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BIOREMEDIATION- POLLUTED SURFACE WATER TREATMENT

BIOREMEDIACJA: METODA UZDATNIANIA ZANIECZYSZCZONYCH WÓD POWIERZCHNIOWYCH

Many countries are currently faced with water scarcity. The causes are due to climate change, higher water consumption, lack of water resources, and especially an increase in water contamination. In developing countries, untreated sewage water is directly discharged to surface water. The self-purification process in the natural surface system can no longer handle itself, and as a result, the surface water system is overloaded and contaminated. Our research focuses on enhancing the self purification process by adding microorganisms into the surface water. From our microorganisms collection, some microorganisms with high growth rates and high adaptability to the normal environment were selected, such as Bacillus subtilis CN2, B. megaterium N20, B. licheniformis P32, Chromobacterium violaceum P2, Ochrobactrum S5, Rhodococcus erythropolis S6. These microorganisms were grown in lab settings, and the biomass was mixed with zeonit to create a product with a cell density of $2.4x10^9$ CFU/g. This product was then used to treat polluted water with an initial density of 10^3 CFU/ml at both bench scale and full scale experiments. Van Lake water, which has BOD_5 of 116 mg/L, was chosen for the bench scale experiments. After seven days, the removal efficiencies of COD, BOD_5, TSS, T-N and T-P at bench scale are 85.13%, 81.65%, 86.67%, 76% and 72.11%, respectively. For full scale conducted at 1.08 ha SaiDong Lake with COD of 82 mg/L, the removal efficiencies of COD and BOD_5 when using the product with aeration were 53% and 51%, respectively.

1. Introduction

Many countries in the world are now struggling with inadequate and contaminated water supply sources. There are several reason, such as climate change causing the sea water invasion of surface water, higher water consumption due to industrialization, lack of water resources due to the uncontrolled exploitation of underground water, and especially an increase in surface water contamination. In developing countries, untreated sewage water is directly discharged to surface water. The self- purification process in the natural surface system can no longer handle itself, and as a result, the surface water

system is overloaded and contaminated. This impacts not only water supply sources but also aquiculture. The treatment of polluted surface water should not contain a disinfection step. Therefore, one solution to solve this problem is to promote the self-purification process by using a number of microorganisms. Those microorganisms must have following characteristics: high capacity to convert contaminant compound into biomass as well as degrade them into smaller compounds or to final products as CO_2 and H_2O, and be harmless to the environment. Those microorganisms will be the first consumer in the food chain and will reduce volumes of sewage sludge produced while also removing associated odours (Szymanski, et al. 2003).

Recently, Effective Microorganisms (EM) Technology has emerged as a promising technology for contaminated environment treatment. The EM concept and technology was developed and introduced by Professor Higa from the of University of Ryukyus, Okinawa, Japan in the 1970's. The EM were defined as groups of microorgan ism which include the coexistence of effective, beneficial and nonpathogenic of both anaerobic and aerobic (Higa, et al. 1995). These microorganisms have the ability to decompose organic compounds. Since EM Technology has been used, it is said that EM do not have any negative effect on the environment, humans, or animals. Moreover, EM can grow naturally in the environment. Therefore, EM Technology is considered a natural and environmentally friendly technology. The main species of EM are lactic acid bacteria, photosynthesis bacteria, yeast, actinomyces and fermenting fungi (Szymanski, et al. 2003). During growth, these EM species produce and secrete organic acids, enzymes, antioxidants, and metallic chelates in the culture and enhance the solid –liquid separation (Higa & Chinen 1998). Szymanski, et al. (2003) used effective microorganisms (EM) in waste water treatment plant and the results showed that there is a clear benefit from the use of EM in wastewater.

Bacillus genus have several interesting characteristics for use in waste water treatment, such as they produce several enzymes: amylase, xylanase, biosorption ions, producing spore,…. Many species belong to *Bacillus* genus have been used for waste water treatment (Nurba, et al. 2002, Obeta, et al. 2008). In this study, some species from the *Bacillus* genus from a natural source were used, such as *Bacillus subtilis* CN2, *B. megaterium* N20, *B. licheniformis* P32. Among them, *B. subtilis* CN2, isolated from Vietnamese fish sauce have some characteristics: produce the enzymes collagenase (Tran, et al. 2002, 2005) and protease (Uchida, et al. 2004), can grow in a wide range of temperature, from 20-40^{o}C, and reduce COD and BOD_5 in polluted water treatment (Tran, et al. 2006).

In this paper we present results from using microorganisms to treat several polluted surface water surrounding Hanoi.

2. Materials and method

2.1. Biological product

Microorganisms used in this study are from the collection of Department of Microbiology and Genetic engineering, Hanoi University of Technology. *B. subtilis* CN2 was isolated from Vietnamese fish sauce (Tran, et al. 2002) and others were isolated from surface water, such as surface water and soil surrounding the city of Hanoi (Nguyen, et al. 2006, Tran, et al. 2006).

Those microorganisms were cultured at optimum conditions (data not shown) to harvest biomass. Then the biomass were mixed with zeonit to create a product with cell density of $2.4x10^9$ CFU/g, humidity of 9% and this product was used to treat polluted surface water. The product was added with initial density of 10^3 CFU/ml at both bench scale and full scale experiments.

