

Title: Evaluation of crude glycerin in swine - **NPB Project #07-165**

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Industry Summary

The objective of the proposed research was to determine the variation in metabolizable energy (ME) content of crude glycerin samples from a several biodiesel production facilities using different feedstock sources (soybean oil, animal fat, and used restaurant grease). Nursery pigs initially weighing 10.4 kg were fed dietary treatments consisting of a basal diet, or diets containing crude glycerin from various biodiesel production facilities supplemented in the diet at approximately 9.1%. Using typical energy balance techniques, each diet was fed twice daily to pigs in individual metabolism crates for a 6-d adjustment period after which a 4-d energy balance trial was conducted. Metabolizable energy for each diet was calculated by subtracting the fecal and urinary energy excretion of each pig from the gross energy consumed, with the ME value of each crude glycerol estimated by the difference between the test and the basal diet. The gross energy (GE) of the crude glycerin samples ranged from 3,173 to 6,021 kcal/kg while the determined ME ranged from 2,535 to 5,206 kcal/kg, each being a reflection of the concentration of glycerol, methanol, and free fatty acids in the crude glycerin. Prediction of each crude glycerin's ME based upon the crude glycerin's composition was poor. In contrast, the prediction of each crude glycerin's GE based upon the crude glycerin's composition high [GE kcal/kg as-is basis = $-306 + (46.65 \times \% \text{ glycerin}) + (54.31 \times \% \text{ methanol}) + (101.83 \times \% \text{ fatty acids})$]. Because the relationship between GE and ME was similar between all crude glycerin samples, averaging 85.4%, ME could be accurately predicted based upon the predicted GE and the average ME:GE of 85.4%. Overall, data presented herein show that the concentration of glycerin, fatty acids, and methanol affect the GE and ME of crude glycerin, and because crude glycerin is easily digested and metabolized, it can be used as a viable source of energy in growing pigs. These data also suggest that the amount of ash and methanol had little to no effect on ME utilization. However, the salt concentration of crude glycerin needs to be accounted for in feed formulation and level of methanol needs to be considered for regulatory reasons.

Abstract

Apparent DE and ME of various crude glycerins from different biodiesel production facilities were empirically determined in nursery pigs (10.4 kg initial BW) in order to predict their DE and ME based on crude glycerin

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composition. Dietary treatments consisted of a basal diet, or diets containing crude glycerin from various biodiesel production facilities supplemented in the diet at approximately 9.1%. Because of bulk density differences, two glycerin sources were supplemented at either 7.7 or 6.9%. In addition, soybean oil and lard were included at 6.7% as dietary treatments to serve as positive controls. Each diet was fed twice daily to pigs in individual metabolism crates. After a 6-d adjustment period, a 4-d balance trial was conducted. During the collection period, feces and urine were collected daily and stored at 0°C until analysis. The GE of each test ingredient, diet, and urine and fecal samples from each pig were determined by isoperibol bomb calorimetry. Digestible energy of the diet was calculated by subtracting fecal energy from the GE in the feed, whereas ME was calculated by subtracting the urinary energy from DE. The DE and ME values of crude glycerol were estimated by difference where the DE and ME content of the basal diet was subtracted from the complete diet containing the test ingredient. Gross energy, DE, and ME of USP-grade glycerin was calculated to be 4,325, 4,443, and 3,664 kcal/kg, respectively. In contrast, GE of the crude glycerin samples ranged from 3,173 to 6,021, DE from 3,022 to 5,228, and ME from 2,535 to 5,206 kcal/kg, reflecting the content of glycerol, methanol, and free fatty acids in the crude glycerin. The GE, DE, and ME of soybean oil and lard was determined to be 9,443, 8,567, 8,469, and 9,424, 8,543, and 8,639 kcal/kg, respectively. The stepwise regression prediction of the ME in crude glycerin exhibited R^2 of only 0.42, whereas prediction of GE achieved a R^2 of 0.99 [GE kcal/kg as-is basis = $-306 + (46.65 \times \% \text{ glycerin}) + (54.31 \times \% \text{ methanol}) + (101.83 \times \% \text{ fatty acids})$]. On average, the ME of crude glycerin was 85.4% of its GE (SD 17.2) and did not differ by glycerin source. Data provided in these experiments indicate that crude glycerin is a valuable energy source with its GE concentration dependent upon the concentration of glycerin, methanol, and fatty acids, and its ME as a percent of GE being constant, at 85.4%.

