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Evaluation of Natural-Based Coagulants Efficacy in Removal of Cyanobacteria from Huruma Wastewater Samples, Eldoret, Kenya

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Abstract

There is water deficit and quality crisis in the world due to water pollution mainly by eutrophic nutrients and heavy metals. Eutrophication is the enrichment of aquatic ecosystems by nutrients resulting in an increase in growth of macrophytes such as duckweed and phytoplanktons such as cyanobacteria that lead to degradation of the water quality. Cyanobacteria bloom changes the odor and taste of water with some producing potent toxins which lower water quality. The objective of this study was to evaluate the efficacy of locally available natural-based coagulants namely, *Moringa oleifera* seeds, *Aloe barbadensis miller* leaf and *Cactus Opuntia stricta* leaf in removal of cyanobacteria from wastewater samples collected from Huruma wastewater stabilization ponds, Eldoret. Wastewater samples were collected from the outlet of the last maturation pond at Huruma wastewater treatment plant, where the final effluents are released to river Sosiani. Physical chemical parameters were determined using multitester pH meter, phosphate were analysed using ammonium molybdate method while nitrate were analysed using brucine method. Cyanobacteria biomass was determined using chlorophyll *a* analysis method. Means of the mentioned parameters were calculated and analyzed using ANOVA and significant means separated using Tukey's test at 5% level. There were significant differences in reduction of physical chemical parameters and nutrients by the locally based coagulants (P = 0.00). The range of cyanobacteria removal efficiency by the coagulants was as follows; *Moringa oleifera* 62%, *Aloe barbadensis miller* leaf 47.3% and *Cactus Opuntia stricta* leaf 52.2%. These coagulants can be used to treat wastewater contaminated by cyanobacteria.

Keywords: Eutrophication, cyanobacteria, moringa, aloe vera, cactus

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Competing interests:
The authors have declared that no competing interests exist

Introduction

Water scarcity is the worldwide risk of greatest concern for humankind and nations (Water, 2016). There is increased demand for water resources as the population tries to acquire water to meet its diversified needs. Approximately 1.2 billion people are unable to acquire safe drinking water (WHO, 2016). Poor urban planning and weak implementation of environmental policies has escalated this problem. This has caused major constraints on the scarce resources besides polluting the available water resources. Polluted water causes detrimental effects on humans who consume it and on aquatic life. The discharge of toxic effluents from various industries, agriculture and domestic sources adversely affects water quality, aquatic organisms and soil fertility, affecting the structure, functions and integrity of ecosystems (Woodward, *et al.,* 2012).

Treatment and reuse of wastewater has become widespread in the world as the source of fresh water diminishes. Wastewater is currently being viewed as an alternative water source that can be treated and reused for various purposes. In this respect, the plight of wastewater is no longer a menace but a solution to the problem of wastewater disposal and water scarcity. Globally, treated wastewater reuse is gaining acceptance (Rebah *et al.,* 2017). Wastewater treatment is paramount in order to recycle the large amount of effluents that are released by the current high human population and the growing urbanization and industrialization (Woodward, *et al.,* 2012).

Various methods are used in the treatment of wastewater including chemical, physical and biological treatments or by using artificial membranes. In the physical and chemical treatment of the wastewater, coagulants are used which play a vital role. Coagulation is an indispensable method which promotes aggregation of pollutants including suspended solid particles (Narmatha *et al.,* 2017). The physicochemical processes such as coagulation and flocculation minimizes pollution and provide clean water for reuse (Manhokwe & Zvidzai, 2019). Chemicals such as aluminium sulphate are commonly used as coagulants for water treatment. However, the use of such chemicals has side effects which include high cost of operation, release of toxic substances to the environment and health effects on humanity (Rebah *et al.,* 2017; Vishali *et al.,* (2015). As a result, natural coagulants are preferred over chemical coagulants as they are locally available, cost effective, eco-friendly and less toxic to the environment (El Bouaidi *et al.,* 2020). The use of natural coagulants in water treatment is paramount for safe and effective water treatment (Nery *et al.,* 2019). There is need to investigate alternative sources of natural coagulants such as *Moringa oleifera* seeds, *Aloe barbadensis miller* leaf and *Cactus Opuntia stricta* leaf in wastewater treatment.

Moringa oleifera seeds contain anti-microbial properties and cationic water-soluble proteins (polyelectrolytes) which possess active coagulative properties that can remove the turbidity and heavy metals from wastewater (Nishi *et al.,* 2011). The seeds are locally available, environmentally friendly and sustainable. They do not alter the pH of the treated water and produce small amounts of sludge that is bio-degradable hence preferred over chemical coagulants. *Moringa Oleifera* seeds contain proteins that produce a positive charge when introduced to water resulting in the electrostatic attraction with the negatively charged particles in water. It has been found that the water purification property of *M. oleifera* seeds is mainly because of the action of proteins present in them. These proteins bind to negatively charged particles present in raw water. These particles then grow in size and form flocks which are then allowed to settle down by gravity or are removed by filtration, thus playing an essential role in water purification (Nishi *et al.,* (2011).

Both aloe vera and Cactus coagulants have natural active components that aids in flocculation and sedimentation of both organic and inorganic compounds in wastewater. Sodium chloride plays a role in activation of this process (Vishali *et al.,* 2015).

Materials and methods

Study area

The study was carried out at the University of Eldoret, Biotechnology lab, located in Uasin Gishu County, about 9 km northeast of Eldoret town. Uasin Gishu County is situated in mid-western Kenya, between 34°55’33” and 36°38’58” E and between 0°2’44” S and 0°55’56” N. Wastewater samples were collected from Huruma wastewater treatment plant located in Eldoret town (Figure 1).

Sample collection

Wastewater samples were collected from the outlet of the maturation pond at Huruma wastewater stabilization ponds. Samples were collected on a weekly basis for four consecutive weeks from 15th Aaugust to 15th September 2022. Samples for the identification of phytoplankton species were taken from the surface and 0.25 m depth and were mixed in equal proportions to produce a composite sample. The composite samples were properly mixed, and 1000 ml aliquots were dispensed into bottles and fixed with Lugol’s iodine (0.01% v/v).

