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Original Research Article

Performance Evaluation of *Moringa oleifera* as a Coagulant for Abattoir Wastewater Treatment

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Abstract: The public's health and surface water are adversely affected by the improper disposal of abattoir wastewater. In this study, 40 litres of abattoir effluent was treated for ten (10) weeks using raw, powdered *Moringa oleifera* seeds as a coagulant. Processed *M. oleifera* seeds in loading quantities of 10, 12, 14, 16, 18 and 20 g were used in a completely randomised design (CRD) for the treatment. There was also a control (abattoir effluent not treated with *M. oleifera*). Before and after treatment, the physical and chemical characteristics of abattoir wastewater were examined. The results demonstrated that the treatments significantly lowered the turbidity value, which fell from 15.40 mg/L to 7.63 mg/L for a 16 g dosage in week 7. For the 20 g treatment in the first week, total alkalinity decreased from 216.67 mg/L to the lowest value of 63.67 mg/L. An amount of 14 g of *M. oleifera* lowered both the total hardness from 116.33 mg/L to 78.40 mg/L and conductivity from 1395.7 mg/L to 520 mg/L within the first week of the experiment. From weeks 2 to 6, the biological oxygen demand (BOD) was discovered to be nil. For the 14 g treatment, the Calcium value decreased from 31.47 mg/L in the first week to 6.23 mg/L in the fifth week. The results generally demonstrated that 16 g/500 mL of *M. oleifera* was capable of treating abattoir effluent, confirming its capacity to coagulate for the treatment of such waste. The study's extract doses did not affect the colour or smell of the treated wastewater. The results have generally highlighted that the natural coagulants could be successfully used for the removal of turbidity, faecal bacteria and all unwanted concentrations of heavy metals including zinc from abattoir wastewater.

Keywords: *Moringa oleifera*; wastewater; abattoir; dissolved oxygen; biological oxygen demand

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1. Introduction

Population expansion and the commercialisation of animal products for human use have led to an increase in abattoir waste production, resulting in pollution and environmental degradation, particularly in surface water bodies. In the northern part of Nigeria, especially in Minna, the state capital of Niger State in the North Central part of Nigeria, 3.92 tonnes of blood, 2.9 tonnes of intestinal content, 4.2 tonnes of bone and 2.2 tonnes of tissues as abattoir waste are generated daily (Ogbonaya *et al.*, 2011). Blood, fat, organic and inorganic solids, salts, and chemicals are frequently found in abattoir wastes, also referred to as slaughter-house wastes, which, if improperly managed and disposed of, pose serious health risks to those who live nearby (Oruonye, 2015; Sindibu *et al.*, 2018; Obidiegwu *et al.*, 2019). The wastewater from abattoirs is typically released directly into ecosystems without adequate treatment, posing serious threats to surface water quality, general environmental safety, and by extension, human health (Adamu & Dahiru, 2020; Aleksic *et al.*, 2020; Ogeleka *et al.*, 2021). The volume of domestic, agricultural, and industrial wastewater entering rivers globally is increasing at an alarming rate, worsening the problem of surface water pollution globally (Shukla *et al.*, 2021). This raw abattoir effluent discharge has a negative impact on water quality, mainly by reducing dissolved oxygen (DO) (Falodun & Rabiu, 2017) and raising the number of heavy metals in the water, which can cause aquatic life to perish (Simeon & Friday, 2017). Before being released into surface water bodies, abattoir wastewater needs to be treated to avoid contamination of water bodies which are used for both domestic and agricultural activities (Olaniran *et al.*, 2019). Liquid effluents discharged from slaughterhouses are extremely difficult to treat or purify due to a number of factors, including the waste's distinctive characteristics, its variable composition at the time of discharge, and a significant amount of mineral, biogenic, and organic matter that is known to provide nutrients for bacterial growth (Pandit *et al.*, 2021; Khan *et al.*, 2021; Meena *et al.*, 2021). According to the National Environmental Sanitation Policy developed by the Federal Ministry of Environment, improper market and abattoir planning; the emergence of illegal markets and abattoirs (including slaughter slabs); a lack of adequate facilities such as potable water; insufficient road networks, institutional regulations, enforcement, and monitoring; and, most importantly, corrupt and sharp practices by market and abattoir supervisors.

