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The Impact of Carbohydrate and Protein Level and Sources on Swine Manure Foaming Properties

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Abstract. *This study explored the impact of swine diet on the composition, methane production potential, and foaming properties of manure. Samples of swine manure were collected from controlled feeding trials with diets varying in protein and carbohydrate levels and sources. Protein sources consisted of corn with amino acids, corn-soybean meal with amino acids, corn-soybean meal, corn-canola meal, corn-corn gluten meal, and corn-poultry meal. Carbohydrate sources consisted of corn-soybean meal, barley, beet pulp, distillers dried grains with solubles (DDGS), soy hulls, and wheat bran. Manure samples were tested for a number of physical and biochemical parameters, including total solids, volatile solids, viscosity, density, methane production rate, biochemical methane potential, foaming capacity, and foam stability. Statistical analyses were performed to evaluate whether different carbohydrate and/or protein ingredients affected these physico-chemical properties or the samples' ability to produce methane gas. After conducting these trials, another feeding trial was performed to evaluate if the addition of Narasin into rations (corn-soybean and DDGS) could reduce the methane production rate or potential of the manure. These samples were also tested for the physical and biochemical parameters mentioned previously. Finally, an additional manure foaming study was conducted involving the addition of specific carbohydrates ground to different particle sizes and corn oil to observe the effects that the additives had on foaming capacity and stability.*

Keywords. *Swine manure, foaming, swine diet, anaerobic digestion, methane production*

Introduction

The accumulation of foam on the surface of deep pit manure storages is a serious concern for pork producers for a number of reasons. On a practical level, excess foam accumulation can significantly reduce the amount of volume available for manure storage in deep pit systems. This reduction of storage space may force farm managers to apply manure during untimely seasonal windows or seek other means of storage. Foam accumulation also has important implications for the impact of swine facilities on the surrounding environment and the overall safety of swine facilities. Along these lines, foam has shown the capacity to trap gases produced by the anaerobic decomposition of swine manure. When the foam layer is broken suddenly, dangerous levels of flammable gases present in the biogas mixture (most significantly methane) can occur. Numerous facilities have reported flash fires or explosions due to the combination of foam layer breakage and an externally introduced spark or flame (Moody et al., 2009).

The goal of this study was to analyze manures produced in controlled diet studies for several key parameters thought to play a significant role in the overall foaming characteristics of deep pit systems. Figure 1 shows some examples of the manure collected, as well as foam-type accumulation after inducing aeration of the manure to simulate enhanced biogas generation. Figure 1 (a) shows foam produced at the surface of manure collected from a single pig during a diet study after aerating the tank with a single disk aeration system developed by this group. Figure 1 (b) shows bubbles accumulated at the surface of manure collected during a similar study after adding a carbohydrate source, allowing it to incubate for a weak, and then stirring gently. In both cases, manure collected from diet studies showed some capacity to capture bubbles that were introduced by external means for the purpose of observation.

(a) **(b)**

Figure 1. (a) Foam accumulation after aeration on the surface of a tank used to collect manure in the diet studies and (b) a picture of the surface of a sample from the bucket additive study after mixing gently.

Foaming manure was considered a “three-phase system” during this study in order to maintain a conceptual framework to better understand the accumulation of foam on deep pits. Davenport and Curtis (2002) originally established this framework as useful means of characterizing the production of foam in municipal anaerobic digesters. In both three-phase systems, the initiation of foam production occurs as a result of both the gas and liquid phases working together to capture bubbles produced by the system at the interface of the liquid layer and the atmosphere. In anaerobic systems, the gas phase is a result of biogas production due to methanogenic activity. When a significant concentration of surface active agents is present in the liquid layer, it facilitates foam production by lowering the surface tension of the solution (Glaser et al., 2007; Davenport et al., 2008). Finally, solids in the form of hydrophobic substances are thought to stabilize the foam that is produced by preventing liquid drainage back into the liquid layer (Bindal et al., 2002; Horozov, 2008; Heard et al., 2009).

As opposed to anaerobic systems in municipal settings, where the input consists of both primary (raw organics) and secondary settled waste (waste activated sludge), the input of deep pit manure storages consists entirely of manures, wasted feed and water, and wash waters. In this way, there is a well-established link between feed composition and the physical and chemical characteristics of manure storages (Kerr et al., 2006; Jarret et al., 2011), so this group hypothesized that the various diets isolated in this trial would have a significant effect on the physical characteristics of the manure, the gas production potential, and the foaming capacity and stability of the manure samples.

Materials and Methods

Three different types of diet studies were conducted at the Iowa State University Swine Nutrition and Management Research Center. Each study consisted of various protein and carbohydrate based diets fed to 24 isolated subjects weighing approximately 100 kg. The diets were distributed evenly depending on the number of diet types involved (i.e. 4 repetitions of each diet for the protein and carbohydrate trials, and 6 repetitions of each diet for the Narasin trial). Table 1 shows the diets and corresponding labels used for each trial. In the protein study, the level and source of dietary protein was varied. Diets A, B, and C for the protein study shown below represented relatively low, medium, and high levels of crude protein, respectively. Diets D, E, and F of the same study utilized alternative protein sources to the soybean meal used in diet C, but at the same overall level. In the carbohydrate study, diet A represented a control diet typically fed in swine producing facilities. The subsequent diets differed in the source of complex carbohydrate at equal crude protein and neutral detergent fiber levels as diet A, thus evaluating the impact that different carbohydrate digestibility had on the manure properties.

Table 1. Diet types with corresponding labels and abbreviations used for each of the three studies.

Protein Study			Carbohydrate Study			Narasin Study		
A	Corn with Amino Acids	C/AA	A	Corn-Soybean Meal	C-SBM	A	Corn-Soybean Meal	C-SBM
B	Corn-Soybean Meal with Amino Acids	C-SBM/AA	B	Barley	B	B	Corn-Soybean Meal with Narasin	C-SBM, Narasin
C	Corn-Soybean Meal	C-SBM	C	Beat Pulp	BP	C	Corn with DDGS	DDGS
D	Corn-Canola Meal	C-CM	D	Distillers Dried Grains with Solubles	DDGS	D	Corn with DDGS and Narasin	DDGS, Narasin
E	Corn-Corn Gluten Meal	C-CGM	E	Soy Hulls	SH			
F	Corn-Poultry Meal	C-PM	F	Wheat Bran	WB			

The protein and carbohydrate studies were conducted in two separate trials of 40 days each, while the Narasin study was conducted in a single 40 day trial. During each trial, subjects were fed 2 kg of the designated feed twice daily. After each feeding session, waste was collected and deposited in an individual tank corresponding to each subject. At the completion of each 40 day trial, samples were collected and stored in a cooler at 4°C.