2.2. Measurement parameters

COD was determined using spectrophotometer HACH 2400 after samples were heated 150^{o}C for 120 min.

BOD_5, total suspended solid (TSS), total Nitrogen (TN) and total phosphate were measured regularly by standard methods.

2.3. Bench scale using Van lake water

The water from Van Lake was taken and in bench scale with 3 l bottle containing 2 l of the water. The experiment was carried out in 3 sets bottles:

Set of bottle 1: containing only lake water

Set of bottle 2: containing lake water and the biological product

Set of bottle 3: containing lake water and biological product with stirring speed of 50 rpm/min.

2.4. Full scale in Saidong Lake

Saidong Lake is located north of Hanoi with 1.08 ha. Nearby Saidong Lake is a small lake with 0.18 ha and the two lakes are separated from each other by a small dike. Domestic waste from local residents is discharged directly into Saidong Lake. Before the experiment was carried out, the lake was thoroughly polluted and does not have aquiculture. Saidong Lake was used for this experiment and the nearby small lake was used as a control. Amount of 3 kg of the product was applied in Saidong Lake and every 5 days samples were taken to analyze all parameters for 40 days. After that, another amount of 3 kg of the product was applied in Saidong lake and stirred by fludization aeration mixing apparatus (Nguyen Van Cach WO/2009/052535). Every 5 days samples were taken to analyze all parameters.

Fig. 1. Location of Saidong Lake and small lake in Hanoi

3. Results and disccussion

3.1. Bench scale using Van Lake water

The experiments were carried out in a 3 l bottle containing 2 l of Van Lake water. In this experiment, set of bottle 1 containing only Van Lake water was used as a control sample. Set of bottle 2 contained Van Lake water and the product and set of bottle 3 contained Van Lake water, the product and stirring with 50 rpm/min. Samples were taken every day to analyze all parameters.

The results shown in fig 2a indicated that COD in all bottles decreased every day during the 7 days experiment and in the set bottle 3, COD value decreased more rapidly, and the self purification for removing COD in the control bottle was 30.41%. The efficiency for treatment COD in the bottle 1, 2, and 3 were 85.1%, 56.8% and 30.41%, respectively.

Fig 2b shows that the efficiency to reduce BOD_5 in bottle 1, 2 and 3 were 28.44%, 53.21% and 81.65%, respectively. The decreasing of contaminants observed through COD and BOD_5 values can be explained several ways, such as that the contaminants were converted into biomass or degraded into the final products. By supplying oxygen through stirring, the efficiency of COD and BOD_5 removal can be increased more than 30% compared to the results without stirring.

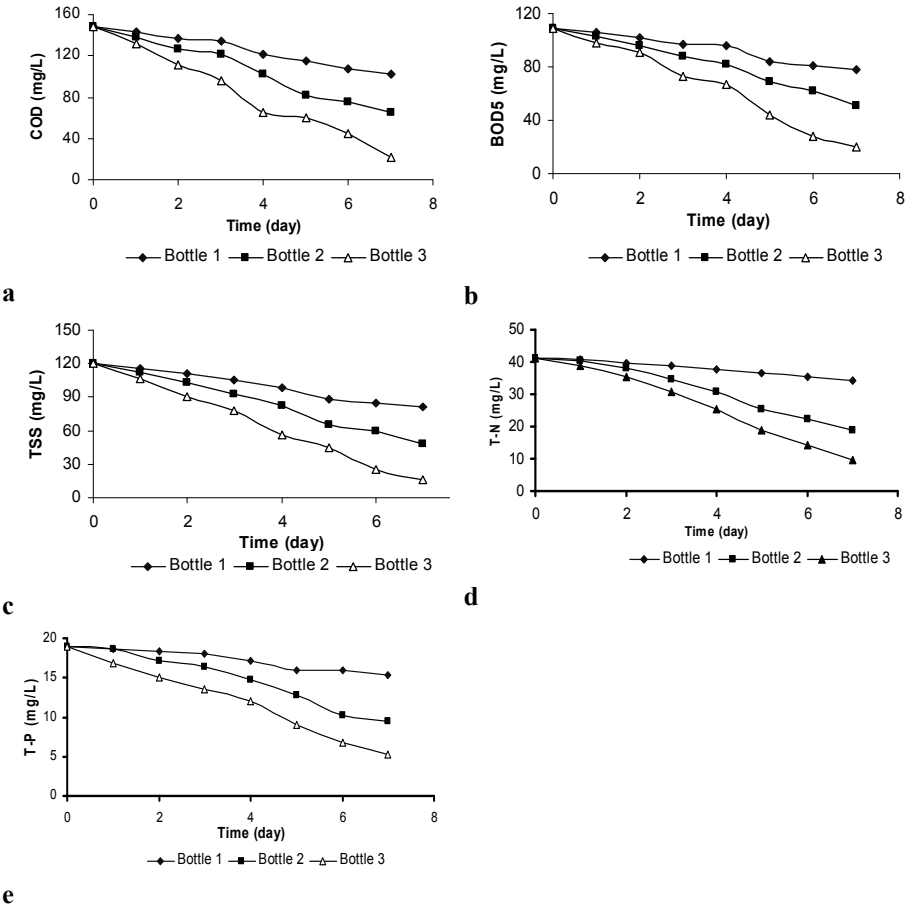

Fig. 2. Changing parameters during experiment a) COD b)BOD_5 c) TSS d) T-N and e) T-P

