

## Optimisation control process of cyanide biodegradation from cassava mill effluent (CME) using indigenous microorganisms

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**SUMMARY.** The cyanide component of cassava mill effluent (CME) is highly toxic to man and its environment. This research was aimed at biodegrading cyanide from cassava mill effluent with various concentrations of cyanide, variable pH values, inoculum size and phenol. The heterotrophic bacterial and fungal counts were  $6.32 \times 10^8 \pm 0.01$ cfu/ml and  $2.87 \times 10^8 \pm 0.11$ cfu/ml, respectively. The microorganisms isolated and characterized were: *Staphylococcus aureus*, *Bacillus* sp., *Escherichia coli*, *Lactobacillus* sp., *Micrococcus* sp., *Klebsiella* sp., *Pseudomonas*, sp. *Salmonella* sp., *Corynebacterium* sp., *Aspergillus niger*, *Penicillium* sp., *Fusarium* sp. and *Saccharomyces* sp. The physicochemical parameters: pH (4.81), electrical conductivity (4860uS/cm), cyanide (17.13 mg/l), chemical oxygen demand (2041.20 mg/l), biological oxygen demand (1490.08mg/l), total dissolved solids (2478.60 mg/l), Chromium (19.44 mg/l), Manganese (136.08mg/l), Iron (340.20 mg/l) and Nickel (121.50 mg/l) were above the Federal Environmental Protection Agency standard for effluent discharge. *Pseudomonas*, *Bacillus* and *Aspergillus* species which had the highest turbidity values with enrichment medium supplemented with 1% cyanide were used for the batch biodegradation studies. *Pseudomonas* sp. had the best degradative ability of all isolates used even in the presence of phenol, an inhibitory substance. However, of all the varied substrate concentration used, 30ppm with other conditions remaining constant gave the highest degradative ability of 32.73% at a residence time of 8 days. Also, the highest biodegradation rate of 74.5% and 71.03% were achieved at pH, 6 and inoculum size of 6ml respectively at a residence time of 8days for 30ppm while other parameters were kept constant. The findings revealed that *Pseudomonas* sp., *Bacillus* sp. and *Aspergillus* sp. could be utilized for remediating cassava mill effluent contaminated environment containing cyanide.

**Keywords:** biodegradation, cyanide degrading microbes, environmental management, optimisation

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## Introduction

Cassava mill effluent (CME) from traditional grating during processing is a major cause of environmental degradation, contaminating agricultural farmlands, streams and affecting biodiversities (Enerijiofi *et al.*, 2017b; Chinyere *et al.*, 2018; Izah *et al.*, 2018). The toxicity of cassava mill effluent discharge is basically connected to its acidic pH and cyanide content. Cyanide compounds are fast acting poison that interferes mainly with cellular respiration process, resulting in a number of illnesses such as nervous instability by lipid peroxidation, poor vision, acute intoxication and sometimes death in humans (Ifeabunike *et al.*, 2017; Izah *et al.*, 2018). The toxicity of cyanide to existing cells is due to these three key mechanisms: resilient chelation to metals in metallo-enzymes; reaction with keto compounds to form cyanohydrin derivatives of enzyme substrates and reaction with Schiff-base intermediates during enzymatic reactions, to form stable nitrile derivatives (Ewa *et al.*, 2017). Long-term discharge of this effluent into the soil could result in a serious imbalance in the microbial population, which in turn could result in alteration of soil fertility towards a negative direction (Akpan *et al.*, 2017). Heavy metals that are discharged into the surroundings during processing with the coated metal machines persist indefinitely, accumulating in living tissues through food chain and causing severe diseases to man (Enerijiofi *et al.*, 2017b).

Cyanide is quite recalcitrant in the sense that it persists so long on any contaminated soil (Ewa *et al.*, 2017). However, various enzymes present in microorganisms aid in the conversion of cyanide to a source of carbon and nitrogen (Ibrahim *et al.*, 2015). The ability of bacteria, fungi, protozoa and other organisms to metabolize xenobiotic and hazardous compounds converting them to less toxic compound have been recognized as potentially effective means of disposal and management. (Eskander and Saleh, 2017). Microorganisms such as *Bacillus* sp., *Brevibacterium nitrophilous*, *Corynebacterium nitrophilous*, *Klebsiella oxytoca*, *Pseudomonas* sp. and *Rhodococcus* UKMP-5M have been reported to be proficient in cyanide degradation (Ibrahim *et al.*, 2015; Razanamahandry *et al.*, 2017; Moradkhani *et al.*, 2018).

