ABSTRACT

The number of anaerobic digester (AD) systems on dairy and swine farms in the U.S. has approximately doubled in the last 5 years, nearly all at larger feedlot operations. Although the odor reductions due to AD can be fairly obvious to the nose, there is a critical need to better understand some of the more subtle environmental and economic impacts of AD systems. A multi-year collaborative effort was begun three years ago to answer some of these questions, and will wrap up next year. The NRCS-funded project is led by the non-profit organization The Minnesota Project and utilizes the research and outreach expertise of the University of Minnesota. The project is composed of three components, focusing on dairy AD systems. The first, augmented with field trial research, compares soil quality and yield response for commercial fertilizer, stored digested manure, and un-digested manure. The second considers the destruction of weed seeds, a widely-claimed, but little studied, benefit of AD. The project tests viability of weed seeds suspended in an anaerobic digester. The final portion of the project considers the economic performance of AD systems, which will be considered in another paper.

Results to date indicate the following key preliminary conclusions: (1) stored, digested manure can result in crop yields equivalent to undigested manure or fertilizer when applied at similar nitrogen rates, while simultaneously allowing the capture of bioenergy; and (2) the digestion process does not appear to significantly destroy weed seed viability, although germination times may be impacted.

BACKGROUND

The need to more fully understand questions posed by anaerobic digesters was spurred in Minnesota by the installation of an anaerobic digester system at Haubenschild Farms, an 800-cow dairy farm an hour north of Minneapolis/St. Paul. In 1999, the farm installed a heated plug flow digester with a 130-kilowatt engine/generator to utilize the biogas. The
successful operation of this facility (the generator has been running over 98% of the time since start-up) has resulted in much interest from policy-makers as to the applicability of digesters to other animal feedlot operations in the state. Also, there were claims by the digester industry of the superiority of digested manure, with only anecdotal evidence to support them, and there was a desire to more fully investigate these claims.

In 2000, The Minnesota Project, a non-profit environmental and rural development group, was the recipient of a 4-year U.S. Department of Agriculture, Natural Resources Conservation Service (USDA-NRCS) grant to look into several questions related to the environmental performance and financial feasibility of anaerobic digesters at dairy farms, as compared to alternative dairy systems. Specifically, the project had three objectives:

1. Compare effects of three nutrient sources - commercial fertilizer, undigested manure and digested manure - on soil quality, crop growth and nutrient uptake;
2. Determine weed seed survival as affected by manure handling; and
3. Conduct an economic evaluation of alternative manure management systems on dairy profitability.

Results from the third objective will be covered in separate paper.

The Minnesota Project contracted with several partners from the University of Minnesota to conduct much of this work. The results presented below are only preliminary (year 3 of a 4-year project), so they should be interpreted as such. Full results will be available at the end of 2004. More information on the Haubenschild Farm and this grant is available at [www.mnproject.org](http://www.mnproject.org).

### DIGESTED MANURE INTERACTIONS WITH SOIL AND CROPS

A number of studies have shown that manure increases yields over fertilizer applications. This advantage may arise from a number of causes, ranging from slow release nutrient availability to increased soil biological activity to enhanced soil physical properties. Regardless of the specific causal agent, the organic fraction of the manure is associated with this beneficial crop response. Anaerobically digesting manure has little effect on manure’s nutrient composition, although some studies have shown that, on a total solids basis, digester effluent has more total nitrogen (N) and a greater percentage of ammonium than undigested manure.\(^2\) Overall, there is a lack of scientific research on the impact of anaerobically digested manure on crop production and soil properties. The objective of this project is to compare and contrast effects of anaerobically digested and undigested manure sources on: a) soil biological and chemical properties; and b) crop yields.

**Materials and Methods**

Three fields with distinct cropping histories on Dennis Haubenschild’s farm were selected as experimental sites. In 2001 when the study began, the Bruce field had been in CRP for the previous two years and had no history of manure application in the past 30

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years. The Lilac field had been in alfalfa for the last two years and had a history of manure application in the rotation, and the Appel field had been in corn for the last two years and had a history of manure application. The treatment comparisons at the three sites were fresh raw manure and stored digested manure from the Haubenschild farm, and conventional fertilizer applied as urea.