According to Adamu and Dahiru (2020), managing pollution at the point of slaughterhouse waste discharge into surface water bodies may be a huge task due to the high cost of technology required and owing to the exorbitant cost of the chemicals often used to treat the wastewater being generated. Alternative materials that are cost-effective, biodegradable, nontoxic, environmentally friendly, and locally accessible are now being investigated as a result of these challenges. Based on their chemical properties, coagulants are frequently used in conventional water treatment procedures. Aluminium and iron (III) salts are the two principal coagulants that are most frequently utilised, according to Bouchareb *et al.* (2021). However, recent studies have shown several disadvantages associated with the use of these salts, including a link between Alzheimer's disease and the use of aluminium salts in wastewater treatment that results in huge volumes of hazardous sludge (Ivanova *et al.*, 2021; Nisar & Koul, 2021). Vigneshwaran *et al.* (2020) have emphasised the drawbacks of the pervasive usage of chemical coagulants such as aluminium. According to the study, the usage of aluminium-based chemical coagulants results in a number of neurological issues, whereas bio-coagulants are dangerous to aquatic life due to their inherent qualities (Onukwuli *et al.*, 2021). The use of natural coagulants like *Moringa oleifera* has attracted a lot of attention lately (Rocha *et al.*, 2020). *M. oleifera* seed kernels are biological coagulants made up primarily of low molecular weight water-soluble proteins with a net positive charge in solution (Saini *et al.*, 2016; Wagh *et al.*, 2022). According to Tunggolou and Payus (2017), *M. oleifera* coagulant is both safe and highly effective in removing pollutants. I It exhibits coagulating qualities that have been employed for turbidity, alkalinity, total dissolved solids (TDSs), and hardness, among other elements of water treatment (Arnoldsson *et al.* 2008).

Muyibi and Evison (1995) first identified the softening property of *M. oleifera* when wastewater turbidity decreased by 99% and hardness was reduced by between 60 and 70%. This was further buttressed by Nand *et al.* (2012) as locally available seeds like corn, cowpeas, and *M. oleifera* were employed for the heavy metal adsorption of substances like lead, chromium, zinc, cadmium, and others, with *M. oleifera* being the most successful. Recent research by Nisar and Koul (2021) also found that *M. oleifera* seeds contain anti-microbial properties as well as cationic water-soluble proteins (polyelectrolytes) that have active coagulative properties. These proteins can effectively treat impure water by removing turbidity and heavy metals like Cu, Pb, Cr, and Zn from wastewater.

According to Rahmadyanti *et al.* (2020), using *M. oleifera* as coagulants in wastewater treatment resulted in pH parameter content that was close to neutral, or 6.65 ± 0.04%; it also decreased TSS to 99.63 ± 0.10%; COD to 98.06 ± 0.04%; and biological oxygen demand (BOD) to 97.67 ± 0.24%. The study also stated that the treated wastewater could be safely discharged into the water body as a result of the successes achieved in the treatment. In some earlier experiments, doses of 0 g/500 mL to 10 g/500 mL of *M. oleifera* seeds were employed to remediate wastewater. These investigations were unsuccessful in determining the optimal doses necessary to sufficiently treat the wastewater (Desta & Bote 2021; Villaseñor-Basulto *et al.*, 2018; Vunain *et al.*, 2019). Therefore, the goal of this study was to establish the best dosage of *M. oleifera* in treating abattoir wastewater while also addressing the physical, chemical, and bacteriological aspects of abattoir wastewater utilising *M. oleifera* seeds as a coagulant.

2. Materials and Methods

2.1 Preparation of Coagulant

Mature *M. oleifera* pods from dried, cracked fruit were selected. The seeds were extracted from the fruits by cracking them, and they were then given two days to air-dry. The outer shells of the seed kernels were removed using a knife, and the kernels were then crushed in a mortar and pestle before being sieved through a 600 m stainless steel sieve size to create a fine powder. The fine powder was maintained in a refrigerator in a sterilised plastic rubber container.

