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Effects of nutrient density and emulsifier supplementation on apparent total tract digestibility, apparent ileal digestibility and urinary nitrogen in weaned pigs

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ABSTRACT. Sixteen weaned pigs with an average body weight of 20.19 ± 0.21 kg were individually fitted with T-cannulas in the distal ileum and randomly assigned to four dietary treatments in a replicated 4 × 4 Latin square design. The dietary treatments were as follows (TRT): 1) TRT1, high nutrient diet (HD); 2) TRT2, HD + 0.05% emulsifier; 3) TRT3, low nutrient diet (LD); 4) TRT4, LD + 0.05% emulsifier. Individual pigs served as the experimental unit to evaluate the effect of emulsifier supplementation in HD and LD diets. Pigs fed the HD diet with 0.05% emulsifier had significantly improved apparent total tract digestibility (ATTD) and apparent ileal digestibility (AID) of dry matter, nitrogen, energy, crude protein and crude fat. Moreover, the ATTD of essential amino acid, including valine, leucine phenylalanine and, methionine, as well as non-essential amino acid: glutamic acid, proline, glycine, alanine, tyrosine, and cystine was significantly increased in pigs fed the HD diet with 0.05% emulsifier ($P < 0.05$). The inclusion of 0.05% emulsifier to the HD diet also significantly increased the AID of essential amino acid like threonine, valine, isoleucine, lysine, arginine and methionine, as well as non-essential amino acid such as serine, glutamic acid, glycine, tyrosine and cystine ($P < 0.05$). Furthermore, pigs receiving the HD diet with emulsifier supplementation showed a significant increase in urinary nitrogen content ($P < 0.05$). These results suggest that feeding weaned pigs a high-nutrient density diet with 0.05% emulsifier is an effective strategy to improve their ATTD and AID.

Introduction

Rearing pigs without antibiotic growth promoters has become increasingly challenging, particularly during the post-weaning phase. As a result, many pig farmers are looking for other solutions that could replace antibiotics. As a result, post-weaning diets are precisely formulated to support the piglets' immature digestive systems (Muniyappan et al., 2022). Lipids are a vital secondary energy source in piglet diets. After early weaning, piglets typically consume less feed, making the inclusion of soybean oil as a vegetable fat an effective strategy to meet their energy demands (Soares and Lopez-Bote, 2002). Dietary fats are considered important ingredients providing more energy than protein and carbohydrates. Previous studies have demonstrated that incorporating fat into piglet diets improves average daily gain (ADG) and the digestibility of dry matter (DM) and crude protein (CP), particularly in the later stages of growth after weaning compared to early stages (Li et al., 1990). However, weaned pigs have difficulties digesting dietary fat, especially in the early post-weaning period (Lewis et al., 2000). There is convincing evidence that piglet diets supplemented with exogenous emulsifiers alleviate the adverse effects of maldigestion caused by hepatic bile acid synthesis (Zhao et al., 2015).

Emulsifiers are compounds composed of two distinct parts: a hydrophilic (water-attracting) component and a hydrophobic (water-repelling) component. The hydrophilic portion of the emulsifier molecule dissolves in oil droplets, while the hydrophobic part does not (Park et al., 2024). This dual property allows emulsifiers to stabilise oil droplets in a dispersed emulsion, facilitating fat absorption and digestion (MaClements, 2004). Emulsifiers function as catalyst by increasing the active surface area of lipids, which improves lipase triglyceride hydrolysis into fatty acids and monoglycerides. This process promotes the formation of micelles that encapsulate the product of lipolysis, improving their absorption in the digestive system (Upadhaya et al., 2018). Commercially available emulsifiers are available in various forms such as soy lecithin, Tween 20, Tween 80, 1,3-diacylglycerol, sodium stearoyl-2-lactylate, or lysophosphatidylcholine. Research indicates that emulsifiers improve growth performance and nutrient digestibility in pigs (Zhao et al., 2015), nutrient digestion in lactating sows (Wang et al., 2017), and fat digestibility in broilers (Upadhaya et al., 2018). The concept of hydrophilic-lipophilic balance serves as the basis for the systematic selection of emulsifiers to produce effective emulsions (Hasenhuettl, 2008). Furthermore, studies demonstrate that combining emulsifiers can improve emulsion stability compared to using individual emulsifiers (Boyd et al., 1972). However, limited research exists on the use of sodium stearoyl-2-lactylate as an emulsifier in pigs, especially during the weaning period.

