Comparative Analysis on Biological Reduction in BOD & COD concentration in Pharmaceutical effluents (Before & After treatment) by using Selective Microbial Strains, Baddi Region, Himachal Pradesh, India

[1]Nidhi Singal, [2]Simerjit Kaur
[1]Research Scholar & Assistant Professor, [2]Professor
[1]USS, Rayat Bahra University, Mohali Campus, Punjab, India
and SOS, Baddi University of Emerging Sciences and Technology, H.P., India,
[2]USS, Rayat Bahra University, Mohali Campus, Punjab, India

Abstract : During these investigations, it was found out that BOD & COD concentration in Pharmaceutical effluents could be reduced to a significant level by using Selective Microbial Strains. After determination of BOD and COD, pharmaceutical effluent samples were further treated biologically using microbial strains viz. *Rhizobium leguminosarum*, *Pseudomonas fluorescens* and *Enterobacter cloacae* procured from IMTECH, Sector-39A, Chandigarh. Observations were done on 3rd Day and 7th Day of microbial inoculation with different strains & results indicated that incubation with *Enterobacter cloacae* on Day-3 and Day-7, was most effective in reducing BOD as compared to other two selected microbial strains in before and after treatment samples. Maximum COD reduction of these samples was recorded with bacterial strain *Rhizobium leguminosarum*. Significant variations were recorded in BOD & COD concentration at 5% level of significance ($P<0.05$) after treatment with different microbial strains.

***IndexTerms* - Water pollution, pharmaceutical effluents, DO, BOD, COD, biological treatment, microbial strains.**

I. Introduction

In today's modern world, one of the major problems of concern is pollution, especially of water. The chief pollution load in the water bodies comprising a mixture of complex organic substances such as carbohydrates, fats and proteins, comes from the discharge of effluents from industries manufacturing animal and plant products besides human and animal wastes [1]. All these substances can be recycled easily as these are readily biodegradable and rapidly decomposed through microbial action when introduced into the environment.

When an organic waste that is biodegradable, discharged into a marine ecosystem, the microorganisms oxidize the organic matter and utilize oxygen dissolved in water during respiration [2]. The rate of oxidation of organic matter and the corresponding consumption of oxygen depends directly on the biodegradability of a waste. Hence the organic content and pollution level of waste waters is usually measured in terms of Biological oxygen demand (BOD).The growth of a large number of species of organisms is supported when the dissolved oxygen (DO) concentration is high in an aquatic system. Depletion of oxygen due to indiscriminate discharge of wastes increases the number of decomposers at the expense of others. Chemical Oxygen Demand (COD) is a measure of the total quantity of oxygen required to oxidize both biologically oxidisable and chemically inert substances into carbon dioxide and water. Also BOD determination takes 5 days whereas COD determination takes only a few hours, hence COD values are always greater than BOD values

The availability of plant nutrients, particularly nitrogen, phosphorus or both may increase the aquatic biological productivity. This in turn depletes dissolved oxygen and causes eutrophication of lakes, ponds, rivers, estuaries and marine ecosystems due to excessive nutrient loading which eventually alters the energy relationship and water balance, disrupting biotic community structure and function. [3]. It not only adds to the nutrient-content of water but the addition of some forms of nitrogen and phosphorus also increased BOD and COD [4]. The discharge of inappropriately treated effluents from WWTPs also results in the deposition of large amounts of organic matter and nutrients, which have major deleterious effects on the health of the existing micro- and macro-fauna and the surrounding environment.

In a recent study, unusually high levels (mg/l) of several drugs were detected in the effluents from local wastewater treatment plant near Visakhapatnam in India [5]. The BOD and COD values in all the three industrial sites in and around Tirupati were above the permissible limits of 100 and 200mg/L by ISI and WHO. DO levels more than 4 mg/L is desirable but all the samples showed very negligible amount of DO [6]. Seasonal variations in DO, BOD, CO were assessed in water of Sirhind Canal passing through Moga, Punjab, India. During summers, Decreased DO and High BOD and COD was recorded, while in monsoon season, DO, BOD and COD concentration indicating the presence of a large number of microorganisms, and hence a high level of pollution [7].

The wastewater from basic drug industry is one of the worst polluting industrial liquid wastes having high amounts of organic matter with COD as high as 48,000 mg/l. The entry of pharmaceutical process wastewaters into the surface waters alters the flow conditions, morphology and the surrounding habitat. They poison the wildlife of the rivers, damage aerobic organisms and their number gets significantly reduced. The lack of oxygen favors anaerobic degradation processes, which can cause biological imbalance [8]. Long-term exposure of lower concentration of complex pharmaceutical mixtures to stream biota may result in acute and chronic damages [9,10], behavioral changes [11,12], accumulation in tissues [13], reproductive damage/abnormalities [14] and inhibition of cell proliferation [15].

