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Anaerobic Digestion of Dairy Manure Combined with Duckweed (*Lemnaceae*)

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Abstract. *Anaerobic digestion (AD) has become an increasingly popular method of treating wastewater or sludges from animal feeding operations. Enhancement of biogas production in anaerobic digesters, through addition of commonly-available and under-utilized biomass, could benefit sustainability of farm-scale anaerobic digesters. Duckweed is a common aquatic plant that aggressively grows in farm ponds, lagoons, and other water bodies that receive agricultural runoff. As such, duckweed is a readily-available biomass that could be easily added to farm-scale anaerobic digesters. Therefore, research aimed to determine if biogas (methane) production could be improved by supplementing digesters with duckweed (e.g., Landoltia punctata). Increases in biogas production and rate of attaining peak biogas production were assessed in batch continuously-stirred reactors at 35^{o}C. Varying concentrations of duckweed were added to dairy manure slurries and gas production was observed for 20-40 days. Additionally, subsequent research will assess changes in chemical oxygen demand (COD), pH, and fatty acids within manure/duckweed slurries in parallel with analyses of biogas production, with time. Preliminary results indicate that addition of duckweed, in the range of 0.5 to 2% (dry mass), enhanced methane and total gas production in dairy manure slurries; however, subsequent increases in methane and total gas production at >2% duckweed were not observed. In conclusion, addition of duckweed biomass, produced during treatment of agricultural wastewaters and runoff, to anaerobic digesters is a promising approach to enhancement of biogas production.*

Keywords. Duckweed, anaerobic digestion, bioenergy, wastewater, dairy

Introduction

Livestock and dairy production produces large quantities of waste that has the potential to spread disease, contaminate groundwater and surface waters, and cause many other environmental consequences. Regulations for managing and treating this waste are becoming stricter in order to decrease the environmental impact of animal production. One method of treating this waste is anaerobic digestion, in which anaerobic bacteria degrade organic material in manure to more stable products, including methane, carbon dioxide, and biosolids that are suitable for fertilization (Sakar, Yetilmezsoy, Kocak, 2009). However, anaerobic digesters are not always economically feasible for small to medium farm operations. Because biogas produced from anaerobic digestion can be used for fuel, increasing the production of methane may increase the return on the initial investment by creating more of a profitable product.

Anaerobic digestion is initiated first by acid-producing bacteria that hydrolyze and ferment complex organic matter (e.g., manure) into volatile fatty acids, which is then utilized by methanogenic bacteria and converted into methane (Marcias-Corral et al., 2009). Anaerobic digestion can considerably reduce the chemical oxygen demand (COD) of the manure, which makes it safer to release into the environment as a soil amendment (Marcias-Corral et al., 2009). Anaerobic digestion systems are generally either a suspended growth process, in which the bacteria is contained in flocs or granules as a sludge, or an attached-growth process, in which the bacteria hold fast to a media that can either be moving or fixed (Sakar, Yetilmezsoy, Kocak, 2009). Microbes in anaerobic digestion systems, especially the methanogens, are sensitive to changes in their environment, such as changes in pH, temperature or increased amounts of ammonia or metals. Fluctuations in any parameter can cause microbes to die, which in return decreases gas production. The optimal range of pH for anaerobic bacteria is 6.5 to 7.5, and the optimal temperature is between 30° C and 40° C (Sakar, Yetilmezsoy, Kocak, 2009).

Natural and constructed wetlands have been used to treat wastewaters since the 1950s. Wetlands are good for wastewater treatment because they are largely aquatic ecosystems with both oxic and anoxic areas where organic matter can be reduced and nutrients can be removed by the wetland vegetation and microorganisms (Verhoeven and Meuleman, 1999). Constructed wetlands are can be designed to handle different loading rates and contain different plants intended to maximize the removal of pollutants such as COD, BOD, and nutrients (Verhoeven and Meuleman, 1999).

