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Various levels of rapeseed meal in weaning pig diets from weaning to finishing periods

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Objective: This experiment was conducted to investigate the influence of rapeseed meal (RSM) supplementation in weaning pig diet on growth performance, blood profile, carcass characteristics and economic analysis on weaning to finishing pigs.
Methods: A total of 120 cross bred ([Yorkshire×Landrace]×Duroc) weaning pigs were allotted to 5 treatments in a randomized complete block design. Each treatment had 4 replications with 6 pigs per pen. Five different levels of RSM (0%, 2%, 4%, 6%, and 8%) were used as dietary treatments.
Results: Overall, no treatment showed significant differences in growth performance with increased dietary RSM levels. The concentration of blood urea nitrogen (BUN) decreased as dietary RSM levels increased in 6 weeks (linear response, $p<0.01$). Total cholesterol, high density lipoprotein cholesterol, low density lipoprotein cholesterol, triiodothyronine, and thyroxine showed no significant differences, neither were there any significant differences in the immune response (IgG and IgA). As the dietary RSM levels of weaning pig diet were increased, no differences were found among dietary treatments upon performing proximate analyses of the pork after finishing. The influence of RSM supplementation on nutrient digestibility and nitrogen retention were not affected by dietary RSM levels either. With increased dietary RSM levels in the weaning pig diet, no differences among dietary treatments were found after performing proximate analyses of the pork's physiochemical properties. In addition, there were no significant differences observed in pork colors, pH levels, and economic benefits.
Conclusion: Consequently, this experiment demonstrated that weaning pig's diet containing RSM influenced BUN concentration, but there were no detrimental effects on the growth performance of weaning pigs with up to 8% RSM in the diet.

Keywords: Rapeseed Meal; Weaning Pigs; Growth Performance; Blood Profiles; Carcass Characteristics

INTRODUCTION

Soybean meal (SBM) is widely used due to good amino acid balance and digestibility [1]. In a European context where potential of forage soybeans cannot be economically produced, there are many alternatives to SBM for protein source in animal feed [2]. However, amino acid availability and profile must be appropriate for animals. Rapeseed meal (RSM) is a by-product of oil extraction from rapeseed and contains 33% to 35% protein, 10% crude fiber but energy is not high [3]. In addition, canola which is acultivar of rapeseed was bred through standard plant breeding techniques to have low levels of erucic acid (<2%) and glucosinolates (<30 μmol/g) [4]. Although lysine content of RSM is lower than SBM, sulphur containing amino acids such as cysteine and methionine are much higher. *Brassica* plants such as RSM use sulphur to synthesise Gls and phytoalexins [5]. Cysteine could reduce sulphur for glucosinolates biosynthesis and for the synthesis of phytoalexins including camalexin [6]. RSM has been used in growing-finishing pig diets because of glucosinolates, which causes thyroid hypertrophy in young animals [7]. When insect herbivory

or tissue damage brings glucosinolates and myrosinase together hydrolysis of glucosinolates into thiocyanates, isothiocyanates, nitriles, oxazolidine-2-thiones and epithionitriles is facilitated [8]. Erucic acid appears to have toxic effects on the heart at high enough doses, an association with the consumption of rapeseed oil. In addition, RSM in the diets could decrease feed consumption because of high content of erucic acid and the bitter taste of sinapine [8]. In previous studies, most of the experiments were carried out on weaning periods, growing and finishing periods separately. Although supplementation of RSM in weaning pig diets has negative effects, there are no evidence regarding how long these effects will continue.

Therefore, the objective of the present study was to evaluate the effect of dietary supplementation levels of RSM on growth performance, blood profiles, carcass characteristics and economic analysis in weaning to finishing pigs.

MATERIALS AND METHODS

All experimental procedures involving animals were conducted in accordance with the Animal Experimental Guidelines provided by the Seoul National University Institutional Animal Care and Use Committee (SNUIACUC;SNU-161004-1).

Animal management and housing

A total of 120 weaning pigs ([Yorkshire×Landrace]×Duroc), 7.28±0.86 kg initial body weight (BW), were used in a 19-week feeding trial at experimental farm of Seoul National University. Pigs were allotted to one of five treatments by BW and sex in 4 replications with 6 pigs per pen in a randomized complete block (RCB) design. All pigs were housed in an environmentally controlled building with plastic-slotted floor facility (1.95 by 1.42 m^2) during weaning periods and fully-concrete floor facility (2.60 by 2.84 m^2) during growing to finishing periods. Each pen was equipped with a feeder and a nipple drinker to provide *ad-libitum* access. The BW and feed intake were recorded at 0, 3, 6, 10, 14, 17, and 19 week to calculate the average daily gain (ADG), average daily feed intake (ADFI) and gain to feed ratio (G:F ratio).

Experimental diets

Dietary treatments were: i) CON: corn-SBM based diet, ii) RSM2: basal diet+2% rapeseed meal, iii) RSM4: basal diet+4% rapeseed meal, iv) RSM6: basal diet+6% rapeseed meal, v) RSM8: basal diet+8% rapeseed meal. After weaning period, all pigs were fed same commercial diet but in a conducted phase feeding method (early growing, lated growing: 8 weeks) (early finishing, late finishing: 5 weeks). All nutrients met or exceeded the requirement of NRC [9]. In addition, experimental diets formula and chemical compositions were presented in Table 1 and 2.