Introduction

Biodiesel can be produced by a variety of esterification technologies using vegetable oil, animal fat, or yellow grease as the initial feedstock (Ma and Hanna, 1999; Van Gerpen, 2005; Thompson and He, 2006). Production of biodiesel from oils other than soybean oil has increased due to the recent increase in soybean oil price. With 0.3 kg of crude glycerin generated for every gallon of biodiesel produced, the potential crude glycerin generated from biodiesel production is large (National Biodiesel Board, 2007). Glycerin is readily absorbed by the gastrointestinal tract (Tao et al., 1983) and the metabolism effect of glycerol has been reviewed previously (Lin, 1977; Brisson et al., 2001). Only limited research has evaluated the ME value of glycerin in monogastrics (Bartelt and Schneider, 2002; Dozier et al., 2008, Lammers et al. 2008ac). In swine, Bartelt and Schneider (2002) utilized pure glycerin in diets containing up to 15% glycerin and reported an average ME of 3,525 kcal/kg. Recently, Lammers et al. (2008a) reported that a high quality crude glycerin (87% glycerin) obtained from a biodiesel facility using soybean oil as the initial feedstock contained 3,207 kcal/kg when fed to starting and finishing pigs, a value which is only slightly higher than the value obtained by Bartelt and Schneider (2002) on an equivalent glycerin basis. Because it was expected that variation in crude glycerin between biodiesel production facilities would vary depending upon sources of feedstock and production efficiencies, a balance experiment was designed to determine the ME concentration of various crude glycerin samples based on different production facilities and different feedstock sources. From this data, an equation estimating the ME concentration was generated based upon the composition of the crude glycerin. A second growth experiment was conducted to evaluate the ability of growing pigs to gain efficiently (gain/Mcal ME) on these diets, regardless of glycerin source.

Objectives

There objective of the proposed research is to determine the variation in metabolizable energy (ME) content of crude glycerin samples from a range of plants and feedstuff sources (soybean oil, animal fat, and used restaurant grease). Only recently have we published data on the ME content of one crude glycerin sample, but there is no data on the variation in crude glycerin from different sources and its impact on ME values in growing pigs. Thus, data is critically needed to understand the impact of crude glycerin's variability on the estimation of its ME in growing pig diets.

Materials & Methods

General Pig Management

The Iowa State University Animal Care and Use Committee approved all experimental protocols, #12-07-6479-S. Crude glycerin was obtained from various biodiesel plants utilizing either soybean oil (both hexane and extruded soybean processing), tallow, yellow grease, or poultry fat. The crude glycerol was characterized courtesy of Ag Processing Inc. (Omaha, NE) using standard techniques employed at biodiesel plants as described previously (Lammers et al., 2008b). The composition of each crude glycerin used in Exp. 1 and 2 is detailed in Table 1.

Dietary treatments consisted of a common basal diet, which met or exceeded the NRC (1998) requirements (Table 2); or diets containing 9.09% crude glycerin (USP = USP-grade glycerin; AGP-IA = Ag Processing Inc., Sergeant Bluff, IA; REG-R = Renewable Energy Group, Ralston, IA; REG-WL = Renewable Energy Group, Wall Lake, IA; REG-MN = Renewable Energy Group, Albert Lea, MN; ADM-MO = Archer Daniels Midland, Mexico, MO; WW-OH = Westway Feed Products, Cincinnati, OH; WW-TX = Westway Feed Products, Houston, TX; IW-AC = Imperial Western Products-acidulated glycerin, Coachella, CA); 7.72% crude glycerin (IW-NA = Imperial Western Products-nonacidulated glycerin, Coachella, CA); or 6.91% crude glycerin (USB-GA = US Biofuels, Rome, GA). The differences in addition of some products were due to their bulk density and our ability to liquefy the product prior to mixer addition because both IW-NA and USB-GA were solid at room temperature. Products obtained were either from hexane-derived soybean oil (AGP-IA, REG-WL, REG-MN, ADM-MO, and WW-TX), expeller derived soybean oil (REG-R), tallow (WW-OH), yellow grease (IW-AC and IW-NA), or poultry fat (USB-GA). We also chose to obtain a product from one plant prior to acidulation (IW-NA) which had an elevated free fatty acid content. In addition, the sample from a plant using poultry fat (USB-GA) also contained a high level of fatty acids. We also included the evaluation of soybean oil or lard, 6.69% of the diet, which were used as ‘high-energy’ controls, in order to validate our methodology as outlined by Adeola (2001). We chose moderate inclusion levels for all products used as there was some concern about the effect of glycerin inclusion level on ME determination (Bartelt and Schneider, 2002; Lammers et al., 2008a) and on feed flowability (Cerrate et al., 2006).

