are occupied, metal desorption is necessary prior to further use.
- The potential for biological process improvement (e.g. through genetic engineering of cells) is limited because cells are not metabolizing.
- There is no potential for biologically altering the metal valency state.
- Release of organic matter and nutrients into the water.
- Most of the biosorption of seed extracts are based on laboratory scale experiments.

2.4 Kinetic Models for Biosorption

The mechanism of adsorption and its potential rate-controlling steps that include mass transport and chemical reaction processes is usually investigated using kinetic models. In addition, information on the kinetics of metal uptake is required to select the optimum condition for full-scale batch metal removal processes (Febrianto*et al*., 2009).

Adsorption kinetics is expressed as the rate of solute removal that controls the residence time of the sorbate in the solid–solution interface. In practice, kinetic studies are carried out in batch reactions using various initial sorbate concentrations, sorbent doses, particle sizes, agitation speeds, pH values and temperatures along with different sorbent and

sorbate types. Then, linear regression is used to determine the best-fitting kinetic rate equation. There are several kinetic models but the most commonly used are the pseudo first and second order rate model in biosorption models(Febrianto*et al*., 2009).

2.4.1 The Pseudo-first-order kinetic model

The Lagergren first-order rate expression based on solid capacity is generally expressed as equation 2.1:

$$\frac{dq}{dt} = k_1\ (q_e - q) \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (2.1)$$

It can also be expressed in the linearized form as in equation 2.2:

$$\log\ (q_e\ - q)\ = \log\ (q_e\ ) - \frac{k_1\, t}{2.303} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (2.2)$$

Where :

q_e (mg g^{-1}) = the solid phase concentration of the metal ions at equilibrium,

q (mg g^{-1}) = the average solid phase concentration of metal ions at contact time t (min) and

k_1 (min^{-1}) = the pseudo-first-order rate constant.

The rate constant k_1 (min^{-1}) could be obtained from the plot of log ($q_e - q$) versus t (Febrianto*et al*., 2009; Njoku, 2011).

2.4.2 The Pseudo-second-order kinetic model

The Ho Pseudo-second-order Kinetic Model is derived on the basis of the sorption capacity of the solid phase and is expressed as in equation 2.3:

$$\frac{dq}{dt} = k_1\ (q_e - q)^2 \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (2.3)$$

It can also be expressed in the linearized form as in equation 2.4:

$$\frac{t}{q} = \frac{1}{k_2\ {q_e}^2} + \frac{1}{q_e}\ t \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (2.4)$$

Where $k_2$is the pseudo-second-order rate constant.

a plot of t/q versus t gives a straight line and the values of the constants k_2 (g mg^{-1}) and q_e (mg g^{-1}) can be calculated (Febrianto*et al*., 2009; Njoku, 2011).

2.5*Adansonia digitata*L.

2.5.1 Classification

Kingdom: Plantae

Division: Magnoliphyta

Class: Equisetopsida

Subclass: Magnoliidae

Superorder: Rosana

Order: Malvales

Family: Malvaceae

Genus : Adansonia

Species: *Adansonia digitata*

(Royal Botanical Gardens, Kew.)

2.5.2 General description

Adansonia digitata is an imposing large tree reaching heights of about 18-25m and producing a rounded crown showing a stiff branching habit (Plate I). It has a characteristic swollen trunk ofup to 10m in diameter, usually tapering or cylindrical and abruptly bottle-shaped; often buttressed. Giant individuals can reach a girth of up to 28m. Branches are distributed irregularly and large; primary branches may be well distributed along the trunk or limited to the apex; young branches are somewhat tomentose but rarely glabrous. The bark is smooth, reddish brown to grey, soft and fibrous. The bark of leaf-bearing branches is normally ashy on the last node. There is a green layer below the outer layer of the bark presumed to photosynthesise when the tree has shed its leaves(Sidibe and Williams, 2002).

The tree produces an extensive lateral root system and the roots end in tubers.Seedlings produce a strong prominent taproot but this is soon replaced by laterals. Roots of mature trees rarely extend beyond 2m and are relatively shallow: one reason explaining why trees are often toppled in old age (Sidibe and Williams, 2002).

Leaves are usually scanty on branches they are 2-3-foliate at the start of the season and they are early deciduous, more mature ones are 5-7(-9)-foliate. Leaves are alternate at the ends of branches or occur on short spurs on the trunk. Leaves of young trees are often simple. Leaflets are sessile to shortly petiolulate, with great variation in size (Sidibe and Williams, 2002). .

Flowers are pendulous, solitary or paired in leaf axils, large and showy and produced during both wet and dry seasons. Pedicels usually vary greatly in length, 15-90cm, with 2 small, caducous bracteoles near the apex of the pedical (Sidibe and Williams, 2002).

Fruits are very variable, they are also pendulous, usually globose to ovoid but sometimes oblong cylindrical, often irregular in shape, 7.5-54cm long x 7.5-20cm wide, apexpointed or obtuse, covered by velvety yellowish to greenish hairs (plate) (Sidibe and Williams, 2002). The hair has a characteristic itch on the skin. The mature fruits have a white pulp containing numerous seeds which are bean shaped with a hard brownish shell (Plate II, III and IV). Actually the meaning of baobab is "fruits of many seeds"(Ajayi*et al.*, 2003).

2.5.3 Distribution

African Baobab occurs naturally in most countries south of the Sahara with notable absence in Liberia, Uganda, Djibouti and Burundi. In some of these countries its

distribution is limited, e.g. in Chad, where it is not found in the east, and South Africa where it is mostly limited to the Transvaal. Essentially, baobab is associated with the savannah, especially the drier parts. However, there are extensions of the distribution into forest areas, probably associated with human habitation. It appears to be introduced into more equatorial areas, such as Gabon, Democratic Congo and Zaire, and to countries with a marked dry season such as São Tomé, Madagascar and Comoros (Sidibe and Williams, 2002).

2.5.4 Uses

The Baobab tree is a tree of multiple uses and it is said that all part of the tree is useful. Generally it is used as food, for providing emergency water, fibres and medicines (Igboeli*et al.,* 1997; Gebauer*et al*., 2002; Sidibe and Williams, 2002; De Caluwé*et al.,* 2010).

2.5.4.1 Food uses

Leaves

The fresh young leaves are used as a staple food in many parts of sub Saharan Africa. Young leaves are widely used, cooked as spinach, and frequently dried, often powdered and used for sauces over porridges, thick gruels of grains, or boiled rice (Sidibe andWilliams, 2002). In northern Nigeria the dried leaves are dried, pounded and used in making a special soup called "MianKuka" which is used in eating "Tuwo" (Corn food). The leaves contain (expressed on dry weight basis):13–15% protein, 60–70%

carbohydrate, 4–10% fat and around 11% fibre and 16% ash (Sidibe and Williams, 2002;De Caluwé*et al.*,2010).

Fruit pulp

The dry pulp is either eaten fresh or used to add to gruels on cooling after cooking. The cattle-owning Fulani and the Hausa of northern Nigeria use the fruit pulp emulsion to mix with milk as a drink (Sidibe and Williams, 2002) (this is sometimes sold as adulterated cow milk known as "nonokarya"). In Tanzania, it is added to aid fermentation of sugar cane for beer making (Fleuret, 1980).

The fruit pulp contains a high amount of carbohydrate, low protein, and extremely low fat (Osman, 2004). It is also high in vitamin C, almost ten times that of oranges (De Caluwé*et al.*, 2010).

Seeds

Generally seeds are used as a thickening agent in soups, but they can be fermented and used as a flavouring agent, or roasted and eaten as snacks (Palmer and Pitman, 1972; Addy and Eteshola, 1984). When roasted, they are sometimes used as a substitute for coffee (Sidibe and Williams, 2002).

Seeds are also a source of cooking oil but this is not widespread, although there has been interest in expanding such use due to deficits of vegetable oils. Oil is extracted by pounding the seeds (Sidibe and Williams, 2002).

The seed contains relatively high amounts of protein, crude fat, and crude fibre, and low levels of carbohydrates (Osman, 2004).

2.5.4.2 Domestic non-food uses, other than medicinal

Fibre Fibre from the inner bark is strong and generally used for making rope, basket nets, snares, fishing lines and is even used for weaving. Fibres are also available from disintegrated wood and have been used for packing. Other fibres used for rope are obtained from root bark (Sidibe and Williams, 2002).

Dye In East Africa the roots are used to make a soluble red dye. The green bark is also used as a dye and for decoration (Dovie, 2003).

Seed shell

The hard fruit shells are used in the manufacture of pots for food and drink (Dovie*et al.*, 2001).

Fuel

The wood is a poor source of fuel; however, fruit shells are used as fuel in Tanzania and they are used as water dippers (Nkana and Iddi, 1991 cited in Sidibe and Williams, 2002).

Animal browse and feed

The leaves, shells from the fruit, and seedcake left after extraction of seed oil, are usually fed to livestock (Sidibe and Williams, 2002).

2.5.4.3 Medicinal uses

- Baobab is used in traditional medicine as an antipyretic or febrifuge to overcome fevers. Both leaves and fruit pulp are used for this purpose.

- Fruit pulp and powdered seeds are used in cases of dysentery and to promote perspiration (i.e. a diaphoretic).
- Powdered leaves can be used as an anti-asthmatic and they are known to have antihistamine and antitension properties. They are variously used to treat fatigue, as a tonic and for insect bites, guinea worm and internal pains, and to treat dysentery.
- Leaves are used for many other conditions: diseases of the urinary tract, opthalmia and otitis.
- Seeds are also used in cases of diarrhoea, and hiccough.
- Oil extracted from seeds is used for inflamed gums and to ease diseased teeth.
- Bark, fruit pulp and seeds appear to contain an antidote to poisoning by Strophanthus species (Sidibe and Williams, 2002).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Collection, Identification and Preparation of Samples

All samples for this study were collected within Ahmadu Bello University (ABU), Zaria. ABU is located in the northern savannah region of kaduna state of Nigeria.The Main

Campus of Ahmadu Bello University is located in Samaru, a suburb of Zaria in Kaduna State, Nigeria. Samaru is situated on latitude 112^{o} 12" N and longitude 07^{o} 37" E, at an altitude of 550-700 meters as reported by ABU (2013).

