

Jimmy University

College of Public Health and Medical Sciences

Department of Environmental Health Sciences and Technology



**Removal of Turbidity and Microbial Load for Household Water
Treatment**

By

Benti Firdissa (B.Sc.)

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Benti Firdissa (B.Sc.)

**Advisors: - Abebe Beyene (PhD, Ass. Prof.)
-Argaw Ambelu (PhD, Ass. Prof.)
-Moa Megersa (PhD candidate)**

June 2013

Jimma, Ethiopia

DECLARATION

I proclaim that this piece of work is my own and all sources of materials used for this thesis work will have been properly acknowledged.

Name: Benti Firdissa

Place: Jimma, Ethiopia

Signature: _____

Date of submission: _____

This thesis was submitted for examination with the approval of university advisors

Name	Signature	Date
Abebe Beyene (PhD, Ass. Prof.)	-----	-----
Argaw Ambelu (PhD, Ass. Prof)	-----	-----
Department Head	-----	-----

Abstract

Background: *The use of inorganic chemicals for the removal of turbidity and bacteria was recognized as one of the public health and environment concern due to disinfection byproduct formation and sludge production. In addition, unsafe drinking water is a paramount concern because of the fact that, 75% of all diseases in developing countries are arising from polluted drinking water especially in rural parts of developing countries. We conducted a series of experiments on the effectiveness of in removing turbidity and microbes by using both synthetic and natural surface water samples in the laboratory of Environmental Health Sciences and Technology, Jimma University from February to April, 2013. A conventional jar test apparatus was used to achieve uniform agitation rate throughout the experiment. The experiments were designed targeting both dose and contact time of plant coagulants and synthetic chemicals while recording major influencing water quality parameters. Spread plating method was employed for microbial test using plant species. Plant coagulants showed relatively lower removal efficiency ($\approx 70\%$) as compared to alum ($\approx 80\%$) at low turbidity (20 NTU) in synthetic water. However, in natural water samples of low turbidity, plant coagulants showed high rate of turbidity removal efficiency ($\approx 90\%$) like that of alum. Plant coagulants can also achieve maximum turbidity removal ($\approx 97\%$) like that of alum in medium turbidity level (200 NTU) in both natural and synthetic water samples. The experimental result revealed that plant coagulants were able to meet World Health Organization standards of drinking water quality (< 5 NTU) in terms of turbidity. The microbial reduction experiment also revealed that plant coagulants can effectively disinfect water at low turbidity but becomes less potent disinfectant as turbidity increases.*

Key words: *Coagulation, Disinfection, Household treatment, Native plants, Turbidity*

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ABBREVIATIONS

BOD - Biochemical oxygen Demand

CDC - Centers for Disease Control and Prevention

CSAE - Central Statistical Authority of Ethiopia

MDG - Millennium Development Goals

MoFED - Ministry of Finance and Economic Development

NOM - Natural Organic Matter

M. subcordata - *Maerua subcordata*

M. stenopetala - *Moringa stenopetala*

NTU - Nephelometric Turbidity Unit

rpm - rotation per minute

TDS -Total Dissolved solid

TTHM -Total trihalomethanes

UNICEF -Unite Nations Children's Fund

WCC - World Chlorine Council

WHO - World Health Organization

CHAPTER ONE: INTRODUCTION

1.1 Background

In developing countries, access to safe water is a crucial issue; because water related diseases are one of the major health problems globally. About 75% the present world lives in developing countries out of which, 1.2 billion people still lack of safe drinking water and more than 6 million children die from diarrhea every year (Action Aid, 2010). About 84% of the populations without access to an improved source of drinking water live in rural areas of developing countries. In Africa, one third of the population no access to safe water, and almost two thirds have no access to sanitation, causing widespread suffering from malaria, typhoid, dysentery and many other diseases that cause loss of productivity (WHO and UNICEF, 2010).

Ethiopia is one of the countries in the world with respect to water resources. Even though there is some improvement concerning access to safe drinking water which increased from 19% in 1990 to 68.5% in 2009/10 the access rate to drinking water and sanitation in Ethiopia is among the lowest in the world. Peoples in Ethiopia without access to safe water depend on surface water sources such as unprotected springs, ponds, streams and rivers in which most of them are located at great distances from their households (up to six hours in some rural areas) where the burden highly rests on women and children. Even in urban areas, where access to safe water is higher, the quality, quantity and irregularity of water supply is far from being adequate (WHO, 2011).



Figure 1 Collection of raw water for drinking purpose from Kersa wereda Kuno kebele (Photo by Benti, 2012).

Natural plant extracts have been used for water purification for many centuries. Various technologies adopted at household level to treat raw water such as SODIS, filtration, and combined coagulant disinfection system. In recent years there has been considerable interest in

the development of usage of natural coagulants which can be produced extracted from plants. These coagulants are biodegradable, less voluminous sludge and are presumed to be safe for human health (Narasiah *et al.*, 2002 and Marina *et al.*, 2006).

Nowadays a number of effective coagulants of plant origin have been identified. Some of the common ones include *Moringa oleifera*, *Solanum incunum*, *Ocimum sanctum*, *Azadirachta indica*, *Triticum aestivum*, *Phyllanthus emblica* and *Strychnos potatorum* and others (Kihampa *et al.*, 2011; Sunil *et al.*, 2011 and Yongabi *et al.*, 2011) of the large number of plant materials that have been used over the years. The seeds from *Moringa oleifera* have been shown to be one of the most effective primary coagulants for water treatment especially in rural communities. Like elsewhere of the world, local people of Ethiopia use their indigenous knowledge to treat their raw water using plants like *M. subcordata* and *M. stenopetala*. So the main aim of this study was to evaluate contaminant removal performance of native plant species interns of turbidity and microbial load reduction in laboratory which were used by local people.

1.2 Statement of the Problem

Getting safe water for rural community is difficult because of the fact that, most of rural dwellers are highly dependent on surface water for their drinking purpose which is untreated and this is evident in developing countries in which much of diarrheal and other water related diseases are reported (WHO, 2011). Aluminum salts are widely used as chemical coagulant in water purification process all over the world. However, resent studies have raised doubts about the advisability of introducing aluminum in to the environment, especially concerning about residuals in the treated water, large production of sludge volume and Alzheimers disease (Diaz *et al.*, 1999 and Okuda *et al.*, 2001). There is also another problem of alum reaction with natural alkalinity present in the water leading to the reduction of pH and low efficiency in coagulation in water (Megat, 2006 and Katayon *et al.*, 2006). These chemicals can be a serious problem on countries economy because they pay high cost for importing the chemicals for water treatment (Diaz *et al.*, 1999).

The use of sophisticated technologies and different chemicals in the context of developing countries for their water supply activities is inappropriate. Wright *et al.* (2003) commented that coagulation and rapid mixing, flocculation, sedimentation, filtration and disinfection are inappropriate in rural areas of developing countries. Scientists have conducted studies on health

effects of exposure to high levels of DBPs on laboratory animals. These studies have shown that several DBPs cause cancer in laboratory animals and some DBPs cause undesirable effects in animal's growth and reproduction.

Similarly few toxicological and epidemiological studies have been carried out examining the effects of DBP on reproductive health outcomes. The main outcomes of interest so far have been low birth weight (Lewis *et al.*, 2006), preterm delivery, spontaneous abortions, still-birth and birth defects in particular central nervous system, oral cleft, and respiratory (Voisin *et al.*, 2010) and neural tube defects (Moser *et al.*, 2004). Similar studies showed that, exposure to very high levels of certain DBPs resulted in kidney and liver damage effects (Chad *et al.*, 2005).

CHAPTER TWO: LITERATURE REVIEW

2.1 General Overview of Water Quality

The access to safe drinking water is a major concern throughout the world. The MDG drinking water target has been reached over 2 billion people gained access to improved water sources from 1990 to 2010, and the proportion of the global population still using unimproved sources is estimated at 11 %. While coverage of improved water supply sources is 90 % or more in Latin America and the Caribbean, Northern Africa and large parts of Asia, it is only 61 % in sub-Saharan Africa. Coverage in the developing world overall stands at 86 %, but it is only 63 % in countries designated as 'least developed'.

Systematically testing the microbial and chemical quality of water at the national level in all countries is prohibitively expensive and logistically complicated, some of these sources may not be adequately maintained and therefore may not actually provide 'safe' drinking water (WHO and UNICEF, 2012). Surface water has become the most common source for raw water, when large quantities of groundwater often are inaccessible and as surface water requires more treatment, simple, cheap and efficient process methods are needed. Turbidity removal is essential for treatment of surface water and is often carried out with coagulation using metal salts as aluminum sulphate. This is also used in Ethiopia but studies suggest that the metal salt can be replaced with a natural coagulant (Arnoldsson and Bergman, 2007).

2.2 Parameters for Drinking Water Quality

When evaluating the quality of drinking water, numerous parameters should be taken into account. Some of them are described below:

2.2.1 Turbidity

The cloudiness of water is referred to as turbidity and has its origin from particles suspended in the water. These are natural contaminants and most often mineral particles such as clay and silt or organic flocs. Turbidity is a major problem in drinking water treatment when the water source is surface water. It is also a key indicator used in assessing the suitability of water for human consumption. The World Health Organization allows drinking water with turbidity below 5 NTU. Deterioration in drinking water quality in distribution networks is also due to an increase

in microbial numbers, an elevated concentration of ion or increased turbidity, all of which affect taste, odor and color in drinking water. Turbidity can provide shelter for opportunistic microorganisms and pathogens. Hence, waters with high turbidity, from organic sources, also give rise to a substantial chlorine demand for disinfection purposes (Sadiq and Rodriguez M. J, 2004).

2.2.2 Microbial quality

The microbial quality has a large effect on the taste and smell of the water and can sometimes be a large problem in river waters. Eutrophication of the waters due to disposal of phosphate and nitrate from agriculture and wastewater among others favors algae and bacteria growth and can cause health risks. Bacteria in waters can cause illnesses as typhoid (*Salmonella typhi*), cholera (*Vibrio cholera*) and diarrhea. Fecal coliforms and streptococci indicate that wastes from humans or animals contaminate the water. Fecal streptococci are the most resistant group of bacteria, and are often analyzed together with total coliforms as an indication of a total bacteriological status. WHO and EPA recommend Total coliform, fecal coliform and *E. coli* to be 0 per 100ml of water.

2.2.2.1 Heterotrophic bacteria

Heterotrophic bacteria are those microorganisms that use organic compounds for most or all of their carbon requirements. Most bacteria, including many of the bacteria associated with drinking water systems are heterotrophic. Unlike other indicators, such as *E. coli* or total coliforms, low concentrations of heterotrophic bacteria will still be present after drinking water treatment. In general, water utilities can achieve heterotrophic bacteria concentrations of 10 colony-forming units (cfu/mL) or less in finished water. Within a distribution system, increases in the density of heterotrophic bacteria are usually the result of bacterial regrowth (Kalibbala, 2007).

