

Biofertilizers from Algal Treatment of Dairy and Swine Manure Effluents: Characterization of Algal Biomass as a Slow Release Fertilizer

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ABSTRACT. An alternative practice to land spreading of manure effluents is to grow crops of algae on the nitrogen (N) and phosphorus (P) present in these liquid slurries. The overall environmental and economic values of this approach depend, in part, on the use and value of the resulting algal byproduct. Among the potential uses of algal biomass from such systems is its use as a slow release fertilizer. The objective of this study was to evaluate the fertilizer value of algae that had been grown in laboratory (indoor) and pilot scale (outdoor) algal turf scrubbers using raw dairy manure effluent, anaerobically digested dairy manure effluent and raw swine manure effluent for vegetable production. Results from a multifactorial N-mineralization experiment using soil amended with eight algal biomass treatments showed that approximately 5% of total

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algal N was present as plant available N at day 0. After 21 and 63 days, the total algal N present as mineral N increased to 25-29% and 36-41%, respectively. Approximately 40% of total algal P was present as Mehlich-3 extractable P throughout the 63-day incubations. Results from plant growth experiments showed that 17 day-old corn (*Zea mays* L.) seedlings grown in algae-amended potting mixes were equivalent to those grown with comparable levels of fertilizer-amended potting mixes with respect to shoot dry weight and nutrient content. There were no differences in the fertilizer value of different batches of algae at the low rate amendment (~ 47 kg of available N·ha⁻¹). However, at the high rate amendment (~ 93 kg of available N·ha⁻¹) shoot biomass, and shoot N and P contents were greatest for treatments containing algae grown using indoor laboratory scale algal turf scrubbers (ATS) units and least for treatments with algae grown outdoors using pilot scale ATS units. Longer term field studies are needed to assess the effect of algal biomass amendment on corn and vegetable yields under a range of growth conditions. doi:10.1300/J484v12n04_08 [Article copies available for a fee from The Haworth Document Delivery Service: 1-800-HAWORTH. E-mail address: <docdelivery@haworthpress.com> Website: <<http://www.HaworthPress.com>> © 2006 by The Haworth Press, Inc. All rights reserved.]

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INTRODUCTION

Controlling the input of nitrogen (N) and phosphorus (P) from dairies and other livestock operations into adjacent aquatic systems, and into the atmosphere poses both technical and economic challenges to the agricultural community (Kaiser, 2001; Van Horn et al., 1994). During storage and land application of manure effluents, large amounts of N are lost to the atmosphere due to volatilization of ammonia (Thompson and Meisinger, 2002). Environmentally and economically sound manure management on farms is vital to sustainability of animal operations and to minimize off-farm losses of valuable plant nutrients.

An alternative practice to land spreading of manure effluents is to grow crops of algae on the N and P present in these liquid slurries and convert manure N and P into algal biomass for use as a fertilizer. When considered in conjunction with anaerobic digestion/co-generation systems where energy is recovered from the manure, algal systems are ideally

sued for capturing N and P from the liquid residuals, and CO₂ from boiler or generator exhaust gases (Wilkie and Mulbry, 2002). Previous studies have demonstrated the use of algal-based treatment systems to remove N and P from dairy and swine manure effluents (Craggs et al., 2003; Goh, 1986; Kebede-Westhead et al., 2003, 2006; Lincoln et al., 1996; Olguin et al., 2001; Wilkie and Mulbry, 2002). However, the overall environmental and economic value of this approach depends, in part, on the use and value of the resulting algal byproduct (Pizarro et al., 2006). The use of blue green algae as soil conditioning amendments and as biofertilizers for rice cultivation has been reported previously (Metting, 1996; Metting et al., 1990). However, there has been very limited research on the fertilizer value of algal biomass derived from the treatment of animal manure. Results using a single sample of algal biomass from laboratory-scale treatment of anaerobically digested dairy manure showed that approximately one-third of algal N was converted to plant available N within 21 days at 25°C (Mulbry et al., 2005). Corn and cucumber (*Cucumis sativus* L.) seedlings grown in potting mixes amended with this algal biomass were equivalent to those grown with comparable levels of fertilizer-amended potting mixes with respect to shoot dry weight and nutrient content. Although these results were encouraging, it was unknown whether these results could be extrapolated to the use of algae that had been grown indoors using other types of animal manure or cultivated outdoors in larger scale systems. The objective of this study was to evaluate and compare the fertilizer values of dried algal biomass produced during algal treatment of different types of manure effluent (raw dairy, anaerobically digested dairy, and raw swine) under laboratory (indoor) and pilotscale (outdoor) conditions.

