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Effect of 24, 48 and 72 hours fermented leaves extract of mistletoe on some physical properties of water

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Abstract

This study evaluated the effects of different fermentation durations of *Tapinanthus bangwensis* extracts on water quality. Water samples were treated with extracts fermented for 24 hrs, 48 hrs, and 72 hrs and monitored over 20 days for pH, electrical conductivity (EC), total dissolved solids (TDS), turbidity, color, and microbial activity. Results showed that 24 hrs fermentation maintained stable pH (6.4–8.5), moderate EC and TDS, low turbidity, and minimal color change, indicating balanced water chemistry and controlled microbial activity. The 48 hrs extract exhibited peak microbial activity and higher pH, EC, and TDS, accompanied by increased turbidity and noticeable darkening of water, reflecting active nutrient release. In contrast, 72 hrs fermentation caused variable pH, fluctuating EC and TDS, high turbidity, strong odor, and pronounced discoloration, indicative of over-fermentation and microbial overgrowth. Days 8–12 consistently corresponded to maximum microbial and physico-chemical changes, while Days 16–20 showed partial stabilization. The findings indicate that short-term fermentation (24 hrs) produces water suitable for aquaculture, 48 hrs fermentation enriches water for irrigation, and prolonged fermentation (72 hrs) deteriorates water quality, rendering it unsuitable for domestic or aquaculture use. This study underscores the importance of optimizing fermentation duration to balance water quality and nutrient release from mistletoe extracts.

Keywords: *Tapinanthus bangwensis*; Fermentation duration; Water quality; PH; Electrical conductivity (EC); Total dissolved solids (TDS); Turbidity; Color change; Microbial activity; Aquaculture; irrigation; Nutrient release; Over-fermentation

1. Introduction

Water is a fundamental component of life, and access to clean, safe drinking water is essential for human health, agricultural productivity, and ecological sustainability. Despite its importance, global water resources face continuous threats from both natural and anthropogenic factors, including industrial discharge, agricultural runoff, domestic waste, and emerging contaminants such as pharmaceuticals and personal care products (Gleick, 2014; Duru et al., 2020).

In Nigeria, the challenges of water pollution are intensified by poor waste management systems, rapid urbanization, and inadequate access to potable water in rural communities (Nwankwoala, 2011). This has necessitated the search for affordable, environmentally friendly, and sustainable water purification techniques that can be adapted to local conditions. One such technique is phytoremediation, which utilizes plant-based materials to remove or neutralize pollutants in water. Water quality is typically assessed using parameters such as pH, electrical conductivity (EC), and total dissolved solids (TDS). These parameters provide critical insights into the chemical balance, ionic concentration, and potability of water (EPA, 2018). High TDS and EC levels may signal the presence of harmful contaminants like heavy metals and nitrates, while abnormal pH levels can affect the solubility and toxicity of waterborne chemicals. Hence, a

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systematic investigation into affordable and natural solutions to manage these parameters is crucial, particularly for rural areas where conventional water treatment technologies are lacking.

This study, therefore, seeks to assess the effectiveness of mistletoe (*Tapinanthus bangwensis*) in improving water quality, specifically focusing on its impact on pH, EC, and TDS. The outcome of this study is expected to contribute to sustainable water management practices and offer an indigenous alternative to conventional purification methods.

Several recent studies have focused directly on the use of mistletoe in water purification. Etukudo et al. (2021) in a study at the University of Uyo treated water samples from community wells in Uyo and Ibesikpo-Asutan with powdered *T. bangwensis* leaves. They observed a 30% drop in TDS, a 38% reduction in EC, and pH stabilization around 7.0 after 24 hours. These findings confirmed the plant's buffering and sedimentation capacity in naturally contaminated water. Ibanga and Udo (2020) examined the effect of mistletoe-treated water on microbial content in river samples from Ikot Abasi. Results showed a significant reduction in total coliform counts, suggesting antimicrobial effects likely due to mistletoe's bioactive compounds.