2.2.4 Total Suspended Solids (TSS)

Total Suspended Solids are the amount of filterable solids in a water sample. Samples are filtered through a glass fiber. The filters are dried and weighed to determine the amount of total suspended solids in mg/l of sample.

2.2.5 Total Dissolved Solids (TDS)

The presence of high levels of TDS in water may be objectionable to consumers owing to the resulting taste and to excessive scaling in water pipes, heaters, boilers, and household appliances. However, it may also indicate elevated levels of ions that do pose a health concern, such as aluminum, arsenic, copper, lead, nitrate, and others. Water with extremely low concentrations of TDS may also be unacceptable to consumers because of its flat, insipid taste; it is also often corrosive to water supply systems. Water containing TDS concentrations below 100mg/L is usually acceptable to consumers, although acceptability may vary according to circumstances. The United States Environmental Protection Agency recommends treatment when TDS concentrations exceed 500 mg/L (US-EPA, 1997).

Small scale (Household water treatment)

HWT applications are any of a range of technologies, devices or methods employed for the purposes of treating water at the household level. HWTS has significant potential to reduce the burden of diarrheal disease by 35-40% which is twice as effective (47%) than improved wells, boreholes and communal stand pipes 27%. In treating diarrheal diseases (Fewtrell *et al* 2005 and Clasen *et al* 2007) HWTS helps vulnerable populations to take charge of their own water security by providing them with the knowledge and tools to treat their own drinking water. Good household water treatment and storage unit should be effective, simple system, easy to use and understand, keeps water stored safely, they should be acceptable to the consumer, adequate training, monitoring and maintenance, replacement (UNICEF, 2008).

2.3 Performance of Native Plant Species in Turbidity Reduction

It was reported in literature that plants have capability of turbidity reduction through their performance varies. Mehdinejad *et al.* (2009) compared efficiency of three plant species namely *Cicer arietinum*, *Moringa oleifera* and *Dolichos lablab* in different turbidity ranges by using 50 to 100 mg/L doses and they found that *Dolichos* reduced maximum turbidity among all coagulants used. It reduced up to 95.89% to 98.64% for highly turbid water which is almost as the same as the reduction capacity of alum. All the study on natural coagulants was efficient in higher-turbidity ranges than lower and medium turbidity water.

CHAPTER THREE: OBJECTIVE OF THE STUDY

3.1 General Objective

The main aim of this study was to evaluate contaminant removal performance of native plant species in terms of turbidity and microbial load.

3.2 Specific Objectives

- 1) To investigate the performance of *M. stenoptala* and *M. subcordata* as coagulant in the removal of turbidity for household water treatment

CHAPTER FOUR: MATERIALS AND METHODS

4.1 Study Area and Period

The study was conducted in Jimma University from February to April, 2013.

4.2 Study Design

Experimental study was carried out in the laboratory of Environmental health Science and Technology Department, Jimma University. The experiment was carried out using synthetic water and natural surface water. The natural surface samples were collected in and around Jimma town.

The bacteriological test of synthetic water was done based on the following diagram.

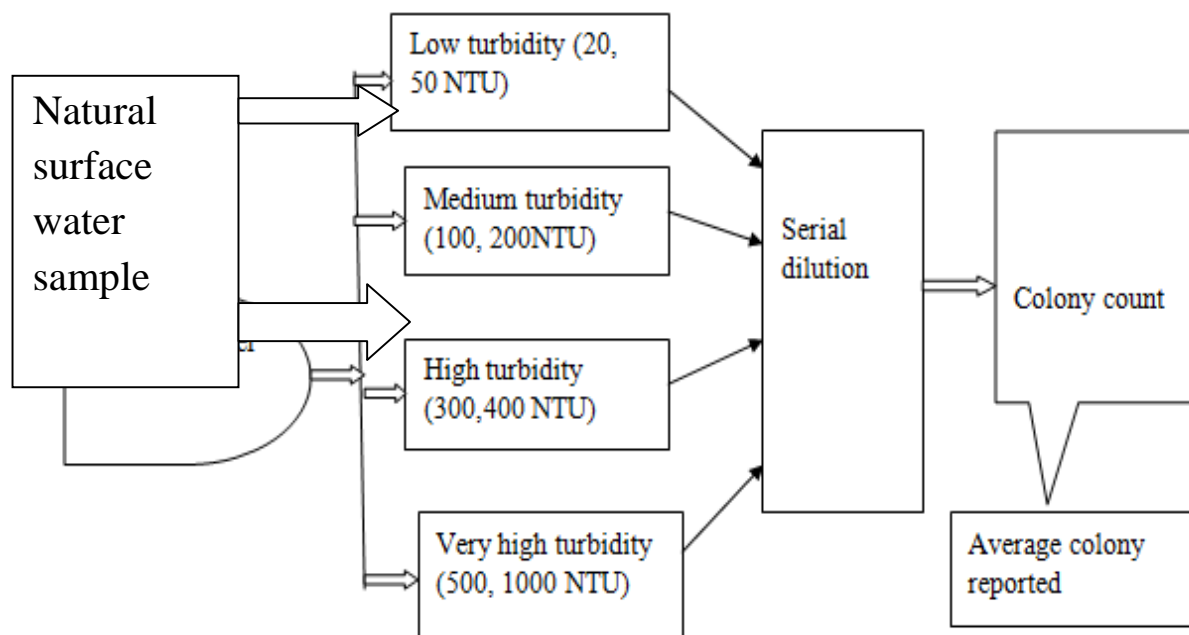


Figure 2. Bacteriological experimental study steps of synthetic water samples

4.2.1 Comparison of plants dose identified with alum & chlorine.

After identifying optimum dose compare with alum and chlorine standards

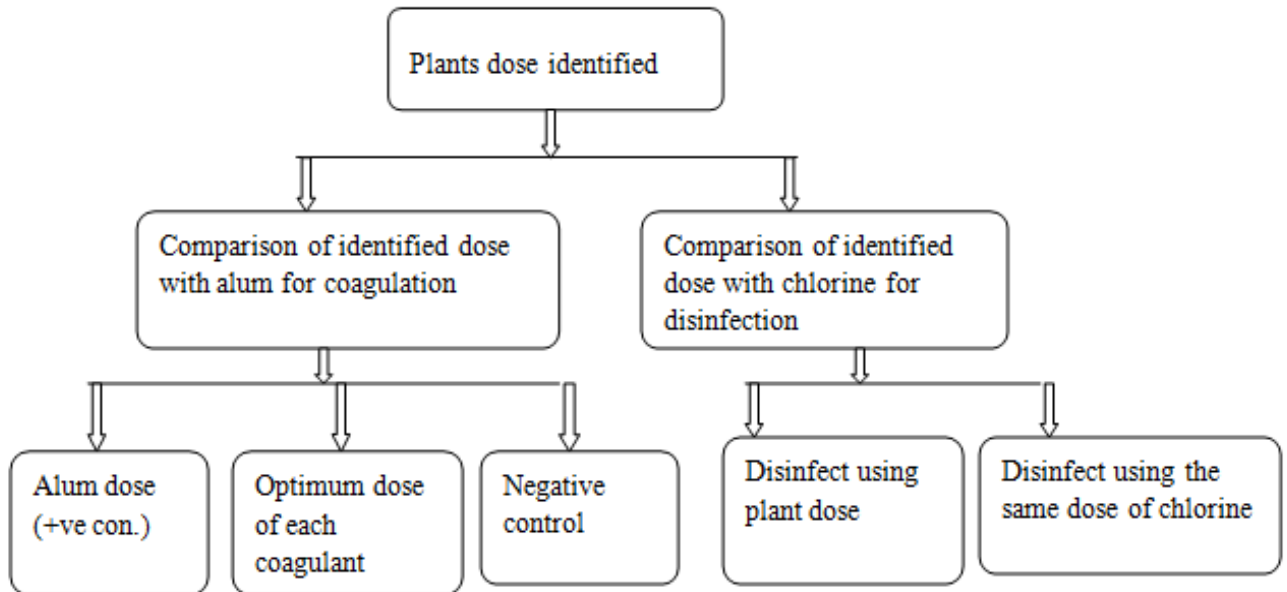


Figure 3. Comparison of identified plant dose with synthetic chemical

4.3 Study Variables

Dependent variables:

Turbidity
Microbial load

Independent variables:

Temperature
Dose of native plant species
Conductivity
pH
Contact time

4.4 Plant Collection and Identification

The plant used traditionally for water purification by local community was collected from selected rural. The plant materials were identified by comparison with the already preserved

specimens kept at the Herbarium in the Department of Biology, Addis Ababa University. The information collected for each plant species is summarized in (Table 2).

4.6 Preparation of Synthetic Water

Kaolin clay used for synthetic turbidity water preparation was collected from Awash Melkassa Aluminum Sulfate and Sulfuric acid factory private PLC. Synthetic turbid water was prepared by adding 10g of kaolin (clay suspension) to 1 liter of distilled water. The suspension was stirred for about 1 hour to achieve a uniform dispersion of kaolin particles. Then it was allowed to settle for 24 hours for complete hydration of the kaolin. After 24 hrs of settling, the turbid-water supernatant was decanted and used as a stable stock solution. This suspension was used as the stock solution for the preparation of turbid water samples desired to use by varying turbidity level for coagulation tests (Okuda *et al.*, 2001). The following turbidity ranges low (L) turbidity (0–125 NTU), medium (M) turbidity (125–250 NTU) and high (H) turbidity (250–375 NTU) are mostly used for coagulation experiment as suggested by Miller *et al.* (2008).

4.7 Natural Water Sampling Technique

4.7.1 Treatment of Sample Containers

Sampling was done with plastic containers. These were cleaned by washing with soap and tap water. The containers were disinfected with HNO₃ and finally rinsed with sterile distilled water several times (Kwame, 2009).

4.7.2 Samples Collection and Transportation

Five natural surface water samples namely Gibe, Ofole, Samiche, Kero and Dolollo with initial turbidities of 195 NTU, 45 NTU, 84 NTU, 22 NTU and 46 NTU were collected respectively in and around Jimma Town using sterilized plastic bottles based on the procedure of American Public Health Association standard (1998). Physico-chemical parameters like pH, turbidity, conductivity and temperature was measured at the sample site. The collected samples were kept in ice box and transported to Environmental Health Science and Technology Department Laboratory.

4.7.3 Storage of Samples

All the samples were temporarily stored in a cold box at the time of sampling until they are finally transferred into a refrigerator and stored at a temperature of below 4 °C (Kwame, 2009).

