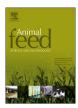
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## Growth performance of weaned pigs fed different levels of starfish meal



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#### ABSTRACT

Starfish meal (SM) was fed in different concentrations to 4952 piglets from 6 kg to assess the effect on animal performance when fed under commercial conditions. Performance was evaluated at low (L-SM), medium (M-SM) and high (H-SM) SM-levels compared with fishmeal (FM). The experimental period was divided into three two-week phases. Pigs received 5, 7.5 and 100 g/ kg SM, and 50 g/kg FM in phase 1. In phase 2, SM and FM were halved. In phase 3, all pigs received the same diet without SM to study compensatory growth. Similar ADG and ADFI was found for pigs fed FM and L-SM in all phases. The ADG of pigs was significantly lower when feeding diets with M-SM and H-SM compared with FM and L-SM in both phase 1 and 2 (P < 0.001), and the ADFI of L-SM-fed pigs was greater than for pigs fed M-SM and H-SM in phase 1 (P = 0.015), whereas in phase 2, the ADFI of pigs receiving M-SM and H-SM was significantly lower compared with the other two treatments (P < 0.001). In phase 3, pig growth was similar on all treatments. M-SM and H-SM gave compensatory growth in phase 3. Piglets can be fed 50 g/kg SM with good results, but greater inclusion levels may cause growth reduction.

#### 1. Introduction

A growing world population and rising incomes are expected to lead to an increased demand for protein sources in the coming decades (Van der Spiegel et al., 2013). Starfish meal (SM) can be a new protein source in animal feed. Aggregations of starfish are considered a pest in Denmark because they predate commercially grown mussels. Therefore, starfish are caught to prevent high losses in mussel yield.

Previous research has demonstrated the potential of feeding weaned pigs diets with SM in pilot-scale experimental settings (Nørgaard et al., 2015; Sørensen and Nørgaard, 2016). Chemical analyses have shown a high protein quality for SM with amino acid profiles comparable to fishmeal (FM) (Nørgaard et al., 2015). FM is frequently used in animal feed and is considered a high quality protein source (Ponce and Gernat, 2002). Furthermore, an experiment with a low number of pigs has proven the feasibility of using SM in weaned pigs (Sørensen and Nørgaard, 2016). Sørensen and Nørgaard (2016) found that growth rate was maintained when substituting 50 g FM/kg for 50 g SM/kg. Including SM at 100 g/kg, however, resulted in reduced growth. It was hypothesized that high calcium (Ca) levels in SM, generating a high Ca:P ratio, could have negatively affected phosphorus (P) digestibility and absorption when feeding 100 g/kg SM (Sørensen and Nørgaard, 2016). Correspondingly, in pigs fed 100 g/kg SM, serum levels were low for P and high for Ca compared with pigs fed FM and 50 g SM/kg (Sørensen and Nørgaard, 2016).

Abbreviations: AA, amino acids; ADFI, average daily feed intake; ADG, average daily gain; CGI, compensatory growth index; CP, crude protein; DM, dry matter; FCR, feed conversion ratio; FM, fish meal; H-SM, high level starfish meal; L-SM, low level starfish meal; M-SM, medium level starfish meal; SM, starfish meal

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The study by Sørensen and Nørgaard (2016) was performed with individually housed pigs in clean and hygienic experimental facilities. Extended research with additional SM levels under commercial conditions will add validation to the use of SM as a potential feedstuff in pig feed. It is expected that there is a limit to the amount of SM that can be added to the diet without reducing growth. However, in cases where available protein sources are already limited, for example in organic pig farming, it would be useful to include a high amount of SM. Therefore, it would be advantageous to evaluate a feeding strategy that allows for inclusion of a high amount of SM during the early part of the growing period. Compensatory growth may be a way to overcome the consequence of P deficiency as a result from the high inclusion of SM in pig feed. Pigs fed diets deficient in a certain nutrient are known to have accelerated growth after changing to a diet sufficient in nutrients. This has been termed compensatory growth (Madsen and Bee, 2015).

In this study, the effect of feeding different SM levels on pig growth was evaluated in commercial conditions. Additionally, compensatory growth in pigs was assessed by removing SM from the diet in the late experimental period.

#### 2. Material and methods

All animal experimental procedures were carried out in accordance with the Danish Ministry of Justice, Act no. 474 of May 15, 2014 concerning experiments with animals and care of experimental animals and license issued by the Danish Animal Experiments Inspectorate, Ministry of Food, Agriculture and Fisheries, the Danish Veterinary and Food Administration.

