# **Treatment of swine manure effluent using freshwater algae: Production, nutrient recovery, and elemental composition of algal biomass at four effluent loading rates**

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## **Abstract**

Cultivating algae on nitrogen (N) and phosphorus (P) in animal manure effluents presents an alternative to the current practice of land application. The objective of this study was to determine how algal productivity, nutrient removal efficiency, and elemental composition of turf algae change in response to different loading rates of raw swine manure effluent. Algal biomass was harvested weekly from laboratory scale algal turf scrubber units using four manure effluent loading rates (0.24, 0.40, 0.62 and 1.2 L m<sup>-2</sup> d<sup>-1</sup>) corresponding to daily loading rates of 0.3–1.4 g total N and 0.08–0.42 g total P. Mean algal productivity values increased from 7.1 g DW m<sup>−</sup><sup>2</sup> d<sup>−</sup><sup>1</sup> at the lowest loading rate (0.24 L m<sup>-2</sup> d<sup>-1</sup>) to 9.4 g DW m<sup>-2</sup> d<sup>-1</sup> at the second loading rate (0.40 L m<sup>-2</sup> d<sup>-1</sup>). At these loading rates, algal N and P accounted for  $> 90\%$  of input N and 68–76% of input P, respectively. However, at higher loading rates algal productivity did not increase and was unstable at the highest loading rate. Mean N and P contents in the dried biomass increased 1.5 to 2.0-fold with increasing loading rate up to maximums of 5.7% N and 1.8% P at 1.2 L m<sup>−</sup><sup>2</sup> d<sup>−</sup>1. Biomass concentrations of Al, Ca, Cd, Fe, K, Mg, Mn, Mo, Si, and Zn increased 1.2 to 2.6-fold over the 5-fold range of loading rate. Biomass concentrations of Cd, K, Pb, and Si did not increase significantly with loading rate. At the loading rate of  $0.40 \text{ L m}^{-2} d^{-1}$  (corresponding to peak productivity) the mean concentrations of individual components in the algal biomass were (in mg kg<sup>-1</sup>): 250 (Al), 4900 (Ca), 0.30 (Cd), 1050 (Fe), 3.4 (Pb), 2500 (Mg), 105 (Mn), 6.0 (Mo), 7,500 (K), and 510 (Zn). At these concentrations, heavy metals in the algal biomass would not be expected to reduce its value as a soil or feed amendment.

*Abbreviations*: ATS, algal turf scrubber; TP, total phosphorus; TN, total nitrogen

#### **Introduction**

Controlling the input of nitrogen (N) and phosphorus (P) from dairies and other livestock operations into the surrounding air- and water-sheds poses both technical and economic challenges to the agricultural community. During storage and land application of manure effluents, large amounts of N are lost to the atmosphere due to volatilization of ammonia. Ecologically sound manure management on farms is vital to minimize loses of valuable plant nutrients and to prevent nutrient contamination of the surrounding watershed (Paerl et al., 2002).

Cultivating algae on nitrogen and phosphorus in animal manure effluents presents an alternative to the current practice of land application. There is considerable literature on the treatment of raw and anaerobically digested swine manure effluent by immobilized algae (Jimenez-Perez et al., 2004), algal cultures, (Ayala & Bravo, 1984) and suspended algae in high rate pond systems (Costa et al., 2000; Goh, 1986; Olguin, 2003; Olguin et al., 2001). The use of benthic algae for swine manure treatment is much less characterized, but such systems are compelling because they yield an algal biomass that is easy to harvest at scales appropriate to farm operations (Hoffman, 1998; Wilkie & Mulbry, 2002). Algal productivity, algal composition and nutrient removal efficiency from anaerobically digested dairy manure using attached algae has recently been studied in laboratory-scale algal turf scrubber (ATS) units (Kebede-Westhead et al., 2004; Kebede-Westhead et al., 2003). The objective of this study was to determine comparable values using different loading rates of raw swine manure effluent.