Total Solids and Volatile Solids

The total solids and volatile solids contents of manure samples were tested according to the Standard Methods for the Examination of Water and Wastewater 2540B and 2540E (APHA, 2000). Approximately 30 mL of a manure sample was poured into a pre-weighed porcelain dish after thorough mixing. After obtaining the weight of the full crucible, the sample was dried in a 104°C oven for approximately 24 hours. After drying the sample was weighed again. The percent of total solids was determined by equation 1 below.

$$\%\ \text{Total Solid} = \frac{\text{Weight of Dried Sample and Dish} - \text{Weight of Crucible}}{\text{Weight of Wet Sample and Dish} - \text{Weight of Crucible}} \times 100 \quad (1)$$

After obtaining the dried weight of the sample, the crucible with the dried contents was placed in a muffle furnace at 550°C for approximately 8 hours. Once cooled, the final weight of the ash and crucible was obtained, and the volatile solids content was determined by equation 2.

$$\%\ \text{Volatile Solids} = \frac{\text{Weight of Dried Sample and Dish} - \text{Weight of Ash and Dish}}{\text{Weight of Wet Sample and Dish} - \text{Weight of Crucible}} \times 100 \quad (2)$$

Biochemical Methane Potential Assay

The biochemical methane potential (BMP) of a sample defines the anaerobic biodegradability of a given material (Owen et al., 1979). Specifically, the BMP test yields the total volume of methane able to be produced over a long-term digestion period and the potential efficiency of anaerobic digestion a particular sample could achieve. Typically the results are normalized to methane produced per gram of volatile solid added.

The procedure in assessing the BMP of the swine manure samples collected for this study was to add 20 to 25 grams of sample to a 250 mL serum bottle (Wheaton Science Products No.:223950), with the exact mass recorded. This mass of sample was selected based on an estimated 300 mL of CH_4 produced per gram of volatile solids added as suggested by Vedreene et al. (2008), Hashimoto (1984), and Burton and Turner (2003) who suggested a range of 244 to 480 L CH_4 per kg volatile solids. Next, 50 mL of inoculum was added from an active anaerobic digester maintained in the Agricultural Waste Management Laboratory (AWML) at Iowa State University. This volume of inoculum was added to approximately achieve a 2:1 mass ratio of volatile solids from the manure to inoculum, with the actually ratio varying due to the exact volatile solids content of the manure. Finally, the solution was diluted to approximately 150 mL with a nutrient medium as per Moody et al. (2011) and sealed with a sleeve stopper septa (Sigma-Aldrich Z564729).

Once the sample was prepared, it was incubated at 35°C while being constantly agitated. The samples were regularly checked for biogas production with a gas-tight syringe (Micro-Mate interchangeable hypodermic Syringe 50cc Lock Tip, Popper & Sons, Inc. New Hyde Park, New York). When the syringe was filled with sampled biogas, it was injected into an infrared methane analyzer (NDIR-CH4 Gasanalyzer University Kiel, Germany) to obtain the percent of methane present in the sample.

For this paper, the amount of methane produced by each sample up to the 40-day incubation mark was used because samples had not finished producing biogas at the time of submission. The volume of methane produced by each sample was normalized the mass of volatile solids added to the sample for comparison purposes.

Methane Production Rate Assay

The goal of the methane production rate assay was to provide a short term biogas production measurement with a relatively simple procedure. While the methane production rate (MPR) test is similar to the BMP assay, it is unique in a number of ways. First, the test is conducted over a much shorter incubation time (approximately 3 to 7 days compared to over 40 days for the BMP assay) to ensure that the sample does not approach substrate limiting conditions. Also, the manure sample used for the MPR assay was not inoculated or diluted; rather, the ability of the endogenous bacteria to produce biogas was evaluated by adding a single volume of the sample. Finally, the sample was incubated at room temperature rather than at 35°C, and the sample was kept stationary rather than agitated. Keeping the sample stationary allowed the observer to record the amount of surface accumulation, foam or otherwise, that developed on the sample.

The procedure for the MPR test involved adding approximately 100 mL of well-mixed sample to a 250 mL serum bottle similar to that used for the BMP assay. Upon the sealing of the sample with a sleeve stopper septa, the exact time was recorded along with the mass of sample added to the bottle. Next, the sample was incubated at room temperature (approximately 23°C). An incubation period of approximately seven days was selected based on preliminary trials by this group. Once the seven day incubation period was over, the sample was checked for biogas production with the gas-tight syringe and analyzed for methane content using the NDIR-CH4 Gasanalyzer. During the analysis of the biogas produced, the accumulation of foam and/or solids on the surface of the sample was observed and recorded. Figure 2 shows a set of samples after seven days of incubation.

Figure 2. A set of samples after seven days of incubation for the methane production rate assay.

The rate of biogas production and the rate of methane production were calculated using equations 3 and 4.

$$BPR(\frac{L}{L*day}) = \frac{\text{Biogas Produced (mL)} \times \rho_{manure}(\frac{g}{mL})}{\text{Mass of sample (g)} \times \text{incubation period(minutes)}} \times \frac{1440 \text{ minutes}}{day} \quad (3)$$

$$MPR\left(\frac{L}{L*day}\right) = \frac{(\text{Methane }\% \frac{1}{100})(\text{Biogas Produced (mL)} + V_{headspace}) \times \rho_{manure}(\frac{g}{mL})}{\text{Mass of sample (g)} \times \text{incubation period(minutes)}} \times \frac{1440 \text{ minutes}}{day} \quad (4)$$

Foaming Capacity and Stability Testing

The foaming capacity and stability apparatus used in this study, as well as the parameters used to evaulate the foaming characteristics of swine manure, were adapted from a number of other studies, including Ross et al. (1992), Bindal et al. (2002), Bamforth (2004), and Hutzler (2011). Air was passed through an in-line gas regulator (Restek Model 21666) directly into a 2-inch diameter clear PVC column. The flow rate of air through the column was measured and controlled with a variable area flow meter (Dwyer RMA-SSV). For the purposes of this experiment, it was determined that a flow rate of 200 cubic centimeters per minute (0.0033 L/s) was appropriate based on preliminary trials. In order to conduct the foaming capacity experiment, a sample volume of approximately 300 mL was poured into the column and the initial level was recorded based on measuring tape placed on the columns. The sample was then aerated through a cylindrical air stone at 0.0033 L/s until a steady state height was reached or the foam layer reached the maximum height of the column. The time of aeration was recorded along with the height of foam produced and the level of the foam-liquid interface. A foaming capacity index was calculated as the height of foam produced divided by the initial manure level and multiplied by a factor of 100.

