



Original Article

BACTERIOLOGICAL AND PHYSICOCHEMICAL ANALYSIS OF WATER FROM MINING PONDS IN BASSA AND JOS SOUTH LOCAL GOVERNMENT AREAS OF PLATEAU STATE

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Submitted: March, 2017; Accepted: April 2017; Published: June, 2017

ABSTRACT

The bacteriological and physicochemical quality of water from mining ponds was carried out in this study. A total of 180 water samples from ten mining ponds in Bassa and Jos South Local Government Areas of Plateau State were collected in wet and dry seasons. The water samples were evaluated for bacteriological quality and physicochemical properties using standard methods. The results showed that the mean total viable bacterial count (TVBC) ranged from 1.3×10^6 - 1.65×10^6 cfu/ml. The highest mean TVBC (1.65×10^6 cfu/ml) was observed in water from Gura-Topp, with least count (1.30×10^6 cfu/ml) observed in Topp tin mining pond. The total coliform count (TCC) ranged from 54 - 493 mpn/100 ml. Variations in TVBC and TCC within ponds and seasons were observed. Predominately, eight bacteria: *Escherichia coli*, *Salmonella Typhi*, *Vibrio cholerae*, *Proteus vulgaris*, *Yersinia enterocolitica*, *Shigella dysenteriae*, *Bacillus subtilis*, and *Salmonella enteritidis*, were isolated from the water samples. Physicochemical properties showed that pH ranged from 7.76 - 7.86, temperature 22.35 - 23.37°C, dissolved oxygen (DO) 5.11 - 8.17 mg/L, total dissolved solids (TDS) 180.6 - 196 mg/L, total suspended solids (TSS) 1.50 - 30.17 mg/L, chemical oxygen demand (COD) 4.63 - 18.8 mg/L and biological oxygen demand (BOD) 6.38 - 8.53 mg/L. Heavy metals detected included zinc, lead, cadmium, cobalt, chromium and arsenic. Lead, arsenic, cadmium and chromium were above the recommended standards. Consistently, physicochemical values obtained were found to significantly ($p < 0.05$) vary among ponds and seasons. This study showed that the tin mining ponds are bacteriologically not safe for drinking purposes and contain high concentration of some heavy metals.

Key words: bacteriological quality, physicochemical quality, heavy metals, tin mining ponds

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INTRODUCTION

Mining is a major source of heavy metals in water has received much attention due to its great health implications (Edema, 2001). Its impact on the environment is so adverse in most of the localities especially in Bassa and Jos South, Local Government Areas of Plateau State, Nigeria. The mining of minerals has been reported to lead to indiscriminate disposal of some toxic substances, such as heavy metals on land and in the aquatic system (Gyang and Ashano, 2010; Ezeaku, 2012). In addition, the inhabitants of the region are living with the mining scar characterized by mounds, ponds, tin-tailings and other hard metals, which make the area vulnerable to erosion and other environmental factors (Wapwera, 2014).

Heavy metals accumulation of in freshwater ecosystem has been a major concern as they may gain entrance into aquatic environment *via* natural or anthropogenic means, which may include industrial effluent, domestic wastewater, mining and agricultural wastes discharge (Vautukuru, 2005). The harmful effect of mining activities can be determined by assessing the concentration and distribution of heavy metals as well as other contaminants in water (Gyang and Ashano, 2010).

Many diseases in developing countries are caused by drinking contaminated water (Fawell and Nieuwenhuijsen, 2013). This is because both quality and quantity of water are affected by an increase in anthropogenic activities and pollution (chemical or physical) that causes changes in the quality of the

recurring water body (Kagoro *et al.*, 2011). In most communities in Plateau State, Nigeria, the supply of pipe borne water is inadequate. Communities in tin mining settlement often rely on the use water from mining ponds for domestic use as is particularly prevalent in those rural areas. The major challenge has been how to make these sources safe for human consumption. Ideally, drinking water should not contain any microorganism known to be pathogenic or indicative of faecal pollution (WHO, 2004). Therefore the aim of this study was to assess the bacteriological and physicochemical properties of water from tin mining ponds in Bassa and Jos South, Local Government Areas (LGA) of Plateau State, Nigeria.