2.2 Experimental Design

The three replications of the experiment used a completely randomised design (CRD). Wastewater from the abattoir in the amount of 40 L was taken from the facility and poured into 18 beakers. Three of the beakers received 500 mL of purified water and 500 mL of abattoir wastewater, but without *M. oleifera*. This was kept around for comparison.

2.3 Sample Preparations

Varying amounts of 10 g, 12 g, 14 g, 16 g, 18 g, and 20 g of *M. oleifera* powder were weighed into beakers containing 500 mL of distilled water each allowed the creation of 6 different amounts of the stock solution for the loading dose. In order to create a clear solution, the contents in the beakers were agitated with an automated stirrer at 125 rpm for 30 min before being left undisturbed to settle for 1 h. By sifting the settled sludge, the solution was extracted and measured into a 500 mL sample of the effluent from the abattoir. To help the coagulant develop, the solution was gently shaken for 2 min. The generated solution was collected and put through a 10-week experimental investigation. After collection, the containers were carefully kept in a cool, dry room, and 2 h later, wastewater analysis began.

2.4 Physico-chemical and Biological Characterisation

The physicochemical parameters that were in this study were temperature, pH, total hardness, turbidity, conductivity, total alkalinity and dissolved oxygen, manganese, zinc.

2.4.1 Turbidity

The turbidity of abattoir wastewater was measured by using an Oakton TL2310 turbidity meter. The turbidity of the wastewater sample was measured before and after treatment. The sample was fed into a simple cell and put into the cell holder. It was placed in the Oakton TL2310 Turbidity meter for measurement and readings were taken after it was stabilised. It was expressed in nephelometric turbidity units (NTU), according to Verma *et al.*, (2020).

2.4.2 pH and dissolved oxygen

A pH meter was used to measure the pH of the effluent both before and after the addition of the coagulant. Winkler's method was used to measure the amount of dissolved oxygen. About 5 L of dilution water was produced by mixing 1 mL of phosphate buffer, magnesium sulphate, calcium chloride, and ferric chloride solution with each litre of distilled water and aerating the mixture for 10 h (Kim *et al.*, 2021).

2.4.3 Conductivity, TDS and temperature

The conductivity and TDS were all measured using a multifunction Oakton CON 550 meter. The wastewater sample was fed into a sample and put into the cell holder before it was placed in the Oakton CON 550 meter for measurements before and after treatment with natural coagulants.

2.4.4 Total hardness, total alkalinity, and zinc

Total hardness, total alkalinity, and zinc were evaluated using the Jeyasekhar (2022) procedure.

2.4.5 Biological oxygen demand (BOD)

BOD was measured using the Verma *et al.* (2020) approach. About 2.5 mL of abattoir wastewater sample before and after the treatment with *M. oleifera* seeds extracts were transferred into each BOD bottle. A volume of 300 mL of diluted water was added to the BOD bottle. Besides that, 300 mL of diluted water was transferred into the BOD bottle and allocated as the control. Each sample was then titrated against N/40 sodium thiosulphate solution with starch as an indicator after ceasing dissolved oxygen using 1 mL of potassium iodide and manganese sulphate. The same numbers of BOD bottles were also incubated in the BOD incubator for 5 days at 20°C, and DO values were measured after 5 days. The difference between the initial DO and the DO after 5 days gave the BOD values.

3. Results and Discussions

Table 1 displays the chemical properties of the *M. oleifera* seed used in this investigation. Table 1 underscores the coagulating nature of *M. oleifera* seeds as it contains cationic proteins, which help in water purification and treatment. *M. oleifera* seeds are also sources of minerals, micronutrients, and bioactive substances such as flavonoids, saponins, steroids, phytates, and trypsin inhibitors (Abdel-Latif *et al.*, 2022). The seed might be classified as an oil seed because its lipid content ranges from 13% to 46% (35.11% as shown in Table 1).

Both as a coagulant and an antibacterial, *M. oleifera*'s chemical characteristics operate on two levels. The positively charged, water-soluble proteins in *M. oleifera* seeds interact with the negatively charged particles in effluent from abattoirs to form bonds that cause flocs to fall to the bottom and be filtered out, thus acting as a coagulant (Baquerizo-Crespo *et al.*, 2021).