Studies have indicated potential for combining the two treatment processes and using anaerobic digestion as a pretreatment for constructed wetlands. For instance, the study by Alvarez, Ruiz, and Soto (2008) found that pretreating animal wastes with anaerobic digestion reduced the amount of organic matter in the influent for a constructed wetland - thereby decreasing the amount of area needed, and therefore construction cost of the constructed wetland. Both technologies are low in construction and operation costs, excess sludge, and energy demand and address the limitations of the other, making the two technologies complimentary to each other (Alvarez, Ruiz, Soto, 2008).

Duckweed (*Lemnaceae*) is a small, floating aquatic plant that is common in temperate wetlands.Duckweed does not have differentiated leaves, but instead have reduced organs referred to as fronds; duckweed is considered the “smallest flowering plant” (Cross, 2002). Duckweed grows rapidly in a range of aquatic environments with a doubling time of approximately 1 week (Landolt 1986). Often, it is used for constructed treatment wetlands and farm ponds because it requires no pretreatment and is adept at removing minerals and nutrients from water (Clark and Hillman, 1996). Duckweed was comparable to water hyacinth in nutrient removal from dairy wastewater – although the water hyacinth had a higher growth rate, and therefore a higher rate of nutrient removal, the duckweed had a higher total amount of phosphorous and nitrogen in its tissues (Debusk et al., 1995). The duckweed also had less seasonal variance than the water hyacinth. The duckweed had a phosphorous uptake of about 20 mg P/m^2-d in both February and July, and a nitrogen uptake of 88 mg N/m^2-d in February and 79 mg N/m^2-d in July, whereas in July the water hyacinth had between 3 and 4 times the amount of phosphorous and nitrogen uptake observed in February (DeBusk, Peterson, Reddy, 1995).

Introducing duckweed to the anaerobic digestion system for waste products may increase methane production for a relatively low price. The addition of iron-enriched duckweed to the anaerobic digestion of poultry manure increased the rate of gas production, though not the net gas production (Clark and Hillman, 1996). The authors concluded that the iron in the duckweed enhanced methane production; however, evidence to any correlation was not provided and the duckweed tissue concentrations of iron was 100X higher than would be expected for naturally-sourced duckweed.

The purpose of the study herein was to quantify the benefits of addition of duckweed from natural sources on methane production from dairy manures.

MATERIALS AND METHODS

MATERIALS COLLECTION

The manure used in this experiment came from the Michigan State University Dairy Farm. The duckweed was collected from local wetlands and the Red Cedar River. In order to dry the duckweed (which was done to avoid difficulties adding the duckweed to the lab-scale reactors), the duckweed was patted dry and placed on paper towel under lights in a well-ventilated room for several days. The duckweed consisted of several species, including *Spirodella sp.*, *Wolffia sp.*, *Landolitia sp.*, and *Lemna sp.*, mostly *Lemna minor*.

Two experiments were carried out to assess the viability of duckweed as a useful supplement to anaerobic digestion, one simulating a summer scenario for an anaerobic digester and one simulating a winter scenario. The summer experiment was carried out in the summer, and the duckweed for that experiment was collected in July and was healthy and green. It was dried and used in the reactors right away. The manure was also collected fresh from the farm and used right away. In the winter experiment, the duckweed was collected in the fall when the growing season was nearly over, causing the duckweed to be less green and healthy. It was then dried and stored for several months before being used in the reactors. The manure used in the winter experiment had also been stored for some time before use.

ANAEROBIC DIGESTION

In order to simulate anaerobic digesters used to treat manure, laboratory-scale batch reactors were used. Glass serum bottles with volume of approximately 125 mL were sealed with rubber septa and used to maintain an anaerobic environment. Five different concentrations of dry mass duckweed were added to 60 mL or 70 mL of manure (winter and summer scenarios, respectively), including 0% as a control, 0.5%, 1%, 2%, and 3% dry mass duckweed. Six sets of these concentrations – making a total of thirty reactors – were used in the analysis. Three of those sets – labeled A, B, and C – were used only for analyzing COD, while the other three sets – labeled D, E, and F – were used to analyze the components of biogases produced from the anaerobic digestion. The reactors were stored in a warm-water bath at 35° C, on a shaker in order to keep the reactors well-mixed.

