Treatment and Reuse of Swine Wastewater

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Abstract

The quality of typical treated swine wastewater does not pass the effluent standard. There are some nutrients and organic matter, which cause deterioration in the water resources. Recycling this wastewater in a sustainable manner presents an important challenge. This study investigates the possibility of changing valuable matter in swine wastewater to algal, *Chlorella sp.*, biomass and then harvesting by small herbivorous aquatic arthropod, *Moina macrocopa*. The effluent quality is also evaluated.

The unicellular form of *Chlorella sp.* grows quite well in treated swine wastewater under normal algal culturing conditions. The amount of biomass reaches the normal range (100-260 mg/L) within a short period (2-4 days). However, the suitable detention time that gives the maximum *Chlorella sp.* removal efficiency and *Moina macrocopa* production was found to be 2 days. The efficiency of *Chlorella sp.* removal increased when the starting algal biomass concentration was increased from 100 mg/L to 260 mg/L. On the other hand, it did not change after varying the starting number of *Moina macrocopa* from 1,500 org./L to 3,000 org./L (P-value > 0.05). The highest efficiency occurred when *Chlorella sp.* concentration was one milligram per 12 organisms of *Moina macrocopa*. The characteristics of swine effluent, including suspended solids, OD_{560}, and COD, were better than the effluent standard. It is recommended that this culturing system is incorporated into the final stage of the wastewater treatment process. The condition of applying 300 g wet weight of *Moina macrocopa* for one cubic meter of wastewater (3,000 org./L) which has dense algal biomass concentration (100-260 mg/L) would yield both the highest wastewater treating efficiency and the maximum *Moina macrocopa* production within 2 days.

Key words: Swine wastewater, *Chlorella sp.*, *Moina macrocopa*.

1. Introduction

Developments in intensive agricultural techniques have been promoted throughout Thailand. They give benefits in terms of earnings, but at the same time can cause many environmental problems. Most agricultural effluents, such as those from swine farming for example, are discharged directly to the waterways and considered to be one of the most serious water pollution sources of major rivers in Thailand [5,13].

The use of only conventional methods in treating swine wastewater, such as anaerobic ponds and anaerobic digesters, cannot succeed in meeting the effluent standard [3]. Using *Chlorella sp.* as a biological treatment process, for further treatment of this wastewater, however, results in high efficiency [2,8]. It not only removes certain amounts of organic substances and nutrients, but also produces useful by-products of algal cell, which have various uses. However, a proper biomass is removal process is needed in order to prevent re-pollution of water resources.

There are various methods for harvesting algal cell from the effluent e.g. microscreen, coagulation-flocculation, dissolved-air flotation and auto-flocculation[5,11,16]. These methods require extra investment costs and operational skills. Herbivorous fishes have also been used for algal biomass harvesting. They are able to remove some amount of algal cell, but there is the disadvantage of producing additional organic substances and suspended solids in the effluent [11,16].

Utilization of small herbivorous aquatic arthropod, *Moina macrocopa*, presents a challenge. It offers high biomass and Biological Oxygen Demand (BOD) removal efficiency as well as requiring negligible investment cost [4,6,10]. Moreover, *Moina macrocopa*, which is easily harvested from the effluent, is a natural feed for various higher aquatic animals especially fish farmed both for human consumption and for decorative aquaria. The average market price of *Moina macrocopa* is 20-50 baht/kg (0.5-1 U.S. dollar/kg).

The present study focused on using *Moina macrocopa* to remove Chlorella sp. in treated swine wastewater. Two major factors affecting the removal efficiency, starting algal biomass and number of *Moina macrocopa*, were determined. Specific objectives of this study were to evaluate: (i) possibility of culturing Chlorella sp. in treated swine watewater, (ii) Chlorella sp. removal efficiency and *Moina macrocopa* production at different starting biomass and number of *Moina macrocopa* and, (iii) wastewater treating efficiency after harvesting *Moina macrocopa*.

2. Materials and Methods

Characterization of Treated Swine Wastewater

Grab sample of swine wastewater was taken from swine farming type A (number of swine 50-100 bodies), Nakorn Phatom province, Thailand. It had already passed the primary treatment process, waste stabilization pond. Sampling the wastewater was carried out once a month, from July to December 1999, a total of 6 months. The samples were characterized immediately. If there was no time for this, they were kept at a cold temperature (4 °C). The characterization of chemical properties was chemical oxygen demand (COD), biological oxygen demand (BOD), total kjeldahl nitrogen (TKN), and total phosphorus (TP).

