Asian Journal of Chemistry Vol. 18, No. 4 (2006), 2533–2537

Physico-chemical and Microbiological Analysis of Dairy Effluents, Nathayyapalem, Gajuwaka, Visakhapatnam, Andhra Pradesh

K.M. ELIZABETH*, K. NAGESH, Y. VIMALA AND P. HEPSIBAH
Department of Microbiology, College of Science
Gandhi Institute of Technology and Management
Visakhapatnam-530 045, India
E-mail: kmelizabeth@yahoo.com

The physico-chemical and microbiological parameters of both untreated and treated effluents of Visakha dairy farm were studied. The physical properties of the effluents such as pH, conductivity, turbidity and temperature were determined. The chemical parameters evaluated were DO, total hardness, calcium hardness, magnesium hardness, chlorides, total solids, total dissolved solids, suspended solids, nitrates, ammonia, hexavalent chromium, nickel, iron, copper and oil and grease. The microbiological parameters employed were MPN test and total plate count. Studies on bioremediation involve removal of some of the pollutants such as ammonia, copper and oil and grease from untreated dairy effluent by viable, killed and immobilized non-pathogenic bacteria.

Keywords: Physico-chemical analysis, Dairy effluents, Bioremediation, Ammonia, Copper, Oil and grease, Bacteria.

INTRODUCTION

Visakhapatnam Dairy Industry is situated in Nathayyapalem, Gajuwaka. It discharges its treated industrial effluents into the sea and on to the land for irrigation of plants. The treated and untreated effluents liberate very bad odour, causing nausea and affecting the health of the surrounding population. The effluents before and after discharge from the treatment plant produce putrefied smell of H_2S, polluting the entire air and land in that area. Therefore an attempt was made to study the physico-chemical and microbiological parameters of Visakha Dairy Farm effluents and the probable mechanisms of removal of few pollutants such as ammonia, copper and oil and grease by non-pathogenic bacterial flora.

EXPERIMENTAL

The dairy effluents of Visakhapatnam were collected in sterile bottles and were refrigerated until microbiological and DO analyses were performed. All the tests were performed according to the method of APHA[1–3]. The physical parameters

such as pH, conductivity, temperature and turbidity were measured by pH-meter, conductivity-meter, thermometer and turbidometer respectively. The microbiological methods employed were MPN test and total plate count test (TPC). For determination of DO Winkler's method was followed. For TH, CaH, MgH and chlorides titrimetric methods were followed. For estimation of TS, TDS and SS gravimetry was used. The nitrates, chromium, ammonia, nickel, iron and copper were estimated by spectrophotometric methods.

Bioremediation of ammonia, copper and oil and grease: The non-pathogenic bacteria such as *Staphylococcus, Pseudomonas, Bacillus* and *Clostridium* species were maintained on nutrient agar medium.

Estimation of ammonia, copper and oil and grease: Ammonia, copper and oil and grease were estimated by Nessler's, neocuproin and petroleum ether methods respectively. Various concentrations of ammonium chloride and copper metal were used to draw the standard curves of ammonia and copper respectively. The amount of these pollutants in the effluent sample was estimated from the standard curve. The viable bacteria and killed bacterial powders and immobilized bacterial cells were incubated in the untreated effluent for different contact periods and with different sorbent concentrations as described earlier[4]. Later, the treated effluents with bacteria were spun at 5000 rpm for 15 min and the culture supernatants were subjected to ammonia, copper and oil and grease assays. Control effluents without inoculation with microbial cells or other forms were also simultaneously incubated whose supernatants were subjected to ammonia, copper and oil and grease assays. The amounts of ammonia and copper present in the effluents were estimated .