We hypothesised that piglets fed diets supplemented with emulsifiers would exhibit higher apparent ileal digestibility (AID) and apparent total tract digestibility (ATTD) of nutrients compared to those fed diets with high- or low-nutrient density feeds alone. The objective of this study was to evaluate the effects of emulsifiers on ATTD, and AID of dietary nutrients, amino acids, nitrogen and energy content in weaned piglets.

Material and methods

All animal procedures used in this experiment were reviewed and approved (Approval number: DK-1-2301) by the Institutional Animal Care and Use Committee of Dankook University (Cheonan, Republic of Korea).

Tested product

The emulsifier, sodium stearoyl lactylate (SSL) consisted of sodium stearoyl-2-lactylate (80%) and Tween 20 (20%), and was purchased from Daeho Co., Ltd. (Seoul, Republic of Korea).

Animals and experimental design

Sixteen [(Duroc × Yorkshire) × Landrace)] weaned pigs with an average body weight of 20.19 ± 0.21 kg, were selected for the study. Simple T-cannulas were surgically implanted 15 cm proximal to the ileocecal junction, following procedures adapted from Sauer et al. (1983). The pigs were randomly assigned to one of four dietary treatments, with assessments conducted over three consecutive 7-day periods. The study utilized a 4 × 4 Latin square design, assigning two pigs per treatment in each period. The dietary treatment (TRT) included: 1) TRT1, high-nutrient diet (HD); 2) TRT2, HD + 0.05% emulsifier; 3) TRT3, low-nutrient diet (LD); 4) TRT4, LD + 0.05% emulsifier. The basal diets were formulated to meet or exceed the nutrient requirements specified by the NRC (2012) (Table 1). Prior to surgery, the pigs were

Table 1. Ingredient composition of experimental diets, as-fed basis

Ingredient, %	High nutrient density	Low nutrient density
Maize	71.55	76.30
Soybean meal, 48%	16.10	7.88
DDGS	6.00	10.00
Tallow	2.65	2.57
MDCP	1.44	0.95
Limestone	0.78	0.72
Salt	0.30	0.30
Methionine (99%)	0.06	0.05
L-lysine (78%)	0.55	0.55
Threonine (99%)	0.10	0.19
Tryptophan (99%)	0.04	0.06
Vit / Mineral Premix[1]	0.40	0.40
Choline (25%)	0.03	0.03
Total	100.00	100.00
Calculated value		
CP, %	15.50	13.00
ME, kcal/kg	3300	3300
fat, %	5.85	6.16
Ca, %	0.70	0.55
P, %	0.60	0.50
lysine, %	1.10	0.90
methionine, %	0.31	0.27

DDGS – Dried distillers grains with solubles, MDCP – monodicalcium phosphate, CP – crude protein, ME – metabolizable energy; [1] provided per kg of complete diet: IU: vit. A 16 800 IU, vit. D_3 2 400; mg: vit. E 108, vit. K 7.2, riboflavin 18, niacin 80.4, thiamine 2.64, D-pantothenic 45. amino acids, apparent ileal digestibility, apparent total tract digestibility emulsifier, nutrient density, weaned pigs 6, cobalamin 0.06, Cu 12 (as $CuSO_4$); Zn 60 (as $ZnSO_4$), Mn 24 (as $MnSO_4$), I 0.6 (as Ca $(IO_3)_2$), Se 0.36 (as Na_2SeO_3)

