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Assessing the Ecological Impact of Cassava Mill Effluent on Soil Microbiology and Physicochemistry

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Abstract

This study examined the effects of cassava mill effluent on soil microbiological and physicochemical properties to evaluate its environmental implications. Soil samples from three sites (A-C) were analyzed for microbial diversity and abundance, with a focus on impacted soil contaminated with cassava effluent. Microbial analysis revealed the predominance of Bacillus sp. and Pseudomonas sp. (28.6 %) among bacterial isolates, while Penicillium sp. (27.2 %) was the most common fungal isolate. Impacted soil exhibited higher bacterial counts (7.8×10^5 cfu/g), but fungal growth was suppressed. Physicochemical analysis indicated significant alterations in soil chemistry due to cassava mill effluent, with elevated levels of nitrogen (797 mg/l), potassium (459 mg/l), and phosphorus (432 mg/l) in impacted soil. However, the study also highlighted substantial heavy metal contamination, including copper and iron, posing toxicity risks. The findings suggest that cassava mill effluent enhances microbial activity but concurrently degrades soil quality. Antibiotic sensitivity testing revealed the resilience of certain isolates, such as Bacillus sp., with Ciprofloxacin exhibiting the highest inhibitory activity. The study concludes that while cassava mill effluent introduces beneficial nutrients for microbial growth, its acidity, heavy metal content, and potential to foster antibiotic-resistant microorganisms pose environmental and health risks. These findings emphasize the urgent need for effective effluent management practices to mitigate soil degradation and ecological imbalances.

Keywords: microbiological, physicochemical, effluent, environmental, contamination.

1. Introduction

Cassava (Manihot esculenta Crantz) is a widely cultivated root tuber crop in tropical regions worldwide. As a primary food source, cassava tubers are rich in carbohydrates (85.9 %) and low in protein (1.3 %), with notable amounts of cyanogenic glucoside (Nwabueze, 2007; Nwinanee, 2021). This crop is a staple food item, particularly for low-income earners in Africa and Asia (Desse, Taye, 2001; Edamisan, 2020).

Originating from South America, cassava was introduced to Africa in the 16th century and has since become a major food source in sub-Saharan Africa (Adewoye, 2005; Nwinanee, 2021). In West Africa and Nigeria, cassava is commonly consumed as garri, a fermented cassava product (Cheesbrough, 2005; Okoye, 2020). Nigeria is currently the world's largest producer of cassava, with significant growth in domestic and international demand (Desse, Taye, 2001; Edamisan, 2020).

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The cassava processing industry has expanded in Nigeria, with many small-scale processing units operating in residential areas. However, traditional garri production generates substantial amounts of wastewater, hydrocyanic acid, and organic waste, posing environmental and health risks (Akinfala, Tewe, 2004; Nwinanee, 2021). The indiscriminate discharge of this effluent contaminates agricultural lands, streams, and underground water, threatening the environment and human health (FAO, 2004; Okafor, 2008; Ehiadgonare et al., 2009; Nwinanee, 2021).

Studies have shown that the toxic concentrations of cassava mill effluent can prevent cereal seed germination and pose significant threats to humans and the environment due to its high cyanide content (Olorunfemi, 2008; Ezeogo et al., 2021). Therefore, investigating the environmental impact of cassava processing and implementing regulations for responsible waste management are crucial.

2. Materials and method

Study Area:

This research was conducted at the Department of Human Nutrition and Dietetics, Federal University of Health Sciences, Azare, Bauchi State. The study sample was collected from two distinct locations within Bauchi and Katagum Local Government Areas, situated in the North-Eastern region of Northern Nigeria.

Experimental details

Sample Collection and Preparation

Soil samples were aseptically collected from three cassava mill dump sites (A-C) in two locations within Bauchi State, Nigeria. Samples were collected at 5m and 50m distances from the dump site, with the latter serving as the control. Samples were transported in ice-cold containers to prevent microbial growth and analyzed accordingly.