The samples were collected randomly between 900 hrs and 1000 hrs. During sampling, care was taken to ensure that no floating debris or large organic materials were collected as well as safety precautions.

Sampling bottles were capped before withdrawing them from the water. The samples were then labeled and transferred into a cooler box kept at 4.0 ± 1.0° C using ice packs to stop biochemical reactions. They were then transported to University of Eldoret biotechnology laboratory for further analysis.

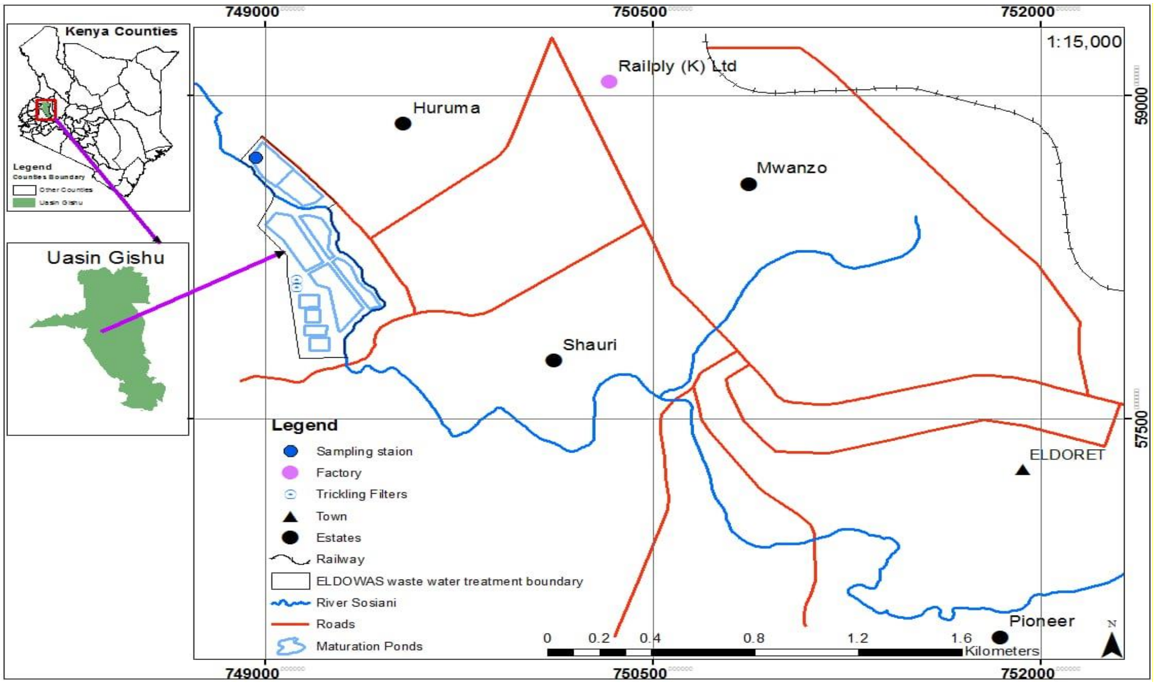

Figure 1: Location of study area showing the general location of Kenya counties, Uasin Gishu County and the sampling site

Preparation of Coagulants

***Moringa oleifera* seeds**

Dry *Moringa oleifera* seeds (MOS) with the pod, a natural coagulant, was purchased from the local markets in Eldoret town, with known sources from the nearby county, Baringo. The healthy seeds (about 1.0 cm long) were de-shelled and oven dried at 105˚C for 2 hrs. The kernels were crushed in a mortar and sieved through 300 µm to produce a powder with particles of 300 µm diameter. The powder was stored in an airtight container ready for use in wastewater analysis.

To extract the active cationic coagulating proteins, 5g of the seed powder was suspended in 100 mL of 1.0 mol/L NaCl solution and stirred using a magnetic stirrer for 30 minutes. The solution (MOS modifier) was filtered through a membrane microfiber filter paper of 0.45 µm pore size, yielding a stock of 2.8 ± 0.032 g/L of MOS extract. An additional stock of 100 mL of 1.0 mol/L NaCl in distilled water was prepared as a control.

***Aloe barbadensis miller* leaf**

Aloe vera leaf was collected from University of Eldoret arboretum. The leaf was washed and rinsed with distilled water. It was then sliced into strips of 1cm, oven dried at 80˚C overnight and then crushed and sieved through 300 µm. Five grams of the seed powder was suspended in 200 mL of 1.0 mol/L NaCl solution and stirred using a magnetic stirrer for 30 minutes.

It was then filtered through a membrane microfiber filter paper of 0.45 µm pore size. An additional stock of 100 mL of 1.0 mol/L NaCl in distilled water was prepared as a control.

***Cactus Opuntia stricta* leaf**

The Cactus Opuntia stricta leaf was collected from the University of Eldoret arboretum. The leaf was properly washed and rinsed with distilled water. It was then sliced into strips of 1cm, oven dried at 80˚C overnight then crushed and sieved through 300 µm. Five grams of the seed powder was suspended in 200 mL of 1.0 mol/L NaCl solution and stirred using a magnetic stirrer for 30 minutes. It was then filtered through a membrane microfiber filter paper of 0.45 µm pore size. An additional stock of 100 mL of 1.0 mol/L NaCl in distilled water was prepared as a control.

Local red soil preparation

Red soil was collected from the University of Eldoret field. It was oven dried at 105˚C for two hours. It was then crushed and sieved through 300 µm to produce a powder with particles of 300 µm diameter. The local red soil was used as an enhancer of stabilization and sedimentation of the cyanobacteria to the bottom of the test tubes.