Adebayo et al. (2019) compared *T. bangwensis* with Moringa oleifera in treating polluted stream water in Oyo State. Although Moringa had stronger coagulating effects, mistletoe proved better at maintaining pH balance and improving taste, indicating potential for community-scale water management. Uwem and Inyang (2022) carried out field applications of mistletoe extracts in rural Esit Eket where residents use stream and shallow well water. The study confirmed that households using mistletoe filtrates reported improved water clarity, less odor, and fewer waterborne illnesses over a 3-month trial period.

Water pollution has become a persistent environmental and public health challenge globally, and Nigeria is no exception. Despite interventions by governmental and non-governmental bodies, many Nigerian communities still rely on untreated surface and groundwater sources (Nwankwoala, 2011; Olalekan et al., 2020). These sources are often contaminated with heavy metals, organic waste, microbes, and dissolved solids, leading to severe health and environmental risks.

The rise in total dissolved solids (TDS), electrical conductivity (EC), and pH irregularities has highlighted the need for improved water quality monitoring and treatment. The World Health Organization (WHO, 2019) warns that elevated levels of TDS and EC may indicate the presence of toxic ions such as arsenic and lead which pose serious health threats. Inconsistent pH values can further complicate the chemistry of water, making it unsafe for consumption.

Contaminated water is a known vehicle for the transmission of diseases such as diarrhea, cholera, typhoid, and hepatitis (WHO, 2019). If *Tapinanthus bangwensis* proves effective in reducing harmful water parameters like TDS and abnormal pH, Its usage can contribute to preventing waterborne diseases, especially in rural areas where access to modern healthcare is limited. This promotes public health awareness and highlights the importance of integrating local plants in community-based water solutions.

This Study offers practical implications by demonstrating that low-cost indigenous plant materials could be harnessed for water treatment, potentially reducing reliance on expensive chemical treatments and imported technologies. This makes water purification affordable and sustainable, especially in resource-poor settings.

This also access the impact of *Tapinanthus bangwensis* extract on the pH level of water samples, determines the effect of the *Tapinanthus bangwensis* extract on electrical conductivity (EC), indicating the ionic content of the water sample, evaluate the ability of the extract to reduce total dissolved solid (TDS) in water which reflect the level of impurity, compare the water quality parameters before and after treatment with mistletoe to determine the plant's effectiveness in regulating water quality parameters.

2. Materials and methods

The African mistletoe (*Tapinanthus bangwensis*) was selected as the plant of interest due to its widespread ethno-botanical applications and relevance to water quality studies. The mistletoe specimens were specifically harvested from citrus trees, as host specificity may influence the chemical composition of the plant. The collection process involved the use of a cutlass to carefully detach the mistletoe from its host tree. Once harvested, the mistletoe was manually processed by separating the leaves from the stems. This step was crucial as the leaves contain the highest concentration of bioactive compounds, making them the primary focus of the study.

Following separation, the leaves were subjected to sun-drying for several days to reduce moisture content. The drying process was conducted under ambient environmental conditions, ensuring gradual dehydration without excessive exposure to direct sunlight, which could degrade certain phytochemicals. The drying duration was carefully monitored until the leaves attained a brittle texture, indicating complete desiccation. Once fully dried, the mistletoe leaves were pulverized using a traditional mortar and pestle. This method of grinding was preferred as it ensures a finer powder consistency, facilitating efficient extraction of soluble compounds when mixed with water. The powdered mistletoe was then stored in an airtight container to prevent contamination and moisture absorption before proceeding to the preparation stage.

2.1. Preparation of Mistletoe Extract for Experimental Purposes

The finely ground mistletoe powder was transported to a controlled greenhouse environment for extract preparation. A total of 40 grams of the powdered mistletoe was accurately measured using an electronic balance to ensure precision. This quantity was dissolved in 2000 mL of distilled water, and the mixture was stirred thoroughly to promote uniform distribution of the plant material. The solution was then allowed to undergo fermentation for 48 hours under room temperature conditions. Fermentation is a crucial step as it enhances the solubility of bioactive compounds, potentially increasing their effectiveness in influencing water quality parameters.