#### 2.1. Animals and housing

A total of 4952 Landrace/Yorkshire  $\times$  Duroc piglets (6.3  $\pm$  0.8 kg; equal sex ratio) were housed in mixed sex groups with castrated males immediately after weaning. Pigs were placed in 11 identical rooms. Each room had 16 pens (2.4 m  $\times$  4.8 m). Two adjacent pens shared a feed dispenser; henceforth, these are termed double-pen. The number of pigs per pen was between 23 and 34 according to the number of weaned piglets available, but similar among treatments. The number of pigs per pen was balanced for each room. Temperature in the rooms was controlled at 23 °C at the start of the experimental period and was gradually decreased to 21 °C at the end of the experimental period. The  $CO_2$  level was controlled at a maximum of 3200 ppm. The flooring of pens was made up of one third concrete and one third slatted floor.

#### 2.2. Dietary treatments

Within each room, pens were assigned to one of four dietary treatments (n=22 double-pens/treatment): fishmeal control diet (FM) and low (L-SM), medium (M-SM) and high (H-SM) SM diets. The experimental period was divided into three phases of two weeks each. During the first two weeks of the experimental period, i.e. phase 1, pigs were fed diets containing 50 g/kg FM and 50, 75 or 100 g/kg SM, respectively (Table 1). In week 3–4, i.e. phase 2, the amount of FM and SM was halved for each treatment, resulting in 25 g/kg FM and 25, 37.5 and 50 g/kg SM, respectively (Table 2). In week 5–6, i.e. phase 3, all pigs received the same diet without SM. Diets were formulated to meet pigs' minimum requirements for all nutrients according to the Danish recommendations for weaned piglets (Tybirk et al., 2016). In both phase 1 and 2, two premixes were used supplementing vitamins, minerals, amino acids and enzymes to optimize diets. In both phases, one premix was adapted to fit the H-SM diet with its high calcium content, while the other premix was adapted to fit the FM control. Premixes contained potato protein concentrate as a carrier compound. Moreover, in phase 1, premix also contained soya concentrate. Eighteen tonnes of starfish were caught with a specialised starfish purse serine in Limfjord, Denmark, in April 2015. Starfish, both the solid and leaked liquid phase, were minced and subsequently dried at an FM processing plant to a water content of 86 g/kg. Dried starfish were ground into meal. All diet ingredients were mixed on farm and fed in meal form directly after mixing using high-precision equipment (Agrisys AirFeed, Herning, Denmark).

#### 2.3. Experimental protocol

Pigs were weaned between 24 and 27 days of age. Experimental treatments were started at day 0 of weaning and lasted 6 weeks. Pigs were distributed to the pens according to body weight to obtain uniform-sized pigs in each pen. Pens that shared the same feed dispenser both had pigs at the same size. There was permanent access to water and diets. Leftover feed was vacuumed out and weighed before transition to a new diet. Feed samples were taken from the pens weekly and stored in a freezer ( $-20\,^{\circ}$ C) for posterior nutrient analyses. Feed samples were pooled per week and per treatment. Pigs were weighed by pen on day 0, 14, 28 and 42 of the experimental period. Average daily gain (ADG), feed conversion ratio (FCR) and average daily feed intake (ADFI) were determined. Diarrhoea incidence was assessed by visual judgement and treated according to veterinary advice. Pens with high prevalence of diarrhoea were treated with antibiotics (Doxx-Sol) in the drinking water. It was not possible to apply antibiotics to drinking water to separate pens within a room, hence treatment of pens had to be applied to an entire room. Individual pigs were treated for diarrhoea with antibiotic injections (Borgal).

#### 2.4. Chemical analyses

Chemical analyses for dry matter (DM), crude protein (CP), crude fat, Ca, P and amino acids (AA) were performed in duplicates on feed samples from the phase 1 and 2 diets. Dry matter content was analysed by oven drying at 103 °C for four hours (European

Table 1
Ingredient composition and calculated nutrient composition (g/kg as-fed) of experimental diets containing fishmeal (FM; 50 g/kg) and low (L-SM; 50 g/kg), medium (M-SM; 75 g/kg) and high (H-SM; 100 g/kg) levels of starfish meal in phase 1 of the experimental period from 6 to 9 kg body weight.

	Treatments					
Item	FM	L-SM	M-SM	H-SM		
Ingredient composition						
Wheat	570	490	531	540		
Barley	50	99	68	50		
Starfish meal	-	50	75	100		
Fishmeal	50	-	_	_		
Soya oil	12	11	10	9		
Extruded SBM <sup>a</sup>	-	56	25	_		
Zinc premix	30	30	30	30		
Starfish premix <sup>b</sup>	-	169	178	208		
Fishmeal premix <sup>c</sup>	288	95	83	63		
Calculated composition <sup>d</sup>						
Dry matter	854	859	858	857		
Crude fat	38	37	38	38		
Net energy, MJ/kg	10.5	10.4	10.4	10.4		
Crude ash	52	53	58	64		
Crude protein	202	204	199	199		
SID <sup>e</sup> crude protein	178	175	171	171		
SID lysine	12.6	12.9	12.6	13.0		
SID methionine + cysteine	6.00	6.13	6.14	6.30		
SID tryptophan	2.72	2.70	2.61	2.59		
SID threonine	7.91	7.91	7.77	7.91		
SID valine	8.82	8.81	8.58	8.61		
Calcium	7.7	7.6	9.8	11.8		
Phosphorus	6.2	5.8	5.9	6.1		
Digestible Phosphorus	3.4	3.2	3.3	3.4		
Phytase Ronozyme NP, U	5760	5280	5220	5420		
β-Xylanase Danisco, FYT	1440	1320	1305	1355		