Soluble organic sources of biochemical oxygen demand (BOD) can be removed by any viable microbial process, aerobic, anaerobic or anoxic. However, aerobic processes are typically used as the principal means of BOD reduction of domestic wastewater because the aerobic microbial reactions are fast, typically 10 times faster than anaerobic microbial reactions. Membrane bioreactors use a combination of the activated sludge process with an additional membrane separation process. The advantages offered by MBRs over traditional activated sludge systems include reduced footprints, a decrease in sludge production, better effluent quality and proficient treatment of wastewaters with varying contamination peaks. These reactors have been used in the treatment of a vast range of municipal or industrial wastewaters such as from pharmaceutical industry [16,17]. Physico-chemical analysis of untreated effluents of textile, automobile and pharmaceutical industries recorded the biological oxygen demand as 322 mg/l, 245 mg/l and 410 mg/l which reduced to 28.0 mg/l, 24.0 mg/l and 14.0 mg/l respectively in the treated effluents. The chemical oxygen demand was recorded as 550 mg/l, 635 mg/l and 548mg/l which also decreased to 236 mg/l, 156 mg/l and 172 mg/l respectively. Therefore, the pharmaceutical effluents could be easily treated and after treatment BOD and COD decreased [18].

The present study aimed to investigate the pollution level of effluents collected from selected pharmaceutical industries (PI-1 to PI-5) from Baddi industrial area, Himachal, India with respect to Dissolved oxygen content (DO), Biological Oxygen Demand (BOD) and Chemical Oxygen Demand (COD) and the subsequent biological treatment with microbial strains to minimize it.

II. MATERIALS AND METHODS

The pharmaceutical effluent samples were collected from the selected five pharmaceutical industrial effluent outlets PI (1-5) before their discharge into Common Effluent Treatment Plant (CETP) followed by collecting the sample of effluent treated in CETP. High-grade polyethylene bottles of 1 litre capacity were used for storing the effluent samples.

2.1 Objective of Sampling

To collect a portion of material small enough in volume to be transported conveniently and handed in the laboratory while still accurately representing the material being sampled. This implied that the relative concentrations of all related components were same in the samples as in the material being sampled and that the sample was handled in such a way that no significant changes in composition occurred before the tests were made.

2.2 Methods

The standard methods were adopted for the analysis of these parameters [19]. Statistical analysis of the data was carried out by using One Way ANOVA at 5% level of significance of various samples before and after treatment in CETP.

2.3 Procedure for Bioremediation

Three microbial strains procured from IMTECH, Sector-39A, Chandigarh, were *Rhizobium leguminosarum*, *Pseudomonas fluorescens* and *Enterobacter cloacae* and revived on prescribed media. For this, each culture was inoculated in MH broth, in flasks. These flasks were incubated in orbital shaker maintained at 37ºC at 180 rpm. After 24 hours, 100µl of the culture was withdrawn and added in BOD bottles, each containing 20 ml of the sample. The bottles were incubated at 37ºC. DO, BOD and COD were determined on 3rd and 7th day after incubation of the sample as per the standard procedures.

III. RESULTS AND DISCUSSION

The results of biochemical analysis of PI effluents were recorded as under-

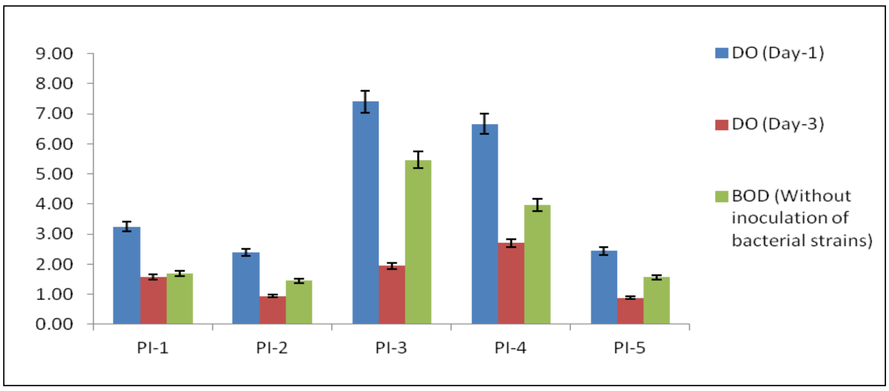

Fig-3.1 Variations in DO & BOD in effluents of selected pharmaceutical industries in Baddi region, H.P., India

The results of Fig.3.1 indicated that DO values on Day-1 ranged from 2.38mg/l to 7.41mg/l, with an average concentration of 4.43mg/l in the untreated PI samples. Maximum DO was recorded in the samples of PI-3 (7.41mg/l) followed by PI-4 (6.67mg/l), PI-1 (3.25mg/l), PI-5 (2.44mg/l) and minimum in the samples of PI-2 (2.38 mg/l) and within permissible limits. After incubation for 3 days, readings were again recorded. On Day-3, dissolved oxygen concentration decreased and ranged from 0.87mg/l to 2.70mg/l, with an average concentration of 1.61mg/l. Maximum DO was recorded for PI-4 (2.70mg/l), followed by PI-3 (1.94mg/l), PI-1 (1.57mg/l), PI-2 (0.94mg/l) and PI-5 (0.87mg/l) respectively. Significant variations were observed in DO concentration on Day-1 and Day-3 in PI-3 & PI-4 samples at 5% level of significance ($P< 0.05$).

