

THE USE OF MORINGA SEED EXTRACT IN WATER PURIFICATION

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ABSTRACT

The high cost of treated water makes most people in the rural communities to resort to readily available sources which are normally of low quality exposing them to waterborne diseases. It is in this light that this research was carried out to confirm the effectiveness of powder extracted from mature-dried *Moringa oleifera* seeds which is commonly available in most rural communities of Africa. This was done using Completely Randomised Design with loading doses of 1, 2, 3, 4, and 6 g/l of the powder processed from *Moringa* seeds, and potash aluminium sulphate (alum) as coagulant. A control (water from the pond with only distilled water without alum and *Moringa* treatments) was also included. The turbidity, pH, and conductivity and total coliform were determined for all the samples. The turbidity for the samples ranged from $\log_{10}0.37$ to $\log_{10}1.00$ NTU while the conductivity ranged from $\log_{10}1.56$ to $\log_{10}2.86$ μ S/cm. The 6 g/l treatment of *Moringa* and 4 and 6 g/l potash alum treatments gave values that are acceptable according to the World Health Organization (WHO) guidelines for safe drinking water. The control sample gave the higher extremes values which are unacceptable. The pH values (7.29 to 7.89) obtained for the treatments were in the recommended range set by World Health Organization (WHO). Comparative studies with potash alum showed that the seed was effective in the sedimentation of inorganic and organic matter in raw water. It reduced the total microbial and coliform counts by 55% and 65%, respectively, after 24 hours whereas potash alum achieved 65% and 85% reduction under similar condition. The Most Probable Number per 100 ml for total coliform counts had values from 3 to 23 at 95% confidence limits. The *Moringa* treatment gave lower counts. Findings of this research lend support to earlier works recommending the use of *Moringa* for water treatment.

KEYWORDS: *Moringa oleifera*, Waterborne diseases, Coliform

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INTRODUCTION

Water is used for several purposes by humans but the level of purity of the water being consumed is very crucial since it has a direct effect on health. The conventional method of water purification using aluminium sulphate (alum) and calcium hypochlorite puts pressure on the nation's over-burdened financial resources since they are imported thereby making treated water very expensive in most developing countries and beyond the reach of most rural folks. Hence, they resort to sources such as dams, dug outs, streams, rivers, and lakes. Water from these sources is usually turbid and contaminated with microorganisms that cause many diseases including guinea worm and bilharzia. Inadequate water services together with sanitation to the rural poor are among the most serious challenges facing the developing world. Every year, approximately 3.4 million people die due to water-borne diseases, with the greatest health burden falling on children¹.

This situation persists and it will continue to cause substantial loss of human lives unless it is seriously dealt with at all levels. In the developing countries treatment plants are expensive, the ability to pay for services is minimal and skills as well as technology are scarce. In order to alleviate the prevailing difficulties, approaches are focus on sustainable water treatment systems that are low cost, robust and require minimal maintenance and operator skills. Locally available materials can be exploited towards achieving sustainable safe potable water supply².

Today, in most industrialized countries, drinking water is ranked as food, and high standards are set for its quality and safety³. The strict requirements for microbiological factors specify that bacterial content should be very low and that no pathogenic microorganisms should be detectable⁴. Guidelines and legislation state that drinking water should contain pathogenic microorganisms only in such low numbers that the risk for acquiring waterborne

infections is below an accepted limit⁵. The fulfilment of these requirements demands resource protection and careful treatment of raw water, as well as accurate quality control of the treatment process⁶.

Biological treatment processes at sewage treatment plants could produce selective elimination and/or changes of proportion, in the bacterial populations⁷. Moreover, the sewage effluent could modify some microbial populations in the reception waters, such as rivers, lakes, or lagoons⁸. This effect could become more important where policies of water re-utilization are applied in regions with poor water resources.

