

# DIETARY PHOSPHORUS EFFECTS ON CHARACTERISTICS OF MECHANICALLY SEPARATED DAIRY MANURE

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**ABSTRACT.** One approach to reduce nutrient losses from livestock farms is to apply biological waste treatment systems such as biological nitrogen (N) removal or enhanced biological phosphorus (P) removal (EBPR) to reduce the nutrient content of land-applied waste. The EBPR process takes advantage of the ability of P-accumulating organisms (PAOs) to sequester excess P as polyphosphate granules in their cytoplasm, yielding a P-depleted liquid effluent and a P-enriched biomass. Biological N removal systems result in the conversion of organic or ammonia-N to innocuous N<sub>2</sub> gas. Understanding the variation in parameters such as chemical oxygen demand (COD), total and volatile suspended solids (TSS and VSS), and ammonia-N (NH<sub>3</sub>-N) is necessary to design these systems. Our objectives were to evaluate the effects of diet and manure separation on parameters important to reactor design. Waste was collected from nine cows fed a high P diet (0.47% P), a low P diet (0.32% P), or low P with exogenous phytase plus cellulase (0.32% P), in a replicated Latin square design (three 3 × 3 squares). Total collection of milk, urine, and feces was conducted on days 19 to 21 of each period, a mixed slurry (urine, feces, and water) was created, and slurry was separated mechanically to generate liquid effluent. Slurry contained more COD, solids, N, and P than liquid effluent, but the COD:P ratio was similar in the two wastes. The ratio of COD:N was higher in slurry than in separator effluent, but the ratio in both wastes was sufficient to support biological N removal. The P content of slurry, liquid effluent, and manure solids from cows fed low P was lower than from cows fed high P, and the COD content of effluent was higher with the low P diet. The COD:P ratio of all wastes was sufficient to support EBPR and biological N removal, but variation was observed with diet. Waste from cows fed low P had a higher COD:P ratio than that of cows fed high P, and waste from cows fed the enzyme-supplemented diet had a lower COD:N ratio than that of cows fed the control diet. Dairy manure slurry and effluent will support EBPR and biological N removal. Dietary effects on parameters important to the design of advanced waste treatment systems were observed, but were not of a magnitude that would affect reactor design.

**Keywords.** Lactating cows, Manure treatment, Research-scale manure separation.

The development of strategies to reduce the nitrogen (N) and phosphorus (P) content of land-applied livestock manure is an important aspect of long-term efforts to reduce nutrient pollution of water resources. Reducing the nutrient content of land-applied waste reduces potential nutrient losses, allows livestock producers to increase the rate of manure application to a fixed land base, and/or reduces the amount of land needed for spreading manure. Approaches to reducing the nutrient content of waste include dietary nutrient management (refining diets to maximize efficiency of utilization of consumed nutrients) and physical or biological nutrient removal systems.

Physical separation of manure solids and liquids via gravity or screening reduces organic loading to the liquid treatment system, removes large particles that could plug or damage nozzles in the irrigation system used in land

application, and results in a fibrous byproduct that can be moved off-site more easily, with or without further processing, if export of nutrients is required. Phosphorus removal from the liquid fraction of dairy slurry via physical separation ranges between 2% and 59% (Holmberg et al., 1983), depending on separator type and influent characteristics (Powers et al., 1995; Zhang and Westerman, 1997).

Even after removal of manure solids through solid-liquid separation, a significant fraction of N and P remains in the wastewater (Van Horn et al., 1994). Biological treatment systems have been used to reduce the N and P content of dilute swine waste (Vanotti et al., 2000; Tilche et al., 2001; Obaja et al., 2003) and waste from dairy processing plants (Bickers et al., 2003). Biological treatment allows significant removal of N, P, solids, and COD from liquid wastes. Examples of biological waste treatment techniques include enhanced biological P removal (EBPR), denitrification, and biological nutrient removal (EBPR coupled with N removal). The EBPR process takes advantage of the ability of P-accumulating organisms (PAOs) to sequester excess P as polyphosphate granules in their cytoplasm (Grady et al., 1999). These systems include sequential aerobic and anaerobic conditions; at the end of the treatment process, the liquid effluent is low in P, while the biomass wasted from the system is enriched in P.

