Scientific Papers. Series E. Land Reclamation, Earth Observation & Surveying, Environmental Engineering. Vol. III, 2014
Print ISSN 2285-6064, CD-ROM ISSN 2285-6072, ISSN-L 2285-6064

BIOREMEDIATION OF CIMUKA RIVER STREAM BY THE CONSORTIUM OF *BACILLUS COAGULANS, BACILLUS PUMILUS, BACILLUS SUBTILIS, PAENIBACILLUS AMYLOLITICUS* AND *NITROSOMONAS* SP.

Bambang PRIADIE[1], Rebiet Rimba RINJANI[1], Zastya Marisa ARIFIN[1], Ratu SAFITRI[2] , Nurhidayah IMANDA[2]

[1] Research and Development Institut for Water Resources, Department of Public Works, Indonesia
[2]Dept. Of Biology, FMIPA, Universitas Padjadjaran, Indonesia

Corresponding author email: bpriadie@yahoo.com

Abstract

Research has been conducted to determine the ability of the bacterial consortium in bioremediation of Cimuka River in terms of reduction in BOD, COD, Ammonia, and TSS. The method used in this study is experimental method with Complete Randomize Design (CRD) consist of 2 factors and 3 replications. The first factor were the type of indigenous of bacteria onsortium (K), the consortium of bacteria are: (k_1) microbial consorsium Bacillus coagulans, Bacillus pumilus, Bacillus subtilis dan Nitrosomonas sp., (k_2) microbial consorsium Bacillus pumilus, Bacillus subtilis, Paenibacillus amylolyticus dan Nitrosomonas sp., (k_3) microbial consorsium Bacillus coagulans, Bacillus subtilis, Paenibacillus amylolyticus dan Nitrosomonas sp., (k_4) sterilized water river without indigenous bacterium neither additional bacterium., (k_5) water river with existing indigenous bacterium. The second factor is Retention time (T) consist of 8 stages, i.e.: (t_0) day- 0, (t_1) day- 4, (t_2) day-8, (t_3) day-12, (t_4) day-16, (t_5) day- 20, and (t_6) day- 25. The results obtained indicated that consortium of Bacillus subtilis, Nitrosomonas, Bacillus pumilus and Bacillus circulans (k1) is capable of effectively reducing BOD values of 77.3 %, COD 76.7 %, Ammonia 81.8%, and TSS 79.5%.

Key words*: bioremediation*, *microbial consortium, BOD, COD, ammonia, TSS*

INTRODUCTION

Bioremediation is a technique that involves the use of organisms to solve problems or environmental pollution using biological organisms, for example, to overcome the soil or groundwater contamination. In other words it is a technology to remove pollutants from the environment so as to restore the original natural environment and prevent further pollution. Currently, bioremediation has grown on waste water treatment containing chemical compounds that are difficult to degrade and is usually associated with industrial activity, such as heavy metals, petroleum hydrocarbons, and halogenated organic compounds such as pesticides and herbicides (Tortora, 2010), and nutrients in the water such as nitrogen and phosphate in stagnant waters (Great Lakes Bio Systems. Inc.. Co. Orb - 3.com).

In fact, at this time, a common flocculant Alum raw material to degrade pollutants river water could have been replaced with the microorganism as bioflocculant isolated from activated sludge process and is known to decrease by 84-94 % turbidity (Buthelezi, SP, et al, 2009). In addition, the reliability of microbes including bacteria, fungi, and protozoa in wastewater treatment and its role in maintaining the ecological balance has been elaborated (Gerardi, 2006). Another example is the bacterium Pseudomonas can reduce the toxicity of detergent on the river ecosystem.. For these reason, indigenous bacteria from Cimuka River was isolated to degrade liquid waste within Cimuka River water that contain industrial and domestic wastewater. Cimuka River which is one part of a sub Regional 3 River Drainage: Cigugur - Cimahi - Cimuka flowing through industrial areas are concentrated in South Cimahi dominated by textiles, clothing and leather industries.