fasted for 16–20 hours. To anaesthetise the animals, 0.2 ml/kg of tiletamine/zolazepam (Zoletil 50, Virbac Laboratory, La Seyne-sur-Mer, France) and azaperone injections (Stresnil, Janssen Pharmaceutica, Beerse, Belgium) were administered. After surgery, the pigs were housed individually in pens (1.2 × 0.6 m) in a temperature-controlled room (28 °C) for a 10-day recovery period. Pre- and post-operative care followed established protocols described by Upadhaya and Kim (2015) and Hossain et al. (2016). The daily feed ration was 0.05 × body weight (kg)$^{0.9}$, as proposed by Muniyappan et al. (2022). Feed was provided twice daily at 12-hour intervals (8:00 and 20:00), and water was available *ad libitum*. Additionally, chromic oxide (Cr_2O_3, 2 g/kg) was added to the diet as an indigestible marker to facilitate digestibility measurements according to the method described by Fenton and Fenton (1979).

Experimental and laboratory analysis

Body weight was determined at the beginning of the experiment. Each experimental period consisted of four days of diet adaptation, followed by one day of faeces collection and one day of ileal digesta collection. Faeces were collected from 08:00 on Day 5 to 08:00 on Day 6. Ileal digesta samples were collected continuously over 24 h, from 08:00 on Day 7 to 08:00 h on Day 8. The pigs were trained to eat their rations in 15 min or less, and the amount of feed offered was adjusted to match the intake of the pig with the lowest consumption, ensuring consistent daily intake for all pigs. Ileal digesta were collected in plastic bags attached to the barrel of the cannula. The bags were removed and replaced promptly once filled with digesta, with no bag remaining attached for longer than 20 min. All the faecal and ileal digesta samples were stored in a freezer at −20 °C until analysis. Before chemical analysis, faecal and ileal digesta samples were thawed and dried at 70 °C for 72 hours and subsequently ground to pass through a 1-mm screen. The samples were analysed for DM and CP according to the AOAC International (2005) procedures. Chromium was analyzed by UV absorption spectrophotometry (UV-1201; Shimadzu, Kyoto, Japan) according to the methods of Muniyappan et al. (2022). Nitrogen was quantified using a Kjeltec 2300 Analyzer (Foss Tecator AB, Hoeganaes, Sweden). Gross energy content was determined using a Parr 6100 oxygen bomb calorimeter (Parr Instrument Co., Moline, IL, USA). The amino acid composition of the samples was measured by insulation using a Beckman 6300 amino acid analyzer (Beckman Coulter, Inc., Fullerton, CA, USA) and high-performance liquid chromatography (HPLC) (Agilent Technologies, Santa Clara, CA, USA) after acid hydrolysis for 24 h in 6 N HCl at 110 °C. Amino acid isolation involved acid hydrolysis for 24 h in 6 N HCl at 110 °C, followed by vaporisation at 50 °C using an evaporator (HAHNSHIN S&T Co. Ltd. Gyeonggi-do, Republic of Korea). Sulphur-containing amino acids were analysed after overnight oxidation with cold performic acid, followed by hydrolysis. ATTD and AID values for nutrients and amino acids were calculated according to the method described by Stein et al. (2007). The apparent ileal amino acid (AA) digestibility was calculated using the formula described by Wu et al. (2020):

$$\text{AID (\%)} = 100 - [100 \times (AA_{digesta} \times Cr_2O_{3diet}) / (AA_{diet} \times Cr_2O_{3digesta})],$$

where: AID – apparent ileal digestibility, AA_{diet} and $AA_{digesta}$ – concentrations (mg/kg DM) of AA in the diet and digesta, respectively; Cr_2O_{3diet} and $Cr_2O_{3digesta}$ – concentrations (mg/kg DM) of the indigestible marker in the diet and digesta, respectively.

The apparent ileal AA digestibility was standardised using average values for basal endogenous AA losses (AAEL) and calculated using the following formula (Muniyappan et al., 2022):

$$\text{AAEL (g/kg)} = AA_{digesta} \times (Cr_2O_{3diet} / Cr_2O_{3digesta}),$$

where: AAEL – average endogenous AA loss in a casein-based diet (g/kg DM); AA – amino acid; Cr_2O_{3diet} and $Cr_2O_{3digesta}$ – concentrations of the indigestible marker in the diet and digesta, respectively.