MATERIALS AND METHODS

Collection of water samples

A total of 180 water samples were collected from ten mining ponds from two Local Government Areas of Jos (Jos South and Bassa) (Figure 1). Three water samples were obtained from each pond every month from August to October 2015, (rainy season) and November to January 2016 (dry season) which add up to six months. Water samples were collected in one liter sterile bottle in duplicates by 'grab' method. This was done by submerging the bottles to a depth of 0.5 meter and immediately placed in a light proof insulated box containing ice-packs with water to ensure rapid cooling. After collection, the water samples were immediately transported to the laboratory where they were analyzed within 6 hours.

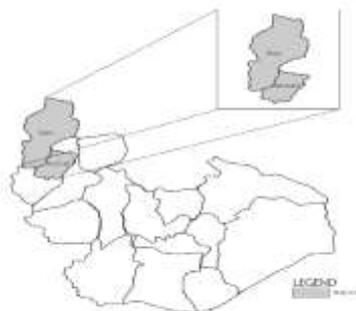


Figure 1. Map of Plateau State indicating Jos South and Bassa Local Government Area. University of Jos, Nigeria (2016)

Bacteriological Analysis of Water Samples

The water samples were serially diluted and 1 ml of the diluted water samples (that is 10^{-3} , 10^{-4} and 10^{-5} dilutions) were inoculated in the respective media. Enumeration and isolation of bacteria Enumeration of total viable bacteria was done on sterile nutrient agar plate using pour plate method. The Petri plates were incubated at 37°C for 24 hours. The colonies, which developed after the incubation were counted and recorded as colony forming units per milliliter (cfu/ml) of the water sample (Chessbrough, 2006). The Most Probable Number (MPN) was adopted for the determination of total coliform bacteria. This was carried out in three stages which were the presumptive test, confirmed test and completed test (WHO, 2004). Isolation of *Salmonella* and *Shigella* species were carried out by inoculating water samples into *Salmonella-Shigella* agar. *Vibrio cholera* was also isolated by inoculating water samples into Thiosulfate Citrate Bile-salts Sucrose (TCBS) (Cheesbrough, 2006).

Detection of Faecal Streptococci

The presence of faecal Streptococci was determined by inoculating water samples on sterile Azide Dextrose Broth (ADB) and incubated at 37°C for 48 hours. The broth was observed for

growth (turbidity) which is positive for faecal streptococci. For confirmatory test, a loopful of the broth after 48 hours incubation was streaked on Bile Esculin Azide Agar (BEAA) plate and further incubated at 37°C for 24 hours. After incubation, BEAA plate was observed for brownish-black colonies with brown halos to confirm the presence of faecal Streptococci (WHO, 2011).

Physicochemical Analysis of Water Samples

The water samples were analysed for the following physicochemical properties: pH, temperature, phosphate, fluoride, chloride, chemical oxygen demand (COD), biological oxygen demand (BOD), total dissolved solids (TDS), turbidity, total hardness, dissolved oxygen (DO) and total suspended solids (TSS) and heavy metals (Cr, Pb, Cd, Zn, As and Co) using the methods of American Public Health Association, APHA (1998).

Characterization and Identification of Bacterial Isolates

The bacteria isolated were subjected to colonial, microscopic and biochemical characterization using the conventional bacteriological methods described by Cheesbrough (2006). The bacterial isolates were identified by comparing their characteristics with those of known taxa (Holt *et al.*, 1994).

Data analysis

Data generated from bacteriological and physicochemical analyses of water samples were expressed as mean \pm standard deviation. To determine variation between mean of samples obtained, data were subjected to one way Analyses of variance (ANOVA) using Microsoft excel package.

RESULTS

Bacteriological quality of water samples

The contamination level of the water sample showed that high TVBC occurred in wet seasons; September in Zawan (2.09×10^6 cfu/ml), Gura-Topp (2.07×10^6 cfu/ml) and Rufam-Gwamna (1.92×10^6 cfu/ml) and October in Gura-Topp (1.94×10^6 cfu/ml) and Zawan (1.74×10^6 cfu/ml) (Table 1). The results also showed that low TVBC counts was observed in dry seasons (for example Du-Kwang had a TVBC count of 8.50×10^5 cfu/ml in January) (Table 1).

Total Coliform Counts (TCC)

The TCC of the water samples (Table 2) showed that highest value (1600 MPN/100ml) occurred in wet seasons; September and October in both Malempe and Zawan mining ponds. The lowest TCC (23 MPN/100ml) were recorded in dry seasons; November and December in Mista-Ali, Du-Kwang and Topp tin mining ponds.