The foam stability measurement occurred immediately after the foaming capacity was determined. Once aeration ceased, the final height of foam became the initial level recorded at time zero. Once this level was established, the descending height of the foam was recorded at expanding time intervals. Simultaneously, the ascending level of the foam-liquid interface was recorded at the same time intervals. The descending height of foam was normalized to percent of initial foam height and plotted as a function of time. A first-order exponential decay model fit the data well in most cases. The half-life of the foam was determined with equation 5 as a measure of the foam stability.

$$t_{\frac{1}{2}}(\text{minutes}) = \frac{\ln(2)}{\text{decay coefficient k}} \quad (5)$$

Bucket Additive Study

An additional study was conducted at the Iowa State University Swine Nutrition and Management Research Center to observe the effects of various feed additives to the ability of a sample to produce foam with the lab-scale apparatus described above. In this study, manure produced in the Narasin study from the corn diet with DDGS was collected in a single tank. Then, about 6000 grams of manure was distributed into 64 buckets and mixed with various types of additives as described in table 2. The order of additives shown in table 2 was repeated for each set of eight buckets to give eight repetitions of each mixture.

Table 2. Additive types with corresponding labels used for each of the three trials.

Bucket	Additive	Abbreviation	Amount Added to Bucket
1	None (Control)	Control	-
2	Ground DDGS	G-DDGS	200 g
3	Unground DDGS	U-DDGS	200 g
4	Ground Wheat Midds	G-WM	200 g
5	Unground Wheat Midds	U-WM	200 g
6	Ground Soybean Hulls	G-SH	200 g
7	Unground Soybean Hulls	U-SH	200 g
8	Corn Oil	CO	30 g

Once all samples were mixed into the 5-gallon buckets, they were allowed to ferment for varying lengths of times before being sampled. Buckets 1-24 were sampled after 7 days, buckets 25-48 were sampled after 14 days, and buckets 49-64 were sampled after 21 days. Once sampled, the manure mixtures were tested with the foaming capacity and stability apparatus.

Statistical Analysis

Statistical analysis was performed using JMP Pro 10 (JMP Pro, Version 10. SAS Institute Inc., Cary, NC, 1989-2012). Fixed factors were established according to the diet type associated with each sample and the trial period. For the bucket additive study, fixed factors included the mixture type as well as the fermentation time in days.

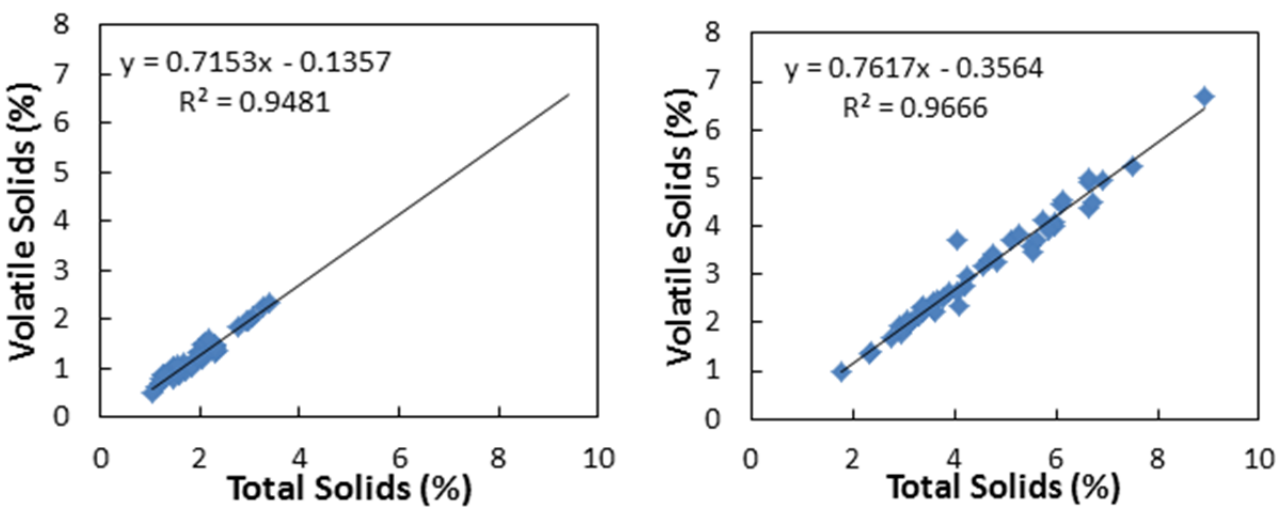
Results and Discussion

Protein and Carbohydrate Studies

The distributions of solids of all samples from the protein and carbohydrate studies are represented below in figure 5. These figures show a relatively consistent volativity (the fraction of volatile solids with respect to total solids) for samples from both trials within each study. The average volativity of samples from the protein and carbohydrate studies are 63.9% and 67.3%, respectively. The regression lines shown in figure 3 (a) and (b) had relatively similar slopes, indicating some baseline inorganic solids content in the manure and then an increase in volatile solids content of approximately 0.7-0.8 units for every unit increase in total solids.