Urine analysis

Urine samples were collected to plastic screw-top containers and refrigerated before energy and nitrogen analyses. The drying method was a factor in the determination of the gross energy (GE) of urine. However, nitrogen was analysed exclusively from undried samples due to the limitations of the thermocombustion equipment used in this experiment. For GE analysis, cellulose pellets were used to absorb and dry the urine for oven-drying procedures, whereas cotton balls and small plastic bags (Jeb Plastics Inc., Wilmington, DE, USA) were used for urine absorption in crucibles for freeze-drying, as described by Dadalt et al. (2017). In both instances, samples were allowed to dry for 48 h. During the drying processes, the oven temperature fluctuated by ±1 °C, whereas the freeze drier showed a variation of ±2 °C.

Statistical analyses

Data were analysed using a completely randomised design with a 2 × 2 factorial arrangement employing GLM procedures (SAS Institute, 1996) when significant differences were observed. Data variability was expressed as the standard error (SE) and significance was set at $P < 0.05$.

Results

Apparent total tract digestibility of amino acids

The effect of emulsifiers supplementation to low- and high-nutrient diets on ATTD of amino acid in the digestive tract of weaned pigs is given in Table 2. Pigs fed a HD diet with 0.05% emulsifier showed a significant increase ($P < 0.05$) in the ATTD of dry matter, nitrogen, crude protein and crude fat; similarly, the ATTD of essential amino acids: valine, leucine, phenylalanine and methionine, and non-essential amino acids: glutamic acid, proline, glycine, alanine, tyrosine, cysteine, as well as total amino acids, was increased compared to those fed the HD diet without emulsifiers. Conversely, a significantly reduction ($P < 0.05$) in the ATTD was recorded for crude protein and following essential amino acids: valine, isoleucine, leucine, phenylalanine and tryptophan, as well as non-essential amino acids: aspartic acid, serine and glutamic acid, in pigs fed the HD compared to the LD diet. No interaction effects were detected between dietary emulsifiers and nutrient density.

Apparent ileal digestibility of amino acids

The effects of emulsifiers added to the low- and high-nutrient diets on the AID of amino acids in the digestive tract of weaned pigs are summarised in Table 3. The AID of crude protein, valine, isoleucine, phenylalanine, lysine, arginine, aspartic

Table 2. Effect of high- and low-nutrient diet with emulsifier supplementation on apparent total tract digestibility in weaned pigs