BOD ranged from 1.44 to 5.47mg/l in the untreated PI samples, with an average concentration of 2.82mg/l. Maximum concentration was recorded in PI-3 (5.47mg/l), followed by PI-4 (3.96mg/l), PI-1 (1.68mg/l), PI-5(1.57mg/l) and PI-2(1.44mg/l) respectively. Results of BOD indicated that the effluent samples of PI-3 were most polluted as compared to other samples, though the value was within limits.

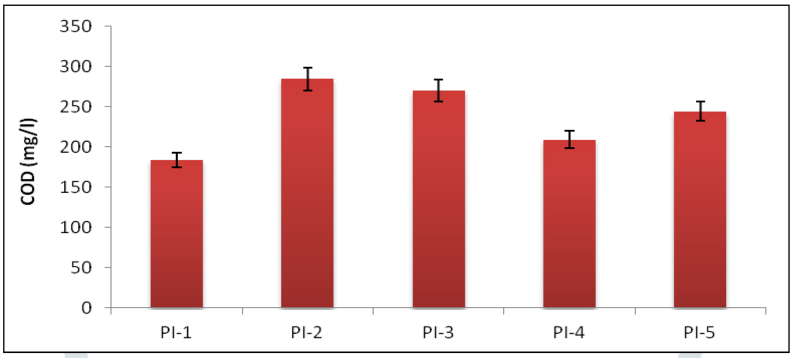

Fig- 3.2 Variations in COD in effluents of selected pharmaceutical industries in Baddi region, H.P., India

The study of Fig.4.24 revealed that COD in selected pharmaceutical industrial samples from Baddi region, H.P., India ranged from 284.8 mg/l to 184.0 mg/l with a maximum value in PI-2 (284.8mg/l) followed by PI-3 (270.4mg/l), PI-5 (244.8mg/l), PI-4 (209.6mg/l) and with a minimum COD in PI-1 (184mg/l) respectively. There were significant variations in COD level in PI-1 and PI-2 samples at 5% level of significance ($P< 0.05$).

Studies conducted by Singh *et al*., 2013 on textile industry of Ludhiana reported very high values of BOD and COD, i.e. 108–790 and 195–3050 mg/l respectively in the effluent samples of 7 different dyeing mills [20]. High level of chemical oxygen demand indicated noxious conditions and presence of resistant organic impurities [21, 22]. The values of BOD and COD of the textile effluent during all the seasons ranged from 121–580 mg/l to 240–990 mg/l, respectively, much higher than WHO water quality standard for BOD (30 mg/l) and COD (250 mg/l) [23].

IV. Results and Discussion

The purpose of seeding (selection of micro-organisms) was to introduce into the sample, a biological population capable of oxidizing organic matter in the waste water. The standard seed material was settled waste water which had been stored for 24-36 hours at 20°C.

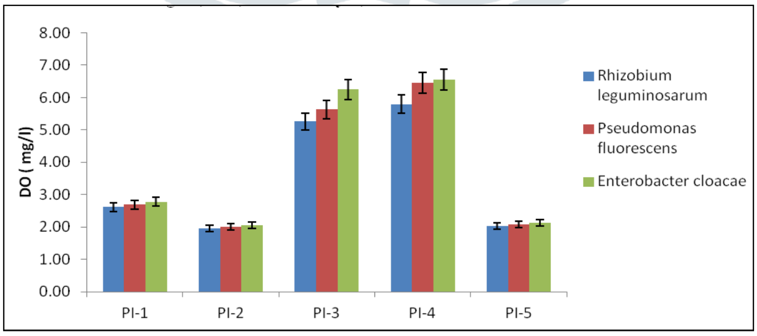

Figure-3.3 Variations in DO in effluents of selected pharmaceutical industries in Baddi region, H.P., India on Day-3, after inoculation with bacterial strains

As indicated in Fig.3.3, after inoculation of effluents from pharmaceutical industries with bacterial strains, dissolved oxygen increased. On 3rd day of incubation with bacterial strain *Rhizobium leguminosarum*, DO increased to 2.61mg/l, with *Pseudomonas fluorescens*, to 2.68mg/l and with *Enterobacter cloacae*, maximum increase in DO i.e. 2.78mg/l was recorded in PI-1 samples. In PI-2 samples, DO increased to 1.95mg/l with *Rhizobium leguminosarum*, 2.0mg/l with *Pseudomonas fluorescens* and 2.05mg/l with *Enterobacter cloacae.* Similarly in PI-3 samples, it increased to 5.26mg/l with *Rhizobium leguminosarum*, 5.63mg/l with *Pseudomonas fluorescens* and 6.25mg/l with *Enterobacter cloacae.* In PI-4, DO increased from 5.80mg/l with *Rhizobium leguminosarum*, to 6.45mg/l with *Pseudomonas fluorescens* and 6.56mg/l with *Enterobacter cloacae*. The concentration in PI-5 increased from 2.03mg/l with *Rhizobium leguminosarum*, to 2.08mg/l with *Pseudomonas fluorescens* and 2.13mg/l with