Experiment 1

In Exp. 1, three groups of 56 barrows weighing (average initial BW 10.4 kg, Cambrough 22 females × L337 sires) were randomly assigned to individual metabolism crates (0.53 × 0.71 m) equipped with screens and trays that allowed for total but separate collection of feces and urine. Pigs were randomly assigned to dietary treatments after pen assignment. Pigs were offered 2,000 g of feed per d, distributed in two equal daily meals (Table 3) with feed consumption and refusal recorded at the end of the experimental period. Water was available from a nipple waterer at all times. A 6-d adjustment period was used to adapt the pigs to the metabolism crate and the dietary treatment prior to the 4-d total fecal and urine collection period. During the collection period, urine was collected once daily into plastic buckets containing 25 mL of 6 N HCl, and stored at 0°C until the end of the collection period. At the end of the collection period, urine was thawed, weighed, and a subsample was collected and stored at 0°C until subsequent analysis. Feces were also collected daily and stored at 0°C.

Experiment 2

In Exp. 2, two groups of 56 pigs (8.7 kg initial BW; Cambrough 22 females × L337 sires), representing 4 replicates of gilts and 4 replicates of barrows, were randomly assigned to individual pens (0.7 × 1.22 m), and then to dietary treatments for a 25 or 18 d (group 1 and 2 respectively) performance study. Feed and water were offered ad libitum at all times and pigs and feeders were weighed at the beginning and end of the experiment to determine ADG, ADFI, and G:F ratio.

Chemical Analyses

Feed samples were ground through a 1-mm screen before energy determination. Fecal samples were thawed, dried at 70°C for 48 h, and weighed to determine the DM content. Fecal samples were ground through a 1-mm screen in preparation for energy determination. For urine energy determination, 2 mL of urine was added to 0.5 g of dried cellulose and subsequently dried at 50°C for 24 h before energy determination. The GE of feed, feces, and urine plus cellulose was determined using an isoperibol bomb calorimeter (model number 1281, Parr Instrument Co., Moline, IL), with benzoic acid used as a standard. Duplicate analyses were performed on all

diets and fecal samples from each pig, whereas triplicate analyses were performed on urine plus cellulose from each pig. Urinary energy was determined by subtracting the energy contained in cellulose from the combined urine plus cellulose energy.

Calculations and Statistical Analysis

Gross energy intake was calculated by multiplying the GE value of the diet fed by feed intake over the 4-d collection period. Apparent DE values were calculated by subtracting fecal energy from intake energy and apparent ME values calculated by subtracting urinary energy from apparent DE. The apparent DE and ME values of the test ingredient fed to the pigs was estimated by difference from the basal diet as described by Adeola (2001). Observations from 151 of the 168 pigs assigned to dietary treatments in Exp. 1 were used for analysis. Observations from 17 pigs were not possible to quantify due to diarrhea, constipation, or feed refusal, such that any data obtained from these pigs were considered outliers. All 112 pigs were used for analysis in Exp. 2. Using the individual pig as the experimental unit, data from each experiment were subjected to ANOVA with group and treatment in the model (SAS Inst. Inc., Cary, NC), with differences between means tested using the PDIFF option. In addition, a stepwise regression model was used to equate the effect of glycerin composition on apparent ME in Exp. 1, with variables having *P* values < 0.15 maintained in the model.

Results & Discussion

Compositional variation in crude glycerin was expected (Table 1). Glycerin content in soybean oil-based glycerin sources were relatively consistent, averaging 83.9%. For crude glycerin obtained from tallow, WW-OH, glycerin content was lower than other crude glycerin sources while water content moved in an opposite manner. Products with high free fatty acid content, IW-NA and USB-GA, had lower glycerin contents. As expected, acidulation increased glycerin content at the expense of fatty acid content (IW-AC vs. IW-NA). Crude glycerin is a viscous liquid and all samples were viscous except for WW-OH which contained high water content, and for IW-NA and USB-GA which were solid at room temperature due their high free fatty acid content. In general, all samples contained low amounts of free fatty acids which are indicative of efficient fatty acid esterification at the biodiesel production facility. Only two samples, IW-NA and USB-GA, had elevated levels of fatty acids. This was expected for the IW-NA sample as it was not acidulated, whereby acidulation is the process in which the soap stock is acidified reducing its emulsifying properties such that there is a reduction in the amount of fatty acids remaining in the crude glycerin (Ma and Hanna, 1999; Van Gerpen, 2005; Thompson and He, 2006). We can not explain why the USB-GA sample contained an elevated level of fatty acids. Likewise, methanol levels were relatively low in all samples, except for the IW-NA and USB-GA samples which contained 3.49 and 14.99% methanol, respectively. Recovery of methanol by biodiesel plants also relates to their production efficiency as methanol recovered is reused in the biodiesel process (Ma and Hanna, 1999; Van Gerpen, 2005; Thompson and He, 2006). Because the IW-NA was not a final product from this location, the high methanol level was not unexpected, as the final product which is acidulated, IW-AC, contained a low level of methanol. Aside from the cost of methanol for the biodiesel production facility, methanol levels are a concern in crude glycerin fed to livestock as described in detail by Lammers et al. (2008ab). All but USP, WW-OH, IW-AC, and USB-GA contained moderate amounts of ash which in most cases was NaCl. We expect that the three products with high ash, but low NaCl (WW-OH, IW-AC, and USB-GA) were from biodiesel facilities utilizing a K-based instead of a Na-based catalyst. If high levels of crude glycerin were to be supplemented in the diet, the level of NaCl or KCl should be considered and balanced accordingly (Lammers et al. 2008b).