Conventional biological removal of N may be achieved separately or in the same reactor system that achieves enhanced biological P removal. Nitrogen exists predomi-

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nantly in the reduced form (organic N or inorganic NH<sub>3</sub>-N) in dairy waste. To biologically remove N by conventional means, it must first be converted aerobically to nitrate by nitrifying bacteria. Then, under anaerobic denitrifying (anoxic) conditions, the nitrate formed by nitrification is used as an alternative electron acceptor, which reduces the nitrate to innocuous N<sub>2</sub> gas.

To fuel these reactions and achieve effective biological removal of N and/or P, appropriate ratios of biodegradable chemical oxygen demand (COD) to N or COD:P are needed (Grady et al., 1999). Designing these systems for dairy manure requires, therefore, additional waste characterization to determine parameters such as COD, degradable COD, total and volatile suspended solids (TSS and VSS), and ammonia-N (NH<sub>3</sub>-N).

The link between diet and nutrient excretion by lactating dairy cows is well documented (Morse et al., 1992; Wu et al., 2000; Knowlton and Herbein, 2002), but the magnitude of the change in composition of separated manure liquids and solids with diet is unknown. In addition, methods to improve digestibility of the diet (i.e., supplementation of the diet with exogenous enzymes) may affect composition of separated dairy manure in a way that influences design of waste treatment systems. The objective of this study was to evaluate the effect of diet and manure separation on compositional parameters important to the design of biological N and P removal reactors for dairy manure.

## MATERIALS AND METHODS

### COWS AND DIETS

Nine early lactation cows were fed diets containing 0.32% or 0.47% P. The low P diets were fed with or without the addition of fibrolytic and phytase enzyme formulations. The fibrolytic enzyme formulation was a commercial preparation from fungal extracts, with 15,000 units of cellulase activity per g (Loveland Industries, Greeley, Colo.), with one unit defined as the cellulase activity that produced a relative fluidity change of 1.0 in 100 min in a 0.2% (w/v) sodium carboxymethyl cellulose (CMC type 7HP, Hercules, Inc., Wilmington, Del.) solution under assay conditions (pH 4.5 and 40 °C) as measured with a calibrated, size 100 Cannon-Fiske viscometer. The phytase formulation contained 5,000 units of phytase activity per g, with one unit defined as the phytase activity that released 1.0 μmol of phosphate per min under assay conditions (pH 5.5 and 37 °C).

The granular enzyme formulations were mixed with a corn grain carrier, and the enzyme-corn grain mixture or control (an equal quantity of corn grain containing no enzyme formulation) was added to the grain portion of the diet prior to mixing of the TMR (200 g of fibrolytic enzyme formulation and 280 g of phytase per tonne DM fed). Dosages of enzymes used are standard for this commercial preparation, originally determined to supply excess enzyme based on typical substrate content of mixed dairy rations. Diets were formulated to meet nutrient requirements (National Research Council, 2001). Ingredient and nutrient composition of diets are listed in tables 1 and 2. This experiment was conducted with approval from the Virginia Tech Animal Care and Use Committee.

**Table 1. Ingredient composition of dairy cow diets (all values in % of diet DM).**

Ingredient	High P	Low P	Low P with Enzyme <sup>[a]</sup>
Corn silage	44.9	45.0	45.0
Corn grain	26.5	26.6	26.6
Soybean meal	14.6	14.7	14.7
Expeller soybean meal <sup>[b]</sup>	1.36	1.36	1.36
Vitamin-mineral mix <sup>[c]</sup>	9.19	9.22	9.21
Calcium carbonate	1.40	1.89	1.88
Dicalcium phosphate	0.81	0.00	0.00
Urea	0.52	0.52	0.52
Sodium bicarbonate	0.52	0.52	0.52
Salt	0.19	0.19	0.19
Enzyme formulation <sup>[a]</sup>	0.00	0.00	0.04

<sup>[a]</sup> Enzyme = cellulase (15,000 units/g) and phytase (5,000 units/g).

<sup>[b]</sup> Soyplus, West Central Soy, Ralston, Iowa.