MATERIALS AND METHODS

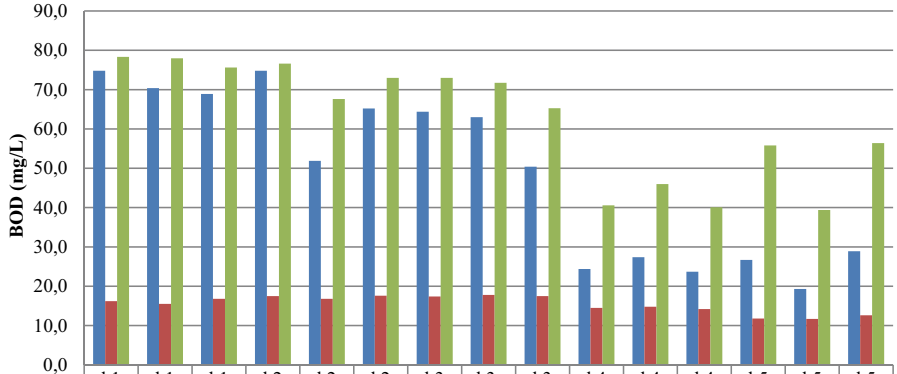
The method used in this study is experimental method with Complete Randomize Design (CRD) consist of 2 factors and 3 replications. The first factor were the type of indigenous of

bacteria consortium (K), the consortium of bacteria are: (k_1) microbial consorsium *Bacillus coagulans, Bacillus pumilus, Bacillus subtilis dan Nitrosomonas sp.*, (k_2) microbial consorsium *Bacillus pumilus, Bacillus subtilis, Paenibacillus amylolyticus* and *Nitrosomonas sp.,* (k_3) microbial consorsium *Bacillus coagulans, Bacillus subtilis, Paenibacillus amylolyticus* and *Nitrosomonas sp*., (k_4) sterilized water river without indigenous bacterium neither additional bacterium., (k_5) water river with existing indigenous bacterium. The second factor is Retention time (T) consist of 8 stages, i.e.: (t_0) day- 0, (t_1) day- 4, (t_2) day-8, (t_3) day-12, (t_4) day-16, (t_5) day- 20, and (t_6) day- 25.

RESULTS AND DISCCUSSION

Reduction of BOD value by bacterial consortium

The decrease in BOD values are influenced by the activity of bacteria through the process of optimizing the continuity of contact time of bacteria with organic matter and the presence of oxygen in wastewater. The nature of the bacteria that spread (disperse) in water (through aeration) optimize suspected bacterial contact with organic materials. Aeration is provided, in addition to supplying oxygen to function, it also serves the wastewater stirring continuously, thus increasing the chances of bacterial contact with organic materials. In general, bacterial consortium (k1) resulted in decreased levels of BOD is highest with an average of 77.30 % (Duncan 's Multiple Range test) on day 20. This suggests that there has been interaction between bacterial consortium B. coagulans, B. pumilus, B. subtilis and Nitrosomonas sp overhaul of organic matter in industrial wastewater, causing a decrease in the levels of BOD in large numbers. B. coagulans is a bacterium that is able to remodel lipids because it produces the enzyme lipase (Hasan et al 2006). B. coagulans can be obtained from soil, activated sludge (Kotay and Das, 2007). B. subtilis is a type of bacteria producing cellulase enzyme that functions decompose cellulose in the waste industry. Decomposition is the process of splitting cellulose anhydroglucose polymers into simpler molecules such as cellobiose, selotriosa, glucose monomers as well as CO2 and water (Lynd et al, 2002). B. subtilis is a bacterium that is most widely used for the production of enzymes such as amylase, and protease (Kosim, 2010).