Drinking water treatment involves a number of processes depending on the quality of the water source, affordability and existing guidelines or standards. The cost involved in achieving the desired level of treatment depends among other things, on the cost and availability of chemicals. Commonly used chemicals for the various treatment units are synthetic organic and inorganic substances. In many places these are expensive and they have to be imported in hard currency. Many of the chemicals are also associated with human health and environmental problems^{9,10,11} and a number of them have been regulated for use in water treatment systems. Natural materials can minimize or avoid the concerns and significantly reduce treatment cost if available locally.

Naturally occurring alternatives to conventionally used chemical agents are generally considered as being safe and have therefore been investigated for decades; such agents include compounds found in plants¹². Of particular interest is the pan-tropical tree *Moringa oleifera*, since grounded seed powder have traditionally been used for the clarification of turbid drinking water in rural areas in the Sudan¹³. The fact however, that the active compounds from seeds can be extracted from the press-cake, an agricultural waste product accumulating after a high quality vegetable oil has been processed, has perhaps been one of the most significant findings that has driven the *Moringa* development as a whole¹⁴.

To ease the problems associated with chemical coagulants, several studies have pointed out the introduction of natural coagulants produced or extracted from microorganisms, animals, or plants^{15,16,17}. Among plant materials that have been tested over the years, the seeds from *Moringa oleifera* have been shown to be one of the most effective primary coagulants for water treatment or water purification. *Moringa oleifera* is a tropical plant belonging to the family of *Moringaceae*. *Moringa oleifera* is a non toxic¹⁸ and natural organic polymer. The tree is generally known in the developing world as a vegetable, a medicinal plant and a source of

vegetable oil¹⁹. The leaves and young seed of *Moringa oleifera* are rich in calcium, iron and vitamin C¹⁹, which serve as nutritious source for communities.

The active ingredients in aqueous extracts are dimeric proteins with a molecular weight of about 13 kDa and iso-electric point of between 10 and 11^{20,21}. Amino acid analysis and sequencing of *Moringa oleifera* showed high contents of glutamine, arginine and proline as well as total of other 60 residues²².²² studied the efficiency and properties of *Moringa oleifera* as a natural coagulant and its mechanism of coagulation on turbid water. They discovered that the active agents in aqueous *Moringa oleifera* are more effective coagulants than alum. Ion exchange column's test further showed that positive charges were a prerequisite for coagulation to be initiated. The zeta potential measurements also indicated that the predominant mechanism of the coagulation with *Moringa oleifera* appeared to adsorption and charge neutralization or adsorption and bridging of destabilized particles, the two assumed to take simultaneously²². They also revealed that *Moringa oleifera* can be either used in shelled or non shelled dry forms seeds, however, shelled seeds are more effective²². Furthermore, sludge produced by *Moringa oleifera* during coagulation is not only innocuous but also four to five times less in volume than the chemical sludge produced by alum coagulation²³.

Generally, coagulants are used for (physical and chemical) purification of the turbid raw waters. At very high turbidity the water can no longer be adequately treated by using filters. Coagulants have to be applied to transform water constituents into forms that can be separated out physically. In large scale treatment plants Aluminium Sulphate is used as a conventional chemical coagulant¹³.

As an alternative to conventional coagulants, *Moringa oleifera* seeds can be used as a natural coagulant (primary coagulant) in household water treatment as well as the community water treatment systems. Natural coagulant properties were found in six (6) different *Moringa* species by laboratory studies. The seed kernels of *Moringa oleifera* contain significant quantities of low molecular weight, (water soluble proteins) which carry a positive charge. When the crushed seeds are added to raw water, the proteins produce positive charges acting like magnets and attacking the predominately negatively charged particles (such as clay, silk, bacteria, and other toxic particles in water). The flocculation process occurs when the proteins bind the negatives forming flocs through the aggregation of particles which are present in water. These flocs are easily removed by settling or filtration. The material can clarify not only highly turbid

muddy water but also water of medium and low turbidity¹³.