<sup>[c]</sup> Each kg contained 1,462 mg Ca, 1,758 mg P, 11,100 mg Mg, 1,270 mg S, 3.45 mg Co, 136 mg Cu, 6.8 mg I, 170 mg Mn, 2.64 mg Se, 340 mg Zn, 46,900 IU Vit A, 16,960 IU Vit D, and 256 IU Vit E.

**Table 2. Nutrient composition of dairy cow diets.**

Nutrient	Values in % of diet DM				P <	
	High P	Low P	Low P plus Enzyme <sup>[a]</sup>	SEm	Low P vs. High P	Enzyme <sup>[a]</sup> vs. Low P Control
CP	17.4	17.1	17.4	0.35	0.69	0.53
ADF	14.1	14.2	14.1	0.06	0.41	0.49
NDF	26.1	26.1	26.0	0.16	0.73	0.62
Ash	8.73	9.02	8.91	0.03	0.55	0.81
Ca	1.11	0.99	0.92	0.07	0.13	0.48
P	0.47	0.32	0.32	0.01	0.01	0.87

<sup>[a]</sup> Enzyme = cellulase (15,000 units/g) and phytase (5,000 units/g).

### EXPERIMENTAL DESIGN AND SAMPLING

Cows were grouped by previous lactation milk yield and assigned to one of three 3 × 3 Latin squares. Squares were balanced for carryover effects. Each experimental period lasted 21 d. Cows were fed in Calan doors for the first 17 d of each period and were moved to individual stalls on d 18 for total collection of feces, urine, and milk. Cows were fed once daily at 0800 h and milked at 0700 h and 1900 h. Feed was offered 5% to 10% in excess of previous day's intake (wet basis).

On day 18, a sterile Foley urine catheter (22 French, 75 cc, C.R. Bard, Inc., Covington, Ga.) was inserted into the urethra for total collection of urine. All excreted urine, feces, and milk were collected on days 18, 19, 20, and 21. Urine was weighed at 4 h intervals and pooled by cow across a 24 h period and used to create a dilute slurry (details below). All excreted feces were collected at 4 h intervals and stored in a sealed container, then weighed, thoroughly mixed, and subsampled daily. Feed ingredients (forages and concentrates) were sampled once each week, and feed refusals were weighed and sampled daily. On days 18, 19, 20, and 21, feed offered and refused were measured, total milk weights were recorded, and milk was sampled at eight consecutive milkings.

All excreted urine and feces were mixed by cow in the mass proportions excreted. Water was added to create a dilute slurry (50% w/w) to represent a typical dilution due to flushed alley removal of waste from barns. Water, feces, and urine were mixed for 3 min with a paint mixing paddle attached to a drill. Slurry from each cow was stored at ambient

temperature for 24 h to mimic accumulation prior to separation, as is common on farms, and then mixed, weighed, subsampled, and pumped (approx. 30 L/min) through a research-scale mechanical solids separator. The separator was a roller press, consisting of two perforated basins (concave screens with 3.18 mm screen opening) with a rotating brush assembly to convey slurry across the screens. Liquids flowed through the screen to a collection basin below the screens. Solids were advanced and forced out of the discharge by the rotating brushes. The mechanical separator used in this study was a research version (1:4 scale) of a commercial available unit (Integrity Nutrient Control System, Nutrient Control Systems, Inc., Chambersburg, Pa.).

Liquid effluent, slurry, and manure solids were weighed and subsampled. A section of pipe fitted with a stopper controlled by a length of wire was used to collect column samples of slurry and liquid separator effluent from the collection containers during mixing. Four subsamples were collected from each collection container, and then pooled and acidified with H<sub>2</sub>SO<sub>4</sub> sufficient to reduce sample pH to 2 to prevent N volatilization. A second pooled sample from each collection container was collected and stored at 4°C to preserve for subsequent analysis (for assays with which acid would interfere). Manure solids were mixed, subsampled, and stored at 4°C until analyzed.