3.1.1 Collection ofwater samples

Surface water sample from the River Kubanni Reservoir, Ahmadu Bello University, Zaria, using proethylene containers at Latitude 11°8′4.39″ N, Longitude 7°39′26.24″ E. The actual location of collection was determined using Global Positioning System (GPS). The first 50 - 100 mL of sample was used to rinse the containers first before the required volume was collected. Sample containers used for biosorption studies were cleaned by immersing them in nitric acid (5% v/v) overnight followed by thorough washing with distilled water (Shrivastava*et al.,* 2013). The water sample used for biosorption studies were preserved using 3ml of (1+1) Nitric acid per Litre of sample. All samples were refrigerated until ready for analysis (Creed and Martins, 1992).

3.1.2 Collection, identification and preparation of plant materials.

Dried fruits of *Adansonia digitata* (Plate II) were collected from a single tree (Plate I) in the Botanical Garden of the Department of Biological Sciences, Ahmadu Bello University, Zaria, and were taken to theherbarium of the Department for verification and confirmation using the herbarium voucher number. For further and proper picture of the plant/parts were snapped and presented as pictographs.

TheThe fruits were broke open (Plate III) by hitting them on a hard surface and seeds (Plate III) were mechanically removed with hands, properly washed with distilled water, sun dried, pulverized into powder (Plate V) using mortar and pestle, sieved through a pore size of about 1mm, and stored in airtight containers. Pictures were also taken using digital camera.

3.1.3 Determination of protein content

Crude protein was estimated using the Kjeldahl method (AOAC, 1980). This was carried out in the Food Science and Technology Unit of the Institute for Agricultural Research (IAR), Ahmadu Bello University, Zaria.

3.1.3.1Digestion

2g of powdered *Adansonia digitata* seed was weighed and put into Kjeldalh flask. One tablet of catalyst (Copper) and 25ml of Concentrated H_2SO_4 were added to the sample. The sample was heated in the fume cupboard till the solution assumed a green colour. The solution was allowed to cool and the black particles at the neck of the flask were washed down back into the solution. After cooling the digest was transferred into a 250ml conical flask with several washings with distilled water.

3.1.3.2 Distillation

The distillation was done using the markham distillation apparatus. A 100ml conical flask containing 5ml of boric indicator was placed under the condenser. 5ml of the digest into the body of the apparatus via the small funnel apparture, and washed down using distilled water followed by 5ml of 60% NaOH solution. The apparatus was allowed to stem through

for about 5-7min to collect enough ammonium sulphate. The receiving flask was then removed and the tip of the condenser was washed down into the flask.

3.1.3.3Titration

The solution in the receiving flask was titrated using 0.01 N Hydrochloric acids until the solution turns light pink which indicated the end point. The titre value was then taken. A blank solution was also run along with the sample. The calculations are carried out using equations (3.1) and (3.2).

Calculations:

$$\%\ Nitrogen = \frac{1.4007\ \times (\ Titre\ value\ for\ sample - Titre\ value\ for\ blank) \times Normality\ of\ HCl}{Weight\ of\ sample} \quad \ldots (3.1)$$

To convert % Nitrogen to % Protein is given by:

$$\%\ Protein = \%\ \text{Nitrogen}(6.25) \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (3.2)$$

3.3 Biocoagulation Studies

3.3.1 Preparation of model turbid water

Turbid water was prepared by adding 10g beneficiated kaolin to 1L tap water. The suspension was stirred for 1h to achieve uniform dispersion of kaolin particles, and then it was allowed to remain for 24 h for completing hydration of the particles. This suspension was used as the stock. Turbid water used for the study was prepared by diluting 50ml of

stock solution in 1000ml of water(This gave an initial turbidity of about 200 nephelometric turbidity units –NTU) (Okuda *et al.,* 2001; Antov*et al.,* 2010)

3.3.2 Extraction of active component from *Adansonia digitata* seeds

The powdered seed sample (*Adansonia digitata* seeds) was de-fatted (Plate VI) using n-hexane in electro-thermal Soxhlet extractor. 30g of powdered seeds was weighed in put into the thimble of the soxlet extractor, the apparatus was mounted and allowed to run for 1 hour, after which the powder is removed and dried over a hot plate at low heat to evaporate the n-hexane. The *Adansonia digitata* cake residue after oil extraction was used in the preparation of the extract. Four different concentrations (50mg/l, 100mg/l, 150mg/l and 200mg/l) of the coagulant reagent were prepared by suspending the required weighed amount of de-fatted powdered seed in 1L of distilled water, which was then stirred for 15 minutes using the flocculator(PCI Ltd England) in order to extract active components. After that, the suspension was filtered through Whatt-man filter paper No. 1 (Bichi*et al.,*2012 ; Choubey*et al.,* 2012). Before each experiment a fresh coagulant regent was prepared, because keeping it overnight reduced the effectiveness.

3.3.3 Jar tests experiment

The Jar test experiment was carried out using a flocculator (PCI Ltd England). The different doses of coagulant reagent (50mg/l, 100mg/l, 150mg/l and 200mg/l) were added to different beakers of the apparatus containing 1L of model turbid water, except to one which which served as the control. Mixing of the coagulant with water was provided by rapid mixing at 100rev/min for approximately 3min followed by slow mixing at 30 rev/min for approximately 17 min. The propellers were stopped and the content of the jars left to

settle for approximately 30 min. After sedimentation, the supernatants were decanted for water quality determination (Arnoldsson*et al*., 2008). The experiment was carried out in triplicate. The same procedures were also carried out for the surface water sample (gotten from KubanniRerservoir.

3.3.4 Measurements of physicochemical parameters

Turbidity, Electrical conductivity (EC), Total Dissolved Solids (TDS), pH, Total Suspended Solids (TSS), alkalinity, hardness and Chemical Oxygen Demand (COD) , were measured according to standard methods for the examination of water and waste water(APHA, AWWA and WPCF, 1985).

3.3.4.1 Turbidity

The turbidity was determined using a turbidimeter (Hach 2100N). Each water sample was poured into the glass cell of the turbidimeter up to the mark, the cell was then inserted into the turbidimeter, and the readings were allowed to stabilize before they were taken.

3.4.4.2 Electronic Conductivity (EC) and Total Dissolved Solid (TDS)

The E.C and T.D.S were taken using the Hanna digital meter (HI 9835). The instrument was set to the appropriate parameter to be taken. 100ml of the water samples were poured into a large test tube of 200ml, the probe of the instrument was inserted, and readings were then allowed to stabilize before they were taken.

3.4.4.3pH

The pH was taken using the Hanna multiphotometer (HI 8320). The instrument was set to the appropriate parameter (pH), 10ml of water sample was poured into the glass cell, 10 drops of phenol red indicator was added, the cell was inserted into the instrument, and readings were allowed to stabilize before taken.

3.4.4.4 Total Suspended Solids (TSS)

Whattman 1 filter paper was folded and fitted into a funnel, and the funnel was inserted into a conical flask. 100ml of distilled water was poured and allowed to drain through the filter paper. The filter papers were carefully removed from the funnel using forceps, and put in an oven at 105°C and allowed to dry for an hour. After that the filter paper was then removed, allowed to cool in a dessicator, and weighed. The filter paper was then replaced in funnel and 100ml of properly shook water sample was poured in and allowed to drain again. After draining the filter paper was carefully removed, put in the oven at 105°C for another one hour, removed and allowed to cool, then weighed again.

3.4.4.5 Total alkalinity

100ml of well shook sample was measured and poured into a conical flask and 3 drops of phenolphthalein and 3 drops of methyl orange indicators were added (this gave an orange colouration). 0.02N sulphuric acid from a burette was used to titrate the sample until the yellow colouration turned orange (indicating the end point).

Calculation:

Total alkalinity was calculated using equation 3.3:

$$Total\ Alkalinity(mg/l\ CaCO_3) = \frac{B \times N \times 50{,}000}{ml\ of\ sample} \ldots\ldots\ldots\ldots\ldots (3.3)$$

Where:

B = Titre value for sample

N =Normality of acid (0.02N).

3.4.4.6 Hardness

25ml of well shook sample was measured into a conical flask and made up to 50ml with distilled water. 2ml of buffer solution and two drops of Eriochrome black indicator was added to sample (this gave a purple colouration). The sample was titrated with standard solution of EDTAfrom a burette until a deep blue colouration was attained (indicating the end point).

Calculation:

Total Hardness calculated using equation 3.4:

$$Total\ Hardness(mg/l\ CaCO_3) = \frac{A \times B \times 1000}{ml\ of\ sample} \ldots\ldots\ldots\ldots (3.4)$$

Where:

A = ml of titrant value

B= mg of $CaCO_3$ equivalent to 1.00ml EDTA titrant

3.4.4.7 Chemical Oxygen Demand (COD)

0.4g of $HgSO_4$ was measured into a refluxing flask. 20ml of well shook sample was added and mixed. 10ml standard $K_2Cr_2O_7$ solution and several pieces of glass beads were also added to the mixture. 30ml of concentrated H_2SO_4 containing Ag_2SO_4 was added and the flask and swirled gently. The flask was attached to the reflux condenser and the mixture was refluxed for 1h. The mixture was diluted to 100ml with distilled water and allowed to cool to room temperature. 3 drops of ferrion indicator was added to mixture. The mixture was then titrated with $Fe(NH_4)_2(SO_4)_2$ and a colour change was observed from blue to green to reddish brown(indicating the end point). A blank containing 20ml of distilled water and reagents was also reflux in same manner.

Calculation:

The COD is calculated using equation 3.5:

$$COD(mg/l) = \frac{(A-B)N \times 1000}{ml\ of\ sample} \ldots\ldots\ldots\ldots\ldots (3.5)$$

Where:

COD = COD from dichromate

A= Titre value for blank in ml

B= Titre value for sample in ml

N= Normality of $Fe(NH_4)_2(SO_4)_2$ = 0.25

The biocoagulation studies was carried out in the Sanitary Chemistry Laboratory of the Department of Water Resources and Environmental Engineering, Faculty of Engineering, Ahmadu Bello University, Zaria.