Aluminum salts are by far the most widely used coagulants in water and wastewater treatment. However, several serious disadvantages of using aluminum salts including Alzheimer's disease and similar health related problems associated with residual aluminum in treated waters have been identified²⁴. There is also a problem of reaction of alum with natural alkalinity present in the water leading to a reduction of pH, and a low efficiency in the coagulation process^{25,26}. A significant economic factor is that many developing countries can hardly afford the high costs of imported chemicals for water and wastewater treatment. Therefore, it is desirable that other cost effective and more environmentally acceptable alternative coagulants be developed to supplement, if not replace alum, ferric salts, and synthetic polymers²³.

MATERIALS AND METHODS

Collection of Samples

The good quality seeds *Moringa oleifera* were obtained in Tunga Minna, Niger state. Good quality seeds were identified from those, which were not rotten, old, infected with diseases, brownish and dried once opened. The seed were de-shelled by hand and the de-shelled seeds were crushed and ground to a medium fine powder with a domestic food blender and sieved using a strainer with a pore size of 2.5mm² to obtain a fine powder.

Preparation of coagulant stock solutions

One gram (1g) of alum and 99ml of distilled water was weigh to make one percent (1%) solution, stirred for thirty minutes (30 min) using glass rod stirrer, to obtain the extract of the substance of potash alum and filtered with Whatman No.1 and 500ml of raw water was dispensed into a beaker and allow for one (1) hour for the dissolved of the solution. And the period of dissolving the equivalent weighs to dissolve easily to know whether the purification and the coliform found in the water sample can be reduces to the very minimal by testing the prepared sample against the media prepare by monitoring the growth in the incubator at 37 and 44°C for 0, 24, 48 and 72 hours.

Effect of *Moringa* on Sedimentation of Total Solids

A clean platinum evaporating dish was dried in an oven at 103°C for one (1) hour until a constant weight was obtained after cooling in desiccators. A 250ml volume of Chanchaga River water sample was thoroughly mixed, from which 100ml was measured into the dish and evaporated in the oven for one (1) hours at 103°C. It was then cooled in the desiccators and weighed. The total solids in ppm were calculated as:

$$\frac{\text{Increase in weight (gm x 1,000,000)}}{\text{ml of sample}}$$

To test the comparative effect of potash alum and *Moringa* powder, 100mg of each substrate weighed into a litre of the river water sample and thoroughly mixed. The total solids contents at 0, 24, 48 and 72 hours were determined as described above by using 100ml from the treated water sample.

Effect of *Moringa* on Total Bacterial and Coliform Load

A total bacterial and coliform count of the raw water was estimated by serial dilution using pour plate method. One milliliter of a 10⁻⁴ dilution was plated on duplication plates of Yeast Extract Agar and incubated at 37°C for 48 hours after which the total coliforms were enumerated on Eosin Methylene Blue agar.

At 0, 24, 48, and 72 hours, 1 ml was carefully measured from the treated portion of water layer, serially diluted and enumerated on the media. The same procedure was followed for potash alum treated water, tap water and distilled water.

Experimental Procedure

A Completely Randomised Design was used for this experiment. The treatments given were the varying concentrations of potash alum, powder produced from *Moringa* seeds, and the control (no potash alum or *Moringa* powder). Each treatment effect on the response (turbidity, pH, conductivity, COD and total coliform counts) was repeated 4 times except the total coliform count which was carried out in triplicates.

Sample Preparation

Twenty (20) litres of sample was fetched from Chanchaga River. This was further dispensed into 25 beakers. The volume of sample in each beaker was 500 ml.

Five different concentrations of the stock solutions for the loading dose were prepared by weighing 1.0, 2.0, 3.0, 4.0 and 6.0 g of potash alum and *Moringa* powder separately into a beaker containing 500 ml of raw water. The mixtures in the beakers were stirred using a glass rod to obtain a clear solution. A 500 ml of raw water with no potash alum or *Moringa* powder was kept as the control treatment.