METHANE MEASUREMENTS

Methane production was measured utilizing manometers to measure displaced volume and gas chromatography (GC) with thermal conductivity detection (TCD) to measure methane concentrations. Gas production was analyzed every two to three days for fortytwo days. The samples were analyzed using a Shimadzu Gas Chromatograph with a molecular sieve column with TCD. Using the ideal gas law, a number of moles of methane produced was calculated for each reactor every few days.

CHEMICAL OXYGEN DEMAND

The effect of duckweed on the change in organic materials in the manure is useful in determining the nutrient value of the manure. Chemical Oxygen Demand (COD) of the reactors was also monitored over the course of the experiment. An alternating schedule of duplicate samples were taken every day for the first three days, and then once a week after that. From each reactor being tested, 2 mL of the manure solution was taken using the syringe. A system of blunt-nosed needles and valves was used for extracting COD samples without introducing oxygen into the digester. It was then diluted with deionized water at a ratio of 1 to 50. Then 2 mL of the diluted sample was transferred to COD vials and heated for 2 hours. After two hours, the samples were tested using a Hach photospectrometry instrument with a high rate COD (HR COD) program.

Results and Discussion

Research results indicated that addition of duckweed increased methane and total gas production in both summer and winter scenarios. Data from the summer scenario is shown in Figure 1. The summer experiment showed that duckweed concentrations of 1-2% dry mass increased the rate of methane production. In controls with no duckweed, no methane was produced over 22 days, while methane production was observed at days >12 in reactors with 0.5 – 2% duckweed. Methane production was greatest at 1 and 2% duckweed; however, increase in methane production may have been undervalued for 2% duckweed at days >20 because greater than the maximum volume of biogas measurable by the manometers was produced. Larger manometers were used for the winter scenario to rectify this error.

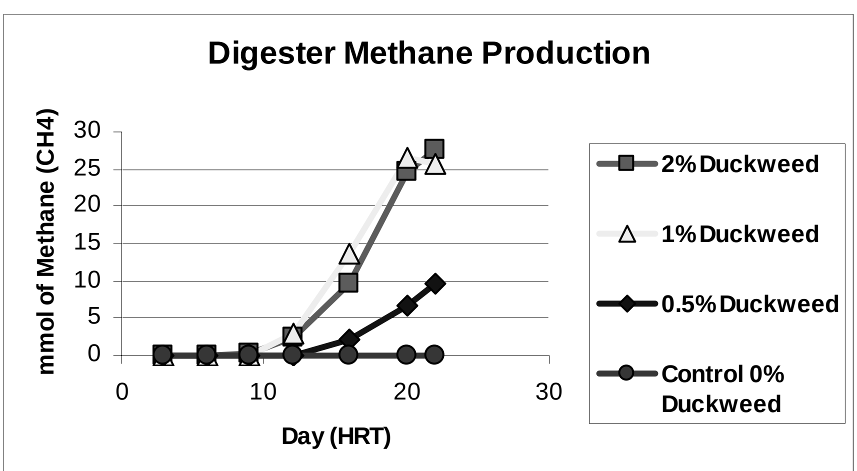

Figure 1. Millimoles of methane produced during summer experiment

In the winter experiment, biogas production from batch digesters performed as expected. A lag phase of 7 - 10 days occurred . Significant methane concentrations (> 50%) appeared on day 14. Peak methane production was achieved by day 20 and then fluctuated around 10 – 17 mmols per day.