Fig 2c showed that the efficiencies of TSS reduction in bottle 1, 2 and 3 were 32.5%, 60% and 86.67%, respectively. It was simple and clear for observation of the TSS reduction in the bottles. The results coincide with the suggestion of Freitag, et. al. (2000), that introducing EM into aerobic treatment facilities helps reduce the unpleasant byproducts of this decomposition and also reduce the production of residual sludge.

Fig 2d and 2e showed that T-N and T-P were removed better in bottle 3 with efficiency of 76.33% and 72.11% respectively compared to the others.

3.2. Full scale experiments Saidong Lake

Before treatment, Saidong Lake and small lake have COD value of 82 mg/l and 88 mg/l and BOD_5 value of 60 mg/l and 68 mg/l, respectively. After 40 days of treatment with the product COD reduced 36% and BOD_5 reduced 34%. After that, another amount

of 3 kg of the product was added and stirred by fludization aeration mixing apparatus (Nguyen Van Cach WO/2009/052535). The results in figs 3a and 3b show that the removal efficiencies of COD and BOD_5 when using the product with aeration were 42% and 38%, respectively. Comparison with COD and BOD_5 before treatment, efficiency of reducing COD and BOD_5 were 52% and 51%, respectively. Interestingly, fish meat from Saidong Lake before treatment had a very strange smell but after treatment fish meat had a normal smell (data not shown).

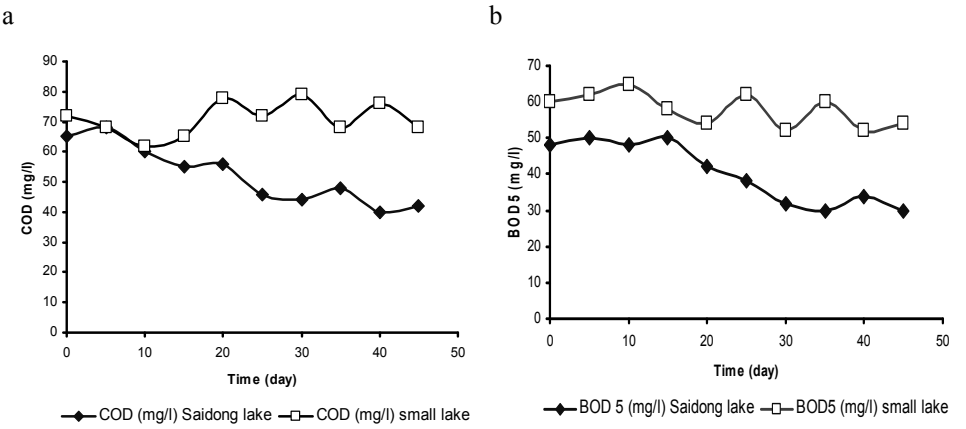

Fig. 3. Changing parameters during experiment in Saidong Lake.

As we know, all organic materials coming to wastewater originate from plants, animals or synthetic organic compounds via a number of routes including human wastes, detergents, and industrial sources. Currently, in biological wastewater treatment processes (either municipal or domestic on-site), microorganisms play a significant role in converting these materials. Many different organisms live within the wastewater itself, assisting in the breakdown of organic pollutants into smaller compound or to final products as CO_2 and H_2O. Microorganisms play a significant role in the decomposition of organic wastes, however, some microorganisms can cause health concerns to humans. These include bacteria and viruses present in the wastes produced (Harris, et al. 2001). Recently, a concept of Probiotic in Progress (PIP) was proposed by Chrisal Company. In this process, microorganisms are used with the aim of establishing a healthy and stable beneficial microbial community and to control unwanted microorganisms, and it does not use disinfectants. Therefore, the PIP approach has some advantages: it provides a safe solution to control pathogens, a friendly solution to the environment and no resistance can be developed. The probiotic bacteria in the PIP product are members of genus *Bacillus* and they belong to biosafety class 1. Vestraete, et al. (2007) had applied PIP product in the patient room of a hospital and results had showed that pathogenic coliform and methicillin resistant *Staphylococcus aureus* were reduced in a large amount and the environment became a healthier microbiological environment. In this experiment, *Bacillus subtilis* CN2 was isolated from fish sauce, considered as biosafety class 1, and it contributed to reduce COD as well BOD_5 in polluted water treatment (Tran, et al.

2006). In Hanoi in 2008, some people were suffered from disease cause by *Vibio chlorella* and Linhquang Lake water in Hanoi was examed and found to contain *Vibio chlorella*. From characteristics of *Bacillus subtilis* CN2, this strain could be considered as EM. However, inorder to apply to it to control pathogenic microorganisms existing in surface water, more experiment should be carried out.

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