3.5 Disinfection Studies

3.5.1 Preparation of *Escherichia coli* culture

Nutrient broth (13g) was dissolved in 1000 ml of distilled water by heating slightly. The mixture was sterilized at 130°C for 15 min in autoclave. The sterilized broth was cooled to room temperature and was used to prepare the *E. coli* culture. Clinical isolates of *E. coli* was obtained from the Department of Pharmaceutics and Pharmaceutical Microbiology, Faculty of Pharmaceutical Sciences,Ahmadu Bello University, Zaria, and was grown in 10 ml nutrient broth at 37°C overnight to obtain an exponential growth phase. The culture was standardized to 1:5000 using normal saline and this was used as the model water for the disinfection experiment (Obire*et al.*, 2005; Bichi*et al.,* 2012).

3.5.2 Disinfection experiment

Four different doses (50mg/l, 100mg/l, 150mg/l and 200mg/l) of the extract was added to four different test tubes containing 10 ml of the *E. coli* suspension and incubated for 2hr without agitation. The various cell survivals was assessed by making dilution series of bacterial suspensions, plating on Mueller Hinton agar dishes, and incubated for 24 h at

37°C. Triplicates were made of every individual assay. Colonies were counted on dishes and the cell survival ratio was estimated by comparison to a control experiment where no extract was added. The same procedure was carried out using the surface water sample (Suarez *et al.,* 2003; Bichi*et al.*, 2012).

The coliform forming unit per ml (cfu/ml) was calculated using equation 3.6:

$$\frac{cfu}{ml} = \frac{Number\ \ of\ Colony}{Dilution\ factor\ \ \times\ \ ml} \ldots\ldots\ldots\ldots (3.6)$$

The biocoagulation and disinfection studies was carried out in the Microbiology Labouratory of the Department of Pharmaceutics and Pharmaceutical Microbiology, Faculty of Pharmaceutical Sciences Ahmadu Bello University, Zaria.

3.6 Adsorption Studies

3.6.1 Preparation of stock solutions of heavy metals (Lead and Cadmium)

All chemicals that were used was of analytical grade. Standard stock solution (30 mg/l) of the nitrate salts of each metal was prepared by weighing 30mg of the salts and dissolving in 1L of deionized water (Sen, 2012). Further dilutions were made from the stock solutions for the study.

3.6.2 Batch adsorption experiments

The effect of pH (2 – 10) (pH was adjusted using Hcl and NaOH), initial concentration of metal salts (5, 10, 20, 30 mg/L), dosage of biosorbent (50mg/l, 100mg/l, 150mg/l and 200mg/l), and contact time (5, 15, 30, 60 min) on adsorption was studied by varying the

parameter of concern and making all others constant. In all the experiments 100ml of metal solution of known concentration was taken in polyethylene bottles along with the required amount of biosorbent (de-fatted powdered *Adansonia digitata*). The bottles were covered properly and agitated with a laboratory shaker (Gallenkamp England) for a particular period of time corresponding to the desired contact time to achieve equilibrium. After agitation the mixture was passed through filter papers (Whattman No. 1) and stored in polyethylene sample bottles. The residue concentrationsof lead and cadmium were determined using Atomic Absorption Spectrophotometer (AA 240 FS Varian).The experiments were carried out in triplicates with their respective controls (Njoku, 2011 ; Sen, 2012; Alam*et al.,* 2012).Theinitial amount of lead and Cadmium weredetermined in the surface water sample and the same procedures werecarried out to determine the rate of adsorption using the various doses of the biosorbent.

The amount of biosorption of heavy metal ions on the biosorbent at equilibrium, $q_e(mgg^{-1})$ was calculated using equation 3.7:

$$q_e = \frac{(C_o - C_e)V}{W} \dots\dots\dots\dots\dots\dots (3.7)$$

Where C_o and C_e (mgL^{-1}) are the liquid-phase concentration of metal ions at initial and equilibrium, respectively, V (L) is the volume of the solution and W (g) is the mass of dry biosorbent (Rajjak and Bapurao, 2012; Mahajan and Sud, 2013)

The removal efficiency R_e (%) of heavy metal ions was calculated using:

$$R_e = \frac{(C_o - C_e)}{C_o} \times 100 \dots\dots\dots\dots\dots\dots. (3.8)$$

3.6.3 Kinetic studies

The Lagergren Pseudo-First-Order and Ho Pseudo-Second-Order Kinetic models was used for this study to determine the mechanism of adsorption.linearized Pseudo-First-Order kinetic model is expressed in equation 3.9:

$$\log (q_e - q) = \log (q_e) - \frac{k_1 t}{2.303} \ldots\ldots\ldots (3.9)$$

Where qe (mg g^{-1}) is the solid phase concentration of the metal ions at equilibrium, q (mg g^{-1}) the average solid phase concentration of metal ions at contact time t (min) and k_1 (min^{-1}) is the pseudo-first-order rate constant.

The rate constant k_1 (min^{-1}) could be obtained from the plot of $\log(q_e - q)$ versus t.

The linearized Pseudo-Second-Order Kinetic model is expressed in equation 3.10:

$$\frac{t}{q} = \frac{1}{k_2 \, q_e{}^2} + \frac{1}{q_e} \, t \ \ldots\ldots\ldots (3.10)$$

a plot of t/q versus t gave a straight line and the values of the constants k_2 (g mg^{-1}) and q_e (mg $-^{1}$) were calculated(Febrianto*et al*., 2009; Njoku, 2011).

The Adsorption studies was carried out in the multiusers laboratory of the Department of Chemistry, Ahmadu Bello University, Zaria.

3.7 Statistical Analysis

The experimental design used was Complete Randomized Design (CRD).ANOVA (Analysis of Variance) was used to compare the means of the various parameters measured, and NDMRT (New Duncan Multiple Range Test) was used to separate means where significant. In the kinetic studies of adsorption, regression analysis was used to examine the relationship between the amount of heavy metal ions adsorbed per unit mass of the biosorbent and the contact time.The confidence level of was taken at 95% ($p < 0.05$).SPSS software(version 20.0) was used to run the analysis.

CHAPTER FOUR

4.0 RESULTS

4.1 Confirmation and Protein Content of *Adansonia digitata* Seed

The result in table 4.1 shows confirmation of the plant with voucher number 2512. The result of protein content determination reveals that the seeds contain a total protein content of 14.26%. Plate I, II, III and IV are pictorial representation of the plant (*Adansonia*

digitata), fruits, broken fruits and seeds respectively. Also plate V and VI shows the powdered seedsand defatted powered seeds respectively.

4.2Biocoagulation Studies and its Efficacy on Physicochemical Parameters on Surface and Model Water

It was observed that an increase in the dosage of biocoagulant caused a significant reduction ($p < 0.05$) in the turbidity of the model turbid water which resulted in an increase in clarity of the treated model water (Plate VIII) compared to the untreated model water (Plates VII). During the jar test experiment flocs began to form (Plate IX) at about 5 minutes after the commencement of stirring. An optimal dose of 150mg/l was able to decrease turbidity of model water from about 20.06NTU to 6.57NTU (Table 4.2 and Appendix I). There was also a decrease in the turbidity of the surface water with an increase in the dosage of biocoagulant but this decrease was not statistically significant ($p > 0.05$) (Table 4.3 and Appendix I).

Table 4.1: Plant verification and protein content

Plant Name	*Adansonia digitata*
Voucher Number	2512
Protein Content (%)	14.26



Plate I: *Adansonia digitata* tree



Plate II: Dried fruits of *Adansonia digitata*



Plate III : Fruits of *Adansonia digitata* broke open



Plate IV: Seeds of *Adansonia digitata*



Plate V: Powered seeds of *Adansonia digitata*



Plate VI: Cake residue of defatted Powdered *Adansonia digitata* seeds

An increase in dosage of biocoagulant caused a decrease in the TSS of model water and surface water, but the decrease was not statistically significant ($p > 0.05$) (Table 4.2 and 4.3; Appendix II).

An increase in dosage of biocoagulant caused a significant increase ($p< 0.05$) in the COD of the model turbid water. A dose of 200mg/l increased the COD of model turbid water from 150mg/l to 230mg/l (Table 4.2; Appendix III). There was also a significant increase ($p< 0.05$) in the COD of the surface water. A dose of 200mg/l of biocoagulant raised the COD of surface water from 70mg/l to 100mg/l (Table 4.3; Appendix III).

An increase in dosage of biocoagulant resulted in a decrease in the hardness of both the model water and surface water. But this decrease was not statistically significant ($p > 0.05$) (Table 4.2 and 4.3; Appendix IV).

There was a significant decrease ($p < 0.05$) in the alkalinity of model turbid water with an increase in dosage of biocoagulant. A dose of 200mg/l reduced the alkalinity from 9.00mg/l to 4.33 mg/l (Table 4.2; Appendix V). There was also a decrease in the alkalinity of the surface water, but this was not statistically significant ($p > 0.05$) (Table 4.3; Appendix V).

An increase in the dose of biocoagulant caused an increase in the TDS of both the model water and surface water. But the increase was not statistically significant ($p > 0.05$) (Table 4.2 and 4.3; Appendix VI).

There was a significant increase ($p < 0.05$) in the E.C of the model water with an increase in the dosage of biocoagulant. A dose of 200mg/l increased the EC from 111.67µs/cm to

119.60µs/cm (Table 4.2; Appendix VII). There was also an increase in the E.C of surface water, but this was not statistically significant ($p > 0.05$) (Table 4.3; Appendix VII).

An increase in the dosage of biocoagulant caused no significant difference ($p < 0.05$) in the pH of the model and surface water (Table 4.2 and 4.3; Appendix VIII).

Table 4.2: Effect of biocoagulant on different parameters of Model turbid water.