Blood sampling and analysis

Blood samples were taken in three times (0, 3, 6 weeks) from anterior vena cava of 6 pigs in each treatment for measuring blood urea nitrogen (BUN), immunoglobulin (IgG, IgA), total cholesterol, low density lipoprotein (LDL) cholesterol, high density lipoprotein (HDL) cholesterol, triiodothyronine (T_3), and thyroxine (T_4). Collected blood samples were centrifuged for 15 min by 3,000 rpm at 4°C. Then, sera samples were aspirated by pipette and stored at –20°C until later analysis. The BUN concentration was analyzed using blood analyzer (Ciba-Corning model, Express Plus Ciba Corning Diagnostics Co., Medfield, MA, USA). IgG and IgA were determined by ELISA assay, according to the manufacture's guidelines (ELISA Stater Accessory Package, Pig IgG ELISA Quantitation Kit, Pig IgA ELISA Quantitation Kit; Bethyl, Montgomery, AL, USA). All samples were assayed in duplicates with 1:20,000 (IgG) or 1:10,000 (IgA) fold dilution. Total cholesterol, LDL cholesterol, HDL cholesterol were measured by enzymatic colorimetric assay (Modular analytics, PE model, Roche, Germany). T_3, T_4 were measured by ELISA assay (Modular analytics, PE model, Germany).

Glucosinolates (Gls) were extracted from RSM (Table 3) with 2 mL of boiling methanol solution (70% vol/vol) and 200 μL internal standard spike solution of glucotropaeolin (ChromaDex, Irvine, CA, USA) was added immediately [10] and extracted Gls were purified on aDEAE Sephadex A-25 anion exchange column (St. Louis, MO, USA). Three types of Gls in RSM were determined by using high performance liquid chromatography (HPLC; Sunnyvale, CA, USA). Desulfo-glucosinolates were separated using a Synergi Fusion-RP 80A (100×3 mm, 4 um, Phenomenex, CA, USA) with a flow rate of 1 mL/min. Glucosinolates (progoitrin, sinigrin, and gluconapin) were confirmed by a Finnigan LCQ Deca XP plus Ion Trap Mass Spectrometer system (Thermo Finnigan, CA, USA) which confirmed by LC-ESIMS in positive mode.

Erucic acid content in RSM (Table 3) was analyzed on a 7890 Agilent Gas Liquid Chromatograph (Agilent Technologies, Palo Alto, CA, USA) and equipped with flame ionization detector and the column was SP-2560 (i.d. 100 m×0.25 mm×0.20 μm film). Nitrogen was used as carrier gas, injector core temperature was 250°C, detector temperature was 260°C, and column temperature was programmed to begin at 170°C and then increase to 250°C then remained at 240°C for 40 min. Chromatography was calibrated with a mixture of 37 different fatty acids (FAME 37; Supelco Inc., Bellefonte, PA, USA) and the standard contained fatty acids ranging from C4:0 to C24:1n9 and samples were added 250 μL of internal standard spike solution (pentadecanoic acid; Sigma, St. Louis, MO, USA) by the method of AOAC [11].

Pork quality and carcass traits

In each treatment, four pigs were slaughtered for the carcass analysis. Longissimus muscles were used from nearby 10th rib on right side of carcass. Because of chilling procedure, 30 minutes after slaughter was regarded as initial time. The times that pH and pork color were measured was 0, 3, 6, 12, and 24 hours. The

Table 1. Formula and chemical compositions of the experimental diets in phase1 (0 to 3 weeks)

Items	Treatments[1]				
	Control	RSM2	RSM4	RSM6	RSM8
Ingredients (%)					
Corn	37.29	36.55	35.79	35.07	34.30
SBM	31.33	29.80	28.28	26.77	25.25
Barley	15.00	15.00	15.00	15.00	15.00
Rapeseed meal	0.00	2.00	4.00	6.00	8.00
Whey powder	4.00	4.00	4.00	4.00	4.00
Lactose	6.00	6.00	6.00	6.00	6.00
Soypeptide	1.81	1.81	1.81	1.81	1.81
Soy-oil	1.14	1.44	1.75	2.04	2.35
Mono-di calcium phosphorus	1.38	1.33	1.30	1.24	1.20
Limestone	1.02	1.02	1.01	1.01	1.01
L-lysine-HCl (78%)	0.28	0.29	0.30	0.30	0.31
DL-methionine (80%)	0.07	0.07	0.06	0.05	0.05
L-threonine (99%)	0.08	0.09	0.10	0.11	0.13
Vit. Mix[2]	0.10	0.10	0.10	0.10	0.10
Min. Mix[3]	0.10	0.10	0.10	0.10	0.10
Salt	0.30	0.30	0.30	0.30	0.30
ZnO	0.10	0.10	0.10	0.10	0.10
Total	100.00	100.00	100.00	100.00	100.00
Chemical composition					
ME (kcal/kg)[4]	3,265.02	3,265.02	3,265.03	3,265.01	3,265.03
Crude protein (%)[5]	19.21	18.47	18.17	18.31	18.59
Crude fat[5]	2.67	2.60	2.88	3.18	3.55
Crude ash[5]	5.86	6.24	6.13	5.98	6.24
Lysine (%)[4]	1.35	1.35	1.35	1.35	1.35
Methionine (%)[4]	0.35	0.35	0.35	0.35	0.35
Calcium (%)[4]	0.80	0.80	0.80	0.80	0.80
Phosphorus (%)[4]	0.65	0.65	0.65	0.65	0.65

RSM, rapeseed meal; SBM, soybean meal; ME, metabolizable energy.
[1] Con, corn-SBM diet; RSM2, basal diet+RSM2%; RSM4, basal diet+RSM4%; RSM6, basal diet+RSM6%; RSM8, basal diet+RSM8%.
[2] Provided the following quantities of vitamins per kg of complete diet: vitamin A, 8,000 IU; vitamin D_3, 1,800 IU; vitamin E, 60 IU; thiamine, 2 mg; riboflavin, 7 mg; calcium pantothenic acid, 25 mg; niacin, 27 mg; pyridoxine, 3 mg; biotin, 0.2 mg; folic acid, 1 mg; vitamin B_{12}, 0.03 mg.
[3] Provided the following quantities of minerals per kg of complete diet: Se, 0.3 mg; I, 1 mg; Mn, 51.6 mg; $CuSO_4$, 105 mg; Fe, 150 mg; Zn, 72 mg; Co, 0.5 mg.
[4] Calculated value. [5] Analyzed value.

pH was determined by pH meter (Model 720, Thermo Orion, Fullerton, CA, USA) and pork color was determined by Commission Internationale de l'Eclairage color L*, a* and b* value using a CR300 (Minolta Camera Co., Osaka, Japan). Proximate of pork samples were analyzed by the method of AOAC [11].