Table 1. Chemical properties of *M. oleifera* seed

Properties	Content (mg/L)	Properties	Content (%)
Iron	7.85	Moisture content	8.24
Manganese	4.42	Crude protein	36.62
Phosphorus	56.83	ASH	6.65
Magnesium	11.33	FATS	35.11
Calcium	4.85	Crude Fiber	4.28
Zinc	51.4	NFE	9.1

3.1 pH

Without *M. oleifera* treatment, the average pH of the abattoir effluent was determined to be 6.96 at the collecting site. In the first week, the pH decreased to 6.80, 6.78, 6.76, 6.71, 6.65, and 6.65 with the addition of 10 g, 12 g, 14 g, 16 g, 18 g, and 20 g of *M. oleifera* treatment, correspondingly. The cationic water-soluble protein found in the seeds of *M. oleifera* has the ability to act as a coagulant, which explains why the solution becomes more alkaline and therefore, increases the pH level of the wastewater. The solution becomes alkaline as a result of the alkaline amino acids in the *M. oleifera* protein absorbing protons in the water and releases hydroxyl groups (Marzougui *et al.*, 2021). The larger dose of *M. oleifera* seed powder employed in this study may have resulted in a lower pH of treated sewage water due to the presence of low molecular weight proteins (Basra *et al.*, 2014). However, as seen in Figure 1, for all doses, the pH value increased during the course of the treatment from week 1 through week 10. This was consistent with the findings of Desta and Bote (2021), who concluded that *M. Oleifera*'s parameters’ removal efficiency rises with an increase in pH value. According to Figure 1, the pH values reached their ideal levels in week 5 for all dosages with a minimum of 6.58 mg/L for the dosage of 16 g of *M. oleifera*. The pH range for industrial wastewater discharge after treatment, as advised by the World Health Organization (WHO), is 7 to 8 mg/L.

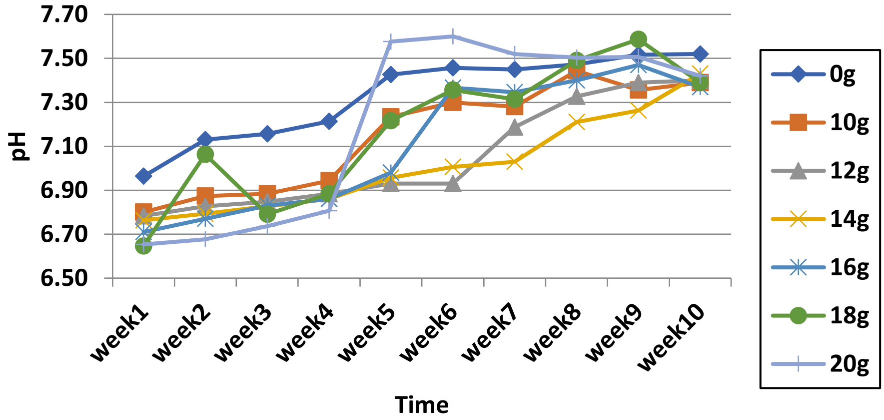

Figure 1. pH values with *M. oleifera* treatments

3.2 Turbidity

From week 2 to week 10, the turbidity of effluent from an abattoir treated with the coagulant *M. oleifera* seed powder significantly decreased (Figure 2). According to Vigneshwaran *et al.* (2020) and Desta & Bote (2021), the decrease in turbidity observed demonstrated that the dosage of cationic protein in *M. oleifera* has begun to react. According to Sotheeswaran *et al.* (2011), the mechanism of coagulation with *M. oleifera* seeds involve the adsorption and neutralisation of the colloidal positive charges that draw the negatively charged contaminants in water. Due to these interactions, the forces that stabilise the particles are weakened, allowing precipitate to form by attaching to minute particulates. As opposed to the 94.4% turbidity reduction reported by Vigneshwaran et al. (2020), the ideal dosage of *M. oleifera* was discovered to be 16 g/500 mL of *M. oleifera* dose in week 7, when it went from 19.73 to 7.53 NTU, or 63.5% turbidity removal from the wastewater (Figure 3). The destabilised particles have a chance of stabilising again, most likely as a result of an overdose. This was clear from Figure 3, which depicts how the turbidity level improved after week 7 to reach 16ng/500 mL. According to Megersa *et al.* (2016), this is a result of the polymer bridge becoming saturated.