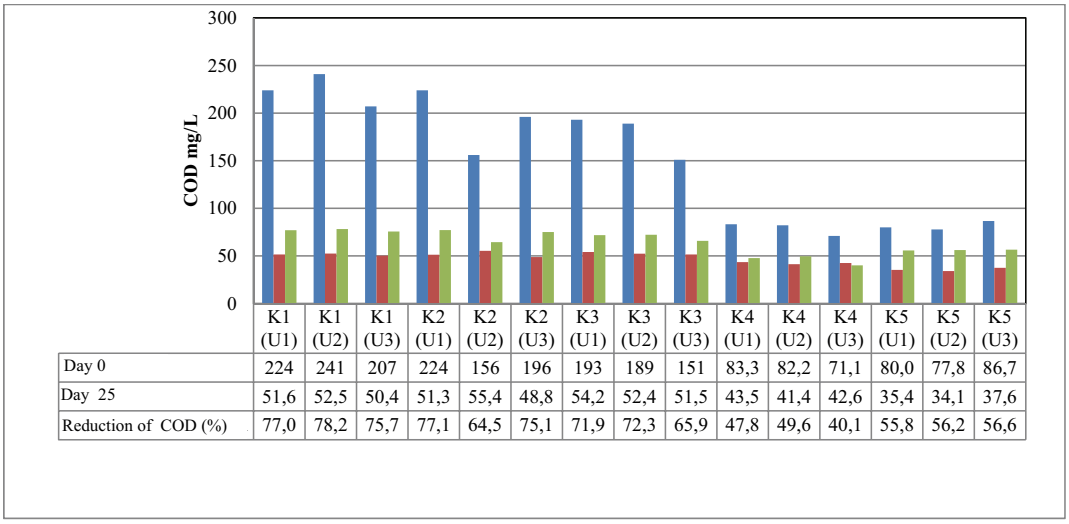
	k1 (U1)	k1 (U2)	k1 (U3)	k2 (U1)	k2 (U2)	k2 (U3)	k3 (U1)	k3 (U2)	k3 (U3)	k4 (U1)	k4 (U2)	k4 (U3)	k5 (U1)	k5 (U2)	k5 (U3)
Day 0	74,8	70,4	68,9	74,8	51,9	65,2	64,4	63,0	50,4	24,4	27,4	23,7	26,7	19,3	28,9
Day 25	16,2	15,5	16,8	17,5	16,8	17,6	17,4	17,8	17,5	14,5	14,8	14,2	11,8	11,7	12,6
Reduction of COD (%)	78,3	78,0	75,6	76,6	67,6	73,0	73,0	71,7	65,3	40,6	46,0	40,1	55,8	39,4	56,4

Figure 1. Decreasing Levels of BOD (%) During Biodegradation Process of Cimuka River Water

Mean of BOD levels after the end of the bioremediation process is 15.5 mg/l (Figure 1) has met the Liquid Waste Quality Standard BOD of (50 mg/l). Thus, decreasing of BOD levels by the addition of bacterial consortium showed that the bacteria used plays a role in lowering levels of BOD.

Reduction of COD value by bacterial consortium

The ability of the consortium (k1) was the highest in the decreasing levels of COD, average of 76.97 % (Duncan 's Multiple Range Test) due to the ability of these microbes to produce three enzymes that can break down the organic matter content of river water. In this consortium, Bacillus sp has an important role in decomposing of lipids and cellulose. According to Roheim (2011), Bacillus sp. can degrade lipids into glycerol and fatty acids that can be easily digested by microbes as a source of nutrients. Industrial wastewater contains organic matter is very high, one of which is the content of cellulose. Cellulose is a polysaccharide that is built by a glycosidic bond that is stable and not easily interrupted, causing the natural decomposition process longstanding industrial wastewater. To speed up the process of decomposition of the waste is necessary cellulase enzymes able to break glycosidic bonds (Howard, 2003). B. coagulans and Bacillus B. subtilis known is a group that is able to produce high amounts of cellulase enzymes, so the use of both of these bacteria strongly supports reform of industrial liquid waste organic materials. On the other hand Nitrosomonas sp. able to decompose ammonia into nitrogen compounds much simpler. Thus it can be said that the more organic materials are broken down by bacteria, the greater the amount of reduction in COD of industrial wastewater. The study results indicated that the average COD concentration obtained is 51.5 mg/l (Figure 2) that already meet the effluent quality standard (100 mg/l). Therefore, decreasing of COD levels by addition of bacterial consortium showed that the bacteria used plays a role in lowering levels of. COD.