A centrifuge method was used for water holding capacity of pork [12]. Longissimus muscle samples were grounded and sampled in filter tube, and heated in water bath at 80°C for 20 min and centrifuged for 10 min at 2,000 rpm and 10°C (Eppendorf centrifuge 5810R, Hamburg, Germany). Then, the longissimus muscles were weight, packed in a polyethylene bag, heated in water bath until core temperature reached 72°C and weighed again after cooking to calculate the cooking loss. After heated, samples were cored (0.5×1.0×1.5 cm) parallel to muscle fiber and the cores were used to measure the shear force using a Warner Bratzler Shear (Norwood, MA, USA). Cooking loss, shear force, and water holding capacity of pork were analyzed by animal origin food science, Seoul National University.

Metabolic trial

A total of 15 pigs ([Yorkshire×Landrace]×Duroc) with an initial BW 13.41±0.32 kg were allotted to five treatments in a completely randomized design. Each pig was housed in an individual metabolic crate (82 by 40 cm^2) in a room of constant temperature (27°C), controlled with a heating lamp. The experimental diets were supplied twice a day at 08:00 h and 20:00 h according to the rate of 2.0 times of the maintenance requirement for metabolizable energy (ME) (106 kcal of ME/kg of $BW^{0.75}$) based on initial BW of pigs. Water was provided *ad libitum*. After 5 days adaptation period, pigs were subjected to 5 days sample collection and 0.5% of chromic oxide and 0.5% of ferric oxide were used as initial and end marker, respectively. The collected urine was strained through glass wool to remove foreign objects and then 10% sulfuric acid (50 mL) was added to prevent volatilization of ammonia. Collected feces and urine were stored at –20°C during the collection period. At the end of trial the feces were dried (65°C, 72 h) and ground (2 mm screen, Wiley mill) for chemical analysis.

Table 2. Formula and chemical compositions of the experimental diets in phase1 (4 to 6 weeks)

Items	Treatments[1]				
	Control	RSM2	RSM4	RSM6	RSM8
Ingredients (%)					
Corn	47.25	46.25	45.79	45.03	44.31
SBM	27.08	25.54	24.03	22.51	20.98
Barley	15.00	15.00	15.00	15.00	15.00
Rapeseed meal	0.00	2.00	4.00	6.00	8.00
Whey powder	2.00	2.00	2.00	2.00	2.00
Lactose	3.00	3.00	3.00	3.00	3.00
Soypeptide	1.81	1.81	1.81	1.81	1.81
Soy-oil	1.01	1.30	1.60	1.91	2.20
Mono-di calcium phosphorus	1.20	1.15	1.10	1.05	1.00
Limestone	0.89	0.89	0.88	0.89	0.88
L-lysine-HCl (78%)	0.18	0.19	0.19	0.20	0.21
DL-methionine (80%)	0.03	0.03	0.02	0.01	0.01
L-threonine (99%)	0.01	0.02	0.03	0.05	0.06
Vit. Mix[2]	0.10	0.10	0.10	0.10	0.10
Min. Mix[3]	0.10	0.10	0.10	0.10	0.10
Salt	0.30	0.30	0.30	0.30	0.30
ZnO	0.05	0.05	0.05	0.05	0.05
Total	100.00	100.00	100.00	100.00	100.00
Chemical composition					
ME (kcal/kg)[4]	3,265.04	3,265.05	3,265.02	3,265.01	3,265.00
Crude protein (%)[5]	17.64	17.91	17.22	17.77	18.88
Crude fat[5]	3.18	2.57	2.89	3.02	3.40
Crude ash[5]	6.01	5.65	5.26	4.94	5.44
Lysine (%)[4]	1.15	1.15	1.15	1.15	1.15
Methionine (%)[4]	0.30	0.30	0.30	0.30	0.30
Calcium (%)[4]	0.70	0.70	0.70	0.70	0.70
Phosphorus (%)[4]	0.60	0.60	0.60	0.60	0.60

RSM, rapeseed meal; SBM, soybean meal; ME, metabolizable energy.
[1] Con, corn-SBM diet; RSM2, basal diet+RSM2%; RSM4, basal diet+RSM4%; RSM6, basal diet+RSM6%; RSM8, basal diet+RSM8%.
[2] Provided the following quantities of vitamins per kg of complete diet: vitamin A, 8,000 IU; vitamin D3, 1,800 IU; vitamin E, 60 IU; thiamine, 2 mg; riboflavin, 7 mg; calcium pantothenic acid, 25 mg; niacin, 27 mg; pyridoxine, 3 mg; biotin, 0.2 mg; folic acid, 1 mg; vitamin B12, 0.03 mg.
[3] Provided the following quantities of minerals per kg of complete diet: Se, 0.3 mg; I, 1 mg; Mn, 51.6 mg; CuSO4, 105 mg; Fe, 150 mg; Zn, 72 mg; Co, 0.5 mg.
[4] Calculated value.
[5] Analyzed value.

Chemical analyses

Diets were ground by a Cyclotec 1093 Sample Mill (Foss Tecator, Hillerod, Denmark) and ground diets were analyzed. All analyses were performed in duplicate samples and analyses were repeated if results from duplicate samples varied more than 5% from the mean. The dry matter of diet samples were determined by oven drying at 135°C for 2 h (method 930.15; AOAC) [11]. Aspartic acid was used as a calibration standard, and crude protein (CP) was calculated as N×6.25 and diets were also analyzed for ash (method 942.05; AOAC) [11]. Crude fat was hydrolized in HCl solution to release bound fat and then extracted with diethyl ether and petroleum ether (method 954.02; AOAC) [11]. Collected excreta were pooled and dried in an air-forced drying oven at 60°C for 72 h, and ground into 1 mm particles in a Wiley mill for chemical analysis include moisture, protein, fat and ash contents AOAC [11]. Total urine was collected daily in a plastic container containing 50 mL of 4 N H_2SO_4 and frozen during the 5 day collection period for nitrogen retention analyses.