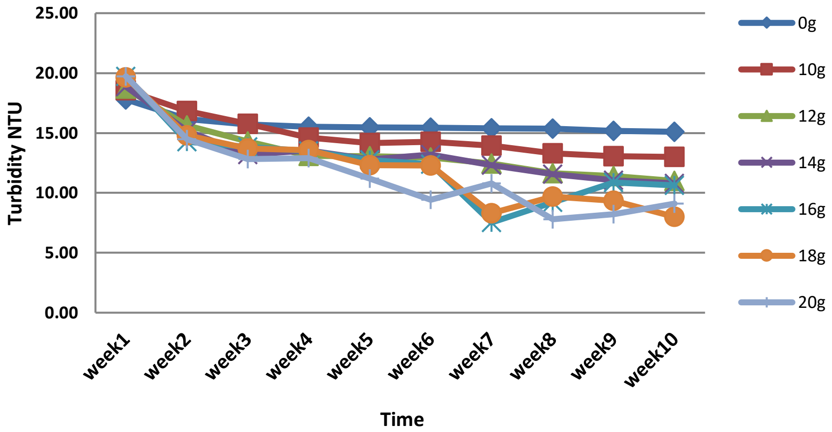

Figure 2. Turbidity values with *M. oleifera* treatments

Figure 3. Turbidity of abattoir effluent before and after treatment with *M. oleifera*

3.3 Conductivity

The conductivity of the wastewater was measured to be 1395.7 s/cm at the time of sampling (week 1) and rose with time to 1878 s/cm in the fifth week following treatment with *M. oleifera*. From the first week, the average conductivity value for all treatments dramatically increased. With 16 g of *M. oleifera* treatment, the value increased by roughly 61% from the point of collection to 1719.67 s/cm (Figure 4). The percentage increase level at 16 g of *M. oleifera*. As seen, the *M. oleifera* treatment is similar to all other therapies. In general, conductivity was seen to rise over time, albeit it gradually fell with treatments using *M. oleifera* (Figure 4). According to Tunggolou and Payus (2017), the increase in conductivity reading has been linked to the ions that develop in the water during the coagulation process. Due to the existence of unbound ions, a higher coagulant dosage in the solution than the recommended dosage will eventually result in an increase in conductivity (Yuliastri *et al*., 2016).