4.7.4 Jar Test Operation

Jar test is the most widely used experimental method for coagulation. A conventional jar test apparatus was used to achieve uniform agitation rate throughout the experiment for both synthetic water and natural surface water with powder native plant species coagulants. It was carried out as a batch test, accommodating a series of six beakers together with six-spindle steel paddles. For natural surface water, before operating the jar test the natural surface water sample was mixed homogeneously. Then, the water samples ought to be measured for physico-chemical; total coliform, fecal coliform, *E. coli* and heterotrophic bacteria count to represent an initial concentration. After the desired amount of coagulant is added to the water sample, agitation takes place, which consisted of (170 rpm) for two minutes followed by 40 rpm for 20 minutes. After the agitation being stopped, the suspensions were allowed to settle for 30 minutes. Effective dose at which the minimum or zero concentration of microbial loads is obtained and maximum turbidity removal point was recorded. Finally, the supernatant of the water sample was withdrawn using a pipette from the middle of the beaker for physico-chemical and bacteriological measurements which represent the final concentration. All tests were performed at an ambient temperature in the range of 20 – 25 °C and for different turbidity ranges.



Figure 4. Jar test apparatus setup

4.8 Sample Analysis

After keeping the agitated sample for the given time, the supernatant samples was collected from each of the six beakers using pipette for physico-chemical analysis in each jar test to reach coagulant and turbidity level. Moreover, for each coagulant and turbidity level, three triplicate jar tests were carried out in order to obtain reliable results (Gidde *et al.*, 2008).

Table 1 Methods and Instruments used for measurement of physico-chemical parameters of natural surface water sample.

S.N ^o	Parameters	Methods/ Measuring Instruments
1	pH	Wagtech International pH meter
2	Turbidity	Wagtech HANNA instruments micro processor Turbidity meter
3	Water temperature	Handheld thermometer
4	BOD	Titration method (Winkler's)
5	Conductivity	Multi-parameter probe(HACH)
6	TSS	Gravimetric Method
7	TDS	Gravimetric Method

4.9 Physico-chemical Analysis

4.9.1 Dose and Contact Time of Native Plant Species.

The effective dose of native plant species was determined or selected after the series of experiments using 0 gm/L, 0.01gm/l, 0.03 gm/L, 0.05 gm/L 0.07 gm/L and 0.09 gm/L dose. To evaluate the effective contact time, different sample was prepared and measured for every 30 minutes consecutively until it fulfills the WHO guideline i.e. < 5 NTU.

4.9.2 Turbidity Measurement

Digital Nephelometric Turbidity Meter Capable of measuring, turbidity from 0.1 NTU to 1000 NTU was used. Natural surface water sample was collected in the middle of the water column without disturbing, for onsite turbidity measurement of natural surface water and removal of turbidity was measured after a 30 minute settling period consecutively.

4.9.3 pH measurement

Both pH of synthetic water and natural surface water were measured before and after the experiment to know the change appeared from the initial after using the coagulant using pH-meter.

4.9.4 Conductivity and Temperature

Conductivity and temperature of both synthetic water and natural surface water was measured using Multi-parameter probe (HACH) and hand heated thermometer respectively.

4.10 Microbiological Analysis

4.10.2 Heterotrophic Bacteria

Spread plate method was used using R2A agar medium and incubates at 20-28 °C for 5-7 days or 35 °C \pm 0.5 °C for 48-72 \pm 2 hours.

4.11 Data Analysis

The data was recorded after each experiment, entered in to computer and analyzed by using Microsoft Excel 2007.

4.12 Quality Control

The procedure of the experiments was done consistently through the whole study to minimize the sources of error and all equipments were calibrated. Triplicate analysis of each parameter was done following the standard protocol in order to get satisfactory result. Moreover, controls were used for every triplicate analysis of each parameter during all the experiment.

4.13 Dissemination of the Result

The final result of the study was presented to Department of Environmental Science and Technology, College of Public Health and Medicinal Science, Jimma University and the result of the study will be published, either in national or international journals in order to reach at the scientific community.

4.15 Operational definitions and definition of terms

Safe water: Potable water free from harmful microorganisms and substances, even if it may have color, odor or taste problem.

Water borne disease: Disease acquired by drinking water contaminated at its source or in the distribution system, or by direct contact with environmental and recreational waters.

Point of use water treatment: systems refer to the treatment of water at the household level as opposed to centralized, larger capacity municipal or private systems that carry out treatment of water for a larger population.

Water related disease: diseases arise simply because of the lack of safe water for drinking and cleaning food. Others are spawned by inadequate sanitation facilities and poor personal hygiene practices that are directly related to a lack of safe water.

Surface water means all water which is open to the atmosphere and subject to surface runoff.

Turbidity: is the cloudiness or haziness of a fluid caused by individual particles (suspended solids) that are generally invisible to the naked eye.

Water disinfection: is the removal, deactivation or killing of pathogenic microorganisms

CHAPTER FIVE: RESULTS

5.1. Preliminary Dose Optimization

Dose optimization of coagulants was done on natural surface water that have initial turbidities of 25.6, 63.3 and 209.3 NTU by using the dose of the coagulant and as well as measuring all the parameters under the study to screen or select the effective coagulant in removing turbidity and microbes. Based on the performance of removing turbidity, the coagulants were selected by checking for each turbidity value. From the experiment conducted on different levels of initial turbidities with different coagulant of native plant species, their turbidity removal efficiency of the coagulants varied from native plant coagulant to coagulants using different dose for each coagulant at different initial turbidity value. From the correspondingly used coagulant dose the effective dose identified at which the effective turbidity removal performance the coagulant seen was 0.03 gm/L for all coagulants. The comparative percentage turbidity removal efficiency experiments to all native plant coagulants were performed for the three turbidity range of natural surface water samples by using the effective dose (0.03 gm/L) to select effective coagulant based on their turbidity removal performance.

The percent turbidity removal efficiency of *Sansevieria ehrenbergii* Schweinfurth, *Sansevieria forskaliiana* (Schult.f) Hepper & Wood, *M. subcordata*, *M. stenopetala* and control free at 0.03 gm/L dose with initial turbidity of 25.6 NTU for natural surface water were 9.76, 11.32, **80.85**, **54.68**, and 7.03% respectively (Figure 9).

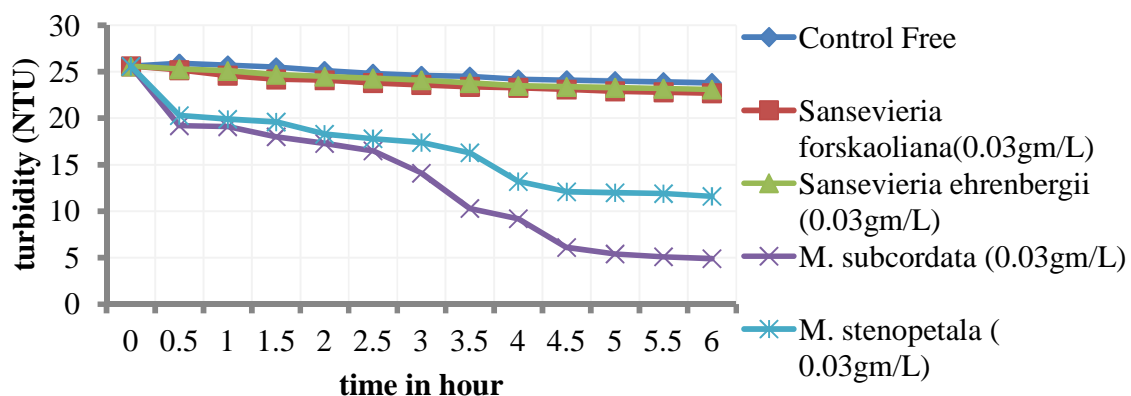


Figure 5. Comparison of coagulants based on turbidity reduction efficiency using natural surface water with initial turbidity of 25.6 NTU

The percent turbidity removal efficiency of *Sansevieria ehrenbergii* Schweinfurth, *Sansevieria forskaoliana*, *M. subcordata*, *M. stenopetala* and control free at 0.03 gm/L dose for natural surface water with initial turbidity of 63.3 NTU were 43.91, 39.33, **92.41**, **79.46** and 11.32% respectively (Figure 10).

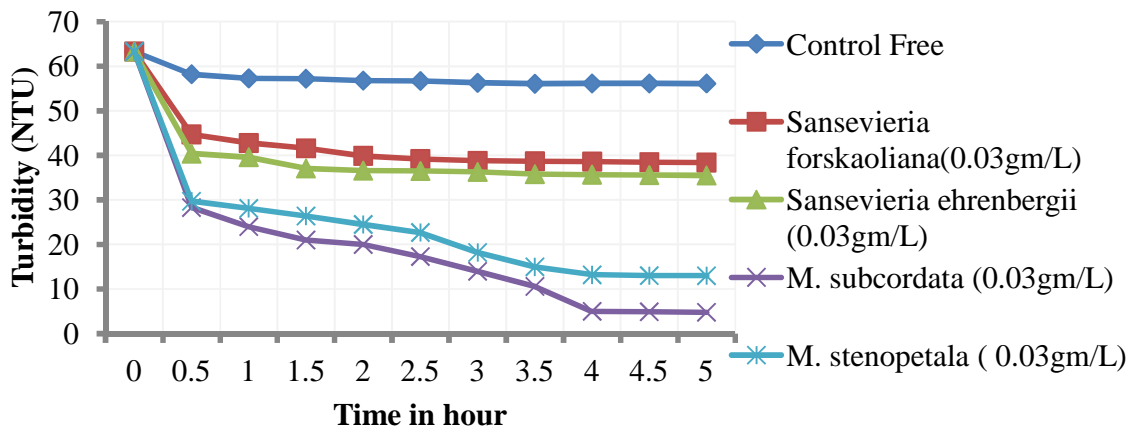


Figure 6. Comparison of coagulants based on turbidity reduction efficiency using natural surface water samples with initial turbidity of 63.3NTU

The percent turbidity removal efficiency of *Sansevieria ehrenbergii* Schweinfurth, *Sansevieria forskaoliana*, *M. subcordata*, *M. stenopetala* and control free at 0.03gm/L dose for natural surface water with initial turbidity of 209.3 NTU were 26.89, 23.93, **97.84**, **96.89** and 2.91% respectively (Figure 11).

