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A comparative study of biocoagulant powers in preparing drinking water: case of *Moringa oleifera* and *Adansonia digitata*

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ABSTRACT

Rivers in the Democratic Republic of Congo are a major source of pollution, with conven-tional methods of producing drinking water employing expensive and unclean chemicals. The addition of aluminum salts for coagulation often leads to high concentrations of dis-solved aluminum, which is suspected to cause Alzheimer's disease. Integrating biological processes for treating drinking water could be a sustainable alternative due to their availa-bility and non-toxicity. This study evaluated the effectiveness of biocoagulants (Moringa oleifera Lam. and Adansonia digitata seeds) in preparing drinking water from surface water. Adansonia digitata seeds were unable to clarify raw river water, whereas Moringa oleifera seeds were. Oil extraction improved biocoagulant power, with hot extracts performing better than cold extracts. The optimal doses for hot extracts were 150, 150, 250, and 600 mg/l for turbidities of 39.6, 108, 380, and 960 NTU, respectively. Cold extracts were retained in the same raw water samples. Settling times of 60 to 90 minutes were found to be sufficient to eliminate most suspended matter and colloids. The chlorine level required for effective dis-infection was 12 to 13 mg/l. The extracts significantly increased the level of organic matter (OM) in the treated water. Moringa oleifera exhibited superior biocoagulant, bio-adsorbent, and bio-disinfectant properties over Adansonia digitata. Hot extracts from Moringa oleifera were more effective than cold extracts.

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1. Introduction

Fresh water is distributed unevenly, and a given use can reduce its quantity available for another, which causes serious water issues (Nsimanda, 2017; Raven et al., 2008). Of the 3% of the fresh water available on the planet, over 2.5% is stored in glaciers and polar ice caps not directly accessible to humans. Thus, the humanity relies on less than 0.5% stored mainly in groundwater, lakes, and rivers for all vital activities requiring fresh water.

The small amount of fresh water available to humans has drastically declined in recent years due to pollution. Polluted waters have serious implications for global health (Edogbanya, 2015). Kabore et al. (2013) believe that access to drinking water remains a major concern in Sub-Saharan Africa, where populations face challenges such as poor management of water points, insufficient hygiene and sanitation, and lack of appropriate household water disinfection methods.

The Democratic Republic of Congo (DRC) is not exempt from this situation, although it hosts the 2nd largest river in the world in terms of flow rate (41,800 m^3/s per mouth), in addition to its numerous rivers, springs, groundwater reserves, and lakes. Recognized worldwide as hydrological paradoxes, they are characterized by an apparently advanced degree of organic, mineral, and bacteriological pollution.

To guarantee human health, water is treated whenever it does not meet the potability criteria defined in standards, mainly these of the World Health Organization (WHO). Drinking water treatment involves multiple procedures employing various chemicals, including synthetic organic and inorganic substances (such as aluminum sulfate, chlorine, acrylamide, or activated charcoal). Typically, these chemicals are expensive and often unavailable, particularly in rural areas. They also pose several health risks, especially associated with aluminum sulfate (Edogbanya, 2016); some synthetic organic polymers, like acrylamide, may exhibit strong neurotoxic and carcinogenic effects (Choubey et al., 2012); chlorine, a strong oxidant, reacts with organic matter during disinfection, forming haloforms, which are likewise carcinogenic (Morris et al., 1992; Cantor et al., 1998; Villanueva et al., 2006; Richardson et al., 2007; Tangou, 2018). According to Pritchard et al. (2009), these chemicals are non-biodegradable.

Given the environmental unsustainability of the conventional water treatment methods utilized in developing countries, there is a need to consider alternative technologies based on natural materials (Pritchard et al., 2011). One of the avenues holding great promise is the use of biocoagulant plant seeds (e.g., Moringa oleifera) in water treatment (Dalen et al., 2009; Subramanium et al., 2011; Edogbanya et al., 2016). Plants with biocoagulant, bio-disinfectant, and bio-adsorbent properties, including Moringa oleifera, Phaseolus vulgaris, cactus species, Ipomea dasysperma, Adansonia digitata, etc. All these plants are found in the DRC. Moringa oleifera is quite widespread, quickly grows and thrives at low altitudes throughout the tropical zone, including arid regions, as well as can grow at medium altitudes on moderate humidity soils. Historically, Sudanese village women have been using such plant species for domestic wastewater treatment.

Adansonia digitata (baobab) belongs to the Bombacaceae family and genus. The baobab, also known as the "tree of life" or "pharmacist tree" possesses numerous medicinal properties, including biocoagulant activity.

In view of the above, the study team conducted this comparative research of the biocoagulant powers specifically of Moringa oleifera Lam. and Adansonia digitata in the preparation of drinking water from surface water to assess and compare their efficiency.

This investigation is particularly relevant given the growing concerns regarding water quality in highly polluted regions. Recent research, such as that of Varsani et al. (2023), indicates that natural biocoagulants can not only reduce water turbidity but also effectively remove pathogenic contaminants, thereby offering an environmentally friendly and cost-effective alternative to conventional chemical methods (Kouniba et al., 2024a). Furthermore, Fguiri et al. (2024) have demonstrated that the use of Moringa oleifera in drinking water treatment can reduce purification costs while simultaneously minimizing the negative ecological impacts associated with chemical coagulants. By integrating these natural processes, this study has aimed to propose sustainable solutions to improve water quality while supporting public health initiatives in vulnerable communities (Sagnon et al., 2023).

2. Materials and methods

Within the framework of this study, a full factorial experimental design with replicates was employed, in which the effects of each plant (Moringa oleifera and Adansonia digitata) extract were evaluated in triplicate using four (4) raw water samples with varying turbidities (low, medium, high, and very high). This approach allowed for a limited number of tests while capturing potential interactions between the parameters. Following dose optimization, the quality analyses were performed in triplicate at each treatment stage (raw water, settled water, and filtered water).