Enterobacter cloacae respectively. It was observed that the increase in DO was maximum with microbial strain *Enterobacter cloacae.* Insignificant variations were recorded in dissolved oxygen concentration in PI-1, PI-2, PI-3, PI-4 and PI-5 samples after inoculation with the three bacterial strains at 5% level of significance (P< 0.05).

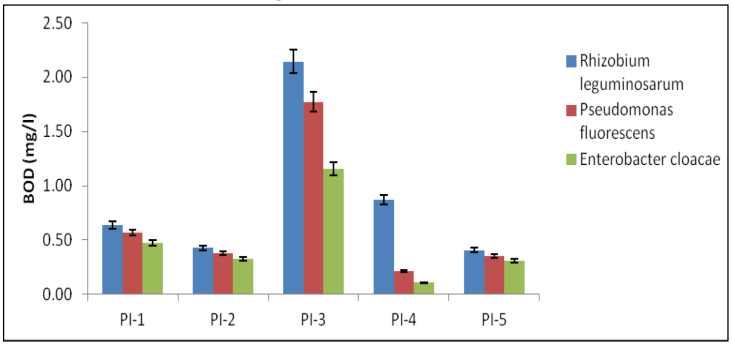

Figure-3.4 Variations in BOD in effluents of selected pharmaceutical industries in Baddi region, H.P., India on Day-3, after inoculation with bacterial strains

The study of Fig.3.4 revealed that BOD concentration decreased after biological treatment of pharmaceutical industrial effluents with microbial strains. On Day-3, after inoculation, BOD decreased to 0.64mg/l with and 0.47mg/l with *E. cloacae* in PI-1 samples. In PI-2, BOD decreased from 0.43mg/l with *R. leguminosarum* to 0.38mg/l with *P. fluorescens* and 0.33mg/l with *E.cloacae*. In PI-3, the decrease was from 2.14mg/l with *R. leguminosarum* to 1.77mg/l with *P. fluorescens* and 1.16mg/l with *E.cloacae.* Similarly in PI-4, BOD decreased from 0.87mg/l with *R. leguminosarum* to 0.22mg/l with *P. fluorescens* and 0.11mg/l with *E.cloacae* and in PI-5 samples, there was a decrease in biological oxygen demand from 0.41mg/l with *R. leguminosarum* to 0.36mg/l with *P. fluorescens* and 0.31mg/l with *E.cloacae* respectively. On comparing the effect of different microbial strains in reducing BOD of PI samples, *Enterobacter cloacae* was found to be most effective. Significant variations were recorded in BOD in PI-4 samples at 5% level of significance (P< 0.05).

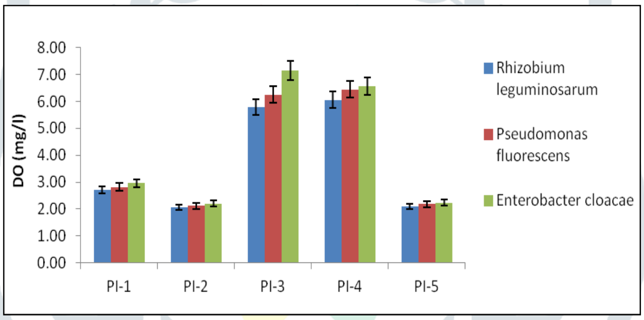

Fig-3.5 Variations in DO in effluents of selected pharmaceutical industries in Baddi region, H.P., India on Day-7, after inoculation with bacterial strains

As indicated in Fig.3.5, DO in PI samples increased further on Day-7, after inoculation with bacterial strains. In PI-1, it increased from 2.70mg/l (*Rhizobium leguminosarum*) to 2.82mg/l (*Pseudomonas fluorescens*) and to 2.96mg/l (*Enterobacter cloacae*). Similarly it increased from 2.06mg/l (*Rhizobium leguminosarum*) to 2.12mg/l (*Pseudomonas fluorescens*) and to 2.20mg/l (*Enterobacter cloacae*) in PI-2 samples. In PI-3, dissolved oxygen increased from 5.80mg/l (*Rhizobium leguminosarum*) to 6.25mg/l (*Pseudomonas fluorescens*) and to a maximum value of 7.14mg/l (*Enterobacter cloacae*), in PI-4 the increase was from 6.06mg/l (*Rhizobium leguminosarum*) to 6.45mg/l (*Pseudomonas fluorescens*) and 6.56mg/l (*Enterobacter cloacae*) and in PI-5 samples, DO increased from 2.09mg/l (*Rhizobium leguminosarum*) to 2.19mg/l (*Pseudomonas fluorescens*) and to 2.23mg/l (*Enterobacter cloacae*) respectively. Insignificant variations were recorded in DO concentration in PI samples after inoculation with the three bacterial strains at 5% level of significance (P< 0.05).