Experimental set up

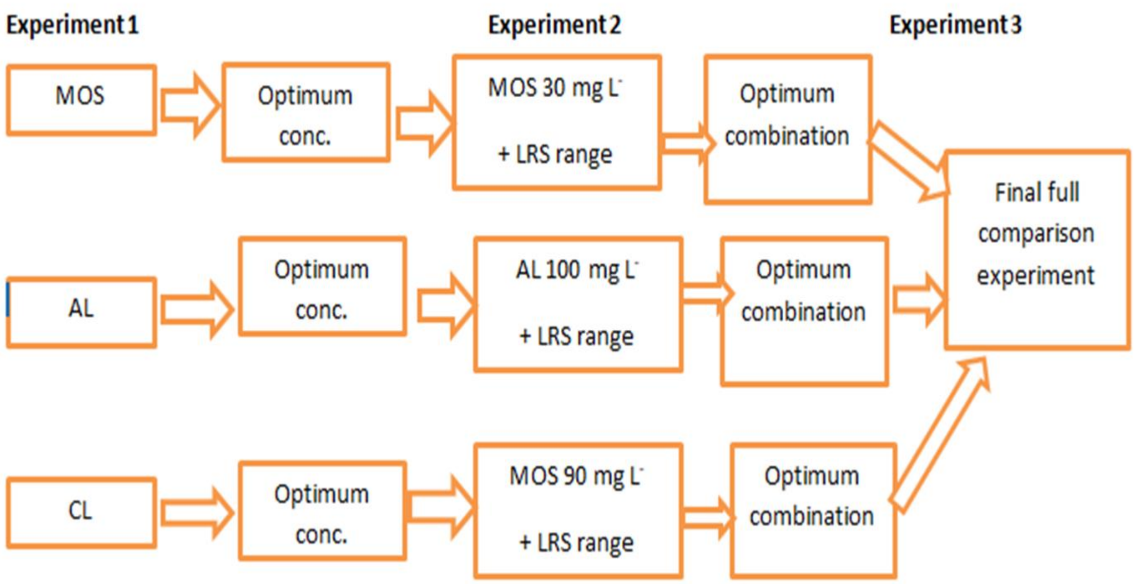

Figure 2: Experimental setup

Determination of cyanobacteria biomass using chlorophyll analysis

The initial volume of the waste water sample was recorded. The cells were separated from the water by filtration method. The filters were placed in a tissue grinder, 2-3 ml of boiling ethanol was added, and grinded until the filter fibers were separated. The ethanol and ground filter were poured into a centrifuge tube, the grinding tube was rinsed out with another 2 ml ethanol which was added to the centrifuge tube. The volume was topped up to a total of 10 ml in the centrifuge tube with ethanol. The cap was placed on the tube, labeled and stored in darkness at approximately 20ºC for 24-48 hours. After 24hrs, the sample was centrifuged for 15 minutes at 3000-5000 gravity to clarify samples. The clear supernatant was decanted into a clean vessel and the volume recorded.

The Spectrophotometer was blanked with 90 percent ethanol solution at 750 nm and 665 nm wavelength. The centrifuged sample was placed in the cuvette and the absorbance recorded at 750 nm and 665 nm (750a and 665a).

0.01 ml of 1 mol I-1 HCl was added to the sample in a cuvette and agitated gently for 1 minute. The absorbance was recorded at 750 nm and 665 nm (750b and 665b).

Calculation

Turbidity was corrected by subtracting absorbance 665a-750a = corrected 665a absorbance. 665b-750b = corrected 665b absorbance. The corrected 665a and 665b absorbance was used to calculate chlophyll *a* as follows;

$$\text{Chlorophyll a} = \frac{29.62(665a - 665b) \times v_e}{v_s \times l} \text{ mg } m^{-3}$$

where:

V_e = Volume of ethanol extract (ml)

V_s = Volume of water sample (litres)

l = Path length of cuvette (cm)

The initial concentration of the cell before the experiment was 238.2µg L-1 which was subjected to treatments with different coagulants in the range of (10,20,30,40,50) mg for *Moringa oleifera* seeds and a dose range of (80,90,100,110 ,120) mg for both Cactus leaf and Aloe vera leaf. Red soil was added at a dose range of (180,220, 260,300,340) mg with respective coagulant doses.

Coagulants efficacy analyses

Criteria to select effective coagulant and red soil dose was based on the percentage of cyanobacteria cells removed from the surface of the test tube, the degree to which the coagulants removed the cyanobacteria without destroying their cells hence removal while alive, percentage of sedimentation at the bottom of the tube and consideration of the coast implications, such that the dose would not be the highest range. To determine whether the cyanobacteria cells were alive or dead, methylene blue dye method was used.

The cyanobacteria removal efficiency was estimated using the formula;

$$Effeciency = \frac{initial\ concentration - final\ concentration}{initial\ concentration}\ x\ 100$$

Data analysis

The data collected was summarized in tables and subjected to statistical analysis using minitab statistical packages to determine the mean and standard error. Descriptive statistics such as means and percentages were calculated. It was further analysed using one-way ANOVA and means separated using Tukeys test at 5% level. Tables, line graphs and bar graphs were used to present the results.

Results and discussion

The mean temperature of Huruma wastewater treatment plant outlet was 21º C, pH 7.3 and conductivity (µs) 410 whereas the mean concentrations of nutrients were phosphates 5.39 mgL^{-1} and nitrates 0.89 mgL^{-1}. The treatment plant had cyanobacteria biomass of 238.2µg L^{-1}

Table 1: Mean concentration of the physicochemical parameters at the outlet of Huruma wastewater treatment plant

Physicochemical parameters	**Concentration**
pH	7.3
Conductivity (µs)	410
TDS (mgL^{-1})	190
Temperature (ºC)	21
Phosphates (mgL^{-1})	5.39
NO3 (mg L^{-1})	0.8966
Cyanobacteria biomass (µg L^{-1})	238.2

The concentration of nutrients in the outlet of the treatment plant was high for both phosphates and nitrates in the effluents that were released from the maturation pond to river Sosiani. The high levels of nutrients may be associated with the low efficiency of the treatment plant and the consequent nutrient-rich wastewater. Moreover, the abundance of cyanobacterial species observed in this study may have resulted primarily from the availability of high levels of nutrients (Lu *et al.,* 2017).