Following fermentation, the stock solution was prepared for dilution. To evaluate the impact of varying mistletoe concentrations on water quality, four different treatment solutions were formulated through a serial dilution process. The treatment levels were structured as follows:

Treatment layout (final volume = 2000 mL):

- 24-hour fermentation series (5 treatments): Control (0 mL extract + 2000 mL water) 40 mL extract + 1960 mL water, 80 mL extract + 1920 mL water, 160 mL extract + 1840 mL water, 320 mL extract + 1680 mL water.
- 48-hour fermentation series (4 treatments): Control (0 mL extract + 2000 mL water) 50 mL extract + 1950 mL water, 150 mL extract + 1850 mL water, 300 mL extract + 1700 mL water.
- 72-hour fermentation series (5 treatments): Control (0 mL extract + 2000 mL water) 40 mL extract + 1960 mL water, 80 mL extract + 1920 mL water, 160 mL extract + 1840 mL water, 320 mL extract + 1680 mL water.

Each unique treatment (fermentation time × extract volume) was prepared in triplicate (n= 3). Total experimental units = (5 treatments for 24 h × 3 replicates) + (4 treatments for 48h × 3 replicates) + (5 treatments for 72 h × 3 replicates) = 42 experimental units.

The solutions were dispensed into 20-liter plastic buckets, which served as experimental containers. These containers were strategically arranged in a randomized block design within an open-field setting to minimize environmental biases and allow for natural inoculation. The experimental setup was maintained under natural environmental conditions for four days, allowing interactions between the mistletoe extract and the surrounding microbial and abiotic components. During this period, close monitoring was conducted to observe any visible changes in water quality, sedimentation, or microbial activity.

2.2. Limnological Procedures for Evaluating the Impact of Mistletoe Extract in Water Quality

To assess the effects of mistletoe extract on water quality, systematic sampling and analysis were conducted. From each treatment, a 50 mL aliquot of solution was collected at designated time intervals. Given that each treatment was prepared in triplicate, this resulted in a total sample volume of 150 mL per treatment per sampling session.

The collected samples were transported to a laboratory for comprehensive water quality assessment. The key parameters analyzed included:

- pH Level: The pH of each solution was measured using a digital pH meter to determine any fluctuations in acidity or alkalinity due to the presence of mistletoe extract. The pH value is crucial in assessing the potential chemical interactions occurring within the water samples.
- Electrical Conductivity (EC): Conductivity measurements were taken using a conductivity meter to evaluate the extent of dissolved ions present in each treatment. Conductivity serves as an indicator of the mineral content and overall ionic strength of the solution, which may be influenced by the mistletoe extract.
- Total Dissolved Solids (TDS): TDS analysis was conducted to quantify the concentration of dissolved substances in the water samples. This parameter provides insights into the degree of solute dissolution contributed by the mistletoe extract, which may impact the physical and chemical properties of the water.

The analysis was repeated at four-day intervals over a total duration of 20days. This time frame allowed for continuous monitoring of changes in water quality and provided sufficient data for trend analysis.

3. Results biological/microbial

Activity across all fermentation's periods (24 hrs, 48 hrs, And 72 hrs) are as follows:

Across the 20-day observation period, water samples from all fermentation periods showed clear progression in physical and biological characteristics.

- 24 hrs fermentation produced water that remained mostly clear, with minimal color change and low turbidity (0.28–2.15 NTU) across all days. Microbial activity was moderate, supporting controlled decomposition and release of nutrients without altering the physical appearance significantly. Slight peaks in turbidity and minor darkening were observed around Day 8–12, coinciding with active microbial metabolism, before settling by Days 16–20.
- 48hrs fermentation showed more pronounced changes. Turbidity increased sharply with concentration and time, peaking around Days 8–12 (e.g., 2.30– 645 mg/L TDS and 17.48 mS/cm EC), reflecting intense microbial activity and release of organic and mineral compounds. Color darkened progressively, indicating accumulation of microbial metabolites and plant-derived chemicals. By Days 16–20, slight reductions in turbidity and settling of suspended particles occurred, suggesting partial microbial stabilization and sedimentation.
- 72hrs fermentation exhibited the most dramatic changes. Water was initially clear, but by Day 4–8, turbidity and discoloration were pronounced, with darker coloration and stronger odor developing due to prolonged microbial decomposition. Peak turbidity and TDS occurred mid-period, followed by partial decline toward Day 20 as microbial activity waned or shifted toward spoilage organisms. The extended fermentation time led to over-decomposition, creating water with high microbial load, strong odor, and poor visual quality.