<sup>&</sup>lt;sup>a</sup> SBM, Soybean meal, AlphaSoy Pig 530.

Commission, 2009). Dried feed samples were milled (Retsch Centrifugal Mill ZM 200) at 1800 rpm with a 1 mm sieve. Thereafter, nitrogen content was evaluated with the Dumas method (AOAC 990.03). Crude protein was estimated as  $6.25 \times \text{nitrogen}$  concentration. Crude fat was hydrolysed with hydrochloric acid and subsequently extracted with petroleum ether (European Commission, 2009). For the determination of AA, samples were oxidised with performic acid and thereafter hydrolysed (European Commission, 2009). Amino acids were separated with ion exchange chromatography and measured by photometric detection after ninhydrin reaction. A correction factor of 1.19 was used for tyrosine to make up for oxidation losses. Tryptophan samples were hydrolysed with saturated barium hydroxide for 20 h at 110 °C. An internal standard was added and tryptophan was quantified with high performance liquid chromatography with fluorescence detection (European Commission, 2009). Before Ca and P determination by ICP-OES, samples were ashed at 550 °C and made into solution with HNO3 and HCL (European Commission, 2009).

#### 2.5. Calculations and statistical analysis

The FCR was calculated by dividing daily feed intake by daily gain. Compensatory growth index (CGI) of ADG and FCR was calculated as described by Hornick et al. (2000). The CGI of M-SM and H-SM was calculated. The following formula was used to calculate CGI:

$$CGI \frac{(FM1^{1} - SM1^{2}) - (FM3^{3} - SM3^{4})}{(FM1^{1} - SM1^{2})}$$

where 1 is ADG or FCR of FM in phase 1, 2 is ADG or FCR of M-SM or H-SM in phase 1, 3 is ADG or FCR of FM in phase 3, and 4 is ADG or FCR of M-SM or H-SM in phase 3.

All statistical analyses were performed using linear mixed models in R (R Core Team, 2016) adopting the package lme4 (Bates

<sup>&</sup>lt;sup>b</sup> Starfish premix contained per kg; 33.2 g SID lysine, 8.50 g SID methionine, 16.7 g SID threonine, 5.10 g SID tryptophan, 16.2 g valine, 208 g lactose, 1.4 g Ca, 2.85 g cloride, 11.4 g phosphorus, 7.43 g sodium, 702 mg iron, 600 mg copper, 187 mg manganese oxide, 468 mg zinc oxide, 94 mg niacin, 47 mg calcium-p-panthothenate, 37400 IU vitamin A, 37000 IU vitamin D3, 655 mg vitamin E, 18.7 mg vitamin K3, 9.4 mg vitamin B1, 18.7 mg vitamin B2, 14.0 mg vitamin B6, 20000 mg benzoic acid, 2000 FYT 6-phytase, 20000 U beta-xylanase.

<sup>&</sup>lt;sup>c</sup> Fishmeal premix contained equal amounts of ingredients to starfish premix except for (values per kg): 30.4 g SID lysine, 7.40 g SID methionine, 17.3 g SID threonine, 5.70 g SID tryptophan, 16.5 g valine, 20.9 g calcium, 8.88 g cloride, 10.7 g P, 5.11 g sodium.

<sup>&</sup>lt;sup>d</sup> Calculated by using a diet optimisation software (WinOpti version. 1.127, AgroSoft A/S, DK-7160 Tørring, Denmark) based on the feed evaluation system and matrix values published by Boisen (2007).

<sup>&</sup>lt;sup>e</sup> SID, standardised ileal digestible.

Table 2
Ingredient and calculated nutrient composition (g/kg as-fed) of experimental diets containing fishmeal (FM; 25 g/kg) and low (L-SM; 25 g/kg), medium (M-SM; 37.5 g/kg) and high (H-SM; 50 g/kg) levels of starfish meal in phase 2 of the experimental period from 9 to 15 kg body weight.