#### LABORATORY ANALYSIS

Samples of feed refusals and feed ingredients were dried to constant weight at 60°C in a forced-air drying oven (Wisconsin Oven, Memmert, Schwabach, Germany). Dried samples were ground through a 1 mm screen in a Wiley Mill (Arthur H. Thomas, Philadelphia, Pa.). Feed and feed refusal samples were analyzed in duplicate for N, P, (AOAC, 1984), and neutral detergent fiber (NDF) and acid detergent fiber (ADF) sequentially with  $\alpha$ -amylase (Van Soest et al., 1991).

Slurry and separated waste samples (separated liquid effluent, separated manure solids) were analyzed for COD and soluble COD, total Kjeldahl nitrogen (TKN), urea plus NH<sub>3</sub>-N (phenate method), and total P (APHA, 1998; AOAC, 1984). Liquid waste samples (slurry, separated liquid effluent) were analyzed for TSS and VSS (APHA, 1998). Manure solids samples were analyzed for total solids (TS) and volatile solids (VS; APHA, 1998). Removal of constituents by mechanical separation was calculated as the concentration of each in solids times the quantity of solids generated, divided by the quantity in slurry.

#### STATISTICAL ANALYSIS

The effect of dietary treatment and manure separation on concentration of components in mixed slurry and effluent was analyzed using the MIXED procedure of SAS (SAS, 1999) with the model:

$$Y_{ijklm} = \mu + S_i + C_j(S)_i + P_k + T_l + D(P_k)_m + M_n + T_l * M_n + e_{ijklm} \quad (1)$$

where

- $\mu$  = overall mean
- $S_i$  = random effect of square ( $i = 1$  to 3)
- $C_j(S)_i$  = random effect of cow within square ( $j = 1$  to 3)
- $P_k$  = fixed effect of period ( $k = 1$  to 3)
- $T_l$  = fixed effect of dietary treatment ( $l = 1$  to 3)

$D(P_k)_m$  = fixed effect of day of sampling within period ( $m = 1$  to 3)

$M_n$  = fixed effect of manure separation ( $n = 1$  to 2)

$T_l * M_n$  = interaction of dietary treatment with manure separation

$e_{ijklmn}$  = residual error, assumed to be normally distributed.

Cow  $C_j(S)_i$  was the error term used to test the effects of diet, manure separation, and their interaction. Residual error was used to test the effects of day of collection; this effect was not significant for any measured parameter, so it is not discussed. Pre-planned contrasts were used to evaluate the effect of dietary P (high P vs. low P and low P-enzyme), enzyme addition (low P vs. low P-enzyme), and manure separation (slurry vs. effluent).

Differences were declared significant at  $P < 0.05$  and trends at  $P < 0.10$ . Results are reported as least squares means.

## RESULTS AND DISCUSSION

Ingredient and nutrient composition of treatment diets are presented in tables 1 and 2. As planned, diets contained approximately 17% protein and 26% NDF, and differed only in P content. Feed intake and milk yield, manure excretion, and P digestion and partitioning were the focus of a concurrent aspect of this experiment, and these data are reported elsewhere (Knowlton et al., in press, 2005). Briefly, neither dietary P content nor exogenous phytase plus cellulase affected DMI (21.8 kg/d), milk yield (39.6 kg/d), or milk composition. Apparent P digestibility tended to increase with the addition of exogenous phytase and cellulase to the diet, but P excretion and retention were not significantly affected.

#### COMPOSITION OF SEPARATED MANURE

Chemical oxygen demand is a measure of the organic matter in a wastewater sample, measured as the oxygen required to oxidize the sample chemically. This measure, as well as the solubility of the COD, TSS, and VSS content of the waste, is important in the design of biological treatment reactors. Dietary effects on these measures are of interest primarily to improve understanding of the overall variation that might be observed on commercial farms interested in the design of waste treatment reactors.

The content of all constituents was higher in slurry than in liquid separator effluent (table 3), but there was no effect of the interaction of diet and manure separation on any parameter. As expected, the P content was lower and the ratio of COD to P higher in all wastes (mixed slurry, liquid effluent, and separated manure solids) from cows fed low P diets (tables 4 and 5).

