EFFECTS OFAERATION ASSOCIATED WITH APPLICATION OF FREE-FLOATING AQUATIC PLANTS SALVINIACUCULLATAROXB. EX BORY AND AZOLLAPINNATA R. BR.) ON PIG-FARM WASTEWATER TREATMENT

[1]PAKAWAT JANYASUPAB, [2]ARUNOTHAI JAMPEETONG

[1]Master's Degree Program in Biology, Faculty of Science, Chiang Mai University, Thailand
[1,2]Department of Biology, Faculty of Science, Chiang Mai University, Thailand
E-mail:[1]mr_pakawat@hotmail.com, [2]arunothai.2519@gmail.com

Abstract - Pig-farm anaerobic effluent, which consists of high ammonium (NH_4^+) and biological oxygen demand (BOD), but low dissolved oxygen (DO), could be improved to reach standard water quality before drained to natural water bodies. Because of high nutrients contamination, so that phytoremediation is one of the interesting and being considered as a high efficiency method. In this experiment, the pig-farm wastewater was aerated for two weeks. After that, it was divided into two groups: undiluted and 50% diluted wastewater. Then, Salviniacucullata and Azollapinnata were planted to each treatment, including controls, totally six treatments. The water parameters, including dissolved oxygen (DO), Biological Oxygen Demand (BOD_5), pH, Electrical Conductivity (EC), Total Dissolved Solid (TDS), Total Suspended Solid (TSS), Nitrate (NO_3^-), Ammonium (NH_4^+), and Orthophosphate (PO_4^{3-}), were determined before an aeration, at the beginning of the experiment (after an aeration for two weeks), and weekly until the sixth week. It was found that pig-farm wastewater could be improved by an aeration. DO had been increased and this high oxygen had reduced NH_4^+, PO_4^{3-}, EC, TDS, and TSS. Furthermore, these two species have highly rate to take up NH_4^+ (70-90% in one week). They could also slightly reduce $NO_3^-$for 3-9% higher than control in undiluted wastewater during the second and fourth week. Furthermore, S. cucullatacould remove a high number of BOD_5 (77-85%) during a week. In addition, TSS in all treatments had been removed for 50-95%. As a result, S. cucullata and A. pinnata were interesting species to reduce these three parameters in the water treatment systems.

Keywords - aeration, Salviniacucullata, Azollapinnata, pig-farm wastewater treatment

I. INTRODUCTION

On the economic expansion nowadays, the products have to be manufactured to meet the demand and for consumption and exportation. In Thailand, agriculture is a basis activities over a century and it makes a good income for this country as well as foreigners have known it as a kitchen of the world. One of the important activitiesis the livestock production, especially pig farming, since porks are needed for consumers in the country andworldwide. As a result, number of pigs in Thailand has been increased from seven millions to ten millions over ten years[1].Hence, the effluent containing high concentrations of nutrients, which drained to waterbodies, has caused eutrophication and low oxygen dissolved so that aquatic organisms have been affected. Hence, wastewater has to be treated before being discharged.The most popular method to treat this effluent is anaerobic digestion, which methane would be produced and organic nitrogenentered into biogas production tank is transformed into ammonium. Aeration is another option which can improve water quality because higher oxygen could activate micro-organisms to degrade organic matters [2,3] and decrease NH_4^+ concentration through nitrification in order to reduce toxicity to organisms. However, nitrate in the system has been increased and needed to be eliminated in order to decrease the risk of eutrophication [4]. Using constructed wetlands for wastewater treatment are high efficiency, cheap, and eco-friendly [5]. In this system, aquatic plants play important roles to improve water quality that they take up excess nutrients particularly nitrogen and phosphorus,release oxygen into rhizosphere and provide habitat for beneficial microorganisms [6]. From previous studies, many species could treat high organic effluent from livestock activities.Olguin et al. [7]found that ammonium in pig-farm wastewater treated with Salvinia minima Baker and Spirodelapolyrrhiza (L.) Schleidwas completely removedwithin 15 days. Furthermore, Eichhorniacrassipes (Mart.) Solms and Salvinianatans (L.) All. planted in aerating wastewater had higher removal rates for biological oxygen demand (BOD_5), chemical oxygen demand (COD), nitrate-nitrogen (NO_3-N), total Kjeldahl nitrogen (TKN), and phosphate-phosphorus (PO_4^{3-}-P) than unplanted treatment[8].Even though nitrogen and phosphorus is necessary for plant growth and development, high-strength effluent could be toxic to the plants. It might stunt their growth rates, reduce size of leaves and stems, and also disturbnutrients uptake rates [9]. Each species has differentrange to tolerate to high nutrient concentration and also potential to improve water quality, so studying their performance in a real wastewater to treat the high-strength wastewater is necessary forplant selection used in wastewater treatment systems. SalviniacucullataRoxb. ex Bory and Azollapinnata R. Br.are free-floating aquatic plants belonging to family Salviniaceae and Azollaceae, respectively. The plants mostly have fast