At each of the three sites with different cropping histories, 18 plots (20’ x 200’) were established. The 18 plots represent three replications of each of three treatments: stored digested manure, fresh raw manure, and conventional fertilizer. There are two subtreatments, which represent an annual application of manure or fertilizer (Annual) or applications made in Year 1 and Year 3 (Biennial). The biennial application is intended to highlight the differences in residual or carryover nitrogen from the two manure sources and conventional fertilizer. In 2002, a second year of this experiment was conducted on the three fields previously evaluated in 2001.

At the Haubenschild farm, manure is typically applied at a rate of 6,000 gallons per acre. In order to maximize yield differences between the treatments we applied nitrogen at less than optimum rates, but at identical rates for stored digested and fresh raw manure. In spring 2001, using Dennis’s manure analysis of 40 lbs total N per 1000 gals of manure (results were the same for both digested and raw manure), and assuming 55% availability in the first year, we applied 3,000 gallons per acre, or approximately 66 lbs available N. Urea fertilizer was applied at 75 lb N/acre. In 2002, application rates were again 3000 gals/A manure for the two manure treatments and 100 lb N as urea fertilizer. Manure samples were taken at the time of application in the spring of 2001 and 2002 (see Table 1). In both years starter fertilizer was applied according to Dennis’s usual practice (190 lbs/A of 9-9-31-6.)

Soil samples were collected in 2001 at three dates (4/23/01, 7/3/01 and 8/23/01). Soil was sampled twice per plot at nine row and interrow positions between rows 3 and 5. The first sampling was to a depth of two feet (split into 0-1’ and 1-2’ sample depths) and was analyzed for soil nitrate. At the second and third sampling times, samples were taken with a soil probe to a depth of one foot, and split into 0-6”and 6-12” sample depths. The samples for each plot at the two different depths were composited into one sample per plot at each depth. Samples were delivered to the lab in coolers and air-dried or refrigerated for subsequent analysis. In 2002, samples were again collected at three dates (5/1/02, 7/18/02, and 8/27/02). Grain yield and silage samples were taken at crop harvest in September of both years.

Soil Analysis
Laboratory analysis of soil samples from July and August of 2001 included available nitrogen (potassium chloride (KCl) extractable ammonium and nitrate), potentially mineralizable nitrogen, total carbon and total nitrogen. In 2002, additional analyses included microbial biomass C and N, and particulate organic carbon and nitrogen (results not yet available). To prepare soils for analysis, the soil was sieved to 4mm, then half was air-dried and the rest stored at 4°C. The air-dried soil was sieved to 2mm after drying. A Leco instrument (Leco Corporation, St. Joseph, MI) was used to determine
total percent nitrogen and carbon using 250 mg of air-dried soil. Available N was measured by extracting 30 g of air dried soil with 150 ml KCl, and analyzing on a Lachat (Lachat Instruments, Milwaukee, WI).

Soil microbial biomass carbon and nitrogen was determined for refrigerated soil by moistening it to 60% water-filled pore space. Two sets of duplicate samples were placed in desiccators; one set was fumigated with chloroform and the other served as controls. After two days of fumigation treatment, the beakers of moistened soil were incubated for 10 days at 25° C in canning jars along with NaOH traps to collect CO2. The trapped C was then analyzed on a Dohrmann C analyzer (Tekmar-Dohrmann, Cincinnati, OH). Carbon dioxide evolved from the controls during the incubation was subtracted from the fumigated samples and the result was multiplied by a correction factor to obtain a value for microbial biomass carbon. Biomass nitrogen was extracted with 30 mls of KCl for 3.5g of soil. After being shaken for 30 min and centrifuged at 4000 rpm for 10 min, 15ml of KCl was pipetted into scintillation vials and frozen, to be analyzed later on the Lachat.

Potentially mineralizable nitrogen (PMN) was determined by using 30g of air-dried soil and adding 8.5mls of water to maintain 60% water filled pore space in a canning jar. A 5ml vial of water was placed in the jar to keep the soil moist. Jars were incubated for 28 days at 25 degrees C. After 28 days the soil was extracted with 150 mls KCl to determine available N.