	Treatment					
Item	FM	L-SM	M-SM	H-SM		
Ingredient composition						
Wheat	596	593	599	605		
Barley	150	150	150	150		
Starfish meal	-	25	37	50		
Fishmeal	25	-	_	_		
Soya oil	25	24	22	21		
Extruded SBM <sup>a</sup>	28	40	28	14		
Soya bean meal	100	100	100	100		
Starfish premix <sup>b</sup>	-	30	44	60		
Fishmeal premix <sup>c</sup>	76	38	20	-		
Calculated composition <sup>d</sup>						
Dry matter	869	870	868	867		
Crude fat	47.0	46.0	45.0	45.0		
Net Energy, MJ/kg	10.1	10.1	10.1	10.1		
Crude ash	59	56	55	54		
Crude protein	190	191	191	190		
SID <sup>e</sup> crude protein	166	166	166	165		
SID lysine	12.1	12.0	12.0	12.0		
SID methionine + cysteine	6.74	6.55	6.55	6.55		
SID tryptophan	2.31	2.39	2.39	2.39		
SID threonine	7.31	7.30	7.30	7.30		
SID valine	7.98	7.98	7.98	7.98		
Calcium	9.2	8.6	8.6	8.6		
Phosphorus	6.4	5.9	5.9	5.8		
Digestible Phosphorus	3.9	3.8	3.8	3.8		
Phytase Ronozyme NP, U	1875	1879	1875	1884		
β-Xylanase Danisco, FYT	4000	4009	4000	4019		

<sup>&</sup>lt;sup>a</sup> SBM, Soybean meal, AlphaSoy Pig 530.

et al., 2014). Two adjacent pens sharing a feed dispenser (double-pen) was considered the experimental unit. Treatments were entered into the model as fixed effects. Initial weight was included as a covariate. Double-pen and pen were added as random effects. Independence of observations was assumed. The assumptions of equal variance and normal distribution were verified using residual plots and QQ plots. Data are presented as least squares means and standard error of means (SEM). Differences among means with  $P \le 0.05$  were considered statistically significant and differences among means with P < 0.10 were considered to represent a tendency.

#### 3. Results

Starfish meal contained 459 g protein/kg DM (Table 3). The Ca content was 99.5 g/kg DM and P content was 10.9 g/kg DM. The analysed nutrient composition of diets is shown separately for phase 1 and 2 (Tables 4 and 5). Crude protein content was comparable between all diets in phase 1 with 199–206 g/kg DM and in phase 2 with 192–194 g/kg DM. The Ca content of the FM and L-SM diets was comparable in phase 1 with 10.9 and 10.3 g/kg feed, respectively. The amount of Ca was increased to 13.0 g/kg for M-SM and to 16.1 g/kg for H-SM. The P levels were comparable between diets, hence Ca:P ratio became wider with increasing starfish content in phase 1. In phase 2, the large differences in Ca content between diets were eliminated.

Pigs were generally in good health. Pig mortality during the study was 1.17%. A little more than 2% of the pigs were taken out of the experiment due to diseases. Arthritis was the most common reason to remove pigs followed by diarrhoea. Most diarrhoea treatments were performed 3–6 days after transition to the phase 2 diets.

Similar ADG was found for pigs fed FM and L-SM in all phases (Table 6). The ADG of pigs was significantly lower when feeding

<sup>&</sup>lt;sup>b</sup> Starfish premix contained per kg; 97.7 g SID lysine, 27.4 g SID methionine, 44.3 g SID threonine, 10.4 g SID tryptophan, 30.8 g SID valine, 49.0 g calcium, 18.2 g cloride, 37.0 g phosphorus, 3.07 g potassium, 29.4 g sodium, 2863 mg iron, 2679 mg copper, 1.73 g magnesium, 764 mg manganese, 2093 mg zinc oxide, 6.68 mg selenium, 382 mg niacin, 191 mg D-pantothenic acid, 95440 IU vitamin A, 9540 IU vitamin D3, 3436 mg vitamin E, 76.36 mg vitamin K3, 38.2 mg vitamin B1, 76.4 mg vitamin B2, 57.3 mg vitamin B6, 83723 mg benzoic acid, 31396 FYT 6-phytase, 66979 U beta-xylanase.

<sup>&</sup>lt;sup>c</sup> Fishmeal premix contained per kg: 76.4 g SID lysine, 23.9 g SID methionine, 35.4 g SID threonine, 6.95 g SID tryptophan, 22.6 g valine, 102.08 g calcium, 61.0 g cloride, 36.17 g phosphorus, 2.39 g potassium, 28.7 g sodium, 2244 mg iron, 2105 mg copper, 0.21 g magnesium, 598 mg manganese, 1644 mg zinc oxide, 5.24 mg selenium, 299 mg niacin, 150 mg D-pantothenic acid, 74800 IU vitamin A, 7480 IU vitamin D3, 2693 mg vitamin E, 59.8 mg vitamin K3, 29.9 mg vitamin B1, 59.8 mg vitamin B2, 44.9 mg vitamin B6, 65789 mg benzoic acid, 24671 FYT 6-phytase, 52632 U beta-xylanase.