Comparative trends across days show that Day 8–12 consistently coincided with peak microbial activity and maximum physical changes across all fermentation periods. Early days (Day 4) displayed moderate microbial growth and slight changes in turbidity and color, while later days (Day 16–20) generally showed partial settling and stabilization, though 72hrs extracts remained darker and more turbid than shorter fermentations.

Table 1 The activity across all fermentation's periods (24hrs, 48hrs, and 72hrs)

Treatment	**pH**			**TDS**			**EC**		
	24 hrs	**48 hrs**	**72 hrs**	**24 hrs**	**48 hrs**	**72 hrs**	**24 hrs**	**48 hrs**	**72 hrs**
Control	7.9	7.8	9.0	57.2	37.0	48.4	1.07	0.83	1.07
40 ml	7.4	–	7.8	76.2	–	40.1	0.95	–	0.95
50 ml	–	9.0	–	–	88.4	–	–	2.44	–
80 ml	7.7	–	7.6	128.6	–	48.2	1.42	–	1.42
150 ml	–	8.4	–	–	186.2	–	–	4.89	–
160 ml	8.0	–	7.7	190.8	–	78.4	2.20	–	2.20
300 ml	–	8.5	–	–	339.8	–	–	8.56	–
320 ml	8.2	–	7.4	246.4	–	102.6	2.95	–	2.95

3.1. Water quality parameters across fermentation periods

3.1.1. pH Dynamics Across All Fermentation Periods

The pH of water fluctuated across the three fermentation periods and over the 20-day observation period. For 24 hrs fermentation, pH values remained generally stable, ranging from 6.4 to 8.5, with mild increases around Day 4–8 at higher concentrations. This indicates moderate microbial activity without causing strong alkalinization. In 48 hrs fermentation, pH values were higher, peaking at 10.3 in the 50 ml sample on Day 8 and 9.3 in the 300 ml sample on Day

12, reflecting more intensive microbial breakdown and release of basic compounds. By Day 16–20, pH declined slightly (7.5– 8.3) in all treatments, showing partial stabilization. For 72 hrs fermentation, pH was highly variable; initial days showed alkalinity up to 9.5, while later days dropped to 6.9–7.5 in some treatments, suggesting over-fermentation and microbial exhaustion. Overall, 24 hrs fermentation maintained the most stable and near-neutral pH across all days, whereas longer fermentation produced fluctuations.

3.1.2. Electrical Conductivity (EC) Trends across Days

EC increased with both fermentation time and mistletoe concentration. For 24 hrs fermentation, values ranged from 0.49 to 6.69 mS/cm, peaking at Day 8–12 in higher concentrations (160- 320 ml), then slightly declining by Day 16–20, suggesting partial sedimentation or microbial assimilation of ions. 48hrs fermentation produced much higher EC, with 300 ml peaking at

17.48 mS/cm on Day 12, reflecting significant ion release. By Day 16, EC dropped in all treatments, indicating possible dilution or precipitation. 72hrs fermentation showed early EC similar to 48hrs but stabilized at lower values (1.32–3.38 mS/cm) by later days, likely due to microbial uptake and sedimentation. In summary, EC rises with fermentation time and concentration, peaking mid-period, thenpartially stabilizing.

3.1.3. Total Dissolved Solids (TDS) Variation across Days

TDS trends mirrored EC patterns. For 24hrs fermentation, TDS ranged from 16– 472 mg/L, peaking around Day 8–12 and declining by Day 16–20, indicating partial sedimentation and microbial uptake of dissolved solids. 48hrs fermentation recorded the highest TDS, with 300 ml reaching 645 mg/L on Day 12, and decreasing by Day 16–20 due to dilution or settling of solids. 72hrs fermentation showed variability, with some peaks (320 ml reached 293 mg/L on Day 8) and stabilization at lower values (58–63 mg/L) in later days, suggesting over-fermentation and nutrient re-uptake. Therefore, 24hrs fermentation maintains moderate and safe TDS, 48hrs enriches water with dissolved solids for nutrient purposes, and 72hrs produces variable results.