Ebelle community of Esan Land is into subsistence farming with main focus on cassava production which are processed into Garri. The cassava mills located in these areas seldom have proper discharge channels for the effluent and upon accumulation are harmful to the environment. The aim of this research is to degrade cyanide from cassava mill effluent using *Pseudomonas* sp., *Bacillus* sp. and *Aspergillus* sp.

## Materials and methods

**Description of the study area.** Ebelle is one of the constituent kingdoms of Igueben Local Government Area of Edo State with geographical coordinates of 6° 30' 0" North, 6° 12' 0" East. It is naturally humid and characterized by a bimodal rainfall pattern particularly in July and September. It is an agrarian setting and the

residents are mainly farmers with cassava tubers production being their leading farm output (Enerijiofi *et al.*, 2017a).

**Collection of Sample.** Raw Cassava mill effluents samples were collected from a FADAMA (III) cassava processing mill site at Ebelle, Edo state. A sterile four (4) litre plastic container was used to collect the samples in triplicate. The samples were immediately conveyed to the laboratory in ice pack containers for physicochemical and microbiological analyses within two hours from sampling.

**Determination of physicochemical parameters and heavy metals concentrations.** The method of APHA, (2011) was used to determine the physicochemical parameters which included pH, electrical conductivity, total dissolved solids, total suspended solid, turbidity, alkalinity, chlorine, ammonium nitrogen, sulphate, nitrate, cyanide, phosphate, chemical oxygen demand, dissolved oxygen and biochemical oxygen demand. The cations ( $\text{Na}^+$ ,  $\text{K}^+$ ,  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$ ) were determined with a Jenway flame photometer, model PFP7, while the heavy metals concentrations were determined using an atomic absorption spectrophotometer, model PG 550 (Enerijiofi *et al.*, 2017b).

**Microbiological Analysis.** Aliquot 1ml of appropriate ten - fold serial dilution ( $10^{-3}$ ,  $10^{-6}$  and  $10^{-9}$ ) of the cassava mill effluent sample was inoculated into Nutrient agar plates containing fusicin and potato dextrose agar plates containing streptomycin in triplicate using pour plate method for bacterial and fungal enumeration, respectively. The inoculated plates were incubated at 37°C for 24 hrs in an incubator and at room temperature of 28°C, for 72hrs, for the enumeration of the total heterotrophic bacterial and fungal counts, respectively. The results were expressed in colony forming units per millilitre of the sample. (Cheesbrough, 2006; Enerijiofi *et al.*, 2017a).

**Isolation of cyanide degrading microbes.** Cyanide - degrading microorganisms were isolated from cassava mill effluent samples and purified by repeatedly transferring the cells to enrichment medium. For enrichment of microorganisms Nutrient broth was used and the sample cultivated in a 500ml Erlenmeyer flask containing 100ml Nutrient broth, with 1% cyanide concentration. To screen for cyanide degrading bacteria, 6ml of culture broth from  $1.5 \times 10^8$  cfu/ml was transferred into 500ml Erlenmeyer flask containing 100ml of buffer medium ( $\text{K}_2\text{HPO}_4$  4.35g, NaOH 4g and 10 ml of trace salts solution;  $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$  300 mg,  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$  180 mg,  $\text{CoCl}_2$  130 mg,  $\text{CaCl}_2$  40 mg,  $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$  40 mg and  $\text{MoO}_3$  20 mg in 1 litre deionized water) and 0.1% yeast extract containing 1% of cyanide was incubated at 30°C, 150rpm. This process was repeated three times by inoculating into fresh medium with 10% (v/v) of the previously grown culture. Cyanide-degrading microbes were isolated on a 2-day interval for 8 days by smearing on nutrient agar medium. Colonies that differed mainly in morphology were selected and pure isolates were obtained by continuous sub-culturing. The bacteria isolated were verified for their Gram staining reactions and biochemical tests (Mirizadeh *et al.*, 2014).