Economic analysis

Economic analysis was calculated by feed cost and feed efficiency (G:F ratio). The total feed cost (Won) per BW gain (kg) was calculated using total feed intake and feed price. The feed cost per weight gain was calculated based on price of raw materials during the time of the experiment. The days to market weight (115 kg) were estimated from the BW at the end of feeding trial and ADG of 19 weeks.

Statistical analysis

The experimental data was analyzed as a RCB design using the general linear model procedure of SAS. For data on growth performance and economic analysis a pen was considered as an experimental unit, while individual pig was used as the unit for data on blood profile, immunological analysis, diarrhea incidence,

Table 3. Anti-nutritional factors and chemical contents of rapeseed meal (as dry matter basis)

Items	Rapeseed meal			
Glucosinolates (μmole/g)				
Progoitrin	0.32			
Sinigrin	8.12			
Gluconapin	29.52			
Total glucosinolates	37.97			
Erucic acid (mg/g)	7.37			
Crude protein (%)	32.80			
Crude fat (%)	1.03			
Crude ash (%)	8.57			
	RSM2[1)]	RSM4	RSM6	RSM8
Glucosinolates in diet (μmol/g)[2),3)]	0.76	1.52	2.28	3.04
Daily glucosinolates intake (μmol/g)[2),3)]	568.48	1,019.92	1,527.60	2,048.96
Erucic acid in diet (mg/g)[2),3)]	0.15	0.29	0.44	0.59
Daily erucic acid intake (mg/g)[2),3)]	112.2	194.59	294.8	397.66

[1)] Con, corn-SBM diet; RSM2, basal diet+RSM2%; RSM4, basal diet+RSM4%; RSM6, basal diet+RSM6%; RSM8, basal diet+RSM8%.
[2)] Glucosinolates content in the diets was equivalent to 0.76, 1.52, 2.28, 3.04 μmol/g for 2%, 4%, 6%, 8% of RSM supplementation respectively. Eucic acid content in the diets was equivalent to 0.15, 0.29, 0.44, 0.59 mg/g for 2%, 4%, 6%, 8% of RSM supplementation respectively.
[3)] Calculated value.

pork quality. Linear and quadratic effects for equally spaced treatments were assessed by measurement of orthogonal polynomial contrast. The differences were declared significant at $p<0.05$ or highly significant at $p<0.01$.

RESULTS

Table 4 and Table 5 show the influence of RSM supplementation on growth performance in weaning pigs and growing to finishing pig period. Overall there was no significant differences in growth performance. In addition, Total cholesterol, HDL cholesterol, LDL cholesterol, T_3 and T_4 were not affected by dietary RSM supplementation in weaning pig diets (Table 6). However, the BUN concentration decreased as dietary RSM level increased in 6 week (linear response, $p<0.01$). Table 7 showed the influence of RSM supplementation on immune response in weaning pigs. There were no significant differences in immune response (IgG and IgA). Influence of RSM supplementation on nutrient digestibility and nitrogen retention were not affected by the dietary RSM level increased (Table 8). As dietary RSM level of weaning pig diet increased, there were no differences in proximate analysis and physiochemical property of the pork after finishing among dietary treatments (Table 9). In addition, there were no significant difference in pork colors L*, a*, b* value (Table 10) and pH at 0, 3, 6, 12, 24 h after finishing (Table 11). In economic benefit, any significant difference was not examined (Table 12) but compared to control treatment, supplemented RSM 2% in weaning pig diet had the greatest economical benefits.

DISCUSSION

In the current study, growth performance between weaning to finishing were not affected by dietary supplementation of RSM of up to 8% in the weaning pig diet. RSM levels in pig diet can be used to replace SBM, and were limited to 5% in young pigs. RSM contain anti-nutrition factors such as Gls and erucic acid. Gls appeared to have negative effects on feed intake due to its high content of progoitrin, which are associated with bitterness [2]. Gill et al [13] reported that 3-week-old weaning pigs were

Table 4. Influence of various rapeseed meal levels in weaning pig diet on growth performance in weaning pigs[1)]

Criteria	Treatment[2)]					SEM	p-value	
	CON	RSM 2	RSM 4	RSM 6	RSM 8		Lin.	Quad.
Body weight (kg)								
Initial	7.28	7.28	7.28	7.28	7.28	0.188	-	-
3 wk	10.75	11.04	10.68	10.55	10.24	0.333	0.27	0.98
6 wk	22.18	22.90	22.40	21.91	21.41	0.566	0.20	0.93
ADG (g)								
0 to 3 wk	165	178	163	155	142	9.0	0.28	0.97
4 to 6 wk	545	565	558	541	532	11.9	0.19	0.90
0 to 6 wk	355	372	361	348	337	9.9	0.20	0.93
ADFI (g)								
0 to 3 wk	305	314	309	293	290	11.8	0.42	0.92
4 to 6 wk	1,042	1,181	1,032	1,047	1,059	33.4	0.21	0.16
0 to 6 wk	673	748	671	670	674	21.8	0.23	0.26
G:F ratio								
0 to 3 wk	0.540	0.562	0.528	0.530	0.471	0.0147	0.16	0.66
4 to 6 wk	0.524	0.479	0.541	0.525	0.508	0.0089	0.50	0.10
0 to 6 wk	0.528	0.497	0.538	0.526	0.501	0.0072	0.96	0.10

SEM, standard error of the mean; Lin, linear; Quad, quadratic; ADG, average daily gain; ADFI, average daily feed intake; G:F, gain to feed.
[1)] A total 120 crossbred pigs was fed from average initial body 7.28 kg and the average final body weight was 22.16 kg.
[2)] Con, corn-SBM diet; RSM2, basal diet+RSM2%; RSM4, basal diet+RSM4%; RSM6, basal diet+RSM6%; RSM8, basal diet+RSM8%.