Results

Manure analysis

Manure samples were collected from the raw and stored digested manure at time of application to the plots in spring 2001 and 2002, as shown below.

Table 1. Manure analysis for samples collected in Spring, 2001 and 2002.

<table>
<thead>
<tr>
<th></th>
<th>Dry matter %</th>
<th>Nitrogen lbs/1000 gals</th>
<th>P2O5</th>
<th>K2O</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Spring 2001</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Raw</td>
<td>8.9</td>
<td>32.9</td>
<td>12.5</td>
<td>32.3</td>
</tr>
<tr>
<td>Stored digested</td>
<td>5.5</td>
<td>30.4</td>
<td>10.5</td>
<td>29.5</td>
</tr>
</tbody>
</table>

| **Spring 2002**| | | |
| Raw            | 7.6          | 30.2                   | 11.9 | 39.6|
| Stored digested| 5.7          | 31.2                   | 9.3  | 35.5|

Total nutrients were similar (on a liquid basis) at the two spring samplings, which suggests that similar amounts of nitrogen were applied to the plots from the raw and stored digested manure sources (about 100 lbs/A of total nitrogen each year).

Manure samples were also collected in October, 2002 from the raw manure mix pit, the fresh digested manure immediately after digestion, and the stored digested manure in the lagoon (at time of mixing and field application). The stored digested samples had lower levels of total and available (ammonium and nitrate) nitrogen than the raw manure or
fresh digestate when compared on a wet basis (per 1000 gallons), but amounts were higher on a dry weight basis (Table 2). These differences are partly due to losses of solids during digestion (10% to 7.3% dry weight), and subsequent dilution in the lagoon (7.3 to 4.4% dry weight).

Table 2. Manure analysis for samples collected in October, 2003.

<table>
<thead>
<tr>
<th></th>
<th>Dry wt</th>
<th>Total N</th>
<th>InorgN</th>
<th>Total N</th>
<th>InorgN</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>%</td>
<td>-----lbs/1000 gal-----</td>
<td>% (dry wt basis)</td>
<td>% (dry wt basis)</td>
<td></td>
</tr>
<tr>
<td>Raw</td>
<td>10.0</td>
<td>33.2</td>
<td>15.7</td>
<td>4.0</td>
<td>1.9</td>
</tr>
<tr>
<td>Fresh digested</td>
<td>7.3</td>
<td>32.7</td>
<td>20.2</td>
<td>5.4</td>
<td>3.3</td>
</tr>
<tr>
<td>Stored digested</td>
<td>4.4</td>
<td>22.3</td>
<td>13.3</td>
<td>6.1</td>
<td>3.6</td>
</tr>
</tbody>
</table>

Grain yields
Results for grain yield are shown in Fig. 1 (2001) and Fig. 2 (2002). Yields did not differ for any of the three soil treatments, although yields did differ among the three field sites due to variations in previous management history and soil properties. Yields were lower than typical yields obtained on the farm (see “typical” bar in Fig. 2) since N availability was limited by design. By avoiding a situation of luxury nutrient supply, we hoped to maximize the differences in effects of the three nutrient sources.

Figure 1. Grain yield for the three field sites on the Haubenschild farm in 2001. Averages of the annual and biennial applications are presented. Error bars represent LSD values for p=0.05. There were no significant differences among yields for any of the three treatments.
Figure 2. Grain yield for the three field sites on the Haubenschild farm in 2002. Averages of the annual and biennial applications are presented. The bar labeled “typical” shows the yields obtained outside the experimental plots. Error bars represent LSD values for $p=0.05$. There were no significant differences among yields for any of the three treatments.

Soil nitrogen

Amounts of available nitrogen (KCl-extractable ammonium and nitrate) measured in air-dried soil samples and released after a 28 day incubation at optimal moisture and temperature (PMN) also were not different for the three treatments (Figs. 3 and 4).
Figure 3. Amounts of available nitrogen (KCl-extractable ammonium and nitrate) and potentially mineralizable nitrogen (extracted after 28 days incubation) are averaged over 0-6” and 6-12” depths for the August, 2001 sampling time. Error bars represent the standard deviation for the mean of 6 replicate samples.
Figure 4. Amounts of available nitrogen (KCl-extractable ammonium and nitrate) and potentially mineralizable nitrogen (extracted after 28 days incubation) are averaged over 0-6” and 6-12” depths for the annual fertilizer or manure application at the August, 2002 sampling time. Error bars represent the standard deviation for the mean of 3 (Bruce and Lilac fields) or 6 (Appel field) replicate samples.