Removal of wet solids by the mechanical manure separator averaged 34% to 40% of the total wet mass input, and this removal efficiency was not affected by diet (table 6). Capture of slurry P, COD, soluble COD, TS, VS, and TKN in solids was not affected by diet, but removal of urea plus NH<sub>3</sub>-N was higher in cows fed low P than in those fed high P diets. The lack of effect of dietary P content on P removal by manure separation is despite the reported increase in the proportion of water-soluble P in feces with high P diets (Dou et al., 2002; Ebeling et al., 2002). This observation is explained by the fact that a significant proportion of the water

**Table 3. Characteristics of separated manure from lactating cows.**

	COD (mg O <sub>2</sub> /L)	Soluble COD (mg O <sub>2</sub> /L)	TSS (mg/L)	VSS (mg/L)	TKN (mg/L)	COD:TKN	Urea + NH <sub>3</sub> -N (mg/L)	Total P (mg/L)	COD:P
Slurry <sup>[a]</sup>	58,040	22,263	60,170	49,734	4,478	14.0	1503	329.4	247.9
Liquid separator effluent	37,588	15,378	32,080	28,334	3,076	12.4	842	230.1	253.3
SEm	1,437	653	1,885	1,593	152	0.83	198	16.9	26.9
Slurry <sup>[a]</sup> vs. effluent, P <	0.01	0.01	0.01	0.01	0.01	0.03	0.02	0.01	0.88
Concentration in municipal wastewater <sup>[b]</sup>	910	.	360	215	75	12.1	35	15	60.7

<sup>[a]</sup> Mixed slurry = feces plus urine mixed in proportions excreted, diluted 50% w/w with water, and stored for 24 h at room temperature.

Composition of slurry and effluent averaged across treatments.

<sup>[b]</sup> Adapted from table 3.2 in Reynolds and Richards (1996).

**Table 4. Effect of diet on mixed dairy manure slurry and liquid effluent following mechanical solids separation.**

	COD (mg O <sub>2</sub> /L)	Soluble COD (mg O <sub>2</sub> /L)	TSS (mg/L)	VSS (mg/L)	TKN (mg/L)	COD:TKN	Urea + NH <sub>3</sub> -N (mg/L)	Total P (mg/L)	COD:P
Mixed slurry <sup>[a]</sup>									
High P	55,924	21,624	59,080	50,003	3,968	14.1	2,150	436.3	162.3
Low P	62,671	23,531	60,650	48,215	4,595	15.3	1,114	280.9	279.3
Low P with enzyme <sup>[b]</sup>	55,798	21,773	61,894	52,297	4,764	12.9	1,198	270.7	298.3
SEm	3,187	1,421	4,069	3,254	335	1.05	659	34.6	46.7
P <									
Low P vs. high P	0.40	0.56	0.66	0.95	0.07	0.95	0.10	0.01	0.02
Enzyme <sup>[b]</sup> vs. low P control	0.13	0.37	0.83	0.38	0.68	0.04	0.90	0.80	0.75
Diet × separation	0.71	0.74	0.80	0.43	0.90	0.73	0.30	0.91	0.83
Liquid effluent									
High P	34,681	14,040	29,762	26,348	3,069	11.8	849.8	317.9	134.8
Low P	39,546	16,072	31,881	27,761	3,106	13.0	829.2	196.7	307.3
Low P with enzyme <sup>[b]</sup>	38,153	15,808	32,815	28,960	3,135	12.3	858.2	179.1	318
SEm	1,571	853	2,217	1,918	120	0.82	121.9	26.1	49.1
P <									
Low P vs. high P	0.04	0.07	0.31	0.35	0.72	0.40	0.97	0.01	0.01
Enzyme <sup>[b]</sup> vs. low P control	0.51	0.82	0.75	0.63	0.86	0.55	0.83	0.58	0.87

<sup>[a]</sup> Mixed slurry = feces plus urine mixed in proportions excreted, diluted 50% w/w with water, and stored for 24 h at room temperature.

<sup>[b]</sup> Enzyme = cellulase (15,000 units/g) and phytase (5,000 units/g).