MATERIALS AND METHODS

Production of algal biomass. Algal biomass was produced using indoor laboratory-scale algal turf scrubbers (ATS) (Adey and Loveland, 1998) with raw and anaerobically digested dairy manure effluent collected from the Dairy Research Unit of the USDA's Beltsville Agricultural Research Center in Beltsville, Maryland and raw swine manure effluent from a swine finishing operation near Richlands, North Carolina (Kebede-Westhead et al., 2006) with manure loading rates corresponding to approximately 0.5-2 g TN/m²/day as previously described (Kebede-Westhead et al., 2003, 2006). In addition, algal biomass was produced from outdoor pilotscale raceways (1 × 30 m surface area, 2% slope,

93 L·min⁻¹ flow rate) using the Beltsville raw and anaerobically digested dairy manure. The characteristics of Beltsville dairy manure effluent have been described (Wilkie and Mulbry, 2002). Wet algal biomass was harvested using a wet/dry vacuum, dewatered by sieving the harvested material through 2 mm mesh nylon netting (Aquatic Ecosystems, Apopka, FL) to approximately 10% solids content, then air dried for 48 h using electric fans to approximately 90% solids content. The dried biomass was ground in a Wiley Mill to pass a 3 mm sieve and stored in sealed plastic bags at 20-25°C. Elemental compositions of algal biomass samples were determined using induced coupled plasma analysis. Garden-tone 4-6-6 fertilizer (Espoma Co., Millville, NJ) used in growth chamber experiments was analyzed for total N and P content by flow injection analysis (model 8000, Lachat Instruments, Milwaukee, WI) after total Kjeldahl block digestion. The product label indicated 4% TN (2.5% ammonia-N; 0.3% other water soluble N; 1.2% water insoluble N, 6% available phosphate (P₂O₅) and 6.0% soluble potash (K₂O).

Algal N mineralization rate determination. Two laboratory incubation studies were conducted to determine the N mineralization rates of dried algal biomass samples in Corderus silt loam soil (Sikora and Enkiri, 2003). The soil was air-dried, crushed and passed through a 2 mm sieve. Its chemical constituents are presented in Table 1. Aliquots of 50 g of this soil were amended with different samples of algal biomass (each sample corresponding to 22.5 mg TN) and transferred to 125 ml flasks (Bartha and Pramer, 1965). Each algal sample was from an individual weekly harvest of either an indoor or outdoor algal turf scrubber (ATS). CaO was added (17 mg per flask) to achieve a soil pH of 7.0. This procedure was repeated for all of the treatment flasks. An equal number of control flasks containing Corderus soil plus CaO were also prepared. All flasks were brought to -33 kPa moisture, weighed and incubated at 25°C. Flasks

TABLE 1. Constituents of Corderus soil.²

Constituent	Value
pH	5.7
Total Organic Carbon	12.6 g·kg ⁻¹
Total Nitrogen	1.0 g·kg ⁻¹
Total Phosphorus	0.37 g·kg ⁻¹
Mehlich-3 Extractable P	0.009 g·kg ⁻¹
Water Extractable P	0.0002 g·kg ⁻¹

²Adapted from Mulbry et al., 2005.

were weighed weekly and adjusted to initial weights with distilled water as needed to maintain constant moisture. Three control flasks and three flasks from each treatment were sampled at 0, 21, 42 and 63 days.

The elemental composition of the algal samples used in these experiments are shown in Table 2 along with the type of manure effluent and ATS (indoor or outdoor) used for their production. The first experiment was conducted using five representative samples of algal biomass obtained from indoor laboratory scale treatment using raw or anaerobically digested dairy manure effluent. The experiment included three samples of algal biomass (containing 3.6, 5.4 or 7.7% algal N) from treatment of raw dairy manure effluent and two samples of algal biomass (containing 3.3 or 6.4% algal N) from treatment of digested dairy manure effluent (Table 2). A second mineralization experiment was conducted to determine mineralization rates of algae grown in outdoor raceways. The experiment included three samples of algal biomass (containing 2, 4 or 6% algal N) from treatment of raw dairy manure effluent (Table 2). In each experiment, the amendments corresponded to total loadings (in $\text{mg}\cdot\text{kg}^{-1}$) of 450 N and 56-82 P (depending on the biomass used).

For determination of nitrate-N and ammonium-N, a 10 g sample from each flask was extracted with 100 ml 2 M KCl on a rotary shaker for 30 minutes. Extracts were filtered using 0.45 μm membrane filters and filtrates were adjusted to pH 3-5 with 2 M H_2SO_4 as needed for preservation. Filtrates were stored frozen until analysis. Ammonium-N and nitrate-N were determined colorimetrically by flow injection analysis (Lachat). For determination of Mehlich-3 extractable P, 3 g soil samples were extracted with Mehlich-3 extractant (Mehlich, 1984; Wolf and Beegle, 1995) following the procedure outlined here, but with no adjustment of extractant pH. Orthophosphate was determined colorimetrically by flow injection analysis (Lachat). Moisture analyses of samples were determined on 10 g samples dried overnight at 105°C.

Growth chamber study. Based on the 21-day net mineralizable N value of 25% determined from the mineralization part of this study (described earlier), a growth chamber experiment using sweet corn was conducted to compare plant growth and nutrient uptake in a potting mix amended with treatments which included either (1) a commercial low-strength fertilizer (Garden tone 4-6-6), or (2) algal biomass from indoor laboratory scale ATS units treating raw swine manure effluent, raw dairy manure effluent and anaerobically digested dairy manure effluent, or (3) algal biomass from outdoor pilot scale ATS units treating raw and anaerobically digested dairy manure effluents. The elemental compositions of the

TABLE 2. Elemental composition (in mg·kg⁻¹) of algal biomass samples used for N-mineralization experiments. Samples were from single weekly harvests.