RESULTS AND DISCUSSION

The results are presented in Tables 1–5. The physical factors of Visakha Dairy Farm (Table-1) showed very high turbidity, *i.e.*, 100.9 NTU in untreated effluent which was reduced to 1.49 NTU after treatment. The pH of untreated effluent was around neutral (7.7) but turned to be alkaline after treatment indicating that it had exceeded the normal permissible limit. Similarly the conductivity was 1.5 μmhos before treatment which was increased to 2.55 after treatment indicating that this value was beyond the permissible limit. The temperatures of both the effluents were beyond the normal limits. The microbiological parameters such as MPN test and total plate counts were very high in both the effluents indicating that these values had exceeded the permissible limits. The dissolved oxygen content in untreated and treated effluents was absent indicating that the effluents were heavily polluted with organic materials. The total hardness of both effluents was very high indicating that their values had crossed the normal limits. The Ca hardness and Mg hardness of both effluents were less, within the permissible limits. The chloride content was very high (389.3 mg/L) before treatment which had come down to 25.5 after treatment. The total solids of untreated and treated effluents were 1900 and 1600 mg/L respectively indicating that they had crossed the limits. There was high content of total dissolved solids (1400 mg/L) in treated

effluent and suspended solid content in untreated effluent resulting in high turbidity. The nitrate contents in untreated and treated effluents were 14 and 16 mg/L respectively, indicating that the nitrate content had increased after treatment but the nitrate content was within the permissible limits. The ammonia contents in untreated and treated effluents were 400 and 4 mg/L respectively indicating that the ammonia content was brought down after treatment. Copper content in untreated effluent was 9 mg/L which was reduced to 3 mg/L after treatment. There was 1000 mg/L of oil and grease in untreated effluent which was reduced to 500 after treatment but the values were beyond the permissible limit. Other pollutants such as iron, nickel, hexavalent chromium and zinc were absent in both the effluents. Similar results were obtained by Pandit and Prajapathi[5] who reported the absence of D.O. in dairy effluent of Bhavnagar, Gujarat. The results of the present study differ with their results in possessing less amount of oil and grease (5500 mg/L), high amounts of turbidity (70.4 NTU) and presence of copper.

TABLE-1
PHYSICO-CHEMICAL AND MICROBIOLOGICAL ANALYSIS OF VISAKHA DAIRY EFFLUENTS

Parameters	Untreated effluent	Treated effluent	Permissible limit
pH	7.7	8.89	6.5–8.5
Conductivity (μmhos)	1.5	2.55	< 2.5
Turbidity (NTU)	100.9	1.49	5.0
Temperature (°C)	29	29	25–27
MPN test (coliforms/1 00 mL sample)	918	1609	0–20
Total plate count (CFU/mL)	2.5×10^8	2.24×10^8	< 100
DO (mg/L)	0.0	0.0	4–8
Total hardness (mg/L)	212	220	75–150
Ca hardness (mg/L)	84	100	75–150
Mg hardness (mg/L)	128	120	75–150
Chlorides (mg/L)	389.3	25.5	250
Total solids (mg/L)	1900	1600	500–1000
Total dissolved solids (mg/L)	700	1400	500–1000
Suspended solids (mg /L)	1200	200	500–1000
Nitrates (mg/L)	14	16	< 45.0
Ammonia (mg/L)	400	4	50.0
Iron (mg/L)	Nil	Nil	0.2
Nickel (mg/L)	Nil	Nil	5.0
Chromium (μg/L)	Nil	Nil	5–10
Copper (mg/L)	9	3	5.0
Oil and grease (mg/L)	1000	500	50

The bioremediation studies revealed the fact that there was (Table-2) highest percentage removal of ammonia (82.7%) by viable *Bacillus* species and least by *Staphylococcus* species (40.5%). The viable *Clostridium* species could remove 72.5% of copper and least removal was by *Staphylococcus* species. There was a range of 80–90% removal of oil and grease by viable cells. The biosorption of ammonia (Table-3) showed that there was 80.6% removal by *Staphylococcus* species within 90 min. *Pseudomonas* could remove 70.6% of ammonia within 1 h. *Bacillus* species removed 44.8% in 1 h by biosorption technique which did not increase rapidly with increasing time. There was 63.2% removal of ammonia by *Clostridium* species in 1 h which was increased to 86.5% in 2 h. The removal of copper, oil and grease were very high (Table-4) by *Clostridium* species (55.6%) and *Bacillus* species (90%) respectively through biosorption technique. There was highest percentage removal of ammonia (Table-5) by *Pseudomonas* (63.2%), copper by *Clostridium* species (66.7%) and oil and grease by *Pseudomonas* species (90%) through immobilization technique.