**Table 5. Effect of diet on manure solids composition following mechanical manure separation.<sup>[a]</sup>**

	COD (mg O <sub>2</sub> /kg)	Soluble COD (mg O <sub>2</sub> /kg)	TS (mg/kg)	VS (mg/kg)	TN (mg/kg)	Urea + NH <sub>3</sub> -N (mg/kg)	Total P (mg/kg)	COD:P
High P	59,867	26,056	130,214	121,445	2,088	611	516.8	127.2
Low P	67,659	30,817	139,987	132,035	2,179	599	338.3	204.3
Low P with enzyme <sup>[b]</sup>	65,215	30,322	139,479	130,326	2,167	657	361.3	193.2
SEm	3,118	2,174	3,317	3,471	80.9	109	25.3	10.9
P <								
Low P vs. high P	0.07	0.10	0.03	0.03	0.40	0.88	0.01	0.01
Enzyme <sup>[b]</sup> vs. low P control	0.54	0.87	0.92	0.72	0.92	0.65	0.49	0.47

<sup>[a]</sup> All data expressed on wet basis.

<sup>[b]</sup> Enzyme = cellulase (15,000 units/g) and phytase (5,000 units/g).

**Table 6. Effect of diet on nutrient removal in the manure solids fraction by mechanical manure separation.**

	Amount Removed by Separation (kg/kg slurry)							
	Wet Solids	COD	Soluble COD	TS	VS	TKN	Urea + NH <sub>3</sub> -N	Total P
High P	0.34	0.44	0.49	0.35	0.39	0.20	0.13	0.50
Low P	0.36	0.43	0.56	0.37	0.43	0.21	0.21	0.58
Low P with enzyme	0.40	0.62	0.69	0.48	0.52	0.21	0.22	0.69
SEm	0.02	0.10	0.09	0.06	0.06	0.02	0.03	0.10
P <								
Low P vs. high P	0.17	0.52	0.21	0.25	0.29	0.43	0.04	0.29
Enzyme <sup>[a]</sup> vs. low P control	0.26	0.18	0.33	0.17	0.19	0.94	0.89	0.45

<sup>[a]</sup> Enzyme = cellulase (15,000 units/g) and phytase (5,000 units/g).

in slurry was retained in the separated solids. Therefore, water-soluble P was not only associated with the liquid effluent fraction; some of the “soluble” P remained in the solids.

Capture of P, COD, and soluble COD in manure solids was more efficient than total solids removal, indicating that manure separation effectively concentrated these constituents in manure solids. In contrast, removal of TKN and urea plus NH<sub>3</sub>-N was less efficient than overall solids capture. Urea and NH<sub>3</sub>-N are primarily in solution, and are not susceptible to concentration in solids.

Removal of significant solids improves performance of anaerobic treatment lagoons, reduces the potential for odor generation, and yields two fractions that are more easily managed than slurry (Zhang and Westerman, 1997). The performance of solid-liquid separators varies in terms of partitioning of nutrients between the liquid and solid fractions, as a function of both separator type and influent characteristics (including total solids concentration and flow rate; Powers et al., 1995; Zhang and Westerman, 1997; Converse et al., 2000). Zhang and Westerman (1997) calculated theoretical maximum total solids separation efficiency by physical separation of 64% to 84% for beef and dairy manure. They concluded that smaller screen opening size and increased flow rate increased total solids separation. Holmberg et al. (1983) reported that P capture from swine manure in the solid fraction varied between 2.5% and 59% as screen size and flow rate varied. Chastain et al. (2001) reported removal of 53.1% of total P from flushed dairy manure by an inclined stationary screen separator with a screen size of 1.5 mm. In the current study, 50% to 69% of the P in slurry was captured in solids.

#### IMPLICATIONS FOR FURTHER TREATMENT OF WASTE

Even after solid-liquid separation, a significant fraction of N, P, and manure solids remains in the separated liquid effluent. Increasingly stringent regulations restricting odor emission and nutrient application to crops provides incentive to treat this waste further prior to land application (Water Quality Improvement Act, 1998; Virginia Poultry Waste Management Program, 1999; EPA, 2003). The growing adoption of waste handling and treatment systems beyond traditional slurry systems makes understanding of the variability of water quality parameters such as COD, TS, and VS in dairy waste important. The concentration of these constituents in manure influences the design of anaerobic and aerobic treatment systems, as well as sludge accumulation within these systems.

