

LAB SCALE STUDIES ON THE EFFICIENCY OF BACTERIAL STRAIN ON THE TREATMENT OF SLAUGHTERHOUSE WASTEWATER

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Key words : Bacterial Strain, Enzyme, Aerobic Treatment, Slaughterhouse Wastewater, Fat.

(Received 19 June, 2012; accepted 25 July, 2012)

ABSTRACT

The objective of this study is to investigate the potency of bacterial strain on improving the quality of slaughterhouse wastewater. Studies on biodegradation of COD, BOD, high protein and fat by selected enzyme producing bacteria were carried out using slaughterhouse wastewater with mixed culture of bacterial strain. At lab scale, it was found that, treatment with bacterial strain in aerobic condition resulted in COD, BOD, protein and fat removal to the extent of 82.12%, 86.33%, 93.33% and 66.66% respectively based on degrading capacity of micro-organism. This article discusses in detail the efficiency of bacterial strain as an amendment to the aerobic system treating slaughterhouse wastewater.

INTRODUCTION

Abattoir wastewater is characterized by the presence of high concentration of blood of the slaughtered animals and suspended particles of the semi digested and undigested foods within the stomach and intestine of slaughtered animals. Recent reports show that zoonotic diseases (i.e. diseases of animals that transmitted to humans) are yet to be eliminated or fully controlled in the Asian countries. Thus they pose serious environmental health risk to the public. Some of the infectious diseases are T.B., Colibacillosis, Salmonellosis, Brucellosis and Helminthosis. Effluent discharge from the slaughter houses cause the deoxygenation of rivers (Quinn and McFarlane, 1989) and the contamination of ground water (Sangodoyin and Agbawhe, 1992). Blood is the major dissolves pollutants in the slaughter house

wastewater and has a COD of 375000mg/L. (Tritt and Schuchardt, 1992). Slaughter house wastewater also contains high concentration of suspended solids including bone pieces, grease, feathers, flesh, manure, grit and undigested food (Bull *et al.*, 1982). Abattoir wastewater contributes high COD and BOD concentration in the range of 4400-18000mg/L. However the composition varies based on the extent of by product recovery, and amount of water consumption during slaughtering. This may be due to the non salvaging of the blood by the slaughter house authorities.

Proteins and lipids (fat, oil and grease) are the major organic matters in slaughter house wastewater and can cause severe environmental pollution. High concentration of these compounds in wastewater often causes major problems in biological wastewater treatment processes. Because of their nature they form

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a scum layer on water surfaces and decrease oxygen transfer rate into the aerobic process. Bioremediation of nutrient rich wastewater of slaughter house, either aerobically or anaerobically, have been investigated. Generally, the protein and fat is degraded by enzymes proteases and lipases produced by the microbial community. Hence, the protein and fat rich wastewater can easily be treated by supplying microbial/bacterial strain which produce proteases and lipases enzymes. The particular benefits offered by enzymes are specificity, mild conditions and reduced waste. It may be possible, by choosing the right enzyme produced by selected bacterial species suitable to treat wastewater of particular composition (Hasan *et al.*, 2005).

Studies on bioremediation of high fat and oil wastewater by selected lipase producing bacteria like *Bacillus subtilis*, *B. licheniformis*, *B. amyloliquifaciens*, *Serratia Marsescens*, *pseudomonas aeruginosa* and *staphylococcus aureus* were carried out in wastewater emanating from palm oil mill, dairy, slaughter house, soap industry and domestic water effluent (Prasad and Manjunath, 2011). The skimmed fat-rich liquid is digested with lipases such as that from *C. rugosa*. Effective breakdown of solids and the clearing and prevention of fat blockage or filming in waste systems are important in many industrial operations (Bailey *et al.*, 1986). Fats in wastewater treatment plants that contains mainly triglycerides is hydrolysed by immobilized lipase (Tschöcke, 1990) *P. aeruginosa* LP602 cells and the lipase were shown to be usable for lipid-rich wastewater treatment (Dharmstithi and Kuhasuntisuk, 1998). High-performance micro-organisms developed for use in the biological wastewater treatment with a high content of greases, fats, and oils. Report further says good efficiency of micro-organisms (<http://www.oasisenviro.co.uk/ww07pproductinfo.html>). Lipases of plant, bacterial and animal (pancreatic) origin have shown to hydrolyse and/or reduce the size of fat particles in slaughterhouse wastewater (SHW) (Masse *et al.*, 2001).