TABLE-2
BIOREMEDIATION OF AMMONIA, COPPER, OIL AND GREASE BY VIABLE BACTERIA

Organism	No. of cells	Percentage removal		
		Ammonia	Copper	Oil and grease
Staphylococcus sp.	10^8	49.5	50.0	90.0
Pseudomonas sp.	10^8	62.5	61.5	80.0
Bacillus sp.	10^8	82.7	72.3	90.0
Clostridium sp.	10^8	73.7	72.5	80.0

TABLE-3
BIOREMEDIATION OF AMMONIA BY BIOSORPTION TECHNIQUE

Contact time (min)	Percentage removal by organisms			
	Staphylococcus sp.	*Pseudomonas* sp.	*Bacillus* sp.	*Clostridium* sp.
30	7.90	45.30	33.20	47.90
60	47.90	70.60	44.80	63.20
90	80.60	77.60	44.80	80.60
120	86.85	73.20	49.50	86.50

These results clearly indicate that the Visakha Dairy industrial effluents (untreated and treated) were highly polluted. The parameters such as pH, conductivity, MPN count, total plate count, DO, total hardness, TS, TDS and oil and grease were beyond the permissible limits even after treatment. The above mentioned properties and other parameters had crossed their respective limits before treatment. Bioremediation of ammonia, copper and oil and grease showed that these pollutants could be removed efficiently by viable, killed and immobilized bacteria which can be utilized in environmental pollution control programmes.

TABLE-4
BIOREMEDIATION OF COPPER, OIL AND GREASE BY BIOSORPTION

Organism	Sorbent concentration (mg/mL)	Contact time (h)	% removal of	
			Copper	Oil and grease
Staphylococcus sp.	20	24	33.4	60.0
Pseudomonas sp.	20	24	0.0	70.0
Bacillus sp.	20	24	44.4	90.0
Clostridium sp.	20	24	55.6	40.0

TABLE-5
BIOREMEDIATION OF AMMONIA, COPPER, OIL AND GREASE BY IMMOBILIZATION TECHNIQUE

Organisms	Contact time (h)	Weight of beads (g /mL)	% removal of		
			Ammonia	Copper	Oil and grease
Staphylococcus sp.	24	1	59.0	52.8	40.0
Pseudomonas sp.	24	1	63.2	22.3	90.0
Bacillus sp.	24	1	15.0	38.9	80.0
Clostridium sp.	24	1	78.6	66.7	70.0

ACKNOWLEDGEMENTS

The authors are thankful to the management of GITAM for providing laboratory facilities. The authors Dr. K.M. Elizabeth and P. Hepsibah are grateful to UGC, (Hyderabad and New Delhi) for its financial assistance in the form of sanctioning a Minor Research Project and JRF respectively.

REFERENCES

1. APHA/AWWA/WEF (American Public Health Association, Water Environment Federation), Standard Methods for the Examination of Water and Wastewater, 19th Edn., United Book Press, Baltimore, pp. 4/90–4/91 (1995).
2. A.I. Vogel, Text Book of Quantitative Chemical Analysis, 5th Edn., C.B.S. Publishers & Distributors, New Delhi (1985).
3. D.S. Ramteke and C.A. Moghe, Manual on Water and Waste Water Analysis, National Environmental Engineering Research Institute, Nagpur, p. 121 (2000).
4. K.M. Elizabeth and G.E. Rani, *Asian. J. Chem.*, **16**, 1269 (2004).
5. B.R. Pandit and S. Prajapati, *Natr. Environ. Pollut. Technol.*, **2**, 341 (2003).

(*Received*: 8 April 2005; *Accepted*: 31 March 2006) AJC-4759