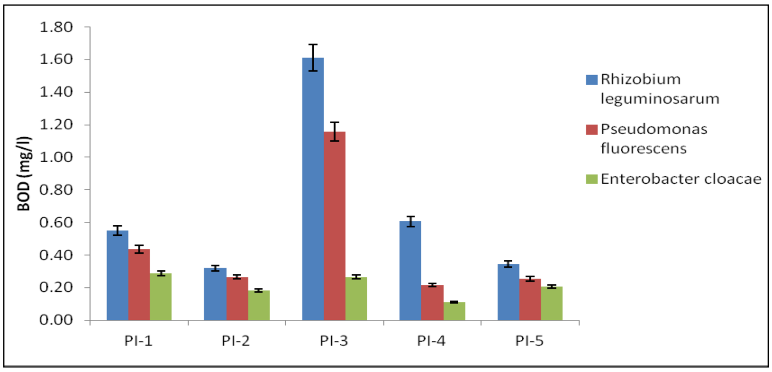

Figure-3.7 Variations in BOD in effluents of selected pharmaceutical industries in Baddi region, H.P., India on Day-7, after inoculation with bacterial strains

The results of Fig.3.7 revealed that BOD concentration decreased after treatment of pharmaceutical industrial effluents with the microbial strains. On Day-7, after inoculation, BOD decreased to 0.55mg/l with *R. leguminosarum*, 0.44mg/l with *P. fluorescens* and 0.29mg/l with *E. cloacae* in PI-1 samples. In PI-2, BOD decreased from 0.32mg/l with *R. leguminosarum* to 0.26mg/l with *P. fluorescens* and 0.18mg/l with *E.cloacae*. In PI-3 samples, the decrease was from 1.61mg/l with *R. leguminosarum* to 1.16mg/l with *P. fluorescens* and 0.26mg/l with *E.cloacae.* Also in PI-4, BOD decreased from 0.61mg/l with *R. leguminosarum* to 0.22mg/l with *P. fluorescens* and 0.11mg/l with *E.cloacae*. Similarly in PI-5 samples, there was a decrease in biological oxygen demand from 0.34mg/l with *R. leguminosarum* to 0.25mg/l with *P. fluorescens* and 0.20mg/l with *E.cloacae* respectively. Again microbial strain *Enterobacter cloacae* was found to be most effective in reducing BOD. Significant variations were recorded in BOD in PI-3 & PI-4 samples at 5% level of significance (P< 0.05) after treatment with different microbial strains.

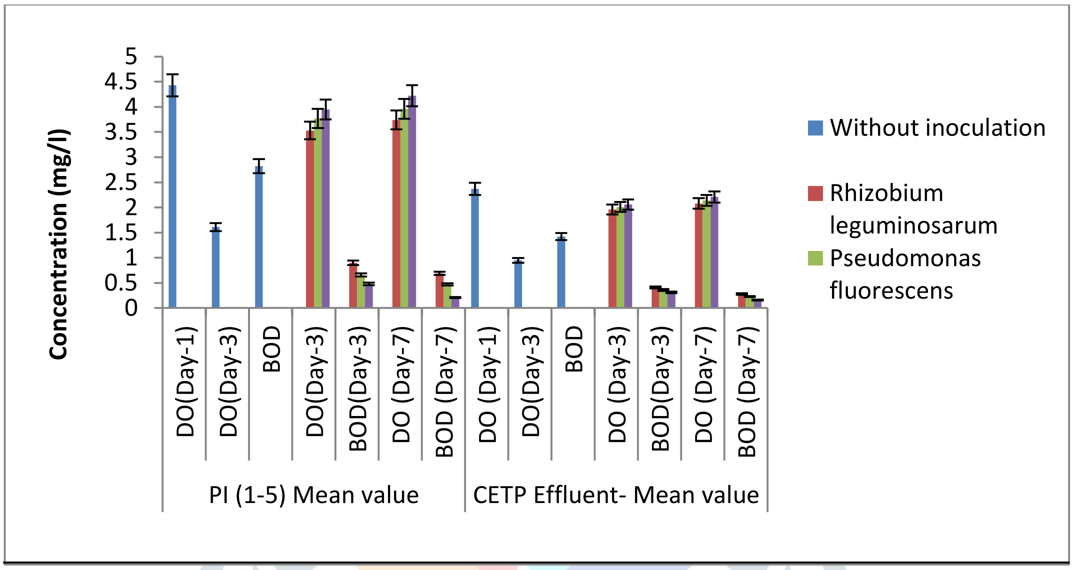

Figure-3.8 Variations in DO & BOD in effluent of selected pharmaceutical industries before and after treatment (CETP) with different bacterial strains