**Cyanide degradation experiment.** Bacterial and fungal isolates with distinct morphology were inoculated into nutrient broth and potato dextrose agar, respectively, for 24 hours. This was used for the purpose of isolates comparison study. Removal of cyanide was determined every 48 hours for 8 days. At regular intervals of 48 hours samples were drawn and tested for cyanide reduction. Non-inoculated medium served as control. The effect of substrate concentration, pH, inoculum size and phenol were determined (Arutchelvan *et al.*, and Mirizadeh *et al.*, 2014). Cell growth was examined by determining the optical density (O.D) of 1 ml culture at 660 nm through Spectrophotometry (model GENESYS 10 UV-Vis Scanning, Thermo Scientific) and expressed as OD<sub>660</sub> nm (Kandasamy *et al.*, 2015).

### Optimisation conditions

**Effect of Initial Substrate concentration on cyanide degradation.** Five different substrate concentrations, 30ppm, 60ppm, 90ppm, 120ppm and 150ppm, were used in the study, while all other given parameters remained constant. To each flask, 100ml of NFG medium was added and the pH was adjusted to 6 before sterilization. To each sterilized and cooled flask 6ml of  $1.5 \times 10^8$  cfu/ml inoculum was added.

**Effect of pH on cyanide degradation.** The effect of pH on degradation of cyanide was determined by maintaining pH ranges. One hundred milliliters (100ml) of NFG medium was prepared at a cyanide concentration of 30ppm in different flasks labelled with pH of 4, 5, 6, 7 and 8. All flasks were subjected to sterilization before adding 6ml of  $1.5 \times 10^8$  cfu/ml inoculum.

**Effect of Inoculum Size on cyanide degradation.** In order to determine the effect of inoculum variation on cyanide degradation, another set of well labelled five flasks with different inoculum loadings: 2ml, 3ml, 4ml, 5ml and 6ml, containing the same species and regulated to a final volume of 100ml of the same medium. The solution was maintained at pH 6 and the initial concentration of cyanide was 30ppm.

**Effect of Phenol on cyanide degradation.** The NFG medium with pH 6 was dispensed in each of the 5 flasks. Subsequently, after sterilization and cooling, the content was inoculated with 6ml of  $1.5 \times 10^8$  cfu/ml culture and added with 0.30%, 0.50%, 0.70%, 0.90% and 1.10% of phenol concentration. Cyanide concentration at 30ppm was maintained and experimentations were carried out.

**Degradation Efficiency.** The degradation efficiency (DE) of cyanide degrading bacterial and fungal isolates were calculated using the formula below:

$$DE(\%) = \frac{Ic - Rc}{Ic} \times 100$$

where DE = Degradation efficiency, Ic = Initial concentration of cyanide (mg/l) and Rc = Residual concentration of cyanide (mg/l).

## Results and discussion

The concentrations of the different physicochemical parameters of the raw cassava mill effluent analysed are shown in Table 1. The cyanide content was 17.13mg/l. The pH of 4.81 reported was highly acidic. Other major pollutants reported were heavy metals, except lead (0.29mg/l). The results of physicochemical quality of the cassava mill effluent revealed high level of pollution, particularly Cyanide (17.13 mg/l) which was far above the 0.2mg/l limit recommended by FEPA (1991). The pH reported was acidic (4.81) which could have resulted from the high cyanide content reported.

**Table 1.**

Physiochemical properties of cassava mill effluent

Physicochemical parameters	Units	Concentration	FEPA Effluent Limitation Guideline (1991) mg/l
pH		4.81±0.21	6-9
Electrical Conductivity	uS/cm	4860±0.02	1000
Chlorine	mg/l	34.02±0.11	600
Alkalinity	mg/l	27.65±0.01	NA
Total Suspended Solid	mg/l	29.65±0.11	30
Total Dissolved Solid	mg/l	2478.60±0.10	2000
Turbidity	NTU	166.74±0.11	300
Chemical oxygen demand	mg/l	2041.20±0.01	40
Dissolved oxygen	mg/l	0.63±0.11	40
Biochemical oxygen demand	mg/l	1490.08±0.14	10
Cyanide	mg/l	17.13±0.01	0.2
Sulphate	mg/l	257.58±0.10	50
Nitrate	mg/l	140.94±0.11	1.0
Phosphate	mg/l	102.06±0.00	5.0
Ammonium nitrogen	mg/l	0.97±0.10	NA
Calcium	mg/l	156.98±0.23	100
Magnesium	mg/l	58.32±0.18	100
Sodium	mg/l	680.40±0.01	200
Potassium	mg/l	1506.60±0.01	NA
Zinc	mg/l	58.32±0.32	1.0
Copper	mg/l	72.90±0.11	1.5
Chromium	mg/l	19.44±0.20	0.5
Lead	mg/l	0.29±0.26	0.5
Manganese	mg/l	136.08±0.22	0.5
Iron	mg/l	340.20±0.01	20
Nickel	mg/l	121.50±0.20	1.0