Occurrence of bacteria in mining ponds

A total of 8 bacterial species, *Escherichia coli*, *Salmonella typhi*, *Salmonella enteritidis*, *Vibrio cholerae*, *Proteus vulgaris*, *Yersinia enterocolitica*, *Bacillus subtilis* and *Shigella dysenteriae* were isolated from tin mining ponds.

Physicochemical Properties of Water

The result obtained for the physicochemical parameters studies is presented in Table 3. The values

obtained showed that the pH, temperature, total hardness, TSS, BOD, COD fall within the recommended values of NSDWQ (2007) and WHO (2011) while the values obtained for TDS, DO, turbidity and Phosphate did not fall within their standards. The results also showed that the zinc, lead, cadmium, cobalt, chromium and arsenic were detected in the water. The values obtained for lead, cadmium, chromium and arsenic were above the recommended limits especially in the months of November, December and January.

The values obtained in Table 4 show that turbidity values in all the ponds are greater than the required limits (NSDWQ, 2007; WHO, 2011). TDS values of 996 mg/ml and 666.17 were obtained in Mista-Ali and Doruwa respectively. These values are above the required limits (NSDWQ, 2007; WHO, 2011). The cadmium value of 0.06 mg/ml in Buji-yelwa is also above the standards. Arsenic was found in Mista-Ali with 0.07 mg/ml above the required limit (NSDWQ, 2007; WHO, 2011)

Table 1: Total Viable Bacterial Counts of Water from Mining Ponds in Bassa and Jos South Local Government Area

Pond	Total Viable Bacterial Counts mean x10 ⁶ (cfu/ml)						Total (mean)
	August	September	October	November	December	January	
Mista Ali	1.57±1.17	1.75±1.06	0.85±0.35	1.20±0.42	1.53±0.64	0.94±0.30	1.31±0.65 ^a
Malempe	1.75±1.05	1.85±1.34	1.45±0.92	1.53±0.67	1.35±0.89	0.90±0.28	1.47±0.75 ^a
RufamGwamna	1.62±0.40	1.92±1.24	1.25±0.78	1.65±0.64	1.51±0.71	1.18±0.23	1.52±0.60 ^a
Buji-Yelwa	1.46±0.45	1.70±1.13	1.37±0.69	1.77±0.96	1.59±0.81	1.17±0.40	1.51±0.61 ^a
Kwang	1.46±0.73	1.88±1.25	0.92±0.45	1.50±0.72	1.40±0.71	0.97±0.33	1.35±0.65 ^a
Du-Kwang	1.78±1.02	1.86±1.01	1.30±0.57	1.71±0.71	1.61±0.76	0.85±0.34	1.52±0.68 ^a
Topp	1.54±0.79	1.75±0.93	1.04±0.22	1.15±0.35	1.33±0.39	0.99±0.19	1.30±0.50 ^a
Gura- Topp	1.81±0.98	2.07±1.03	1.94±1.17	1.71±0.98	1.15±0.50	1.20±0.42	1.65±0.75 ^a
Doruwa	1.61±0.83	1.53±0.67	1.44±0.79	1.43±0.88	1.51±0.74	0.96±0.33	1.41±0.58 ^a
Zawan	1.86±0.91	2.09±1.10	1.74±0.93	1.33±0.94	1.33±0.39	0.86±0.33	1.53±0.73 ^a

Values are mean ± standard error of mean of duplicate determinations. Means having the same superscripts are not significantly different ($p > 0.05$)

Table 2: Total Coliform Counts in Mining Ponds in Bassa and Jos South Local Government Area

Name of Pond	Total Coliform Counts (MPN/100ml)/ Month						Mean Count
	Wet season			Dry season			
	August	September	October	November	December	January	
Mista Ali	90	90	30	23	23	70	54 ^a
Malempe	170	1600	110	90	40	23	339 ^c
Rufam Gwamna	140	280	140	110	90	30	132 ^b
BujiYelwa	280	350	90	70	40	90	153 ^b
Kwang	50	280	110	80	50	170	123 ^b
Du-Kwang	40	90	90	40	23	90	62 ^a
Topp	50	90	90	80	23	140	79 ^a
Gura Topp	280	110	170	110	90	50	135 ^b
Doruwa	350	170	170	90	40	70	148 ^b
Zawan	350	350	1600	350	170	140	493 ^c
Mean	180	341	260	104.3	58.9	87.3	171

Values are mean \pm standard error of mean of duplicate determinations. Means with different superscripts are significantly different ($p < 0.05$) from each other while means with the same superscripts are not significantly different ($p > 0.05$).