growth, adapt well in various environments, and have high efficiency to treat domestic, human, and pig wastes [7,10]. Moreover,Azolla contains symbiosis cyanobacteria Anabaena azollae[11], which is able to fix nitrogen gas from the air and transformed into ammonia, amino acid and protein.These two genera could also use as a high nutrient source to feed animals or fertilizer after treatment [12], so they are interesting to use as a biological treatment for pig-farm wastewater after an aeration mainly to take up nutrients. Therefore, this study aims to evaluate efficiency of these two species to treat aerated wastewater. The results would be beneficial for selecting aquaticmacrophytes for a treatment in constructed wetland systems.

II. MATERIALS AND METHODS

A. Wastewater Sampling

The wastewater was collected from pig-farm belonging to Department of Animal and Aquatic Science, Faculty of Agriculture at Mae Hia campus, Chiang Mai University, Thailand. It was then aerated by air pump continuously for two weeks to reduce ammonium concentration so that the plants would not receive high toxic.

B. Plant Cultures and Experimental Design

S. cucullata and A. pinnata were collected from natural ponds in Chiang Mai and Lamphun provinces, Thailand.The plant stocks was cultivated using a standard growth medium modified from Smart and Barko [13] solution with an addition of 500 µM KNO_3, 500 µM $(NH_4)_2SO_4$, and 100 µM KH_2PO_4. All plants were grown under greenhouse conditions at the Department of Biology, Faculty of Science, Chiang Mai University, Thailand for two weeks. For the experiment, 25L of aerated pig-farm wastewater was transferred into 27L buckets. The treatments were: undiluted and 50% diluted wastewater treated with each species including two controls. Water surface were 50% covered by these two species, while the controls, the water surface was covered by black polyethylene sheet to prevent water evaporation and algae bloom. All the buckets were placed in the greenhouse where light regimes and temperature were 13 h light/11 h dark and $33\pm5^{O}C$: $25\pm5^{O}C$ for six weeks.

C. Water Quality Analysis

Wastewater was analysed before an aeration (Table 1). After a transferring to the buckets, approximately 600 mL wastewater was withdrawn from each buckets and analysed at the beginning of the experiment and weekly until the sixth week. Electrical Conductivity (EC), Total Dissolved Solid (TDS), and pH were measured with a multi-parameter analyzer (CyberScan PC 300, Eutech Instruments Pte Ltd., Singapore). Dissolved Oxygen (DO) was determined by an azide modification method (4500-O C) [14]. Biological Oxygen Demand (BOD_5) was calculated by the differences between DO_0 and DO_5 [14]. Total Suspended Solid (TSS) was measured through a standard method 2540 D [14]. After that, the filtered water samples were analyzed for ammonium(NH_4^+), nitrate(NO_3^-), and orthophosphate(PO_4^{3-}) concentrations. These three nutrients were measured by modified salicylate method (Quikchem Method no. 10-107-06-3; Lachat Instruments, Milwaukee, WI, USA), UV- method [15], and stannous chloride method (4500-P D) [14], respectively. The water qualitymeasurement was also compared to the PCD standard [16].

III. RESULTS AND DISCUSSIONS

After an aeration for two weeks, DO had dramatically increased, from 0.9 to 12.62 mg/L (Table 1).This high level of oxygen had played an important role for nitrifying bacteriaso that NH_4^+concentrations had been reduced for almost ten times during two weeks, however, this increased concentrations of $NO_3^-$from 200 µM to 1,261.5 µM. Furthermore, PO_4^{3-} had been decreased for 25.55%, probably due to a transformation intoinsoluble organic phosphorus [17].This study were similar to Luo et al. [18] who found that NH_4^+ and PO_4^{3-} in aerated pig manure had been 32.3% and 75% reduced, respectively.In addition, an aeration had helped the microorganisms to degrade organic matter[19], so the TDS, TSS, and EC had been decreased for29.77%, 38.85%, and 29.74%, respectively.