Estimation of the effective dose of Moringa, Aloe vera and Cactus leaf coagulates in removing cyanobacteria

For effective estimation, the initial concentration of the cyanobacteria cells before the experiment was analyzed using chlorophyll *a* formula which gave a biomass of 238.2µg L-1, the dose range of (10,20,30,40,50) mgL^{-1} was used for *Moringa Oleifera* Seeds, (80,90,100,110 ,120) mgL^{-1} for both Cactus leaf & Aloe Vera leaf. The dose range of local red soil was (180,220, 260,300,340) mgL^{-1}.

The following results were obtained for both top concentrations and the bottom concentrations of chlorophyll *a* in the test tubes as per the described procedures. In *Moringa oleifera* series a concentration of 30 and 40 mg/L reduced Chl-a in the top of the tubes by 19% and 5%, respectively, while in the bottom of the tubes, the Chl-a concentration increased by 7% and 3.7% respectively (Figure 3).

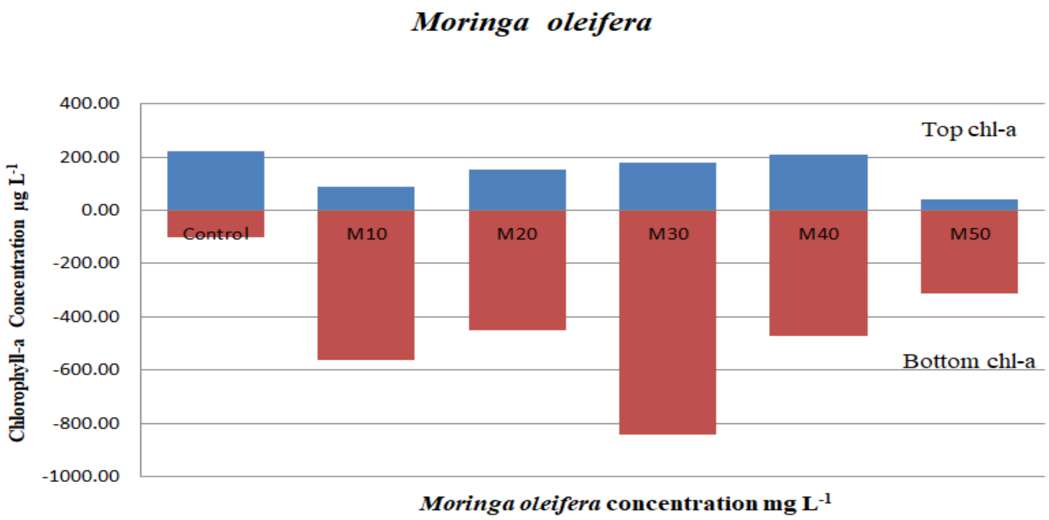

Figure 3: Cyanobacteria concentration on top and bottom of the tube after treatment with *Moringa oleifera*

In this study, Moringa seeds concentration of 30 mgL^{-1} was effective in coagulation and settling the cyanobacterial species in the waste water treatment plant effluent without damaging the cell. According to Habtemariam *et al.,* (2021) a similar dose of Moringa seeds concentration (30 mgL^{-1}) effectively coagulated and settled cyanobacteria from Legedadi reservoir in Ethiopia.

The interaction between the algal cells and the active protein molecule in the Moringa seeds is thought to be the cause of the Moringa seeds capacity to flocculate the cells (Camacho *et al.,* 2017). Due to the chemical components that make up their cell walls, cyanobacteria responds negatively, whereas the active cationic protein molecule in the Moringa seeds responds positively when it comes in contact with the cell wall and causes the cyanobacteria to flocculate (Hajar *et al.,* 2016).

In *Cactus* series, a concentration of 90 and 100mg/l reduced Chl-a in the top of the tubes by 61.6% and 85 % respectively while in the bottom of the tubes, Chl-a concentration increased by 1.41% and 0.06 % respectively (Figure 4).

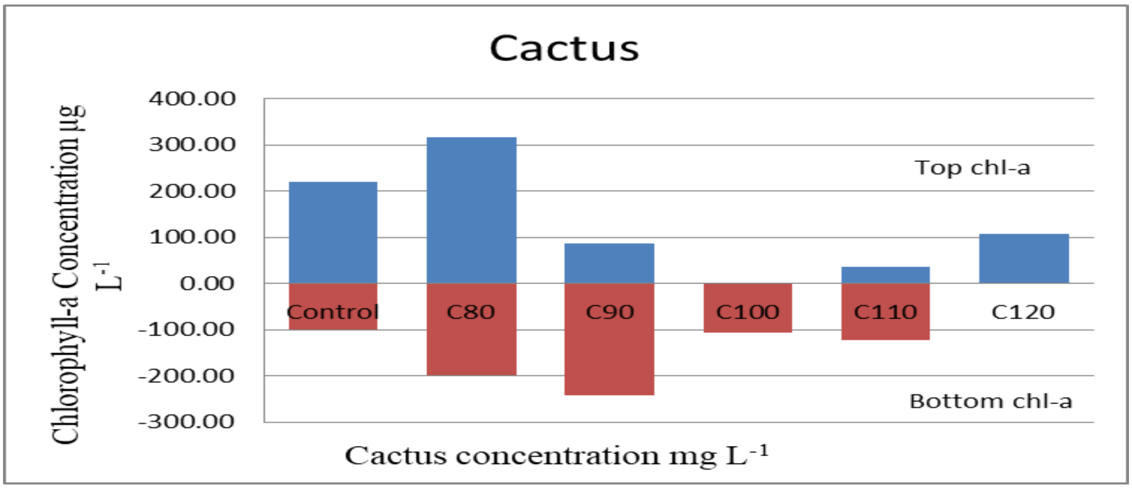

Figure 4: Cyanobacteria concentration on top and bottom of the tube after treatment with *Cactus opuntia leaves*

Cactus concentration 90 mgL^{-1} was found to be effective in coagulation and settling the cyanobacterial species in the wastewater treatment plant effluent without damaging the cell. However, Ayat *et al.,* (2021) used much higher dose 2800 mgL^{-1} in wastewater treatment plant effluent and obtained 98.3 % removal efficiency.

In the Aloe *vera* series, a concentration of 100 and 110 mgL^{-1} reduced Chl-a in the top of the tubes by 33% and 45%, respectively while in the bottom of the tubes, the Chl-a concentration increased 4.89% and 3.68 % respectively (Figure 5).