Both EBPR and denitrification rely on organic matter (typically measured as COD content) to fuel the nutrient removal process. The content of COD, its relative biodegradability, and its content relative to N and P are all critical factors in reactor design (Grady et al., 1999; Tasli et al., 1999). The COD and soluble COD content observed in the liquid fraction of separated dairy manure in the current study (tables 3 and 4) are similar to the values observed in raw piggery wastewater (Tilche et al., 2001), but are more than tenfold higher than is typical in municipal waste treated with biological nutrient removal systems. All constituents in separated liquid effluent were much higher than in untreated municipal wastewater. Concentrations of COD, TSS, VSS, TKN, NH<sub>3</sub>-N, and total P in municipal wastewater are reported in table 3. Separated liquid effluent is from 15 to

130 times more concentrated than municipal waste for these parameters.

The ratio of COD to TKN, important in designing biological N removal, was lower in liquid separator effluent than in mixed slurry (12.4 vs. 14.0; table 3). While statistically significant, this difference is of little importance. Approximately 4.3 g of COD are required per g of N removed by aerobic autotrophic NH<sub>3</sub> oxidation and anoxic heterotrophic denitrification (Grady et al., 1999). Successful biological N removal was achieved in swine wastewater with a COD:TKN ratio of 1.8 (Vanotti et al., 2000). Assuming that the COD in dairy waste is readily dissimilable by denitrifying bacteria, good to excellent biological N removal should be achieved by sequentially exposing liquid dairy manure to anoxic and aerobic conditions.

The COD to total P ratio observed in slurry and effluent (tables 3 and 4) is quite high relative to typical municipal waste streams, and was unaffected by mechanical separation, indicating similar efficiencies of capture of these parameters in manure solids. Enhanced biological P removal systems work best when P is limiting (Grady et al., 1999), as would be the case for the wastes in the current study. However, too much excess COD relative to P can encourage growth of non-phosphorus accumulating organisms in the biological treatment system, and reduce EBPR efficiency. Piggery waste of similar COD content was successfully treated with EBPR, but dilution was required (Tilche et al., 2001; Obaja et al., 2003).

## CONCLUSIONS

Effects of dietary P content, exogenous dietary cellulase and phytase, and mechanical separation on chemical composition of dairy manure were observed, but these are of little practical significance in the design of advanced waste treatment systems. The observed COD:P and COD:N ratios of dairy manure slurry and separated liquid effluent were well above the minimum required for biological N and P removal.

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## REFERENCES

- AOAC. 1984. *Official Methods of Analysis*. 14th ed. Arlington, Va.: Association of Official Analytical Chemists.
- APHA. 1998. *Standard Methods for the Examination of Water and Wastewater*. 20th ed. Washington, D.C.: American Public Health Association.
- Bickers, P. O., R. Bhamidimarri, J. Shepard, and J. Russell. 2003. Biological phosphorus removal from a phosphorus-rich dairy processing wastewater. *Water Sci. Tech.* 48(8): 43-51.