4. Discussion

The pH values fluctuated across the three fermentation periods (24, 48, and 72 hrs). At 24 hrs, the pH remained closer to neutrality, making it more stable for aquaculture species that thrive in pH 6.5–8.5. After 48 hrs, the pH tended to increase significantly, indicating alkalinization due to metabolic breakdown and release of basic compounds. By 72 hrs, the pH either stabilized or showed irregular shifts, suggesting over- fermentation and microbial competition. This shows that 24 hrs is the most stable for aquaculture. This is in line with the findings of Boyd and Tucker (2012), who reported that aquaculture systems perform best under stable pH conditions, and corroborates earlier studies on microbial fermentation dynamics (Okpokwasili and Nweke, 2005).

Electrical conductivity (EC) is a measure of dissolved ions in water. At 24hrs, EC values remained relatively low, showing moderate mineralization that supports physiological balance. At 48hrs, EC values increased sharply, suggesting higher ion release from the mistletoe extract. While this may enhance nutrient availability, excessive conductivity can stress aquatic life. At 72hrs, EC values declined or fluctuated, possibly due to precipitation and microbial re-assimilation of ions. Thus, 48hrs favors plant growth systems but not aquaculture. This is consistent with the work of Ayoola (2010), who observed that high EC levels impair aquatic balance but support agricultural nutrient enrichment.

TDS showed a similar trend to EC. At 24hrs, TDS remained within FAO (2019) recommended safe limits for aquaculture (<500 mg/L). At 48hrs, TDS rose drastically, indicating high organic matter release and dissolved compounds. By 72hrs, TDS reduced, likely due to sedimentation and microbial uptake. Hence, 24hrs is best suited for aquaculture, while 48hrs may be more beneficial in agriculture. This corroborates findings by El-Sayed (2006), which highlighted that aquaculture water requires low TDS levels, while agriculture benefits from higher dissolved nutrients.

Fermentation for 24hrs promoted active microbial decomposition without overgrowth. At 48hrs, microbial activity peaked, releasing metabolites such as organic acids, phenolic, and alkaloids. At 72hrs, microbial populations may have declined or shifted towards spoilage organisms, reducing water quality. This suggests 48hrs is optimal for bioactive compound extraction but not for water quality stability. Similar observations were made by Osadebe and Ukwueze (2004), who reported peak metabolite release at intermediate fermentation stages.

Aquaculture species generally prefer water with stable pH, moderate EC, and low TDS. From the results, 24hrs fermentation produced the most stable water chemistry, whereas 48 or 72hrs fermentation water may cause stress due to ion imbalance and pH fluctuations. Therefore, 24hrs is best suited for aquaculture. This finding is in line with FAO (2019), which emphasizes that aquaculture water must be stable and balanced to support survival and growth.

In contrast to aquaculture, crops benefit from nutrient-rich water. The 48 hrs fermentation extract, with higher EC and TDS, may enhance plant growth by supplying dissolved minerals and organic compounds. Thus, the 48 hrs treatment is superior for irrigation. This corroborates findings by Okwu and Josiah (2006), who noted that plant-derived extracts enrich soil fertility when adequately mineralized.

For domestic water use, the 24hrs extract remained safest due to its acceptable physical properties. In contrast, the 72hrs fermentation produced discoloration and odor, making it unsuitable for household applications. This supports observations by Aworinde et al. (2018), who reported that prolonged plant fermentation reduces domestic water acceptability.

The changes in odor, color, and taste across treatments reflect progressive biochemical decomposition. At 24hrs, the extract remained clear and mild. By 48hrs, turbidity and odor increased due to microbial metabolites, while at 72hrs, advanced decomposition produced darker coloration and stronger odor. This is in line with Ajaiyeoba (2002), who linked prolonged fermentation of mistletoe to increased turbidity and undesirable odor.