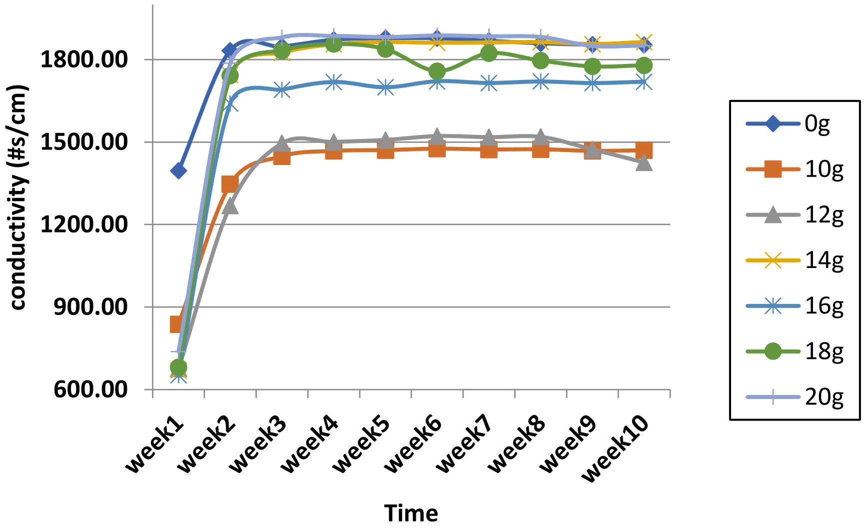

Figure 4. Conductivity values with *M. oleifera* treatments

3.4 Total Hardness

The abattoir effluent has greater total hardness values because it contains hardness from calcium, magnesium, and other hardness-causing chemicals. Hardness, particularly when magnesium sulphate is present, can create scaling in pipelines and develop a laxative effect on new customers (Egbueri, 2023). Cooking utensils and water heaters frequently develop incrustations as a result of calcium salts. The portable water must therefore be softened. This merely suggests that the necessary doses of *M. oleifera* will rise as the sample's hardness-causing species count rises. In Week 1, it was estimated that the samples' initial total hardness at the point of collection was 141.3 mg/L. From weeks 1 to 10, as the doses of *M. oleifera* rose, there was an overall decrease in total hardness, as depicted in Figure 5. This is because as the adsorption properties of *M. oleifera* seeds in wastewater help in aggregating the particles of dissolved ions, including calcium and magnesium ions, which by coagulation and flocculation, are removed from the wastewater thereby reducing total hardness. The outcome therefore demonstrated that the amount of hardness eliminated increased with the amount of *M. oleifera* administered. Because of this, 14 g of *M. oleifera* had the greatest ability to reduce wastewater hardness.

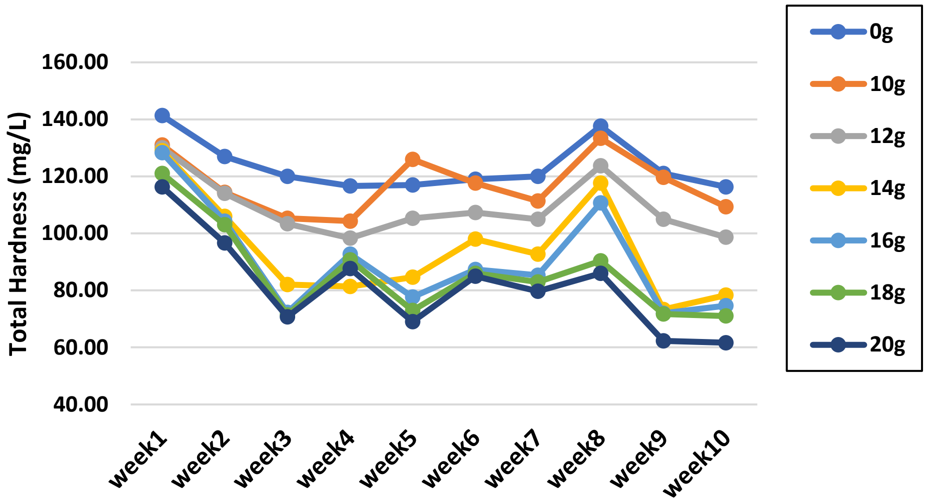

Figure 5. Total hardness values with *M. oleifera* treatments

3.5 Total Alkalinity

The collection point's alkalinity was 216.67 mg/L. In the first week, it dropped sharply, from 216.67 mg/L for the control to 86 mg/L for the 14 g *M. oleifera* treatment, a 60.3% reduction, as shown in Figure 5. Figure 6 shows that overall total alkalinity levels decreased throughout all weeks as the amount of *M. oleifera* rose. The reduction of total alkalinity of abattoir wastewater by *M. oleifera* is due to the adsorption properties of *M. oleifera* seeds (Desta & Bote, 2021) Through adsorption, *M. oleifera* seeds can reduce the concentration of dissolved organic compounds, ions, and other substances present in the wastewater, thereby decreasing total alkalinity. Because of this, 12 g of *M. oleifera* had the greatest ability to lower wastewater's overall alkalinity.