Dosage	Turbidity (mg/l)	T.S.S (mg/l)	COD (mg/l)	Hardness (mg/l)	Alkalinity (mg/l)	T.D.S (µs/cm)	E.C	pH	(mg/l)	(NTU)
0.00	20.06±1.63^{c}	0.07±0.01^{b}	150.00±0.58^{a}	35.35±5.83^{a}	9.00±1.00^{b}	57.70± 0.79^{a}	111.67±0.58^{a}	6.63±0.03^{a}		
50.00	10.12±0.53^{b}	0.07±0.03^{b}	170.00±0.58^{b}	35.35±15.42^{a}	8.00±1.00^{b}	58.40± 1.06^{a}	111.60±1.48^{a}	6.60±0.10^{a-}		
100.00	7.51±0.58^{ab}	0.05±0.00^{ab}	190.00±5.77^{c}	43.77±11.03^{a}	4.67±0.67^{a}	58.87± 1.19^{a}	113.53±2.25^{ab}	6.50±0.00^{a}		
150.00	6.57±1.57^{a}	0.05±0.01^{ab}	220.00±1.00^{d}	20.20±5.05^{a}	5.00±0.58^{a}	59.03± 0.97^{a}	118.76±2.29^{b}	6.50±0.00^{a}		
200.00	7.86±0.94^{ab}	0.04±0.00^{a}	230.00±1.00^{e}	26.93±7.34^{a}	4.33±0.58^{a}	59.80± 1.38^{a}	119.60±5.17^{b}	6.50±0.00^{a}		

Mean ± S.E.M; n = 3, means in the same column with the same alphabets are not significantly different at $p < 0.05$

TSS = Total Suspended Solids; COD = Chemical Oxygen Demand; TDS = Total Dissolved Solids; EC= Electronic Conductivity

Table 4.3: Effect of biocoagulant on different parameters of surface water

Dosage (NTU)	Turbidity (mg/l)	T.S.S (mg/l)	COD (mg/l)	Hardness (mg/l)	Alkalinity (mg/l)	T.D.S (µs/cm)	E.C	pH	(mg/l)
0.00	311.00±1.15[a]	0.01±0.00[a]	70.00±0.58[a]	67.34±14.96[a]	29.67±1.15[a]	57.70± 0.79[a]	72.60±2.36[a]		
6.50±0.00[a]	50.00	290.33±2.33[a]	0.01±0.00[a]	80.00±0.58[b]	35.35±5.05[a]	33.67±1.53[a]	58.40± 1.06[a]		
	68.67±1.99[a]	6.57±0.07[a]	100.00	284.33±12.35[a]	0.01±0.00[a]	130.00±0.58[c]	48.82±1.68[a]	34.67±4.04[a]	58.87± 1.19[a]
	70.37±2.70[a]	6.50±0.00[a]							
150.00	294.33±16.95[a]	0.01±0.00[a]	120.00±0.58[d]	57.23±4.45[a]	30.67±2.52[a]	59.03± 0.97[a]	71.76±2.56[a]	6.50±0.00[a]	
200.00	281.00±5.69[a]	0.01±0.00[a]	100.00±1.00[e]	53.87±1.68[a]	27.67±13.05[a]	59.80± 1.38[a]	78.80±3.54[a]	6.50±0.00[a]	

Mean ± S.E.M; n = 3; means in the same column with the same alphabets are not significantly different at $p < 0.05$

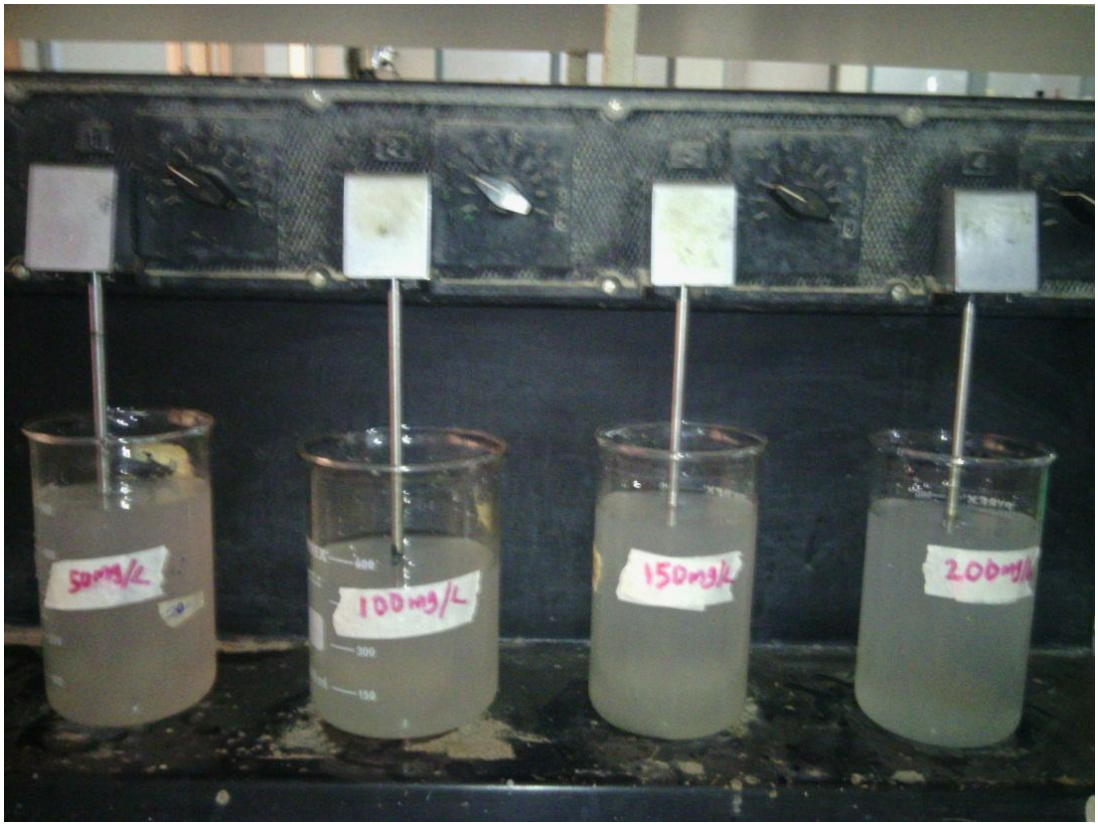


Plate VII: Model turbid water before treatment with *Adansonia digitata* seed biocoagulant



Plate VIII: Model water after treatment with *Adansonia digitata* seed biocoagulant

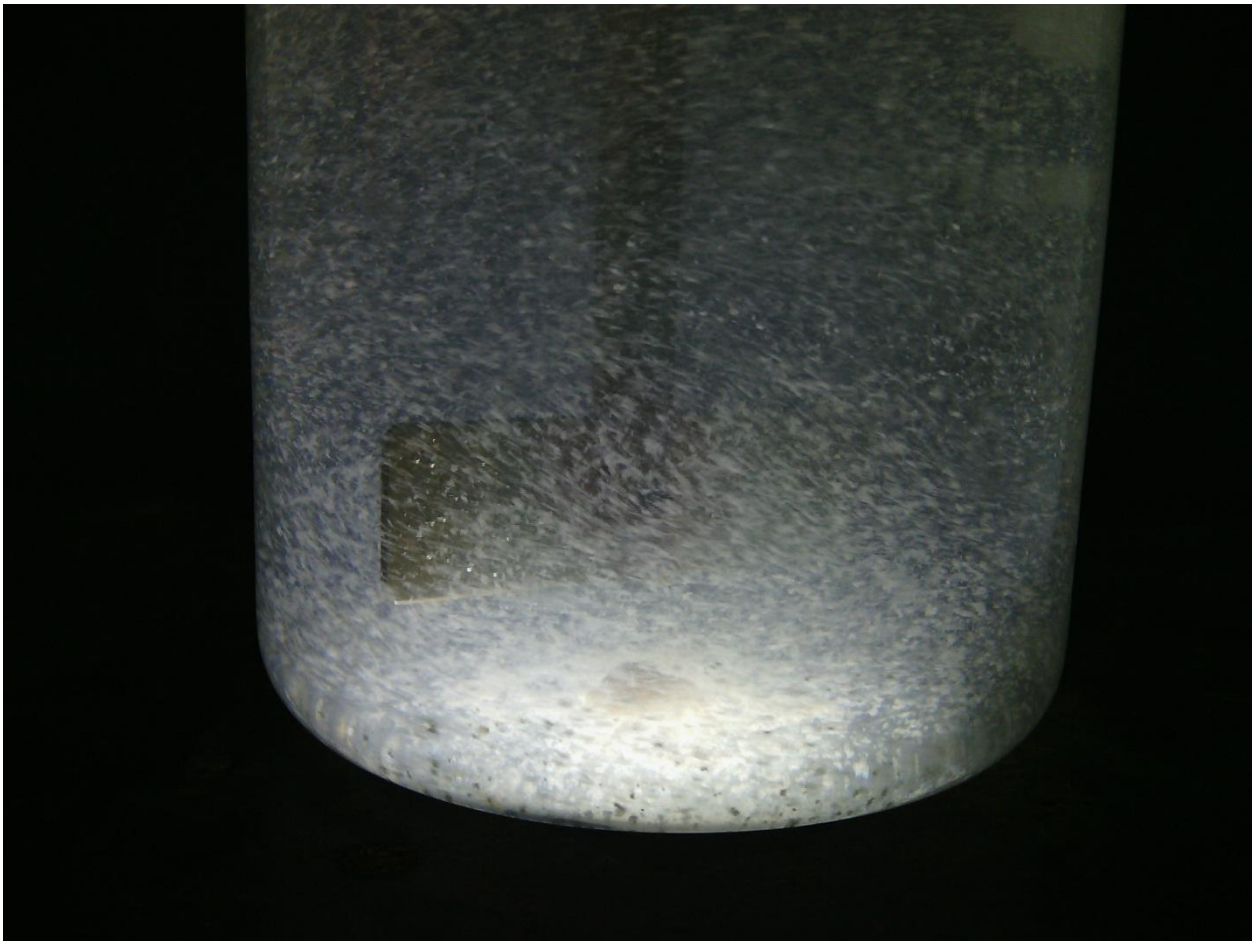


Plate IX: Formation of flocs during jar experiment after about 5 minutes of stirring model water with a concentration of 150mg/l of Adansonia digitata seed biocoagulant

4.3 Disinfection Studies

An increase in the dose of biocoagulant resulted in a significant decrease ($p < 0.05$) in the Total Coliform Counts of both the model and surface water.As a disinfectant a dose of 200 mg/l was able to reduce Total Coliform Count of both model and surface water from 1.65×10^4 cfu/ml and 4.27×10^2 cfu/mlto 5.00×10^2 cfu/ml and 6.67×10^1 cfu/ml respectively (Table 4.4; Appendix IX).

4.4 Adsorption Studies

Adansonia digitata as a biosorbent was able to adsorb the metal ions (Cd^{2+} and Pb^{2+}) from solution. Variation of various parameters (pH, initial concentration of metal ions, dosage of biosorbent, and contact time) had significant effects ($p < 0.05$) on the biosorption efficiency of the biosorbent.