The study of Fig.3.8 revealed that mean DO concentration, in effluents of selected pharmaceutical industries PI (1-5), decreased after incubation from 4.43mg/l (Day-1) to 1.61mg/l (Day-3). Hence calculated value of BOD was 2.82mg/l. After treatment of effluent in CETP, there was a decrease in DO from 2.37mg/l (Day-1) to 0.95mg/l (Day-3). So after treatment in CETP, BOD concentration decreased to 1.42mg/l.

When biodegradation assay was carried out with microbial strains, average DO concentration increased on Day-3 from 3.53mg/l (*Rhizobium leguminosarum*) to 3.77mg/l (*Pseudomonas fluorescens*) and to 3.95mg/l (*Enterobacter cloacae*) and hence BOD decreased from 0.9mg/l (*Rhizobium leguminosarum*) to 0.66mg/l (*Pseudomonas fluorescens*) to 0.48mg/l respectively in PI samples. In CETP effluent samples, on Day-3, DO concentration increased from 1.96mg/l (*Rhizobium leguminosarum*) to 2.01mg/l (*Pseudomonas fluorescens*) and 2.06mg/l (*Enterobacter cloacae*) and hence BOD decreased from 0.41mg/l (*Rhizobium leguminosarum*) to 0.36mg/l (*Pseudomonas fluorescens*) to 0.31mg/l respectively. After inoculation of samples with microbial strains for 7 days, readings were again recorded. The observations revealed that DO concentration increased on Day-7 from 3.74mg/l (*Rhizobium leguminosarum*) to 3.96mg/l (*Pseudomonas fluorescens*) and to 4.22mg/l (*Enterobacter cloacae*) and hence BOD decreased from 0.69mg/l (*Rhizobium leguminosarum*) to 0.47mg/l (*Pseudomonas fluorescens*) to 0.21mg/l respectively.

On comparing the results with CETP treated effluent samples, the observations revealed that DO concentration increased further on Day-7 from 2.08mg/l (*Rhizobium leguminosarum*) to 2.14mg/l (*Pseudomonas fluorescens*) and 2.21mg/l (*Enterobacter cloacae*) and therefore BOD decreased from 0.28mg/l (*Rhizobium leguminosarum*) to 0.23mg/l (*Pseudomonas fluorescens*) to 0.16mg/l respectively. So it can be concluded that biological treatment of CETP treated effluent sample reduced BOD level significantly at 5% level of significance (P<0.05). Strain *Enterobacter cloacae* was found to be most effective in reducing BOD.

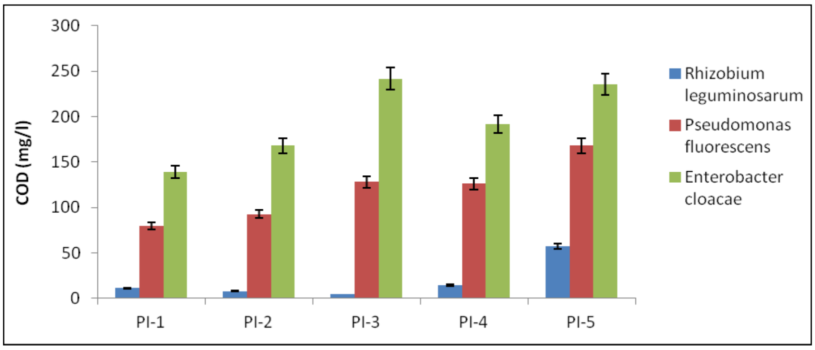

Figure-3.9 Variations in COD in effluents of selected pharmaceutical industries in Baddi region, H.P., India on Day-7, after inoculation with bacterial strains

The results of Fig.3.9 indicated that after biological treatment with microbial strains *Rhizobium leguminosarum*- MTCC99, *Pseudomonas fluorescens*- MTCC103 and *Enterobacter cloacae*- MTCC509, the COD level decreased to 11.2mg/l with MTCC99, 80mg/l with MTCC103 and 139.2 mg/l with MTCC509 for PI-1 samples. For PI-2 samples, COD reduced to 8.0mg/l *with* MTCC99, 92.8mg/l with MTCC103 and 168mg/l with MTCC509. Similarly it was found that there was a visible decline in chemical oxygen demand for PI-3 samples where COD decreased to 4.8mg/l, 128.0mg/l and 241.6mg/l when treated with microbial strains *Rhizobium leguminosarum*, *Pseudomonas fluorescens* and *Enterobacter cloacae* respectively and in PI-4, it decreased from 14.4mg/l with *Rhizobium leguminosarum*- MTCC99, 126.4mg/l with *Pseudomonas fluorescens*- MTCC103 and 192mg/l with *Enterobacter cloacae*- MTCC509. For PI-5 samples, it reduced to 57.6mg/l when inoculated with *Rhizobium leguminosarum*, 168mg/l with *Pseudomonas fluorescens* and 235.2mg/l when biodegradation assay was carried out with *Enterobacter cloacae.*