**Legend:** NA - Not Available; Values are in Mean ± standard error of triplicate samples

Other physicochemical parameters, particularly heavy metals recorded apart from Lead (0.29 mg/l), were higher than the FEPA (1991) limit for effluent discharge. These high heavy metals concentrations could be traced to the anthropogenic inputs, such as corrosion of the metal parts of equipment used in harvesting and milling into the environment, as reported by earlier authors (Ebukiba, 2010).

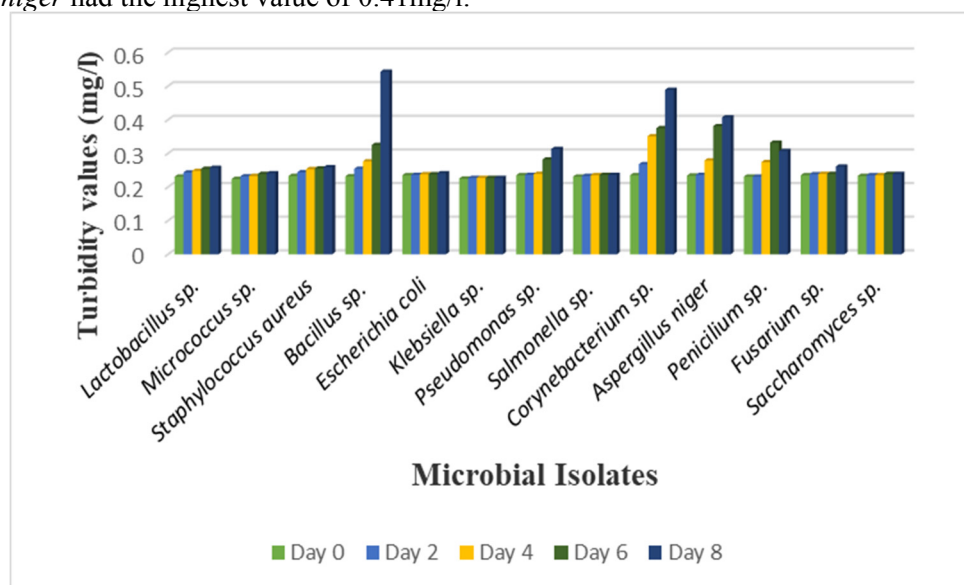
The results in Table 2 shows the bacterial and fungal counts. The total heterotrophic bacterial count ( $6.32 \times 10^8 \pm 0.01$  cfu/ml) was higher than the total heterotrophic fungal count of ( $2.87 \times 10^8 \pm 0.11$  cfu/ml). The isolation of high numbers of cassava mill effluent utilising bacterial and fungal isolates from cassava mill effluent was an indication these organisms are active cassava mill effluent degraders in the environment (Enerijiofi *et al.*, 2017a).

**Table 2.**

Mean bacterial and fungal counts ( $\times 10^8$ cfu/ml)		
	THBC (cfu/ml)	THFC (cfu/ml)
Cassava mill effluent	6.32±0.01	2.87±0.11

**Legend:** THBC – Total Heterotrophic Bacterial Count; THFC – Total Heterotrophic Fungal Count; Values are in Mean  $\pm$  standard error of triplicate samples

Figure 1 shows the bacterial and fungal taxa that could utilise cyanide as substrate for growth. However, *Bacillus* and *Pseudomonas* species had turbidity values of 0.543mg/l and 0.31mg/l among the bacterial isolates while *Aspergillus niger* had the highest value of 0.41mg/l.



**Figure 1.** Isolation of cyanide degrading microbes (bacteria and fungi) with mineral salt medium containing 1% cyanide

The microorganisms surviving in such environment are those that have developed enzymatic and physiological responses that allow them to use cyanide as substrate for growth and subsequent proliferation. This agreed with the earlier findings (Izah *et al.*, 2018; Orji and Ayogu, 2018). A similar observation was made by Enerijiofi *et al.* (2017a) on biodegradation potentials of cassava mill effluents using indigenous microorganisms.