<sup>&</sup>lt;sup>d</sup> Calculated by using a diet optimisation software (WinOpti version. 1.127, AgroSoft A/S, DK-7160 Tørring, Denmark) based on the feed evaluation system and matrix values published by Boisen (2007).

<sup>&</sup>lt;sup>e</sup> SID, Standardised ileal digestible.

Table 3
Analysed chemical composition (g/kg DM) of starfish meal.

Dry matter, g/kg         914           Crude protein         459           Crude fat         92           Crude ash         320           Calcium         99.5           Phosphorus         10.9           Alanine         24.0           Arginine         24.7           Aspartic Acid         38.4           Cystein + cystine         5.02           Glutamic acid         50.6           Glycine         52.0           Histidine         7.75           Isoleucine         16.5           Leucine         26.9           Lysine         29.1           Methionine         10.2           Phenylalanine         16.3           Proline         17.8           Serine         18.8           Threonine         19.6           Tryptophan         4.74           Tyrosine         13.5           Valine         19.6	Item	Starfish meal
Crude fat       92         Crude ash       320         Calcium       99.5         Phosphorus       10.9         Alanine       24.0         Arginine       24.7         Aspartic Acid       38.4         Cystein + cystine       5.02         Glutamic acid       50.6         Glycine       52.0         Histidine       7.75         Isoleucine       16.5         Leucine       26.9         Lysine       29.1         Methionine       10.2         Phenylalanine       16.3         Proline       17.8         Serine       18.8         Threonine       19.6         Tryptophan       4.74         Tyrosine       13.5	Dry matter, g/kg	914
Crude ash 320 Calcium 99.5 Phosphorus 10.9 Alanine 24.0 Arginine 24.7 Aspartic Acid 38.4 Cystein + cystine 5.02 Glutamic acid 50.6 Glycine 52.0 Histidine 7.75 Isoleucine 16.5 Leucine 26.9 Lysine 29.1 Methionine 10.2 Phenylalanine 16.3 Proline 17.8 Serine 18.8 Threonine 19.6 Tryptophan 4.74 Tyrosine 13.5	Crude protein	459
Calcium 99.5 Phosphorus 10.9 Alanine 24.0 Arginine 24.7 Aspartic Acid 38.4 Cystein + cystine 5.02 Glutamic acid 50.6 Glycine 52.0 Histidine 7.75 Isoleucine 16.5 Leucine 26.9 Lysine 29.1 Methionine 10.2 Phenylalanine 16.3 Proline 17.8 Serine 18.8 Threonine 19.6 Tryptophan 4.74 Tyrosine 13.5	Crude fat	92
Phosphorus 10.9 Alanine 24.0 Arginine 24.7 Aspartic Acid 38.4 Cystein + cystine 5.02 Glutamic acid 50.6 Glycine 52.0 Histidine 7.75 Isoleucine 16.5 Leucine 26.9 Lysine 29.1 Methionine 10.2 Phenylalanine 16.3 Proline 17.8 Serine 18.8 Threonine 19.6 Tryptophan 4.74 Tyrosine 13.5	Crude ash	320
Alanine       24.0         Arginine       24.7         Aspartic Acid       38.4         Cystein + cystine       5.02         Glutamic acid       50.6         Glycine       52.0         Histidine       7.75         Isoleucine       16.5         Leucine       26.9         Lysine       29.1         Methionine       10.2         Phenylalanine       16.3         Proline       17.8         Serine       18.8         Threonine       19.6         Tryptophan       4.74         Tyrosine       13.5	Calcium	99.5
Arginine 24.7 Aspartic Acid 38.4 Cystein + cystine 5.02 Glutamic acid 50.6 Glycine 52.0 Histidine 7.75 Isoleucine 16.5 Leucine 26.9 Lysine 29.1 Methionine 10.2 Phenylalanine 16.3 Proline 17.8 Serine 18.8 Threonine 19.6 Tryptophan 4.74 Tyrosine 13.5	Phosphorus	10.9
Aspartic Acid 38.4  Cystein + cystine 5.02  Glutamic acid 50.6  Glycine 52.0  Histidine 7.75  Isoleucine 16.5  Leucine 26.9  Lysine 29.1  Methionine 10.2  Phenylalanine 16.3  Proline 17.8  Serine 18.8  Threonine 19.6  Tryptophan 4.74  Tyrosine 13.5	Alanine	24.0
Cystein + cystine       5.02         Glutamic acid       50.6         Glycine       52.0         Histidine       7.75         Isoleucine       16.5         Leucine       26.9         Lysine       29.1         Methionine       10.2         Phenylalanine       16.3         Proline       17.8         Serine       18.8         Threonine       19.6         Tryptophan       4.74         Tyrosine       13.5	Arginine	24.7
Glutamic acid 50.6 Glycine 52.0 Histidine 7.75 Isoleucine 16.5 Leucine 26.9 Lysine 29.1 Methionine 10.2 Phenylalanine 16.3 Proline 17.8 Serine 18.8 Threonine 19.6 Tryptophan 4.74 Tyrosine 13.5	Aspartic Acid	38.4
Glycine 52.0  Histidine 7.75 Isoleucine 16.5 Leucine 26.9 Lysine 29.1  Methionine 10.2  Phenylalanine 16.3  Proline 17.8  Serine 18.8  Threonine 19.6  Tryptophan 4.74  Tyrosine 13.5	Cystein + cystine	5.02
Histidine       7.75         Isoleucine       16.5         Leucine       26.9         Lysine       29.1         Methionine       10.2         Phenylalanine       16.3         Proline       17.8         Serine       18.8         Threonine       19.6         Tryptophan       4.74         Tyrosine       13.5	Glutamic acid	50.6
Soleucine   16.5   Leucine   26.9   Lysine   29.1   Methionine   10.2   Phenylalanine   16.3   Proline   17.8   Serine   18.8   Threonine   19.6   Tryptophan   4.74   Tyrosine   13.5	Glycine	52.0
Leucine 26.9  Lysine 29.1  Methionine 10.2  Phenylalanine 16.3  Proline 17.8  Serine 18.8  Threonine 19.6  Tryptophan 4.74  Tyrosine 13.5	Histidine	7.75
Lysine       29.1         Methionine       10.2         Phenylalanine       16.3         Proline       17.8         Serine       18.8         Threonine       19.6         Tryptophan       4.74         Tyrosine       13.5	Isoleucine	16.5
Methionine     10.2       Phenylalanine     16.3       Proline     17.8       Serine     18.8       Threonine     19.6       Tryptophan     4.74       Tyrosine     13.5	Leucine	26.9
Phenylalanine       16.3         Proline       17.8         Serine       18.8         Threonine       19.6         Tryptophan       4.74         Tyrosine       13.5	Lysine	29.1
Proline 17.8 Serine 18.8 Threonine 19.6 Tryptophan 4.74 Tyrosine 13.5	Methionine	10.2
Serine         18.8           Threonine         19.6           Tryptophan         4.74           Tyrosine         13.5	Phenylalanine	16.3
Threonine         19.6           Tryptophan         4.74           Tyrosine         13.5	Proline	17.8
Tryptophan 4.74 Tyrosine 13.5	Serine	18.8
Tyrosine 13.5	Threonine	19.6
-,	Tryptophan	4.74
Valine 19.6	Tyrosine	13.5
	Valine	19.6