Microbial biomass
As an indicator of differences in biological activity, we measured microbial biomass carbon. Digested manure had equivalent microbial biomass to fertilizer and raw manure treatments except in the Bruce field, where the raw manure treatment had a higher biomass (Figure 5).
Summary
Results from two growing seasons have shown that use of stored, digested manure can result in crop yields equivalent to undigested manure or fertilizer when applied at similar nitrogen rates, while simultaneously allowing the capture of bioenergy. Neither were there significant differences in the amounts of available nitrogen or microbial biomass measured in soil samples from field plots treated with these three amendments. Further testing is being carried out to see whether nitrogen supplied by mineralization is different for these amendments when they are incubated with the same, homogeneous soil under ideal conditions in the laboratory.
WEED SEED SURVIVAL AS AFFECTED BY MANURE HANDLING

Introduction
The potential for weed seed introduction to cropland through the application of manure is a question faced by many farmers. One potential benefit of anaerobic manure digestion includes a reduction in weed seed germination and viability. In a field study, this project will determine the effect of anaerobic digestion on germination of weed seeds common to this system. Because of the uncertainties of weed seeds origin and health, and to work within the limited funding available, weed seeds were obtained form a known source and were characterized for potential germination and viability to establish a baseline prior to inclusion in the study. To track this “needle in a haystack” weed seed lot, the seed needed to be contained and retrievable and so were placed in nylon mesh bags. For the two manure storage treatments, mesh bags were placed in either the anaerobic digester or in raw, undigested manure prior to its entering the digester. Seed bags were in manure treatments for 20 days (length of time for one batch of manure to pass through the digester though weed seeds may pass through at different rates depending on size, solution/suspension density, etc.). Seeds were placed in the end of the anaerobic digester where the manure exits prior to entering the storage lagoon, as this position was the only available internal access from which to introduce the seeds and ensure they could be retrieved again. Though not ideal, we feel this approach most closely reflects anaerobic digester conditions, considering limitations of internal access to the digester and the need to track a seed lot know origin and viability in lieu of using a simulated digester.

Materials and Methods
Six weed species were chosen in part to reflect species typically found in manure in the region and to include representatives of weed seed groups. Species include grass and broadleaf species, large and small seeded species, true seeds and achenes (smartweeds), and species with known impermeable, protective seed coats (velvetleaf). Weed Species included in this experiment are as follows:

1. velvetleaf (*Abutilon theophrasti*)
2. common lambsquarters (*Chenopodium album*)
3. redroot pigweed (*Amaranthus retroflexus*)
4. wild proso millet (*Panicum miliaceum*)
5. giant foxtail (*Setaria faberi*)
6. ladysthumb smartweed (*Polygonum persicaria*)

Weed seeds were collected from the Rosemount Experiment Station during the Fall of 2001. Seeds were cleaned and stored at room temperature until used in experiments. Seed germination was tested by placing seed in a petri dish between moistened filter paper at 24 C for 14 days in the light. Seeds with emerged radicals were counted as germinated (Buhler et al. 1999). Viability of 400 of seed from each species was determined by placing seed in a petri dish between moistened filter paper for a minimum of 48 hours in a germinator at 24 C, then treated with a 1% (w/v) solution of tetrazolium. Seeds were considered viable if the embryo stained red.
For each treatment, all six species of weed seeds were combined into one mesh bag. One hundred seed per species was added to each bag. Each treatment was replicated six times. Since most, (but not all) of the weed seed in the system will pass through a cow, we subjected seed to an in vitro rumen fermentation procedure (Marten and Barnes, 1980) which has been proven to simulate conditions of a cow’s digestive system. In this procedure, weed seeds were soaked in rumen fluid for 48 hours in an Ankom Daisy fermentor oven. Next, the weeds were immersed in a pepsin and hydrochloric acid solution for 24 hours to simulate passage through the stomach.