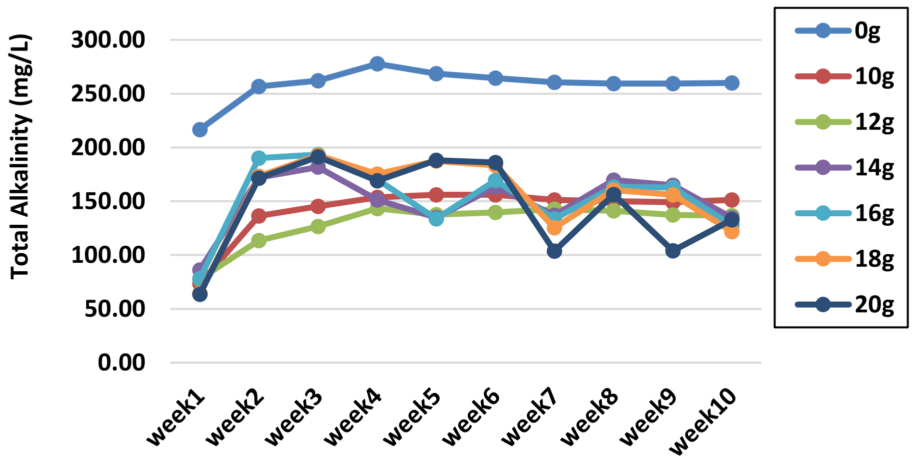

Figure 6. Total alkalinity values with *M. oleifera* treatments

3.6 Zinc

Figure 7 illustrates the concentration of zinc, one of the heavy metals in wastewater, which was reported to be 1.42 mg/L at the collection location. From week 1 to week 5, zinc concentrations generally decreased (see Figure 7). The reduction in zinc level in abattoir wastewater by *M. oleifera* can also be attributed to the adsorption properties of *M. oleifera* seeds. With 14 g of *M. oleifera* present, the lowest value of 0.62 mg/L was noted in week 5. This demonstrates how effective *M. oleifera* is at removing heavy metals. However, it was discovered that the zinc concentration at the place of collection was below the 5 mg/L maximum allowed by the WHO. The significant volume of blood discovered in wastewater has been linked to the high zinc level.

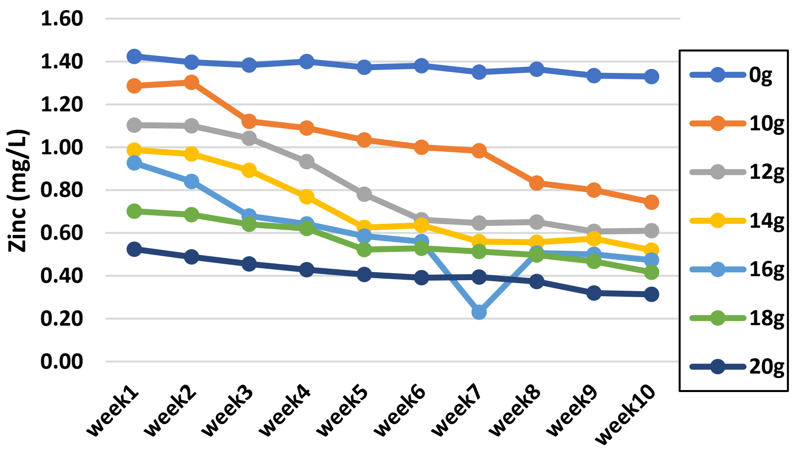

Figure 7. Zinc values with *M. oleifera* treatments

3.7 Dissolved Oxygen (DO)

The DO at the moment of collection ranged from 1.67 mg/L to 2.0 mg/L, according to Figure 8. With treatments of 10 g, 12 g, 16 g, 18 g, and 20 g of *M. oleifera*, it increased to 2.87 mg/L at the end of Week 5, except for the 16 g treatment that gave 2.93 mg/L of DO. There was also a considerable improvement in the DO right from Week 1. The improvement in DO in the samples has been completely achieved by the *M. oleifera* doses in the abattoir effluent by week 5, resulting in complete treatment across the samples, from 1.67 mg/L to 2.87 mg/L representing a total increase of 71.86% of the treatment, which is consistent with Yuliastri *et al*., (2016). This improvement in DO level of the wastewater has been attributed to the removal of organic compounds from the wastewater by the *M. oleifera* which are responsible for the consumption of DO during microbial decomposition (Ogunshina *et al*., 2023). The presence of natural and organic chemicals in *M. oleifera* seeds have been attributed to the rise in DO levels (Verma *et al*., 2020).