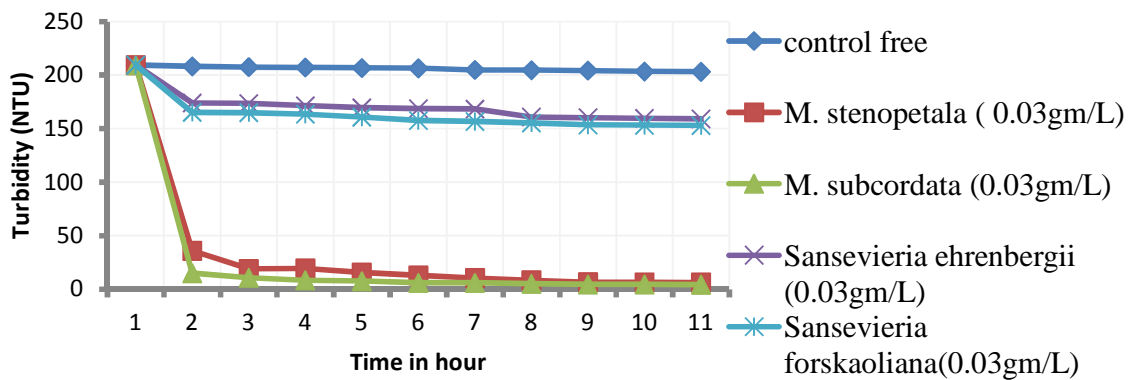


Figure 7. Comparison of coagulants based on turbidity reduction using natural surface water sample with initial turbidity of 209.3 NTU

Generally, as it can be seen in Figure 9, 10 and 11, among the four coagulants *M. subcordata* and *M. stenopetala* are very effective in removing turbidity from natural surface water when compared with *Sansevieria ehrenbergii* Schweinfurth and *Sansevieria forskoliana* that shows to select *M. subcordata* and *M. stenopetala* for detail study or further analysis. The performance of *Sansevieria ehrenbergii* Schweinfurth and *Sansevieria forskoliana* in removing turbidity was not effective.

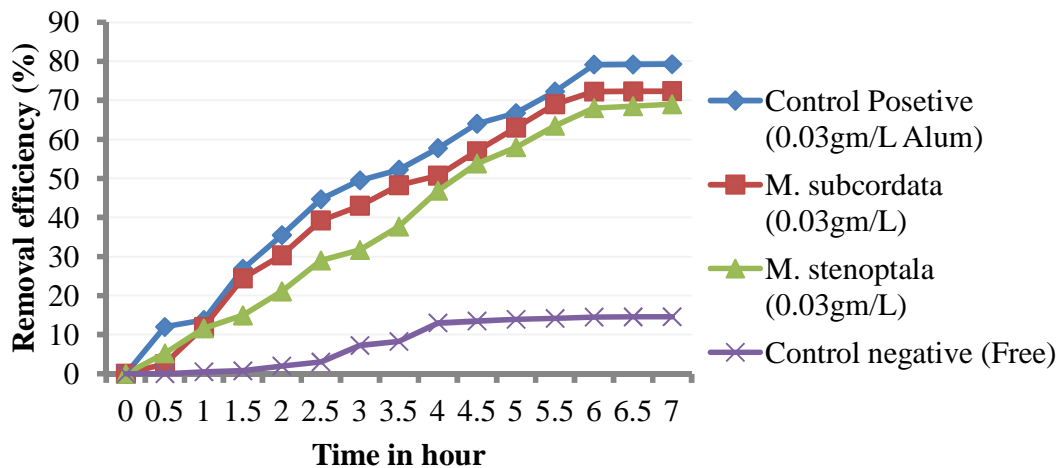


Figure 8. Removal efficiency of *M. subcordata* and *M. stenopetala* (0.01 gm/L) dose at initial turbidity of 20 NTU

Synthetic water with initial turbidity of 50 NTU, the effective dose of *M. subcordata* and *M. stenopetala* identified for effective removal of turbidity was 0.03 gm/L. This effective dose was compared with positive control (Alum) with the same dose from the corresponding dose used of both coagulants. After treatment using this coagulant the turbidity was decreased to 4.37 NTU, 6.71 NTU and 4.76 NTU for Alum, *M. stenopetala* and *M. subcordata* respectively. The turbidity removal efficiency of Alum, *M. subcordata* and *M. stenopetala* were 90.48%, 91.26% and 86.58% respectively (Figure 15).

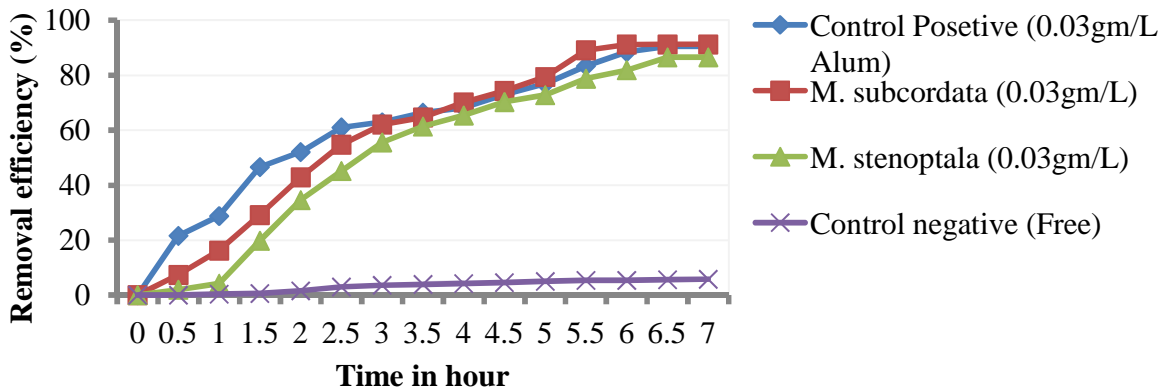


Figure 9. Removal efficiency of *M. subcordata* and *M. stenoptata* (0.03 gm/L) dose at initial turbidity of 50 NTU

5.2.2 Medium Turbidity (100 NTU, 200 NTU)

The synthetic water with initial turbidity of 100 NTU, the effective dose of *M. subcordata* and *M. stenoptata* identified for effective removal of turbidity were 0.03 gm/L and 0.05 gm/L respectively which were compared with positive control (Alum) of 0.03 gm/L dose from the corresponding dose used of both coagulants. The turbidity removal efficiency of Alum, *M. subcordata* and *M. stenoptata* were 94.23%, 95.38% and 92.2% respectively (Figure 17).

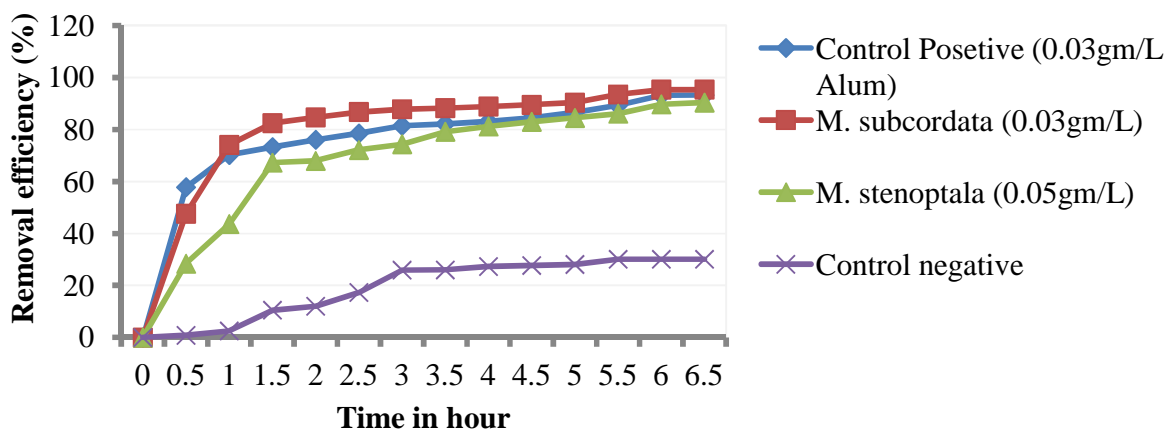


Figure 10. Removal efficiency of *M. subcordata* and *M. stenoptata* (0.03 gm/L and 0.05 gm/L) dose respectively for synthetic water with initial turbidity of 100 NTU

Synthetic water with initial turbidity of 200 NTU, the effective dose of *M. subcordata* and *M. stenopetala* identified for efficient removal of turbidity were 0.05 gm/L which was compared with positive control (Alum) of 0.05 gm/L dose from the corresponding dose used of both coagulants. The turbidity removal efficiency of Alum, *M. subcordata*, *M. stenopetala* and negative control were 97.34%, 97.5%, 96.28% and 12.92% respectively. At this turbidity range, the turbidity removal efficiency of *M. subcordata* was very effective even greater than the synthetic chemical (Alum) Figure 19.

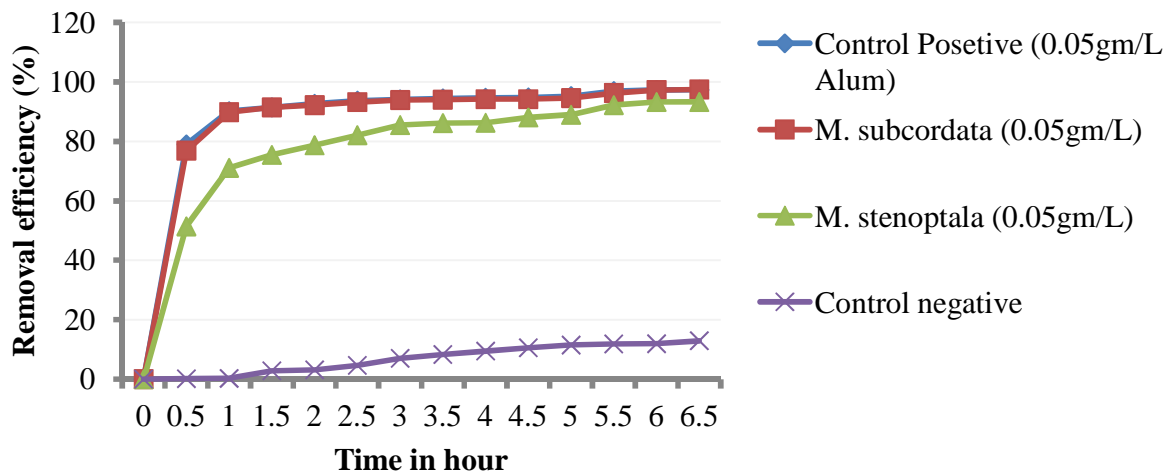


Figure 11. Removal efficiency of *M. subcordata* and *M. stenopetala* (0.05 gm/L) dose for synthetic water with initial turbidity of 200 NTU

The graph of Alum and *M. subcordata* was overlapped because of their turbidity removal efficiency at this turbidity range was not significantly difference when compared them.

5.2.3 High Turbidity (300 NTU, 400 NTU)

For synthetic water with initial turbidity of 300 NTU the effective dose identified from the corresponding dose applied for efficient removal of turbidity was observed at 0.05 gm/L dose for both *M. subcordata* and *M. stenopetala* which was compared with positive control (Alum) at the same dose. The turbidity removal efficiency of Alum, *M. subcordata*, *M. stenopetala* were 98.31%, 98.3%, and 97.69% respectively. Minimum turbidity removal was found for control free with turbidity of 264.65 NTU. At this turbidity range the turbidity removal efficiency of

Alum, *M. subcordata*, *M. stenopetala* coagulant was very effective and almost similar in performance to each other (Figure 21).