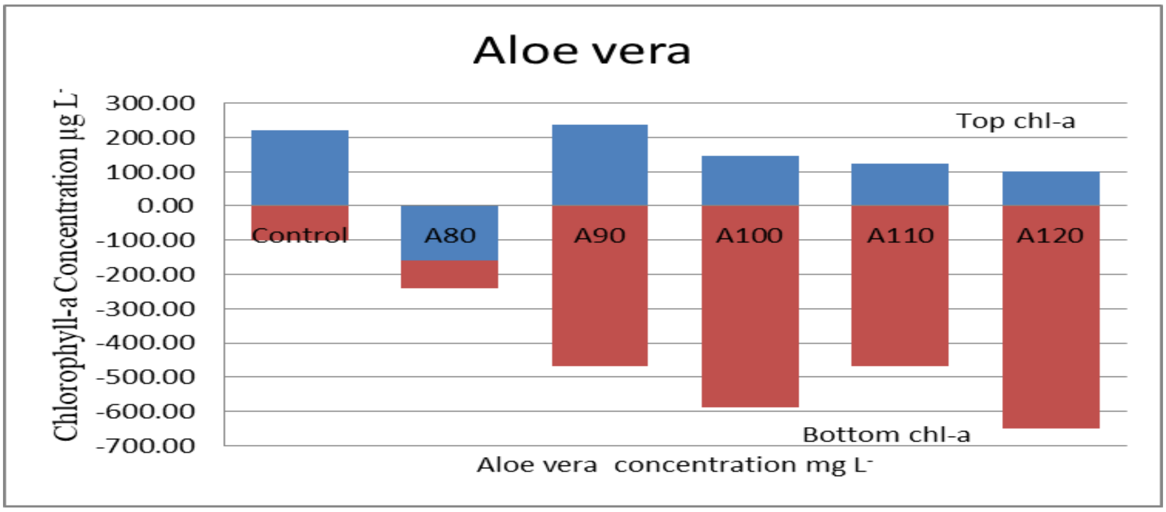

Figure 5: Cyanobacteria concentration on top and bottom of the tube after treatment with *Aloe vera leaves*

Aloe vera leaves concentration of 100 mgL^{-1} was found to be an effective dose in coagulation and settling the cyanobacterial species in the wastewater treatment plant effluent without damaging the cells. *Aloe vera* has potent antibacterial, antifungal, and antiviral properties (Ramasubramanian *et al.,* 2010)

Effect of the coagulant doses on pH

In *Moringa oliefera* series, the pH dropped gradually in all doses. In the Cactus series the pH remained more or less constant in dosage of 80 to 110 however in Cactus concentration 120 the pH increased gradually.

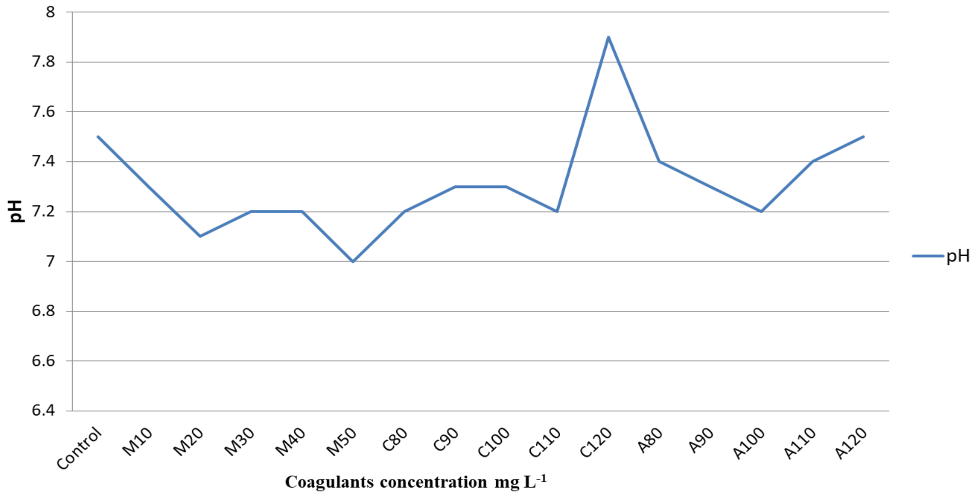

Figure 6: pH levels of wastewater after treatment with different concentrations of coagulants

The same observation was made by Beyene *et al.,* (2016), who reported that as the dosage of Cactus increased the pH also increased rapidly. In the case of Aloe vera, the pH remained constant in all dosages compared to the control as indicated in figure 6.

Impact of red soil in removal of cyanobacteria with effective dose of different coagulants

The effective dose of different coagulants was further tested with locally available red soil that was used as an enhancer of stabilization and sedimentation of the cyanobacteria to the bottom of the test tubes. The following results show the enhancement effects of red soil for all the coagulants.

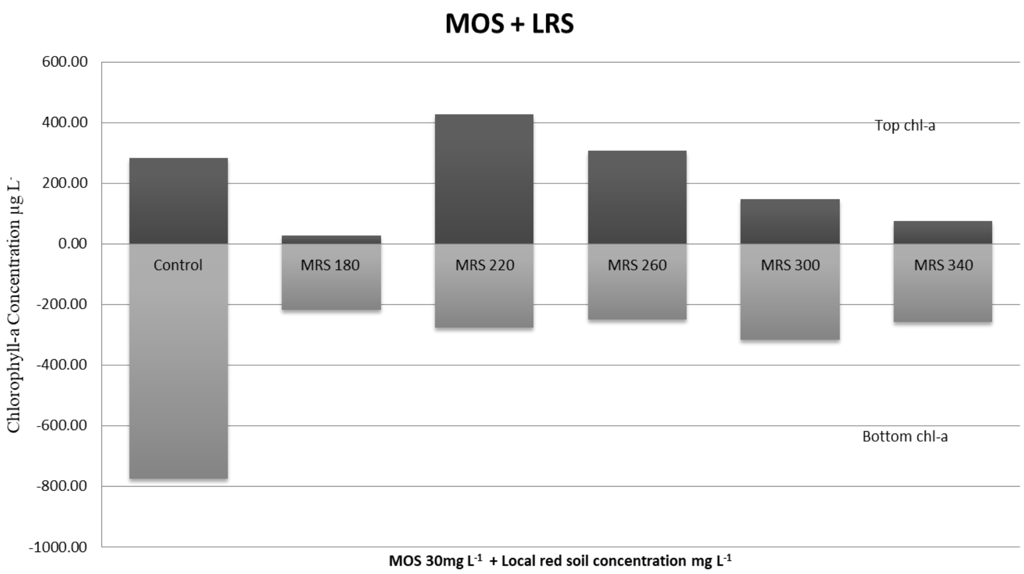

Figure 7: Effects of Local Red soil on settling of cyanobacteria in different concentrations of *Moringa oleifera* seeds coagulant