2.1. Pilot treatment system

The pilot system comprised three primary components:

- 30-liter flocculator equipped with an agitator to ensure mixing and promote floc formation. The agitation speed was approx. 180 revolutions per minute (rpm);

- 25-liter decanter fitted with a bottom drain to remove deposited flocs. The retention time ranged from 60 to 90 minutes;

- 100-liter filter incorporating a strainer and a single filter medium composed of quartz particles with a diameter ranging between 0.7 and 1.2 mm. The filter medium was 60 cm thick.

Fig. 1 illustrates the overall pilot treatment system setup used to evaluate the efficiency of Moringa oleifera seeds as a biocoagulant, as well as indicates the sampling points for decanted and filtered water. The conical decanter design facilitated efficient sludge decantation and removal.

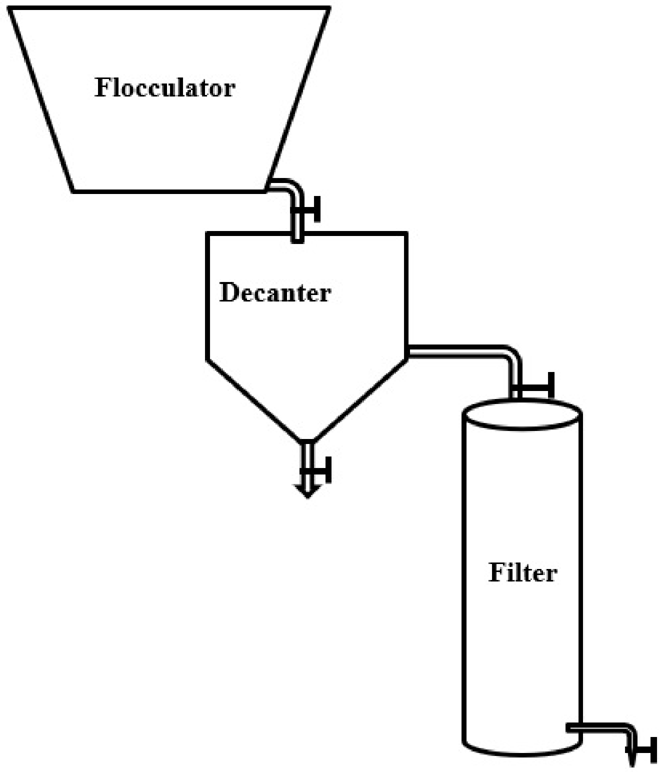

Figure 1. Processing pilot system

2.2. Biocoagulant solutions
2.2.1. Seed collection and grinding

Moringa oleifera Lam. seeds used in this study were collected in Kinshasa, specifically in the commune of N'selé at the SETA military camp opposite the N'djili International Airport. Ripe pods were harvested directly from the trees. Once dried, the seed coats and light wings were easily removed, revealing the white seeds. The seeds were thoroughly sun-dried for one (1) week, then sorted and finely pulverized using Butterfly B-592 crusher in the organic chemistry laboratory at the Faculty of Sciences, University of Kinshasa. The resulting powder was sieved through a fine mesh and de-oiled prior to use in water treatment.

Fully ripe and dry Adansonia digitata seeds, which had naturally dehisced on the ground, were collected in the commune of Lemba at the University of Kinshasa.

The seeds were extracted mechanically, thoroughly washed with clean water, sun-dried, and pulverized into a fine powder using the mortar and pestle, followed by grinding with the aforementioned crusher. The powder was then sifted and stored in hermetically sealed containers for subsequent use.

The quantity of crushed seeds required to treat river water depends on the concentration of suspended solids and seed quality.

2.2.2. Powder defatting

The study's hypothesis was that the presence of oil in aqueous solution could induce flotation and phase separation due to the inherent properties and density of oil relative to water. Thus, if the target coagulants contained a significant amount of oil, that may have impeded the flocculating activity of the extracts, resulting in incomplete floc decantation. For this reason, the seeds were de-oiled prior to use.

Furthermore, to preserve undenatured crude proteins in the meal, the extraction was conducted applying a cold solvent or at the lowest possible temperature. Additionally, a hot extraction was performed to compare efficiency (Rakotoniriana et al., 2015).

The materials and equipment employed included a Rotavopor Büchi RE 120, water bath, vacuum pump, two (2) round-bottom flasks, ice, electronic balance, soxhlet extractor, hot plate, filter glass, Bucney 25 G3, and rubber tubing. The solvent was 96% pure n-hexane, with the boiling point of 68.2°C and density of 0.6548 g/cm^3.

2.2.3. Process

Solvent distillation. The solvent was distilled to obtain pure n-hexane, preventing material contamination. That involved placing a quantity of the crude solvent in the flask, and positioning it in the Rotavapor to collect the distillate (pure solvent). The vacuum pump was connected and activated to create vacuum, reducing system pressure. The water bath was filled, heated to 68°C, and the cold-water valve was opened. Finally, the Rotavapor was activated, and the rotation speed was adjusted to initiate distillation.

Oil extraction (hot and cold). For hot extraction, 75g of the initial powder (P1) was weighed, formed into cartridges, and placed in the soxhlet extractor. A sufficient quantity of distilled n-hexane was added to the soxhlet flask, passing through the cartridges. The soxhlet condenser was then attached, and the cooling water was turned on. The hot plate and stirrer were activated, and the system was allowed to run for 24 hours until the yellow color – indicating the presence of oil – completely disappeared. The delipidated cartridges were removed, and the powder was soared in the open air to evaporate any residual solvent. Finally, the resulting powder (P2) was weighed.

Cold extraction. For cold extraction (maceration), 150g of the initial powder (P1) was placed in a clean 2.5-liter bottle, to which one (1) liter of n-hexane was added. The bottle was then sealed, shaken thoroughly, and allowed to macerate for at least 24 hours. Subsequently, the mixture was filtered using the vacuum filter apparatus, and the powder was collected in the bucney, spread in the open air to evaporate any remaining solvent, and weighed again to get the final powder (P2).

The extraction yield was calculated using the following formula:

$$\mathbf{Re} = \frac{(P2-P1)}{P1} x\ \mathbf{100} \quad (1)$$

, where P1 represents the weight of the powder before extraction, and P2 represents the weight of the powder after extraction.