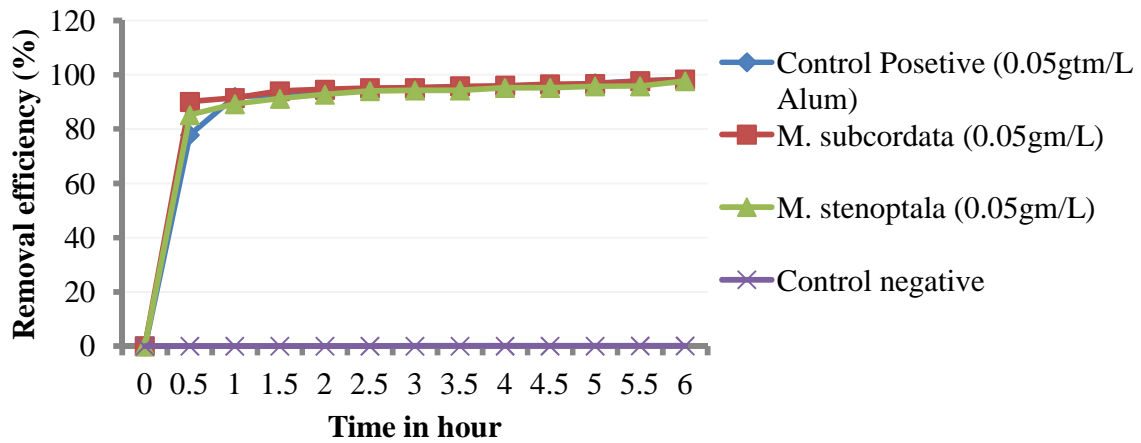


Figure 12. Removal efficiency of *M. subcordata* and *M. stenopetala* (0.05 gm/L) dose for synthetic water with initial turbidity 300 NTU

The graph of Alum, *M. subcordata*, and *M. stenopetala* was overlapped because of their turbidity removal efficiency at this turbidity range was similar.

Synthetic water with initial turbidity of 400 NTU the effective dose of *M. subcordata* and *M. stenopetala* identified for the efficient removal of turbidity were 0.05 gm/L dose for both coagulants which was compared with positive control (Alum) with the same dose. The turbidity removal efficiency in percent of Alum, *M. subcordata* and *M. stenopetala* and negative control were 98.73%, 98.9%, 98.55%. 28.1% respectively. This can indicate *M. subcordata* and *M. stenopetala* coagulant were very effective in removing turbidity as synthetic chemical

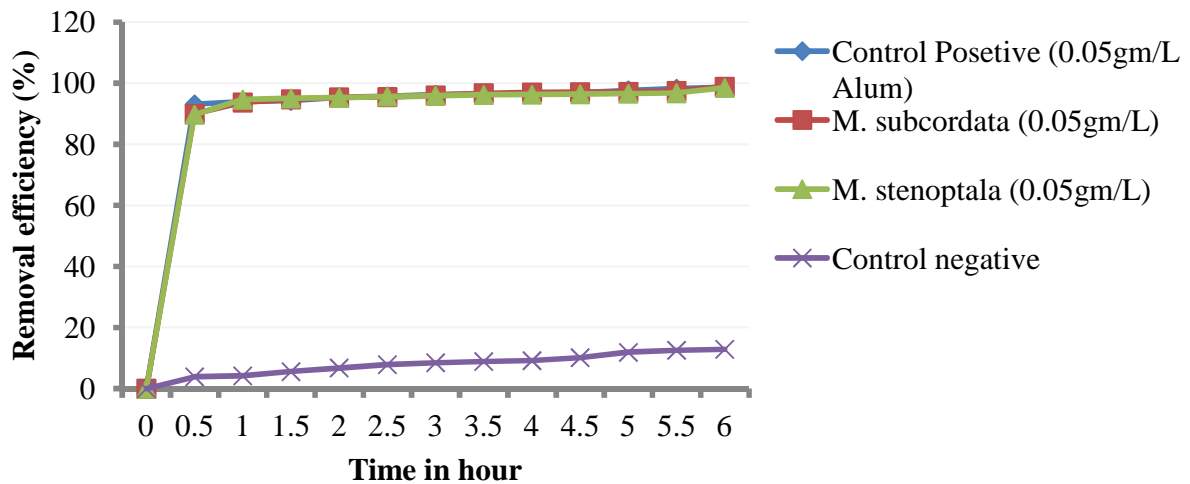


Figure 13. Removal efficiency of *M. subcordata* and *M. stenoptala* (0.05 gm/L) dose for synthetic water with initial turbidity of 400 NTU

The graph of Alum, *M. subcordata*, and *M. stenoptala* were overlapped because of their turbidity removal efficiency at this turbidity range were no significant difference.

5.2.4 Very High Turbidity (500 NTU, 1000 NTU)

Synthetic water with initial turbidity of 500 NTU the effective dose of *M. subcordata* and *M. stenoptala* identified for the efficient removal of turbidity was 0.07 gm/L as compared with positive control (Alum) of 0.07 gm/L dose from the corresponding dose used for both coagulants. The turbidity removal efficiency in percent of Alum, *M. subcordata*, *M. stenoptala* and negative control for this turbidity range were 98.91%, 99.11%, 98.99% and 2.05% respectively. Their turbidity removal performance of these two coagulants was almost similar and hence the graph line were overlapped (Figure 25). The result also indicates that as initial turbidity range increase the turbidity removal efficiency of both coagulants also increases in similar way.

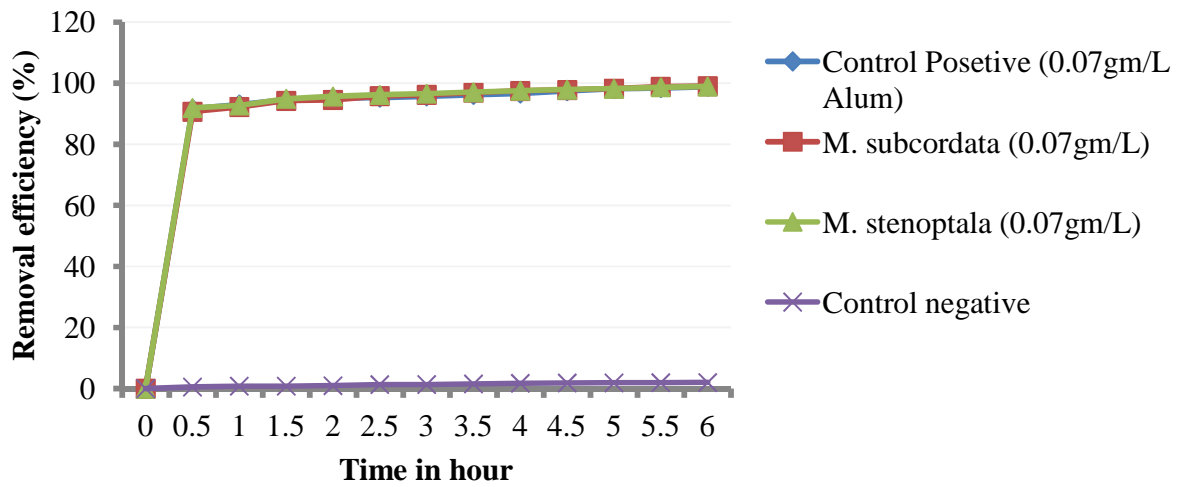


Figure 14. Removal efficiency of *M. subcordata* and *M. stenoptala* (0.07 gm/L) dose for synthetic water with initial turbidity 500 NTU

The graph of Alum, *M. subcordata*, and *M. stenoptala* was over lapped because of their turbidity removal efficiency at this turbidity range was no significant difference.

For synthetic water with initial turbidity of 1000 NTU the effective dose identified from the corresponding dose applied for effective turbidity removal were seen at 0.07 gm/L for both *M. subcordata* and *M. stenoptala* which was compared with positive control (Alum) dose of 0.07 gm/L. The turbidity removal efficiency in percent of Alum, *M. subcordata*, *M. stenoptala* and negative control were 99.45%, 99.49%, 99.41% and 3.68% respectively. Minimum turbidity removal was found for control free with turbidity of 963.2 NTU (Figure 27).

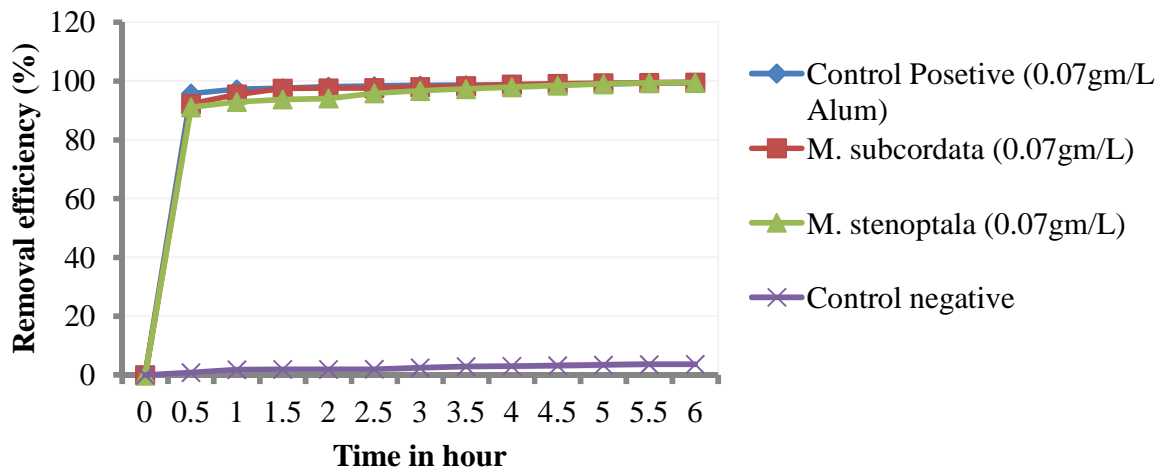


Figure 15. Removal efficiency of *M. subcordata* and *M. stenoptala* (0.07 gm/L) dose for synthetic water with initial turbidity 1000 NTU

The graph of Alum, *M. subcordata*, and *M. stenoptala* was overlapped because of their turbidity removal efficiency at this turbidity range was no significant difference.

The graph of Alum, *M. subcordata*, and *M. stenoptala* was overlapped because of their turbidity removal efficiency was no significant difference when coagulant dose was increased.

5.3 Microbial Removal in Synthetic Water

M. subcordata and *M. stenoptala* reduced the microbial loads (Total coliform, fecal coliform, and *E. coli* and Heterotrophic bacteria) like that of Cl_2 . For instance *M. subcordata* reduced total coliform from 175 colonies to zero at 20 NTU (Table 3 and 4).