When the wastewater was planted with S. cucullata and A. pinnata.It is obvious that NH_4^+ concentrations was dramatically reduced for 70-90% removal, in a week compared with the controls which had been removed only 35% during two weeks in undiluted wastewater and 47% during a week in diluted wastewater(Fig.1b). This is because nitrogen is the most important element that is absorbed by plants and these two species preferred in form of NH_4^+ rather than NO_3^-due to the lower energy requirement for the assimilation [20-21]. In contrast, NH_4^+ in controls were gradually decreased by transforming into NO_3^- through nitrification [22]. In addition,S. cucullata has higher efficiency to remove NH_4^+ than A. pinnata since it eliminated almost 100% of ammonium in both two treatments in one week while A. pinnata in undiluted wastewater had reduced for 72%. However, nitrification caused an increasing of NO_3^- during first two weeks (Fig.1a), butit lowered on week 2-4 because of denitrification under low dissolved oxygen (DO) in two control treatments (Fig.2a), while NO_3^- in the planted treatments was reduced by plant uptake, which is slightly higher removal (4-10%) than the control (almost 2%) in 100% wastewater.For 50% wastewater, each treatmenthad nearly equal removal rate, at 21-23%, between the second and fourth week (Fig.1a). In contrast,NO_3^-concentrations were higher

during week 5-6 as thesetwo species have lower efficiency for wastewater treatment. Similarly, a study by Toledo and Penha [23] found that S. auriculata and A. caroliniana had ability to remove all NH_4^+ in fish farming effluent during a month while NO_3^- had been increased.

For PO_4^{3-} measurement, it hasn't been decreased in any treatment (Fig.1c). On 100% wastewater, PO_4^{3-} in unplanted treatments had increased approximately 20% during six weeks, probably because of a transformation of organic phosphorus into PO_4^{3-} through mineralization [24], while S. cucullata and A. pinnata had absorbed very little.However, each species could take up phosphorus in different degrees [25]. S. cucullata and A. pinnata are small free-floating plants so that their efficiency were lower than larger species, such asEichhorniacrassipes [26] and emergentAcoruscalamus [27], which could remove 90% of total P in dairy farm and over 80% of PO_4^{3-} in pig-farm effluent, respectively.

During the first week of the experiment, DO in unplanted treatments decreased 5-6 mg/L (Fig.2a). For 100% wastewater treated with S. cucullata and A. pinnata, DO was dropped to nearly zero and became higher in the second week. It is possible that microorganisms in these two treatments had grown and degraded organic matters so that the oxygen in water lowered. On the other hand, these two species had released oxygen from their roots, whichlead to DO increasing [28], especially in 50% wastewater planted with S. cucullata. As a result, this species is the most efficient in removing BOD_5, at as high as 85% removal rate in undiluted effluent during the first week, while BOD_5 in a half dilution had been 77% reduced (Fig.2b).A similar result had been discovered in S.natans, which could remove 38-78% of BOD_5 while unplanted had removed only 27% [8]. However, BOD_5 in planted treatments had slightly increased after a month since the oxygen had been used for plant's organic matter degradation. At that time, the fronds' color of these two species gradually changed from green to yellow and brown.

Regarding the other water physical properties,the pH were constantly at 6.8-7.3 from the beginning until the sixth week (Fig.2c). Likewise, EC and TDS in unplanted wastewater treatmentsduring six weeks were nearly equal from the beginning. (Fig.2d and 2e). However, 10-20% increasing of the two parameters presented in the four planted treatments, which is probably due to nutrients release into water because of plant decay, increasing dissolved ions and hence the conductivity. Similar to the wastewater in constructed wetland with Typhalatifolia [29], their conductivity and suspended solid had been increased. However, in this study, TSSin all treatmentswere reduced by 70-90% after 4 weeks (Fig.2f) which probably caused by sedimentation ofthe suspended solids to the bottom of the buckets.

CONCLUSIONS

In conclusion, two weeks of aeration had improved all parameters of pig-farm wastewater to become lower than a PCD standard [16; Table 1]. However, the water quality is much better when plants were applied.BothS. cucullataand A. pinnatahave a high potential for reducing NH_4^+ in aerated pig-farm wastewater during in one week. These two species could also decrease NO_3^- concentration between the second and fourth week although their proportion in this function is low. Furthermore, S. cucullatagreatly removed BOD_5 that may because of the ability to release O_2 from its roots, so this species can be a one candidate to use for wastewater treatment systems.