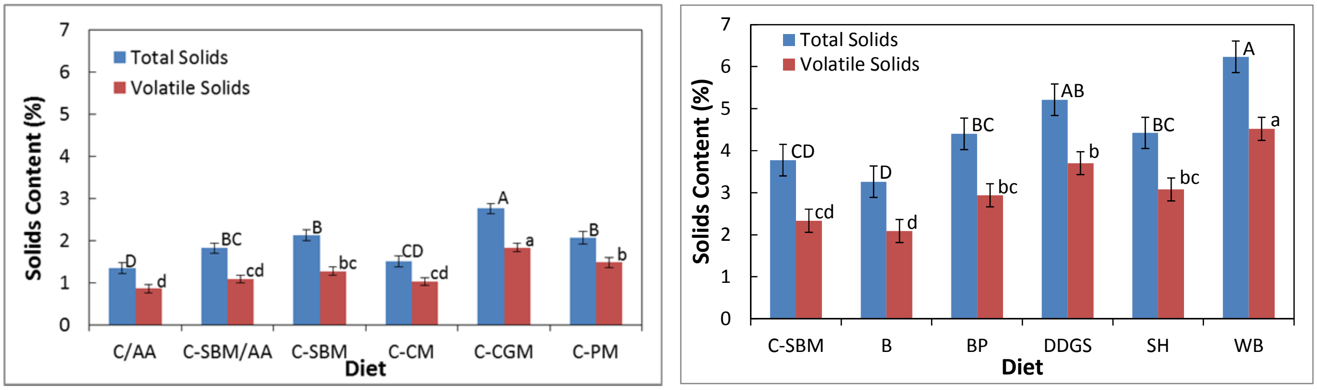
The statistical analysis of the protein study total solids concentrations indicated that the trial wasn't significant ($p = 0.0748$), but that diet was ($p < 0.0001$). Analysis of volatile solids indicated that both trial ($p = 0.0116$) and diet ($p < 0.0001$) were significant. Trial was used as a blocking variable that controlled for external temperature and water consumption by the pigs. The results indicated that the corn-corn gluten meal had significantly higher total and volatile solids concentrations than manures generated from the other diets, while the corn with amino acids diet had the lowest total and volatile solids content of all the diets. Results are summarized in figure 4 (a).

Statistical analysis of total solids concentrations in the carbohydrate diet indicated that both trial and diet were significant ($p < 0.0001$). In this analysis, trial was used as a blocking variable that was related to weather conditions (temperature) at the time of the study. The results indicated lower total solids concentrations on average during the first trial (3.8%) than the second trial (5.3%). A similar trend was seen for volatile solids concentrations with both trial and diet being significant ($p < 0.0001$) while the interaction of trial and diet were not significant ($p = 0.1764$). Treatment effects of diet on both total and volatile solids are shown in figure 4 (b). With respect to the diet types, the wheat bran and distillers grain diets showed the highest percentage of total solids while the corn-soybean meal and barley diets yielded the lowest solids content. In general, the carbohydrate study yielded samples with higher solids contents then the protein diets, as well as a much greater range of values.



(a) **(b)**

Figure 3. (a) Volatile Solids vs. Total Solids for protein trials 1 and 2 (combined) and (b) carbohydrate trials 1 and 2 (combined).

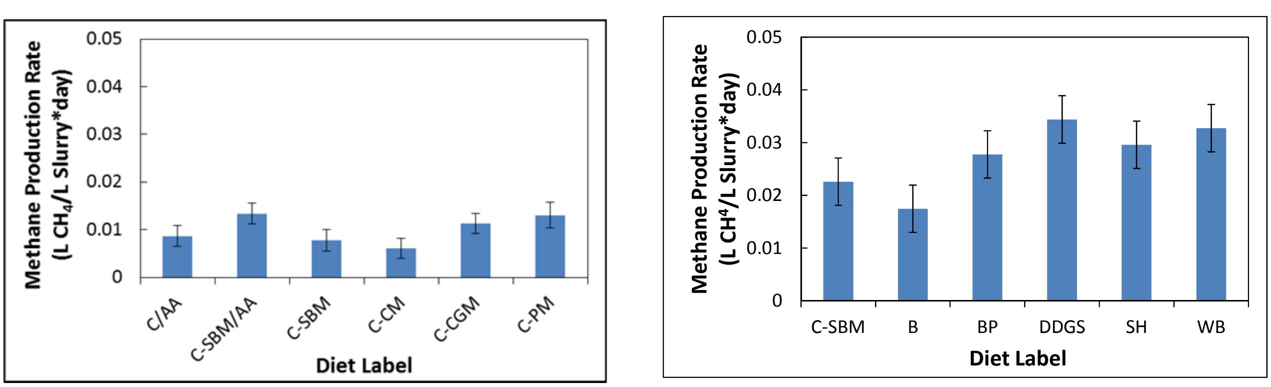


(a) (b)

Figure 4. Average total and volatile solids concentrations for swine feed diets of differing (a) proteins and (b) carbohydrates levels and sources with abbreviations from table 1. Error bars represent the standard error of the mean. Capital letters indicate differences among total solids concentrations of the differing diets and lower case letters represent differences among volatile solids concentrations.

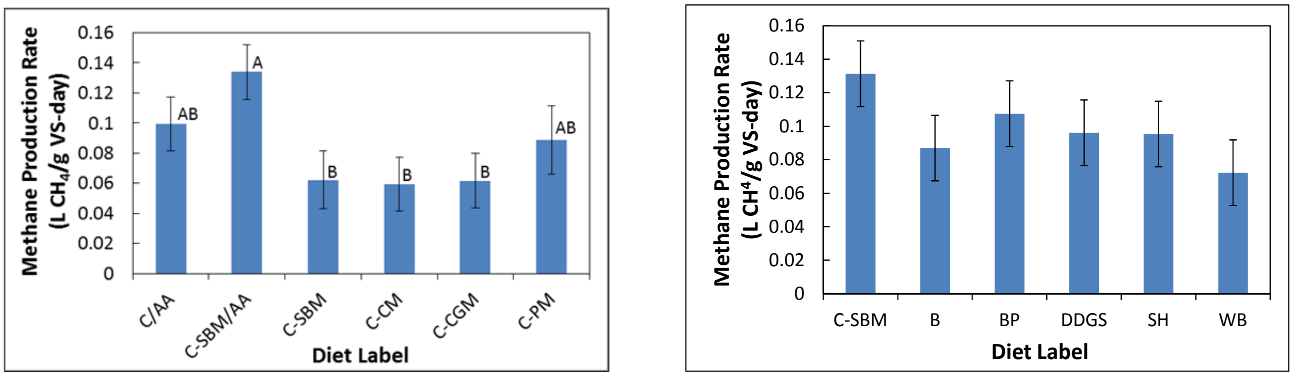
The average methane production rates of both studies are shown in figure 5. On average, the samples from the protein study had smaller magnitudes of methane production rates than those from the carbohydrate study. This group believes that this was due to the lower solids concentration that diluted the strength of the manure. The statistical analysis did not find a difference for the impact of diet on MPR ($p = 0.1465$). However, results were slightly different if the manures were normalized to the rate of methane production per gram of volatile solids. In this case the impact of diet was significant ($p = 0.0391$) with the corn-soybean meal with amino acids diet have significantly greater ($\alpha = 0.05$) rates of methane production than the corn-soybean meal diet, the corn-corn gluten meal, and the corn-canola meal diet (figure 6 (a)). For the carbohydrate diet study, no statistical differences were found for methane production rate on a whole sample basis or when normalized per gram of volatile solids. However, the methane production rates for the DDGS and wheat bran diets had higher values than the barley-based diet on average. These differences may speak to the efficiency at which certain diets are digested. That is, DDGS and wheat bran contain more hemicellulose and lignin than other diets studied, which are digested more slowly by microbes.