Constituent	Indoor Algal biomass					Outdoor algal biomass		
	Raw Dairy			Manure type		Raw Dairy		
	AT ^z -RD-3	AT ^z -RD-5	AT ^z -RD-8	AT ^z -DD-3	AT ^z -DD-6	OR-RD-2	OR-RD-4	OR-RD-7
N	35,900	54,300	76,600	32,700	63,600	22,900	39,700	72,300
P	6400	7700	9500	6000	9100	4300	5500	11,800
K	12,500	11,500	9000	8900	11,100	n.d. ^y	n.d.	n.d.
Ca	5500	5400	5600	5500	5800	n.d.	n.d.	n.d.
Mg	2700	2600	2800	2400	2800	n.d.	n.d.	n.d.
Fe	1420	790	1060	1500	1700	n.d.	n.d.	n.d.
Al	590	430	360	510	680	n.d.	n.d.	n.d.
Zn	410	360	450	560	540	n.d.	n.d.	n.d.
Mn	370	230	290	270	440	n.d.	n.d.	n.d.
Cu	110	110	100	120	130	n.d.	n.d.	n.d.
Si	90	35	80	135	140	n.d.	n.d.	n.d.
Pb	1.9	4.5	3.3	3.9	6.8	n.d.	n.d.	n.d.
Mo	0.8	1.6	1.5	2.2	1.9	n.d.	n.d.	n.d.
Cd	0.2	0.6	0.3	0.3	0.5	n.d.	n.d.	n.d.
Ash (%)	10	11	11	10	11	14	18	16

^zSamples AT^z-RD-3, AT^z-RD-5, and AT^z-RD-8 were grown in indoor laboratory scale AT^z units using raw dairy manure effluent; AT^z-DD-3 and AT^z-DD-6 were grown in indoor laboratory scale AT^z units using digested dairy manure effluent; OR-RD-2, OR-RD-4, and OR-RD-7 were grown in outdoor pilot scale raceways using raw dairy manure effluent.

^yn.d.= not determined.

fertilizer and algal biomass samples are presented in Table 3 along with the type of manure and ATS (indoor or outdoor) used for algal production. In these experiments, 15 cm plastic pots containing a peat-based potting mix (6.1 g CaO, 5 g perlite, 240 g peat moss (Premier Horticulture, Quebec, Canada) per pot) were amended with two rates of algal biomass or two rates of fertilizer. Each mixture was wetted with 550 ml distilled water to achieve approximately -33 kPa, and the wetted mix was added

TABLE 3. Elemental composition (in mg·kg⁻¹) of algal biomass samples and Garden-tone 4-6-6 fertilizer used in plant growth experiments.

Constituent	Manure effluent type					Fertilizer
	Indoor algal biomass			Outdoor algal biomass		
	Raw Dairy ^z	Digested Dairy ^z	Raw Swine ^z	Raw Dairy ^y	Digested Dairy ^y	
	ATS-RD ^x	ATS-DD	ATS-RS	OR-RD	OR-DD	
N	45,300	58,800	59,000	49,100	47,800	48,000
P	7900	7400	12,500	8000	7400	25,200
K	8800	6100	7800	7200	6400	49,800
Ca	6600	9500	5100	11,900	11,900	30,000
Mg	2900	3500	2700	3100	5200	5000
Fe	1050	1700	1200	3000	3500	10,000
Al	500	690	440	1980	3050	n.d. ^w
Zn	350	460	620	560	430	500
Mn	280	380	140	450	540	500
Cu	100	120	105	155	135	500
Si	90	90	50	80	95	n.d.
Pb	3.2	5.1	7.6	8.4	6.2	n.d.
Mo	2.0	2.0	5.4	1.1	1.6	5
Cd	0.4	0.5	0.5	0.8	0.7	n.d.
Ash (%)	10	12	7	15	19	n.d.

Note. Algal biomass was grown in indoor laboratory scale ATS units (using raw dairy, anaerobically digested dairy, and raw swine manure effluents) and in outdoor raceways (using raw and anaerobically digested dairy manure effluents). Constituent values for ATS biomass were determined in our laboratory. Fertilizer N and P values were determined in our laboratory. Other fertilizer constituent values are from the product label.

^z Values are averages of two pooled samples. Each pooled sample was composed of biomass from 3-6 weekly harvests.

^y Values are averages of two samples. Each sample was from a single weekly harvest.

^x Sample designation.

^w n.d.= not determined.

to pots. Pots (five per treatment) containing five corn seeds ("How sweet it is," Wetsel Inc., Harrisonburg, VA) each were placed in a growth chamber maintained on a 25°C/20°C 16 h day/8 h night cycle. Light intensity from a mixture of high pressure sodium and metal halide lamps was approximately $600 \mu\text{mol}\cdot\text{s}^{-1}\cdot\text{m}^{-2}$. After seven days, only two seedlings/pot were maintained. The 23 treatments consisted of two rates (370 and 740 mg algal TN/pot, corresponding to field application rates of 47 and 93 $\text{kg}\cdot\text{ha}^{-1}$ of available N, respectively) of five different algal biomass products, two rates of fertilizer (3.3 and 6.6 g fertilizer per pot, corresponding to field application rates of 47 and 93 $\text{kg}\cdot\text{ha}^{-1}$ of available N, respectively), and control pots containing non-amended potting mix.

Pots were manually watered every 24–48 h and adjusted to initial weights with distilled water as needed to maintain initial moisture content. After 17 days, shoots were harvested by cutting at the soil surface, dried for 48 h at 70°C in a forced-air oven, weighed and ground in a Wiley Mill. Ground samples were analyzed by flow injection analysis for total N and P content after total Kjeldahl block digestion.