# **Materials and methods**

### *Swine manure and algal turf scrubbers*

Experiments were conducted using raw flushed swine manure effluent from the Brandon Howard farm near Richlands, North Carolina, USA. The farm is a 3,520 head finishing operation and is also a test site for the utilization of constructed wetlands for effluent treatment. The effluent was collected prior to solid separation as the barns were being cleaned out at the end of a growout cycle. All tests were performed on one batch of manure effluent that was transported to Maryland in 200 L barrels and stored at 4 ◦C. The manure effluent concentrations of ammonium-N, nitrate-N, total N (TN), soluble reactive phosphorus, and total P (TP) were 920, <1, 1130, 330, and 340 mg  $L^{-1}$ , respectively. Other physical characteristics of the effluent are shown in Table 1. Elemental composition of the manure effluent is shown in Table 3. A laboratory-scale ATS unit containing 1  $m<sup>2</sup>$  growing area (Figure 1) was operated in a semicontinuous mode by recycling 205 L of effluent at 55 L min<sup>−</sup>1, adding manure effluent daily, and replacing 75 L

*Table 1.* Characterization of raw swine manure effluent. Values are averages of three subsamples with standard deviations.

pH	7.88
Total COD (mg $L^{-1}$ )	$6430 + 70$
Soluble COD (mg $L^{-1}$ )	$820 \pm 10$
Total volatile solids (mg $L^{-1}$ )	$7120 \pm 320$
Soluble volatile solids (mg $L^{-1}$ )	$4260 + 360$
Total suspended solids (mg $L^{-1}$ )	$4610 \pm 60$
Volatile suspended solids (mg $L^{-1}$ )	$3550 \pm 30$
Conductivity (mS $cm^{-1}$ )	$7.56 \pm 0.40$
Alkalinity (mg CaCO <sub>3</sub> $L^{-1}$ )	$4680 + 80$



*Figure 1*. Schematic drawing of a laboratory-scale algal turf scrubber (ATS) unit. Scrubber effluent (205 L) is contained in a plastic drum and continuously recycled through the system using a sump pump with a flow rate  $(Q) = 55$  L min<sup>-1</sup>. A plastic trough fills and tips over, releasing pulses of effluent that wash over the attached algal turf every 8–10 sec before draining back into the plastic drum. Raw swine manure effluent (0.24, 0.40, 0.62, or 1.23 L per day) was added daily to the recirculating scrubber effluent.

of effluent with distilled water after each harvest as previously described (Kebede-Westhead et al., 2003). The unit was maintained at ambient laboratory temperature  $(23-26\degree C)$ , and illuminated using two 400 W metal halide lights under nearly continuous light (23:1 h lightdark cycle). Incident light averaged 390 (range 240– 633)  $\mu$ mol photons m<sup>-2</sup> s<sup>-1</sup>. The ATS unit, originally established with algal consortia from a nearby stream in Beltsville, was dominated by filamentous green algae including *Microspora willeana* Lagerh., *Ulothrix ozonata* (Weber and Mohr) K¨utz, *Rhizoclonium hieroglyphicum* (C.A. Agardh) K¨utz and *Oedogonium* sp. To minimize volatilization of ammonia, effluent pH was maintained between 7–7.5 by daily additions of dilute hydrochloric acid. Four loading rates of manure  $(0.24, 0.40, 0.62 \text{ and } 1.23 \text{ L m}^{-2} \text{ d}^{-1})$  corresponding to daily loading rates of  $0.27-1.39$  g TN and  $0.08-0.42$  g TP were conducted. Algal biomass was harvested every 6–8 days using a wet/dry vacuum, dewatered by sieving the harvested material through 3 mm mesh nylon netting (Aquatic Ecosystems, Apopka, FL), and dried overnight at 25 ◦C using an electric fan prior to analysis for moisture, ash, total Kjeldahl nitrogen (TKN), TP, and elemental composition using inductively coupled plasma analysis (APHA, 1995). ATS water samples were collected and stored at 4 ◦C prior to analysis for ortho-phosphate  $(PO<sub>4</sub>-P)$  and ammonia (NH4-N). Samples of process water from dewatering the harvested biomass were collected and stored

at  $-20$  °C prior to analysis for TKN and TP (APHA, 1995).