- Chastain, J. P., M. B. Vanotti, and M. M. Wingfield. 2001. Effectiveness of liquid-solid separation for treatment of flushed dairy manure: A case study. *Applied Eng. in Agric.* 17(3): 343-354.
- Converse, J. C., R. G. Koegel, and R. J. Straub. 2000. Nutrient separation of dairy manure. In *Proc. 8th Int. Symposium: Animal, Agricultural, and Food Processing Wastes*, 118-131. St. Joseph, Mich.: ASAE.
- Dou, Z., K. F. Knowlton, R. A. Kohn, Z. Wu, L. D. Satter, G. Zhang, J. D. Toth, and J. D. Ferguson. 2002. Phosphorus characteristics of dairy feces affected by diets. *J. Environ. Qual.* 31: 2058-2065.
- Ebeling, A. M., L. G. Bundy, J. M. Powell, and T. W. Andraski. 2002. Dairy diet phosphorus effects on phosphorus losses in runoff from land-applied manure. *SSSA J.* 66(1): 284-291.
- EPA. 2003. National pollutant discharge elimination system permit regulation and effluent limitation guidelines and standards for concentrated animal feeding operations: Final rule. 40 CFR Parts 9, 122, 123, and 412. Washington, D.C.: U.S. Environmental Protection Agency.
- Grady, C. P., G. T. Daigger, and H. C. Lim. 1999. *Biological Wastewater Treatment*. New York, N.Y.: Marcel Dekker.
- Holmberg, R. D., D. T. Hill, T. J. Prince, and N. J. V. Dyke. 1983. Potential of solid-liquid separation of swine wastes for methane production. *Trans. ASAE* 26(6): 1803-1807.
- Knowlton, K. F., and J. H. Herbein. 2002. Phosphorus partitioning during early lactation in dairy cows fed diets varying in phosphorus content. *J. Dairy Sci.* 85(5): 1227-1236.
- Knowlton, K. F., C. M. Parsons, C. W. Cobb, and K. F. Wilson. 2005. Exogenous phytase plus cellulase and phosphorus excretion in lactating cows. *The Professional Animal Scientist* (in press).
- Morse, D., H. Head, C. J. Wilcox, H. H. V. Horn, C. D. Hissem, and B. Harris, Jr. 1992. Effects of concentration of dietary phosphorous on amount and route of excretion. *J. Dairy Sci.* 75(11): 3039-3049.
- National Research Council. 2001. *Nutrient Requirements of Dairy Cattle*. 7th rev. ed. Washington, D.C.: National Academy of Sciences.
- Obaja, D., S. Mace, J. Costa, C. Sans, and J. Mata-Alvarez. 2003. Nitrification, denitrification, and biological phosphorus removal in piggery wastewater using a sequencing batch reactor. *Bioresource Tech.* 87(1): 103-111.
- Powers, W. J., R. E. Montoya, H. H. VanHorn, R. A. Nordstedt, and R. A. Bucklin. 1995. Separation of manure solids from simulated flushed manures by screening or sedimentation. *Applied Eng. in Agric.* 11(3): 431-436.
- Reynolds, T. D., and P. A. Richards, eds. 1996. *Unit Operations and Processes in Environmental Engineering*. 2nd ed. Washington, D.C.: PWS Publishing.
- SAS. 1999. SAS User's Guide, version 8.0. Cary, N.C.: SAS Institute, Inc.
- Tasli, R., D. Orhon, and N. Artan. 1999. The effect of substrate composition on the nutrient removal potential of sequencing batch reactors. *Water SA* 25(3): 337-344. Available at: www.wrc.org.za.
- Tilche, A., G. Bortone, F. Malaspina, S. Piccinini, and L. Stante. 2001. Biological nutrient removal in a full-scale SBR treating piggery wastewater: Results and modelling. *Water Science and Tech.* 43(3): 363-371.
- 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(7): 2008-2030.
- Vanotti, M. B., J. M. Rice, S. L. Howell, P. G. Hunt, and F. J. Humenik. 2000. Advanced treatment system for liquid swine manure using solid-liquid separation and nutrient removal unit processes. In *Proc. 8th Int. Symposium: Animal, Agricultural, and Food Processing Wastes*, 393-400. St. Joseph, Mich.: ASAE.
- Van Soest, P. J., J. B. Robertson, and B. A. Lewis. 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *J. Dairy Sci.* 74(10): 3583-3597.
- Virginia Poultry Waste Management Program. 1999. In *Virginia Code*. Vol. 62.1-44.17:1.1.
- Water Quality Improvement Act of 1998. 1998. In *Maryland Agriculture Code*. Vol. 8-801.1.
- Wu, Z., L. D. Satter, and R. Sojo. 2000. Milk production, reproductive performance, and fecal excretion of phosphorus by dairy cows fed three amounts of phosphorus. *J. Dairy Sci.* 83(5): 1028-1041.
- Zhang, R. H., and P. W. Westerman. 1997. Solid-liquid separation of animal manure for odor control and nutrient management. *Applied Eng. in Agric.* 13(5): 657-664.

## NOMENCLATURE

- COD = chemical oxygen demand  
P = phosphorus  
N = nitrogen  
TSS = total suspended solids  
VSS = volatile suspended solids  
TS = total solids  
VS = volatile solids  
TKN = total Kjeldahl N  
EBPR = enhanced biological P removal