Culturing of Stock *Chlorella sp.* in Treated Swine Wastewater

The treated swine wastewater was stored in the reactor, a transparent plastic container 3 liters in volume, and 14x14x15 cm in size. Water level was maintained at 10 cm. Lighting was provided for 12 hours/day by a constant artificial source, fluorescent lamps with light intensity equal to 5,000 lux and continuous mixing. The sample was tested daily for *Chlorella sp.* biomass by filtering with GF/C filter paper and drying in an oven at 103°C. Typically, the algal biomass ranges from 100 to 260 mg/L [14]. This study involved culturing *Chlorella sp.* in the reactor at different detention times until the design concentrations of 100 mg/L, 180 mg/L, and 260 mg/L respectively were obtained.

Culturing of Stock *Moina macrocopa*

Seeding of *Moina macrocopa* had been cultured in the reactor with stock *Chlorella sp.* in treated swine wastewater. The Pipette method [12,14] was used for measuring *Moina macrocopa* organisms. The ratio of *Moina macrocopa* to wastewater was 20 organisms/L. Culturing was carried out for 2 days then the mature and strong cultures were chosen for use in the experiment.

Removal Efficiency of *Chlorella sp.* by *Moina macrocopa*

Two major factors which affect the *Chlorella sp.* removal efficiency, starting algal biomass (100, 180, 260 mg/L) and starting number of *Moina macrocopa* (0, 1,500, 3,000 organisms/L) were studied at a detention time of 2 days. The experiment was run in triplicate. The total experimental unit was 27 (3x3x3).

Characterization of Effluent

Effluent from the reactor after the harvesting of *Moina macrocopa* was characterized on a daily basis for algal biomass, optical density at wavelength 560 nm (OD_{560}), chemical oxygen demand (COD), and *Moina macrocopa* organisms. The characteristic of final effluent, biological oxygen demand (BOD), total kjeldahl nitrogen (TKN), and total phosphorus (TP) was analyzed and compared with the influent.

Statistical Analysis

One way ANOVA and non-parametric test (Kruskal-Wallis test) at 0.05 level of significance were used to compare the *Chlorella sp.* removal efficiency of the experimental unit in reactor of 100, 180, and 260 mg/L starting algal biomass concentration in treated swine wastewater.

The T-test at 0.05 level of significance was used to compare the *Chlorella sp.* removal efficiency of the experimental unit in the reactor of 1,500 and 3,000 org./L starting number of *Miona macrocopa.*

3. Results and Discussion

Characteristics of Treated Swine Wastewater

Characteristics of treated swine wastewater are presented in Table 1. Although the wastewater had been pre-treated in the waste stabilization pond, it still contained high organic pollutants, which were expressed in terms of high BOD, COD and TKN values. The ratio of COD:BOD (3:1) was in the normal range of general agricultural waste in which most organic pollutants are simply biodegradable. Most of these organic pollutants come from feces and urine of swine and resulted in high TKN, TP and SS values.

The characteristics of BOD, COD, TKN and SS exceeded the Thai Industrial Effluent Standard of 20, 120, 100 and 50 mg/L respectively (Ministry of Industry, 1996). It confirms that swine wastewater needs additional treating process before being discharged into the natural water resource.

Table 1

Characteristics of Treated Swine Wastewater

Parameter	Mean Value
Chemical Oxygen Demand (COD), mg/L	301
Biochemical Oxygen Demand (BOD), mg/L	127
Total Kjeldahl Nitrogen (TKN), mg/L	315
Total Phosphorus (TP), mg/L	17
Suspended Solid (SS), mg/L	153

Possibility of Culturing *Chlorella sp.* in Treated Swine Wastewater

Chlorella sp. was cultured, without seeding, in the reactor under the designed laboratory conditions. It was measured daily for algal biomass quantity as mg/L. The cultured biomass at different detention times is shown in Table 2.