Table 5. Influence of various rapeseed meal levels in weaning pig diet on growth performance in growing-finishing pigs

Criteria	Treatment[1)]					SEM	p-value	
	CON	RSM2	RSM4	RSM6	RSM8		Lin.	Quad.
Body weight (kg)								
6 wk	22.18	22.90	22.40	21.91	21.41	0.566	0.20	0.93
10 wk	43.32	45.02	44.43	41.81	42.63	0.822	0.18	0.52
14 wk	67.20	72.75	69.42	65.62	67.59	1.415	0.13	0.18
17 wk	90.23	96.26	93.06	89.11	91.42	1.546	0.17	0.20
19 wk	106.63	111.85	109.42	105.37	108.27	1.567	0.28	0.22
ADG (g)								
7 to 10 wk	755	790	787	716	753	12.9	0.20	0.26
11 to 14 wk	853	990	892	789	891	31.0	0.28	0.11
7 to 14 wk	803	890	840	780	822	17.0	0.12	0.10
15 to 17 wk	1,097	1,119	1,126	1,110	1,127	17.9	0.93	0.77
17 to 19 wk	1,171	1,113	1,169	1,161	1,205	20.4	0.27	0.89
15 to 19 wk	1,127	1,117	1,143	1,131	1,162	14.4	0.52	0.84
ADFI (g)								
7 to 10 wk	1,778	1,744	1,711	1,738	1,679	31.9	0.72	0.57
11 to 14 wk	2,357	2,373	2,408	2,365	2,376	67.7	0.94	0.99
7 to 14 wk	2,068	2,059	2,059	2,052	2,027	45.6	0.87	0.89
15 to 17 wk	2,999	2,273	2,990	3,058	2,883	86.0	0.81	0.35
17 to 19 wk	3,735	3,861	4,154	3,812	3,872	81.0	0.45	0.94
15 to 19 wk	3,294	3,231	3,456	3,359	3,279	76.6	0.90	0.50
G:F ratio								
7 to 10 wk	0.425	0.456	0.461	0.412	0.449	0.0072	0.29	0.08
11 to 14 wk	0.368	0.418	0.375	0.331	0.376	0.0117	0.25	0.07
7 to 14 wk	0.392	0.433	0.410	0.382	0.406	0.0007	0.13	0.05
15 to 17 wk	0.373	0.403	0.385	0.371	0.391	0.0110	0.70	0.37
17 to 19 wk	0.316	0.290	0.284	0.304	0.311	0.0059	0.11	0.89
15 to 19 wk	0.350	0.348	0.335	0.339	0.354	0.0072	0.65	0.49

SEM, standard error of mean; Lin, linear; Quad, quadratic; ADG, average daily gain; ADFI, average daily feed intake; G:F, gain to feed.
[1)] Commercial diet was fed in growing-finishing periods.

particularly sensitive to RSM and showed reductions in feed intake in their first week. In addition, pigs (weighing 23 to 45 kg) fed with 5% to 10% of RSM showed a 17% reduction in daily gain and 7% less feed intake [3]. Hanne [14] recommended the use of up to 15% RSM (glucosinolates content 23 μmol/g) in weaning pig diet without negatively affecting productivity. This is due to the low content of 4-hydroxy-glucobrassicin in RSM, which are the molecules responsible for negatively affecting the productivity of pigs, and the presence of which indicates damage by heat-treatment and the disintegration of glucosinolates. Generally, RSM is not used in weaning pig diets up to 20 kg bodyweight because RSM has a negative effect on weaning pig performance. It was also documented that increasing supplementation of RSM linearly reduced ADG and ADFI [15]. The typical tolerance level for glucosinolates was 2.0 μmol/g for growing pigs, while the maximum tolerable level for weaning pigs remains to be proven [14]. Inclusion of up to 25% solvent-extracted canola meal (glucosinolates contents 4 to 9.5 mol/g) in diets fed to weaning pigs (6 to 23 kg BW) did not affect ADG or voluntary feed intake [15]; supplemented canola meal (glucosinolates contents 10 to 22 mol/g) of up to 15% to 20% in weaning pig diets also did not result in negative effects on ADG, ADFI, and G:F [16]. The present study supports previous evidence indicating that RSM supplementation of up to 8% in weaning pig diets do not affect growth performance for the duration of weaning to finishing.

RSM has more sulfur containing amino acids such as methionine and cysteine than SBM. Although SBM contains more lysine than RSM, its amino acid balance compares well with SBM. In contrast, the digestibility of RSM containing diets cannot be well compared with SBM [17]. Nutrient digestibility is influenced by a number of factors including the fiber from rapeseed hulls, anti-nutritional factors (tannin, sinapine, erucic acid, and glucosinolates) and dietary formulation [17]. In the present study, BUN concentration decreased as dietary RSM levels increased. This result supports that of Fenwick and Curtis [18] in which a combination of SBM and RSM shows some clear advantages. Proteins from RSM are less digestible than those of SBM (72% vs 88%), but the amino acid balance is better than in a SBM (for the sulfur amino acids) [19]. Clandinin et al [5] suggested that lysine is sensitive to excessive heat, which leads to undesirable reactions and reduced availability of amino acids. Any reduction in lysine availability will seriously affect the competitive position of RSM for monogastric use. Upon supplementation of RSM in animal diet, minimum heat is necessary to inactivate enzymes (especially

Table 6. Influence of various rapeseed meal levels in weaning pig diet on blood profiles in weaning pigs[1)]