The extracted oil was recovered using the Rotavapor, following the procedure described earlier. The flask was weighed prior (P1 = 194.94 g), and after removing the solvent – the second weighing (P2) was performed to determine the extraction yield and quantify the extracted oil.

The de-oiled powders were stored in clean, dry, and tightly closed bottles for later use.

Preparation of biocoagulative solutions and conservation. 20g of the delipidated powder was weighed and added to a 1-liter beaker containing distilled water. The beaker was placed on a stirrer set to 1,000 rpm, and the water volume was adjusted to 1,000 ml. The solution was stirred for 15 minutes, then filtered through a pre-weighed clean cloth (P1). The cloth containing the residue was placed in the oven at 95°C to expel the water, and then weighed again (P2) to determine the quantity of the total extract filtered in the water (solution) by making the difference between the two weights.

The prepared solution was stored at 4°C and used within one (1) week. The prepared solutions and concentrations included:

- Hot moringa solution – 10 g/l;
- Cold moringa solution – 11 g/l;
- Hot baobab solution – 13 g/l;
- Cold baobab solution – 12 g/l.

2.3. Sampling procedures

Approximately 50 liters of raw water from the N'djili River were sampled at the site within REGIDESO using thoroughly cleaned containers rinsed at least twice with raw water. The samples were transported to the central lab for parameter measurements prior to use in the pilot treatment system. The samples from the system (decanted and filtered water) were collected in clean bottles rinsed with the same water for physico-chemical analyses, and in sterilized bottles for bacteriological analyses. All analyses followed the standard operating procedures.

The collected samples were preserved according to the specific requirements of each target parameter. Generally, they were stored at 4°C, if analyses were to be performed within 24 hours of collection.

2.4. Experimental testing

Flocculation (jar) test. The test involved determining the optimal biocoagulant dosage required to clarify 1 m3 of water, using the AQUALYTIC flocculator with 6 stations, one of which served as a control. Several clarification trails were performed.

Quality analyses were performed only on samples where floc formation occurred. The fast speed was 150 rpm for 3 minutes, followed by the slow speed of 40 rpm for 17 minutes. Floc formation was observed, and the samples were left to settle for 60-90 minutes. Afterwards, siphoning was carried out, and turbidity was measured. The beaker with the lowest turbidity determined the treatment dose employed in the pilot system.

To determine the optimal settling time, the turbidity of each beaker was measured every 30 minutes until a turbidity between 0 and 5 NTU was achieved.

Chlorine demand was assessed to determine the chlorine dosage required to disinfect 1m3 of water treated with the biocoagulant. That was performed using the filtered water from the pilot treatment system. The granular chlorine (Titrating 70%) solution was prepared at 1 g/l in distilled water as follows: adding increasing amounts of the solution; shaking well; immediately placing in the dark; and after 30 minutes, determining the residual chlorine. The difference between the injected dose and residual chlorine gave the chlorine demand value. The dose corresponding to the sample the residual chlorine of which was within the recommended limit (0.8-1.2 mg/l) according to the WHO (2017) was retained.

The physico-chemical parameters – including turbidity, pH, conductivity, and temperature – were measured using HANNA HI 2211 multi-parameter instrument. The free chlorine was measured colorimetrically using a disk comparator with DPD (diethyl paraphenylenediamine) tablets. Nitrate, iron, and manganese concentrations were determined by spectrophotometric absorption using HACH DR 3900 spectrophotometer. Organic matter was quantified by manganese oxidizability; CAS (complete alkaline strength) and HT (hydrotimetric titer) were determined by acid-base titration to measure alkalinity and hardness, respectively. All reagents applied were of analytical grade and specific to each parameter.

Bacteriological parameters, including total coliforms (TC) and fecal coliforms (FC), were quantified using the membrane filtration method with 0.45 µm filter membranes. A dilution of 10-3 was prepared to facilitate colony counting, and the results were multiplied by the inverse of the dilution factor. 100 ml of the diluted sample were filtered through the membrane using a vacuum filtration ramp. The membrane was placed on solid culture medium: m-endo agar (pH 7.4), then incubated at 37°C for the TC, and m-FC agar (pH 7.5) at 44°C for 18-24 hours. All solid culture media utilized were extremely homogeneous and sterile.

Statistics. The data obtained for each parameter (turbidity, color, pH, conductivity, suspended solids, nitrates, iron, manganese, organic matter, and total and fecal coliforms) were analyzed using one-way ANOVA to compare the effects of Moringa oleifera extracts according to the extraction method (hot and cold). The null hypothesis (H_0) tested stipulated that there was no significant difference between the treatment means.

The analyses were performed with a significance threshold of $\alpha = 0.05$. When the p-value was < 0.05, the null hypothesis was rejected, indicating a significant difference between the treatments. In cases where significant differences were observed, a post-hoc multiple comparison (Tukey HSD) test was applied to identify homogeneous groups of means. All statistical analyses were performed using the R software version x64 4.1.3. The results were presented as mean ± standard deviation (SD).

3. Results and discussion

3.1. Extraction yield results

Table I shows the extraction yields for Moringa oleifera and Adansonia digitata seeds, demonstrating higher oil yields from hot extraction (32.32 and 20.06 %) compared to cold extraction (25.27 and 16.11 %).

Table I. Extraction yields and quantities of Moringa oleifera and Adansonia digitata oil

	Moringa oleifera		*Adansonia digitata*	
	Hot extraction	Cold extraction	Hot extraction	Cold extraction
Weight before extraction (P1)	300g	300g	300g	300g
Weight after extraction (P2)	203.05g	224.18g	239.82g	251.67g
Yield	32.32%	25.27%	20.06%	16.11%
Quantity of oil extracted	98 ml	87 ml	63 ml	58 ml

From Table I, it is evident that hot extraction produced higher yield (32.32%) than cold extraction (25.27%), due to the fact that the high temperature (68°C) of the soxhlet allowed for better extraction, because it made the oil lighter. Similarly, hot extraction for Adansonia digitata also showed higher yield (20.06%) than cold extraction (16.11%), likewise due to soxhlet temperature (68°C). Compared to Adansonia digitata (63 ml for hot and 58 ml for cold extraction), Moringa oleifera seeds contained more oil (98 ml for hot and 87 ml for cold extraction).