Therefore, these two species are interesting for decreasing inorganic nitrogen in constructed wetland. Furthermore, S. cucullata could be used to remove BOD_5 in one week, so it is interesting for rising oxygen in wastewater. However, the time of water treatments is also important, these two species should not be planted longer than a month so that their efficiencies would be worse, leading to lower quality of water.

ACKNOWLEDGEMENTS

This research is financially supported by Science Achievement Scholarship of Thailand (SAST) and Faculty of Science, Chiang Mai University. The authors thankMr. VisutSirinupongsanun, Department of Animal and Aquatic Science, Faculty of Agriculture, Chiang Mai University, for his kind support and a water sampling permission.

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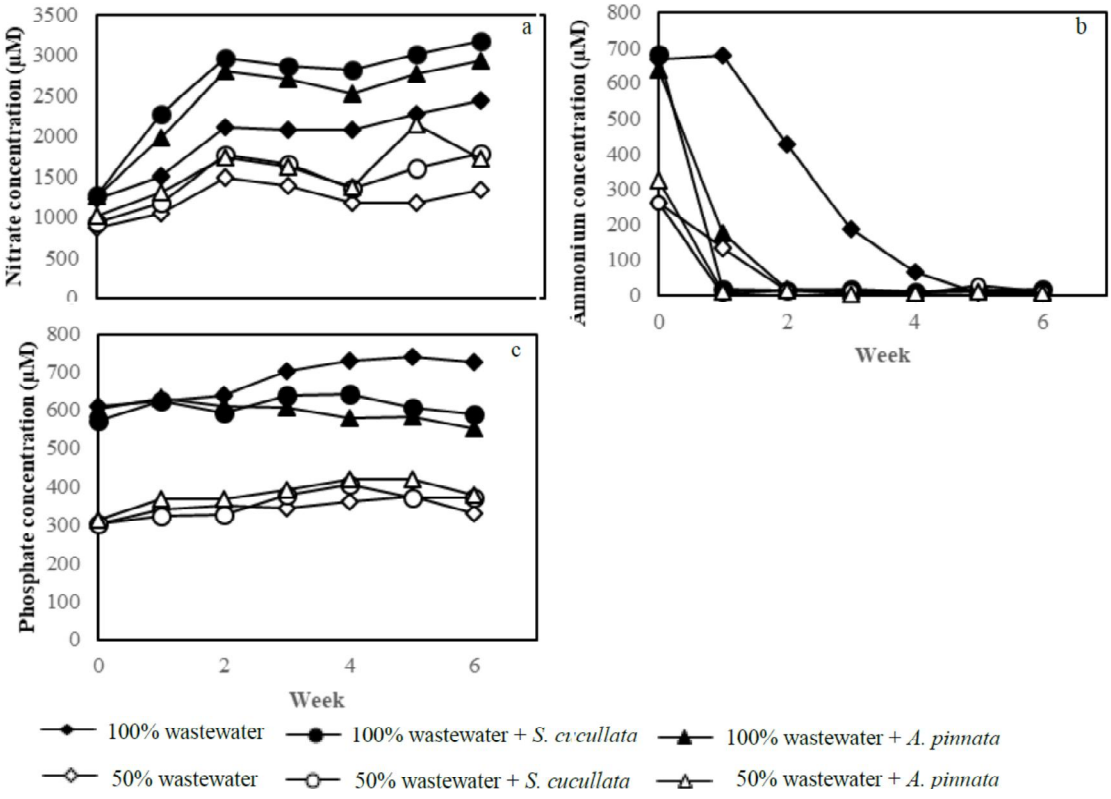


Fig.1. NO_3^- concentration (a), NH_4^+ concentration (b), and PO_4^{3-} concentration (c) in aerated pig-farm wastewaterduring six weeks of the experiment