Also it was observed that as compared to strains *Pseudomonas fluorescens* and *Enterobacter cloacae*, strain *Rhizobium leguminosarum* - MTCC99 was found to be most effective and reduced COD to the maximum extent. There were significant variations in the PI-samples treated with *Rhizobium leguminosarum*, *Pseudomonas fluorescens* and *Enterobacter cloacae* respectively at 5% level of significance ($P< 0.05$).

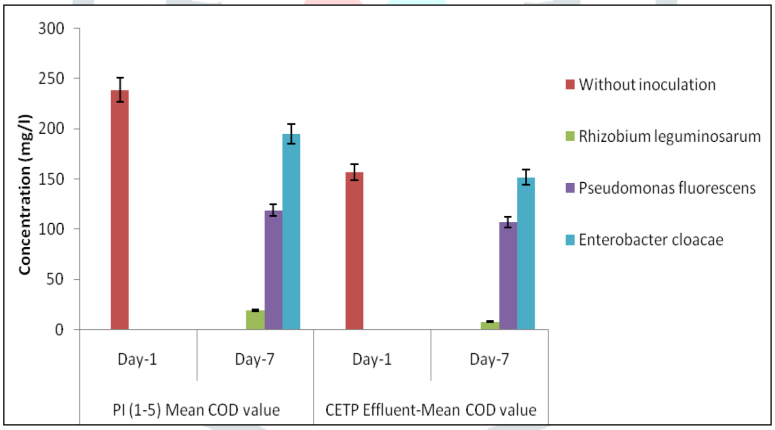

Figure-3.10 Variations in COD in effluent of selected pharmaceutical industries before and after treatment (CETP) with different bacterial strains

The observations of Fig.3.10 indicated that COD concentration decreased from an average value of 238.72mg/l in untreated PI samples to 156.8mg/l in treated pharmaceutical effluent, without inoculation with bacterial strains on Day-1. On Day-7 of biodegradation assay with microbial strains, COD concentration in PI decreased on an average to 19.2mg/l (*Rhizobium leguminosarum*), 119mg/l (*Pseudomonas fluorescens*) and 195mg/l (*Enterobacter cloacae*) while in PI-CETP, it decreased to 8.0mg/l (*Rhizobium leguminosarum*), 107.2mg/l (*Pseudomonas fluorescens*) and 152mg/l (*Enterobacter cloacae*) respectively. The investigations revealed that biological treatment with *Rhizobium leguminosarum* was most effective in causing COD reduction in comparison to *Pseudomonas fluorescens* and *Enterobacter cloacae.* Significant variations were recorded in COD concentration after biological treatment with microbial strains at 5% level of significance ($P<0.05$). Similar results were obtained on treatment of industrial effluents with bacterial strains to reduce BOD and COD [24-26].

V. CONCLUSION

It is concluded that Biological treatment of Pharmaceutical effluents and CETP treated effluent samples, after inoculation with *Enterobacter cloacae* on Day-3 and Day-7, was most effective in reducing BOD as compared to other microbial strains i.e. *R. leguminosarum and P. fluorescens.* However maximum COD reduction of these samples was recorded with bacterial strain *Rhizobium leguminosarum.* Therefore, it's advisable for the pharmaceutical industries to opt out for biological reduction methods before disposing of the treated effluent in to any adjacent water body or in to any open field to control the environmental pollution.

REFERENCES

[1] Danida. 1998. Environmental Profile of Mwanza Municipality, Mwanza Municipal Council. CBEM Kampsax.

[2] Davies. B. R. and Walker. K. F. 1986. The Ecology of River Systems. John Wiley & Sons, New York.

[3] Perry. R. H., Green. D. W. and Maloney. J. O. 2007. Perry's Chemical Engineers. Handbook, Eighth Edition.

[4] Mahdieh. E. and Amirhossein. M. 2009. Water Quality Assessment of Bertam River and its Tributaries in Cameron Highlands, Malaysia. World Applied Sciences Journal, **7**(6):769-776.

[5] Chanti Babu Patneedi and Prasadu. K. D. 2015. Impact of Pharmaceutical Wastes on Human Life and Environment. Rasayan J. Chem. 8(1):67-70.

[6] Ramesh. P., Abraham. K., Suresh. B. and Damodharam, T. 2016. Analysis of Physico-chemical Characteristics of Industrial Effluents in Tirupati, Andhra Prades. Journal of Research in Environmental and Earth Science, **2**(9):01-06.