3.2. Results of flocculation (jar) tests

Tables II-V below illustrate the results of jar tests for both biocoagulants for various turbidity levels. The tables show that Moringa oleifera consistently outperformed Adansonia digitata in clarifying raw water of the N'djili River. The tables identify the optimal dosages for Moringa oleifera extracts across different turbidity levels, ranging from 150 mg/l (for low turbidity water) to 600 mg/l (for highly turbid water).

Table II. Jar test results for Moringa oleifera and Adansonia digitata extracts on raw water of the N'djili River with low turbidity (below 100 NTU) – 39.6 NTU, pH 5.7, 350 Hazen color

Beaker (B)	*Moringa oleifera* extracts						*Adansonia digitata* extracts					
	Hot extract			Cold extract			Hot extract			Cold extract		
	Doses injected (mg/l)	Turbidity (NTU)	Flock appearance	Doses injected (mg/l)	Turbidity(NTU)	Flock appearance	Doses injected (mg/l)	Turbidity (NTU)	Flock appearance	Doses injected (mg/l)	Turbidity (NTU)	Flock appearance
B1	50	26.1	+	55	22.4	+	0	26.8	-	0	29	-
B 2	100	13.2	++	110	11.3	++	65	31.4	-	60	33.6	-
B 3	150	4	+++	165	3.6	+++	130	34.2	-	120	34.8	-
B 4	200	1.62	++++	220	1.2	++++	195	36.3	-	180	40.5	-
B 5	250	1.62	++++	275	0.9	++++	260	41.9	-	240	44.1	-
B 6	300	1.45	++++	330	1.1	++++	325	44.8	-	300	49.3	-

The minimum doses of 150 and 165 mg/l of hot and cold de-oiled Moringa oleifera extracts were enough to reduce raw water turbidity to 89.9 and 90.9%, respectively. That was because the hot extract exhibited a slightly higher degree of purity than the cold one. No Adansonia digitata extract formed flocs. That failure may be due to the lack of biocoagulant proteins in the seeds, or that they were denatured. The observed drop in turbidity was due to the mechanical action of the flocculator propellers.

Table III. Jar test results for Moringa oleifera and Adansonia digitata extracts on raw water of the N'djili River with average turbidity (100-200 NTU) – 108 NTU, pH 6.1, 500 Hazen color

Beaker (B)	*Moringa oleifera* extracts						*Adansonia digitata* extracts					
	Hot extract			Cold extract			Hot extract			Cold extract		
	Doses injected (mg/l)	Turbidity (NTU)	Flock appearance	Doses injected (mg/l)	Turbidity(NTU)	Flock appearance	Doses injected (mg/l)	Turbidity (NTU)	Flock appearance	Doses injected (mg/l)	Turbidity (NTU)	Flock appearance
B1	0	88.9	-	0	92.7	-	0	-	-	0	-	-
B 2	50	57.3	+	55	65.4	+	65	-	-	60	-	-
B 3	100	13.8	++	110	35.5	++	130	-	-	120	-	-
B 4	150	4.0	+++	165	6.9	+++	195	-	-	180	-	-
B 5	200	2.9	++++	220	4.2	++++	260	-	-	240	-	-
B 6	250	3.1	++++	275	5.2	++++	325	-	-	300	-	-

Table III shows that the rate of 150 mg/l of the hot extract remained 96.3% effective for average turbidity, while the dose of the cold extract had to be 220 mg/l to clarify the water to 96.1%. The increase depended on the degree of purity. Regardless of the extraction method employed, Adansonia digitata extracts did not clarify the water. This suggests that the proteins contained in the extracts produced did not have biocoagulant power, or were all denatured.

Table IV. Jar test results for Moringa oleifera and Adansonia digitata extracts on raw water of the N'djili River with high turbidity (200-500 NTU) – 380 NTU, pH 5.9; 500 Hazen color

Beaker (B)	*Moringa oleifera* extracts						*Adansonia digitata* extracts					
	Hot extract			Cold extract			Hot extract			Cold extract		
	Doses injected (mg/l)	Turbidity (NTU)	Flock appearance	Doses injected (mg/l)	Turbidity(NTU)	Flock appearance	Doses injected (mg/l)	Turbidity (NTU)	Flock appearance	Doses injected (mg/l)	Turbidity (NTU)	Flock appearance
B1	0	278	-	0	289	-	0	-	-	0	-	-
B 2	50	189	+	55	191.4	+	65	-	-	60	-	-
B 3	100	129	++	110	157.8	++	130	-	-	120	-	-
B 4	150	36	++	165	41.1	++	195	-	-	180	-	-
B 5	200	7.5	++++	220	11	++++	260	-	-	240	-	-
B 6	250	4.1	++++	275	4.9	++++	325	-	-	300	-	-

The doses of 250 and 275 mg/l reduced raw water turbidity to 98.9 and 98.7%, respectively, for the hot and cold Moringa oleifera extracts. The higher the turbidity, the more effective the extracts became. That is explained by the fact that high turbidity allows the biocoagulant proteins to react well, forming many flocs resulting from the attraction of the (negative) charges of the colloidal materials responsible for this high turbidity and positive charges of the biocoagulant proteins.

The observations were quite the opposite for the Adansonia digitata powders. Despite the higher turbidity observed, no flocs formed in the beakers. The explanation for that is the same as above.

Table V. Jar test results for Moringa oleifera and Adansonia digitata extracts on raw water of the N'djili River with very high turbidity (above 500 NTU) – 960 NTU, pH 6.2, 500 Hazen color

Beaker (B)	*Moringa oleifera* extracts						*Adansonia digitata* extracts					
	Hot extract			Cold extract			Hot extract			Cold extract		
	Doses injected (mg/l)	Turbidity (NTU)	Flock appearance	Doses injected (mg/l)	Turbidity(NTU)	Flock appearance	Doses injected (mg/l)	Turbidity (NTU)	Flock appearance	Doses injected (mg/l)	Turbidity (NTU)	Flock appearance
B1	400	19.4	++	440	27	++	0	-	-	0	-	-
B 2	500	9.2	+++	550	16.1	++	520	-	-	480	-	-
B 3	600	5.2	++++	660	5.7	+++	650	-	-	600	-	-
B 4	700	5.5	++++	770	6.1	++++	780	-	-	720	-	-
B 5	800	7.6	++++	880	7.3	++++	910	-	-	840	-	-
B 6	900	9.2	++++	990	11	++++	1040	-	-	960	-	-

Table V shows that 600 mg/l of hot extract and 660 mg/l of cold extract of Moringa oleifera clarified the most turbid raw water up to 99.5 and 99.4%, respectively. The high dose of biocoagulants required was directly related to high turbidity. In contrast, no Adansonia digitata extract was able to form the flocs in the same raw water.