Table 2

Produced Algal Biomass at Different Detention Times

Algal Biomass (mg/L)	Detention Time (Day)
100	2
180	3
260	4

Through microscopic scanning, almost all algal biomass found in the reactor was *Chlorella sp.* and a negligible amount was *Euglena sp.*. This agrees with Udomsinroj [14] who found that algal biomass cultured in an algal pond is mainly in microscopic unicellular form. Therefore, total algal biomass found in this study was assumed to be the biomass of *Chlorella sp.* only.

From Table 2, algal biomass can be seen to have increased dramatically. The quantities were increasing twofold at detention times of 1.2 and 1.3 days respectively. These results were similar to a study of Assavaree and Pechmanee [1] which found that *Chlorella sp.* has a mean doubling time of 1.2 days when cultured at light intensity of 5,000 lux.

It can be concluded that there is a possibility of culturing the unicellular form of *Chlorella sp.* in treated swine wastewater under normal algal culturing conditions. Moreover, the amount of biomass reached the normal range (100-260 mg/L) within a short period (2-4 days).

Removal of *Chlorella sp.* by *Moina macrocopa*

A. Suitable Detention Time

In order to determine the suitable detention time, a pretest study was done under conditions of 200 mg/L of starting algal biomass and 2,000 org./L of *Moina macrocopa*. Results are shown in Table 3.

Table 3

Removal Efficiency and *Moina macrocopa* Production at Different Detention Times

Parameter	Detention time			
	1 day	2 days	3 days	4 days
% of algal biomass removal	68.30	80.65	81.25	81.00
% of OD560 reduction	73.33	83.33	80.00	77.77
% of COD reduction	52.47	60.65	49.27	59.11
Total number of *M. macrocopa*, ,org./L	12,550	13,950	5,850	2,750

The removal efficiency and *Moina macrocopa* production were quite similar. They rapidly increased the number to six fold and provided removal efficiency over fifty percent from the start within only one day. On the second day of incubation, the maximum numbers of *Moina macrocopa* and removal efficiency were reached. These may be caused by asexual reproduction of *Moina macrocopa* as an abundant food condition [12]. On the other hand, where numbers of *Moina macrocopa* were over the food supply, the population was sharply decreased while removal efficiency was slightly changed. It can be concluded that the suitable detention time, where maximum treating efficiency and *Moina macrocopa* production was met, is just two days of culturing.

B. Removal Efficiency and *Moina macrocopa* Production at Different Starting Algal Biomass

The relationship between the efficiency of *Chlorella sp.* removal and *Moina macrocopa* production was evaluated from using various starting algal biomass (100, 180, 260 mg/L) and the starting number of *Moina macrocopa* (0, 1,500, 3,000 org./L) at two days retention time. Tables 4 and 5 show the statistical analysis, at 0.05 level of significance, of the mean removal efficiency and the total number of *Moina macrocopa* production at different starting algal biomass concentrations (100-260 mg/L) in treated wastewater.

The starting algal biomass concentration in treated synthetic swine wastewater could affect the removal efficiency by *Moina macrocopa*. The denser starting algal biomass concentration in treated swine wastewater (260 mg./L) gave the higher *Chlorella sp.* removal efficiency than the less dense starting algal biomass concentration (180 and 100 mg/L). The reason was that *Moina macrocopa* filtration rate depended largely on the algal biomass concentration and animal size, which could be directly measured by the number of suspended cell concentration [12]. Moreover, both filtration rate and food intake of *Moina macrocopa* rose with the increase of cellular concentrations of *Chlorella sp.*, which determined algal biomass concentration, OD_{560}, and COD.

Far from increasing the treatment efficiency, the highest starting algal biomass concentration (260 mg/L) in treated swine wastewater gave a lower number of *Moina macrocopa* than the treatment of algal biomass concentration 100 and 180 mg/L. However, the decreasing number of *Moina macrocopa* according to increased algal biomass concentration showed no statistical difference.

This means that the reproduction rate of *Moina macrocopa* does not seem to depend on the range of starting algal biomass concentration 100 to 260 mg/L.