One last note with respect to the values of MPR reported in this paper is that they show substantially lower values compared to swine manure collected from deep pit storages. In fact, the numbers in this study were approximately one order of magnitude smaller than those collected from a deep pit and tested similarly by this group (Van Weelden et al., 2013). When normalized per gram of volatile solids, the protein and carbohydrate diets had similar methane production rates, indicating that differences in gas production from these two studies may have been due to water consumption and dilution of the manures during the protein trials. When methane production rates from both the field sampling efforts and the diet study are normalized per gram of volatile solids, diet study values were approximately 33-50% of values from field samples. This may be due to differences in manure age and development of microbial populations within deep pits as compared to the tanks used in this study.



(a) (b)

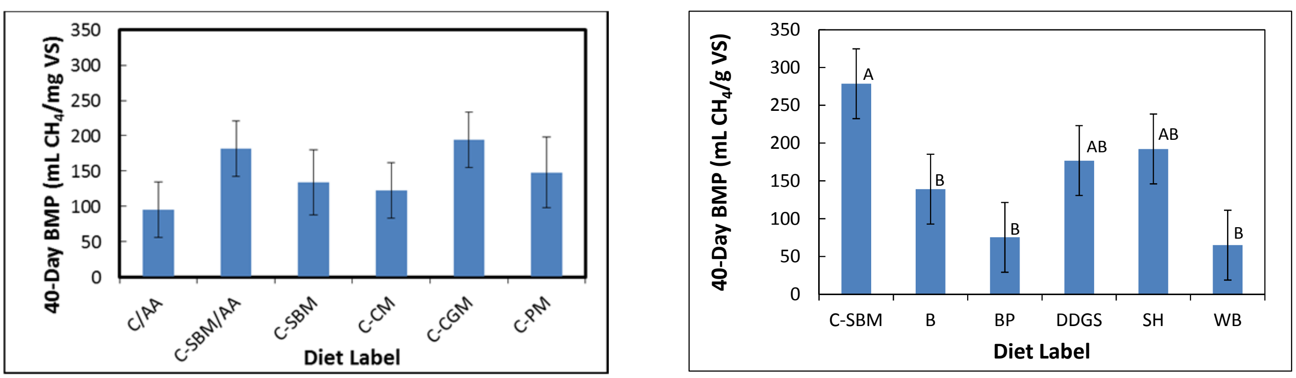
Figure 5. (a) Average methane production rates for protein diets and (b) carbohydrate diets with abbreviations from table 1. Error bars represent the standard error of the mean.



(a) **(b)**

Figure 6. (a) Average methane production rates for protein trials 1 and 2 (combined) and (b) carbohydrate trials 1 and 2 (combined) with abbreviations from table 1. One standard deviation is shown.

The results of the abbreviated biochemical methane production test (40 day incubation period as test is ongoing) proved to be an interesting comparison to the MPR rates discussed above. Average values with the standard error of the mean are shown in figure 7. The average values reported after 40 days of incubation are comparable, and often significantly higher than numbers reported by samples from deep pits studied by this group in a different study after a full incubation period (Van Weelden et al., 2013). This suggests that in relation to manure that has been stabilized in deep pits, the manure collected in these studies had a higher anaerobic biodegradability remaining. The results are also comparable to results from research done by King et al. (2011), who suggest that fresh swine manure should have an approximate methane production potential in the range of 250-270 mL CH_4/g VS. The protein study did not yield any significant differences between diets ($p = 0.5381$). On average the samples had a biochemical methane production potential of 144 mL CH4/g VS. On the other hand, the carbohydrate trial had significant differences between diets ($p = 0.0227$). The control diet, corn-soybean meal, had a significantly higher BMP values (279 mL CH_4/g VS) than barley (139 mL CH_4/g VS), beat pump (75 mL CH_4/g VS), and wheat bran (65 mL CH_4/g VS) diets.



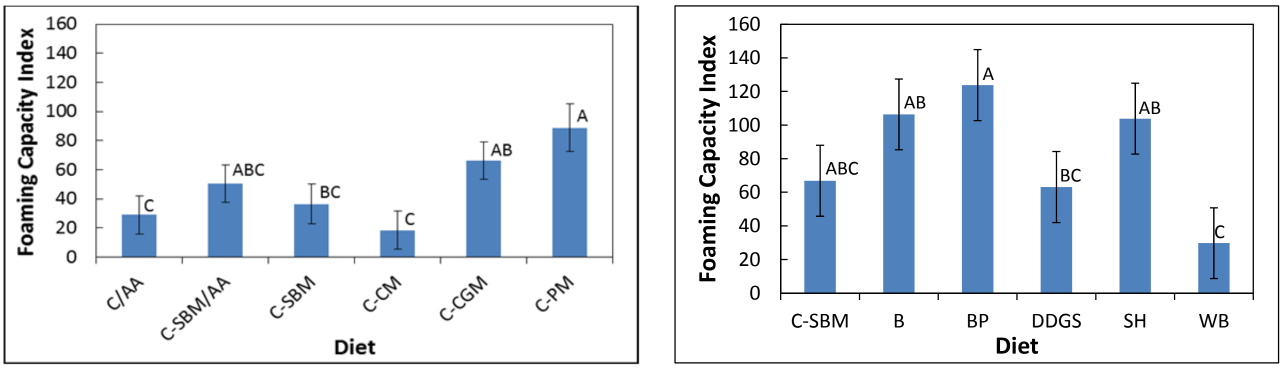
(a) **(b)**

Figure 7. (a) Average BMP for protein diets and (b) carbohydrate diets with abbreviations from table 1. Error bars represent the standard error of the mean. In (b), graph bars not connected by the same letter are significantly different at $\alpha = 0.05$.