In Exp. 1, the removal of 17 pigs for various reasons was unexpected as the herd health at the Iowa State University swine farm was normal during both experiments (Exp. 1, January through March; Exp. 2, March through April). It was noted, however, that several pigs exhibited a fair amount of looseness or feed refusal in Exp. 1, but this could not be attributed to any particular diet. One could speculate this to not using any antibiotics in the basal diet (Table 2), but pigs in Exp. 2 did not exhibit this same condition and were fed the same diets. In addition, in past experiments we have not used an antibiotic in the basal diet and have not experienced these problems (Lammers et al., 2008a). During Exp. 1, it was noted, however, that pigs were noticeably excitable when moved to the metabolism crates and during the entire experiment. This occurred even though each cage allows visual access to other pigs, a pen 'toy' is included in each crate, and pigs were

managed appropriately during the trial. We have no reason for the excitability of pigs used in Exp. 1, but this may have contributed to the looseness of stools in some of the pigs.

Caloric values of the various test ingredients are shown in Table 4. For comparative purposes, the ME determined for the basal diet was 3,352 kcal/kg which is similar to the calculated value of 3,325 kcal/kg based upon the NRC (1998). Similarly, the DE and ME determined for soybean oil (8,567 and 8,469 kcal/kg, respectively) and lard (8,524 and 8,639 kcal/kg) are close to that reported for soybean oil (8,750 and 8,400 kcal/kg, respectively) and lard (8,285 and 7,950 kcal/kg) as reported in the NRC (1998) where DE was predicted from free fatty acid concentration and the unsaturated:saturated ratio, and ME predicted as 96% of the DE (Powles et al., 1995). In Exp.1, ME as a percent of DE for soybean oil and lard averaged 99.85% which is similar to that reported by Powles et al. (1995). All of the above comparisons provided confidence in our balance techniques and the use of the difference method (Adeola, 2001) for determination of DE and ME in these products.

Digestible energy as a percent of GE varied between sources ($P < 0.01$) averaging 96.6% across all crude glycerin sources. This is higher than found previously (92%, Lammers et al., 2008a), but similar to the $> 97\%$ obtained for pure glycerin digested prior to the cecum reported by Bartelt and Schneider (2002). The observations where DE as percent of GE was greater than 100% is not logical, but we speculate this may be due to the inherent variation associated with metabolism trials which was 17% for this parameter in the current study.

The glycerin products containing low free fatty acid levels had a lower ME as a percent of DE ($P < 0.05$) compared to the two samples with high amounts of fatty acids, averaging 86.0 and 101.3%, respectively. Because ME as a percent of DE was basically similar for all low-fatty acid crude glycerin products (averaging 86.0%), albeit lower than the 96% reported by Lammers et al. (2008a), we have elected not to discuss actual DE values, but will focus on the actual ME values and on ME as a percent of GE. It is worthy to note, however, that in the two samples with high amounts of fatty acids (IW-NA and UAB-GA), ME as a percent of DE were the same as that determined for soybean oil and lard, and similar to the 96% reported for fats and oils (Powles et al., 1995).

There was no difference in ME as a percent of GE across treatments ($P = 0.19$). The average for all crude glycerin products was 85.4% (Table 4) which compares favorably with the 88.4% reported for crude glycerin reported by Lammers et al. (2008a). Consequently, with differences in the GE between crude glycerin products, it is only logical to assume that actual ME values would differ. Soybean oil and lard had the highest actual ME, followed by the two products with high free fatty acid content (IW-NA and USB-GA), followed by all the remaining crude glycerin products. There did not appear to be any difference in ME relative to feedstock source used at the biodiesel plant, although the crude glycerin obtained from the biodiesel plant using tallow as their feedstock (WW-OH) had a lower ME which can be attributed to its composition, not source. We were somewhat surprised with the 2,535 kcal ME/kg determined for AGP-IA which appeared to be low given that we had previously obtained a sample from this same plant and determined a ME value of 3,207 kcal/kg (Lammers et al., 2008a). However, it should be noted that the composition of samples did vary [glycerin (83.58 vs. 86.95%), water (13.40 vs. 9.22%), methanol (0.1137 vs. 0.028%), free fatty acid (0.07 vs. 0.29%), ash (2.93 vs. 3.19%) and GE (3,601 vs. 3,625 kcal/kg), current vs. Lammers et al., 2008a, respectively] which would likely affect the caloric values of the product.