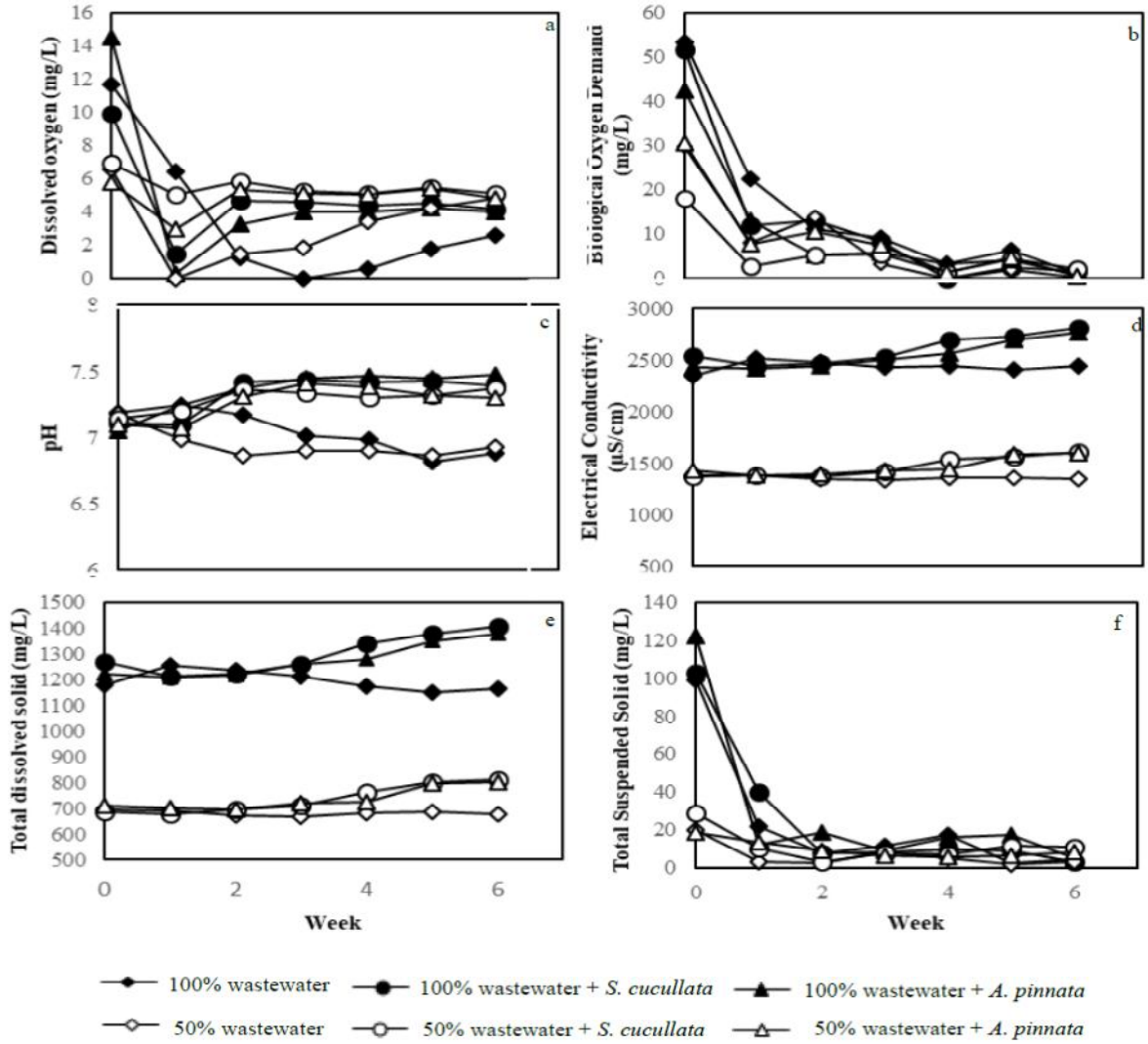


Fig.2. Dissolved Oxygen (a), Biological Oxygen Demand (b), pH (c), Electrical Conductivity (d), Total Dissolved Solid (e), and Total Suspended Solid (f) in aerated pig-farm wastewaterduring six weeks of the experiment

Table. 1. Wastewater properties before and after an aeration for two weeks

	DO (mg/L)	BOD_5 (mg/L)	TSS (mg/L)	TDS (ppm)	pH	EC (µS/cm)	NO_3^- (µM)	NH_4^+ (µM)	PO_4^{3-} (µM)
Before an aeration	0.9	52.75	185	1,740	7.18	3,480	205.88	4,982.54	800
After an aeration	12.62	47.78	113.13	1,222	7.12	2,445	1,261.5	662.61	595.64
PCD[a] [25]	-	60	150	-	5.5-9	-	-	120 mg/L TKN	-

[a]General standards for discharge of pig-farm into inland surface water by PCD [16]

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