Table 4

Algal Removal Efficiency and *Moina macrocopa*_Production at Different Starting Algal Biomass Concentration and Starting Number of *Moina macrocopa*, Two Days Detention Time

(oneway ANOVA analysis)

Starting Algal Biomass Conc. (mg/L)	Dependent Variable	Mean*	S.D.	F-test	P-value
100	Algal biomass removal efficiency	76.16 [a]	2.55	32.041	< 0.001
180		85.09 [b]	3.89		
260		88.59 [c]	1.21		
100	Total number of *Moina Macrocopa*	7,733 [a]	1,269	0.693	0.516
180		7,533 [a]	1,372		
260		6,833 [a]	1,520		

* Mean of the treatment using starting number of 1,500 and 3,000 org./L *Moina macrocopa* in the column followed by same letters is not significant different at P < 0.05 using DMRT.

Table 5

Removal Efficiency at Different Starting Algal Biomass

Non parameteric test (Kruskal-Wallis test)

Starting Algal Biomass Conc. (mg/L)	Depen-dent Variable	Mean*	S.D.	X^2	P-value
100	OD_{560} removal efficiency	70.10 [a]	8.46	15.221	< 0.001
180		85.52 [b]	3.08		
260		92.22 [c]	0.94		
100	COD removal efficiency	53.70 [a]	3.54	12.931	0.002
180		63.58 [b]	4.94		
260		68.53 [c]	0.66		

* Mean of the treatment using starting number of 1,500 and 3,000 org./L *Moina macrocopa* in the column followed by same letters is not significant different at P < 0.05 using DMRT.

C. Removal Efficiency of *Chlorella sp.* and *Moina macrocopa* Production at Different Starting number of *Moina macrocopa*

The analysis of the *Chlorella sp.* removal efficiency and total number of *Moina macrocopa* from various starting algal biomass concentrations (100-260 mg/L) and starting number of *Moina macrocopa* (1,500 and 3,000 org./L), two days detention time, by using t-test analysis at 0.05 level of significance is shown in Table 6.

Table 6

Removal Efficiency and *Moina macrocopa* Production at Different Starting number of *Moina macrocopa* and Starting Algal Biomass Concentration, Two Days Detention Time

(T-test analysis)

Starting number of *Moina macrocopa* (org./L)	Dependent Variable	Mean*	S.D.	t-test	P-value
1,500	Algal biomass Removal efficiency	83.19 [a]	6.79	-0.065	0.949
3,000		83.38 [a]	5.47		
1,500	OD_{560} Removal efficiency	83.22 [a]	9.92	0.234	0.818
3,000		82.00 [a]	12.05		
1,500	COD removal efficiency	61.50 [a]	7.89	-0.250	0.806
3,000		62.37 [a]	6.80		
1,500	Total number of *Moina macrocopa*	6,333 [a]	1,082	-4.964	< 0.001
3,000		8,400 [b]	624		

* Mean of the treatment using starting number of 100, 180, and 260 mg/L algal biomass concentration in the column followed by same letters is not significant different at P < 0.05 using DMRT.

It was found that the starting number of *Moina macrocopa* did not affect the *Chlorella sp.* removal efficiency in treated swine wastewater (Table 6). The reason was that the filtration rate of *Moina macrocopa* depends largely on algal biomass concentration [12]. A study by Pechmanee et al [10] found that the amounts of *Chlorella sp.* filtered by *Moina macrocopa* varied with the existing amounts of *Chlorella sp.* The consumption rate is in the range of 0.5 – 1.5 x 10^6 cells of *Chlorella sp.* /day/organism.

The starting number of *Moina macrocopa* could affect the total number of produced *Moina macrocopa*. Production rate per individual of the treatment with 1,500 org./L starting number of *Moina macrocopa* was higher than those treatments of 3,000 org./L. However, the final population number of *Moina macrocopa* in the reactor with a starting number of 3,000 org./L

Moina macrocopa was significantly, the highest and met the objective of aquatic production. From the experimental condition where food was abundant for growth and reproduction, *Moina macrocopa* would reproduce parthenogenically. Each parthenogenetic female could produce 4 - 13 larvae for breeding [1]. Thus, the higher starting number of *Moina macrocopa* gave a higher total number of *Moina macrocopa* than the lower starting number. Wongrad et al [15] suggested that when abundant food is available and environmental conditions are suitable, the population consists almost exclusively of parthenogenic females. Such females are capable of producing their young without the services of a male. If environmental conditions remain satisfactory, the new generation will also be parthenogenic females.