Total Coliform Using Most Probable Number (MPN) Procedure

In determining the most probable number of coliforms that were present in each of the treated water samples, the multiple tube fermentation method was adopted. Lactose broth was used as the medium for the bacteria growth. Two types of the lactose broth were prepared. These were the single strength lactose broth (SSLB) and the double strength lactose broth (DSLb).

In the single strength, 13.0 g of the lactose powder was weighed and dissolved in 1000 ml of distilled water. The

solution was then stirred gently for 10 min on a glass rod stirrer to dissolve and mix well.

The double strength was prepared using exactly a double of each of the weights of the reagents used. This solution was put on a glass rod and stirred gently for 10 min.

A volume of 1.0 ml of the control, 4.0 and 6.0 g of both *Moringa* and potash alum treatments supernatants were measured and introduced into test tubes containing 10 ml of the double strength lactose broth and 10 ml of the single strength lactose broth. Another volume of 0.1 ml of the same supernatants above was measured and introduced into another set of test tubes containing 10 ml of the single strength lactose broth. The test tubes were then incubated for 24 hours at 37°C after which they were analysed. The results obtained were compared with ²⁷ to obtain the most probable number at 95% confidence level.

RESULTS AND DISCUSSION

In Figure 1, the turbidity values ranged from $\log_{10}0.37\text{NTU}$ to $\log_{10}1.00\text{NTU}$ for all the treatments used. Turbidity may be caused when light is blocked by large amounts of silt, microorganisms, plant fibres, sawdust, wood ashes, chemicals and coal dust. Any substance that makes water cloudy will cause turbidity. The treatments used gave significant differences ($p < 0.001$) on turbidity. The declared WHO guideline for conductivity provided for safe drinking water is 5NTU ($\log_{10}0.700\text{NTU}$). The 4.0, 6.0 g of potash alum, 6.0 g of *Moringa* and tap water of 6.0 g give treatments recorded values that were acceptable according to the guideline for drinking water (Fig 1).

In Figure 2 and 3, conductivity which is a measure of total dissolved solids (TDS) in water varies considerable in different geographical regions owing to differences in the solubility of minerals; hence there is no standard value for it but high levels of it in drinking water maybe objectionable to consumers therefore, the quality of water for consumption for rural communities in Minna can be improved by first adding powder from *Moringa* before the generally recommended "boil before use" strategy. As expected, the control treatment gave the highest turbidity value of $\log_{10}1.04\text{NTU}$. It is clearly seen that higher concentrations of *Moringa* powder of 6.0 g/500 ml loading dose as coagulant gives similar effect on turbidity compared with alum of loading doses of 4.0 g/500 ml and 6.0 g/500 ml. This shows that *Moringa* can be adopted for water purification. This will likely to lead to cost reduction in the conventional water treatment using alum and no threat to human life in case of overdose.

In Figure 4, at 95.0% confidence level, there was significant difference ($p < 0.001$) among all the

treatments at the varying loading dose concentrations on the pH. The conductivity ranged from $\log_{10}1.56$ to $\log_{10}2.86\mu\text{S/cm}$ for the varying concentrations of all the coagulum used. The F-test p-value less than 0.001 indicate that there are significant differences among the means due to the concentrations of the different coagulants use. The conductivity value of $\log_{10}2.86\mu\text{S/cm}$ recorded for the control was extremely high indicating the presence of dissolved impurities (Fig 3). This indicates that turbid water which is allowed to stand with no treatment is an inadequate procedure for removing dissolved and floating particles. It could be efficient if the turbid water is left to stand for a very long time. The conductivity measurements followed a similar pattern as the turbidity measurements. Increasing concentrations of both the potash and *Moringa* treatments led to decrease in conductivity values. It can be deduced that higher loading dose other than the ones used for *Moringa* in this work can be adopted to decrease water conductivity and turbidity in water meant for consumption in most rural communities in Minna. The *Moringa* treatment values ranged from $\log_{10}1.15$ to $\log_{10}2.38\mu\text{S/cm}$; and $\log_{10}2.56$ to $\log_{10}2.70\mu\text{S/cm}$ when the alum was used.