Items, %	High nutrient density		Low nutrient density		SEM	*P*-value		
	0% emulsifier	0.05% emulsifier	0% emulsifier	0.05% emulsifier		nutrient emulsifier	emulsifier effect	interaction
Apparent total tract digestibility								
dry matter, %	76.37^{ab}	79.32^{a}	74.32^{b}	77.54^{ab}	0.86	0.1228	0.0143	0.2011
nitrogen, %	77.72^{ab}	81.02^{a}	75.99^{b}	78.56^{ab}	0.84	0.0817	0.0160	0.0813
energy, %	77.07	80.11	76.40	77.65	0.86	0.2011	0.0813	0.4636
crude protein	77.75^{ab}	81.71^{a}	76.70^{b}	77.64^{ab}	0.62	0.0138	0.0172	0.1143
crude fat	79.55^{ab}	83.68^{a}	78.69^{b}	82.41^{a}	0.93	0.4355	0.0118	0.8800
phosphorus	51.04	53.02	49.29	51.21	0.92	0.1955	0.1589	0.9774
Essential amino acid								
threonine	75.94	78.92	75.82	77.20	0.82	0.4424	0.0848	0.5028
valine	74.72^{ab}	77.97^{a}	72.37^{b}	75.07^{a}	0.92	0.0674	0.0418	0.8374
isoleucine	76.87	79.21	72.69	74.54	0.80	0.0005	0.3040	0.5894
leucine	82.07^{b}	84.79^{a}	81.40^{b}	83.52^{ab}	0.75	0.3814	0.0424	0.7812
phenylalanine	81.26^{a}	83.39^{a}	77.35^{b}	80.49^{ab}	0.71	0.0053	0.0222	0.6216
histidine	87.66	86.46	87.10	87.89	0.76	0.6936	0.8492	0.3722
lysine	85.00^{a}	86.89^{a}	82.16^{b}	82.99^{b}	0.51	0.0006	0.0862	0.4824
arginine	90.81	92.09	90.76	91.09	0.48	0.2955	0.1196	0.3442
methionine	76.16^{b}	80.41^{a}	78.09^{b}	80.17^{a}	0.86	0.4989	0.0227	0.3881
tryptophan	76.76^{b}	78.05^{b}	83.98^{a}	83.69^{a}	0.88	0.0002	0.6917	0.5383
TEAA	80.73	82.82	80.17	81.66	0.64	0.3211	0.0853	0.8194
Non-essential amino acid								
aspartic acid	80.87	83.49	77.53	77.82	0.84	0.0026	0.2437	0.3456
serine	82.71	84.60	80.62	81.84	0.61	0.0160	0.0980	0.7025
glutamic acid	86.59^{ab}	89.09^{a}	84.55^{b}	86.64^{ab}	0.53	0.0107	0.0094	0.7861
proline	84.87^{ab}	87.44^{a}	82.87^{b}	85.98^{ab}	0.64	0.0791	0.0084	0.7656
glycine	74.51^{ab}	77.71^{a}	72.19^{b}	74.64^{ab}	0.89	0.0536	0.0449	0.7704
alanine	73.28^{b}	77.81^{a}	73.03^{b}	76.03^{ab}	0.95	0.4651	0.0162	0.5836
tyrosine	77.12^{ab}	78.85^{a}	74.15^{b}	79.58^{a}	0.69	0.2715	0.0031	0.0818
cystine	80.22	83.62	81.26	81.66	0.56	0.5719	0.0329	0.0814
TNEAA	80.02^{ab}	82.83^{a}	78.27^{b}	80.52^{ab}	0.68	0.0558	0.0216	0.7766
Total amino acid	80.41^{ab}	82.82^{a}	79.33^{b}	81.16^{ab}	0.66	0.1523	0.0461	0.7988

emulsifier – sodium stearoyl-2-lactylate (80%) and Tween 20 (20%), TEAA – total essential amino acid, TNEAA – total non-essential amino acid, SEM – standard error of the mean; ^{ab} – means in the same row with different superscript are significantly different at $P < 0.05$

Table 3. Effect of high- and lownutrient diet with emulsifier supplementation on apparent ileal digestibility in weaned pigs