Statistical analyses. The mineral N data for 21, 42 and 63 days in the first mineralization experiment was analyzed as a three-factor general linear mixed model where treatment and day were the fixed factors and variance group was a random factor. The assumptions of the general linear model were tested and the variance grouping technique was used to correct for variance heterogeneity. The biomass and plant N data from the plant growth experiment were analyzed separately as four-factor general linear mixed models using PROC Mixed in SAS (SAS Institute, 2004). The fixed factors included AN (applied nitrogen), Env (ATS or outdoor raceway (OR)) and Manure. Variance group was a random factor. Mean comparisons were done with Sidak adjusted p-values to hold the experiment-wise error at 0.05 (SAS Institute, 2004).

RESULTS

Mineralization of ATS biomass N and P in Codorus soil. Algal biomass production and nutrient content generally increase with increasing manure effluent loading rate, but are also influenced by the type of effluent being treated and incident light levels, photoperiod and flow rate. As a first step in comparing the fertilizer value of algal samples that had been grown using different effluents and environmental conditions, soil incubation studies were conducted to determine net mineralizable N and plant

available P of representative algal samples. The first experiment was conducted using five representative samples of algal biomass obtained from indoor laboratory-scale treatment using raw or anaerobically digested dairy manure effluent (Table 4). The results showed that approximately 1 to 4% of the total algal N was present as mineral N at day zero (Table 5). After 21, 42 and 63 days, mean values of total algal N present as mineral N increased to 25, 32 and 41%, respectively (Table 5, Figure 1A). Although there were statistically significant differences ($P = 0.05$) between the mineralization values from different treatments at each

TABLE 4. Two-way ANOVA testing effects of amendment and time on levels of mineral N (NH_4 and NO_3) in a limed Codorus soil amended with one rate of five samples of algal biomass grown in laboratory scale algal scrubbers using raw or anaerobically digested dairy manure effluent. Day zero values were not included in the statistical analysis.

Source	df	F-value	p-value
Treatment (T)	5	219.31	<0.0001
Day (D)	2	27.42	<0.0001
T × D	10	16.55	<0.0001

TABLE 5. Comparison of mean levels of mineral N (NH_4 and NO_3) (mg N/flask) over time in a limed Codorus soil amended with algal biomass samples (corresponding to 22.5 mg algal N/flask) grown in indoor laboratory scale algal scrubbers using raw or anaerobically digested dairy manure effluent (sample designations are given in Table 2).

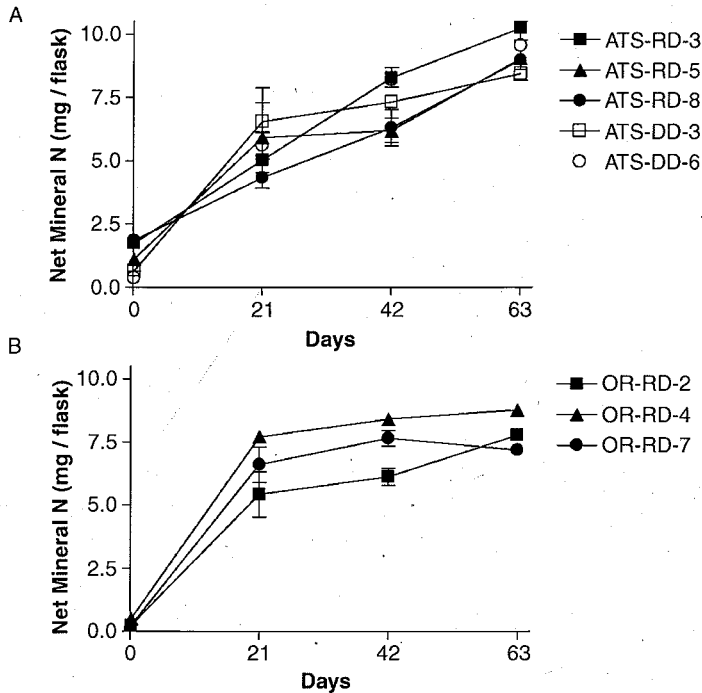
Treatment	Day ^z			
	0	21	42	63
ATS-RD-3	5.3	9.9a ^y y ^x	13.2axy	14.8ax
ATS-RD-5	4.7	10.9axy	11.2ay	13.8ax
ATS-RD-8	5.5	9.1az	11.3ay	13.7ax
ATS-DD-3	4.3	11.3ax	13.3ax	13.3ax
ATS-DD-6	4.0	10.5ay	14.1ax	14.4ax
0	3.6	4.8bx	5.0bx	4.7bx

^z Day zero values were not included in the statistical analysis.

^y Treatment means within Day with different a, b letters are different at the 0.05 significance level.

^x Day means within Treatment with different x, y, z letters are different at the 0.05 significance level.