With differences in crude glycerin composition likely driving the ME determination, we chose to evaluate whether the composition of crude glycerin could predict the ME values experimentally determined in Exp. 1. Using stepwise regression to characterize the relationship of glycerin, water, methanol, fatty acids, and ash content in the crude glycerin to ME yielded the equation: ME, kcal/kg = $3,580 - (40.52 \times \% \text{ water}) + (48.55 \times \% \text{ fatty acids})$, ($R^2 = 0.42$, $P < 0.01$). Because water and glycerin appear to be inversely related in crude glycerin, removing water from the list of variables subsequently yielded the equation: ME, kcal/kg = $464 + (31.54 \times \% \text{ glycerin}) + (87.78 \times \% \text{ fatty acids})$ ($R^2 = 0.41$; $P < 0.01$). Because the amount of fatty acids appear to be a large driver of the ME in crude glycerin, elimination of the IW-NA and USB-GA samples from the analysis (each exhibited higher levels of fatty acids than the other crude glycerin products) did not improve the ability of estimating ME from the composition of crude glycerin. Because of these relatively low R^2 estimates, we chose to evaluate the value of crude glycerin in a different manner. Estimation of GE from the composition

of crude glycerin by regression was extremely accurate where: $GE \text{ kcal/kg} = -306 + (46.65 \times \% \text{ glycerin}) + (54.31 \times \% \text{ methanol}) + (101.83 \times \% \text{ fatty acids})$ ($R^2 = 0.998$, $P < 0.01$). Because there was no difference in ME as a percent of GE ($P = 0.19$, Table 4), application of the average ME as a percent of GE (85.4%) to the predicted GE could then be used to predict ME. Likewise, one could also apply conversion estimates obtained from complete diets (Noblet and Perez, 1993) to estimate dietary DE, ME, or NE. Applying the prediction equation for GE and ME to GE conversion of 85.4% on the data of Lammers et al. (2008a) results in a GE and ME prediction of 3,781 and 3,229 kcal/kg, respectively, which compares favorable with the actual values of 3,625 and 3,207 kcal/kg, respectively.

A biological approach to calculating the caloric value of crude glycerin could be to estimate the GE of crude glycerin based upon the GE for glycerin (4,325 kcal/kg, Table 4), oil (9,443 kcal/kg, Table 4), and methanol (5,425 kcal/kg, Bossel, 2003). Using these values and the composition of crude glycerin listed in Table 1, the predicted GE concentrations would be 4,310, 3,628, 3,657, 3,682, 3,702, 3,633, 3,270, 3,549, 4,150, 5,881, and 5,516 kcal/kg for USP, AGP, REG-R, REG-WL, REG-AL, ADM-MO, WW-OH, WW-TX, IW-AC, IW-NA, and USB-GA, respectively. These values compare favorably with the analyzed GE values shown in Table 4. Not surprisingly, the slope values obtained by stepwise regression are relatively close to the biological GE estimates for glycerin (4,665 vs. 4,325, respectively), methanol (5,431 vs. 5,425, respectively), and oil (10,183 vs. 9,443, respectively). As indicted above, one could then apply the average ME as a percent to GE (85.4%) to this data to predict ME. Applying this approach to the data of Lammers et al. (2008a) results in a GE and ME of 3,790 and 3,237 kcal/kg, respectively, which compares favorable with the actual values of 3,625 and 3,207 kcal/kg, respectively, or the regression GE and ME estimates of 3,781 and 3,229 kcal/kg, respectively.

Performance of nursery pigs fed these same diets is presented in Table 5 (Exp. 2). There was no difference in ADG ($P = 0.25$) or ADFI ($P = 0.34$) due to any source of crude glycerin, soybean oil, or lard added to the basal diet. There were differences in G:F ratio ($P = 0.01$), but due to differences in the ME determined for each complete diet (Exp. 1, data not presented), we felt it was more appropriate to represent gain per Mcal intake. Expressed in this manner, pigs fed the control diet had a greater gain:Mcal ME intake than pigs fed any other diet ($P = 0.01$), with no differences between pigs fed any other diet. Differences in G:F ratios were not expected as past (Mourot et al., 1994; Kijora et al., 1995, 1997; Kijora and Kupsch, 2006) and recent studies (Grosebeck et al., 2008; Hansen et al., 2008; Lammers et al., 2008b) have reported similar G:F between pigs fed diets with or without crude glycerin. Only one data set has reported a small decline in G:F ratio with crude glycerin supplementation (Casa et al., 2008). The reduced G:F ratio in pigs fed all but the control diet could be a function of dilution of nutrients in the diet as in these experiments, the test ingredient was added on top of the basal diet. For pigs fed crude glycerin, the 9% dilution would have lowered the true ileal digestible Lys to approximately 1.05%, although this still should have been adequate according to the NRC (1998). Although soybean oil and lard were added at a lower rate, because oils and fats have 2.25 times the energy of starch (and basically glycerin, Table 4), one should have expected that the change in nutrient:energy ratio would have had a greater effect on gain:Mcal ME in the diets containing more ME (IW-NA, USB-GA, soybean oil, and lard), but this was not the case. Consequently, we do not have a clear explanation for these data.