Items, %	High nutrient density		Low nutrient density		SEM	*P*-value		
	0% emulsifier	0.05% emulsifier	0% emulsifier	0.05% emulsifier		nutrient effect	emulsifier effect	Interaction
Apparent ileal digestibility								
dry matter, %	73.39[ab]	75.80[a]	72.19[b]	74.09[ab]	0.84	0.0897	0.0128	0.7641
nitrogen, %	72.62[ab]	75.30[a]	71.03[b]	73.57[ab]	0.87	0.0535	0.0037	0.9361
energy, %	72.10[ab]	75.04[a]	71.30[b]	73.23[ab]	0.94	0.1675	0.0117	0.5903
crude protein	69.69[a]	73.71[a]	64.96[b]	66.95[b]	1.69	0.0053	0.1002	0.5576
crude fat	73.55[ab]	77.52[a]	72.82[b]	75.27[ab]	0.91	0.1258	0.0041	0.4170
phosphorus	45.86[b]	47.88[a]	44.25[b]	46.16[ab]	0.87	0.0805	0.0438	0.9564
Essential amino acid								
threonine	66.27[b]	73.49[a]	65.52[b]	68.67[b]	1.46	0.0811	0.0041	0.1891
valine	68.64[b]	72.51[a]	65.00[b]	68.57[b]	1.52	0.0279	0.0306	0.9216
isoleucine	77.47[a]	80.84[a]	71.34[b]	74.27[b]	1.39	0.0006	0.0422	0.8759
leucine	79.69	82.32	78.67	80.89	1.17	0.3154	0.0606	0.8666
phenylalanine	76.81	79.76	72.74	74.73	1.29	0.0042	0.0792	0.7159
histidine	70.41[b]	78.76[a]	73.66[b]	75.82[ab]	1.71	0.9297	0.0095	0.0951
lysine	82.80[a]	85.53[a]	77.58[b]	79.77[ab]	0.96	0.0001	0.0247	0.7797
arginine	86.71[ab]	88.95[a]	84.15[b]	85.90[b]	0.56	0.0003	0.0039	0.6737
methionine	74.98[b]	80.39[a]	75.77[b]	79.89[ab]	1.59	0.9291	0.0110	0.6901
tryptophan	66.34	73.02	64.77	70.33	2.88	0.4735	0.0550	0.8476
TEAA	75.01[ab]	79.56[a]	72.92[b]	75.88[ab]	1.05	0.0180	0.0039	0.4668
Non-essential amino acid								
aspartic acid	73.54[a]	78.08[a]	66.54[b]	69.13[b]	2.04	0.0020	0.1058	0.6402
serine	73.26[a]	78.71[a]	69.70[b]	72.71[b]	1.20	0.0018	0.0042	0.3283
glutamic acid	82.19[ab]	84.41[a]	79.54[b]	81.21[ab]	0.76	0.0023	0.0248	0.7284
proline	76.74	79.63	76.89	78.65	1.78	0.7324	0.0723	0.6417
glycine	62.28[b]	69.28[a]	58.46[b]	62.11[b]	2.16	0.0233	0.0271	0.4435
alanine	70.11	74.61	69.07	71.38	1.64	0.2173	0.0606	0.5190
tyrosine	72.08[b]	78.29[a]	73.23[a]	74.69[ab]	1.23	0.3395	0.0089	0.0774
cystine	58.32[ab]	66.88[a]	61.92[b]	66.91[a]	1.83	0.3393	0.0030	0.3483
TNEAA	71.07[ab]	76.23[a]	69.42[b]	72.10[ab]	1.36	0.0545	0.0135	0.3765
Total amino acid	73.26[b]	78.08[a]	71.36[b]	74.20[ab]	1.16	0.0290	0.0065	0.4103

emulsifier – sodium stearoyl-2-lactylate (80%) and Tween 20 (20%), TEAA – total essential amino acid, TNEAA – total non-essential amino acid, SEM – standard error of the mean; [ab] – means in the same row with different superscript are significantly different at $P < 0.05$

acid, serine, glutamic acid, glycine and total amino acids was significantly lower ($P < 0.05$) in pigs fed the LD diet compared to the HD diet. The inclusion of the emulsifier to the HD diet significantly increased ($P < 0.05$) the AID of dry matter, nitrogen, energy, crude fat, and phosphorus, threonine, valine, isoleucine, histone, lysine, arginine, methionine, serine, glutamic acid, glycine, tyrosine, cysteine, and total amino acids. No interaction effects were observed between dietary nutrient density and emulsifiers on the apparent ileal digestibility of amino acids.

Urine content

An increase ($P < 0.05$) in urine volume was observed in piglets fed emulsifier-supplemented diets. However, urine energy content remained unaffected ($P > 0.05$) by the supplementation of emulsifiers, or the type of diet (Table 4).

Table 4. Effect of high- and low-nutrient diet with emulsifier supplementation on nutrient digestibility in weaned pigs

Items, %	High nutrient density		Low nutrient density		SEM	*P*-value		
	0% emulsifier	0.05% emulsifier	0% emulsifier	0.05% emulsifier		nutrient effect	emulsifier effect	Interaction
Urine								
nitrogen, %	9.02[ab]	9.64[a]	8.46[b]	9.17[ab]	0.30	0.0889	0.0283	0.8858
energy, cal/g	437	458	421	441	13	0.1910	0.1089	0.9648

emulsifier – sodium stearoyl-2-lactylate (80%) and Tween 20 (20%), SEM – standard error of the mean; [ab] – means in the same row with different superscript are significantly different at $P < 0.05$