Table 2 Removal of TC, FC and *E. coli* using *M. subcordata*, *M. stenopetala* powder and chlorine in colony counting form Synthetic water

Turbidity (NTU)	Microbial load before treatment in colony(initial)			pH of solution	Microbial reduction after treatment in colony															
					Negative control			pH of solution	<i>M. stenopetala</i> (shifera)			pH of solution	<i>M. subcordata</i> (gulf)			pH of solution	Positive control (Cl ₂)			pH solution
	TC	FC	EC		TC	FC	EC		TC	FC	EC		TC	FC	EC		TC	FC	EC	
20	175	180	179	6.9	174	175	161	6.8	0	0	0	7.3	0	0	0	7.4	0	0	0	6.7
50	189	185	189	7.2	172	177	179	6.7	2	0	0	7.4	0	0	0	7.4	0	0	0	6.9
100	188	176	195	6.8	177	172	174	7.1	0	0	1	7.1	2	0	0	7.2	1	1	0	7.2
200	200	197	187	7.1	174	181	172	6.9	3	1	0	7.2	0	0	0	7.5	0	0	1	6.8
300	200	198	199	6.9	181	188	189	6.8	1	0	2	7.3	3	1	0	7.4	1	0	0	6.8
400	175	197	199	6.7	169	186	187	7.2	0	1	0	7.4	1	0	1	7.2	1	0	2	7.2
500	199	185	189	7.2	188	183	174	7.3	2	0	2	6.9	0	1	0	7.1	0	1	2	7.6
1000	200	180	180	7.3	197	179	178	6.9	4	1	1	7.2	3	0	1	7.3	4	1	1	6.9

pH of the medium is 6.99 for TC=total coliform, 7.04 for FC=fecal coliform, 7.2 for EC=*E. coli*, (checked before sterilization).

Table 3 Removal of heterotrophic bacteria using *M. subcordata*, *M. stenopetala* and chlorine from Synthetic water

Initial turbidity (NTU)	Dose of <i>M. subcordata</i> (gulf) used(gm/L)	Dose of <i>M. stenopetala</i> (shifera) used(gm/L)	Dose of positive control (Cl ₂) used (gm/L)	Microbial loads before treatment incolony(initial)	pH of solution	Microbial reduction after treatment in colony							
						Negative control	pH of solution	<i>M. subcordata</i> (Gulf)	pH of solution	<i>M. stenopetala</i> shifera	pH of solution	Cl ₂	pH of solution
20	0.01	0.01	0.01	155	6.9	148	6.8	0	7.4	0	7.3	0	6.7
50	0.03	0.03	0.03	169	7.2	165	6.7	0	7.4	0	7.4	0	6.9
100	0.03	0.05	0.03	172	6.8	169	7.1	0	7.2	0	7.1	0	7.2
200	0.03	0.05	0.05	178	7.1	173	6.9	1	7.5	1	7.2	2	6.8
300	0.05	0.05	0.05	183	6.9	180	6.8	0	7.4	0	7.3	0	6.8
400	0.05	0.05	0.05	187	6.7	183	7.2	1	7.2	2	7.4	1	7.2
500	0.07	0.07	0.07	188	7.2	185	7.3	2	7.1	1	6.9	2	7.6
1000	0.07	0.07	0.07	189	7.3	187	6.9	4	7.3	5	7.2	2	6.9

pH of the medium is 7.05 (checked before sterilization)

5.4 Natural Surface Water Results

5.4.1 Natural Surface Water Physico-Chemical Characteristics

Five natural surface water samples namely Gibe, Ofole, Samiche, Kero and Dolollo were tested for physico-chemical water quality parameters on the day of testing before any treatment was initiated (**Table 6**). Initial turbidity of this natural surface water sample was ranged from 22 NTU to 195 NTU. The turbidity of natural surface water was found in the ranges of 20 NTU to 200NTU synthetic water desired in the laboratory to find the effective dose and contact time from the corresponding doses used.

5.4.2 Natural Surface Water Turbidity Removal

M. subcordata and *M. stenopetala* coagulant demonstrated adequate coagulation capacity for natural surface water almost with the same performance as synthetic water. The reduction of turbidity indicates because of the coagulation performance of *M. subcordata* tuber and *M. stenopetala* seed powder coagulants.

Natural surface water initially having minimum and maximum turbidities of 22 NTU and 195 NTU were treated with an optimum dose of 0.01 gm/L and 0.03 gm/L for both *M. subcordata* tuber and *M. stenopetala* seed powder. Both of the coagulants were very effective in removing turbidity when compared with the positive control (Alum) with the same dose. At optimum dosage, the percentage of turbidity removal was found to increase with increasing initial turbidity. The performance of positive control (Alum) in turbidity removal was also found to increase with increase initial turbidity. The observed percentage turbidity removal between *M. subcordata* tuber and *M. stenopetala* seed powder did not show significant difference on turbidity removal with positive control (Alum). The natural surface water pH, conductivity and temperature were not significantly changed when coagulants were added for water treatment.

Natural surface water with initial turbidity of 22 NTU the effective dose identified or desired in synthetic water from the corresponding dose applied for efficient removal of turbidity was seen at 0.01 gm/L for both *M. subcordata* and *M. stenopetala* which was compared with positive control (Alum) at 0.01 gm/L dose. The turbidity removal efficiency in percent of Alum, *M. subcordata* and *M. stenopetala* were 92.52%, 89.52% and 87.21% respectively.

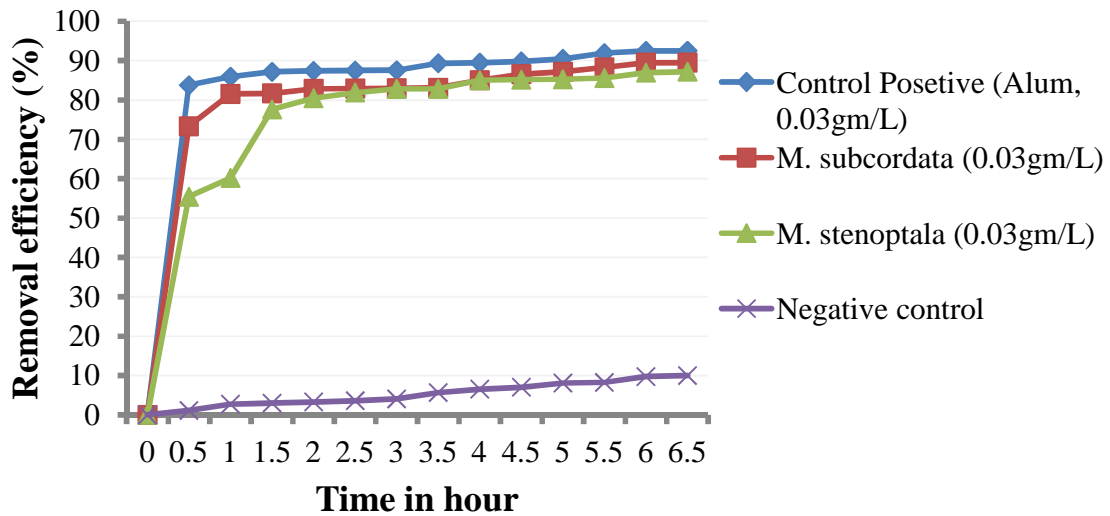


Figure 16. Removal efficiency of *M. subcordata* and *M. stenoptala* (0.01 gm/L) dose for natural surface water with initial turbidity 22 NTU

Natural surface water with initial turbidity of 45 NTU the effective dose identified or desired in synthetic water from the corresponding dose applied for efficient removal of turbidity was seen at 0.03 gm/L for both *M. subcordata* and *M. stenoptala* which was compared with positive control (Alum) at 0.03 gm/L dose. The turbidity removal efficiency in percent of Alum, *M. subcordata*, *M. stenoptala* and negative control were 90.06%, 93.28%, 90.53% and 11.66% respectively. The turbidity was decreased to 4.47, 3.02, 4.26 and 39.75 NTU after treating with Alum, *M. subcordata*, *M. stenoptala* and negative control respectively. This result was agreed with WHO standard for drinking water. Generally the performance of these two coagulants in turbidity removal was very effective as synthetic chemical coagulants (Alum). When this result was compared with synthetic water with 50 NTU turbidity range the efficiency was almost similar with each other except *M. stenoptala*. The percent removal of Alum, *M. subcordata*, *M. stenoptala* and negative control for synthetic water at turbidity of 50 NTU were 90.48%, 91.26%, 86.58% and 5.8% respectively. This description also represents the natural surface water Kero with initial turbidity of 46 NTU.

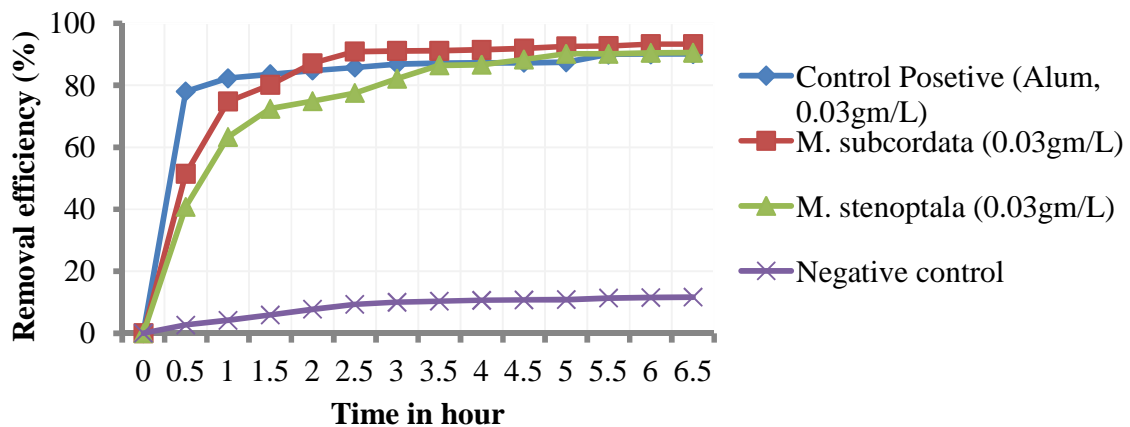


Figure 17. Removal efficiency of *M. subcordata* and *M. stenoptala* (0.03 gm/L) dose for natural surface water with initial turbidity 45 NTU

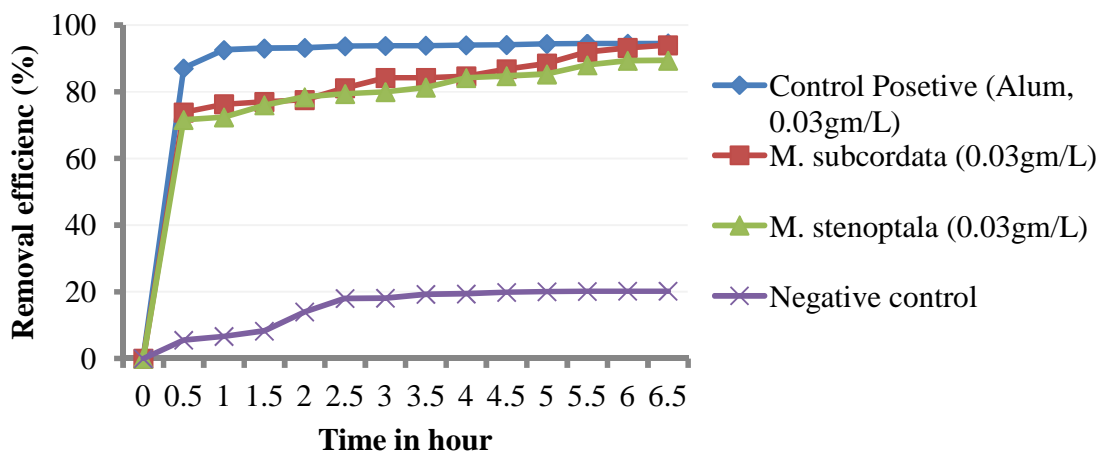


Figure 18. Removal efficiency of *M. subcordata* and *M. stenoptala* (0.03 gm/L) dose for natural surface water with initial turbidity 84 NTU, 4.83, 6.06 and 182.72 NTU after treatment with Alum, *M. subcordata*, *M. stenoptala* and negative control respectively. This turbidity level was agreed with WHO standard for drinking water only for *M. subcordata*, coagulant. Generally the performance of *M. subcordata* coagulants in turbidity removal was very effective as synthetic chemical coagulants (Alum). When this result was compared with synthetic water with initial turbidity of 200 NTU and the turbidity removal efficiency was almost similar with each other.