The result from Table 6 shows that the treatment with a starting number of 1,500 org./L *Moina macrocopa* gave equal treatment efficiency, OD_{560} and COD reduction, to those of 3,000 org./L starting number of *Moina macrocopa*. Therefore, the recommended design criteria is the latter because it yielded both the highest treatment efficiency of wastewater and maximum production within 2 days.

Wastewater Treating Efficiency as Measured after the Harvest of *Moina macrocopa*

After culturing *Chlorella sp.* in the swine wastewater until the desired concentration of 260 mg/L was obtained, the amount of *Moina macrocopa* 3,000 org./L was applied to the reactor. Wastewater treating efficiency after harvesting of *Moina macrocopa* at detention time 0,1,2 days was observed as shown in Table 7.

The process of culturing *Chlorella sp.* causes the transformation of organic matter and nutrients in the wastewater to algal biomass. At 0 days of detention time (without *Moina macrocopa* activity), therefore, the wastewater quality was better than the original (Table 1).

Moina macrocopa had been working as an algal biomass harvester. The more algal production and harvesting the better the achieved water quality. The filter-feeding activities involve the removal of finely suspended materials from the water and result in a reduction in turbidity, BOD and total bacteria [4,15,16]. A study by Navanaraset [9] found that using *Moina macrocopa* for *Chlorella sp.* removal could reduce BOD, total bacteria and E.coli by 83.7%, 91.2% and 98.2% respectively. The reduction rates of TKN and TP were rather high (55% and 67% respectively) due to most of these nutrients being changed to solid organic forms of algal cell which facilitate feeding to *Moina macrocopa*. However, a study by Kawai et al [6] agreed that *Moina macrocopa* could remove TKN and TP in the effluent by 59% and 44% respectively.

Table 7

Removal Efficiency and *Moina macrocopa* Production at 260 mg/L of Starting Algal Biomass Concentration and 3,000 org./L of *M.macrocopa*

Parameter	Detention time		
	0 day	1 day	2 day
mg/L of algal biomass, (% removal)	260,(0)	63,(75.64)	41,(84.11)
OD_{560}, (% removal)	0.44,(0)	0.11,(75.76)	0.07,(84.85)
mg/L of COD, (% removal)	323,(0)	110,(65.91)	95,(70.52)
mg/L of BOD, (% removal)	53,(0)	24,(54.28)	13,(74.49)
mg/L of TKN, (% removal)	17,(0)	10,(41.11)	7,(55.56)
mg/L of TP, (% removal)	10,(0)	4,(64.46)	3,(67.02)
Total number of *Moina macrocopa*, org./L	3,000	5,260	7,267

The results prove that this culturing system could successfully treat swine wastewater and return beneficial organisms to the environment. The characteristics of the effluent obtained were also under the level of the effluent standard [7].

4. CONCLUSIONS

1) Efficiency of *Chlorella sp.* removal, using *Moina macrocopa*, depends on the starting algal biomass concentration. The denser the starting algal biomass concentration, the higher *Chlorella sp.* removal efficiency. The highest removal efficiency occurs when there is 260 mg/L of starting algal

biomass concentration for 3,000 organisms of *Moina macrocopa* per liter.

2) The increased number of *Moina macrocopa* does not depend on the starting algal biomass concentration e.g. the total number of *Moina macrocopa* in systems of 100, 180, and 260 mg/L starting algal biomass concentration were similar (Table 4).
3) Efficiency of *Chlorella sp.* removal does not depend on the starting number of *Moina macrocopa*. The *Chlorella sp.* removal efficiency in systems of 1,500 and 3,000 org/L starting number of *Moina macrocopa* was similar (Table 6).
4) The increased number of *Moina macrocopa* depends on the starting number of *Moina macrocopa* in the system, since total number of *Moina macrocopa* in system of 3,000 org/L starting number of *Moina macrocopa* was more than that of 1,500 org/L starting number of *Moina macrocopa* (Table 6).
5) The variation of the starting algal biomass concentration from 100, 180, and 260 mg/L resulted in increasing *Moina macrocopa* product from 3,000 org./L to 8,600, 8,400, and 8,200 org./L (from the mean of 8,600 org./L in Table 6), respectively. The production rates of the *Moina macrocopa* were similar under these conditions. This confirmed that excess food condition and the highest *Chlorella sp.* removal efficiency exists when there is approximately 1 mg of *Chlorella sp.* per 12 organisms of *Moina macrocopa*.

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6. References

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