In terms of gas phase, no single diet stood out as one that exhibited enhanced rates of biogas production. However, the variability in solids content of each trial seems to have had a direct impact on the gas production parameters. In this way, the relative magnitudes and high variability of the gas production values could be attributed to the comparatively variable amount of solids in the samples.

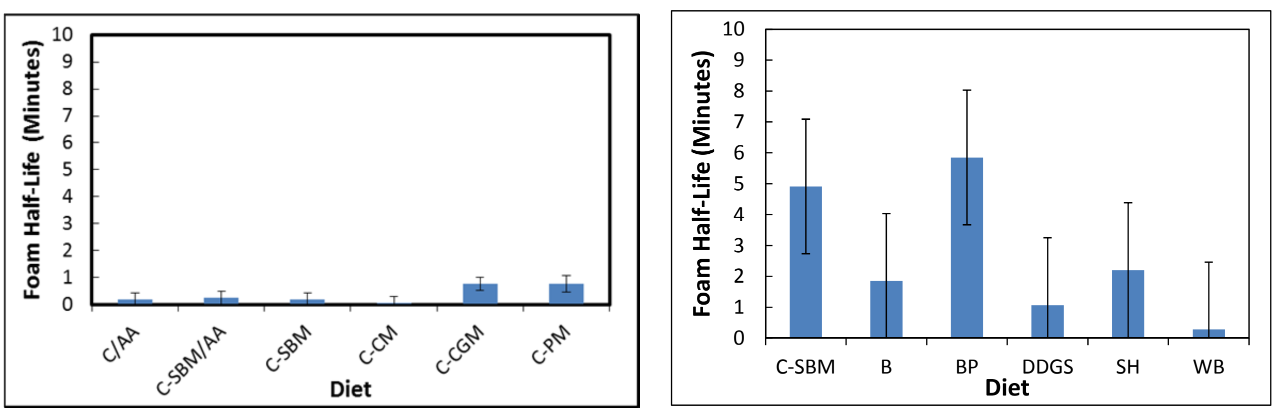
The results of the foaming capacity experiment are shown in figure 8 for both the protein and carbohydrate trials. Statistical analysis of the protein diets indicated significant differences in foaming capacity ($p = 0.0153$). The results indicated that the corn-poultry meal diet had a significantly greater foaming potential than the corn with amino acids, the corn-canola meal, or the corn-soybean meal diets, which showed minimal capacity to foam. The carbohydrate study indicated that diet again had a significant impact on the foaming capacity of the manure ($p = 0.0311$). The beat pulp diet had the highest average foaming capacity index, which was significantly higher ($\alpha = 0.05$) than the wheat bran diet. In the carbohydrate study, the beat pulp diet showed a significantly higher capacity to foam than the DDGS and wheat bran diets. In both studies, there are a number of results that showed different values between trial 1 and trial 2. One possible explanation for these differences has to do with the solids characteristics of samples, particularly the particle size. The samples with the largest particles (the thickest samples) often showed a lower capacity to foam, possibly due to the inhibiting effect of the large particles and their interaction with the liquid interface. For example, the samples collected from the wheat bran diet of the carbohydrate study were notably thicker than others, and showed the lowest statistical foaming capacity for that study.

The corresponding stability of foams is shown in figure 9. The protein study showed no significant differences in foam half-life ($p = 0.2078$) and all half lives were very short, indicating that no mechanism was present to stabilize the bulbbles despite some capacity to foam. Similarily, samples from the carbonhydrate study showed no significant differences between the diets. Some of the samples exhibited substancially longer foam half lifes than samples from the protein study, but there was great variability among values. In general, then, the carbohydrate study showed a greater ability to stabilize the foam, with a few samples exhibiting substantially higher stabilities than most others.



(a) (b)

Figure 8. (a) Average foaming capacity for protein trials 1 and 2 and (b) carbohydrate trials 1 and 2 with abbreviations from table 1. Error bars represent one standard error of the mean.



(a) (b)

Figure 9. Average half-life of foam for protein diets (a) and carbohydrate diets (b) with abbreviations from table 1. Error bars represent one standard error of the mean.

Narasin Studies

The average volativity of samples in the Narasin feeding trial was 73.1%, which is higher than the averages reported for either the carbohydrate or protein studies discussed previously. The distribution of samples with respect to total and volatile solids content is shown in figure 10. In this study, the overall solids contents were substantially higher than in either of the previous studies, and the results indicated that for every unit increase in total solids there was a 0.9 unit increase in volatile solids. This experiment was analyzed as a factorial experiment with factors of Narasin addition (yes or no) and diet (corn-soybean meal or DDGS). Results of the statistical analysis indicated that DDGS manures had significantly greater solids concentrations ($p < 0.0001$) than the corn-soybean meal manures, but that Narasin did not impact total solids content. The same results were found for volatile solids.

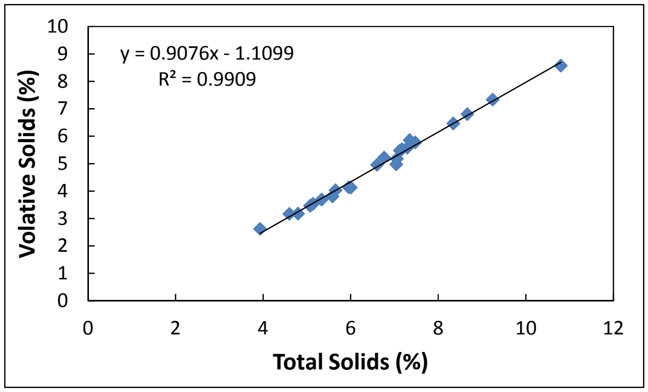


Figure 10. Volatile Solids vs. Total Solids for Narasin diet study.

Both the density and dynamic viscosity were measured on samples obtained from these diets (Density measured with Weight Per Gallon Cup, Gardco, Pompano Beach, Florida; Viscosity measured with DV-II+ Pro Viscometer, Brookfield Engineering Laboratories, Middleboro, Massachusetts). The results again indicated that DDGS and corn-soybean meal diets were different. Manures from pigs fed the DDGS diet were more dense (1.032±0.001 g/mL, mean ± standard error) than manures from pigs fed the corn-soybean meal diet (1.024±0.001 g/mL) at a p-value of 0.0011. In the case of viscosity, diet again had an impact; average viscosity was 314 ± 36 Pa-s for the DDGS diet as compared to 53.7 ± 28 Pa-s for the corn-soybean meal diet. In this way, the values of both density and viscosity increased with total solids.