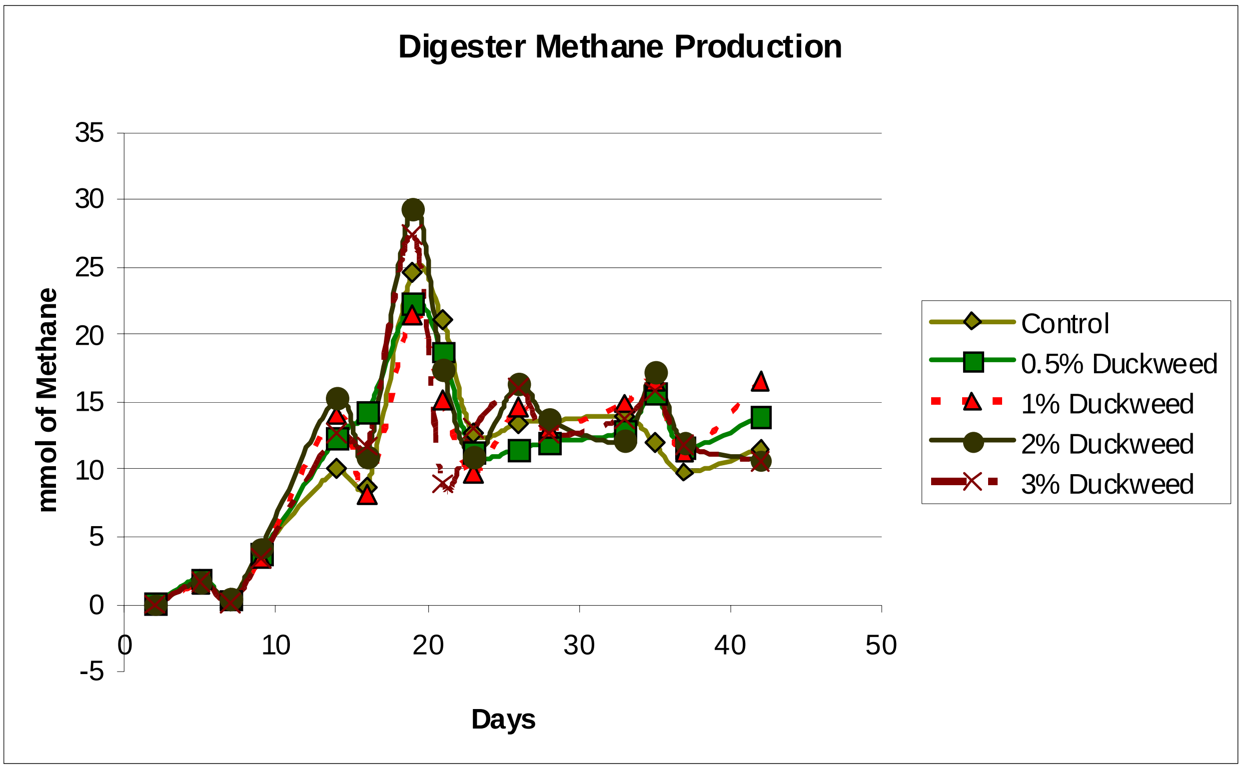

Figure 2. Millimoles of methane produced in winter experiment.

