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Summary and Implications

Carcass characteristics, meat quality indices, and tissue histology of growing pigs fed crude glycerol were determined after a 138-d feeding trial. Dietary treatments were 0, 5, and 10% crude glycerol inclusion in corn-soybean meal based diets. Diets were offered ad libitum in meal form and formulated to be equal in metabolizable energy (ME), sodium, chloride, and Lys, with other amino acids (AA) balanced on an ideal AA basis. At the end of the feeding trial, all pigs were scanned using real time ultrasound and pigs were processed at a commercial abattoir. Blood samples were collected pre-transport and at the time of harvest. Loins were removed from the carcass for meat quality, sensory evaluation, and fatty acid profile analysis. Kidney, liver, and eye tissues were collected at harvest and examined for lesions characteristic of methanol toxicity. Loin chop fatty acid profile was slightly changed by diet with the loin chops from pigs fed 10% crude glycerol having less linoleic acid and more eicosapentaenoic acid than pigs fed the 0 or 5% crude glycerol treatments. Dietary treatment did not affect blood metabolites or frequency of lesions in the examined tissues. This experiment demonstrates that pigs can be fed up to 10% crude glycerol with little to no effect on carcass composition, meat quality, or lesion scores.

Introduction

The production of biofuels, namely ethanol and biodiesel, is increasing dramatically. Biodiesel sales in the United States have grown exponentially since 1999 with existing U.S. production capacity at approximately 5.3 billion liters. A co-product of the biodiesel industry is crude glycerol, with 79 g of crude glycerol generated for every 1.0 liter of biodiesel produced. Consequently, current biodiesel production capacity could generate approximately

420,000,000 kg of crude glycerol annually. The objectives of the current study were to evaluate effects of crude glycerol supplementation on carcass composition, meat quality, fatty acid profile of the longissimus dorsi muscle lipid, and histology of the eye, liver, and kidney in growing pigs.

Materials and Methods

Crude glycerol was obtained from a biodiesel production facility (AG Processing Inc., Sergeant Bluff, IA) utilizing soybean (*Glycine max*) oil as its feedstock. Pigs (Cambrough 22 females × L337 terminal sires) were weaned at 21 d of age and fed a commercial starter diet for one week. Eight days post-weaning, 96 pigs (48 gilts, 48 barrows) with an average BW of 7.9 ± 0.4 kg were allotted to 24 pens (4 pigs/pen) with gender distribution and pen weight balanced at the start of the experiment. Dietary regimes were randomly assigned to each pen, with dietary treatments being 0, 5, and 10% crude glycerol inclusion in corn-soybean meal diets. There were five dietary phase changes over the 138-d trial. Within each phase, diets were offered ad libitum in meal form and were formulated to be equal in ME, sodium, chloride, and Lys with other AA balanced on an ideal AA basis.

During the course of the experiment, six pigs were removed from the trial due to health issues with no pattern of pig removal based on dietary treatment and no individual pen having more than one pig removed. On d-138, all pigs were individually weighed (133 ± 6 kg BW) for the termination of the performance period, bled for metabolite analysis, and scanned using real-time ultrasound. Pigs remained in their respective pens with access to feed and water until transport to the abattoir on d-139.

On the morning of d-139, 90 pigs were transported to the abattoir (Sioux-Preme Packing Co., Sioux Center, IA) with one pig dying in transit. On d-140, pigs were electrically stunned, exsanguinated, and blood, eye, and liver samples were harvested for further analysis. Pigs were chilled overnight (0°C) and on the following morning, one loin was removed from the left side of each carcass (10th rib to posterior tip), vacuum packaged, placed on ice, transported to Iowa State University, and stored at 0°C. Tissue and loin samples from two pigs were not collected at the abattoir due to operator error. Loin and chop marbling, moisture, drip loss, purge loss, color scores, and fatty acid profile were determined using standardized techniques. The loins of two pigs from each pen (1 barrow, 1 gilt) were randomly selected for sensory evaluation. Following 12 d of storage, two 2.54 cm thick loin chops were removed from the center of the loin for sensory and instrumental texture analysis.

On the day prior to slaughter, blood samples for plasma analysis were collected via jugular venapuncture into vacutainers containing sodium heparin. Blood samples were also collected on the day of harvest at the time of exsanguination into 50 ml centrifuge tubes containing sodium heparin (14.3 USP units/ml). Plasma samples were stored at -80°C pending analysis. Blood urea nitrogen, cortisol, glucose, glycerol, lactate, and creatine phosphokinase concentration were measured using commercially available kits and referred methods. All of the plasma metabolites were measured in duplicate. One eye, liver, and kidney per pig were collected at the time of slaughter and were prepared using standard techniques. All tissue samples were read for lesions twice by a single person versed in lesion evaluation.