Table 3. Monthly Physicochemical properties of Water from Tin Mining Ponds in Bassa and Jos South Local Government Areas of Plateau State

Parameters	Months						NSDWQ standards 2007	Standard values (WHO, 2011)
	August	September	October	November	December	January		
Temperature (°C)	21.83 ^a	23.39 ^a	23.39 ^b	23.37 ^b	21.50 ^a	22.52 ^b	Ambient	Variable
pH	8.17 ^a	7.75 ^b	7.63 ^a	7.49 ^b	7.64 ^a	7.76 ^b	6.5-8.5	6.5- 9.5
Turbidity (NTU)	46.90 ^b	110.80 ^b	73.00 ^a	59.00 ^b	1.50 ^b	2.70 ^b	5	<5
TSS (mg/L)	15.80 ^b	26.40 ^b	9.80 ^b	11.60 ^a	1.00 ^b	5.60 ^b	-	100
TDS (mg/L)	325.80 ^a	328.80 ^a	267.40 ^b	244.10 ^b	655.10 ^b	475.40 ^a	500	<300
DO (mg/L)	5.26 ^a	8.55 ^a	9.58 ^a	8.31 ^a	4.87 ^a	5.18 ^a	-	5.00
Total Hardness (mg/L)	22.27 ^b	35.02 ^b	40.16 ^a	17.38 ^a	12.93 ^b	8.69 ^a	150	100- 500
COD (mg/L)	13.21 ^a	5.52 ^a	5.33 ^b	5.33 ^b	13.41 ^b	12.93 ^a	-	350
BOD (mg/L)	3.75 ^b	13.3 ^b	8.40 ^b	11.91 ^b	5.33 ^a	3.27 ^b	-	30
Phosphate (mg/L)	0.29 ^b	0.29 ^b	0.22 ^a	0.16 ^a	0.99 ^b	1.12 ^a	-	1.00
Chloride (mg/L)	9.97 ^a	17.42 ^a	68.10 ^a	22.91 ^a	9.75 ^a	10.46 ^a	250	25- 60
Fluoride (mg/L)	<0.1 ^a	1.5	<1.5					
Zinc (mg/L)	0.031 ^a	0.186 ^a	0.154 ^a	0.176 ^a	0.063 ^a	0.065 ^b	3.0	<3.0
Lead (mg/L)	0.026 ^b	0.002 ^a	0.005 ^a	0.005 ^a	0.199 ^a	0.196 ^a	0.01	0.01
Cadmium (mg/L)	0.025 ^a	0.021 ^a	0.040 ^b	0.039 ^b	0.017 ^a	0.018 ^a	0.003	0.05
Cobalt (mg/L)	0.060 ^b	0.012 ^b	0.016 ^a	0.031 ^a	0.004 ^a	0.006 ^b	-	0.05
Chromium (mg/L)	0.013 ^a	0.051 ^a	0.003 ^b	0.001 ^b	0.016 ^a	0.017 ^b	0.05	0.05
Arsenic (mg/L)	-	-	-	-	0.07 ^a	-	0.01	0.01

NTU: Nephelometric turbidity unit, TSS: total suspended solids, TDS: total dissolved solids, DO: dissolved oxygen, mg/L: milligram per liter, -: not present, NSDWQ: Nigerian Standard for Drinking Water Quality. Values are mean of duplicate determinations. Means with different superscripts are significantly different ($p < 0.05$) from each other while means with the same superscripts are not significantly different ($p > 0.05$)

Table 4. Physicochemical properties of Water from Tin Mining Ponds in Bassa and Jos South