3.3. Moringa oleifera settling time (TD) monitoring

Table VI presents data on the settling times for Moringa oleifera extracts, indicating that 60-90 minutes were sufficient for effective clarification.

Table VI. Monitoring of settling time (TD) for Moringa oleifera extracts

T.D. (min)	Moringa hot-extract Solution: 10 g/l				Moringa cold-extract Solution: 11 g/l			
	Raw water turbidity				Raw water turbidity			
	39.6 NTU	108 NTU	380 NTU	960 NTU	39.6 NTU	108 NTU	380 NTU	960 NTU
30	16.6	11.3	36	47	19	14.3	34.1	68
60	4.2	5.1	6.5	19	5.4	6.7	9.6	28
90	4	4	4.1	5.2	4.2	4.2	4.9	5.7
120	3.6	2.9	3.1	3.4	3.7	3.2	3.6	2.8

The results in Table VI show that a necessary time between 60 and 90 minutes was sufficient to obtain turbidity below 5 NTU. The settling time was reduced because Moringa oleifera extracts were de-oiled, which prevented the floating effect of oils and favored proper settling.

3.4. Physico-chemical parameters

Table VII and Fig. 2-4 show the values for physico-chemical parameters (turbidity, conductivity, color, pH, temperature, complete alkaline strength (CAS), hydrotimetric titer (HT), suspension materials (SM), nitrates, iron (Fe), manganese (Mn), and organic matter (OM)) of the raw, decanted, and filtered water of the N'djili River water samples, treated with the hot and cold extracts of Moringa oleifera seeds. The figures demonstrate the influence of each of the steps on the parameters.

Table VII. Means and standard deviations of the effects of Moringa oleifera extracts on physico-chemical parameters

		Moringa hot-extract		Moringa cold-extract		P	WHO standard (2017)
Parameters	Raw Water	Decanted water	Filtered Water	Decanted water	Filtered Water		
Turbidity (NTU)	204.5±13.3	3.8±0.6	3.3±1.3	4.3±0.8	3.9±0.5	0.00	5
pH	6.3±0.1	6.0±0.3	6.2±0.3	6.0±0.2	6.1±0.1	0.05	6.5-8.5
Color (Hazen)	500.0±0.0	23.3±15.3	16.7±5.8	26.7±5.8	23.3±5.8	0.00	20
Temperature (°C)	28.3±1.2	29.3±1.4	28.3±2.5	29.0±1.7	29.2±2.7	0.22	
Conductivity (µS/cm)	31.6±0.6	57.3±9.6	65.4±2.4	55.5±4.2	65.0±4.0	0.7	125
CAS (°F)	1.2±0.4	0.6±0.1	0.7±0.3	0.7±0.2	0.9±0.1	0.03	
HT (°F)	5.1±2.9	1.3±0.2	1.3± 0.6	1.3±0.3	1.4±0.7	0.00	

Table VII. Cont.

SM (mg/l)	246.0±143.8	0.0±0.0	0.0±0.0	0.0±0.0	0.0±0.0	0.02	0.0
OM (mgd'O2/L)	83.3±2.9	146.7±60.5	164.0±80.0	208.3±76.4	158.3±55.8	0.03	5
Fe (mg/l)	6.1±0.4	0.3± 0.2	0.1±0.0	0.6±0.8	0.3±0.4	0.973	0.1
Mn (mg/l)	1.2±1.7	0.1±0.1	0.1±0.0	0.3±0.4	0.3±0.4	0.01	0.05
Nitrates (mg/l)	28.5±28.1	4.0±3.0	4.7±3.5	5.3±3.6	4.2±2.9	0.00	25