Table 4

Analysed chemical composition (g/kg as-fed) of experimental diets containing fishmeal (FM; 50 g/kg) and low (L-SM; 50 g/kg), medium (M-SM; 75 g/kg) and high (H-SM; 100 g/kg) starfish meal in phase 1 of the experimental period from 6 to 9 kg bodyweight.

Item	Treatment					
	FM	L-SM	M-SM	H-SM		
Dry matter	888	889	888	888		
Crude protein	206	205	202	199		
Crude fat	42.5	43.0	41.5	42.0		
Calcium	10.9	10.3	13.0	16.1		
Phosphorus	6.85	6.19	6.24	6.48		
Calcium:phosphorus	1.59	1.67	2.08	2.48		
Lysine	14.5	14.8	14.5	13.8		
Methionine	4.76	4.91	4.68	5.17		
Cystein + cystine	3.10	3.09	2.97	2.94		
Threonine	9.71	9.38	9.50	9.15		
Valine	9.81	9.73	9.66	9.47		

Table 5

Analysed chemical composition (g/kg as-fed) of experimental diets containing fishmeal (FM; 25 g/kg) and low (L-SM; 25 g/kg), medium (M-SM; 37.5 g/kg) and high (H-SM; 100 g/kg) starfish meal in phase 2 of the experimental period from 9 to 15 kg bodyweight.

Item	Treatment					
	FM	L-SM	M-SM	H-SM		
Dry matter	876	875	873	877		
Crude protein	192	194	194	195		
Crude fat	50.5	48.0	48.5	48.5		
Calcium	11.3	11.1	10.5	10.2		
Phosphorus	6.92	5.52	5.20	5.76		
Calcium:phosphorus	1.62	2.02	2.02	1.77		
Lysine	13.0	11.0	10.2	11.9		
Methionine	4.53	3.60	3.38	3.78		
Cystein + cystine	2.90	3.10	3.11	2.96		
Threonine	8.51	9.02	9.03	9.04		
Valine	8.82	8.94	8.76	8.93		

Table 6

Body weight (BW ± SD) in kg, average daily gain (ADG) and average daily feed intake (ADFI) in kg/day and feed conversion ratio (FCR) of pigs fed diets containing fishmeal (FM) and low (L-SM), medium (M-SM) and high (H-SM) levels of starfish meal.