Serial Dilution and Inoculation

Serial dilution of the samples was performed, and 0.1ml of the appropriate dilution was inoculated into nutrient agar and potato dextrose agar media in triplicate (Cheesbrough, 2005; Okoye, 2020). The inoculated plates were incubated at 37°C for 24h for bacterial enumeration and at room temperature for 5-7 days for fungal enumeration.

Characterization and Identification of Microbial Isolates

Discrete colonies were purified and identified based on cultural parameters, microscopic, and biochemical analysis. Biochemical tests, including Gram staining, motility test, oxidase test, catalase test, citrate test, indole test, methyl red test, and Voges Proskauer test, were conducted to further characterize the isolates (Fawole, Oso, 2004; Cheesbrough, 2005; Okoye, 2020).

Statistical Analysis

Simple ratios and percentages were used to determine relationships, while ANOVA was used to determine statistically significant differences between independent groups.

3. Results

A total of three soil samples were analyzed, with two samples from cassava mill effluent dumpsites and one from fertile, unimpacted farmland. Microbiological analysis revealed the presence of various bacterial and fungal species.

Bacterial Isolates

The identified bacterial isolates included Bacillus sp. (28.6 %), Pseudomonas sp. (28.6 %), Enterobacter sp. (7.1 %), Corynebacterium sp. (14.3 %), Proteus sp. (7.1 %), and Escherichia coli (14.2 %).

Fungal Isolates

The identified fungal isolates included Aspergillus sp. (18.2 %), Penicillium sp. (27.2%), Mucor sp. (9.1 %), Candida sp. (18.2 %), Saccharomyces sp. (18.2 %), and Rhizopus sp. (9.1 %).

Microbial Enumeration

The average microbial count of bacteria and fungi is presented in Figure 1. The total mean heterotrophic bacterial count ranged from $7.0\pm0.11\text{x}10^5$ cfu/g for the control soil, while the effluent and polluted soil were $5.1\pm0.28\text{x}10^5$ cfu/g and $7.8\pm0.19\text{x}10^5$ cfu/g, respectively. The fungal mean ranged from $4.2\pm0.57\text{x}10^5$ cfu/g for the control soil, while the effluent and polluted soil were $4.4\pm0.41\text{x}10^5$ cfu/g and $3.9\pm0.42\text{x}10^5$ cfu/g, respectively.

The impacted soil had the highest bacterial count of 7.8±0.19x10^5 cfu/g. Bacterial counts were higher than fungal counts in all samples.

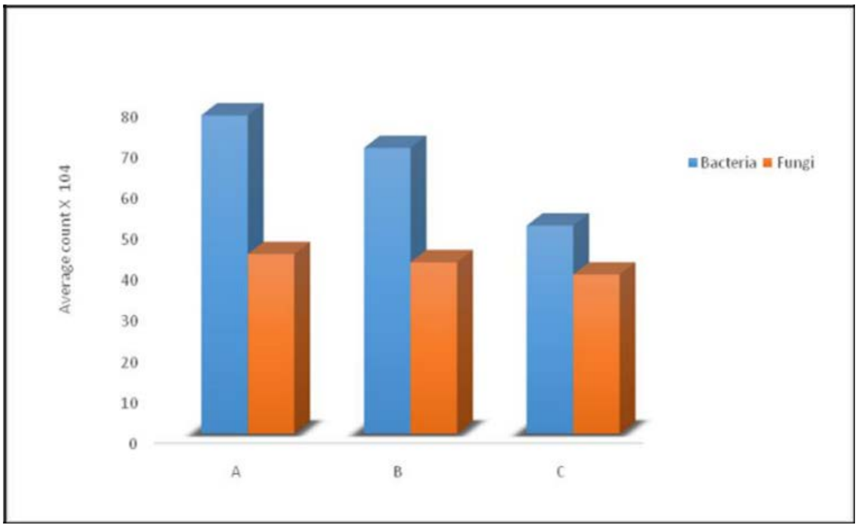

Fig. 1 Bacterial and fungal counts at the impacted soil, un-impacted soil and cassava effluent (A – Impacted soil, B – Un-impacted soil and C – Cassava effluent)