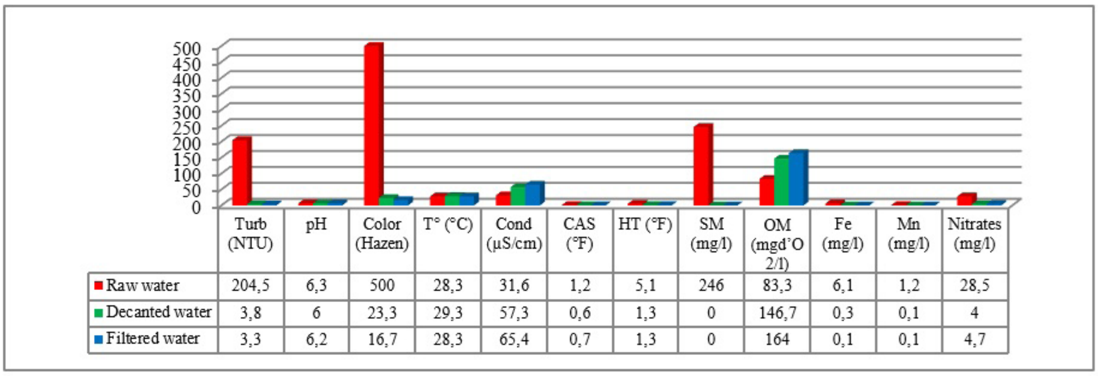

Figure 2. Effect of Moringa oleifera hot extracts on physico-chemical parameters

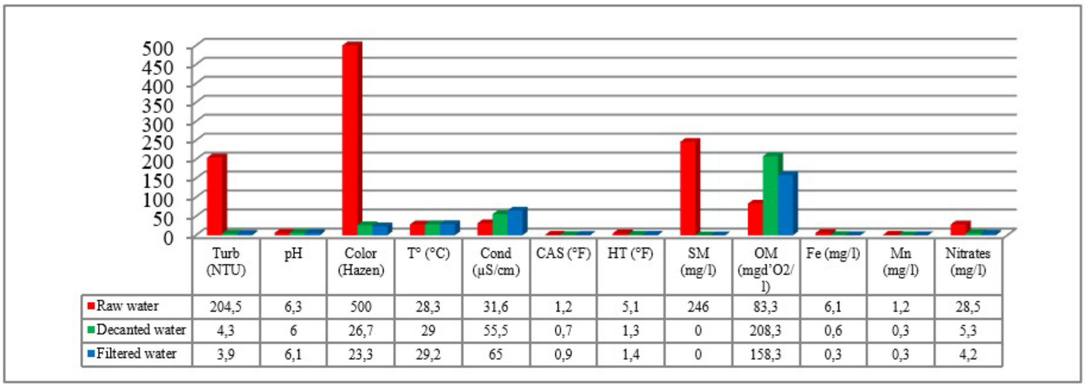

Figure 3. Effect of Moringa oleifera cold extracts on physico-chemical parameters

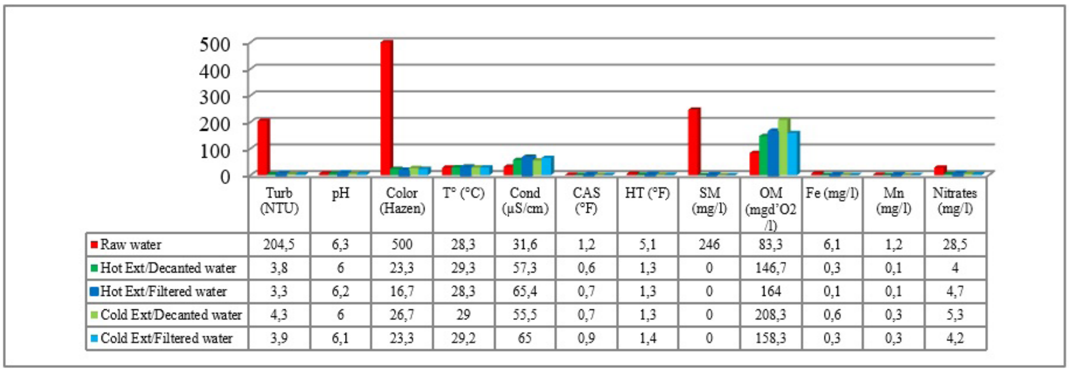

Figure 4. Effect of Moringa oleifera extracts (hot and cold) on physico-chemical parameters

The turbidity of raw water was significantly reduced by the target hot and cold extracts (98.1 and 97.9%, respectively) within the recommended limits (5 NTU according to WHO, 2017). That was due to the fact that the proteins contained in the extracts effectively neutralized the colloids responsible for initial turbidity. The filter did not significantly reduce turbidity because its particle size (0.7-1.2 mm) and bed height (60 cm) were insufficient to retain the remaining colloids. A significant difference between the effects of moringa extracts on turbidity ($p = 0.000$) was observed, i.e. the hot extract demonstrated a higher biocoagulant power than the cold one.

The initial conductivity almost doubled after adding the extracts and passing through the filter but remained below the recommended value (125 µS/cm) in all cases. This variation may be due to the fact that the extracts contained ions that they released into the water to agglomerate the colloids; the rest of the ions were added to the number of the initial ones and thus increased the value; and by passing through the filter, the water carried other ions that remained in the latter. There was no significant difference between the effects of the target extracts on this parameter ($p = 0.7$).

All Moringa oleifera extracts reduced the color of raw water (95.4% -- hot, and 95.6% -- cold) within the recommended limits (20 Hazen). This is due to the fact that the colloidal particles responsible for color were removed after their coagulation and decantation by the active principle released by the extracts into the raw water. The difference between the effects of moringa extracts on color ($p = 0.000$) was significant.

The extracts and the filter had little influence on pH. Although their values remained slightly below the recommended range (6.5-8.5), they did not make the treated water unsafe to drink. This is due to the fact that no H^+ ions were released into the water after adding the extracts; yet, some initial ions had reacted with other negatively charged compounds.

The temperature values did not vary much. However, they could be influenced by the ambient environment (laboratory and filter). No significant difference was noted between the effects of moringa extracts on temperature.

The extracts reduced the CAS and HT values by half. This could be due to the fact that some calcium and magnesium carbonates and bicarbonates reacted with the active principle during coagulation-flocculation, precipitated and then were removed from the water after floc decantation. The p-values (0.03 and 0.000, respectively) were higher than that of α in the first case and lower in the second, so the effects of the extracts on the total water alkalinity were the same; although, they were significantly different in total hardness.

The extracts (hot and cold) were able to remove 100% of suspended matter, bringing the water back to the standard (0.0 mg/l). This is due to the fact that the SM all agglomerated after adding the active ingredient. The difference between the effects of the target extracts on this parameter ($p = 0.002$) was significant. The nitrate values (NO_3^-) dropped to 86.3% for the hot extract, and to 81.9% for the cold extract, both bringing the parameter back to the set limit (25 mg/l). This is attributable to coagulation flocculation. They acted the same way on this parameter ($p = 0.000$)

Moringa oleifera extracts significantly reduced iron (94.4% -- for the hot extract, and 88.9% -- for the cold extract), allowing the filter to bring these values back to the standard (0.1 mg/l). This is attributed to the bio-adsorbent power of the active ingredient contained in the extracts – iron, being a metal, was adsorbed by the flocs and removed after coagulation and decantation. No significant difference between the extracts on the adsorbent power of iron ($p = 0.973$) was observed. The manganese (Mn) values of the raw water were reduced by the hot extract to 90.9% and the cold extract to 72.7% without being within the standard (0.05 mg/l). This reduction is attributed to the adsorption of manganese by the active ingredient. A significant difference was observed between the effects of the extracts on manganese adsorption ($p = 0.01$).