Statistical analysis of the methane production rates indicated a significant diet effect (DDGS versus corn soybean, p = 0.0026), but no impact of Narasin (p = 0.8368). Results for each diet are summarized in figure 11. Interestingly, the samples collected from diets including Narasin dosages were not different than those that didn't, suggesting that the metabolized Narasin dosage had no effect on the ability for samples to produce methane. When normalized to methane production rate per gram of VS, the average result measured for the corn-soybean meal diet (0.095 ± 0.007 L CH_4 per g VS per day) was similar to those found in the carbohydrate and protein studies, but the value measured for the DDGS diet (0.033 ± 0.007 L CH_4 per g VS per day) was substantially lower.

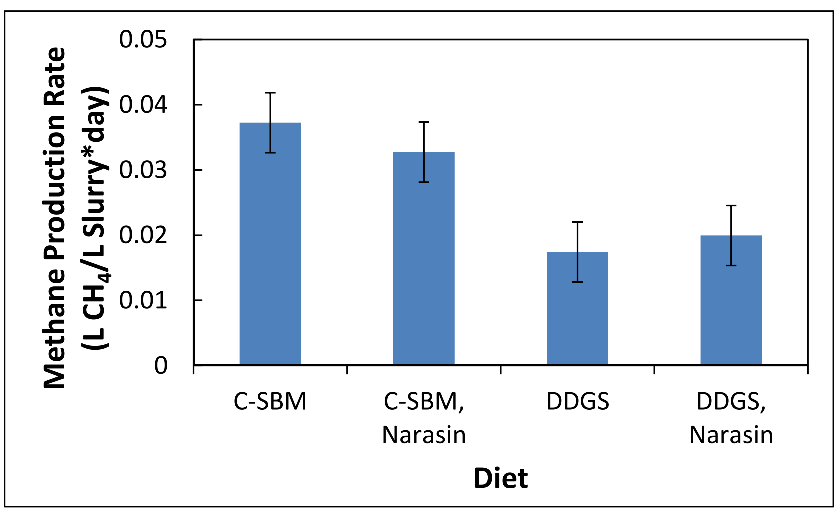


Figure 11. Average methane production rates for Narasin study with abbreviations from table 1. Error bars represent one standard error of the mean.

Analysis of the BMP values indicated that both Narasin (p = 0.0417) and the interaction of Narasin and diet (p = 0.0235) were significant, but that diet itself was not (p = 0.6931). Figure 12 shows the 40-day biochemical methane production rate of samples collected from the Narasin study. The corn-soybean meal with no Narasin addition showed the greatest potential to make additional methane, while the methane production potential of the samples collected from the corn-soybean diet including Narasin was significantly lower. At the same time, the corn-DDGS diets did not show statistical differences between samples collected from diets with Narasin and those that weren't. In this way, it is difficult to make any strong conclusions about the impact on Narasin addition to swine diet on the potential for reduced methane production.

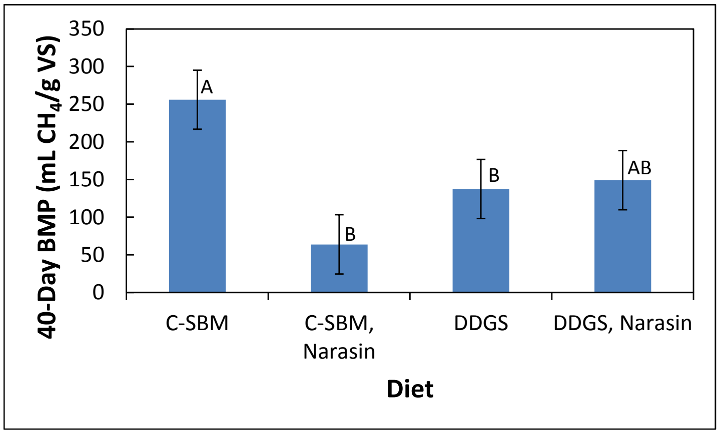
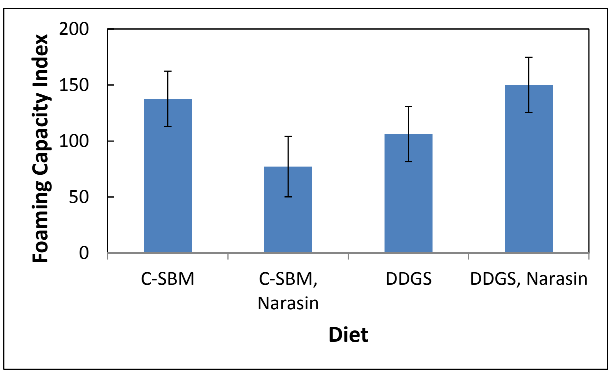
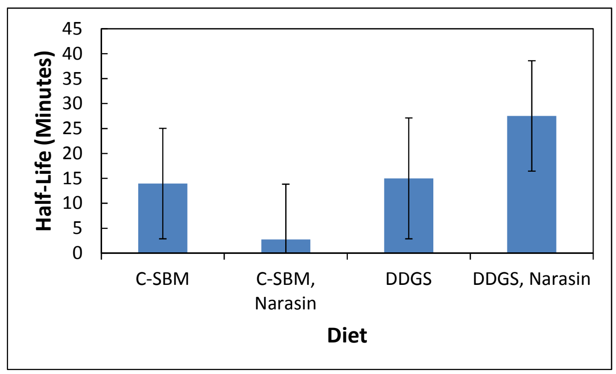


Figure 12. Average biochemical methane potential for the Narasin study with abbreviations from table 1. Error bars represent one standard error of the mean. Columns not connected with the same letter are significantly different at α = 0.05.