4.4.1 Effect of pH on biosorption efficiency

It was observed that as pH increased from 2 – 10 the biosorption efficiency of the biosorbent increased significantly ($p < 0.05$) for both Cd^{2+} and Pb^{2+}. Maximum biosorption efficiency was obtained at pH 10, with a dose of 200mg/l of biosorbent; 74.19% and 94.57% for Cd^{2+} and Pb^{2+} respectively (Table 4.5; Appendix X).

4.4.2 Effect of initial concentration of Cd^{2+} and Pb^{2+} biosorption of efficiency

As the initial concentration of metal ions increased significantly ($p < 0.05$) from 0 – 30mg/l the biosorption efficiency also increased significantly for both Cd^{2+} and Pb^{2+}. Maximum biosorption efficiency was observed at an initial concentration of 30mg/l of metal ions,

Table 4.4: Effect of biocoagulant on Total Coliform Counts in model and surface water

	Coliform count (cfu/ml)	
Dosage (mg/l)	**Modelwater**	**Surface water**
0.00	$1.65\times10^{4} \pm 2.67\times10^{3\ c}$	$4.27\times10^{2} \pm 5.33\times10^{2\ d}$
50.00	$1.80\times10^{3} \pm 2.00\times10^{2\ ab}$	$3.13\times10^{2} \pm 1.30\times10^{1\ cd}$
100.00	$5.87\times10^{3} \pm 1.87\times10^{3\ b}$	$1.60\times10^{2} \pm 2.00\times10^{1\ c}$
150.00	$8.33\times10^{2} \pm 1.33\times10^{2\ a}$	$3.97\times10^{2} \pm 2.33\times10^{2\ b}$
200.00	$5.00\times10^{2} \pm 5.77\times10^{-2\ a}$	$6.67\times10^{1} \pm 6.67\times10^{0\ a}$

Mean±S.E.M; n =3; means in the same column with the same alphabets are not significantly different ($p>0.05$)

with a dose of 200mg/l of biosorbent; 61.59% and 97.49% for Cd^{2+} and Pb^{2+} respectively. (Table 4.6; Appendix XI).

4.4.3 Effect of contact time on biosorption efficiency

As the contact time of biosorbent with metal solution increased, the biosorption efficiency increased significantly ($p < 0.05$) for both Cd^{2+} and Pb^{2+}. Maximum biosorption was observed at a contact time of 60min, with a dose of 200mg/l of biosorbent; 74.25% and 99.97% for Cd^{2+} and Pb^{2+} respectively (Table 4.7; Appendix XII).

4.4.4 Effect of dosage of biosorbent on biosorption efficiency

As the dose of biosorbent increased biosorption efficiency also increased significantly ($p < 0.05$) for both model water and surface water. Maximum biosorption was observed at a dose of 200mg/l; 72.98% and 97.49% for Cd^{2+} and Pb^{2+} respectively, in the model water (Table 4.8; XIII); 93.52% and 100% for Cd^{2+} and Pb^{2+} respectively, in the surface water (Table 4.9; Appendix XIX).

Table 4.5: Effect of pH on the Adsorption of Cd^{2+} and Pb^{2+} from model water onto *Adansonia digitata* seed biosorbent

	Removal Efficiency (%)	
pH	**Cd**	**Pb**
2	8.09±0.00[a]	3.72±0.34[a]
4	38.80±0.72[b]	77.23±4.86[b]
7	57.79±1.50[c]	93.57±2.05[c]
8	68.27±0.82[d]	85.18±2.68[d]
10	74.19±1.97[e]	94.57±0.94[e]

Mean±S.E.M; means in the same column with the same alphabets are not significantly different($p > 0.05$)

Table 4.6: Effect of initial concentration of ions on the adsorption of Cd^{2+} and Pb^{2+} from model water onto *Adansonia digitata* seeds

	Removal Efficiency (%)	
Conc. (mg/l)	**Cd**	**Pb**
0.00	0.00 ± 0.00^{a}	0.00 ± 0.00^{a}
5.00	53.51 ± 4.58^{b}	42.75 ± 1.28^{c}
10.00	57.31 ± 4.58^{b}	21.50 ± 5.63^{b}
20.00	66.74 ± 0.23^{bc}	57.16 ± 2.46^{d}
30.00	61.59 ± 1.21^{c}	97.49 ± 0.35^{e}

Mean±S.E.M; means in the same column with the same alphabets are not significantly different ($p > 0.05$)

Table 4.7: Effect of dosage of biosorbent on the removal of Cd^{2+} and Pb^{2+} from model water onto *Adansonia digitata* seed biosorbent

	Removal Efficiency (%)	
Dosage (mg/l)	**Cd**	**Pb**
0.00	0.00 ± 0.00^{a}	0.00 ± 0.00^{a}
50.00	57.25 ± 3.36^{b}	42.75 ± 0.68^{b}
100.00	53.23 ± 1.61^{b}	21.50 ± 0.89^{b}
150.00	65.75 ± 0.41^{c}	57.16 ± 0.23^{b}

200.00	72.98±1.05[d]	97.49±0.00[b]

Mean±S.E.M; means in the same column with the same alphabets are not significantly different ($p > 0.05$)

Table 4.8: Effect of contact time on the adsorption of Cd^{2+} and Pb^{2+} from model water onto *Adansonia digitata* seed biosorbent

	Removal Efficiency (%)	
Time (min)	**Cd**	**Pb**
0.00	0.00±0.00[a]	0.00±0.00[a]
5.00	54.43±1.83[b]	92.07±1.22[b]

15.00	66.41±0.48^{c}	87.48±1.08^{c}
30.00	60.90±4.61^{bc}	93.84±0.54^{c}
60.00	74.25±0.64^{d}	99.97±0.00^{d}

Mean±S.E.M; means in the same column with the same alphabets are not significantly different ($p < 0.05$)

Table 4.9: Effect of dosage of biosorbent on the adsorption of Cd^{2+} and Pb^{2+} from surface water onto Adansonia digitata seed biosorbent

Removal Efficiency (%)

Dosage (mg/l)	Cd	Pb
0.00	0.00±0.00[a]	0.00±0.00[a]
50.00	94.16±0.11[b]	100.00±0.00[b]
100.00	93.95±0.00[c]	100.00±0.00[b]
150.00	94.27±0.18[c]	100.00±0.00[b]
200.00	93.52±0.11[c]	100.00±0.00[b]

Mean±S.E.M; means in the same column with the same alphabets are not significantly different ($p < 0.05$)

4.5 Kinetic Studies

4.5.1 Adsorption of Cd^{2+}

The R^2 value from the regression line of the Pseudo 1st and 2nd order kinetic models for the adsorption of Cd^{2+} were 0.85 (Appendix XV) and 0.99 (Appendix XVI) respectively. The Pseudo 1st and 2nd order constants obtained from the regression equation were $k_1 = 0.14$ and $k_2 = 2.29 \times 10^{-5}$ respectively (Table 4.10).

4.5.2 Adsorption of Pd^{2+}

The R^2 value from the regression line of the Pseudo 1st and 2nd order kinetic models for the adsorption of Pb^{2+} were 0.812 (Appendix XVII) and 0.998 (Appendix XVIII) respectively. The Pseudo 1st and 2nd order constants obtained from the regression equation were $k_1 = 0.16$ and $k_2 = 5 \times 10^{-6}$ respectively (Table 4.10).

Table 4.10: Pseudo 1st and 2nd order constants for the Adsorption of Cd^{2+} and Pb^{2+} onto *Adansonia digitata* seed biosorbent

		1st Order			2nd Order		
	Metal ion	**K_1** (min^{-1})	**$q_{e(\ predicted)}$** (mg g^{-1})	**R^2**	**K_2**(g mg^{-1})	**$q_{e(\ predicted)}$** (mg g^{-1})	**R^2**
Cd^{2+}	0.15	2.49×10^{4}	0.853	2.29×10^{-5}	2.50×10^{4}	0.993	
	Pb^{2+}	0.16	9.07×10^{4}	0.813	5.00×10^{-6}	2.00×10^{5}	0.998

CHAPTER FIVE

5.0 DISCUSSION

5.1 The Protein Content of *Adansonia digitata* seeds and its Effect as Biocoagulant

Presence of crude protein in *Adansonia digitata* seeds indicate that it contains biocoagulant properties. Biocoagulant proteins present in the seed actually brought about a significant decrease ($p < 0.05$) in the turbidity of the model water. This may be due to the fact that the seeds contain coagulant proteins which have charged ions and as a result when put in turbid water at a particular concentration, they released oppositely charged ions which led to the adsorption and neutralization of the charged particles (colloids) which are responsible for the turbidity in water. This finding is similar to the findings of Ndabigengesere*et al.*(1995) and Muyibi and Alfugara(2003) who worked on the use of *Moringaoleifera* as biocoagulant in turbid water, and reported that it caused a significant decrease in the turbidity of water.

The significant decrease ($p < 0.05$) in TSS observed was due to the settling of suspended particles, as a result of the introduction of the biocoagulant in water, thereby leading to the decrease in the turbidity(Arnoldsson*et al.*, 2008).

The significant increase ($p < 0.05$) in COD is due to the fact that the introduction of the biocoagulant resulted in an increase in the organic load of water, thereby more oxygen was required for the oxidation of these organic compounds. The increase in COD is a major challenge in the use of natural coagulants in the treatment of water for it tends to reduce the

shelf life of stored water, because the organics released in water begin to favour microbial activities after a period of time(*Ghebremichael, 2004).*

The significantdecrease ($p < 0.05$) in alkalinityobservedmaybe due to the factthat the cations introducedinto the water reactswith the anions responsible for alkalinity (OH^- , CO_3, $HCO3^-$) in water, therebyreducingitsability to reactwithHydrogen ions (H^+) and hencecausing a decrease in alkalinity (Muyibi and Evison, 1994).

There was an increase in the TDS althoughthiswas not statisticallysignificant($p< 0.05$), but thismayberesponsible for the slightsignificantincrease ($p < 0.05$) in the EC of water (Arnoldsson*et al.,* 2008).