Physico-Chemical Analysis of Cassava Effluent and Soil Samples

The results of the physico-chemical analysis of cassava impacted soil, un-impacted soil, and cassava effluent samples are presented in Table 1. Duplicate data for each parameter were analyzed using One-Way ANOVA at a 0.05 (95 %) significance level.

pH and Nutrient Levels

The cassava effluent sample had an acidic pH of 3.13, while the un-impacted and impacted soil samples had slightly alkaline pH values of 7.87 and 7.21, respectively. The cassava effluent sample contained the highest concentration of nitrogen (797.00 mg/l), followed by the un-impacted soil (95.15 mg/l) and impacted soil (7.51 mg/l). Potassium levels were significantly higher in the cassava effluent sample compared to the impacted and un-impacted soil samples.

Metal Concentrations and Other Parameters

Magnesium levels were similar in the cassava effluent (657.75 mg/l) and impacted soil (660.85 mg/l) samples, but lower in the un-impacted soil sample (36.95 mg/l). Calcium, sodium, and electrical conductivity levels were higher in the effluent and un-impacted soil samples compared to the impacted soil sample. The biological oxygen demand (BOD5) was highest in the impacted soil sample (7.18), followed by the cassava effluent sample (5.73) and un-impacted soil sample (4.54). Dissolved solids were highest in the effluent sample (478.75 mg/l) and lowest in the impacted soil sample.

Heavy Metal Concentrations

The concentration of copper was two- to three-fold higher in the effluent sample compared to the un-impacted and impacted soil samples. Iron, cobalt, and ammonia concentrations were highest in the cassava effluent sample. Zinc concentrations were relatively constant across all three samples. Nitrate levels were highest in the cassava effluent sample (12.01 mg/l), while nitrite levels were highest in the un-impacted soil sample (88.05 mg/l).

Table 1. Physicochemical analysis of cassava effluent, impacted and un-impacted soils samples

Parameters	Un-impacted soil	Impacted soil	Cassava effluent
pH	7.87±0.01a	7.21±0.01b	3.13±0.01c
Nitrogen (mg/l)	95.15±0.10	7.51±0.01	797.00±1.41
Phosphorus (mg/l)	288.85±0.10	79.51±0.01	432.00±1.41
Potassium (mg/l)	66.65±0.10	56.25±0.10	56.25±0.10
Magnesium (mg/l)	36.95±0.10	660.85±0.10	657.75±0.21

Parameters	Un-impacted soil	Impacted soil	Cassava effluent
Calcium (mg/l)	34.15±0.10	6.71±0.01	64.30±1.41
Sodium (mg/l)	53.75±0.10	4.51±0.01	30.90±0.14
Conductivity(μs/cm)	26.35±0.10	8.99±0.01	46.20±0.01
BOD (28 °C)	4.54±0.01	7.18±0.01	5.73±0.01
TDS (mg/l)	57.12±0.01	4.51±0.01	478.75±0.01
Copper (mg/l)	8.40±0.01	14.73±0.01	29.34±0.01
Zinc (mg/l)	9.76±0.01	11.13±0.01	10.17±0.00
Nickel (mg/l)	0.66±0.00	0.68±0.00	1.75±0.00
Cobalt (mg/l)	BDLd	0.78±0.00	8.80±0.01
Iron (mg/l)	13.95±0.01	18.09±0.01	31.95±0.01
Ammonia (mg/l)	3.76±0.01	3.50±0.00	10.50±0.01
Nitrate (mg/l)	BDLd	1.10±0.01	12.01±0.01
Nitrite (mg/l)	88.05±0.01	0.61±0.01	6.10±0.01

Notes: a, b, c Represent Mean±SD readings that are significant ($p < 0.05$) across the rows for each of the parameters and samples while BDLd equals below detection level, respectively.