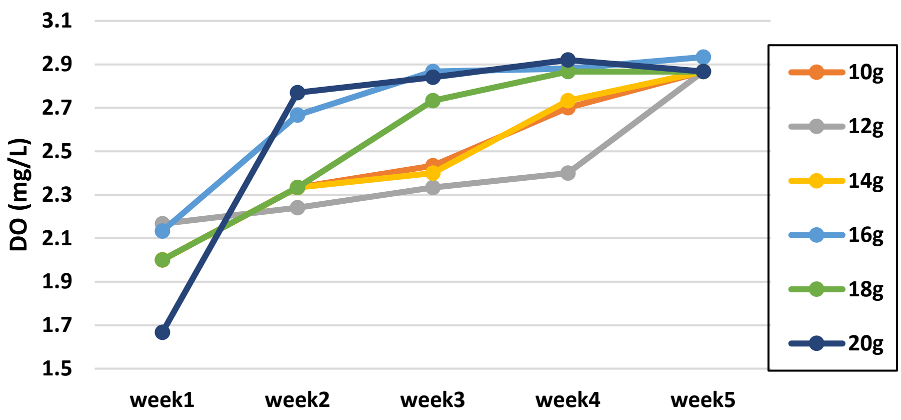

Figure 8. Dissolved oxygen values with *M. oleifera* treatments

3.8 Biological Oxygen Demand (BOD)

At the time of sampling, a BOD value of 4.33 mg/L was initially recorded without any coagulant added. When the *M. oleifera* was added at different doses in Week 2, the BOD value varied from 3.67 mg/L to 5.55 mg/L. BOD removal was made possible by the interaction between the cationic protein molecules with negatively charged particles present in abattoir wastewater. These proteins act as coagulants causing the particles to clump together and form bigger flocs which can then be easily removed from water thereby reducing the BOD. By comparing the sample's dissolved oxygen levels before and after 5 days of incubation (BOD_5) in the dark, the BOD level was calculated. The required amount of oxygen for the breakdown of organic compounds in the wastewater was determined by the difference between the two DO levels. The high blood volume in the wastewater was attributed to the higher BOD contents observed at the point of collection. This is consistent with Ma *et al*. (2020) and Iloms *et al*. (2020) findings. The lower BOD values recorded at the end of Week 2 and beyond indicate that all of the organic materials present in the wastewater samples have been completely degraded, as indicated in Table 1. It was, however, observed that at Week 8, the values of BOD started rising, though, insignificantly which could be attributed to the exhaustion of interaction between positively charged protein molecules in *M. oleifera* seeds and negatively charged wastewater particles. This, therefore, underscores the need for proper monitoring of BOD removal by *M. oleifera* as the BOD levels in wastewater samples tend to rise with time. This was the case between Weeks 7 and 10 (Table 1).

Table 1. BOD Values For the wastewater samples

M. oleifera Content (g)	**0 g**	**10 g**	**12 g**	**14 g**	**16 g**	**18 g**	**20 g**
week1	4.33	4.87	4.33	5.33	3.67	4.00	3.67
week2	0.00	0.00	0.00	0.00	1.00	0.00	1.00
week3	0.00	0.00	0.00	0.00	0.00	0.00	0.00
week4	0.00	0.00	0.00	0.00	0.00	0.00	0.00
week5	0.13	0.00	0.00	0.00	0.00	0.00	0.00
week6	0.00	0.00	0.00	0.00	0.00	0.00	0.00
week7	0.13	0.10	0.18	0.40	0.27	0.33	0.60
week8	0.19	0.20	0.35	0.27	0.30	0.33	0.63
week9	0.13	0.13	0.20	0.20	0.18	0.22	0.20
week10	0.17	0.20	0.32	0.30	0.30	0.38	0.40

4. Conclusions

The effectiveness of *M. oleifera* as a natural coagulant that is favourable to the environment has been evaluated. *M. oleifera* has been shown to be useful in purifying wastewater that has unwanted concentrations of heavy metals including zinc and manganese. The following conclusions can be taken from the experimental analysis and the results:

M. oleifera is eco-friendly and economically advantageous and locally available. The physicochemical properties of wastewater, including pH, turbidity, total alkalinity, total hardness, dissolved oxygen, BOD, calcium, and conductivity, can be improved by using *M. oleifera*, a powerful natural coagulant. As 16 g/500 mL of *M. oleifera* was able to treat abattoir wastewater and thereby confirm the suitability of using *M. oleifera* for abattoir wastewater treatment, *M. oleifera* seeds present a more effective and affordable method of treating abattoir wastewater that should be adopted in treating them before being discharged into the surface water body. The water-soluble, positively charged proteins found in *M. oleifera* seeds are thought to be responsible for the plant's reported coagulant activity.

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