Carcass composition and meat quality traits were evaluated using a regression model to test for effect of dietary treatment, pig gender, and diet \times gender interaction. Plasma metabolites pre-transport and at the time of harvesting were compared using a regression model to test for effect of dietary treatment. Differences in the frequency of histological lesions among dietary treatment groups were also evaluated using a simple regression model. Individual pigs were the experimental unit for analysis of carcass composition, meat quality, plasma metabolites, and lesion data.

Results and Discussion

The influence of diet \times gender, diet, and gender on carcass characteristics are described in Tables 1 and 2. There was no diet \times gender interaction for any trait examined. Dietary treatment did not affect estimated 10th rib backfat, hot carcass weight, percentage lean, or chop lipid percentage. These measures differed between barrows and gilts, with gilts producing a smaller carcass with a greater percentage lean and less backfat and total chop lipid ($P \leq 0.05$).

Pigs fed 10% crude glycerol had lower levels of linoleic acid than the other dietary treatments ($P < 0.01$). Eicosapentaenoic acid increased with increasing crude glycerol supplementation ($P = 0.02$). There was a trend ($P = 0.06$) for ultimate pH to increase with crude glycerol level. Meat quality and sensory evaluation of loin chops was not affected by diet, gender, or diet \times gender interaction (Table 3). Cooking loss was not affected ($P = 0.90$) by crude glycerol supplementation.

There was no effect of diet on any plasma metabolite measured nor was there any diet \times time interaction (Table 4). Levels of most plasma metabolites were different between pre-transport and at the time of harvest ($P < 0.01$) indicating the pigs were less calm following transport to the abattoir. Blood urea nitrogen was not affected by time of collection or diet, supporting the conclusion that neither kidney function nor lean tissue growth were not altered by feeding up to 10% crude glycerol. The absence of a dietary treatment effect on blood glycerol levels indicates

metabolism of dietary glycerol was not affected at levels less than or equal to 10% of the diet.

Current biodiesel processing techniques utilize methanol which is not completely recovered, and thus, methanol is found in crude glycerol at very low levels. An intermediate in the metabolism of methanol to carbon dioxide and water, is formaldehyde and formate. The metabolism of formate to carbon dioxide and water is slow in some species, which results in accumulation of formate, the metabolic product principally responsible for the toxic effects of methanol. Clinical consequences of methanol poisoning are central nervous system depression, vomiting, severe metabolic acidosis, blindness, and Parkinsonian-like motor disease. During the course of the study, no pig demonstrated clinical symptoms of methanol toxicity. The six animals that were removed during the trial were removed for respiratory disease or lameness, with no attribution to a specific dietary treatment. Of the 87 pigs harvested, no gross lesions were observed at the time of collection. In addition, frequency of histological lesions in kidney, liver, and eye, the pharmacological targets for methanol toxicity, were not influenced by dietary treatment (Table 5).

Overall, the results from this study demonstrate that up to 10% crude glycerol can be fed to pigs with little to no effect on carcass composition, meat quality, or lesion scores in the eye, liver, or kidney tissue. Up to 10% crude glycerol fed to pigs does not seem to impact lean tissue growth and energy balance if fed diets are equal in nutrient density. While this study was not designed to examine the toxicology of methanol fed to pigs, the results of this trial indicate that pig performance and health were not negatively affected by the levels of methanol fed. With noted effects on pH and fatty acid profiles, however, the affect of crude glycerol on meat quality may warrant further examination through method and length of administration. Crude glycerol is a viable source of dietary energy that is well utilized by pigs. Inclusion of crude glycerol in pig diets may be determined by the relative availability and price of other dietary energy sources.

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Table 1. Effect of crude glycerol on estimated carcass characteristics¹.

	Diet ²			SEM	Gender			P-value		
	0	5	10		Barrow	Gilt	SEM	Diet	Gender	D × G
Pigs, no.	30	29	31		44	46				
Initial BW, kg	8.0	8.0	7.9	0.2	7.9	8.0	0.2	0.80	0.78	0.69
Final BW, kg	133	134	133	2.0	137	129	2.0	0.93	< 0.01	0.92
10 th rib backfat, mm	18.8	21.0	20.7	0.8	22.0	18.3	0.7	0.14	< 0.01	0.13
LM area, cm ²	48.6	49.0	46.6	0.9	48.0	48.1	0.7	0.12	0.92	0.33
Fat free lean, %	52.0	51.8	50.6	0.8	51.9	51.1	0.6	0.37	0.34	0.78
Lean gain, g/d	365	363	355	5.0	364	358	4.0	0.37	0.30	0.70

¹From ultrasound scan data.