Criteria	Treatment[2)]					SEM	p-value	
	CON	RSM2	RSM4	RSM6	RSM8		Lin.	Quad.
BUN (mg/dL)								
Initial	9.90	9.50	11.70	12.50	13.50	0.283	-	-
3 wk	14.35^{A}	13.08^{A}	11.80^{AB}	12.68^{A}	9.10^{B}	0.632	0.15	0.11
6 wk	10.21^{a}	11.08^{a}	9.55^{ab}	7.47^{bc}	7.00^{c}	0.443	<0.01	0.63
Total cholesterol (mg/dL)								
Initial	139.00	163.00	131.00	105.00	200.00	5.979	-	-
3 wk	59.33	60.17	68.67	60.50	66.83	2.007	0.77	0.76
6 wk	82.17	80.67	86.83	73.71	85.17	1.941	0.76	0.33
LDL cholesterol (mg/dL)								
Initial	86.00	167.00	109.00	150.00	67.00	7.025	-	-
3 wk	29.17	31.00	33.33	31.50	33.83	2.394	0.85	0.68
6 wk	45.67	44.17	45.67	40.00	43.50	3.790	0.88	0.71
HDL cholesterol (mg/dL)								
Initial	51.00	61.00	55.00	54.00	51.00	0.683	-	-
3 wk	23.00	23.33	27.50	22.33	28.00	0.941	0.46	0.33
6 wk	28.83	29.67	32.67	31.33	33.33	0.767	0.55	0.79
T3 (ng/mL)								
Initial	0.11	0.08	0.20	0.15	0.34	0.017	-	-
3 wk	0.14	0.11	0.24	0.25	0.16	0.027	0.96	0.14
6 wk	0.14	0.25	0.29	0.15	0.23	0.032	0.40	0.42
T4 (μg/dL)								
Initial	25.05	31.43	30.07	24.06	20.35	0.756	-	-
3 wk	23.97	21.60	22.09	23.28	20.76	1.290	0.99	0.53
6 wk	19.91	23.33	21.12	23.18	16.57	1.220	0.11	0.48

SEM, standard error of mean; Lin, linear; Quad, quadratic; BUN, blood urea nitrogen; LDL, low density lipoprotein; HDL, high density lipoprotein; T_3, triiodothyronine; T_4, thyroxine.
[1)] Least squares means for 6 pigs per treatment.
[2)] Con, corn-SBM diet; RSM2, basal+RSM2%; RSM4, basal diet+RSM4%; RSM6, basal diet+RSM6%; RSM8, basal diet+RSM8%.
[AB] Means in a same row with different superscript letters significantly differ ($p<0.05$).
[abc] Means in a same row with different superscript letters significantly differ ($p<0.01$).

myrosinase) and to avoid denaturation of proteins [5]. In this study, RSM supplementation of up to 8% in the weaning pig diet had no negative effect on BUN concentration.

The hydrolysis product of glucosinolates is known to depress iodine metabolism in the thyroid gland and inhibit the synthesis of thyroid hormones T_3 and T_4 [20]. When these compounds, especially thiocynates, interfere with iodine uptake, hypothyroidism and enlargement of the thyroid gland ensue. In addition, these changes affect the metabolism in all tissues, including reproductive organs [21]. Maison [22] suggested that the dietary content of glucosinolates (9 to 10 μmol/g) induced iodine deficiency, hypothyroidism, reduced bone and serum zinc content and alkaline phosphatase activity. Consequently, RSM supplementation of up to 8% did not show any significant effect on T_3 and T_4 hormones (glucosinolates content 3.04 μmol/g).

The fatty acid composition of RSM consists of oleic (51%), lin-

Table 7. Influence of various rapeseed meal levels in weaning pig diet on immune response in weaning pigs[1)]

Criteria	Treatment[2)]					SEM	p-value	
	CON	RSM2	RSM4	RSM6	RSM8		Lin	Quad
Serum IgG (mg/mL)								
Initial	2.44	2.70	2.51	2.46	2.33	0.022	-	-
3 wk	2.67	2.50	1.97	2.37	2.54	0.088	0.23	0.32
6 wk	2.78	3.23	2.73	3.38	3.25	0.180	0.94	0.65
Serum IgA (mg/mL)								
Initial	1.76	1.30	1.90	1.22	1.35	0.050	-	-
3 wk	3.50	3.36	3.43	3.80	4.16	0.242	0.37	0.82
6 wk	0.56	0.57	0.54	0.55	0.51	0.036	0.77	0.93

SEM, standard error of mean; Lin, linear; Quad, quadratic; Ig, immunoglobulin.
[1)] Least squares means for six pigs per treatment.
[2)] Con, corn-SBM diet; RSM2, basal dietA+RSM2%; RSM4, basal diet+RSM4%; RSM6, basal diet+RSM6%; RSM8, basal diet+ RSM8%.

Table 8. Influence of various rapeseed meal levels in weaning pig diet on nutrient digestibility in weaning pigs[1)]

Criteria	Treatment[2)]					SEM	p-value	
	CON	RSM2	RSM4	RSM6	RSM8		Lin	Quad
Nutrient digestibility (%)								
Dry matter	91.28	89.94	90.02	88.66	89.79	0.727	0.88	0.91
Crude protein	88.97	86.73	85.98	84.91	87.61	1.026	0.59	0.71
Crude ash	73.82	72.99	71.91	68.73	70.59	2.113	0.87	0.85
Crude fat	86.80	76.39	81.50	83.77	86.13	2.182	0.12	0.54
Nitrogen retention (g/d)								
N intake	20.14	19.93	19.58	19.03	19.57	0.197	0.85	0.22
Fecal N	2.22	2.63	2.72	2.88	2.42	0.195	0.60	0.76
Urinary N	3.75	3.20	3.15	4.17	3.57	0.196	0.31	0.29
N retention[3)]	14.17	14.10	13.71	11.97	13.58	0.329	0.79	0.19

SEM, standard error of mean; Lin, linear; Quad, quadratic.
[1)] Least squares means for three pigs per treatment. Initial BW: 13.22 kg.
[2)] Con, corn-SBM diet; RSM2, basal diet+RSM2%; RSM4, basal diet+RSM4%; RSM6, basal diet+RSM6%; RSM8, basal diet+RSM8%.
[3)] N retention = N intake – Fecal N – Urinary N.