FIGURE 1. Net mineral N ($\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$) in Codorus soil amended with one rate (corresponding to 22.5 mg TN) of representative samples of algal biomass grown using dairy manure and indoor laboratory scale ATS units (A) or outdoor pilot scale raceways (B). Samples ATS-RD-3, ATS-RD-5, and ATS-RD-8 were grown in indoor laboratory scale ATS units using raw dairy manure effluent; ATS-DD-3 and ATS-DD-6 were grown in indoor laboratory scale ATS units using digested dairy manure effluent; OR-RD-2, OR-RD-4, and OR-RD-7 were grown in outdoor pilot scale raceways using raw dairy manure effluent. Net values (mean \pm std error) were calculated at each time point as treatment values minus values from flasks containing no amendment.



timepoint (Table 5), there was no consistent pattern in these values relative to algal N content, or to the type of manure effluent used to grow the algae. In all amended flasks approximately 38% (\pm 5%) of the total algal P was present as Mehlich-3 extractable P (a measure of plant available P) at day zero and this value increased only slightly (to 41 \pm 5%) of the total algal P was 4%) during the 63-day incubation period (data not shown).

Results from the experiment to determine mineralization rates of algal samples grown using outdoor raceways indicated that initial rates and amounts of N mineralization were similar to those obtained from indoor-produced algal biomass ($29 \pm 4\%$ N mineralized within 21 days) (Table 6, Figure 1B). However, after 21 days, the level of N mineralization increased only slightly to $36 \pm 5\%$ on day 63. Results also indicated lower levels of plant available P compared to results from experiments using indoor-grown algal biomass. Approximately $25\% (\pm 4\%)$ of the total algal P was present as Mehlich-3 extractable P at day zero; and this value increased to $35 \pm 5\%$ on day 63 (data not shown).

Seedling growth. In all treatments, shoot mass and nutrient content increased with increased algal or fertilizer amendment (Tables 7-11, Figure 2). Shoot mass and N content values were highest in potting mixes amended with algal biomass from indoor ATS units. Although these values were also influenced by the type of manure effluent used to grow the algae in indoor ATS units (with raw dairy manure > digested dairy manure > swine manure), the type of manure effluent (raw or digested dairy manure) had no effect with regard to values from mixes using algae grown in outdoor raceways (Tables 8-11). With one exception, there was no significant difference ($P < 0.05$) in shoot biomass or shoot N between algal or fertilizer-amended potting mixes based on available N at a low amendment rate (93 mg available N per pot; equivalent to approximately $47 \text{ kg}\cdot\text{ha}^{-1}$ of available N) (Table 7, Figure 2). At a high amendment rate (185 mg available N per pot; equivalent to approximately $93 \text{ kg}\cdot\text{ha}^{-1}$ of available N algal amendment yielded equal or higher levels of shoot biomass and N content compared to that in the

TABLE 6. Comparison of mean levels of mineral N (NH_4 and NO_3) (mg N/flask) over time in limed Codorus soil amended with algal biomass samples (corresponding to 22.5 mg algal N/flask) grown in pilot scale outdoor raceways using raw dairy manure.

Treatment	Day ^z			
	0	21	42	63
OR-RD-2	1.9	9.4a ^y x [*]	10.7by	12.7bx
OR-RD-4	2.2	11.7ay	13.0ax	13.7ax
OR-RD-7	1.9	10.6ax	12.3abx	12.1bx
0	1.6	4.0bz	4.6cy	4.9cx

^z Day zero values were not included in the statistical analysis.

^y Treatment means within Day with different a, b letters are different at the 0.05 significance level.

^x Day means within Treatment with different x, y, z letters are different at the 0.05 significance level.

TABLE 7. Mass and nutrient values of corn shoots grown in potting mix amended with two rates of algal biomass from indoor algal turf scrubber units (ATS biomass), algal biomass from outdoor raceways (OR biomass), or fertilizer (mean \pm std error).

	Amendment												None
	----- ATS biomass -----						----- OR biomass -----						
	Manure source												
	--- Raw dairy ---	- Digested dairy -	-- Raw swine --	-- Raw dairy --	- Digested dairy -	---	---	---	---	---	---	---	
--- ATS-RD ^z ---	--- ATS-DD ---	--- ATS-RS ---	--- OR-RD ---	--- OR-DD ---	---	---	---	---	---	---	---	---	---
Amount applied (g/pot)	9.2	18.4	7.2	14.4	6.6	13.2	8.1	16.2	8.3	16.6	3.3	6.6	0
Applied N (mg/pot)	370	740	370	740	370	740	370	740	370	740	132	264	0
Available N (mg/pot) ^y	93	185	93	185	93	185	93	185	93	185	93	185	0
Applied P (mg/pot)	67	135	49.5	99	77	155	58	116	56	113	56	112	0
Biomass (g DW/pot)	1.26 \pm 0.06	2.12 \pm 0.16	0.81 \pm 0.04	1.63 \pm 0.09	0.74 \pm 0.03	1.36 \pm 0.06	0.80 \pm 0.03	1.22 \pm 0.06	0.92 \pm 0.03	1.20 \pm 0.05	0.66 \pm 0.07	1.42 \pm 0.05	0.35 \pm 0.01
N Content (%)	4.4 \pm 0.4	4.1 \pm 0.3	5.8 \pm 0.2	5.2 \pm 0.2	5.6 \pm 0.2	5.2 \pm 0.4	5.4 \pm 0.4	5.1 \pm 0.4	5.6 \pm 0.4	5.2 \pm 0.3	4.5 \pm 0.4	3.5 \pm 0.4	3.2 \pm 0.2
Total Plant N (mg/pot)	55.3 \pm 2.1	87.5 \pm 5.4	47.8 \pm 3.3	82.0 \pm 5.5	42.0 \pm 2.0	70.9 \pm 4.6	44.3 \pm 1.2	64.4 \pm 2.9	51.6 \pm 1.1	59.2 \pm 1.3	29.1 \pm 1.9	47.2 \pm 1.1	11.4 \pm 0.24