The result indicated that *Moringa Oleifera* seeds concentration of 30 mgL^{-1} with 180 mgL^{-1} local red soil (Figure 7) reduces the top cyanobacteria by 90%. Cactus seed effective dose of 90 mgL^{-1} with addition of local red soil concentration 220 mgL^{-1} reduced the top cyanobacterial concentration by 32% compared to cactus without red soil (Figure 8).

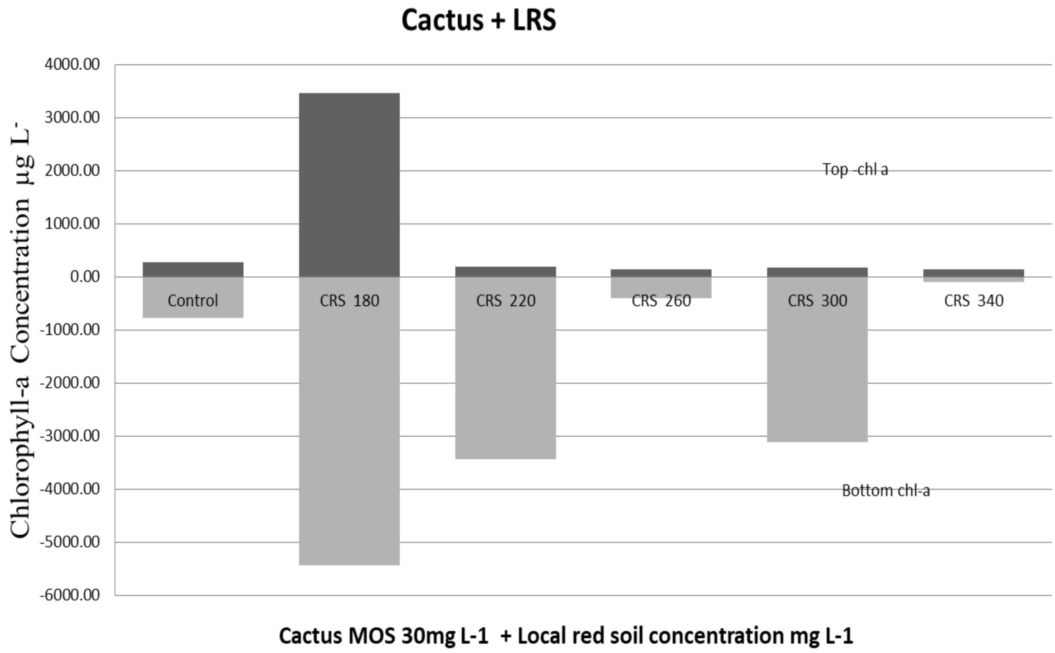

Figure 8: Effects of Local Red soil on settling of cyanobacteria in different concentrations of Cactus leaves coagulant

Cactus with red soil showed lower removal efficiency. The effective dose of Aloe vera leaf concentration of 100 mgL^{-1} with local red soil concentration of 180 mgL^{-1} was able to reduce the top cyanobacterial by 58% (Figure 9). In the case of *Moringa oliefera* and Aloe vera coagulants, addition of the local red soil improved the settling of buoyant cyanobacteria. Similar findings were observed by Peng *et al.,* (2019) who reported that the red soil-based flocculant can significantly remove cyanobacterial biomass and reduce concentrations of nutrients including total nitrogen, nitrate, ammonia, total phosphorus, and orthophosphate.

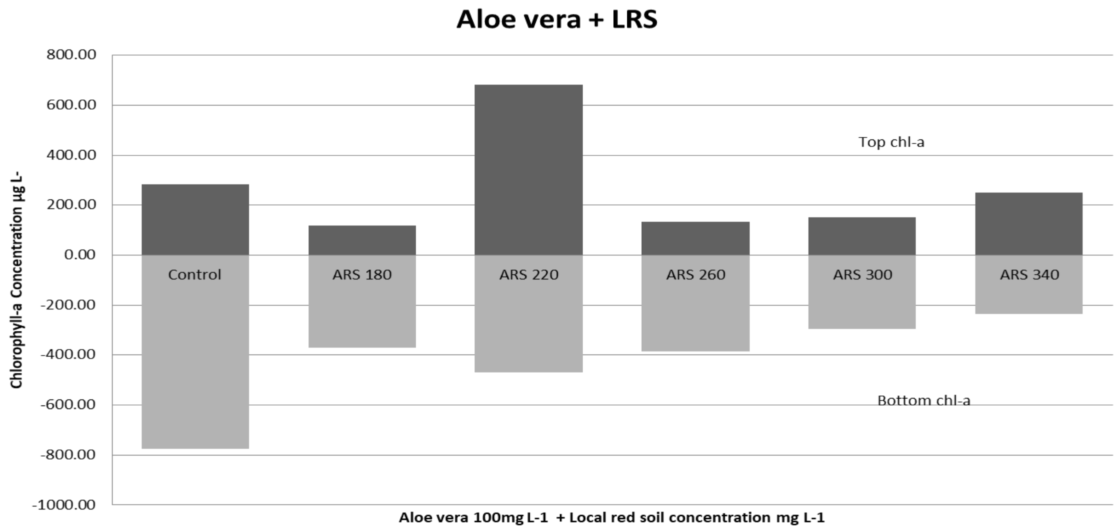

Figure 9: Effects of Local Red soil on settling of cyanobacteria in different concentrations of Aloe vera leaves coagulant

Effect of introduction of red soil on pH

The addition of red soil to Aloe vera and cactus didn't affect the pH of the solution however, addition of soil to *Moringa oleifera* showed significant reduction of the pH. This is in agreement with another study done by Habtemariam *et al.,* (2021) at Legedadi reservoir Ethiopia, who reported that *Moringa oleifera* reduced the pH of the solution.