Table 9. Influence of various rapeseed meal levels in weaning pig diet on pork quality of longissimus muscle[1)]

Criteria	Treatment[2)]					SEM	p-value	
	CON	RSM2	RSM4	RSM6	RSM8		Lin	Quad
Proximate analysis (%)								
Moisture	74.31	74.31	73.52	73.70	73.65	0.131	0.57	0.53
Crude protein	24.64	24.41	23.72	23.51	24.11	0.134	0.72	0.12
Crude fat	1.68	1.39	2.12	1.87	1.46	0.150	0.81	0.21
Crude ash	1.17	1.34	1.20	1.25	1.17	0.032	0.17	0.51
Physiochemical property								
Cooking loss (%)	30.05	31.97	32.24	31.88	31.85	0.357	0.34	0.59
Shear force ($kg/0.5 \times 1.0 \times 1.5\ cm^2$)	7.64	7.44	5.89	6.71	6.71	0.275	0.89	0.59
WHC (%)	68.81	63.67	66.13	66.32	65.48	0.665	0.20	0.12

SEM, standard error of mean; Lin, linear; Quad, quadratic; WHC, water holding capacity.
[1)] Least squares means for three pigs per treatment.
[2)] Phase 2 diet was fed (Con, corn-SBM diet; RSM2, basal diet+RSM2%; RSM4, basal diet+RSM4%; RSM6, basal diet+RSM6%; RSM8, basal diet+RSM8%).

oleic (25%), and linolenic (14%) acids. When pigs are fed with an abundance of monounsaturated fatty acids, specifically oleic acid, these biomolecules can be exchanged for saturated fatty acids to increase serum HDL cholesterol levels without affecting those of LDL [23]. Kaneko [24] suggested that an increase in cholesterol concentrations is an indicator of hypothyroidism because of the important modulatory role of thyroid hormones in the intermediary metabolism of hypercholesterolemia, which is associated with increased serum LDL cholesterol concentrations. Consequently, total cholesterol, HDL, LDL, and thyroid hormone concentrations were not affected in any of our experiments.

IgA is the major antibody present in mucosal secretions, with many functional roles such as the prevention of bacteria and viruses from breaching the mucosal barrier [25]. IgG is generally

Table 10. Influence of various rapeseed meal levels in weaning pig diet on pork pH after slaughter[1)]

Time after slaughter (h)	Treatment[2)]					SEM	p-value	
	CON	RSM2	RSM4	RSM6	RSM8		Lin	Quad
0	5.53	5.88	5.90	5.66	5.71	0.056	0.10	0.39
3	5.44	5.73	5.54	5.59	5.42	0.049	0.08	0.74
6	5.46	5.63	5.56	5.49	5.46	0.043	0.23	0.69
12	5.70	5.61	5.61	5.63	5.57	0.026	0.99	0.40
24	5.79	5.68	5.69	5.68	5.63	0.028	0.95	0.36

SEM, standard error of mean; Lin, linear; Quad, quadratic.
[1)] Least squares means for four pigs per treatment.
[2)] Con, corn-SBM diet; RSM2, basal diet+RSM2%; RSM4, basal diet+RSM4%; RSM6, basal diet+RSM6%; RSM8, basal diet+RSM8%.

Table 11. Influence of various rapeseed meal levels in weaning pig diet on pork color after slaughter[1)]

Criteria	Treatment[2)]					SEM	p-value	
	CON	RSM2	RSM4	RSM6	RSM8		Lin	Quad
Hunter value, L[3)]								
0 h	40.26	38.78	40.28	40.40	38.92	0.391	0.91	0.06
3 h	39.45	39.13	40.57	41.54	38.02	0.521	0.40	0.08
6 h	42.06	40.25	43.84	42.83	41.26	0.634	0.96	0.15
12 h	43.69	42.03	43.21	44.09	43.46	0.418	0.33	0.31
24 h	45.52	44.53	45.02	46.39	45.97	0.437	0.37	0.60
Hunter value, a[4)]								
0 h	1.96	1.98	1.62	1.89	1.40	0.085	0.16	0.36
3 h	2.52	2.28	2.11	2.96	1.75	0.175	0.49	0.06
6 h	3.46	2.87	3.15	3.61	3.14	0.158	0.27	0.06
12 h	4.08	3.59	4.11	4.68	4.74	0.210	0.07	0.52
24 h	4.93	4.68	4.99	4.96	5.07	0.178	0.38	0.73
Hunter value, b[5)]								
0 h	4.20	4.24	4.08	3.88	4.74	0.078	0.97	0.28
3 h	4.03	4.08	4.24	4.73	3.77	0.112	0.27	0.05
6 h	5.31	4.74	5.51	5.44	5.20	0.148	0.53	0.20
12 h	5.65	5.13	5.74	5.95	6.18	0.164	0.09	0.61
24 h	6.21	6.29	6.68	6.78	6.86	0.165	0.58	0.91

SEM, standard error of mean; Lin, linear; Quad, quadratic.
[1)] Least squares means for four pigs per treatment.
[2)] Con, corn-SBM diet; RSM2, basal diet+RSM2%; RSM4, basal diet+RSM4%; RSM6, basal diet+RSM6%; RSM8, basal diet+ RSM8%.
[3)] L, luminance or brightness (vary from black to white). [4)] a, red · green component (+a = red, –a = green). [5)] b, yellow · blue component (+b = yellow, –b = blue).