Pond location	Mean Value of Physicochemical Parameters																
	pH	Tem	Turb	TSS	TDS	DO	Cl	Hard	COD	BOD	Phos	Zn	Pb	Cd	Co	Cr	Ar
Mista-Ali	7.68	22.35	*43.83	10.33	*996.00	7.12	33.32	22.27	18.8	8.08	0.76	0.16	*0.24	0.02	0.05	0.03	*0.07
Malempe	7.70	22.40	*34.33	30.17	203.67	6.57	32.72	35.02	9.08	8.53	0.43	0.17	*0.02	0.03	0.05	0.03	-
Rufam Gwamna	7.81	22.67	*36.00	23.00	347.33	7.57	32.57	40.16	8.04	7.63	0.58	0.11	*0.15	0.05	0.06	0.03	-
Buji-yelwa	7.68	22.38	*32.83	15.67	180.67	8.17	21.69	17.38	7.66	7.17	0.18	0.06	0.01	*0.06	0.01	0.01	-
Kwang	7.74	22.65	*80.00	5.33	336.33	6.75	14.53	12.93	9.95	7.43	0.57	0.07	*0.07	0.02	0.01	0.01	-
Du-Kwang	7.73	22.58	*74.17	7.00	301.50	7.00	15.17	8.69	8.74	7.48	0.41	0.07	0.01	0.02	0.02	0.01	-
Topp	7.86	22.55	*32.00	1.50	199.83	8.01	21.99	9.29	4.63	7.75	0.15	0.07	0.01	0.02	0.01	0.02	-
Gura-Topp	7.73	23.70	*17.00	3.33	220.83	7.19	19.59	15.17	6.4	7.67	0.20	0.07	*0.02	0.01	0.01	0.01	-
Doruwa	7.64	22.65	*66.33	2.83	*666.17	5.11	16.50	12.08	17.3	11.62	1.18	0.21	*0.18	0.03	0.02	0.01	-
Zawan	7.86	22.37	*73.33	2.83	375.33	6.10	22.94	25.37	7.86	6.28	0.72	0.14	*0.01	0.02	0.02	0.01	-
Mean values	7.74	22.63	48.98	10.20	382.77	6.96	23.10	19.84	9.85	7.96	0.52	0.11	0.07	0.02	0.03	0.02	-

Tem: Temperature Turbid: Turbidity, TSS: Total Suspended Solid, TDS: Total Dissolved Solid, DO: Dissolved Oxygen, Cl: Chloride, Hard: hardness, COD: Chemical Oxygen Demand, BOD: Biological Oxygen Demand, phos: phosphate, Zn: Zinc, Pb: Lead, Cd: Cadmium, Co: Cobalt, Cr: Chromium, Ar: Arsenic. Values with * show high values than standard, -: not detected

DISCUSSION

Based on the total plate count, water from tin mining ponds were contaminated by the presence of bacterial population considered to be unfit for drinking purposes. The total bacterial count exceeded the limit of 1.0×10^2 cfu/ml standard limits for drinking water (Environmental Protection Agency, EPA, 2015). According to EPA (2015), for water to be considered no risk to human health, the total viable bacteria count should not exceed 500 cfu/ml. The present study provides evidence of higher microbial contamination in pond water and consistent with previous result on assessment of surface water sources (Odeyemi *et al.*, 2011). In most cases this has been attributed to high rate of human activities such as swimming, agricultural activities, washing and defecation within and around these water sources (EPA, 2015). Presence of coliform provides a definite evidence of faecal contamination in water, which also indicates the capacity of the mining pond to transmit diseases relating to water pollution. Variation in contamination level among the ponds with higher viable counts encountered during the rainy seasons agrees with the findings of Avotri *et al.* (2002).

In this study, a positive and significant relationship was observed between viable counts of bacteria cell and season of the year. This is a pattern observed globally in most surface water attributed to agricultural usage, wastewater resulting from human activities, and surface water runoff (Avotri *et al.*, 2002; Odeyemi *et al.*, 2011; EPA, 2015). *Escherichia coli*, *Shigella dysenteriae*, *Vibrio cholerae*, *Proteus vulgaris*, *Yersina enterocolitica*, *Bacillus subtilis*, *Salmonella enteritidis*

and *Salmonella Typhi* were predominant in this study. Similar reports by Ichor *et al.* (2014) on microbial contamination of surface water sources in rural areas reported the isolation of *E. coli*, *Klebsiella* species, *Shigella* species, and *S. aureus*. Therefore, the presence of these organisms as predominant bacteria species in the water pond indicates that the water is polluted with human and animal faeces (EPA, 2015). Water contaminated with these bacterial species is regarded as a greater risk to human health. When *E. coli* is found in water, it suggests the presence of pathogens causing waterborne illnesses (Olsen *et al.*, 2002). Infection of persons with *E. coli* serotype 0157:H7 may lead to haemorrhagic colitis, gastroenteritis and haemolytic-uraemic syndrome and (Bahiru *et al.*, 2013).