	K1 (U1)	K1 (U2)	K1 (U3)	K2 (U1)	K2 (U2)	K2 (U3)	K3 (U1)	K3 (U2)	K3 (U3)	K4 (U1)	K4 (U2)	K4 (U3)	K5 (U1)	K5 (U2)	K5 (U3)
Day 0	224	241	207	224	156	196	193	189	151	83,3	82,2	71,1	80,0	77,8	86,7
Day 25	51,6	52,5	50,4	51,3	55,4	48,8	54,2	52,4	51,5	43,5	41,4	42,6	35,4	34,1	37,6
Reduction of COD (%)	77,0	78,2	75,7	77,1	64,5	75,1	71,9	72,3	65,9	47,8	49,6	40,1	55,8	56,2	56,6

Figure 2. Decreasing Levels of COD (%) During Biodegradation Process of Cimuka River Water

Reduction of ammonia level by bacterial consortium

Nitrification process is one of methods apply to overcome the accumulation of ammonium in the wastewater. Nitrification process may occur due to the activity of ammonium oxidizing bacteria and nitrite or also called nitrifying bacteria. Nitrifying bacteria was applied in the biodegradation process in this study is derived from the genus Nitrosomonas sp.

Bacteria of the genus Nitrosomonas sp. a bacterium capable of oxidizing ammonium to nitrite. An autotrophic bacteria are those bacteria which use CO2 as a carbon source and can grow at a pH optimum ranging from 7.5 to 8.5 (Ratledge, 1994). The addition of the genus Bacillus bacteria also have an important role to decrease ammonia levels. Edwards (2011), states that the genus Bacillus can reduce levels of ammonia due to its ability to oxidize ammonia content in the waste and to utilize these in heterotrophic ammonia as a source of nutrients. Organic compounds break down into simple compound can be used as a source of nutrition for nitrifying bacteria in the wastewater industry. According to Adhi (2008), based on the nitrogen cycle, nitrification bacteria are bacteria found in wastewater containing organic compounds, so the nitrifying bacteria that gets the extra nutrients able to work more effectively in the decomposition of ammonia in the wastewater industry. The addition of bacteria Nitrosomonas sp. assist the process of nitrification and ammonia monooxygenases produce enzymes to break down ammonia into nitrite with the help of oxygen. Initially the ammonia is oxidized to hydroxylamine compound by enzymes produced ammonia monooxygenases Nitrosomonas sp

The decrease in ammonia also occurs in sterile waste control (k4) and non-sterile waste (k5). In the sterile waste (k4) a decrease in ammonia levels may be due to evaporation during the biodegradation process is aided by the presence of aeration. Meanwhile, in the non-sterile wastewater (k5) are also given aeration, decreased levels of ammonia can be caused due to the persistence of indigenous bacteria in the waste can reduce levels of ammonia. As is known, Nitrosomonas sp. spread on fresh water, sea water and soil (Holt et al., 1994). The percentage decrease in ammonia levels high enough on industrial wastewater that has been treated and control allegedly due to change ammonia into a gas that is released into the air through evaporation process. Figure 3 shows the curve of changes in levels of ammonia and its the percentage decline during river water biodegradation process