Based on the detail literature survey, it is preferable to use specialized bacterial strain for the treatment of industrial wastewaters because it is ecofriendly, cost effective and sustainable technique. Specialized strain of selective bacterial species is the answer which gives high degradation of COD, BOD, protein, fat and other nutrient in effluent wastewaters.

MATERIALS AND METHODS

Initially work has been initiated for the isolation,

culturing and identification of required microbial strain.

Isolation of Microbes

For isolation of microbes, Bacterial species are collected from campus of National chemical laboratory, Pune and then the sample was collected and stirred in double distilled water. The serially diluted samples were plated on tributyrin agar plates. The formation of clear zone around the colony on the plate was considered as microbes.

Identification

Microbes which formed clear zones around the colony were identified on morphological and physiological characters according to '*Bergey's manual of determinative bacteriology*' (Doudoroff and Palleroni, 1974); all species are gram +ve in nature.

Bacterial species identified are:

- *Lignocellulosic Bacteria -Bacillus subtilis*
- *Pseudomonas aeruginosa*
- *Megaterium family*
- *Acetobactor*
- *Clostridium*
- *Thermophilic bacteria*
- *Sulphur reducing bacteria*

Pure culture of these microorganisms was maintained on nutrient agar slant.

Biodegradation of slaughter house effluent by isolated specialized bacterial strain (i.e. mix culture) was carried out for 24 hrs and then BOD, COD value, and lipid content were analyzed.

After isolation of specific bacterial strain, their role in treatment was studied on laboratory scale experiments. Lab scale studies indicated very good results in terms of COD, BOD, Protein and Fat removals.

Slaughterhouse Wastewater Treatment

Wastewater needed for the experimental work was collected and composited. The wastewater was collected from a large animal slaughterhouse situated at Solapur

The slaughterhouse wastewater sample (10 L) taken in 15 L container and allow to settle for 30 min for removal of suspended and floating impurities in the range of 70-71%. From that 5 L was inoculated with 1% (v/v) bacterial culture. After vigorous shaking, it was divided into 20 portions of 250 mL each in 500 mL flask. All the cultures were incubated at 30°C at 200 rpm; the same incubation was done for remaining 5 L settled raw wastewater without

addition of bacterial culture. Samples were taken at regular interval of 24 hrs from 500 mL flask for physicochemical analysis.

The lab scale experiment was performed and physicochemical analysis was done as per the standard methods (APHA, 1998).

RESULT AND DISCUSSION

The purpose of the present study was to evaluate the potency of mixed culture of bacterial species to reduce the BOD, COD and high protein and fat content from slaughter wastewater. In the present study, bacterial colonies are isolated which resulted in large clear zone tributyrin agar media. All the isolates were then cultured on tributyrin agar plates and observed. The prepared bacterial strain is then subject to wastewater treatment at lab scale experiment to observe its potency in aerobic conditions. Lab scale studies indicated very good results in terms of COD, BOD, Protein and Fat removals.

The characteristics of untreated raw wastewater of slaughter house rich in BOD, COD, protein and fats is showed in Table 1. The pH in the range of 7.4 to 8.2 was observed and average value of BOD, COD, protein and per cent fat was 1428mg/L, 2630 mg/L, 6412 mg/L and 0.48% respectively. The wastewater subjected to simple settling for 30 min., resulted removal of 70%-71% in settleable solids, 38%-42% in BOD and 32%-36% in COD, protein reduction 1%-2% and fat reduction 4%-5% achieved.

The settled effluent further treated with and without mixed culture of bacterial strain in aerobic system. At lab scale, It was found that, treatment with bacterial strain resulted in COD, BOD, protein and fat removal to the extent of 82.12%, 86.33%, 93.33% and

66.66% respectively based on degrading capacity of micro-organism. Whereas, the treatment without bacterial strain resulted in COD, BOD, protein and fat removal to the extent of 67.04%, 70.54%, 62.28% and 31.25% respectively showed in Figure 1. It is observed that in treatment with bacterial strain, the COD, BOD, protein and fat removal is greater by 16.08%, 15.79%, 31.09% and 31.10% respectively showed in Figure 2. From the lab scale results, it is clear that; the microbial strain is very useful in improving the treatment efficiency of slaughterhouse wastewater.