considered to be the most common antibody in blood circulation, and plays important roles in controlling bacterial infections in the body; it can also function to control diarrheal infections by binding multiple pathogenic antigens [25]. Upon dietary exposure of animals to glucosinolates, negative effects such as low growth performance and impaired fertility were observed. Some

Table 12. Influence of various rapeseed meal levels in weaning pig diet on economic benefits[1)]

Criteria	Treatment2)					SEM	p-value	
	CON	RSM2	RSM4	RSM6	RSM8		Lin	Quad
Feed cost per weight gain (won/kg)								
0 to 3 wk	965	928	993	984	1,142	31.7	0.11	0.45
4 to 6 wk	885	972	859	899	928	16.6	0.59	0.08
7 to 10 wk	1,215	1,133	1,117	1,252	1,147	19.8	0.33	0.10
11 to 14 wk	1,394	1,220	1,371	1,592	1,349	51.0	0.41	0.06
15 to 17 wk	1,356	1,245	1,324	1,382	1,272	43.3	0.78	0.28
18 to 19 wk	1,532	1,666	1,705	1,588	1,545	31.5	0.10	0.97
0 to 19 wk	7,347	7,165	7,368	7,697	7,382	93.6	0.50	0.17
Total feed cost per pig (won/head)								
0 to 3 wk	3,298	3,414	3,371	3,201	3,176	128.0	0.45	0.90
4 to 6 wk	10,138	11,500	10,081	10,261	10,410	325.3	0.25	0.16
7 to 10 wk	25,595	22,205	24,625	25,019	24,160	858.1	0.42	0.30
11 to 14 wk	33,400	33,626	34,115	33,512	33,657	959.6	0.94	0.99
15 to 17 wk	31,242	29,287	31,145	31,847	30,033	896.2	0.81	0.35
18 to 19 wk	25,098	25,944	27,916	25,620	26,018	551.0	0.45	0.94
0 to 19 wk	128,771	125,976	131,252	129,460	127,455	2,836.6	0.98	0.62
Total feed cost per pig (won/head, reached 115 kg)	141,565	132,891	141,808	145,260	138,945	2,162.0	0.47	0.10
Days to market weight (reached 115 kg)	141	137	138	142	138	1.4	0.43	0.12

SEM, standard error of mean; Lin, linear; Quad, quadratic.
[1)] Least squares means of six observations per treatment.
[2)] Weaning periods (Con, corn-SBM diet; RSM2, basal diet+RSM2%; RSM4, basal diet+RSM4%; RSM6, basal diet+RSM6%; RSM8, basal diet+RSM8%). Growing-finishing periods was fed commercial diet.

breakdown products of glucosinolate, especially nitriles, might lead to mucosal irritation in the gastro-intestinal tract and transient impairment of liver and kidney functions [26]. Hydrolysis of glucosinolates can occur in the presence of isothiocyanates, which cause significant effects on the synthetic bio-fumigants of the intestinal mucosa [27]. In the current study, IgG and IgA were not affected by increased RSM in the weaning pig diet.

Sarwar et al [27] suggested that the protein content of hulls were highly indigestible. Compared with SBM, RSM have poorly digestible non-starch polysaccharides and oligosaccharides [28]. Notably, glucosinolates levels have presented a problem in situations where RSM is used as a dietary ingredient [8]. The digestibility of CP and amino acids in canola meal could vary depending on the age of pigs and the quality of proteins [29]. In addition, nutrient digestibility of RSM could be affected by many factors such as different genetic selections, environments, and the process of oil extraction [22]. In this study, no significant difference in nutrient digestibility were observed as dietary RSM levels increased.

In the present study, no differences were found among dietary treatments upon performing proximate analyses and physiochemical property studies of the pork after butchery. Growth performance of weaning pigs were influenced by feed nutrients, while the chemical composition of finishing pigs carcasses were not [30]. The water holding capacity (WHC) of the meat can increase with time after finishing due to the proteolytic action of cathepsins, which break down enzymes of the myofibrillar structure and influence physioelectrical charges. These changes increase the absorption of ions such as potassium, calcium and sodium [31], while maturation time affects meat tenderness. During the period after finishing, shear force may decrease due to proteolysis of the myofibrillar structural components. When WHC decreases, shear force was observed to increase [31].

RFN (reddish, firm, and non-exudative), pale, PSE (pale, soft, and exudative), and of DFD (dark, firm, and dry) meat are intimately related with pH. Time after maturation periods, pH values decreased. This result can explain about growth of lactic acid bacteria, which optimally grow at pH <6 [31]. Redness is closely associated with the state and amount of myoglobin in the meat. Low-pH conditions, it causes a denaturation of globin, leaving the heme function unprotected and accelerate oxidation of the metmyoglobin [31]. The increase in the time after finishing of the meat could accelerate darker and yellowness tends to increase over time [31]. In present experiment, there were no significant differences in pork color L*, a*, and b* value and pH at 0, 3, 6, 12, 24 h after slaughter.

The raw material price for the analysis of the experiment was based on the cost of the feed supply at the time of the experiment and was compared based on the feed price due to the raw material feed except for processing costs and labor costs of the experiment etc. Statistical analysis showed no significant differences between treatments but treatment RSM6 showed the highest numerical value in weight gain, feed cost, and days to market weight compared to other treatments and treatment RSM2 was 3% lower in weight gain and feed cost compared to control treatment. Also, days to market weight (reached 115 kg BW) reached 4 days earlier than control treatment. These results showed RSM in weaning pig diet was considered to be the most economical one when it was supplemented up to 2%.

IMPLICATIONS

Increasing RSM levels in weaning pig diets were not observed to affect growth performance, blood profiles, pork quality and carcass traits. Economic analysis showed that supplementation of RSM 2% was the most beneficial effect. Consequently, RSM could be used for weaning pig up to 8% without any detrimental effect on growth performance but highest economic profit was achieved in 8% of RSM treatment.

CONFLICT OF INTEREST

We certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

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