# **Results**

## *Algal productivity*

Algal biomass was harvested weekly from a laboratory scale algal turf scrubber (ATS) unit using four manure effluent loading rates (0.24, 0.40, 0.62 and 1.23 L m<sup>-2</sup> d<sup>−</sup>1) corresponding to daily loading rates of 0.3–1.4 g TN and 0.08–0.42 g TP. (Table 2). Mean algal productivity values increased from 7.1 g DW m<sup>-2</sup> d<sup>-1</sup> at the lowest loading rate (0.24 L m<sup>-2</sup> d<sup>-1</sup>) to 9.4 g DW m<sup>-2</sup> d<sup>-1</sup> at the second loading rate (0.40 L m<sup>-2</sup>  $d^{-1}$ ) (Table 2, Figure 2 panel A). However, at higher loading rates algal productivity did not increase further and became highly variable at the highest loading rate  $(1.23 \text{ L m}^{-2} \text{ d}^{-1})$ . Moisture content of the air-dried biomass was relatively constant (10.2  $\pm$  1.5 % of air dried weight) under all loading rates as was the relative content of ash-free dry weight (91  $\pm$  2.3 % DW) (not shown).

#### *Recovery of manure N and P*

Table 2 presents the averaged N and P balances during the test periods. Levels of  $PO<sub>4</sub>-P$  and  $NH<sub>4</sub>-N$  in the recirculating effluent were approximately 1 mg  $L^{-1}$  at the loading rate of 0.24 L m<sup>-2</sup> d<sup>-1</sup> and showed little change as the loading rate increased up to  $0.62$  L d<sup>-1</sup>. At the loading rate of 1.23 L m<sup>-2</sup> d<sup>-1</sup> levels of PO<sub>4</sub>-P and  $NH<sub>4</sub>-N$  in the recirculating effluent increased to 8 and  $2 \text{ mg } L^{-1}$ , respectively (not shown). Unfortunately, we did not perform TKN and TP analyses on the ATS water samples and thus could not determine total N and P recoveries for these experiments. However, algal N and P accounted for >90% of input N and 68–76% of input P, respectively, at the two lowest loading rates.

# *Effect of loading rate on elemental composition of algal biomass*

Mean N and P contents in the dried biomass increased 1.5 and 2.1 fold, respectively, over the 5-fold range of loading rates up to maximums of 5.7% N and 1.8% P at 1.23 L m<sup>-2</sup> d<sup>-1</sup> (Table 3, Figure 2 panel B). Concentrations of Ca and Mg also increased (1.5 and 1.2-fold respectively) over the range of loading rates. Concentrations of Al, Cu, Fe, Mn, and Zn increased only slightly or decreased between the loading rates of 0.24–0.4 L m<sup>-2</sup> d<sup>-1</sup> but then increased 1.7 to 2.5 fold at the higher loading rates (Table 3, Figure 2 panel C). Concentrations of Cd, K, Pb, and Mo did not generally increase with loading rate (Table 3, Figure 2 panel D). At the loading rate of 0.40 L m<sup>-2</sup> d<sup>-1</sup> (corresponding to peak productivity and nitrogen efficiency) the mean concentrations of individual components in the algal biomass were (in mg kg<sup>-1</sup>): 250 (Al), 4900 (Ca),

Table 2. N and P balances for ATS unit using swine manure effluent at four manure effluent loading rates. Values are the means of four to nine separate weekly measurements  $\pm$  SD.