During the experiment, rainfall reduced pH, EC, and TDS in some treatments, indicating that environmental factors can strongly affect fermentation outcomes. This suggests that outdoor aquaculture systems are vulnerable to such fluctuations, hence controlled fermentation before application is recommended. This observation corroborates findings by Boyd and Tucker (2012), who noted rainfall events as key modulators of aquaculture water quality.

Studies on *Tapinanthus bangwensis* have shown that it contains alkaloids, flavonoids, and tannins. These compounds leach more effectively at 48hrs fermentation, explaining the increased EC and TDS. However, prolonged exposure (72hrs) may lead to toxicity. This is in agreement with earlier reports by Ajaiyeoba (2002) and Okwu and Josiah (2006), who confirmed the presence of these compounds and their extraction dynamics.

The increased EC and TDS at 48hrs suggest higher water hardness. While hard water is unsuitable for delicate aquaculture species, it can enhance shell formation in crustaceans. Thus, the 48hrs extract may be useful in prawn culture but not for finfish. This corroborates findings by Boyd (2015), who emphasized the importance of hardness in crustacean aquaculture.

Over-fermentation (72hrs) may release excess phenolic compounds, making water harmful to aquatic organisms. This agrees with Osadebe and Ukwueze (2004), who noted that mistletoe's bioactive compounds can be beneficial in moderation but toxic in excess.

The extracts demonstrated clear effects on nutrient cycling. At 24hrs, nutrients remained balanced; at 48hrs, enrichment occurred, making it useful for fertilization; while at 72hrs, nutrient imbalance posed risks of eutrophication. This corroborates findings by Ayoola (2010), who observed that organic extracts promote eutrophication if allowed to ferment excessively.

Using mistletoe extracts fermented for 24hrs can improve aquaculture sustainability by maintaining water quality, while the 48hrs extract may be better integrated into agricultural systems. However, the 72hrs extract must be avoided as it reduces water quality. This aligns with FAO (2019) guidelines for sustainable aquaculture practices.

Based on the findings:

- For aquaculture: 24hrs extract is best, as it maintains balanced water parameters.
- For agriculture: 48hrs extract is more beneficial, being nutrient-rich.
- For domestic use: only the 24hrs extract is safe.

The 72hrs extract should be avoided in both aquaculture and domestic applications due to poor water quality and potential toxicity. These findings are consistent with previous studies (Ajaiyeoba, 2002; Okwu and Josiah, 2006; Osadebe and Ukwueze, 2004; Aworinde et al., 2018), which emphasize controlled fermentation periods to maximize benefits while minimizing risks.

5. Conclusion

The study demonstrates that fermentation duration of *Tapinanthus bangwensis* extracts significantly influences the physico- chemical, physical, and biological properties of water. Water treated with 24 hrs fermented extracts maintained stable pH, moderate electrical conductivity, low total dissolved solids, minimal turbidity, and controlled microbial activity, making it most suitable for aquaculture applications. In contrast, 48 hrs fermentation enhanced nutrient release, increased EC and TDS, and promoted microbial metabolite accumulation, making it beneficial for agricultural irrigation but less ideal for aquaculture. Prolonged fermentation for 72hrs led to over-decomposition, excessive turbidity, pronounced discoloration, strong odor, and potential toxicity, rendering it unsuitable for both aquaculture and domestic use.

The findings underscore the importance of optimizing fermentation time to balance water quality, nutrient availability, and microbial activity. Short-term fermentation (24hrs) supports sustainable aquaculture by maintaining water quality, intermediate fermentation (48hrs) can be harnessed for crop irrigation due to nutrient enrichment, and extended fermentation (72hrs) should be avoided due to deterioration of water quality and potential hazards. These results provide practical guidance for the use of mistletoe extracts in aquaculture and agricultural systems, emphasizing controlled fermentation as a key factor for safe and effective application.

Compliance with ethical standards

Disclosure of conflict of interest

If two or more authors have contributed in the manuscript, the conflict of interest statement must be inserted here.

The authors declare no conflict of interest.

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