Net Plant N/Applied N (%)	11.9 ± 0.6	10.3 ± 0.7	9.8 ± 0.9	9.5 ± 0.7	8.3 ± 0.5	8.0 ± 0.6	8.9 ± 0.3	7.2 ± 0.4	10.9 ± 0.3	6.5 ± 0.2	13.5 ± 0.9	13.6 ± 0.3	- ^x
P Content (%)	1.2 ± 0.1	1.3 ± 0.1	1.3 ± 0.2	1.3 ± 0.1	1.7 ± 0.1	1.7 ± 0.1	1.3 ± 0.1	1.3 ± 0.1	1.3 ± 0.1	1.4 ± 0.1	1.4 ± 0.1	1.2 ± 0.1	0.2 ± 0.01
Total Plant P (mg/pot)	14.4 ± 0.4	26.2 ± 1.2	10.1 ± 0.3	20.3 ± 0.8	12.3 ± 0.6	22.6 ± 0.9	10.0 ± 0.26	15.7 ± 0.4	11.5 ± 0.4	16.2 ± 0.5	8.9 ± 0.6	16.7 ± 0.5	0.81 ± 0.02
Net Plant P/ Applied P (%)	19.6 ± 0.9	18.8 ± 0.8	18.2 ± 0.7	20.9 ± 0.8	13.5 ± 0.7	14.0 ± 0.6	15.9 ± 0.5	12.8 ± 0.4	19.2 ± 0.8	12.9 ± 0.8	14.5 ± 1.2	14.2 ± 0.5	-

^z Sample designation.

^y Available N values were calculated as 25% and 70% of TN for algal biomass (ATS and OR) and fertilizer, respectively.

^x - = Non-amended control.

TABLE 8. Three-way ANOVA testing effects of applied available nitrogen, algal growth environment (indoors or outdoors), and type of dairy manure effluent used to grow algae on corn shoot biomass grown using potting mix amended with two rates of algal biomass from indoor algal turf scrubber units (ATS biomass) or algal biomass from outdoor raceways (OR biomass).

Source	df	F-value	p-value
Available N (AN)	1	103.70	<0.0001
Environment (E)	1	50.87	<0.0001
Manure (M)	1	18.55	<0.0001
AN × E	1	21.26	<0.0001
AN × M	1	1.74	0.1937
E × M	1	18.18	<0.0001
AN × E × M	1	0.13	0.7238

TABLE 9. Means comparisons of corn shoot biomass using potting mix amended with two rates of algal biomass from indoor algal turf scrubber units (ATS biomass) or algal biomass from outdoor raceways (OR biomass). Algae was grown using raw or anaerobically digested dairy manure effluent.

	93 mg N per pot	185 mg N per pot	Raw dairy	Digested dairy
ATS	0.996a ^z y ^y	1.850ax	1.607a ^x x ^w	1.176ay
OR	0.851ay	1.172bx	1.013bx	1.010ax

^z Env means within AN with different a, b letters are different at the 0.05 significance level.

^y AN means within Env with different x, y letters are different at the 0.05 significance level.

^x Env means within Manure with different a, b letters are different at the 0.05 significance level.

^w Manure means within Env with different x, y letters are different at the 0.05 significance level.

comparable rate of fertilizer amendment (Figure 2). Net corn shoot N represented 26-47% of available algal N and 19% of available fertilizer N. Net corn shoot P represented 13-21% of algal P and 14% of fertilizer P (Table 7).

DISCUSSION

Previous mineralization and plant growth experiments were based on a single algal biomass sample produced in an indoor laboratory scale ATS unit treating digested dairy manure effluent. Mineralization results from a flask study using two soils amended with that algal biomass showed that plant available N increased from 3% of total algal N at day

TABLE 10. Three-way ANOVA testing effects of applied available nitrogen, algal growth environment (indoors or outdoors), and type of dairy manure effluent used to grow algae on corn shoot nitrogen levels grown using potting mix amended with two levels of algal biomass from indoor algal turf scrubber units (ATS biomass) or algal biomass from outdoor raceways (OR biomass).

Source	df	F-value	p-value
Available N (AN)	1	76.47	<0.0001
Environment (E)	1	23.11	<0.0001
Manure (M)	1	0.68	0.4136
AN × E	1	14.36	0.0004
AN × M	1	0.44	0.5093
Env × M	1	2.25	0.1389
AN × E × M	1	1.12	0.2939

TABLE 11. Means comparisons of corn shoot nitrogen levels using potting mix amended with two rates of algal biomass from indoor algal turf scrubber units (ATS biomass) or algal biomass from outdoor raceways (OR biomass). Algae was grown using raw or anaerobically digested dairy manure effluent.