Methanol has warranted special consideration in the use of crude glycerin as it is not completely removed at the biodiesel production facility. Methanol is a potentially toxic compound and can elicit a variety of acute and chronic symptoms (Roe, 1982; Medinsky and Dorman, 1995; Skrzydlewska, 2003). Even though gastrointestinal disturbances are one of the chronic exposure symptoms to methanol, there was no apparent effect of dietary methanol on pigs fed IW-NA (3.49% methanol) or USB-GA (14.99% methanol) because no more pigs were removed, relative energy utilization indices (i.e., ME as a percent of GE) did not differ, and G:F was not reduced compared to pigs fed other glycerin sources. In the US, there is no Generally Recognized As Safe regulation or American Association of Feed Control Officials definition listing specifications for crude glycerin use in animal feeds such that specifications for pure glycerin defined under United States Pharmacopeia (USP) and Food and Chemical Codex (FCC) specifications are used for FDA guidance. Because methanol levels are not specifically listed in the USP or FCC specifications, the US FDA has decided to address free methanol levels under CFR573.640, regulation 21, requiring that levels of methanol in methyl esters of higher fatty acids should not exceed 0.015% or a level shown to be safe for use in animal diets. In addition to

the current experiment, it is noteworthy that Lammers et al. (2008b) did not report any increased incidence in frequency of lesions associative with methanol toxicity in eye, kidney, or liver tissue in growing-finishing pigs fed 5 or 10% crude glycerin containing 0.32% methanol.

Overall, data presented herein show that the concentration of glycerin, fatty acids, and methanol affect the GE of crude glycerin, and because crude glycerin is easily digested and metabolized, it can be used as a viable source of energy in growing pigs. In addition, these data also suggest that the amount of ash and methanol has little to no effect on ME utilization. However, the ME and salt concentration of crude glycerin needs to be accounted for in feed formulation and level of methanol needs to be considered for regulatory reasons.

Table 1. Chemical analysis of crude glycerin samples¹

<u>Sample</u> ²	<u>Source</u> ³	<u>Glycerin</u>	<u>Moisture</u>	<u>Methanol</u>	<u>pH</u>	<u>NaCl</u>	<u>Ash</u>	<u>Fatty acids</u>
USP	-	99.62	0.348	ND	5.99	0.013	0.01	0.02
AGP-IA	SB	83.58	13.397	0.1137	5.53	2.838	2.93	0.07
REG-R	SB	84.06	9.363	0.0072	5.82	6.346	6.45	0.22
REG-WL	SB	84.48	9.201	0.0309	5.73	6.000	5.90	0.28
REG-AL	SB	85.56	8.347	0.0260	6.34	6.065	5.87	ND
ADM-MO	SB	83.72	10.161	0.0059	6.30	5.997	5.83	0.12
WW-OH	TA	75.49	24.367	0.0290	3.99	0.073	1.91	0.04
WW-TX	SB	81.89	11.406	0.1209	6.59	6.577	7.12	0.01
IW-AC	YG	95.57	4.071	0.0406	6.10	0.162	1.93	0.15
IW-NA	YG	55.53	4.157	3.4938	8.56	1.977	4.72	34.84
USB-GA	PF	55.73	4.989	14.9875	9.28	0.011	4.20	24.28

¹ Samples analyzed as described in Lammers et al. (2008b) courtesy of by Ag Processing Inc., Omaha, NE, 68154. All values are percents. ND = not detected.

² USP=USP grade glycerin; AGP-IA=Ag Processing Inc., Sergeant Bluff, IA; REG-R=Renewable Energy Group, Ralston, IA; REG-WL=Renewable Energy Group, Wall Lake, IA; REW-MN=Renewable Energy Group, Albert Lea, MN; ADM-MO=Archer Daniels Midland, Mexico, MO; WW-OH=Westway Feed Products, Cincinnati, OH; WW-TX=Westway Feed Products, Houston, TX; IW-AC=Imperial Western Products-acidulated glycerin, Coachella, CA; IW-NA=Imperial Western Products-nonacidulated glycerin, Coachella, CA; USB-GA=US Biofuels, Rome, GA.