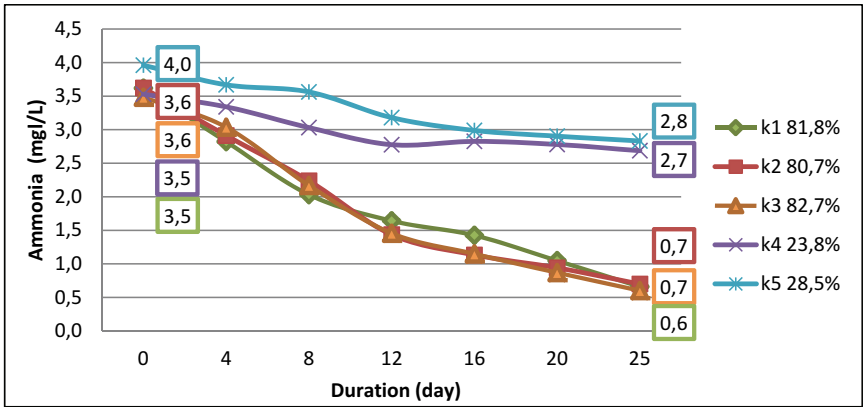


Figure 3. Curve of changes in levels of ammonia and its the percentage decline during During Biodegradation Process of Cimuka River Water

Consortium of bacteria (k1) can reduce ammonia levels by 81.8% This shows that there is a synergistic interaction between B. coagulans, B. pumilus, B. subtilis and Nitrosomonas sp. resulting in decreasing of ammonia levels is high. Sastrawijaya (2000) consider that the concentration of TSS in the water generally consists of phytoplankton, zooplankton, human waste, animal waste, sludge, crop residues and animal, as well as industrial waste. The materials were suspended in natural waters are not toxic, but if excessive amounts can increase turbidity which further inhibits sunlight penetration into the water column (Effendi, 2000).

The decrease in TSS by bacterial consortium (k1) B.coagulans, B. pumilus, B. subtilis and Nitrosomonas sp. not significantly different from the bacterial consortium (k2) B. pumilus, B. subtilis, P.amylolyticus and Nitrosomonas sp. resulted in decreased levels of TSS best. This can be caused by the ability of the bacteria contained in the consortium is able to remodel the organic material in the suspended substances in the wastewater industry. B. coagulans and B. subtilis has a high cellulolytic ability, thus containing cellulose dissolved solids will be described. Besides B. coagulans and B. the genus Bacillus subtilis includes groups that have the ability to decipher crude fiber and lignin are difficult to decompose lignin and delignification process through hydrolysis of cellulose so solid that dissolved organic matter in the form of lignin, lipids, and cellulose in industrial wastewater can be reduced.

Figure 4 shows a decreasing of TSS level, during biodegradation process of Cimuka River Water by bacterial consortium (k1) 79.5 % and effectively occurred on day 25.

The survey results revealed that the average TSS concentration obtained is 131.8 mg / l TSS has met the quality standards of industrial wastewater is 150 mg / l. Thus, a decrease in TSS levels in the addition of bacterial consortium showed that the bacteria used effectively contribute to lower levels of TSS.

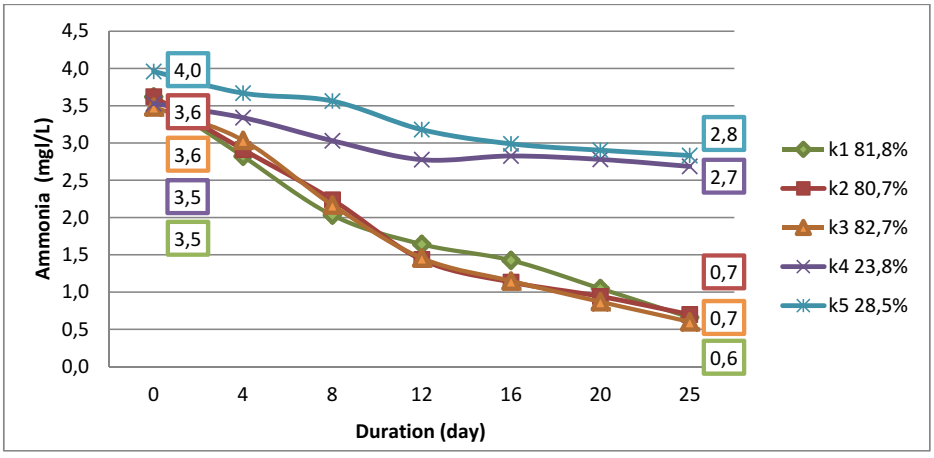


Figure 4. Changes in levels of TSS during During Biodegradation Process of Cimuka River Water