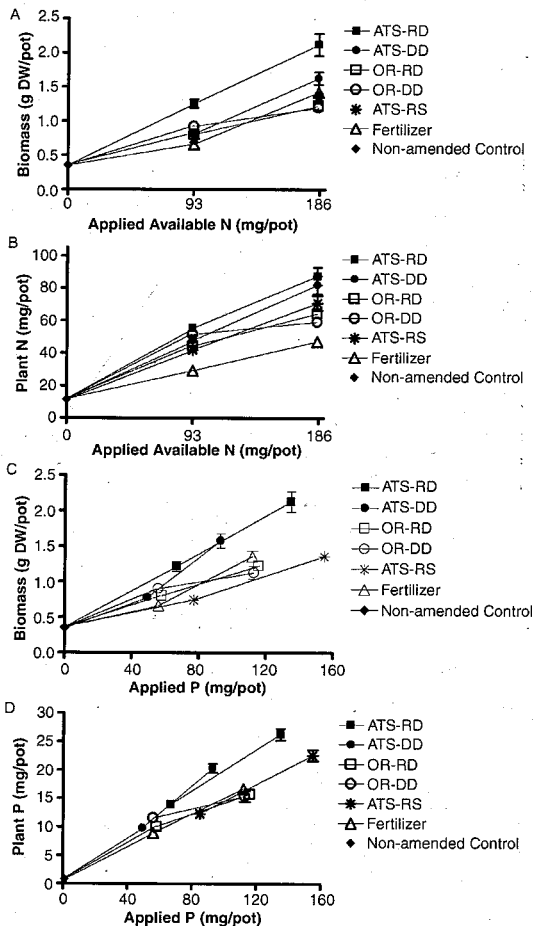
	93 mg N per pot	185 mg N per pot
ATS	49.42a ^z y ^y	84.76ax
OR	46.55ay	60.52bx

^z Env means within AN with different a, b letters are different at the 0.05 significance level.

^y AN means within Env with different x, y letters are different at the 0.05 significance level.

0 to 33% within 21 days at 25°C in both soils. Levels of water soluble P and Mehlich-3 extractable P in the two soils were influenced by existing soil P levels but increased as the rate of algal amendment increased. In a phosphorus-deficient Codorus soil (also used in this study), water soluble P and Mehlich-3 extractable P accounted for approximately 5 and 39% of applied algal P, respectively (Mulbry et al., 2005). In the present study, comparable amounts of N and P mineralization were obtained in Codorus soil using a broad set of representative algal samples grown using different types of dairy and swine manure in indoor and outdoor systems. Taken together, results from these two studies suggest that algal biomass from ATS manure treatment systems may have consistent and predictable mineralization values in several soils. Although this prediction remains to be tested using a variety of soils under different environmental conditions, if correct, it would stand in contrast to the

FIGURE 2. Shoot mass (A, C), seedling N (B), and seedling P (D) values of 17-day old corn seedlings grown in an unamended potting mix and in potting mix amended with two rates of fertilizer or two rates of algal biomass from indoor laboratory scale (ATS) and outdoor pilot scale (OR) algal units using raw dairy, anaerobically digested dairy and raw swine manure effluents (mean \pm std error). Algal samples ATS-RD, ATS-DD, and ATS-RS were grown in indoor laboratory scale ATS units using raw dairy, digested dairy, and raw swine manure effluents, respectively. Algal samples OR-RD and OR-DD were grown in outdoor pilot scale raceways using raw dairy and digested dairy manure effluents, respectively.



highly variable and unpredictable results found using manured soils (Van Kessel and Reeves, 2002; Calderon et al., 2004). The current unpredictability of N mineralization potentials among manures severely limits the ability of producers to efficiently use the organic fraction of manure N.

Results from corn growth experiments demonstrate that algal biomass cultivated in indoor or outdoor systems using dairy or swine manure was equal to fertilizer in supplying N and P to corn seedlings in 17-day growth chamber studies. Algal fertilizers possess a number of distinct environmental advantages compared to conventional fertilizers and manure. In addition to the potential predictability of algal fertilizers in different soils (discussed previously), algal biomass constitutes a stable, transportable and highly concentrated form of transformed manure nutrients. For example, compared to the relatively concentrated manure effluents (containing 0.1 to 0.25% TN) used to cultivate the algae described in this study, the resulting dry algal biomass typically contained 5-7% N (a 20- to 70-fold concentration of N compared with the effluents). Moreover, algal N would have much less potential for leaching or for loss in run-off since only about 5% of the algal N would be available as mineral N at the time of application. Applying dried algal biomass to soils would not result in NH_3 volatilization as is the case with manures (Thompson and Meisinger, 2002). Preliminary results from other experiments show that the fertilizer value of algal biomass applied to the soil surface is roughly 60-70% of the value of algal biomass incorporated into soil. This benefit may allow algal biomass to be side-dressed into established crops. However, longer term field studies are needed to assess the effect of algal biomass amendment on corn and vegetable yields under a range of growth conditions.

A recent assessment of the nutrient recovery potential and economic cost of an on-farm algal treatment system to treat dairy manure effluent reported that under the best case, the yearly operational costs per cow, per kg N, per kg P, or per kg of dried biomass were \$454, \$6.20, \$31.10 and \$0.70, respectively (Pizarro et al., 2006). These costs did not include any value for the algal biomass. For perspective, a recent survey of 36 Maryland dairy farms found long-term annual profits of about \$500 per cow (Johnson et al., 2005). Based on this relative value, projected manure treatment costs are very high and would consume or exceed most profit. However, the economic balance becomes more favorable if values from algae as a byproduct (e.g., fertilizer sale) and/or the value of nutrient recovery from the watershed can be realized. In regard to the value of manure grown algae as an organic fertilizer, the retail, bagged-product

consumer market is the most attractive with respect to price with retail prices of \$2-3·kg⁻¹ for comparable organic fertilizers. However, the pricing at which the dried algal biomass could successfully penetrate this market is unknown.