The experiment had the following three treatments:
1. anaerobic digested manure, +/- weed seeds
2. conventional lagoon stored manure, +/- weed seeds
3. inorganic fertilizer control, +/- weed seeds

Wooden frames, 15 x 18 inches, were placed in an area previously in sod. The sod was removed and the soil worked to a depth of approximately 3 inches. A soil sample was taken and analyzed. Wooden frames were placed 2 inches into the soil. In late November, 2001, weed seeds were removed from the digester or conventional manure storage, placed in the appropriate wooden frame and incorporated into the soil. Digested or non-digested manure was added to the wooden frames at a rate of 6000 gal/A, a rate used by Dennis Haubenschild in fields where corn will be planted the following year.

In mid-April of 2002, ammonium nitrate (34-0-0) at a rate of 529 lb/A was added to each plot requiring inorganic fertilizer. The number of germinating weed seeds was recorded on a monthly basis and weeds were hand pulled from the plot. In April of 2003, soil within each frame will be mixed to a depth of two inches and germinating seedlings will again be counted and removed. Each treatment was replicated six times and the experiment is being repeated. Results from the experiment were analyzed as a randomized complete block design and means were separated with a Least Significant Difference test at the 0.05 level of significance.

Results and Discussion
Viability of weed seed used in the experiment ranged from 99% for velvetleaf to 81.5% for wild proso millet (Table 1). No giant foxtail or wild proso millet seeds germinated in plots from any treatment during spring or summer of 2001. This is most likely due to treatment of the weed seed in rumen fluid and simulated stomach acid. There were no differences in number of broadleaf seeds germinating between conventional lagoon and anaerobic digestion manure treatments, with one exception. Velvetleaf seeds had higher rates of germination after the anaerobic manure treatment (Table 2). However, it must be noted that of 100 seeds of each weed species added to every plot, only smartweed seeds had a high rate of germination. The remaining weed seeds may still be dormant and may germinate next summer. Counting and removing all weed seedlings in each plot next summer will give us additional information about the effect of each manure treatment on weed seed germination. It must also be noted that there were many smartweed seeds present in the soil in addition to those added for this experiment, which will be accounted
for through the use of control treatments without weed seed added. These results are preliminary as the study will be repeated in time and space.

**Table 1.** Percent germination and viability of weed seed used in anaerobic digestion experiment, prior to rumen digestion. Fall, 2001.

<table>
<thead>
<tr>
<th>Species</th>
<th>Germination* (%)</th>
<th>Viability* (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>pigweed</td>
<td>2.3</td>
<td>91.8</td>
</tr>
<tr>
<td>common lambsquarters</td>
<td>11.3</td>
<td>88.3</td>
</tr>
<tr>
<td>wild proso millet</td>
<td>0</td>
<td>81.5</td>
</tr>
<tr>
<td>Ladysthumb smartweed</td>
<td>0</td>
<td>94.5</td>
</tr>
<tr>
<td>velvetleaf</td>
<td>0</td>
<td>99.3</td>
</tr>
<tr>
<td>giant foxtail</td>
<td>0.8</td>
<td>88.3</td>
</tr>
</tbody>
</table>

*Mean of 4 lots of 100 seeds for each species.

**Table 2.** Numbers of weed seed germinating in the spring and summer after 20 days of fall storage in different manure storage systems. Haubenschild Farms. 2002.

<table>
<thead>
<tr>
<th>Manure system</th>
<th>With weed seed added</th>
<th>Without weed seed added</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CLQ</td>
<td>PW</td>
</tr>
<tr>
<td>Anaerobic Digestion</td>
<td>21</td>
<td>4</td>
</tr>
<tr>
<td>Lagoon Storage</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>Inorganic Fertilizer</td>
<td>11</td>
<td>5</td>
</tr>
<tr>
<td>LSD (0.05)</td>
<td>NS</td>
<td>NS</td>
</tr>
</tbody>
</table>

1 No giant foxtail and wild proso millet seed germinated in plots.
2 CLQ = common lambsquaters
   PW = pigweed spp.
   VL = velvetleaf
   SW = smartweed spp.