It was established that *Pseudomonas* sp. performed best because it was able to reduce the substrate concentration of 30ppm and 60ppm by 32.73% and 17.62%, respectively, at a residence time of 8 days while *Bacillus* sp. gave 16.93% and 15.29% reduction for 120ppm and 150ppm, respectively, at residence time of 8 days. In summary, Table 3 revealed that the smaller the concentration, the better the degradation efficiency.

**Table 3.**

Effect of substrate concentration of cyanide; Inoculum size = 6ml of  $1.5 \times 10^8$ cfu/ml, pH =6

<b>Day 0</b>	<b>30ppm</b>	<b>60ppm</b>	<b>90ppm</b>	<b>120ppm</b>	<b>150ppm</b>
<i>Bacillus</i> sp.	29.98	60.14	90.07	120.03	149.98
<i>Pseudomonas</i> sp.	29.96	60.07	89.96	120.05	149.95
<i>Aspergillus niger</i>	29.99	60.09	89.99	120.02	150.04
Control	29.97	60.02	90.03	120.04	149.99
<b>Day 2</b>	<b>30ppm</b>	<b>60ppm</b>	<b>90ppm</b>	<b>120ppm</b>	<b>150ppm</b>
<i>Bacillus</i> sp.	27.65	58.76	90.01	120.01	149.98
<i>Pseudomonas</i> sp.	26.78	58.32	89.92	120	149.95
<i>Aspergillus niger</i>	28.94	59.89	89.98	120.02	150.04
Control	30.01	60.01	90.03	120.04	149.99
<b>Day 4</b>	<b>30ppm</b>	<b>60ppm</b>	<b>90ppm</b>	<b>120ppm</b>	<b>150ppm</b>
<i>Bacillus</i> sp.	25.58	58.46	89.11	112.81	146.98
<i>Pseudomonas</i> sp.	22.13	57.79	86.32	117.60	148.45
<i>Aspergillus niger</i>	26.98	59.56	86.38	114.02	147.04
Control	29.97	59.03	90.01	120.02	150.01
<b>Day 6</b>	<b>30ppm</b>	<b>60ppm</b>	<b>90ppm</b>	<b>120ppm</b>	<b>150ppm</b>
<i>Bacillus</i> sp.	24.05	57.29	87.33	106.04	139.63
<i>Pseudomonas</i> sp.	20.80	54.32	83.73	115.25	146.97
<i>Aspergillus niger</i>	26.17	57.18	85.52	107.18	142.63
Control	29.97	59.03	90.01	120.02	150.01
<b>Day 8</b>	<b>30ppm</b>	<b>60ppm</b>	<b>90ppm</b>	<b>120ppm</b>	<b>150ppm</b>
<i>Bacillus</i> sp.	22.60	55.57	86.45	99.68	127.06
<i>Pseudomonas</i> sp.	20.18	49.43	80.38	112.94	145.50
<i>Aspergillus niger</i>	24.60	56.03	82.10	101.82	138.35
Control	29.98	59.01	89.99	120.01	150.02

The result revealed that the smaller the substrate concentration, the better the degradation efficiency. *Pseudomonas* sp. degraded cyanide best to the tune of 32.73% at a residence period of 8 days. From this, it can be concluded that increased

substrate will inhibit the degradation process as reported by earlier researchers (Kandasamy *et al.*, 2015; Mekuto *et al.*, 2013; Mirizadeh *et al.*, 2014).

Cyanide serves as a nutrient for bacterial growth, acting as nitrogen source. Some bacteria make use of cyanide compounds both as carbon and nitrogen source. Therefore, external supply of carbon is no longer necessary for these bacteria. In the presence of cyanide other bacteria required glucose as carbon source for their survival (Bouari, 2012).

Table 4 revealed the ability of *Pseudomonas* sp., *Bacillus* sp. and *Aspergillus niger* to degrade cyanide at varying pH, while other parameters remained constant. pH 6 gave the highest reduction in cyanide concentration by *Pseudomonas* sp. at 63.17%, 73.70%, 73.97% and 74.50% at day 2, 4, 6 and 8 respectively. The pH concentration also plays a major role in the biological activity of the degradation process. *Pseudomonas* sp. gave the maximum of 74.50% degradation at pH 6 at a residence time of 8 days. From this study, it can be concluded that the bacterial and fungal isolates performed best in the slightly acidic condition of pH 6.