Parameter Pha		Treatment	Treatment					
	Phase*	FM	L-SM	M-SM	H-SM	SEM	P-values	
Initial BW	1	6.3 ( ± 0.8)	6.3 ( ± 0.8)	6.3 ( ± 0.8)	6.3 ( ± 0.8)	_	_	
ADG	1	0.203 <sup>b</sup>	$0.202^{\rm b}$	$0.178^{a}$	$0.169^{a}$	0.009	< 0.001	
ADFI	1	$0.278^{ab}$	$0.287^{\rm b}$	$0.272^{a}$	$0.265^{a}$	0.015	0.015	
FCR	1	1.38 <sup>a</sup>	1.43 <sup>a</sup>	1.53 <sup>b</sup>	1.57 <sup>b</sup>	0.08	< 0.001	
ADG	2	$0.472^{c}$	0.447 <sup>c</sup>	$0.382^{b}$	$0.307^{a}$	0.017	< 0.001	
ADFI	2	0.718 <sup>c</sup>	$0.732^{c}$	0.671 <sup>b</sup>	$0.614^{a}$	0.031	< 0.001	
FCR	2	1.55 <sup>a</sup>	1.67 <sup>b</sup>	1.76 <sup>b</sup>	2.03 <sup>c</sup>	0.08	< 0.001	
ADG	3	$0.797^{a}$	$0.804^{a}$	$0.801^{a}$	$0.812^{a}$	0.024	0.835	
ADFI	3	1.242 <sup>a</sup>	1.258 <sup>a</sup>	1.239 <sup>a</sup>	1.206 <sup>a</sup>	0.034	0.387	
FCR	3	1.56 <sup>b</sup>	1.56 <sup>b</sup>	1.54 <sup>ab</sup>	1.48 <sup>a</sup>	0.03	0.030	
ADG	1–3	0.487 <sup>c</sup>	0.483 <sup>c</sup>	0.448 <sup>b</sup>	$0.427^{a}$	0.012	< 0.001	
ADFI	1–3	0.746 <sup>bc</sup>	0.759 <sup>c</sup>	$0.727^{ab}$	$0.698^{a}$	0.021	0.0005	
FCR	1-3	1.52 <sup>a</sup>	1.57 <sup>b</sup>	1.60 <sup>c</sup>	1.63 <sup>c</sup>	0.016	< 0.001	

 $<sup>^{</sup>a,b,c}$  Values within a row with different superscripts are significantly different (P  $\leq$  0.05).

diets with M-SM and H-SM compared with FM and L-SM in both phase 1 and 2 (P < 0.001). Moreover, in phase 2, ADG was significantly lower when feeding pigs H-SM compared with M-SM (P < 0.001). In phase 3, however, ADG did not significantly differ between treatments. The calculated CGI of ADG was 1.16 for pigs fed M-SM and 1.44 for pigs fed H-SM.

The ADFI of pigs given FM and L-SM was comparable in each phase. The ADFI of L-SM-fed pigs was increased compared with pigs fed M-SM and H-SM in phase 1 (P = 0.015). Furthermore, ADFI of pigs receiving M-SM and H-SM was significantly lower in phase 2 compared with the other two treatments (P < 0.001). Additionally, in phase 2, feeding pigs M-SM resulted in significantly higher ADFI compared with feeding H-SM (P < 0.001). All treatments caused similar ADFI in phase 3.

In phase 1, FCR was equivalent for pigs fed FM and L-SM, but significantly lower for pigs fed M-SM and H-SM (P < 0.001). The FCR of pigs on the H-SM diet in phase 2 was significantly higher than the FCR of pigs on all other treatments (P < 0.001). Moreover, FCR was increased when feeding pigs the M-SM and L-SM diets compared with feeding pigs the FM diet (P < 0.001). In phase 3, FCR of pigs fed H-SM was lower compared with pigs fed FM and L-SM (P = 0.030). The CGI of FCR was 1.13 for pigs fed M-SM and 1.42 for pigs fed H-SM. The difference in performance of pigs fed M-SM and H-SM compared with pigs fed FM and L-SM was larger in phase 2 than in phase 1.