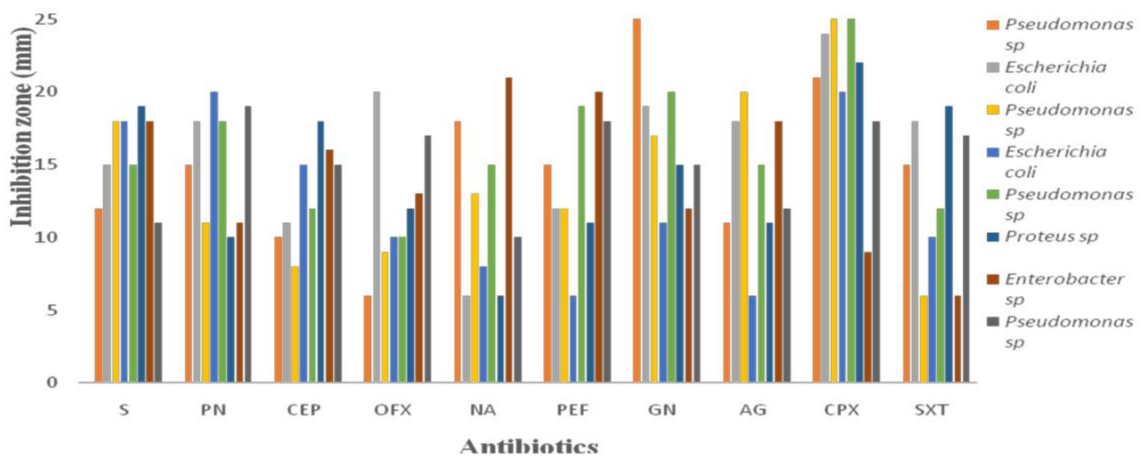

Fig. 2. The sensitivity of different gram negative organisms and antibiotics

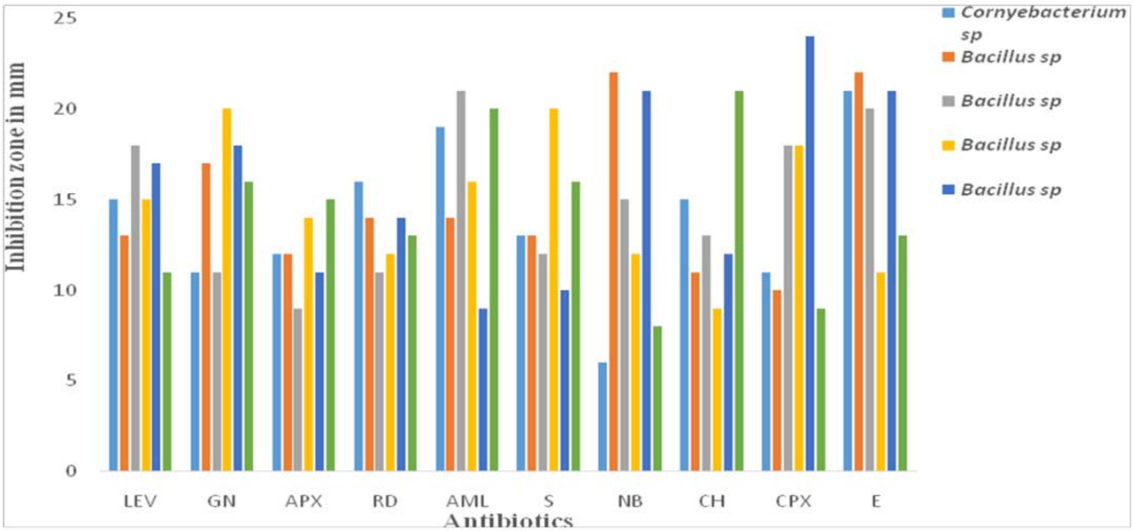

Fig. 3. The sensitivity of different gram positive organisms and antibiotics

Statistical Significance and Antibiotic Susceptibility Testing

Values marked with a, b and c represent significant mean ± standard deviation readings ($p < 0.05$) across rows for each parameter and sample. The symbol BDLd indicates values below the detection level.