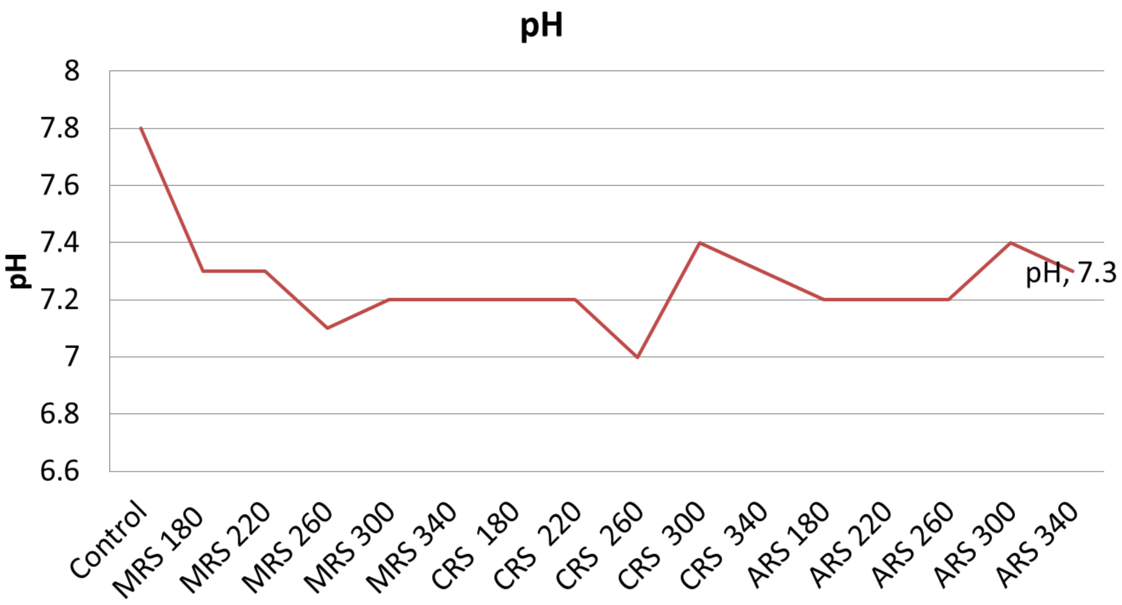

Figure 10: Effects of Red soil on pH of the treated wastewater

Evaluation of the efficient coagulant in removing the cyanobacterial from the wastewater suspension

A comparison of the most effective coagulant was achieved through comparison with the control, which was the initial concentration of the cyanobacteria in the waste water.The comparison of the three coagulants showed that *Moringa oleifera* had a better cyanobacteria removal efficiency than Aloe vera and cactus. *Moringa oliefera* seed removed buoyant cyanobacteria by 62%, cactus by 52.2 % and Aloe vera by 47.3%.

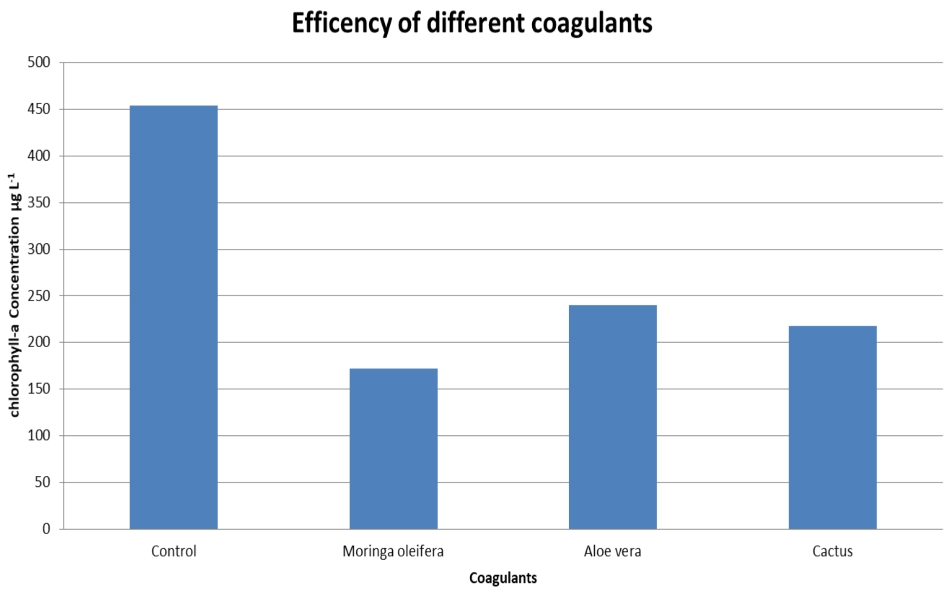

Figure 11: Efficiency of the coagulants in removal of cyanobacteria in the wastewater

Therefore, Moringa is a better nature-based coagulant. Moreover, *Moringa oleifera* can also reduce the concentration of extracellular cyanotoxins (microcystins) by 50% (Gouvea-Barros *et al.,* 2019). However, there are concerns regarding the use of biodegradable flocculants like Moringa due to their limited availability only in certain regions with tropical and subtropical climates (Bamishaiye *et al.,* 2011). Furthermore, *Moringa oleifera* is a multipurpose tree. Thus, its use in the removal of cyanobacteria leads to a conflict of interest. Besides, applying Moringa in the reservoir might increase the organic load of the reservoir, thereby affecting the level of dissolved oxygen (Vieira *et al.,* 2010).