LITERATURE CITED

- Adey, W. H. and K. Loveland. 1998. *Dynamic Aquaria: Building Living Ecosystems*. Academic Press, New York.
- Bartha, R. and D. Pramer. 1965. Features of a flask and method for measuring the persistence and biological effects of pesticides in soil. *Soil Sci.* 100:68-70.
- Calderon, F. J., J. B. Reeves, G. W. McCarty, and J. S. Van Kessel. 2004. Carbon and nitrogen dynamics during incubation of manured soil. *Soil Science Society America Journal* 68(5):1592-1599.
- Craggs, R. J., C. C. Tanner, J. P. Sukias, and R. J. Davies-Colley. 2003. Dairy farm wastewater treatment by an advanced pond system. *Water Sci. Technol.* 48:291-297.
- Goh, A. 1986. Production of microalgae using pig waste as a substrate, pp. 235-244. In: W. R. Barclay, R. P. McIntosh (eds.). *Algal Biomass Technologies*. Cramer, Berlin.
- Johnson D. M., S. W. Fultz, and M. R. Bell. 2005. Maryland dairy business summary information series 2004-03 (rev 2005). Dept. Agric. Res. Eng., University of Maryland, College Park, MD.
- Kaiser, J. 2001. The other global pollutant: Nitrogen proves tough to curb. *Science* 294: 1268-1269.
- Kebede-Westhead, E., C. Pizarro, and W. Mulbry. 2003. Production and nutrient removal by periphyton grown under different loading rates of anaerobically digested flushed dairy manure. *J. Phycol.* 39:1275-1282.
- Kebede-Westhead, E., C. Pizarro, and W. Mulbry. 2006. Treatment of swine manure effluent using freshwater algae: Production, nutrient recovery, and elemental composition of algal biomass at four effluent loading rates. *J. Appl. Phycol.* 18:41-46. DOI: 10.1007/s10811-005-9012-8.
- Lincoln, E. P., A. C. Wilkie, and B. T. French. 1996. Cyanobacterial process for renovating dairy wastewater. *Biomass and Bioenergy* 10:63-68.
- Mehlich A. 1984. Mehlich-3 soil test extractant: A modification of Mehlich-2 extractant. *Commun. Soil Sci. Plant Anal.* 15(12):1409-1416.
- Metting, B. 1996. Biodiversity and application of microalgae. *J Ind. Microbiol.* 17: 477-489.
- Metting, B., W. J. Zimmerman, I. Crouch, and J. Van Staden. 1990. Agronomic uses of seaweed and microalgae, pp. 589-627. In: I. Akatsuka (ed.). *Introduction to Applied Phycology*. SPB Academic Publishing, The Hague.
- Mulbry W., E. K. Westhead, C. Pizarro, and L. Sikora. 2005. Recycling of manure nutrients: Use of algal biomass from dairy manure treatment as a slow release fertilizer. *Bioresour. Technol.* 96(4):451-458.
- Olguin, E. J., S. Galicia, O. Angulo-Guerrero, and E. Hernandez. 2001. The effect of low light flux and nitrogen deficiency on the chemical composition of *Spirulina* sp. (*Arthrospira*) grown on digested pig waste. *Bioresour. Technol.* 77:19-24.

- Pizarro, C., W. Mulbry, D. Blerish, and P. Kangas. 2006. An Economic Assessment of Algal Turf Scrubber Technology for Treatment of Dairy Wastewater. *Ecol. Eng.* 26(4):321-327.
- SAS Institute Inc. 2004. *SAS/STAT® 9.1 User's Guide*, Cary, NC: SAS Institute Inc., 5124 pp.
- Sikora, L. J. and N. K. Enkiri. 2003. Availability of poultry litter compost P to fescue compared with triple super phosphate. *Soil Sci.* 168:192-199.
- Thompson, R. B. and J. J. Meisinger. 2002. Management factors affecting ammonia volatilization from land-applied cattle slurry in the Mid-Atlantic USA. *J Environ. Qual.* 31:1329-1338.
- Van Horn, H. H., A. C. Wilkie, W. J. Powers, and R. A. Nordstedt. 1994. Components of dairy manure management systems. *J Dairy Sci.* 77:2008-2030.
- Van Kessel, J. and J. Reeves. 2002. Nitrogen mineralization potential of dairy manures and its relationship to composition. *Biol. Fertil. Soils* 36:118-123.
- Wilkie, A. C. and W. Mulbry. 2002. Recovery of dairy manure nutrients by benthic freshwater algae. *Bioresour. Technol.* 84:81-91.
- Wolf, A. M. and D. B. Beegle. 1995. Recommended soil tests for macronutrients: Phosphorus, potassium, calcium, and magnesium, pp. 25-34. In: J. T. Sims and A. Wolf (eds.) *Recommended Soil Testing Procedures for the Northeastern United States*. Northeast Regional Bull. #493. Agricultural Experiment Station, University of Delaware, Newark, DE.

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