CONCLUSIONS

1. Consortium (k1) consisting of B. coagulans, B. pumilus, B. subtilis and Nitrosomonas sp. can reduce levels of contaminants of Cimuka River streams 77.3% of BOD, COD 76.97%, 81.8% and TSS Ammonia 79.5%.
2. Consortium of bacteria (k1) may reduce organic wastes 70% of the initial content of industrial wastewater streams Cimuka River streams

REFERENCES

Adhi, I. 2008. *Daur Biogeokimia*. http://gurungeblog.wordpress.com/2008/11/17/ daur biogeokimia/ diakses pada 10 Oktober 2012 pukul 15.00 WIB

Buthelezi, S. P., Olaniran, A. O. and Pillay, B., 2009, *Turbidity and microbial load removal from river water using bioflocculants from indigenous bacteria isolated from wastewater in South Africa,* African Journal of Biotechnology Vol. 8 (14), pp. 3261-3266, 20 July, 2009. ISSN 1684–5315 © 2009 Academic Journals

Edwards, V.A. 2011, *The Nitrogen Cycle-Control Ammonia and Nitrite in Ponds, Lakes Lagoons, Rivers and Wastewater Treatment*. CEO Alken-Murray Corporation.

Effendi, H. 2003, *Telaah Kualitas Air bagi Pengelolaan Sumber daya dan Lingkungan Perairan*. Penerbit Kanisius. Yogyakarta.

Great Lakes Bio Systems. Inc..co Orb-3.com/LakeAndPond Orb-3 Professional Enzymes & Bacteria are the total solution.

Gerardi, M.H. 2006. *Wastewater Bacteria.* New Jersey. John Willey

Gerard J. Tortora, Berdell R. Funke, Christine L. Case. - 10th ed, 2010, *Microbiology: an introduction*. Microbial Technology 39.

Hasan, F., A. A. Shah dan A. Hameed. 2006. *Industril applications of microbial lipases.* Enzyme and Microbial Technology 39.

Holt, G., Kreig, N.R., Sneath, P. H. A., Staley, J. T., Williams, S. T. 1994. *Bergey's Manual of Determinative Bacteriology, 9th edition*. Lippincott Williams and Wilkins, Philadelphia, USA.

Howard, R.L, E. Abotsi, and E.L.J.V. Rensburg, S. 2003. African Journal of Biotechnol, 602-619 p.

Kosim M, Putra SR. 2010. *Pengaruh Suhu Pada Protease dari Bacillus subtilis.*Jurusan Kimia FMIPA ITS Surabaya.

Kotay, S.M. and D. Das. 2007. *Microbial hydrogen production with Bacillus coagulans IIT-BT S1 isolated from anaerobic sewage sludge*. Bioresources Technology. 98: 1183– 1190.

Lynd, L.R., P.J. Weimer, W.H. van Zyl, and I.S. Pretorius. 2002. *Microbial cellulose utilization: fundamentals and biotechnology*. Microbiology and Molecular Biology Reviews 66 (3): 506-577

Ratledge, C. 1994. *Biochemistry of Microbial Degradation*. Kluwer Academic Publishers, London.

Roheim, J. R. 2011. Low *Cost Automated On Site System For Growing And Dispending Vegetative Bacteria.* http://www. repository.usu.a.id/bitstream/ 123456789/19890/4/Chapter%20II.pdf diakses pada 10 Oktober 2012.

Sastrawidana,I.D.K. 2008. *Pengolahan Limbah Tekstil Menggunakan Biofilm Konsorsium Bakteri Pada Reaktor Dengan Sistem Anaerobik-Aerobik*. Disertasi Program Studi Pengelolaan Sumbedaya Alam and Lingkungan. IPB.