An increase in organic matter (OM) after adding the extracts was observed – the cold moringa extract brought more, but the difference between the two was not significant ($p = 0.03$). This could be due to the fact that the target biocoagulants, themselves being organic matter, were added to that present in the raw water. The reduction in the filter was linked to the low bacteriological activity following the non-significant residence time (69 minutes). All those figures remained higher than the standard (5 mg/l), requiring an adjuvant treatment by extending residence time.

3.5. Bacteriological parameters

Table VIII and Fig. 5 show the values of bacteriological parameters (Total and Fecal Coliform) for the raw and decanted water from the N'djili River samples, as well as the influence of Moringa oleifera extracts on them.

Table VIII. Means and standard deviations of the effects of Moringa oleifera extracts on Total coliforms (TC) and Fecal coliforms (FC)

		Hot moringa extract	Cold moringa extract	p	WHO standard (2017)
Parameters	Raw water	Decanted water	Decanted water		
TC (CFU).10^3	43.667±6.7	5.3±3.1	7.667±4.0	0.000	0
FC (CFU).10^3	43.667±6.7	5±2.6	4±2	0.005	0

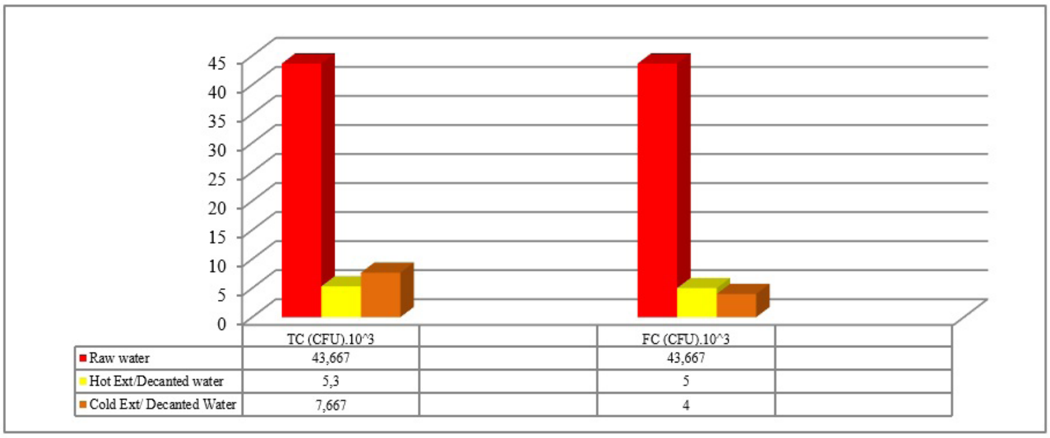

Figure 5. Effects of Moringa oleifera extracts on Total coliforms (TC) and Fecal coliforms (FC)

Table VIII and Fig. 5 demonstrate the effects of the moringa extracts on the raw water, with the number of TC reduced to 88% by the hot extract, and to 82% by the cold extract. This is due to the adsorption of these germs on the colloids and suspension materials (SM), coagulated and decanted later on. The number of FC dropped by over half after decantation (55.9% with the hot extract, and 64.7% with the cold extract). This reduction is attributed to the adsorption of bacteria onto suspended matter and colloids, subsequently removed during coagulation decantation. The results remained above the WHO standard (0colonie/liter). The extracts reduced coliforms significantly differently in the raw water (p = 0.00; 0.005, respectively, for TC and FC).

3.6. Chlorine demand

Table IX highlights the disinfection rates retained for each water type treated respectively with moringa hot and cold extracts, as well as shows fairly high chlorine consumption.

Table IX. Result of chlorine demand for Moringa oleifera extracts

Settings	Moringa hot extract						Moringa cold extract					
	F1	F2	F3	F4	F5	F6	F1	F2	F3	F4	F5	F6
Doses injected (mg/l)	10	11	12	13	14	15	10	11	12	13	14	15
Residual chlorine	0.1	0.2	0.3	0.2	0.2	0.1	0.1	0.2	0.3	0.4	0.3	0.2
Demand	9.9	10.8	11.7	12.8	13.8	14.9	9.9	10.8	11.7	12.6	13.7	14.8

Table IX indicates that with the chlorine levels of 12 and 13 mg/l, the clarified water can be disinfected both with Moringa oleifera hot and cold extracts. However, the residual chlorine levels did not reach the WHO recommended levels for domestic tap water. Such disinfection requires high chlorine doses, likely due to reactions with oxidizable materials forming haloforms (trihalomethanes) before killing the bacteria.

4. Discussion

The oil extraction results match these of Edogbanya et al. (2015) and Rakotoniriana et al. (2015), as well as reveal that hot extraction generates better yields than cold extraction, on the one hand, and on the other hand, Moringa oleifera gives a better yield than Adansonia digitata.

The jar test results obtained within the study showed that whatever the concentrations, doses, and turbidities of the raw water, Adansonia digitata powders did not clarify it. This may be due to the fact that these plant seeds did not contain biocoagulant proteins, or that they were denatured by oil extraction techniques. These findings contradict these of Edogbanya et al. (2013), who reported clarification of synthetic water with this plant's extracts.

Hot and cold de-oiled Moringa oleifera seed extracts effectively reduced the turbidity of surface waters within the standard (5 NTU) set by WHO (2017).

During the jar test, flocs appeared in the cloudy water approximately 5 minutes after agitation began. The observed drop in water turbidity may be due to the seeds containing coagulating proteins and/or polysaccharides with charged ions, which – once released into the water – absorb and neutralize oppositely charged colloids responsible for water turbidity. This results in the agglomeration of colloids and floc formation, which are then decanted, favoring the clarification of the cloudy water. These findings are similar to these of Fatombi et al. (2008) and Kabore et al. (2013).

The optimal doses of 150, 150, 250, and 600 mg/l were retained for turbidities of 39.6, 108, 380, and 960 NTU for hot extracts, respectively. For cold extracts, the doses of 165, 220, 275, and 660 mg/l were also retained in the same raw waters. As the turbidity increased, the dose of biocoagulants grew as well – this probably leads to an increase in the availability of ionic charges responsible for clarifying water. These findings are consistent with these of Edogbanya et al. (2013). These quantitative results provide precise data on the optimal doses required for different turbidity levels, contributing to practical recommendations for water treatment. The doses are lower (900-1,000 mg/l) than these of walnut husk biocoagulants (Zourif et al., 2024).