Discussion

The present study evaluated sodium stearoyl-2-lactylate and Tween 20 with hydrophilic properties as fat emulsifiers in diets fed to weaned pigs. Two primary objectives of pig feeding systems are to supply the necessary amino acids in adequate amounts to promote optimal and efficient growth, and to reduce nitrogen excretion, thereby mitigating environmental pollution (Dadalt et al., 2017). Ileal digestibility tests in pigs are recognised methods for assessing the digestibility of amino acids in feed components (Kim et al., 2000). Just et al. (1985) demonstrated that digestible protein and amino acids levels determined in faecal samples showed lower correlations with carcass protein compared to digestible protein and amino acids obtained from ileal digesta. According to Yin et al. (2000), formulating diets for growing pigs based on the content of ileal digestible lysine, methionine, isoleucine, leucine, and arginine, rather than total amino acid content, may improve the feed-to-gain ratio and provide economic benefits (Hodgkinson et al., 2020). Energy and nitrogen inputs are essential to sustain growth and nutrient digestion in rapidly weaning pigs. To meet these needs, animal fats and vegetable oils are often added to diets to increase nutrient content (Boontiam et al., 2017). Fat is considered the most nutrient-dense energy source, providing 2.25 times higher nutritional value compared to carbohydrates (Field and Robinson, 2019). However, fat has low metabolic efficiency because it is insoluble in water and difficult to digest and absorb in the gastrointestinal tract (Olsen et al., 2021). Dietary nutrient utilisation reflects appropriate dietary nutrient density, and use of nutrients may be prioritised for efficient growth and then for maintenance of life functions. According to this hypothesis, high-nutrient density diets provide suitable growth as they result in relatively higher ATTD and AID values of crude protein and amino acids compared to low-nutrient density diets, as observed in the present study. Sun et al. (2019) reported that supplementation with high-nutrient density diets could reduce crude protein and crude fat in pigs. Conversely, nutrient digestibility has been previously shown to decrease due to low dietary nutrient density (Massuquetto et al., 2020; Ahmadi-Sefat et al., 2022). Similarly, Ko et al. (2023) reported that the inclusion of nutrient-dense diets reduced crude fat, crude protein and amino acid digestibility in broilers.

Emulsifiers generally improve the utilisation of fat-soluble nutrients and digestion of dietary fat, thereby increasing nutrient density (Sun and Kim, 2019). Previous research has demonstrated that supplementing pigs with emulsifiers increased their fat absorption capacity (Zhao et al., 2015), with similar effects observed in broilers (Upadhaya et al., 2018). Zhao (2015) found that adding an emulsifier to the diet improved fat digestibility, effectively increasing the apparent nutrient density of the diets (Solbi et al., 2021). In the present study, the inclusion of emulsifiers in HD significantly improved ATTD and AID of dry matter, nitrogen, crude protein, crude fat, and amino acids. The results are consistent with the findings of Zhao et al. (2015), who reported that diets supplemented with emulsifiers significantly improved the ATTD of dry matter and nitrogen compared to control diets. Similarly, Sun and Kim (2019) demonstrated that the inclusion of an emulsifier to the diet increased the digestibility of dry matter and crude fat in pigs compared to low-nutrient-density diets. Ko et al. (2023) observed a significant increase in the ATTD and AID values of dry matter, crude protein, crude fat, and both essential and non-essential amino acids in chickens fed diets with emulsifiers. Kaczmarek et al. (2015) found that emulsifier addition improved the digestibility of dietary fat and oil in both basal diets and low-energy diets in a broiler feeding trial. Xing et al. (2004) showed that dietary supplementation of emulsifiers increased the nutrient digestibility of dry matter, crude fat, crude protein, and energy in pigs. Previously, Muniyappan et al. (2022) also reported a significant increase in the ATTD and AID of dry matter, crude protein and energy in pigs fed a fermented soybean diet supplemented with coconut oil. Øverland et al. (1993) observed a significant increase in the ATTD of dry matter, energy, crude fat, and crude protein with the inclusion of soybean oil. In contrast, Soares and Lopez-Bote (2002) found no significant difference in the digestibility of dry matter, nitrogen, and energy in pigs fed diets supplemented with emulsifiers (lysophospholipids). According to Cera et al. (1989) and Li et al. (1990), vegetable oils such as coconut oil, maize oil, and soybean oil are more easily digested than animal fats like tallow. This disparity may explain why some earlier studies failed to observe significant effects of emulsifier supplementation. The addition of sodium stearoyl-2-lactylate and Tween can enhance the emulsifying capacity of bile salts and enzymes during the emulsification process *in vivo*, positively affecting digestion, contributing to improved nutrient digestibility. These findings support the incorporation of emulsifiers into low-energy pig diets, offering the potential to reduce feed costs and increase profitability for producers.