The biocoagulant caused no significantincrease ($p > 0.05$) in hardness and pH ; thisis an advantage of biocoagulants over conventional coagulants likeAlumwhich tend to cause an increase in the hardness and acidity(due to reduction of pH) (Arnoldsson*et al.,* 2008).

The biocoagulant had no significantefffect ($p > 0.05$) on most of the parameters of the surface water except on the COD, thiscouldbe to the very high intitialturbidity (about 300NTU) of the surface water.

5.2 *Adansonia digitata* Seeds asDisinfectant

As a disinfectant*Adansonia digitata*seed biocoagulant caused a significantdecrease($p < 0.05$) on the total coliform count of both model water (culture) and surface turbid water. This issimilar to the findings of *Ghebremichael (2004) who worked on the disinfectant characteristics of Moringaoleifera.* The antimicrobialeffect of the biocoagulant maybeattributed to flocculation or bacteriacidaleffect of the coagulant protein. By

flocccculation the biocoagulant proteins causes aggregation of the microorganismscausingthem to besettle in the sludgeformedaftertreatment. By bacteriacidaleffect the biocoagulant maycontainsome compounds (such as antinutrientsand antimicrobialpeptides)whichmay cause a distruption in the normal biologicalfunction of the cellstherebyleading to inhibition of theirgrowth. Generally the mechanism of action of biocoagulant proteins have not yetbeingfullyunderstood (Sidibe and Williams, 2002; *Ghebremichael, 2004)*.

5.3 *Adansonia digitata*Seeds as Biosorbent

As a biosorbent*Adansonia digitata*seedssignificantly($p< 0.05$) reducedheavymetal ions (Cd^{2+} and Pb^{2+}) from water. This findingissimilar to that of Kumari*et al.*(2006) and Oboh and Aliyor(2008)whoworked on the adsorption of heavymetals by *Moringaoleifera* and *Annona muricata*seeds respectively.

The biosorption for agricultural by-products like the residual seed cake of defatted *Adansonia digitata* seeds, have been attributed to two main terms; coulombic interaction and intrinsic adsorption (Gang and Weixing, 1998). The coulombic term results from the electrostatic energy of interactions between the adsorbents(biosorbent) and adsorbates (metal ion solution). The strength of charges on biosorbent and biosorbate are mostly responsible for the intensity of the interaction. Coulombic interaction is observed from the adsorption of cationic species versus anionic species on adsorbents (Gang and Weixing, 1998). The intrinsic adsorption is due to the nature of the surface area of biosorbent, which is observed by the effect of different sizes of biosorbent on adsorption capacity (Igwe*et al.*, 2003; Igwe and Abia, 2006).

Variation of different parameters had a significant effect ($p < 0.05$) on the biosorption efficiency of *Adansonia digitata* seeds.

A change in pH from 2-10 caused a significant increase in the biosorption efficiency of *Adansonia digiatata*seeds . At a lower pH (acidic) the hydrogen ions (H^+) from the acid make the overall surface of the biosorbent to be positively charge and this leads to the repelling of the metal ions and hence a reduction in the biosorption efficiency of the biosorbent. At higher pH (basic) the hydroxide ions (OH^-) introduced by the base causes the overall surface of the biosorbent to be negatively charge leading to the attraction of more metal and hence an increase in the biosorption efficiency of the biosorbent. This is similar to the findings of Kumari*et al.* (2006).

It was observed that as the initial concentration of the metal ions increased the biosorption efficiency increased significantly ($p < 0.05$). This May be due to an increase in the concentration gradient that serves as a driving force (Han *et al.*, 2006). This was similar to the findings of Njoku*et al.*(2011) that worked on the adsorption of heavy metals using cocoa pod husk.

The effect of contact time on biosorption of Cd^{2+} and Pb^{2+} revealed that adsorption took place in two phases. A rapid phase which took place within 5 minutes and a slow phase that took until 60 minutes. The rapid biosorption was probably due to the initial availability of many binding sites and as the binding sites became exhausted biosorption took place at a slower rate. This is similar with the findings of Aravind*et al.* (2013) that worked on the adsorption of Nickel ions (Ni^{2+}) onto *Cajanuscajan* pods.

As the dosage of biosorbent increased the biosorption efficiency increased significantly ($p< 0.05$). This is probably due to the fact that an increase in dosage makes available more

binding sites for the metal ions, hence an increase in the efficiency of their removal from solution.

5.4 Kinetic Studies.

Better data fit with the pseudo-second-order model implies a marked chemisorption contribution to the metal uptake process and a similar observation was reported by Ho (2003) for copper ions onto tree fern.

CHAPTER SIX

6.0 SUMMARY, CONCLUSION AND RECOMMENDATION

6.1 Summary

Adansonia digitata seed had a crude protein content of14.26%.

As a biocoagulant, an optimal dose of 150mg/l*Adansonia digitata* seed coagulant was able to decrease turbidity of the model water from about 20.06NTU to 6.57NTU. An increase in the dosage of biocoagulant also caused a significant increase ($p< 0.05$) in the COD and EC; a significant decrease ($p< 0.05$) in TSS and alkalinity; while there was no significant difference ($p > 0.05$) in hardness, TDS, and pH.The biocoagulant had no significant effect ($p > 0.05$) on the physicochemical parameters of the surface turbid water except an increase in the COD.

As a disinfectant a dose of 200 mg/l significantly reduced ($p< 0.05$) Total Coliform Count of model and surface water from 1.65×10^4 cfu/ml and 4.27×10^2 cfu/ml to 5.00×10^2 cfu/ml and 6.67×10^1 cfu/ml respectively.

As a biosorbentthe seeds of *Adansonia digitata*was able to significantly reduce($p< 0.05$) the concentration ofthe heavy metal ions (Cd^{2+} and Pb^{2+}). Maximum biosorption was observed at a dose of 200mg/l:72.98% and 97.49% for Cd^{2+} and Pb^{2+} respectively,in the model water; 93.52% and 100% for Cd^{2+} and Pb^{2+} respectively,in the surface water. An increase in pH, initial concentration of metallic ions, dosage and contact time caused a significant increase ($p< 0.05$) in the biosorption efficiency of the biosorbent.The biosorption of Cd^{2+} and Pb^{2+}were better fitted to the Pseudo 2^{nd} order Kinetic model.

6.2 Conclusions

- *Adansonia digitata* seeds contain coagulant proteins.
- *Adansonia digitata* seeds possess potentials as a biocoagulant.It was not significantly effective when used as a biocoagulant for highly turbid water, since it was unable to bring turbidity down within the acceptable limit of less than 5NTU(USEPA, 2012).
- *Adansonia digitata* seeds possess potentials as a disinfectant.It was not significantly effective when used as a disinfectant for highly contaminated water, since it was unable to bring the total coliform count down within the accepted limit of a maximum of 10cfu/ml (USEPA, 2012).
- *Adansonia digitata* seeds possess biosorptive potentials.Its potential for the adsorption of $Pb^{2+}$was higher than that of Cd^{2+}.

6.3 Recommendation and Further Studies

- The efficiency of *Adansonia digitata* seeds as a biocoagulant and disinfectant may be improved by isolation and purification of the biocoagulant protein.
- The efficiency of *Adansonia digitata* seeds as a biosorbent may be improved by the chemical or physical modification of the nature of the biosorbent(e.g by treatment withacids and heat to form activated carbon).
- *Adansonia digitata* seed biocoagulant may be used to complement other conventional coagulants like alum.

- Scanning Electron Microscope (S.E.M) studies may be carried out to understand the morphology of the *Adansonia digitata* seed biosorbent

- Fourier Transform Infrared Spectroscopy (FTIR) studies may be carried out to understand the functional groups available on the surface of *Adansonia digitata* seed biosorbent.

REFERENCES

Ahmadu Bello University (ABU) (2013). Zaria at a glance. Retrieved from http://www.abu.edu.ng/info/zaria.php

Ahalya, N., Ramachandra, T.V. and Kanamadi, R.D. (2005). Biosorption of Chromium (VI) from aqueous solutions by the husk of bengal gram (*Cicerarientinum*). *Electronic Journal of Biotechnology*, 3: 258-264.

Ahluwalia, S.S. and Goyal, D. (1998). Advances in the biosorption of heavy metals, *Trends in Biotechnology*, 16: 291-300.

Ajayi, I.A., Dawodi, F.A., Oderinde, R.A. (2003). Fatty acid composition and metal content of *Adansonia digitata* seeds and seed oil. *La RivistaItalianadelleSostanze Grasse,* 80, 41-43.

Alam, M., Nadeem, R., and Jilani, M. I. (2012). Pb (II) removal from wastewater using Pomegranate waste biomass. *International Journal of Chemical and Biochemical Sciences,* 1(2012): 24-29.

Amagloh F.K. and Benang A.(2009). Effectiveness of *Moringa oleifera* seed as coagulant forwater purification. *African Journal of Agricultural Research*, 4 (1):119-123.

American Public Health Association, American Water Works Association and Water Pollution Control Federation (APHA, AWWA and WPCF) (1985). *Standard methods for the examination of water and wastewater*, 16th ed. Port City Press, Baltimore, Maryland. 1268 p.

Antov, M.G., Šc´iban, M.B. and Petrovic´ N.J. (2010). Proteins from common bean (*Phaseolusvulgaris*) seed as a natural coagulant for potential application in water turbidity removal. *Bioresource Technology*,101(2010): 2167–2172.

Andy, E.O. and Eteshola, E. (1984) Nutritive value of a mixture of tigernut tubers (*Cyprus esculentus*) and Baobab seeds. *Journal of the Science of Food and Agriculture*, 35 (1984): 437 – 440.

Arnoldsson, E., Bergman, M., Matsinhe, N., and Persson, K.M. (2008). Assessment of drinkingwater treatment using *Moringaoleifera* natural coagulant. *VATTEN*, 64:137–150.

Aravind, J., Muthusamy, S., Sunderraj, S. H., Chandran, L. and Palanisamy, K. (2013). Pigeon pea (*Cajanuscajan*) pod as a novel eco-friendly biosorbent: a study on equilibrium and kinetics of Ni (II) biosorption. *International Journal of Industrial Chemistry*, 4(25): 1-9

Asmatullah, Q. S. N. and Shakoori, A.R. (1998). Hexavalent chromium induced congenital abnormalities in chick embryos. *Journal of Applied Toxicology*, 18 (3):167-171.