²Dietary treatments were 0, 5, or 10% crude glycerol inclusion in corn-soybean meal diets fed in five phases over a 138-d feeding trial.

Table 2. Carcass characteristics and fatty acid profile of loin chops from pigs fed crude glycerol.

	Diet ¹			SEM	Gender			P-value		
	0	5	10		Barrow	Gilt	SEM	Diet	Gender	D × G
Loins, no.	27	29	31		43	44				
Hot carcass wt, kg	95.2	97.2	97.3	1.8	98.7	94.5	1.4	0.61	0.03	0.97
Lean, %	55.8	54.7	55.5	0.5	54.7	56.0	0.4	0.21	0.02	0.61
Moisture, %	74.0	73.9	74.0	0.1	73.8	74.1	0.1	0.78	< 0.01	0.78
Total lipid, %	1.30	1.31	1.25	0.03	1.30	1.27	0.02	0.31	0.30	0.47
Ultimate pH	5.57	5.65	5.65	0.03	5.63	5.62	0.02	0.06	0.77	0.59
Drip loss, %	0.85	0.73	0.81	0.10	0.79	0.80	0.08	0.67	0.96	0.87
Loin purge, %	1.67	1.84	1.62	0.17	1.77	1.65	0.13	0.61	0.54	0.43
Chop purge, %	3.72	3.84	3.90	0.30	3.70	3.94	0.20	0.90	0.46	0.24
Chop lipid, %	2.15	2.07	2.08	0.07	2.19	2.02	0.06	0.71	0.04	0.70
Fatty acids ²										
14:0	1.29	1.31	1.25	0.03	1.30	1.27	0.02	0.06	0.03	0.04
16:0	24.10	24.14	24.15	0.19	24.29	23.97	0.16	0.98	0.15	0.48
16:1 (n - 7)	3.73	3.87	3.82	0.08	3.79	3.83	0.07	0.45	0.65	0.29
17:0	0.28	0.29	0.25	0.02	0.28	0.27	0.01	0.28	0.64	0.68
17:1 (n - 10)	0.27	0.30	0.30	0.01	0.29	0.29	0.01	0.08	0.68	0.59
18:0	11.68	11.77	12.00	0.18	11.86	11.78	0.14	0.41	0.69	0.39
18:1	39.47	38.92	40.18	0.44	39.90	39.14	0.36	0.12	0.13	0.75
Unknown	5.12	5.24	5.10	0.10	5.08	5.22	0.08	0.57	0.26	0.23
18:2 (n - 6)	10.34	10.34	9.27	0.26	9.68	10.28	0.21	< 0.01	0.04	0.63
18:3 (n - 3)	0.27	0.29	0.27	0.02	0.26	0.30	0.02	0.78	0.17	0.65
20:0	0.13	0.13	0.13	0.01	0.13	0.13	0.01	0.55	0.82	0.75
20:4 (n - 6)	2.96	3.00	2.90	0.10	2.78	3.13	0.08	0.76	< 0.01	0.76
20:5 (n - 3)	0.09	0.10	0.11	0.01	0.09	0.10	0.01	0.02	0.05	0.90
22:5 (n - 6)	0.28	0.29	0.28	0.01	0.26	0.30	0.01	0.08	< 0.01	0.75

¹Dietary treatments were 0, 5, or 10% crude glycerol inclusion in corn-soybean meal diets fed in five phases over a 138-d feeding trial.

²Fatty acids are expressed as g/100g total fatty acids. Fatty acids are designated by the number of carbon atoms followed by the number of double bonds. The position of the first double bond relative to the methyl (n) end of the molecule is also included.

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Table 3. Meat quality and sensory evaluation of loin chops from pigs fed crude glycerol.