The recommended acceptable range of pH for drinking water specified is between 6.0 and 8.0. The treatments gave a range of 6.92 to 8.06 (Fig 4) which falls within the reduced as the concentrations of the dosing solutions were increased for *Moringa* and tap water. Whereas the potash alum give the treatment range from 9.33 to 11.78 with much alkalinity level which mean it's require much water to be treated. This could be explained by the fact that the solutions were becoming more acidic. This was attributed to the fact that the potash alum in the treatment procedure produced sulphuric acid which lowered the pH levels. The increase in acidity could be due to the trivalent cation aluminium which serves a Lewis acid. Thus it can accept a lone pair of electrons. The reverse was observed with the *Moringa* treatment. The pH increases with increasing concentrations of the *Moringa* coagulant. It has been reported that the action of *M. oleifera* as a coagulant lies in the presence of water soluble cationic proteins in the seeds. This suggests that in water, the basic amino acids present in the protein of *Moringa* would accept a proton from water resulting in the release of a hydroxyl group making the solution basic. This accounted for the basic pH values observed for *Moringa* treatments compared with alum treatments. Significant degrees of sedimentation of total dissolved solids were effected by the *Moringa* seed extract, comparative to potash alum. After a holding time of 24 hours, 37% reduction was achieved by the seed extract as

against 50% recorded by potash. These corresponded with 58% and 65% reduction in the seed extract and potash alum respectively in (Fig. 5).

Similarly, the bacterial load of raw water treated with the seed and alum were reduced after 24 hours by 55% and 65% respectively. The coliform populations of raw sample were also reduced by 65% and 85% by the seed extract and potash alum (Fig. 6, 7).

A lower count of 3 for the Most Probable Number (MPN) of total coliform per 100 ml with both the 4.0 and 6.0g *Moringa* treatment and the conventional recorded a count of 23 coliforms per 100 ml (Table 1). This supports findings that suggest that the process of flocculation removes about 90 -99% of bacteria which are normally attached to the solid particles. The control treatment had the highest counts of coliform (150/100 ml). This affirms earlier stated recommendation above that raw water without treatment is not safe for drinking.

CONCLUSION

The results obtained showed that powder from seed kernels of *Moringa oleifera* contains some coagulating properties at loading doses of 6g/500ml and above that have similar effect as the conventional coagulum, potash. This lends support to earlier findings of the use of powder processed from *Moringa* seeds as a coagulant in water purification system. *Moringa* coagulum has an added advantage of having antimicrobial properties. Considering the fact that *Moringa* coagulum can be locally produced, its use in water purification should be encouraged. This is likely to reduce the high cost of the current water treatment systems.

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Table 1. Total coliforms counts per 100ml of the samples

Sample	DSLB 10ml	SSLB 1ml	SSLB 0.1ml	MPN/100ml
P3	3	0	0	23
M3	0	1	0	3
P4	3	0	0	23
M4	1	0	0	4
C	3	2	1	150

Source: Most Probable Number (MPN) at 95% confidence level DSLB Double Strength Lactose Broth, SSLB Single Strength Lactose Broth