**Table 4.**

Effect of pH on cyanide degradation; Cyanide concentration = 30ppm,  
Cell suspension=6 ml of  $1.5 \times 10^8$ cfu/ml

<b>Day 0</b>	<b>pH 4</b>	<b>pH 5</b>	<b>pH 6</b>	<b>pH 7</b>	<b>pH 8</b>
<i>Bacillus</i> sp.	15.34	15.42	15.33	15.37	15.44
<i>Pseudomonas</i> sp.	15.28	15.36	15.41	15.38	15.42
<i>Aspergillus niger</i>	15.43	15.31	15.35	15.42	15.38
Control	15.33	15.41	15.42	15.37	15.41
<b>Day 2</b>					
<i>Bacillus</i> sp.	12.54	11.89	11.55	13.33	15.11
<i>Pseudomonas</i> sp.	14.11	11.43	11.05	13.97	15.25
<i>Aspergillus niger</i>	14.68	13.75	12.76	14.78	15.31
Control	15.14	15.38	15.19	15.36	15.4
<b>Day 4</b>					
<i>Bacillus</i> sp.	10.89	10.35	8.63	12.78	14.98
<i>Pseudomonas</i> sp.	13.77	10.85	7.89	12.43	15.06
<i>Aspergillus niger</i>	14.11	12.98	12.04	14.31	15.34
Control	15.05	15.33	15.14	15.31	15.39
<b>Day 6</b>					
<i>Bacillus</i> sp.	10.67	9.83	8.46	12.27	14.68
<i>Pseudomonas</i> sp.	13.63	10.63	7.81	11.81	14.76
<i>Aspergillus niger</i>	13.83	12.59	11.68	14.17	15.03
Control	14.90	15.02	14.84	15.00	15.08
<b>Day 8</b>					
<i>Bacillus</i> sp.	10.46	9.64	8.29	12.02	14.39
<i>Pseudomonas</i> sp.	13.36	10.42	7.65	11.57	14.46
<i>Aspergillus niger</i>	13.55	12.34	11.45	13.88	14.73
Control	14.89	15.01	14.83	14.89	15.07



However, Kwon *et al.* (2002) and Ibrahim *et al.* (2015) reported that microbial isolates did perform best at acidic, slightly acidic and neutral conditions.

The findings in Table 5 show that inoculum size of 6ml and 5ml, *Pseudomonas* sp. gave a degradation efficiency of 71.03% and 68.73% at day 8. It was observed that with increased incubation time, degradation efficiency increases. It was evident that the degradation percentage increased with increased biomass concentration. At inoculum size of 6ml, *Pseudomonas* sp. gave the optimum degradation capacity of 71.03% at a residence time of 8 days.

**Table 5.**

Effect of Inoculum size on cyanide degradation; pH = 6, Cyanide Concentration = 30ppm

<b>Day 0</b>	<b>2ml</b>	<b>3ml</b>	<b>4ml</b>	<b>5ml</b>	<b>6ml</b>
<i>Bacillus</i> sp.	15.34	15.42	15.33	15.37	15.44
<i>Pseudomonas</i> sp.	15.28	15.36	15.41	15.38	15.42
<i>Aspergillus niger</i>	15.43	15.31	15.35	15.42	15.38
Control	15.33	15.41	15.42	15.37	15.41
<b>Day 2</b>					
<i>Bacillus</i> sp.	13.76	13.24	12.88	11.69	11.05
<i>Pseudomonas</i> sp.	12.93	12.55	10.48	9.97	9.36
<i>Aspergillus niger</i>	14.97	14.16	13.97	13.26	12.98
Control	15.33	15.39	15.4	15.37	15.41
<b>Day 4</b>					
<i>Bacillus</i> sp.	12.93	12.84	12.75	10.99	10.06
<i>Pseudomonas</i> sp.	12.54	11.42	10.06	9.77	9.27
<i>Aspergillus niger</i>	14.07	13.88	13.41	12.60	12.59
Control	15.31	15.39	15.4	15.35	15.39
<b>Day 6</b>					
<i>Bacillus</i> sp.	12.16	12.59	12.11	10.33	9.55
<i>Pseudomonas</i> sp.	11.79	10.74	9.66	9.58	9.17
<i>Aspergillus niger</i>	13.65	13.32	13.28	11.84	12.21
Control	15.31	15.34	15.38	15.34	15.37
<b>Day 8</b>					
<i>Bacillus</i> sp.	11.43	12.21	11.99	9.71	9.08
<i>Pseudomonas</i> sp.	11.44	9.77	9.27	9.38	8.69
<i>Aspergillus niger</i>	12.83	13.06	12.75	11.25	11.85
Control	15.31	15.39	15.38	15.33	15.35