	Diet ¹			SEM	Gender		SEM	P-value		
	0	5	10		Barrow	Gilt		Diet	Gender	D × G
Loins, no.	16	16	16		24	24				
Loin marbling score ²	2.0	2.1	2.1	0.1	2.1	2.0	0.1	0.8	0.6	0.6
Cook loss, %	18.3	17.9	18.6	0.9	18.7	17.9	0.7	0.9	0.5	0.8
Japanese color score ³	2.6	2.7	2.8	0.8	2.7	2.7	0.1	0.8	0.8	0.5
Hunter L* ⁴	53.4	53.0	53.4	0.9	53.7	52.9	0.8	0.9	0.5	0.4
Minolta L* ⁴	55.6	55.3	55.6	0.8	55.8	55.1	0.7	0.9	0.5	0.4
Minolta a* ⁴	17.5	17.4	17.4	0.2	17.3	17.6	0.1	0.9	0.2	0.1
Minolta b* ⁴	4.9	5.1	4.6	0.4	4.9	4.9	0.3	0.7	0.9	0.1
Instron, kg force ⁵	2.0	2.1	2.1	0.1	2.1	2.0	0.1	0.8	0.6	0.6
Juiciness score ⁶	5.5	5.7	5.5	0.4	5.4	5.7	0.3	0.9	0.5	0.4
Tenderness score ⁶	6.1	6.1	5.9	0.4	5.8	6.3	0.3	0.9	0.2	0.3
Chewiness score ⁶	3.6	3.4	3.3	0.3	3.5	3.3	0.2	0.7	0.4	0.3
Pork flavor score ⁶	2.2	2.2	2.2	0.1	2.2	2.2	0.1	0.9	0.6	0.1
Off-flavor score ⁶	3.5	3.4	3.1	0.3	3.2	3.5	0.3	0.7	0.4	0.2

¹Dietary treatments were 0, 5, or 10% crude glycerol inclusion in corn-soybean meal diets fed in five phases over a 138-d feeding trial.

²Evaluated 12 d postmortem according to National Pork Board Standards (Berg, 2000). The marbling standards correspond to % intramuscular lipid.

³Japanese color bar 1–6 scale, 1 = extremely light, 6 = extremely dark.

⁴Higher L* values indicate a lighter color, higher a* values indicate a redder color, and higher b* values indicate a more yellow color.

⁵Average of 3 maximum force peaks.

⁶Scores on a 1–10 scale. Lower scores represent low degrees of characteristics, high scores represent high degrees of characteristics.

Table 4. Effect of crude glycerol on plasma metabolites pre-transport and at the time of harvest.

Diet ²	Pre-transport ¹			Harvest			SEM	P-value		
	0	5	10	0	5	10		Diet	Time	D × T
BUN mg/dL ³	14.7	14.5	13.6	14.0	14.6	13.8	0.5	0.24	0.67	0.59
Cortisol, µg/dL	6.7	6.6	6.1	15.1	11.8	13.6	1.6	0.56	< 0.01	0.59
Glucose, mg/dL	101.8	99.0	98.0	138.6	143.4	140.3	4.6	0.91	< 0.01	0.70
Glycerol, µM	0.04	0.04	0.04	417.5	410.3	444.8	34.7	0.87	< 0.01	0.87
Lactate, mM	4.0	4.7	4.1	12.4	12.3	12.2	0.6	0.86	< 0.01	0.83
CPK, IU/L ⁴	720.2	683.3	678.0	1,844.2	2,212.7	1,954.8	110.3	0.29	< 0.01	0.19

¹Blood samples for plasma analysis were collected prior to transport to the abattoir and at the time of harvest immediately after electrical stunning.

²Dietary treatments were 0, 5, or 10% crude glycerol inclusion in corn-soybean meal diets fed in five phases over a 138-d feeding trial.

³BUN = blood urea nitrogen

⁴CPK = creatine phosphokinase

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Table 5. Frequency of histological lesions in tissue of pigs fed crude glycerol¹.

Lesion, % of tissues with lesion	Diet ²			SEM	P-value
	0	5	10		
Hepatocellular pleomorphism	93.1	96.6	96.8	4.0	0.75
Portal hepatitis	41.3	34.5	45.1	9.2	0.70
Periportal fibrosis	27.6	17.2	12.9	7.3	0.34
Lymphoplasmacytic interstitial nephritis	41.4	41.4	48.4	9.4	0.82
Lymphoplasmacytic hepatitis	3.4	3.4	3.2	3.4	0.99
Lymphohistiocytic perineuritis	0.0	3.4	0.0	2.0	0.36
Hepatic lipidosis	3.4	0.0	0.0	2.0	0.35

¹No gross lesions were observed in tissues harvested. One eye, liver, and kidney were collected from 29, 29, and 31 pigs for Diet 0, 5, and 10, respectively.

²Dietary treatments were 0, 5, or 10% crude glycerol inclusion in corn-soybean meal diets fed in five phases over a 138-d feeding trial.