The necessary settling times of 60-90 minutes made it possible to settle the flocs until obtaining a turbidity below 5 NTU as per the WHO standard, which differs from the findings of Fatombi et al. (2008), Kabore et al. (2013), Ngbolua et al. (2016), and Rakotoniriana et al. (2015). This outcome can be attributed to lipid removal in the extracts under this study, that further purified the biocoagulants and reduced their floating character, confirming the hypothesis of Rakotoniriana et al. (2015).

The results for turbidity, color, CAS, HT, SM, nitrates, iron, Mn, TC, and FC revealed that Moringa oleifera extracts were capable of reducing these parameters and bringing them within the recommended limits. However, the variance analyses demonstrated significant differences between the effects of Moringa oleifera extracts on turbidity, color, iron, TC, and FC, on the one hand, and no significant difference in the effects of these extracts on CAS, HT, SM, nitrates, and manganese, on the other hand. These findings are similar to those of Kouniba et al. (2024a), and Kouniba et al. (2024b), who demonstrated that biocoagulants such as banana peel and watermelon powder can remove turbidity up to 97% and 99.21%, respectively – i.e. that biocoagulants can well replace conventional coagulants. The findings of this investigation are also confirmed by these of Huynh et al. (2020), who – in their work on the use of Moringa oleifera seed powder as biocoagulants for surface water treatment – evaluated the effectiveness of surface water treatment in Vietnam using Moringa oleifera seed by-product as a biocoagulant to eliminate turbidity and natural organic matter. Indeed, this study shows that Moringa oleifera as a biocoagulant has more advantages than others, including no chemicals used, low cost, ease of implementation, and equivalent effectiveness and safety for health.

In addition, water analyses showed that Moringa oleifera extracts increased electrical conductivity and organic matter values.

The ANOVA indicated no significant difference in their effects on conductivity, with an opposite effect on organic materials. It could be caused by the biocoagulants themselves being organic materials and containing releasable ions. These findings are confirmed by these of Fatombi et al. (2008), Kabore et al. (2013), Ngbolua et al. (2016), Rakotoniriana et al. (2015). Thus, such water can only be consumed in an emergency; otherwise, an adjuvant treatment should be considered to make it drinkable and ensure storage.

The extracts did not demonstrate a noticeable effect on pH and T°, because no H+ ions were released into water after their addition; however, some initial ions reacted with other negatively charged compounds. As mentioned above, after coagulation and decantation of the raw water with Moringa oleifera extract, the reduction of 82-87% (for TC) and 56-65% (for FC) – mainly linked to the adsorption of these germs on SM and other colloids (Tangou, 2018) – did not render the water bacteriologically safe for drinking.

The findings for the chlorine demand revealed high consumption of the disinfectant. This is explained by the presence of large amounts of organic matter, oxidized by chlorine, likely forming haloforms before killing the germs.

The rate of 12 mg/l with the demand of 11.7, and 13 mg/l with the demand of 12.6 mg/l, respectively, for the hot and cold extracts were retained for good disinfection, resulting in residual chlorine levels between 0.2-0.3 mg/l.

Based on the results of clarification (turbidity), adsorption (ions), and disinfection (TC), it appears that there is a significant difference between Moringa oleifera hot and cold extract. This could be due to the fact that the extractive yield of oil when hot is higher than that when it is cold (32.32 and 25.27%), which thus increases the biocoagulant power. This finding contradicts Rakotoniriana et al. (2015), who suggested that hot extraction destroyed biocoagulant proteins.

However, the two extraction methods gave the same results arithmetically, i.e. either of them can be used to bring target parameter values within the limits set by the WHO standard.

Biocoagulants, such as Moringa oleifera and Adansonia digitata seeds, have several significant advantages. First, being of natural origin, they are ecologically friendly and reduce environmental impact compared to synthetic chemicals. Their non-toxicity makes them safe alternatives for drinking water treatment, without risks to human health. In addition, these biocoagulants can be less expensive to produce, especially in the Democratic Republic of Congo, where these plants are abundant, thus facilitating access to drinking water, especially in rural areas. Their effectiveness in reducing water turbidity is also well established, and, being biodegradable, they do not leave harmful residues in the environment.

However, there are also disadvantages to using biocoagulants. Their efficiency can vary depending on seed quality and environmental conditions, making their performance less predictable compared to chemical coagulants. The preparation of biocoagulant solutions can be complex and require additional steps, which can complicate their application. In addition, the optimal doses required to treat very turbid water can be high, posing availability challenges. Settling times can likewise be longer, requiring a prolonged wait before water is ready for consumption. Moreover, they drastically increase the organic matter in the treated water, making disinfection (with chlorine) and water conservation difficult. Finally, the lack of knowledge/ training on the effective use of such biocoagulants can limit their adoption in some regions.

The target biocoagulants, particularly Moringa oleifera, are economically more advantageous than chemical coagulants such as aluminum sulfate. They offer a lower purchase cost, do not leave toxic residues, and reduce expenses related to waste management and disinfection.

Conclusion

This study evaluated the effectiveness of biocoagulants produced from Moringa oleifera and Adansonia digitata seeds in preparing drinking water from surface water. The study's findings show that Adansonia digitata seeds have low extraction efficiency and cannot clarify raw water from the N'djili River; whereas Moringa oleifera seeds are effective and can replace aluminum sulfate in water treatment. Oil extraction enhanced biocoagulant activity, with hot extraction performing better than cold extraction. The optimal doses to treat different turbidities range between 150-600 mg/l for hot extracts and from 165 to 660 mg/l for cold extracts, highlighting that the consumption of active ingredient depends on the initial turbidity. The required settling time was identified between 60 and 90 minutes, and a chlorine rate of 12 to 13 mg/l was necessary for disinfection.

In conclusion, Moringa oleifera extracts demonstrate superior biocoagulant power and can replace chemical coagulants. Further research is recommended to explore other local plants and optimize the use of biocoagulants in water treatment.

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