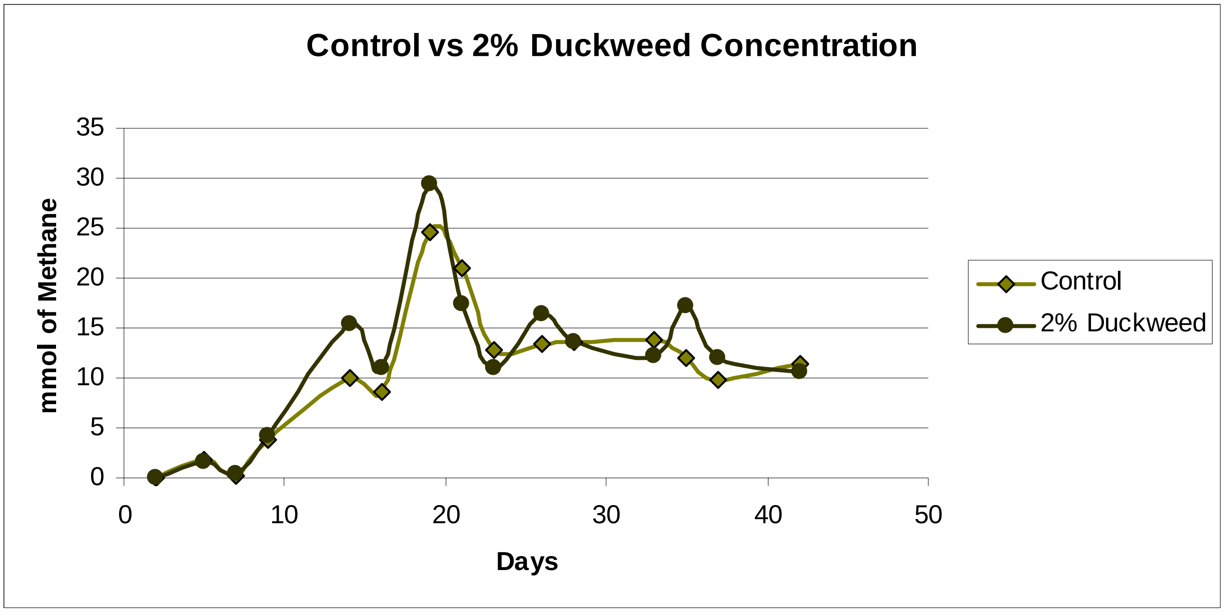

Figure 3. Millimoles of methane produced in winter experiment. Control and 2% Duckweed groups only.

As in previous experiments a concentration of 2% dry mass duckweed appeared to perform best; however, the distinction was not as clear as in the first experiment carried out in June - August 2008. In the winter experiment, all concentration varieties behaved with relative similarity, unlike the summer experiment in which the control group did not produce significant amounts of methane at all for the duration of digestion.

Concentrations of COD in the experimental reactors was generally similar for all concentrations of duckweed, as shown in Figure 4. The initial concentration of COD was 57.6 ± 4.6 g/L and final concentrations were 31.8 ± 6.4 g/L, indicating approximately 45% removal of COD over 35 days. High variability of COD ratios was observed in duckweed reactors due to sampling of duckweed fronds during collection. Additionally, high variability was observed in the initial measurement of COD, indicating that manures used for experiment were not homogeneous. For future batch studies utilizing duckweed and manures, results suggest that maceration of manures and duckweed prior to COD measurements may improve results by decreasing variability. However, to more accurately represent field conditions, measurements should also include non-macerated duckweed and manures (as was completed for this study).

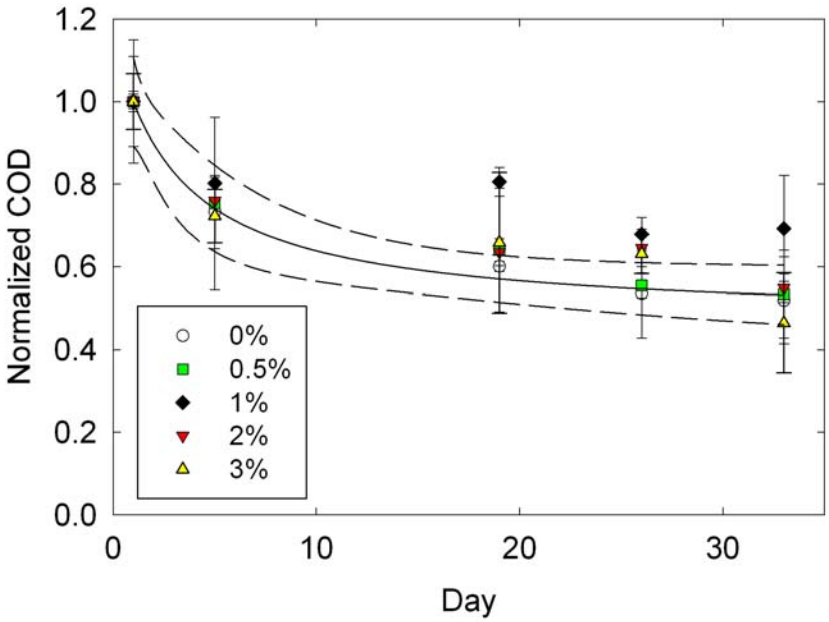

Figure 4. Normalized concentration of chemical oxygen demand (COD) with time for winter scenario. Line represents exponential decay fit of 0% (control) data, with dashed lines representing 95% confidence intervals.