³ SB = soybean oil from hexane soybean crush plant except for REG-R where the soybean oil was obtained from extruded soybeans, TA = tallow, YG = yellow grease, PF = poultry fat.

Table 2. Composition of basal diet, as-is basis

<u>Ingredient</u>	<u>%</u>
Corn	57.50
Soybean meal	24.50
Dried whey	10.00
Fish meal, select	5.00
Plasma, appetin ¹	1.25
Soybean oil	0.15
Defluoriated phosphate	0.61
Limestone	0.14
Sodium chloride	0.35
Vitamin mix ²	0.30
Trace mineral mix ³	0.20
DL-methionine	0.005
<u>Calculated composition, %</u>	
ME, kcal/kg	3,325
CP	21.00
True ileal digestible Lys	1.15
Calcium	0.74
Phosphorus _{available}	0.40

¹ American Protein Corp., Ankeny, IA, 50011.

² Provided the following per kg of diet: vitamin A, 6,614 IU; vitamin D₃, 1,653 IU; vitamin E, 33 IU; vitamin B₁₂, 33 µg; riboflavin, 10 mg; niacin, 50 mg, d-pantothenic acid, 26 mg.

³ Provided the following per kg of diet: Zn, 225 mg as ZnO; Fe, 263 mg as Fe₂SO₄; Cu, 26 mg as CuO; Mn, 90 mg as MnO₂; I, 3.0 mg as CaI; Se, 0.3 mg as Na₂SeO₃.

Table 3. Allowance of basal and test supplement in pigs fed various energy containing feedstuffs over the 4 d balance trial, as-is basis

<u>Diet¹</u>	<u>Basal, g²</u>	<u>Test supplement, g and (%)³</u>
Control	2000	-
USP	1818	182 (9.09%)
AGP-IA	1818	182 (9.09%)
REG-R	1818	182 (9.09%)
REG-WL	1818	182 (9.09%)
REG-AL	1818	182 (9.09%)
ADM-MO	1818	182 (9.09%)
WW-OH	1818	182 (9.09%)
WW-TX	1818	182 (9.09%)
IW-AC	1818	182 (9.09%)
IW-NA	1846	154 (7.72%)
USB-GA	1862	138 (6.91%)
Soybean oil	1866	134 (6.69%)
Lard	1866	134 (6.69%)

¹ Control diet or source of glycerin or lipid: USP = USP grade glycerin; AGP-IA = Ag Processing Inc., Sergeant Bluff, IA; REG-R = Renewable Energy Group, Ralston, IA; REG-WL = Renewable Energy Group, Wall Lake, IA; REG-MN = Renewable Energy Group, Albert Lea, MN; ADM-MO = Archer Daniels Midland, Mexico, MO; WW-OH = Westway Feed Products, Cincinnati, OH; WW-TX = Westway Feed Products, Houston, TX; IW-AC = Imperial Western Products-acidulated glycerin, Coachella, CA; IW-NA = Imperial Western Products-nonacidulated glycerin, Coachella, CA; USB-GA = US Biofuels, Rome, GA.

² Refers to amount of complex basal diet offered to the pig in addition to test supplement.

³ Refers to the amount of test supplement mixed with the amount of basal diet shown in the proceeding column. Number in parentheses represents the percent of test supplement relative to the total feed offered.

Table 4. Caloric values of various crude glycerin sources and lipids in starting swine, Exp. 1¹

Diet ²	N ³	GE		DE		ME	
		kcal/kg	kcal/kg	% of GE	kcal/kg	% of GE	% of DE
Control	11	3,945	3,469 ^{efg}	88.0 ^{def}	3,352 ^{de}	85.0	96.7 ^b
USP	11	4,325	4,457 ^c	103.1 ^{abc}	3,682 ^d	85.2	81.7 ^d
AGP-IA	11	3,601	3,022 ^g	84.0 ^{ef}	2,535 ^f	70.5	84.0 ^{cd}
REG-R	11	3,670	3,517 ^{efg}	95.9 ^{abcd}	3,024 ^e	82.5	86.2 ^{cd}
REG-WL	11	3,751	3,690 ^{def}	98.3 ^{abcd}	3,274 ^{de}	87.3	88.3 ^c
REG-AL	10	3,676	3,789 ^{cdef}	103.4 ^{abc}	3,299 ^{de}	89.9	86.9 ^{cd}
ADM-MO	10	3,627	3,928 ^{cde}	108.2 ^{ab}	3,389 ^{de}	93.4	85.7 ^{cd}
WW-OH	10	3,173	3,128 ^{fg}	98.5 ^{abcd}	2,794 ^e	88.0	89.0 ^c
WW-TX	11	3,489	3,815 ^{cde}	109.4 ^a	3,259 ^{de}	93.5	84.4 ^{cd}
IW-AC	11	4,153	3,919 ^{cde}	94.4 ^{bcde}	3,440 ^{de}	82.9	87.7 ^c
IW-NA	10	6,021	5,228 ^b	89.6 ^{def}	5,206 ^b	86.6	99.5 ^{ab}
USB-GA	12	5,581	4,336 ^{cd}	77.7 ^f	4,446 ^c	79.7	103.1 ^a
Soybean oil	10	9,443	8,567 ^a	90.8 ^{cdef}	8,469 ^a	89.8	98.2 ^{ab}
Lard	10	9,456	8,524 ^a	89.9 ^{cdef}	8,639 ^a	91.2	101.5 ^{ab}
Standard deviation		-	779	16.4	812	17.2	6.8
P value		-	0.01	0.01	0.01	0.19	0.01