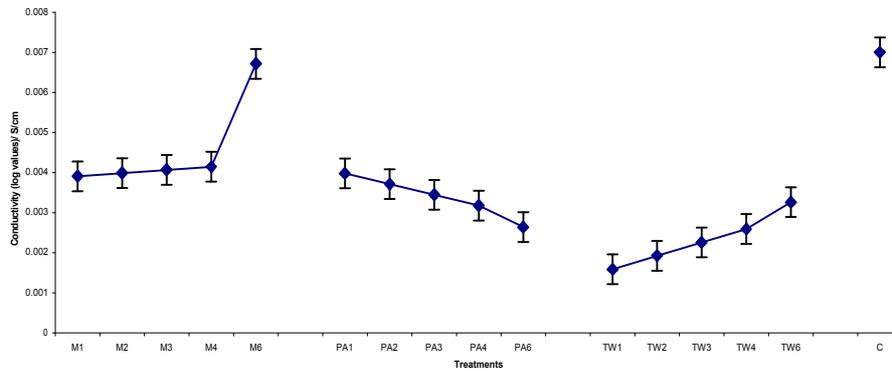


Figure 1: Effect of moringa, potash alum and tap water as coagulant at varying concentration on conductivity of pond water at river Chanchaga.

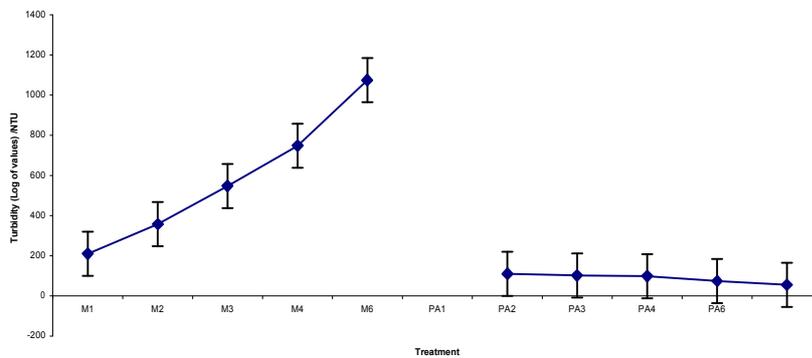


Figure 2 Effect of moringa (M) and potash alum (PA) as coagulant at varying concentration on turbidity of pond water at river Chanchaga

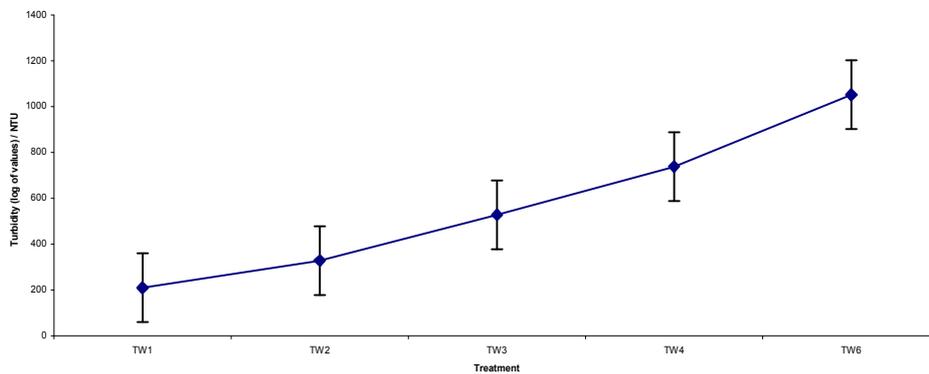


Figure 3: Effect of tap water as coagulant at varying concentration on turbidity of pond water at river Chanchaga

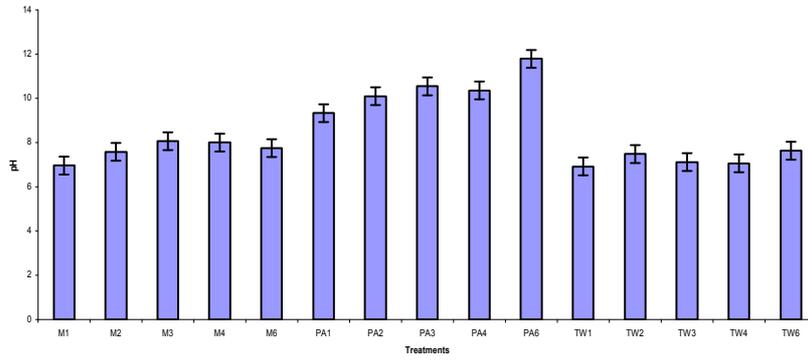


Figure 4: Effect of moringa (M) potash alum (PA) and tap water as coagulant at varying concentration on pH of pond water (TW) at Chanchaga river