Association of Analytical Chemistry (A.O.A.C) (1980). *Official methods of analysis of the Association of Analytical Chemistry* ,13th ed. Washington D.C. pp. 376 – 384.

Banerjee, k., Ramesh, S.T., Gandhimathi, R., Nidheesh, P.V. and Bharathi, K.S., (2012) A novel agricultural waste adsorbent, watermelon shell for the removal of copper from aqueous solutions. *Iranica Journal of Energy and Environment*, 3 (2): 143-156.

Bichi, M. H., Agunwamba, J. C. and Muyibi, S. A. (2012). Optimization of operating conditions for the application of *Moringaoleifera* (Zogale) seeds extract in water disinfection using response surface methodology. *African Journal of Biotechnology*, 11(92): 15875-15887.

Bodlund, I. (2013). Coagulant protein from plant materials: Potential water treatment agent. School of Biotechnology, Royal Institute of Technology (KTH), AlbaNova University Center, Stockholm, Sweden, pp. 1- 37.

Bremer, B., Bremer, K., Chase, M.W., Reveal, J.L., Soltis, D.E., Soltis, P.S...Zmarzty, S. (2003). An update of the angiosperm phylogeny group classification for the orders and families of flowering plants: APG II. *Botanical Journal of the Linnean Society*, 141, 399-436.

Cantor, K.P., Lynch, C.F., Hildesheim, M.E., Dosemeci, M., Lubin, J., Alavanja, M. and Craun, G. I. (1998) Drinking water source and chlorination by-products: Risk of bladder cancer. *Epidemiology*, 9:21–28.

Celik, A. and Demirbas, A. (2005). Removal of heavy metal ions from aqueous solutions via adsorption onto modified lignin from pulping wastes, *Energy Sources*, 27: 1167-1177.

Chen, L. and Dixon, K. (1998). Analysis of repair and mutagenesis of chromium induced DNA damage in yeast mammalian cells and transgenic mice. *Environmental Health Perspective*, 106:1027-1032.

Choubey, S., Rajput, S.K. and Bapat, K.N.(2012). Comparison of efficiency of some natural coagulants-bioremediation. *International Journal of Emerging Technology and Advanced Engineering*, 2(10):430-434

Colwell, R.R. and Sasson, A. (1996). Biotechnology and development. In World Science Report 1996 .UNESCO. Retrieved from www.unesco.org/bpi/science/.../10.htm

Creed, J.T. and Martin, T.D. (1992). *Determination of trace elements in marine waters by off-line chelation*. Environmental Monitoring Systems Laboratory Office of Research and Development U.S. Environmental Protection Agency, Cincinnati, OH 45268. pp. 1-20.

Dada, A.O., Olalekan, A.P., Olatunya, A.M., and Dada, O. (2012). Langmuir, Freundlich, Temkin and Dubinin–Radushkevich isotherms studies of equilibrium sorption of Zn^{2+} unto phosphoric acid modified rice husk. *Journal of Applied Chemistry.* 3 (1): 38-45

Dalziel, J.M. (1973). The useful plants of West.Tropical Africa. Crown agents for the colonies, London,pp.18-22.

Dalen, M.B., Pam, J.S., Izang, A. and Ekele, R.(2009). Synergy between *Moringaoleifera* seedpowder and alum in the purification of domestic water. *Science World Journal*, 4, 6-10.

Das, N., Vimala, R. and Karthika, P. (2008). Biosorption of heavy metals- An overview. *Indian Journal of Biotechnology*, 7: 159-169.

De Caluwé, E., Halamová, K. and Van Damme, P. (2010). *Adansonia digitata* L. – a review of traditional uses, phytochemistry and pharmacology. *Afrika focus*, 23 (1): 11-51.

Delle, A.S. (2001). Factors affecting sorption of organic compounds in natural sorbent/water systems and sorption coefficients for selected pollutants. A review. *Journal of Physical and Chemical Reference Data*, 30:187–439.

Diaz, A., Rincon, N., Escorihuela, A., Fernandez, N., Chacin, E. and Forster, C.F. (1999). A preliminary evaluation of turbidity removal by natural coagulants indigenous to Venezuela. *Process Biochemistry*, 35: 391-395

Diop, A.G., Sakho, M., Dornier., M., Cisse, M. and Reynes, M. (2005). Le baobab africain (*Adansonia digitata* L.): principals caractéristqueset utilisations. *Fruits*, 61, 55-69.

Doerr, B. (2005). *Moringa* water treatment, an echo technical note. Retrieved from http://miracletrees.org/moringa_water_purification.html

Dovie, B. D. K. (2003). Rural economy and livelihoods from the non-timber forest products trade. Compromising sustainability in southern Africa. *International Journal of Sustainable Development and World Ecology*, 10:247-262

Dovie, B. D., Shackleton, C. M. and Witkowski, E. T. F. (2001) Involving local people: Reviewing participatory approaches for inventorying the resource base, harvesting and utilization of non-wood forest products. In Harvesting of Non-Wood Products. Proceedings of FAO/ECE/ILO Internaitonal Seminar, Ministry of Forestry, Turkey, pp. 175-187.

Edogbanya, P.R.O., Ocholi, O.J. and Apeji, Y. (2013a). A review on the use of plants' seeds as biocoagulants in the purification of water. *Continental Journal of Biological Sciences*, 6(2): 26-32.

Edogbanya, P.R.O., Ocholi, O.J. and Apeji, Y. (2013b). A review on the use of plants' seeds as biosorbents in the removal of heavy metals from water. *Advances in Agriculture, Science and Engineering Research*, 3 (8): 1036-1044.

Egila J. N., Dauda, B. E. N., Iyaka, Y. A. and Jimoh, T. (2011). Agricultural waste as a low cost adsorbent for heavy metal removal from wastewater. *International Journal of Physical Science*, 6 (8): 2152-2157

Febrianto, J., Kosasiha, A.N., Sunarsob, J., Jua, Y., Indraswatib, N. and Ismadjia, S.(2009). Equilibrium and kinetic studies in adsorption of heavy metals using biosorbent: A summary of recent studies. *Journal of Hazardrous Materials*, 162, 616–645.

Fleuret, A. (1980). Non-food uses of plants in Usambara. *Economic Botany*, 34: 320-333.

Food and Agricultural Organisation (FAO). (2007).Coping with water scarcity, 2007 world waterday,22ndmarch,2007.Retrieved from: http://www.fao.org/nr/water7docs/wwd07brochure.pdf

Food and Agriculture Organization of the United Nations (FAO). (2014). Water – The Most Basic Resource but also the Most Essential. Retrieved from http:// www.fao.org/zhc/detail-events/en/c/231215/

Gang, S. and Weixing, S. (1998). Sunflower stalks as Adsorbents for the removal of metal ions from wastewater. *Indian Engineering Chemistry Research*, 37: 1324-1328.

Gavrilescu, M. (2004). Removal of Heavy metals from the environment by biosorption. *Engineering in life Sciences*, 4(3): 219-232.

Gebauer, J., El-Siddig, K. and Ebert, G. (2002).Baobab (*Adansonia digitata* L.): a review on a multipurpose tree with promising future in the sudan. *Gartenbauwissenschaft*, 67, 155-160.

Ghebremichael, K.A., Gunaratna, K.R., Henriksson, H., Brumer, H. and Dalhammar, G. (2005). A simple purification and activity assay of the coagulant protein from Moringaoleifera seed. *Water Research*. 39 (11): 2338-2344

Gohulavani, G. and Andal, N.M. (2013) Removal of nickel from aqueous solution using *Azaridachtaindica* seed shell powder as adsorbent. *Research and Reviews: Journal of Chemistry*, 2 (1):78-84.

Gupte, A., Karjikar, M. and Nair, J. (2012) Biosorption of copper using mucilaginous seeds of *Ocimumbasilicum*. *ActaBiologicaIndica,* 1(1):113-119

Hadi, N.A., Rohaizar, N.A. and Wong, C.S. (2011). Removal of Cu (II) from water by adsorption on papaya seed. *Asian Transactions on Engineering*, 1 (5): 49-55.

Han, R., Zhang, J., Zou, W., Xiao, H., Shi, J. and Liu, H. (2006). Biosorption of copper (II) and lead (II) from aqueous solution by chaff in a fixed-bed column. *Journal of Hazardous Materials*, 133: 262-268.

Ho, Y.S. (2003). Removal of copper ions from aqueous solution by tree fern. *Water Resources*, 37: 2323-2330.

Igboeli, L.C., Addy, E.O.H. and Salami, L.I. (1997). Effects of some processing techniques on the antinutrient contents of baobab seeds (*Adansonia digitata*).*Bioresource Technology*, 59, 29-31.

Igwe, J.C. and Abia, A.A. (2005). Sorption kinetics and intraparticulate diffusivities of Cd, Pb and Zn ions on maize cob. *African Journal of Biotechnology*, 4(6):509-512.

Igwe, J. C. and Abia, A.A. (2006). A bioseparation process for removing heavy metals from waste water using biosorbents. *African Journal of Biotechnology*. 5 (12):1167-1179.

Mahajan, G. and Sud, D. (2013). Bioremediation of Cr (vi) metal ion from aqueous solutions using modified lignocellulosic agricultural waste biomass. *International Journal of Advanced Technology & Engineering Research*, 3(2): 35 – 43.

Jahn, S.A.A. and Dirar, H. (1979). Studies on natural water coagulants in the Sudan, with special reference to *Moringaoleifera* seeds. *WATER SA* 5:90–97.

Jimoh, T. O., Yisa J., Ajai, A. I. and Musa, A. (2013). Kinetics and Thermodynamics Studies of the Biosorption of Pb(II), Cd(II) and Zn(II) Ions from Aqueous Solution by Sweet Orange (*Citrus sinensis*) Seeds. *International Journal of Modern Chemistry*, 4, 1, 19-37

Kale, A.A. (2013). Uptake of Pb^{2+} by Sulphonated Biomass of Cicerarietinum; batch studies. *International Scientific Research Network: Environmental Chemistry*, doi: 320649.

Kumari, P., Sharma, P., Shalini, S. and Srivastava, M.M. (2006). Biosorption studies on shelled *Moringaoleifera* Lamarck seed powder: removal and recovery of arsenic from aqueous system, *International Journal of Mineral Processes,* 78: 131- 139.