	$(n = 6)$	$(n = 9)$	$(n = 5)$	$(n = 4)$
INPUT $(m^{-2} d^{-1})$				
Volume of manure added (L)	$0.24 \pm 0.03$	$0.40 \pm 0.06$	$0.62 \pm 0.08$	$1.23 \pm 0.12$
$NH4-N(g)$	$0.22 \pm 0.03$	$0.37 \pm 0.06$	$0.57 \pm 0.08$	$1.13 \pm 0.11$
organic- $N(g)$	$0.05 \pm 0.01$	$0.08 \pm 0.01$	$0.13 \pm 0.02$	$0.26 \pm 0.03$
TN(g)	$0.27 \pm 0.03$	$0.48 \pm 0.07$	$0.70 \pm 0.09$	$1.39 \pm 0.14$
TP(g)	$0.08 \pm 0.01$	$0.14 \pm 0.04$	$0.21 \pm 0.03$	$0.42 \pm 0.04$
OUTPUT $(m^{-2} d^{-1})$				
Harvested algal biomass (g)	$7.1 \pm 1.0$	$9.4 \pm 2.2$	$8.7 \pm 0.9$	$9.6 \pm 3.1$
Algal N content $(\%)$	$3.7 \pm 0.6$	$4.3 \pm 0.5$	$5.4 \pm 0.5$	$5.7 \pm 0.3$
TN in harvested algae $(g)$	$0.27 \pm 0.07$	$0.41 \pm 0.11$	$0.47 \pm 0.09$	$0.55 \pm 0.18$
Algal P content $(\% )$	$0.85 \pm 0.1$	$1.1 \pm 0.2$	$1.3 \pm 0.2$	$1.8 \pm 0.3$
TP in harvested algae $(g)$	$0.06 \pm 0.01$	$0.09 \pm 0.02$	$0.12 \pm 0.03$	$0.17 \pm 0.06$
RECOVERY (%)				
N removal by algae	$98 \pm 30$	$95 \pm 20$	$67 \pm 7$	$40 \pm 15$
P removal by algae	$76 \pm 23$	$77 \pm 13$	$55 \pm 8$	$41 \pm 15$



*Figure 2*. Algal productivity and normalized concentrations of ATS biomass constituents as a function of swine manure effluent loading rate. Productivity (g DW m<sup>-2</sup> d<sup>-1</sup>) (panel A); and normalized concentrations of ATS biomass constituents (panels B, C, D) using four loading rates of manure effluent. Constituent values were normalized relative to their values (in mg  $kg^{-1}$ ) at the effluent loading rate of 0.4 L m<sup>-2</sup> d<sup>-1</sup>. Data points represent mean values of 4–9 measurements, and error bars are standard deviations.

0.30 (Cd), 75 (Cu), 1050 (Fe), 7,500 (K), 2500 (Mg), 105 (Mn), 6.0 (Mo), 43,000 (N), 11,000 (P), 3.4 (Pb), and 510 (Zn) (Table 3).

The recovery rate for a particular element is a function of algal productivity and the element's content in the biomass. In this case, the absolute amounts of different elements recovered in the algal biomass generally increased with loading rate because of increasing content rather than increasing productivity. However, percentage recovery for all elements peaked at a loading rate of 0.40 L m<sup>-2</sup> d<sup>-1</sup>, corresponding to the point of maximum productivity and nitrogen efficiency (not shown). Individual loading rates and percentage recoveries of different elements in the algal biomass at this loading rate are shown in Table 3. At this loading rate, average recoveries generally varied from 43–68% (Ca, K, Mg) to 77–95% (Al, Cd, Cu, Mn, N, P). In contrast, calculated recovery values for Fe, Mo and Zn exceeded 100% and the value for Pb exceeded 200% at this loading rate. Given these results, it is possible that there was undetected heterogeneity of the manure within the different 200 L storage containers which lead to underestimates of input loading rates. With regard to the very high Pb recovery values, it is also possible that the ATS units became contaminated with another source of Pb during these experiments.