[7] Simerjit Kaur & Jasvir Kaur. 2015. Assessment of Seasonal variations in oxygen demanding parameters (DO, BOD, COD) along Sirhind Canal passing through Moga, Punjab, India. International Journal of Innovative Science, Engineering & Technology. 2(5):697-700.

[8] Lapara. T. M., Nakatsu. C. H. , Pantea. L.M. and Alleman. J. E. 2001. Aerobic Biological Treatment of a Pharmaceutical Process Wastewater: Effect of Temperature on COD Removal and Bacterial Community Development. Water Research. 35:4417-4425.

[9] Crane. M., Watts. C. and Boucard.T. 2006. Chronic Aquatic Environmental Risks from Exposure to Human Pharmaceuticals. Science of the Total Environment. 367:23-41.

[10] Quinn. B., Gagne. F. and Blaise. C. 2008. An Investigation into the Acute and Chronic Toxicity of Eleven Pharmaceuticals (and their solvents) found in Wastewater Effluent on the Cnidarian, Hydra Attenuate. Science of the Total Environment. 389(2-3):306-14.

[11] Gaworecki. K. M. and Klaine. S. J. 2008. Behavioral and Biochemical Responses of Hybrid Striped Bass during and after Fluoxetine Exposure. Aquatic Toxicology. 88:207-213.

[12] Stanley. J. K., Ramirez. A. J., Chambliss. C. K. and Brooks. B. W. 2007. Enantiospecific Sublethal Effects of the Anti-depressant Fluoxetine to a Model Aquatic Vertebrate and Invertebrate. Chemosphere. 69(1):9-16.

[13] Brooks. B. W., Turner. P. K., Stanley. J. K., Weston. J. J., Glidewell. E. A., Foran. C. M., Slattery. M., La Point T. W. and Huggett, D. B. 2003. Waterborne and Sediment Toxicity of Fluoxetine to Select Organisms. Chemosphere, 52(1):135-142.

[14] Nentwig. G. 2007. Effects of Pharmaceuticals on Aquatic Invertebrates. Part II: The Antidepressant Drug Fluoxetine. Archives of Environmental Contamination and Toxicology. 52:163-170.

[15] Pomati. F., Castiglioni. S., Zuccato. E., Fanelli. R., Vigetti. D., Rossetti. C., & Calamari. D. 2006. Effects of a Complex Mixture of Therapeutic Drugs at Environmental Levels on Human Embryonic Cells. Environmental Science and Technology. 40(7):2442-2447.

[16] Livingston. A. G. 1992. A Novel Membrane Bioreactor for Detoxifying Industrial Waste Water.II: Biodegradation of 3-chloronitrobenzene in an Industrial Produced Waste Water. Biotechnology and Bioengineering. 41:927-936.

[17] Alberti. F., Bienati. B., Bottino. A., Capannelli. G. 2007. Hydrocarbon removal from industrial waste water by hollow fibre membrane bioreactors. Desalination. 204:24-32.

[18] Rohit. K. C. and Ponmurugan. P. 2013. Physico-chemical Analysis of Textile, Automobile and Pharmaceutical Industrial Effluents. International Journal of Latest Research in Science and Technology. 2(2):115-117.

[19] APHA. 2000. Standard methods for the examination of water and waste water.

[20] Singh. D., Singh. V. and Agnihotri. A. K. 2013. Study of Textile Effluent in and around Ludhiana District in Punjab, India. International Journal of Environmental Science. 3(4):1271-1278.

[21] Kumar. R. S., Swamy. R. N., Ramakrishnan. K. 2001. Pollution Studies on Sugar Mill Effluent-Physico-chemical Characteristics and Toxic Metals. Pollution Research. 20(1):93–97.

[22] Rafeeq. M. A., Khan. A. M. 2002. Impact of Sugar Mill Effluents on the Water Quality of the River Godavari near Kandakurthi Village, Nizamabad District, Andhra Pradesh. Journal of Aquatic Biology. 17(2):33–35.

[23] Bhatia. D., Sharma. N. R., Kanwar. R. and Singh. J. 2018. Physicochemical Assessment of Industrial Textile Effluents of Punjab (India). Applied Water Science. 8:83.

[24] Adebayo. G. B. and Kolawol. O. M., Ajijolakewu. A. K. and Andulrahaman. S. O. 2010. Assessment and Biological Treatment of Effluent from a Pharmaceutical Industry. Annals of Biological Research. 1(4):28-33.

[25] Surti. H. S. 2016. Physico-chemical and Microbial Analysis of Waste Water from Different Industry and COD Reduction Treatment of Industrial Waste Water by using Selective Microorganisms. International Journal of Current Microbiology and Applied Sciences. 5(6):707-716.

[26] Ibegbulam-Njoku. P. N., Chijioke–Osuji. C.C. and Imo. E. O. 2013. Physicochemical Characteristics and Biodegradation of Pharmaceutical Effluent. International Journal of Scientific & Engineering Research. 4(11):29-38.