CONCLUSIONS

In the present study, bacterial colonies are isolated which resulted in large clear zone tributyrin agar media. All the isolates were then cultured on tributyrin agar plates and observed. The prepared bacterial strain is then subject to wastewater treatment at lab scale experiment to observe its potency in aerobic system. From the detailed study using specialized bacterial strain in aerobic system, the following conclusions can be drawn;

Slaughterhouse wastewater is highly emanable to biological treatment. Simple settling reduces the COD, BOD, suspended solids to the tune of 37%- 40%, 38% -42% and 70-72% respectively. Bacteria were isolated and cultured from soil sample. Bacterial strain addition improves the treatment efficiency by manifolds. Apart from COD and BOD; reduction in Protein and fats is also achieved. Enzymatic treatment technique is clean and ecofriendly application technique. Hence, it is necessary to treat the slaughterhouse wastewater by biological technique in combined with bacterial strains in secondary aerobic system. Based on these results, Pilot plant study can also be undertaken to

Table 1. Physico-Chemical Characteristics of Solapur Slaughter House Raw Wastewater

Sr. No.	Parameter	Values of raw wastewater	Values After 30 min. settling
1.	pH	7.4-8.2	7.0-7.2
2.	Alkalinity as CaCO ₃	1738 ± 2.94	1628 ± 2.94
3.	TSS	1200 ± 1.82	356 ± 3.51
4.	TDS	2610 ± 2.94	895 ± 2.58
5.	Chemical Oxygen Demand (COD)	2488 ± 1.82	1598 ± 5.47
6.	Biological Oxygen Demand (BOD)	1428 ± 4.76	852 ± 4.08
7.	Sodium as Na	240 ± 2.58	236 ± 2.58
8.	Potassium as K	171 ± 2.94	168 ± 2.94
9.	Chloride as Cl ⁻	274 ± 6.08	262 ± 1.82
10.	Total Kjeldahl's Nitrogen	1026 ± 2.94	1012 ± 2.94
11.	Protein	6412 ± 2.58	6325 ± 2.94
12.	Fat (%)	0.48 ± 0.02	0.46 ± 0.03

*All values except pH are expressed in mg/L; Values are the average of four sets of reading.

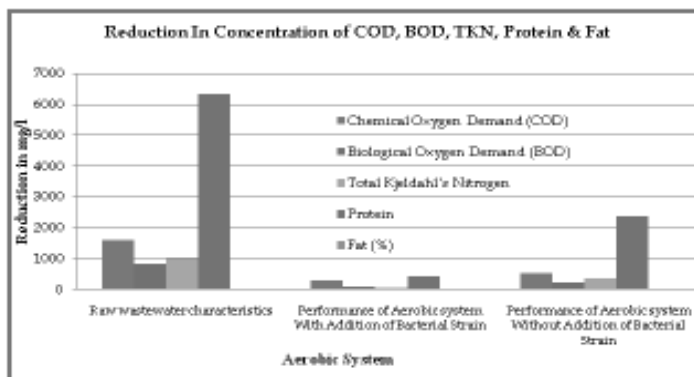


Fig. 1 Comparison between Aerobic System with and without Addition of Bacterial Strain showing Concentration Reduction in COD, BOD, Total Nitrogen, Protein & Fat

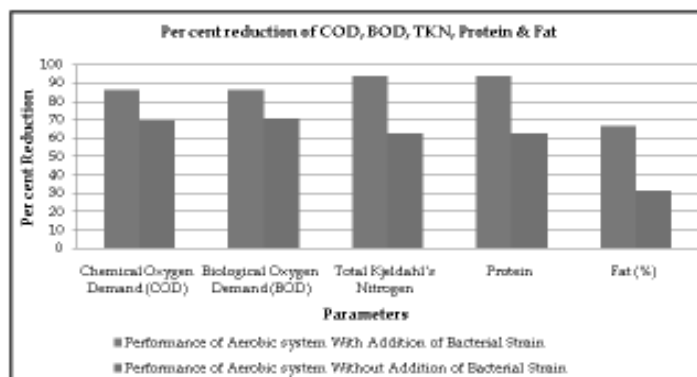


Fig. 2 Performance of Bacterial Strain in Wastewater Treatment

evaluate the potency of bacterial strain in secondary aerobic treatment system of Effluent treatment plant.

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