The natural coagulants such as Aloe vera, cactus and *Moringa oleifera* are dominant plants in tropical/ subtropical climates that are used for many purposes (El-Azazy *et al.,* 2019). Wastewater treatment and treatment of water sources affected by cyanobacterial using these natural coagulants is proven to be a cost- effective, efficient, renewable and safe option for developing countries that have suitable climates to cultivate them (Habtemariam *et al.,* 2021). Moreover, the Cactaceae family has exceptional adaptability to severe and contrasting situations, including drought, high and low temperatures, nutrient and organic poor soils make it suitable coagulant to be used to adapted the impact of climate change in tropical countries.

Conclusion and recommendation

Conclusion

Different coagulants had different cyanobacteria removal efficiency. *Moringa oleifera* had cyanobacteria removal efficiency of 62%, *Aloe barbadensis miller* leaf had 47.3% while *Cactus Opuntia stricta* leaf had 52.2%. Compared to other coagulant *Moringa oleifera* showed higher cyanobacteria removal with low dose of local red soil. *Aloe barbadensis miller* leaf showed higher percent of cyanobacteria removal without the addition of local red soil

Recommendation

The three local coagulants can be used to treat wastewater contaminated by cyanobacteria. *Moringa oleifera* can remove cyanobacteria with low dose of local red soil while *Aloe barbadensis miller* leaf can remove cyanobacteria without the addition of local red soil.

References

Ayat A, Arris S, Abbaz A, Bencheikh-Lehocine M, Meniai AH (2021). Application of Response Surface Methodology for Modeling and Optimization of a Bio Coagulation Process (Sewage Wastewater Treatment Plant). Environ Manage. https://doi.org/10.1007/s00267-020-01407-0

Bamishaiye EI, Olayemi FF, Awagu EF, Bamshaiye OM. Proximate and phytochemical composition of Moringa oleifera leaves at three stages of maturation. Adv. J. F. Sci. and Tech. 2011; 3(4): 233–237.

Beyene, H. D., Hailegebrial, T. D., & Dirersa, W. B. (2016). Investigation of coagulation activity of cactus powder in water treatment. *Journal of Applied Chemistry*, *2016*, 1-9.

Camacho FP, Sousa VS, Bergamasco R, Teixeira MR. The use of Moringa oleifera as a natural coagulant in surface water treatment. Chemical Engineering Journal. 2017; 313:226–237.

El-Azazy, M., Dimassi, S. N., El-Shafie, A. S., & Issa, A. A. (2019). Bio-waste Aloe vera leaves as an efficient adsorbent for titan yellow from wastewater: Structuring of a novel adsorbent using Plackett-Burman factorial design. Applied Sciences, 9(22), 4856

El Bouaidi, W., Essalhi, S., Douma, M., Tazart, Z., Ounas, A., Enaime, G., & Loudiki, M. (2020). Evaluation of the potentiality of Vicia faba and Opuntia ficus indica as eco-friendly coagulants to mitigate Microcystis aeruginosa blooms. Desalination Water Treat, 195, 198-213.

Gouvea-Barros S, Bittencourt-Oliveira MD, Paterniani JE. Moringa-Seed-Based Coagulant Removes Microcystins Dissolved in Water. CLEAN–Soil, Air, Water. 2019; 47(6):1800465.

Habtemariam H, Kifle D, Leta S, Mucci M, Lu¨rlingM (2021) Removal of cyanobacteria from a water supply reservoir by sedimentation using flocculants and suspended solids as ballast: Case

of Legedadi Reservoir (Ethiopia). PLoS ONE 16(4): e0249720. https://doi.org/10.1371/journal. Pone.0249720

Lu H, Campbell E, Campbell DE, Wang C, Ren H (2017) EPA public access. Atmos Environ 23:248–258. https ://doi.org/10.1016/j. Hal.2017.06.001

Nery, J. F., & Nery, G. K. (2019). Potential of Opuntia stricta Haw (Mexican elephant ear) in removing cyanobacteria in surface water. African Journal of Biotechnology, 18(29), 875-881.

Nishi, L., Madrona, G. S., Guilherme, A. L. F., Vieira, A., Araújo, Á. A. D., Ugri, M. C. B. A., & Bergamasco, R. (2011). Cyanobacteria removal by coagulation/flocculation with seeds of the natural coagulant Moringa oleifera Lam.

Peng, L., Lei, L., Xiao, L., & Han, B. (2019). Cyanobacterial removal by a red soil-based flocculant and its effect on zooplankton: an experiment with deep enclosures in a tropical reservoir in China. *Environmental Science and Pollution Research*, *26*, 30663-30674

Ramasubramanian TS, Sivakumar VT, Thirumalai AV. (2010). Antimicrobial activity of *Aloe vera* (L.) Burm. f. against pathogenic microorganisms. J Bio Sci Res.;4: 251–258.

Rebah, F. B., & Siddeeg, S. M. (2017). Cactus an eco-friendly material for wastewater treatment: A review. Journal of Materials and Environmental Sciences, 8(5), 1770-1782.

Vieira AM, Vieira MF, Silva GF, Arau´ joA´ A, Fagundes-Klen MR, Veit MT, et al. Use of Moringa oleifera seed as a natural adsorbent for wastewater treatment. Water, air, and soil pollution. 2010; 206(1):273–281.

Vishali, S., & Karthikeyan, R. (2015). Cactus opuntia (ficus-indica): an eco-friendly alternative coagulant in the treatment of paint effluent. Desalination and Water Treatment, 56(6), 1489-1497.

Water, U. (2016). WWAP (United Nations World Water Assessment Programme) Paris, France: The United

Nations World Water Development Report. (pp. 1–148).

WHO. (2016). Sanitation Safety Planning: Manual for Safe Use and Disposal of Wastewater, Greywater and Excreta. Geneva, Switzerland, WHO. www.who.int/water_sanitation_health/ publications/ssp-manual/en/.

Woodward, G., Gessner, M. O., Giller, P. S., Gulis, V., Hladyz, S., Lecerf, A., & Chauvet, E. (2012). Continental-scale effects of nutrient pollution on stream ecosystem functioning. Science, 336(6087), 1438-1440.