The foaming capacity and stability trends of samples from the Narasin study are shown in figure 13 (a) and (b), respectively. Results indicated that there were no differences in foaming capacity based on diet ($p = 0.4237$), Narasin addition ($p = 0.7453$), or the interaction of Narasin and diet ($p = 0.0539$). Similar results were seen for the foam stability values, with no significant differences for any of the factors ($p = 0.9546$, $p = 0.2697$, and $p = 0.3088$ for the impact of Narasin, diet, and their interaction, respectively). The averages shown for foaming capacity and foam stability show a similar trend, where samples with a greater capacity to foam also had a greater capacity to stabilize foam.,



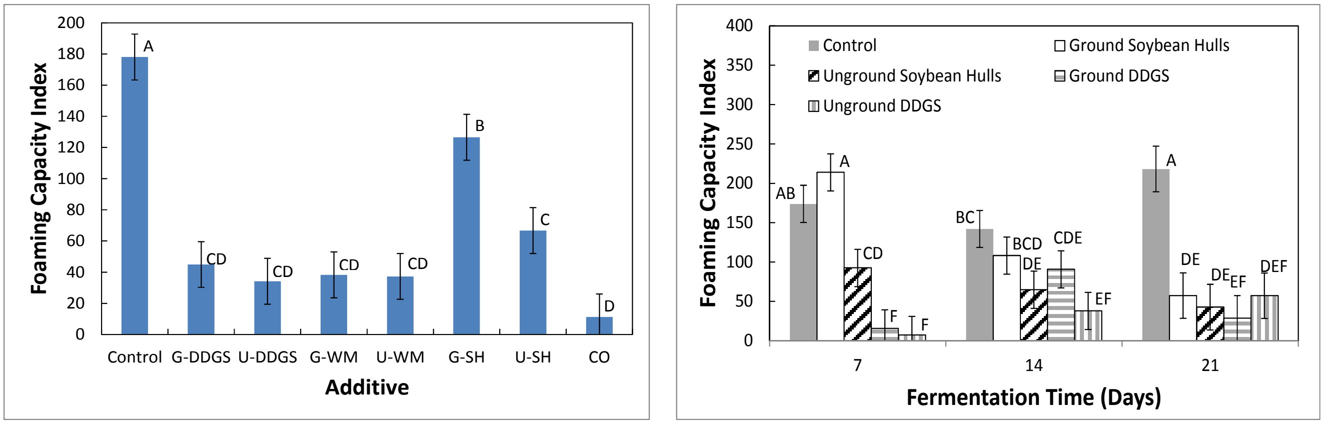


(a) **(b)**

Figure 13. Average foaming capacity (a) and foam stability (b) of the diets used in the Narasin study with abbreviations from table 1. Error bars represent one standard error of the mean.

Bucket Additive Study

The manure collected from dietary studies did not accumulate foam layers during storage in the collection tanks (foam could be generated upon aeration, but generally showed only limited stability). One explanation for the lack of foam production has to do with the relatively low methane production rates measured by volume. In an effort to observe foam production without external aeration, differing carbohydrate sources and particle sizes were added directly to the manure to provide readily available carbon. The results of the subsequent foaming capacity tests indicated that incubation length was not significant ($p = 0.5779$), but that both the additive ($p < 0.0001$), and incubation length by additive interaction were significant ($p = 0.0179$). In this case, the interaction was mostly due to the soybean additive (ground form) which showed a decreasing foaming capacity with time. Also, the DDGS additives showed greater foaming capacities in the second and third weeks of incubation. On the other hand, the control samples and other additives maintained relatively consistent foaming capacities throughout. This data is summarized in two ways: first the overall means for foaming capacity are provided in figure 14 (a); however, due to the interaction, the impact of incubation time on foaming capacity is shown for several select additives in figure 14 (b). The main effects indicated that the control (no additive) had the largest foaming capacity, with both soybean hull treatments showing the next highest foaming capacity averages. The interaction by time graph indicates that ground soybean hulls initially had the highest foaming capacity, but that the capacity decreased with time. A contrast statement testing differences between ground and unground additions indicated no difference in foaming capacity ($p = 0.2765$).



(a) **(b)**

Figure 14. Impact of carbohydrate source and particle size on (a) foaming capacity and (b) the interaction of carbohydrate source and particle size relative to incubation time with abbreviations from table 2. Error bars represent the standard error of the mean. Columns not connected with the same letter are different at α = 0.05.

An analysis of foam stability indicated that incubation length ($p = 0.0004$), additive ($p < 0.0001$), and the incubation length by additive interaction ($p < 0.0001$) were all significant. Foam stability was the greatest in the control manure on day 21 with a half-life of 76 minutes. The next highest stability was shown by manure with soybean additives at day 7, which had half-lives of 35 and 24 minutes for the ground and unground additives, respectively. All other incubation lengths by additive combinations had similar half-lives of 0-4 minutes.

The differences in the behavior of the control mixture, the soybean hull mixture, and other samples provide interesting evidence with respect to a foaming mechanism. In particular, the efficiency of fermentation of the different additives, or lack thereof, may have directly affected the foaming characteristics of the sample. In the case of the soybean hull mixture, the initial spike of the foaming capacity followed by a gradual decline by week may reflect the relatively high cellulose composition of soybean hulls, which are more readily digested by the microbial population. In fact, this elevated microbial response to the additive addition could have led to the accumulation of some "biosurfactant" as described by Ganidi et al. (2009). This hypothesis may also support the increasing foaming trend in the control diet as the weeks progressed if the control diet was less quickly utilized by the bacteria in the bucket.

Conclusion

The diet trials conducted in this study sought to better understand the impact of different feed mixes on both the physical and biochemical characteristics of swine manures. The two main studies employed various protein and carbohydrate based diets as fixed factors in multiple trials. Many of the key parameters observed in both studies varied significantly for the protein and carbohydrate studies, but may have been affected by the dissimilar solids composition of manures from each study. Within each study, no single diet stood out as one that exhibited both enhanced biogas production and foaming capacity. However, it was observed that in general, manure tested in this study exhibited lower MPR values and higher BMP values when compared to manure collected from deep pit storages from various facilities in Central Iowa that have been studied by this group.

A subsequent trial observing the effects of Narasin on the same key parameters also did not yield any statistically significant results in terms of a single diet that produced more biogas or foaming capacity. In addition, it was not shown definitively that Narasin played a significant role in the inhibition of biogas production of samples. However, the solids content of samples were relatively variable based on the results of the feeding trials, which may have played a large factor in the values obtained for several of the key parameters in this study.

A final study observed the effects of diet-based additives on the foaming characteristics of samples. One additive (soybean hulls) showed enhanced foaming characteristics after one week of fermentation with a decreasing trend in the weeks to follow. On the other hand, the control samples with no additive showed an increasing trend in both foaming capacity and stability. A possible explanation for these phenomena includes an accumulation of a "biosurfactant" due to microbial activity in these systems.

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