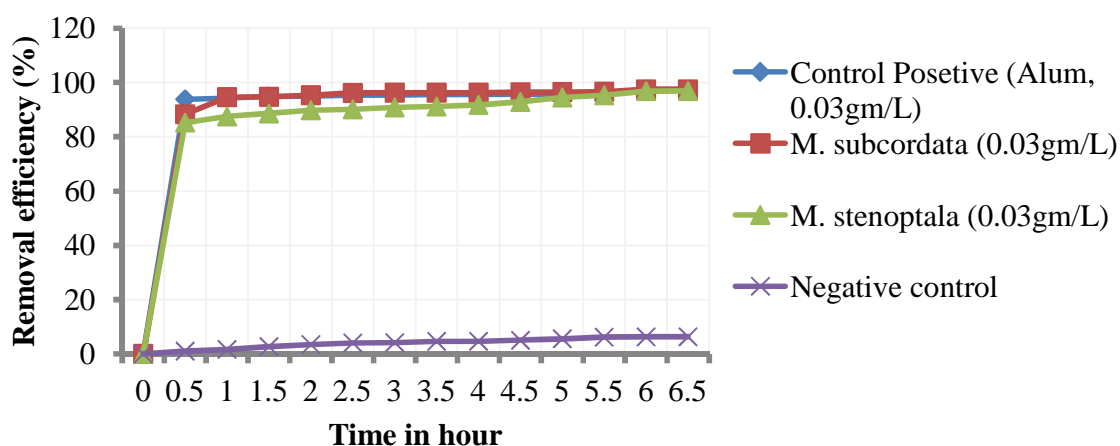


Figure 19 Removal efficiency of *M. subcordata* and *M. stenoptala* (0.03 gm/L) dose for natural surface water with initial turbidity 195 NTU

The line graphs of Alum and *M. subcordata* were overlapped because of their turbidity removal efficiency at this turbidity range was not significantly different.

Table 4 Turbidity removal comparison of *M. subcordata* and *M. stenoptala* with positive control (Alum) on natural surface water

Sample site mane	Initial turbidity(NTU)	<i>M.st.</i> (shifera) dose used gm/L	<i>M.su.</i> (gulf) dose used gm/L	PC(Alum) dose used gm/L	<i>M.st.</i> (shifera) Removal efficiency (%)	<i>M.su.</i> (gulf) removal efficiency (%)	PC (Alum) removal efficiency (%)
Gibe	195	0.05	0.03	0.03	96.89	97.52	97.42
Ofole	45	0.03	0.03	0.03	90.53	93.28	90.06
Samiche	84	0.03	0.03	0.03	89.41	94.05	94.54
Kero	22	0.01	0.01	0.01	67.39	74.89	75.86
Dolollo	46	0.03	0.03	0.03	87.21	89.52	92.52

M.st. = *M. stenoptala*, M.su. = *M. subcordata*, PC=positive control

5.4.4 Natural surface water microbial removal

Natural surface water microbial removal by using *M. subcordata* tuber and *M. stenopetala* seed was compared with the same water disinfected by chlorination after treatment. From Table 10 and 11 the microbial removal was observed for *M. subcordata* tuber and *M. stenopetala* seed powder as positive control. For all water samples tested for indicator microbial listed in the table after 24 hrs of safe storage, high microbial loads reduction was seen for *M. subcordata* tuber and *M. stenopetala* seed as chlorination. A comparison of data obtained revealed that at each sampling station for all water samples treatment using *M. subcordata* tuber and *M. stenopetala* seed powder the colony count ranged from 1 to 4 CFU. There was no significant difference on microbial loads reduction found between *M. subcordata* tuber and *M. stenopetala* seed powder and positive control treatment after 24 hrs of treated water storage for all of indicator microbial and heterotrophic bacteria

Table 5 Removal of TC, FC and *E. coli*. Comparison of *M. stenopetala*, and *M. subcordata* with control (chlorine) for natural water

Turbidity (NTU)	Microbial load before treatment in colony(initial)			pH of solution	Microbial reduction after treatment in colony															
					Negative control			pH of solution	<i>M. stenopetala</i> (shifera)			pH of solution	<i>M. subcordata</i> (gulf)			pH of solution	Positive control (Cl ₂)			pH of solution
	TC	FC	EC		TC	FC	EC		TC	FC	EC		TC	FC	EC		TC	FC	EC	
195	189	178	175	7.9	182	170	171	7.7	4	1	0	7.1	3	0	0	7.3	2	1	2	6.8
45	174	173	169	7.3	172	169	167	7.4	0	0	2	6.8	0	0	0	6.9	0	0	0	7.3
84	187	186	175	7.8	182	181	169	7.8	4	0	0	7.2	2	1	0	7.3	1	2	1	7.2
22	168	167	165	7.3	165	163	161	7.1	0	0	0	7.4	0	0	0	7.3	0	0	0	7.5
46	172	169	171	7.4	170	167	169	7.3	01	1	2	7.1	0	0	1	7.2	1	2	0	7.4

pH of the medium is 6.99 for TC=total coliform, 7.04 for FC=fecal coliform, 7.2 for EC=*E. coli*, (checked bore sterilization).

Table 6 Removal of heterotrophic bacteria Comparison of *M. stenopetala*, and *M. subcordata* with positive control (chlorine) for natural surface water

Initial turbidity (NTU)	Dose of <i>M. subcordata</i> (gulf)	Dose of <i>M. stenopetala</i> (shifera)	Dose of positive control (Cl ₂) used (gm/L)	Microbial loads before treatment in colony(initial)	pH of solution	Microbial reduction after treatment in colony							
						Negative control	pH of solution	<i>M. subcordata</i> (Gulf)	pH of solution	<i>M. stenopetala</i> shifera	pH of solution	Cl ₂	pH of solution
195	0.03	0.05	0.03	187	7.9	160	7.7	4	7.3	4	7.1	3	6.8
45	0.03	0.03	0.03	176	7.3	179	7.4	0	6.9	0	6.8	0	7.3
84	0.03	0.03	0.03	183	7.8	170	7.8	2	7.3	4	7.2	2	7.2
22	0.01	0.01	0.01	164	7.3	183	7.1	1	7.3	0	7.4	0	7.5
46	0.03	0.03	0.03	172	7.4	185	7.3	1	7.2	3	7.1	2	7.4

pH of the medium is 7.05 (checked before sterilization).

Microbial removal efficiency of these coagulants was also checked for initial turbidity level of 4 NTU following the same procedure used for higher initial turbidity level. The result found was efficient than the higher initial turbidity level (Table 12, 13). This indicates that the removal of microbes both from synthetic water and natural surface water was effective at low turbidity. But the removal of these microbes was either because of the coagulant was kill them or removed with the particles was need further experiment. This means the toxicity of the coagulant to microbial was need further investigations.

Table 7 Effectiveness of *M. stenopetala* and *M. subcordata* in removing TC, FC and *E. coli* comparing with positive control (chlorine) at 4 NTU

Turbidity (NTU)	Mi.load before treatment in colony(initial)			pH solution	Microbial reduction after treatment in colony															
					Negative control			pH solution	<i>M. stenopetala</i> (shifera)			pH solution	<i>M. subcordata</i> (gulf)			pH solution	Positive control (Cl ₂)			pH solution
	TC	FC	EC		TC	FC	EC		TC	FC	EC		TC	FC	EC		TC	FC	EC	
4	104	101	99	6.7	102	100	98	6.8	2	3	1	7.2	3	2	1	7.1	1	2	2	6.9

pH of the medium is 6.99 for TC=total coliform, 7.04 for FC=fecal coliform, 7.2 for EC=*E. coli* (checked before sterilization).

Table 8 Effectiveness of *M. stenopetala* and *M. subcordata* in removing heterotrophic bacteria comparing with positive control (chlorine) at 4 NTU

Initial turbidity (NTU)	Dose of <i>M. subcordata</i> (gulf) Used (gm/L)	Dose of <i>M. stenopetala</i> (shifera) used (gm/L)	Dose of positive control (Cl ₂) used (gm/L)	Microbial loads before treatment in colony(initial)	pH	Microbial reduction after treatment in colony							
						Negative control	pH	<i>M. subcordata</i> (Gulf)	pH	<i>M. stenopetala</i> shifera	pH	Cl ₂	pH
4	0.001	0.001	0.001	105	6.7	98	6.8	2	7.1	4	7.2	1	6.9

pH of the medium is 7.05 (checked before sterilization)

CHAPTER SIX: DISCUSSION

6.1. Preliminary dose optimization

Preliminary dose optimization is very important to identify the effective coagulant for further investigations. The preliminary experimental results of this study revealed that from the four native plant coagulants *M. stenopetala* and *M. subcordata* has high performance in turbidity removal efficiency when compared with the other two native plant coagulants (*Sansevieria ehrenbergii* Schweinfurth and *Sansevieria forskaoliana*). This may be due to the high content of coagulant in nature. The dose used to check the performance of turbidity removal for all native plant coagulants for this preliminary dose optimization were 0.03 gm/L for all natural surface water sample used for the study. The performance of *Sansevieria ehrenbergii* Schweinfurth and *Sansevieria forskaoliana* in removing turbidity was not effective; this may be due to their natural low content of coagulants. treatment. Over optimal amount coagulant could cause the aggregated particles to re-stabilize in the suspension and would also disturb particle settling (Diyaakaran and Sivasanakra, 2002 and

Alsameraiy, 2012).