In the current study, the supplementation of emulsifier to the diet improved the ATTD and AID of amino acids in weaned pigs. Previous studies have demonstrated beneficial effects on the ATTD and AID of amino acids with the inclusion of ingredients such as soybean meal, fermented soybean meal, fermented soybean meal with coconut oil, maize, distiller's dried grains with solubles or rapeseed meal. Kim et al. (2017) reported a significantly increase indispensable amino acids and dispensable amino acids in weaned pigs fed diets supplemented with soybean meal and distiller's dried grains with solubles. Muniyappan et al. (2022) reported a significant increase in the AID of lysine, arginine, tryptophan, valine, isoleucine, glutamic acid, proline, and cysteine of weaned pigs fed a diet supplemented with fermented soybean meal and coconut oil. In broilers feeding trial, Ko et al. (2023) found that dietary supplementation of an emulsifier increased the ATTD and AID of both essential and non-essential amino acids. An *in vitro* experiment of Martín et al. (2007) demonstrated that bile acids destabilised protein structure, leading to increased protein digestion. Similarly, Casterlain and Genot (1994) showed that when bovine serum albumin was adsorbed on the oil/water interface, its tryptic residue became hydrophobic. These findings indicate that the structural modifications of protein complexes, mediated by the emulsification effect, affect the digestion of hydrophobic amino acids (Casterlain and Genot, 1994). The ATTD and AID values obtained in the present study suggest that protein consumption, rather than fat intake, provides the energy necessary for the growth and maintenance of weaned pigs in the emulsifier-supplemented group. In addition to previous studies demonstrating how emulsifier supplementation can counteract high nutrient density by improving fat digestion efficiency, this study further shows that emulsifier supplementation plays a significant role in energy metabolism, reducing the reliance on proteins and amino acids for energy.

In digestion studies, urine analysis is used to identify metabolite profiles, the estimation of gross energy (GE) and nitrogen (N) being the most commonly measured parameters. This is important because N excretion pathways can be affected by variations in the content of urine acidifying substances and dietary fibre (Shriver et al., 2003). As noted by Pedersen et al. (2007) urine acidification is a standard practice in dietary supplementation during nutrient balance experiments, preventing N losses due to microbial growth. Here, the supplementation of emulsifiers to the diet improved urinary nitrogen content compared to animals fed nutrient-dense diets alone. Similarly, Muniyappan et al. (2022) reported a significant increase in urinary nitrogen content in pigs fed a diet supplemented with fermented soybean meal and coconut oil. Ribeiro et al. (2001) observed higher N content in faeces of broilers administered nutrient-dense diets. No interaction was observed between emulsifiers and diets with different nutrient densities in any of the measured parameters, indicating no additive effects.

Conclusions

The present experiments indicate that the apparent total tract digestibility and apparent ileal digestibility of dry matter, nitrogen, crude protein, crude fat and amino acids were improved by emulsifiers compared to nutrient-dense diets alone. However, dietary supplementation of emulsifiers also led to increased urinary nitrogen content. Additionally, a high-nutrient density diet resulted in elevated levels of crude protein and amino acids compared to a low-nutrient density diet.

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Conflict of interest

The Authors declare that there is no conflict of interest.

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