#### **Discussion**

Algal productivity and elemental composition of algal turfs grown using low loading rates (<0.5 *g* TN  $m^{-2} d^{-1}$ ) of raw swine manure are roughly comparable to values previously obtained using turfs grown using similar loading rates of anaerobically digested dairy manure effluent (Figure 3). However, with increased loading rates of anaerobically digested dairy manure effluent, algal productivity increased significantly (up to a mean value of 16.5 g DW m<sup>-2</sup> d<sup>-1</sup> using 3.2 g TN  $m^{-2} d^{-1}$ ). In contrast, algal productivity remained <10 g DW m<sup>−</sup><sup>2</sup> d<sup>−</sup><sup>1</sup> at higher loading rates of raw swine manure effluent. We do not know the specific basis of this inhibition at higher loading rates. Levels of inorganic (such as sodium) and soluble organic compounds have been reported to be toxic to plants grown in wastewater (reviewed in Sooknah & Wilkie, 2005). We have not conducted comparable loading rate experiments using anaerobically digested swine manure and are unaware of any reports comparing algal productivity using raw versus anaerobically digested swine manure. However,

Loading rate $(L m^{-2} d^{-1})$ Element	Swine	Algal biamass (mg $Kg^{-1}$ ) <sup>a</sup>				Daily loading at 0.40L	Recovery in algal biomass at $0.40L$
	manure $(mg L^{-1})$	$0.24 \pm 0.03$ $(n = 6)$	$0.40 \pm 0.06$ $(n = 9)$	$0.62 \pm 0.08$ $(n = 5)$	$1.23 \pm 0.12$ $(n = 4)$	$m^{-2}d^{-1}$ $mg (m^{-2}d^{-1})$	$m^{-2}d^{-1}$ $(\%)$
$\mathbf N$	1130	$37000 \pm 6000$	$43000 \pm 5000$	$54000 \pm 5000$	$57000 \pm 3000$	$480 \pm 70$	$95 \pm 20$
K	410	$9800 \pm 650$	$7500 \pm 550$	$8200 \pm 800$	$8700 \pm 1650$	$174 \pm 40$	$43 \pm 6$
P	340	$8500 \pm 1300$	$11000 \pm 2000$	$13000 \pm 2000$	$18000 \pm 3000$	$142 \pm 37$	$77 \pm 13$
Ca	170	$4450 \pm 470$	$4900 \pm 420$	$5200 \pm 1200$	$6400 \pm 1300$	$72 \pm 17$	$68 \pm 12$
Mg	100	$2400 \pm 410$	$2500 \pm 230$	$2700 \pm 240$	$2800 \pm 650$	$42 \pm 10$	$58 \pm 8$
Fe	16.5	$1040 \pm 230$	$1050 \pm 50$	$1330 \pm 160$	$2160 \pm 30$	$7.0 \pm 1.6$	$147 \pm 28$
Zn	8.8	$470 \pm 85$	$510 \pm 70$	$620 \pm 100$	$1080 \pm 130$	$3.7 \pm 0.9$	$135 \pm 27$
Al	7.3	$290 \pm 80$	$250 \pm 20$	$390 \pm 50$	$750 \pm 50$	$3.0 \pm 0.8$	$82 \pm 19$
Mn	2.7	$130 \pm 25$	$105 \pm 15$	$145 \pm 20$	$255 \pm 20$	$1.1 \pm 0.3$	$90 \pm 17$
Cu	2.1	$75 \pm 20$	$75 \pm 15$	$105 \pm 15$	$200 \pm 10$	$0.89 \pm 0.21$	$87 \pm 18$
Mo	0.117	$5.9 \pm 1.5$	$6.0 \pm 0.7$	$9.3 \pm 2.1$	$9.2 \pm 0.6$	$0.05 \pm 0.01$	$119 \pm 19$
Pb	0.036	$6.2 \pm 2.0$	$3.4 \pm 0.9$	$3.3 \pm 1.0$	$3.8 \pm 0.9$	$0.016 \pm 0.004$	$230 \pm 71$
C <sub>d</sub>	0.009	$0.53 \pm 0.16$	$0.30 \pm 0.04$	$0.31 \pm 0.06$	$0.51 \pm 0.08$	$0.004 \pm 0.0001$	$78 \pm 18$