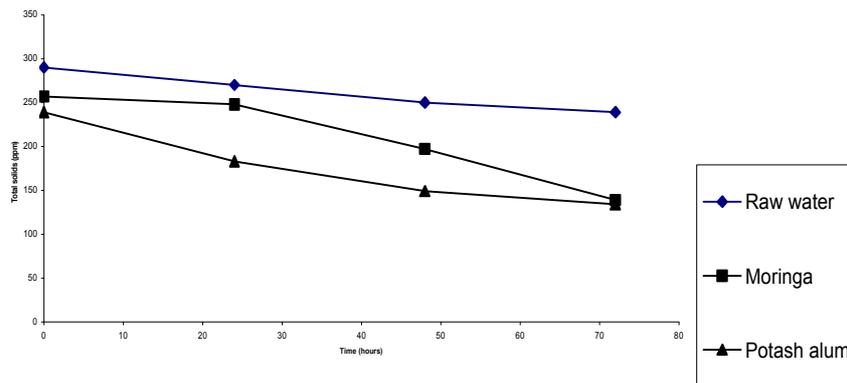


Figure 5: Sedimentation rate of Total solids in water treated with moringa seed and potash alum.

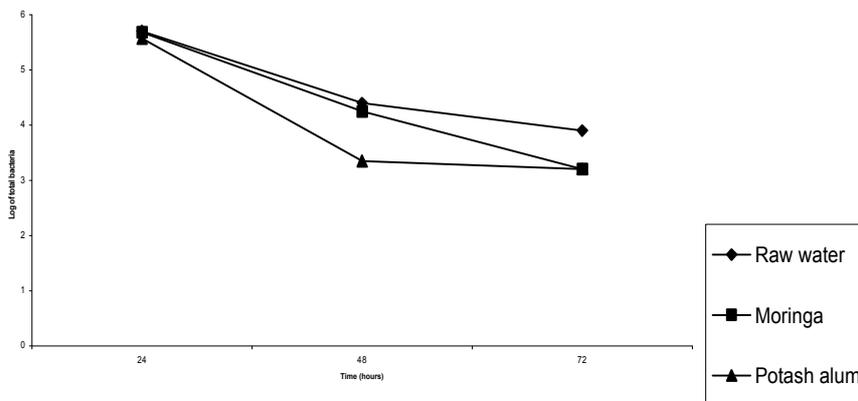


Figure 6: Effect of moringa seed and potash alum on total bacteria in Chanchaga river water.

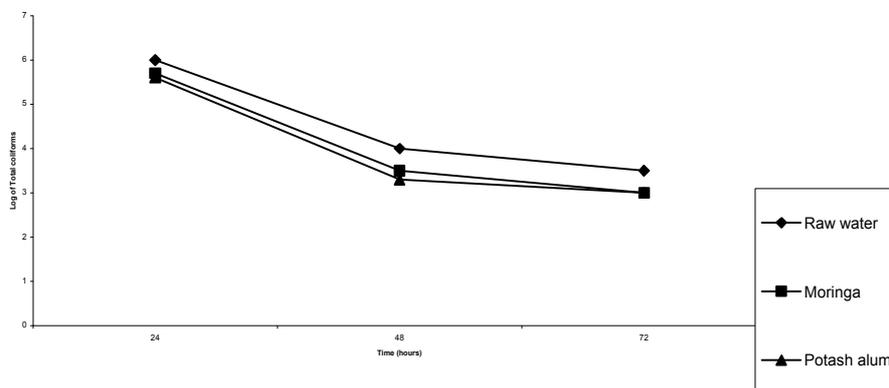


Figure 7: Effect of Moringa seed and potash alum on total coliforms in Chanchaga river water

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