Lima, E. C., Royera, B., Vaghettia, J.C.P., Brasila, J.L., Simona, N.M., dos Santos, A.A., Pavana, F.A., Diasa, L.P.S., Benvenuttia, E.V. and da Silvab, E.A. (2007). Adsorption of Cu(II) on Araucaria angustifolia wastes: Determination of the optimal conditions by statistic design of experiments. *Journal of Hazardous Materials*, 140: 211–220.

Mancy, T.B., Mathew, M.M. and Stanley, S.A. (2013). Comparative analysis of cost effective biosorbents in the removal of chromium and iron from aqueous solution. *Advances in Biotecnology*. 12(12):01-05.

Marobhe, N.J., Dalhammar, G. and Gunaratna, K.R. (2007). Simple and rapid methods for purification and characterization of active coagulants from the seeds of *Vigna unguiculata* and *Parkinsoniaaculeata*. *Environmental technology,* 28:671–681.

Meneghel, P.A., Conclaves A.C., Fernanda, R., Dragunski, D.C., lindino A.C., Strey L. (2013). Biosorption of cadmium (Cd) using *Moringaoleifera* (lam) seeds. *Water Air Soil Pollution,* 224 (1383): 1-13.

Morris, R., Audet, A., Angelillo, I., Chalmers, T. and Mosteller, F. (1992). Chlorination, chlorination by-products, and cancer - a meta-analysis. *American Journal of Public Health*, 82:955–963.

Mudgal,V., Madaan,N., Mudgal, A., Singh, R.B. and Mishra, S. (2010). Effect of toxic metals on human health. *The Open Nutraceuticals Journal*, 3:94-99.

Muyibi, S.A. and Alfugara, A.M.S. (2003). Treatment of surface water with *Moringaoleifera*seed extract and alum—A comparative study using a pilot scale

water treatment plant. *International Journal of Environmental Study,* 60 (6): 617–626.

Muyibi, S.A. andEvison, L.M. (1994). *Moringaoleifera* seeds for softening Hardwater.*Water Resources,* 29 (4): 1099-1105.

Ndabigengesere, A., Narasiah, K.S. and Talbot, B.G. (1995). Active agents and mechanism of coagulation of turbid waters using *Moringaoleifera*. *Water Resources,*29 (2): 703–710.

Nkana, Z. G. and Iddi, S. (1991). *Utilization of Baobab (Adansonia digitata) in Kondoa District, Central Tanzania.* Faculty of Forestry, Sokoine University of Agriculture Morogoro. pp. 1- 50.

Njoku, V.O., Ayuk, A.A., Ejike, E.E., Oguzie, E.E., Duru, C.E. and Bello, O.S. (2011). Cocoa pod husk as a low cost biosorbent for the removal of Pb(II) and Cu(II) from Aqueous solutions. *Australian Journal of Basic and Applied Sciences,* 5 (8): 101-110.

Obire, O., Tamuno, D.C. and Wemedo, S.A. (2005). Bacteriological water quality of elechi creek in portharcourt, nigeria. *Journal of Applied Science and Environmental Management*. 9(1):79-84.

Oboh, O. I. and Aluyor, E.O. (2008). The removal of heavy metal ions from aqueous solutions using soursop seeds as biosorbent. *African Journal of Biotechnology*. 7 (24): 4508-4511.

Okuda T., Baes A. U., Nishijima W. and Okada, M. (2001). Isolation and characterization of coagulant extracted from *Moringaoleifera* seed by salt solution. *Water Resources,* 35 (2): 405–410.

Omorogie, M.O., Babalola J.O., Unuabonah, E.I. and Gong, J.R. (2012). Kinetics and thermodynamics of heavy metal ions sequestration onto novel *Naucleadiderrichii* seed biomass. *Bioresource Technology*, 118: 576–579.

Osman, M.A. (2004). Chemical and Nutrient Analysis of Baobab (*Adansonia digitata*) Fruit and Seed Protein Solubility. *Plant Foods for Human Nutrition*, 59: 29-33.

Palmer, E. and Pitman, N. (1972). *Trees of Southern Africa*. A.A BalKema, Cape Town. Vol. 2.

Pritchard, M., Mkandawire, T., Edmondson, A., Neill, J.G.O. and Kululanga, G. (2009). Potential of using plant extracts for purification of shallow well water in Malawi, *Physics and Chemistry of the Earth*, 34: 799-805.

Proll, J., Petske, K.J., Ezeagu, I.E., Metges, C.C. (1998). Low nutritional quality of unconventional tropical crop seeds in rats. *Journal of Nutrition*, 128: 2014-2022.

Raj, K.R., Kardam, A. and Srivastava, S. (2013). PEI modified *Leucaenaleucocephala* seed powder, a potential biosorbent for the decontamination of arsenic species from water bodies: bioremediation. *Applied Water Science*, 3:327–333.

Rajjak, S.P. and Bapurao, B.A. (2012). Biosorptive behaviour of neem (*Azadirachtaindica*) bark for Iron removal from aqueous solutions. *Journal of Applied Phytotechnology in Environmental Sanitation*, 2 (1): 31- 36.

Richardson, S.D., Plewa, M.J., Wagner, E.D., Schoeny, R. and Demarini, D.M. (2007). Occurrence, genotoxicity, and carcinogenicity of regulated and emerging disinfection by-products in drinking water: a review and roadmap for research. *Mutation research*, 636:178–242.

Royal Botanical Gardens, Kew. *Adansonia digitata* (Baobab). retrieved from http://www.kew.org/science-conservation/plants-fungi/adansonia-digitata-baobab

Saefudin, S., Aryani, M., Safaria T. (2012). Effectivity of broadbean (*Viciafaba*) as a biocoagulant to rehabilitate physic and chemical character of pulp and paper mill wastewater. Retrieved fromhttp://file.upi.edu/Direktori/SPS/PRODI.PENDIDIKAN_IPA/196307011988031-SAEFUDIN/Vicia_faba_Saefudin_dkk_%5B1%5D.pdf

Saif, M.M.S., Kumar, N.S. and Prasad, M.N.V. (2012). Binding of cadmium to Strychnospotatorum seed proteins in aqueous solution: adsorption kinetics and relevance to water purification. *Colloids and Surfaces :Biointerfaces,* 94:73–79.

Sen, T.K. (2012). Agricultural by-product biomass for removal of pollutants from aqueous solution by adsorption, *Journal of EnvironmentalResources and Development*, 6 (3): 524-533.

Sharma, P. Kumari, P. Srivastava, M.M. and Srivastava, S. (2007). Ternary biosorption studies of Cd(II), Cr(III) and Ni(II) on shelled *Moringaoleifera* seeds, *Bioresource Technology*, 98:474-477.

Baobab - *Adansonia digitata*. International Centre for Underutilised Crops, Southampton, United Kingdom. pp. 14 – 16

Shrivastava, A.K. and Khaladkar, H.S. (2013). Novel method for micro determination of lead in water samples. *Scientific Reviews Chemical Communication*, 3(1): 43-53.

Singh, U., Kausal, R.K. (2013). Treatment of waste water with low cost adsorbent – A review, *VSRD International Journal of Technical and Non-Technical Research*, 4,3,33-42.

Stanley, S.A., Mathew, M.M. and Mancy, T.B. (2013). Removal of chromium (VI) and iron (III) from aqueous solution using agricultural byproducts. *Journal of Chemical and Pharmaceutical Resources*, 4:301-309.

Subramanium, S., Vikashni, N., Maata, M. andKoshy, K.(2011). Moringa oleifera and other local seeds in water purification in developing countries .*Research Journal of Chemistry and Environment,* 15 (2): 135-138

United Nations World Water Assessment Programme (UN WWAP) (2003). The world water development report 1: water for people, water for life. Retrieved from: http*://unesdoc.unesco.org/images/0012/001295/129556e.pdf.*

United States Environmental Protection agency (USEPA). (2004). Drinking water treatment. Retrieved from water.epa.gov/.../2009_08_28_sdwa_fs_30ann_treatment_web.pdf

United States Environmental Protection agency (USEPA). (2012). Drinking water standards and health advisories. Retrieved from water.epa.gov/action/advisories/drinking/upload/dwstandards2012.pdf

Villanueva, C.M., Cantor, K.P., Grimalt, J.O., Malats, N., Silverman, D., Tardon, A., Garcia-Closas, R., Serra, C., Carrato, A., Castano-Vinyals, G., Marcos, R., Rothman, N., Real, F.X., Dosemeci, M. and Kogevinas, M. (2006). Bladder Cancer and Exposure to Water Disinfection By-Products through Ingestion, Bathing, Showering, and Swimming in Pools. *American Journal of Epidemiology*, 165:148–156.

World Health Organization (WHO). (2004). Water, Sanitation and Hygiene Links to Health, Facts and Figures retrieved from http://www.who.int/water_sanitation_health/publications/facts2004/en/

World Business Council for Sustainable Development (WBCSD). (2005). Water: facts and trend. Retrieved from http://www.unwater.org/downloads/Water_facts_and_trends.pdf.

Yao, Z.Y., Qi, J.H. and Wanga, L.H. (2010). Equilibrium, kinetic and thermodynamic studies on the biosorption of Cu(II) onto chestnut shell. *Journal of Hazardous Materials*,174: 137–143.

Yazzie, D., VanderJagt, D.J., Pastuszyn, A., Okolo, A. and Glew, H. (1994). The amino acid and mineral content of baobab (*Adansonia digitata* L.) Leaves. *Journal of Food Composition and Analysis*, 7, 189-193.

Yongabi, K., Lewis, D. and Harris, P. (2012). Water Pollution: Natural Materials for http://www.intechopen.com/books/water-pollution/natural-materials-for-sustainable-water-pollution-management

Yongabi, K.A, Lewis, D. and Harris, P.L. (2011). Indigenous plant based coagulants/ disinfectants and sand filter media for surface water treatment in Bamenda, Cameroon,*African Journal of Biotechnology*, 10 (43): 8625-8629.

Zamani, A., Shokri, R., Yaftian, M. and Parizanganeh, A. (2013). Adsorption of lead, zinc and cadmium ions from contaminated water onto *Peganumharmala* seeds as biosorbent. *International Journal of Engineering*. 10 (1): 93-102.