The results of this study showed variations in mean pH with higher values observed in wet season- an indication of marked significant seasonal fluctuations ($P < 0.05$). The seasonal variation in pH values agrees with results obtained by Usman *et al.* (2014), with highest pH in the wet season and lower values in late rainy season. The seasonality in the pH of water may be due to the influx and decay of debris in the area as well as imbalance level of H^+ ions input from surface run-offs during the rains. Significant variations ($p < 0.05$) in TSS level in the water sample from mining ponds and seasons observed agree with earlier reports (Usman *et al.*, 2014). The higher TSS during wet season may be due to the continuous discharge of effluents from houses that carries many materials into the ponds. Bilotta and Brazier (2008) reported that fine particles sometime act as food source for filter feeders, which are

part of the food chain, leading to biomagnification of chemical pollutants in fish and ultimately, in man.

In this study, 100% of the mining ponds had TDS above recommended maximum value of <300 mg/L for drinking water (WHO, 2011). Palatability of water with a TDS level of less than 500 mg/L is generally considered to be good (NSDWQ, 2007) hence, drinking water needs treatment when TDS concentrations exceed 500 mg/L, or 500 parts per million (ppm). High TDS concentration may indicate elevated levels of ions such as aluminum, arsenic, copper, lead, nitrate and others that do pose a health concern (EPA, 2015). The turbidity values obtained varied between ponds and exceeded the WHO (2011) guidelines of <5 NTU for domestic water use. The relatively high level of turbidity during the wet season is expected since the period is characterized by heavy rainfall. Generally during wet season, suspended particles in the water are always in motion due to high rate of water circulation whereas in the dry season, the particles tend to settle on submerged logs as there is little turbulence (Avotri, 2002).

If the concentration of dissolved oxygen falls below 5 mg/L, it may have adverse effects on how biological communities function and survive and if it is below 2 mg/L, it may lead to death of fishes (Chapman, 1996). Generally, the finding of this study revealed that the water from the mine ponds was well oxygenated during wet and dry seasons within allowable limit for use as raw water. Higher values of dissolved oxygen obtained in the wet seasons could be due to agitation and frequent wind current in

the water (Kolo and Yisa 2000; Ramulu and Benarjee, 2013). The BOD value observed throughout the study period indicates that the tin-mine ponds are polluted and contain high content of easily degradable organic materials.

Heavy metals found in the environment may occur from natural processes and as pollutants from human activities (Franca *et al.*, 2005). In this report however, low levels of heavy metals were observed among the mining ponds and were within acceptable limits (World Health Organization, 2004; Nigeria Standard for Drinking Water Quality, NSDWQ, 2007; Standards Organization of Nigeria, SON, 2007). The only exception to the heavy metals concentration was lead. Study has also reviewed high lead in Zamfara, Nigeria which results to the killing of estimated 400 children (MSF, 2012). The concentration of cadmium and chromium also exceeded the recommended limits in the months of December and January.

Arsenic was also observed in one of the mining pond (Mista-Ali, 0.07 mg/L) which is also above the recommended standards of 0.01 mg/L (WHO, 2004; NSDWQ, 2007). Arsenic is known for its toxicity and carcinogenic properties (NSDWQ, 2007) and exposure to arsenic may occur *via* inhalation, ingestion, dermal contact, and parenteral routes (WHO, 2004). Contamination of water with arsenic may also be due to inappropriate disposal of arsenical chemicals within the mining ponds (Singh *et al.*, 2007).

CONCLUSIONS

Water from the tin mining ponds were heavily contaminated with bacteria

higher than the recommended standards of WHO and NSDWQ for domestic and drinking purposes. The study also showed significant seasonal variation in the bacterial loads, with months of September and October (wet seasons) having highest contamination level. *Escherichia coli*, *Bacillus subtilis*, *Vibrio cholerae*, *Salmonella enteritidis*, *Shigella dysenteriae*, *Yersinia enterocolitica*, *Proteus vulgaris* and *Salmonella Typhi* were more consistently isolated from the mining ponds.

There were seasonal influences on the physicochemical properties of the ponds with the highest impact occurring in the wet season during the month of August to October. Heavy metal contaminants detected in the tin mining ponds were lead, cadmium and arsenic. However, lead occurred most and significantly higher than recommended limits though variations existed in their levels between ponds and seasons.

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