¹ Pigs were adapted to diets and feeding regimens for 6 d prior to a 4 d collection period. Initial and final BW of 10.4 and 12.8 kg, respectively.

² Control diet or source of glycerin or lipid: USP = USP grade glycerin; AGP-IA = Ag Processing Inc., Sergeant Bluff, IA; REG-R = Renewable Energy Group, Ralston, IA; REG-WL = Renewable Energy Group, Wall Lake, IA; REG-MN = Renewable Energy Group, Albert Lea, MN; ADM-MO = Archer Daniels Midland, Mexico, MO; WW-OH = Westway Feed Products, Cincinnati, OH; WW-TX = Westway Feed Products, Houston, TX; IW-AC = Imperial Western Products-acidulated glycerin, Coachella, CA; IW-NA = Imperial Western Products-nonacidulated glycerin, Coachella, CA; USB-GA = US Biofuels, Rome, GA.

³ Number of observations (pigs) per dietary treatment.

Table 5. Impact of diet containing crude glycerin, soybean oil, or lard on pig performance, Exp. 2¹

Diet ²	ADG	ADFI	G:F	Gain:Mcal ME ³
Control (0%)	0.479	0.683	0.712 ^a	212 ^a
USP (9.09%)	0.383	0.652	0.584 ^{cd}	173 ^b
AGP-IA (9.09%)	0.364	0.594	0.609 ^{bcd}	186 ^b
REG-R (9.09%)	0.354	0.624	0.564 ^d	170 ^b
REG-WL (9.09%)	0.342	0.612	0.561 ^d	168 ^b
REG-AL (9.09%)	0.436	0.718	0.612 ^{bcd}	183 ^b
ADM-MO (9.09%)	0.420	0.696	0.602 ^{cd}	180 ^b
WW-OH (9.09%)	0.423	0.703	0.607 ^{cd}	184 ^b
WW-TX (9.09%)	0.436	0.702	0.621 ^{bcd}	186 ^b
IW-AC (9.09%)	0.394	0.660	0.597 ^{cd}	178 ^b
IW-NA (7.72%)	0.386	0.637	0.607 ^{cd}	174 ^b
USB-GA (6.91%)	0.429	0.675	0.631 ^{bc}	185 ^b
Soybean oil (6.69%)	0.361	0.580	0.620 ^{bcd}	168 ^b
Lard (6.69%)	0.371	0.547	0.676 ^{ab}	183 ^b
Pooled SEM	0.035	0.049	0.024	7.1
P value	0.25	0.34	0.01	0.01

¹ Data represents 8 replications of individually penned pigs (4 barrow and 4 gilt replicates, 8.70 kg initial BW) in 2 groups (25 d and 18 d trials, respectively). There were no 3- or 2-way interactions between group, gender, or diet; or for gender for any performance parameter.

² Control diet or source of glycerin or lipid: USP = USP grade glycerin; AGP-IA = Ag Processing Inc., Sergeant Bluff, IA; REG-R = Renewable Energy Group, Ralston, IA; REG-WL = Renewable Energy Group, Wall Lake, IA; REG-MN = Renewable Energy Group, Albert Lea, MN; ADM-MO = Archer Daniels Midland, Mexico, MO; WW-OH = Westway Feed Products, Cincinnati, OH; WW-TX = Westway Feed Products, Houston, TX; IW-AC = Imperial Western Products-acidulated glycerin, Coachella, CA; IW-NA = Imperial Western Products-nonacidulated glycerin, Coachella, CA; USB-GA = US Biofuels, Rome, GA. Values in parenthesis represent the concentration of test ingredient in the final diet.

³ Derived using ADG (g basis) divided by the ME intake (Mcal) obtained from the complete diet in Exp. 1.