In this experiment, the optimum dose found for low turbidity (20 and 50 NTU) was 0.01 mg/L and 0.03 mg/L by which (79% and 91%) turbidity reduction was achieved by powder of *M. subcordata*. Similar turbidity reduction (72% and 86.5%) was also exhibited by *M. stenopetala*. Diaz *et al.* (1999) found similar result by which extract of *Prosopis juliflora* reduced initial turbidity of 30 NTU to 5 NTU with optimum dose of 40 mg/L. In the same fashion for initial turbidity of 300 NTU and 400 NTU the optimum dose found for both *M. subcordata* and *M. stenopetala*, powder was 0.05gm/L with turbidity removal efficiency of 98.3%, 97.69% and 98.9%, 98.55% respectively. This result is nearly in agreement with Gide and Malusare *et al.* (2011) in which the protein extraction of *Moringa oleifera* powder reduced 96.33 % of 150 NTU and 98.51 % of 450 NTU with the dose of 40 mg/L and 100 mg/L. A slight difference of findings may be because of difference in *Moringa* seed extract species that seeds from different sources (geographic locations) exhibit varying coagulation performance (Nwaiwu *et al.*, 2012). Another study regarding *Moringa oleifera* showed the effectiveness of *Moringa oleifera* for

turbidity removals of up to 97% for high turbid water and lower removals of 86% for low turbidity water (58 NTU) Abaliwano *et al.* (2008).

The optimum dose found for initial turbidity of 500 NTU and 1000 NTU was 0.07gm/L for both *M. subcordata* and *M. stenopetala* powder with turbidity removal efficiency of 98.11%, 98.01% and 99.41%, 99.01% respectively. This result is in line with the finding of Zhang *et al.* (2006) where the optimum dosage of *opuntia spp.* used for turbidity removal of seawater (980 NTU) was 60 mg/L with removal efficiency of 99%. So, these natural coagulants (*M. subcordata* and *M. stenopetala*) might be considered as excellent option of traditional chemicals like alum and very efficient coagulants for high turbidity ranges. Gebremichael *et al.* (2009) also recommended the use of *Moringa* plant as coagulant in developing countries.

Turbidity removal efficiency of *M. subcordata* and *M. stenopetala* on natural surface water with initial turbidity of 22 NTU using the optimum dose (0.01gm/L) was 89.52% and 87.21%. When this value was compared with synthetic water with initial turbidity of 20 NTU it was greater in efficiency. This is may be due to the natural water was no interference which inhabited the performance of the coagulant. When the two values were compared they were almost similar in turbidity removal performance.

Turbidity removal efficiency in percent of *M. subcordata* powder on natural surface water with initial turbidity of 46 NTU and synthetic water with initial turbidity of 50 NTU were 89.52% and 91.26%, respectively. In the same fashion turbidity removal efficiency of *M. stenopetala* seed powder on natural surface water for initial turbidity of 46 NTU and synthetic water with initial turbidity 50 NTU was 87.21% and 90% respectively where as the turbidity removal efficiency of positive control (Alum) on natural surface water at 46 NTU and synthetic water at 50 NTU initial turbidity was 90.12% and 92.52% respectively. This result revealed that the turbidity removal efficiency of *M. subcordata* and *M. stenopetala* powder in synthetic turbid water was better performance than on natural surface water. This phenomenon probably is due to the fact that the surface water is likely to contain different substances like color, organic and inorganic compound, etc., which may inhibit the coagulation performance. The turbidity removal performance of *M. subcordata* and *M. stenopetala* for natural surface water with initial turbidity of 45 NTU and 46 NTU was different using the same dose of coagulant (0.03gm/L). This phenomenon is may be due to the nature of the natural surface water, i.e. in natural surface water

with 46 NTU initial turbidity there may be coagulation interference that decrease the efficiency of the coagulants to coagulate than natural surface water with initial turbidity of 45 NTU. The turbidity removal performance of *M. subcordata* and *M. stenopetala* for natural surface water with initial turbidity of 84 NTU and 195 NTU was 94.05%, 89.41% and 97.52%, 96.89% respectively. When this result was compared with synthetic water with initial turbidity of 100

6.3. The Relative Performance of Indigenous Plant Species as Disinfectant

With regards to microbial result the colony counts were drastically decreased with both *M. subcordata* and *M. stenopetala* powder treatments for both synthetic water and natural surface water (Table 5, 6, 10, 11 and 12). As the results of average colony count of bacteria showed there was no significant difference between *M. subcordata* and *M. stenopetala* powder treatment with respect to all types of bacteria (Total coliform, fecal coliform, *E. coli* and heterotrophic bacteria) as chlorine treatment in both natural surface water and synthetic water. In percent about 99.9% of microbial load removal was observed for both natural surface water and synthetic water after treating the water using these two coagulants.

The percentage microbial load reduction after treatment with *M. subcordata* and *M. stenopetala* for both synthetic and natural surface water was ranged from 97.6% to 99.9% for the first 0.5 hour. This finding is in agreement with the finding of Bina *et al.* (2010) who found effect of *Moringa oleifera* crude protein extract on microbial in different turbidities show rapid reduction of 99.2% – 99.97% was observed in the first 0.5 hour process. This might be due to *M. subcordata* and *M. stenopetala* powder treatment was reduced microbial with turbidity. This was supported by findings of Atieno *et al.* (2011) that the process of coagulation by *M. oleifera* extract removes about 90-99% of bacteria which are normally attached to the solid particles. Therefore, the use of *M. subcordata* and *M. stenopetala* powder can be considered advantageous and a promising step towards improving the processes of water coagulation to remove these microbial. For 24 hr observation period no regrowth of coliform and heterotrophic bacteria was observed. No significant difference was found on microbial reduction for all water samples with different turbidities before and after treatment using *M. subcordata* and *M. stenopetala* powder treatment for total coliform, fecal coliform, *E. coli* and heterotrophic bacteria respectively. The results of the microbial reduction seen in laboratory studies demonstrated that *M. subcordata* and *M. stenopetala* powder treatment was consistent with WHO drinking water guidelines and

USEPA standards of coliforms and *E. coli* concentration in suggesting that an effective treatment is possible under a wide variety of conditions. Finally, applying *M. subcordata* and *M. stenopetala* powder as household water treatment technology could have importance in developing countries where people are used to drink contaminated turbid water.

6.4. Optimum conditions of indigenous plant species for coagulation and disinfection

The optimum dose of the coagulant found for effective removal of turbidity and microbial in synthetic water was seen in the range of 0.01 gm/L to 0.07 gm/L and for the natural surface water the dose ranged from 0.01 gm/L to 0.03 gm/L. The pH and temperature of the water after treatment using the effective dose of the two coagulants was ranged 6.89 to 7.04, 25 °C to 27 °C respectively which shows almost neutral. The stirring time used for coagulation in this study was 170 rpm for 2 min and followed 40 rpm for 20 min and measurement of turbidity was done for every 30 min consecutively for 6 hr for each turbidity range both in synthetic and natural surface water. This stirring time was agreed with (Wang, 2002.) which says synthetic water samples (600 ml) were stirred at 125 rpm for 2 min and coagulants were added into the samples during this time. Then the samples were stirred at 70 rpm for 30 min. After the agitation, the samples would stand for 30 min and then the turbidity of the supernatant liquors was measured using a turbid meter (HACH 2100P).

CHAPTER SEVEN: CONCLUSION AND RECOMMENDATION

7.1 CONCLUSION

In general, the experimental result indicated that *M. subcordata* and *M. stenopetala* plants were very effective in reduction of turbidity and microbial load. The pH, conductivity and temperature of the water did not significantly changed as compared to chemical based coagulants when both *M. subcordata* and *M. stenopetala* coagulant was added for both synthetic and natural surface water after treatment. Since indigenous plant species has similar performance with synthetic chemical in removing turbidity and microbial, it can be concluded that *M. subcordata* and *M. stenopetala* has the potential to be utilized for household water treatment applications.

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ANNEXES

Annex 1 Steps for Preparation of synthetic water

- Synthetic turbid water was prepared by adding 10 g/L kaolin (Sigma Aldrich) to 1L distilled water solution and mixing thoroughly.
- The suspension was stirred using magnetic stirrer for 1hr to achieve uniform dispersion of kaolin particles, and then allowed to remain for 24hr for complete hydration of the particles.
- After 24 hrs of settling, the turbid water supernatant will be decanted and used as a stable stock solution.
- This stock solution was diluted with distilled water to achieve the desired turbidity
- The desired pH was attained by adding 1 M HCl or 1 M NaOH (Abaliwano, Ghebremichael and Amy, 2008).

4. 0.1 ml from each tube will be plated.

SAMPLE ANALYSES

- 1) Assemble filtering apparatus and filter and begin suction.
- 2). Filter a measured volume of well mixed sample through the glass fiber filter.
- 3). Wash with three successive 10 mL volumes of distilled water, allowing complete drainage between washings and continue suction for about 3 minutes after filtration is complete.
- 4) Remove the crucible and filter combination from the crucible adapter if a Gooch crucible is used.
- 5) Dry for at least 1hour at 103 to 105⁰C in an oven,
Cool in desiccators to balance temperature, and weigh.

5) Calculation

mg suspended solids/L = (A-B) x1000

ML sample

Where:

A= Weight of filter + dried residue, mg

B= Weight of filter, mg

Annex 2. Dissolved oxygen test step

The Azide Modification of the Winkler Method

- 1) Collection the sample in glass-stopper BOD bottle of 250-300 mL capacity. Write down the volume of the bottle.
- 2) Remove the glass stopper from the sample bottle, using a measuring pipet, add 1 ml of manganous sulfate solution followed by 1 ml alkali-iodide-azide reagent. Place the tip of the pipet below the surface of the water so as to allow the heavy solution to flow in without contact with the air
- 3) Stopper carefully to exclude air bubbles and mix by inverting the bottle a few times
- 4) Allow the resulting precipitate to settle at least to one half the bottle volume to leave clear supernatant above the manganese hydroxide floc.
- 5) Remove the stopper again, and with measuring pipet, add 1ml conc. Sulphuric acid
- 6) Re stopper carefully to prevent air from entering the bottle Mix by inverting several times until the precipitate completely dissolves and the brown or yellow color is distributed uniformly.
- 7) Titrate with 0.025 N sodium thiosulfate solutions a volume corresponding to 200 ml original sample after correction for sample loss by displacement with reagents. Thus for a total of 2 ml of reagents (1 ml each of MnSO₄ and alkali-iodide- azide reagents) in a 300-ml, titrate $\frac{200 \times 300}{298} = 201$ ml

298

- 8) Gradually add small portions of the sodium thiosulfate titrant while constantly swirling the liquid in the flask, until the sample changes to a pale yellow or straw color
- 9) Add a few drops of starch indicator solution and continue the titration to the first disappearance of the blue color.

10) Calculation

$$\text{Mg/L DO} = \frac{A \times N \times 8000}{\text{Ml of sample}}$$

Where:

A = ml sodium thiosulfate

N= Normality of sodium thiosulfate