#### 4. Discussion

This study showed that feeding FM and L-SM diets resulted in similar growth, but M-SM and H-SM diets caused reduced growth during phase 1, which is in agreement with Sørensen and Nørgaard (2016). They found that including 100 g/kg SM reduced growth and resulted in high Ca:P ratio, which coincided with reduced serum P (Sørensen and Nørgaard, 2016). Similarly, in the current study, a high Ca:P ratio for M-SM and H-SM can have led to reduced growth. Calcium can form complexes with phytate, making it unavailable for phytase-activity (Lei et al., 1994). Furthermore, Ca can compete for the active sides of phytase (Brady et al., 2002). Lastly, Ca and P can form insoluble complexes (Selle et al., 2009). Several studies have found a reduction in growth when Ca:P ratio exceeds 2.0:1 with P included below requirement (Qian et al., 1996; Liu et al., 1998; Liu et al., 2000). Moreover, Reinhart and Mahan (1986) found reduced growth for diets with a Ca:P ratio above 2.0:1 when feeding P at 1.0 g/kg above requirement. In the current study, the Ca:P ratio in the M-SM and H-SM diets exceeded 2.0:1, and P was supplied at minimum requirement level. Therefore, we may assume that in the current study, the combination of P-level and the Ca:P ratio may have lowered P absorption. Low P availability impairs development of muscular and skeletal tissue (Alexander et al., 2008).

The high Ca:P ratio observed for L-SM and M-SM may account for reduced growth found in phase 2. However, the growth reduction for H-SM was unexpected as Ca:P ratio was reduced in the second phase. The exact reason for this is unknown, but a possible explanation can be that growth recovery is not immediate. According to Hornick et al. (2000) sparing mechanisms occurring at reduced growth can be maintained for some time. Moreover, in the case of P deficiency, P and energy may have been used to restore bone mineral content, therefore being unavailable for improved growth. A high Ca:P ratio has been known to reduce bone ash levels (Qian et al., 1996; Liu et al., 1998; Liu et al., 2000). A two-week period might not have been long enough to observe a positive effect of reducing the amount of SM in the diets. Furthermore, most diarrhoea incidences were observed shortly after transition to the phase 2 diets. Frequency of diarrhoea was not treatment specific (data not shown). Diarrhoea may indicate a general difficulty in adapting to the diet.

Compensatory growth is the ability of a pig to show accelerated growth after restoring a deficiency in the diet. Compensatory growth observed in phase 3 of the current study is believed to be a result of a resolved high Ca:P ratio. Compensatory growth response is mostly studied with different feeding levels, but rarely with different mineral levels. Varley et al. (2011) tested compensatory growth from low P inclusion in diets. They found a compensatory growth response in the form of an improved FCR and increased

<sup>\*</sup>In phase 1, FM and L-SM were included at 50 g/kg of the diet. M-SM was 75 g/kg and H-SM was 100 g/kg of the total diet. In phase 2, the inclusion percentages of FM and SM were halved for all treatments. In phase 3, SM was removed from the diets.

ADG in gilts after feeding diets low (4 g total P/kg) and sufficient (6 g total P/kg) in P in either the grower and/or the finisher phase. Pigs fed H-SM were able to improve growth to a larger extent than M-SM fed pigs. The extent of compensatory growth is dependent on the length, severity and type of nutrient restriction and is more pronounced if restriction is not too severe (Fernández and Nørgaard, 2009). The higher compensatory growth for pigs fed H-SM in this study compared with M-SM was therefore unexpected. Possibly, compensatory growth was at a different stage for both treatments. In phase 2, compensatory growth mechanisms may have already started for H-SM, whereas they were not started for M-SM due to the Ca:P ratio remaining high. In the current study, compensatory growth was observed for a short period of time. Determining the maximum duration of feeding H-SM and M-SM, after which compensatory growth is sufficient to generate normal growth levels, is a necessary prerequisite for the successful implementation of this feeding strategy. Hornick et al. (2000) described a pattern of compensatory growth where accelerated growth persists until a maximum and after some time returns to normal levels. Maximum compensatory growth and the duration of compensatory growth will influence the ability of SM-fed pigs to reach normal slaughter weights.

#### 5. Conclusion

Feeding weaned pigs  $50\,\text{g/kg}$  SM gave similar growth to feeding  $50\,\text{g/kg}$  FM under commercial conditions. The ADG, ADFI and FCR were negatively affected when feeding a diet with  $75\,\text{g/kg}$  SM or  $100\,\text{g/kg}$  SM. Growth recovery did not start directly after halving SM levels. Complete removal of SM from the diets during phase 3 resulted in similar growth for pigs on all treatments. Compensatory growth of pigs was observed for both the  $75\,\text{g/kg}$  SM and the  $100\,\text{g/kg}$  SM treatment, and was strongest for the  $100\,\text{g/kg}$  SM treatment. Piglets responded well to a dietary inclusion of up to  $50\,\text{g/kg}$  SM, but caution should be taken for higher inclusion levels due to risk of too high dietary Ca:P.

#### Conflict of interest

No potential conflict of interest was reported by the authors.

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