Antibiotic Susceptibility Testing Results

Figures 2 and 3 present the results of the antibiotic susceptibility testing on Gram-negative and Gram-positive organisms isolated from different soil samples. The antibiotics used in this study were: Streptomycin; Ciprorex; Ampicillin; Tarivid; Nalidixic Acid; Perflacine; Gentamicin; Augmentin; Ciprofloxacin; Septrin; Levoflacin; Ampiclox; Rifampin; Amoxil; Norfloxacin and Erythromycin.

Pseudomonas sp., Proteus sp., and Escherichia coli isolated from all three sites were tested against these antibiotics.

Key Findings

1. Ciprofloxacin efficacy: Ciprofloxacin demonstrated consistent activity against the test isolates, with zones of inhibition of 25 mm and 24 mm against Pseudomonas and Escherichia coli, respectively.
2. Least effective antibiotics: Tarivid and Septrin showed the least zones of inhibition (6 mm) against Pseudomonas, E. coli, and Proteus species.
3. Pseudomonas sensitivity: Pseudomonas species isolated from impacted and effluent soil samples were most sensitive to the test antibiotics.
4. Gram-positive organism sensitivity: The highest zone of inhibition (24 mm) was observed for Bacillus sp. isolated from cassava effluent-impacted soil with Ciprofloxacin.

Antibiotic Efficacy Ranges

1. Levoflacin, Streptomycin, and Gentamicin: Showed zones of inhibition between 10 mm and 20 mm on all test isolates.
2. Remaining antibiotics: Had zones of inhibition between 6 mm and 24 mm.

Most Isolated Species

1. Bacillus sp.: Most frequently isolated from all three locations.
2. Corynebacterium and Enterobacter: Followed in terms of frequency of occurrence.

4. Discussion

Microbial Isolates and Counts

The study revealed the presence of various bacterial and fungal species in the soil samples. The bacterial isolates included Bacillus sp., Pseudomonas sp., Enterobacter sp., Corynebacterium sp., Proteus sp., and Escherichia coli. Eleven species of fungi were also isolated, including Aspergillus sp., Penicillium sp., Mucor sp., Candida sp., Saccharomyces sp., and Rhizopus sp. These findings are consistent with previous studies (Ehiagbonare et al., 2009; Igbinosa, Igiehon, 2015).

Microbial Counts and Comparison

The impacted soil had the highest microbial count of 7.8±0.19x105 cfu/g. Bacterial counts were higher than fungal counts in all samples. The total mean heterotrophic bacterial count ranged from 7.0±0.11x105 cfu/g to 7.8±0.19x105 cfu/g, while fungal counts ranged from 4.2±0.57x105 cfu/g to 3.9±0.42x105 cfu/g. These findings suggest that fungal counts were significantly lower than bacterial counts ($p < 0.05$), consistent with previous reports (Aiyegoro et al., 2007).

Effects of Cassava Mill Effluent on Soil Microorganisms

The study revealed that soil samples not affected by cassava mill effluent had a higher microbial population compared to those impacted by the effluent. This difference can be attributed to the harmful effects of cyanide acid content in cassava mill effluent on soil microorganisms. These findings support previous reports (Akubuenyi, 2022; Dike et al., 2022; Ukaegbu-Obi et al., 2018).

5. Conclusion and Recommendations

The study highlights the adverse environmental effects of cassava mill effluents on soil microorganisms and physicochemical properties. To mitigate these effects, it is recommended that:

1. Relocation of Cassava Mills: Cassava mills should be relocated to areas away from residential and agricultural areas.

2. Regulations and Enforcement: Governments should establish and enforce regulations for the disposal of cassava mill effluents.

3. Public Awareness and Education: Public awareness campaigns should be conducted to educate cassava farmers and processors about the dangers of cassava mill effluents and proper disposal methods.

4. Further Research: Additional research should be conducted on the effects of cassava mill effluents on surrounding water bodies and human health.

6. Acknowledgements

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