While conditions in both experiments were almost identical, there were some differences which might have led to the observed differences between the results from the summer and winter results. In the second experiment, dry duckweed was used that was not as fresh. It had been left out to dry for several months as opposed to only several days, before being mixed into the reactors. Also in the winter experiment, a shaker bath was used to maintain mixing and temperature instead of an incubator with a platform shaker. The water bath was not as reliable and may not have provided enough mixing. Weekly hand shaking was required to prevent the duckweed from forming a floating mat on top of the manure in the digester.

Conclusion

This study has shown that anaerobic digestion could potentially benefit from the addition of common duckweed species to animal waste. A sustainable cycle in which digestate is treated in a wetland system with duckweed then used to enhance the digestion process could prove beneficial to both farmers and ecosystems. By supplementing with duckweed, digesters could be designed and operated with lower hydraulic retention times. An increased rate of gas production would be directly related to a more efficient energy recovery.

The reason behind duckweed's impact on microbial populations is still unknown, but may be due to provision of minerals and nutrients (Clark and Hillman, 1996). Other hypotheses include the fact that fresh duckweed is sourced from an ecosystem where microbial degradation is prevalent and addition of dry duckweed may shift the microbial community within the digestion reactors. Future studies on the effect of iron on anaerobic digestion could also prove beneficial.

Iron can bond to sulfide and possibly reduce the amount of hydrogen sulfide produced from degrading organic material. Reductions in odor from hydrogen sulfide make the construction of digesters even more appealing to small and large farm operations as urban sprawl becomes increasing common in rural areas. To more fully investigate the role of duckweed in anaerobic digestion, future research includes a continuous digester combined with a constructed wetland in which duckweed can be used to treat digestate and then subsequently harvested for use in digestion.

Acknowledgements

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References

Alvarez, J.A., I. Ruiz, and M. Soto. "Anaerobic Digesters as a Pretreatment for Constructed Wetlands." *Ecological Engineering* 33 (2008): 54-67. In Science Direct[database online]. (accessed February 17, 2009).

Clark, PhD, BSc, P.B., and P.F. Hillman, MSc, BSc, CEng, MICE. "Enhancement of Anaerobic Digestion Using Duckweed (Lemna minor) Enriched with Iron." *Journal of Chartered Institution of Water and Environmental Management* 10 (1996): 92-95.

Cross, J.W., Ph.D. *The Charms of Duckweed.* Missouri Botanical Garden. (2002). http://www.mobot.org/jwcross/duckweed.htm. (April 9, 2009).

DeBusk, T.; Peterson, J.; Reddy, K. “Use of Aquatic and Terrestrial Plants for Removing Phosphorus from Dairy Wastewaters.” *ECOLOGICAL ENGINEERING* 5 (2-3) (1995): 371-390.

Landolt, E. (1986). Biosystematic investigations in the family of duckweeds (Lemnaceae). Zurich, Switzerland, Veroffentlichungen des Geobotanischen Institutes.

Marcias-Corral, Maritza, Zohrab Samani, Adrain Hanson, Geoffrey Smith, Paul Funk, Hui Yu, and John Longworth. "Anaerobic Digestion of Municipal Solid Waste and Agricultural Waste and the Effect of Co-digestion with Dairy Cow Manure." *Bioresource Technology* 99 (2008): 8288-8293. In Science Direct[database online]. (accessed March 18, 2009).

Sakar, Suleyman, Kaan Yetilmezsoy, and Emel Kocak. "Anaerobic Digestion Technology in Poultry and Livestock Waste Treatment - a Literature Review." *Waste Management & Research* 27 (2009): 3-18, http://wmr.sagepub.com/cgi/content/abstract/27/1/3.

Verhoeven, Jos T.A., and Arthur F. M. Meuleman. "Wetlands for Wastewater Treatment: Opportunities and Limitations." *Ecological Engineering* 12 (1999): 5-12.