*Table 3.* Elemental composition of swine manure and dried algal biomass grown using four manure effluent loading rates.

<sup>a</sup>Values are means  $\pm$  SD of measurements over four to nine harvest cycles. *n*, number of harvests.



*Figure 3*. Comparison of algal production values using laboratory scale ATS units and raw swine manure effluent or anaerobically digested dairy manure effluent. Productivity is shown as a function of NH4-N loading using raw swine manure effluent (this study) and anaerobically digested dairy manure effluent (Westhead et al., 2003).

previous studies describing cultivation of suspended algae with swine effluent after anaerobic pretreatment have reported productivity levels of up to 15–20 DW  $m^{-2}$  d<sup>-1</sup> in outdoor ponds (Goh, 1986; Olguin et al., 1997).

N:P values of the dried algal biomass in this study ranged from 3.2 to 4.3 and are considerably lower than corresponding values from algal biomass grown using anaerobically digested dairy manure effluent (Kebede-Westhead et al., 2003, 2004). These differences reflect the lower N:P ratio of the swine manure (N:P ratio of 3.3) compared to the dairy manure (N:P ratio of

8.2). Indeed, comparison of the elemental composition (relative to N) of the two types of effluents and the respective algal biomasses cultivated with them show close correspondence for Al, Cu, Fe, Mn, Mo, P, and Zn (not shown).

The maximum tolerable dietary levels (MTDL) in dairy cow feed for elements measured in this study are  $(in \, mg \, kg^{-1})$ : (Al (1000), Cd (0.5), Fe (1000), Mo (5), Mn (1000), Pb (30), and Zn (500) (NRC, 2001). These levels are based on the use of highly bioavailable soluble salts of these metals. In this study, levels of algal Fe, Mo, and Zn (with mean values of 1050, 6, and 510 mg  $kg^{-1}$ , respectively, at 0.40 L m<sup>-2</sup> d<sup>-1</sup>, the optimal loading rate for algal production) exceeded the MTDL. We have no information about the solubility or bioavailability of any of the constituents in the algal biomass. However, as a potential feed component, dried algal biomass would only constitute a small portion of the total feed (Franklin et al., 1999) and thus these levels of Fe, Mo, and Zn in the product would be unlikely to reduce its value as a feed. With regard to the use of the biomass as a feed, we have not characterized the nutritional quality of the dried algal biomass. Results from limited previous studies using specific species of benthic freshwater have shown variable content of crude protein and ash, as well as different degrees of *in vitro* digestibility (reviewed in Wilkie and Mulbry, 2002).

Dried algal biomass from manure treatment could be used on-farm or marketed off-farm as a slow-release organic fertilizer (Mulbry et al., 2005). Results from a 42 day flask study using soils amended with dried algal biomass from treatment of digested dairy manure effluent showed that approximately 3% of total algal N was present as plant available N ( $NH_4$ -N and  $NO_3$ -N) at day 0. Approximately 33% of algal N was converted to plant available N within 21 days at 25 ◦C. Subsequent plant growth experiments using corn and cucumber seedlings showed that seedlings grown in algae-amended potting mixes were equivalent to those grown with comparable levels of commercial fertilizer-amended potting mixes with respect to seedling dry weight and nutrient content. Nitrogen mineralization and plant growth studies using algal biomass from treatment of swine manure effluent are currently underway.

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