This result also revealed that with increased inoculum size and residence time, degradation efficiency also increased, which agreed with previous report (Arutchelvan *et al.*, 2005).

Table 6 records the cyanide degradation efficiency of the bacterial and fungal isolates at varied concentrations of phenol with other parameters kept constant. *Pseudomonas* sp. gave the best result, of 12.04, at 0.30% phenol concentration, *Bacillus* sp. at 0.50% and 0.70% gave 13.68 and 14.46, respectively, while at 0.90% and 1.10% *Pseudomonas* sp. gave a reduced value of 14.18 and 14.43, respectively.

**Table 6.**

Effect of inhibitory substance (phenol); Inoculum size = 6ml of  
 $1.5 \times 10^8$ cfu/ml, pH =6, Cyanide Concentration = 30ppm

<b>Day 0</b>	<b>0.30%</b>	<b>0.50%</b>	<b>0.70%</b>	<b>0.90%</b>	<b>1.10%</b>
<i>Bacillus</i> sp.	15.34	15.42	15.33	15.37	15.44
<i>Pseudomonas</i> sp.	15.28	15.36	15.41	15.38	15.42
<i>Aspergillus niger</i>	15.43	15.31	15.35	15.42	15.38
Control	15.33	15.41	15.42	15.37	15.41
<b>Day 2</b>					
<i>Bacillus</i> sp.	13.88	15.01	15.27	15.31	15.43
<i>Pseudomonas</i> sp.	13.25	14.95	15.25	15.29	15.4
<i>Aspergillus niger</i>	14.96	15.13	15.33	15.41	15.38
Control	15.33	15.41	15.42	15.37	15.41
<b>Day 4</b>					
<i>Bacillus</i> sp.	12.78	14.69	15.06	15.28	15.41
<i>Pseudomonas</i> sp.	12.41	14.25	15.19	15.23	15.03
<i>Aspergillus niger</i>	13.99	15.21	15.28	15.39	15.36
Control	15.32	15.39	15.41	15.36	15.41
<b>Day 6</b>					
<i>Bacillus</i> sp.	12.52	13.96	14.76	14.67	15.10
<i>Pseudomonas</i> sp.	12.29	13.97	15.04	14.47	14.73
<i>Aspergillus niger</i>	13.71	14.75	14.82	15.24	15.05
Control	15.32	15.37	15.42	15.35	15.41
<b>Day 8</b>					
<i>Bacillus</i> sp.	12.27	13.69	14.46	14.38	14.80
<i>Pseudomonas</i> sp.	12.04	13.68	14.74	14.18	14.43
<i>Aspergillus niger</i>	13.44	14.46	14.53	14.93	14.75

It was recorded that, with increase of phenol concentrations, the degradation of cyanide reduced due to the inhibitory action of phenol and the residence time increased with interference of phenol. *Pseudomonas* sp., *Bacillus* sp. and *Aspergillus niger* were more active in degrading the phenol rather degrading the cyanide, which corroborated with earlier findings (Neetu and Chandrajit, 2016; Singh *et al.*, 2018).

### Conclusions and recommendations

*Pseudomonas* sp., *Bacillus* sp. and *Aspergillus* sp. were the most dominant microbial isolates which shown the highest ability of improving cassava mill effluent by reducing the cyanide content; under precise cultural conditions. However, *Pseudomonas* sp. had the best degradative ability. This study has unveiled the potentials of biodegradation of cyanide from cassava mill effluent.

It is recommended that further leaps be taken in a bid to exploring newer, more effective, less costly and better satisfactory methods of cyanide management from cassava mill effluent prior to eventual discharge into the environment.

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