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Effect of supplemental milk replacer and liquid starter diet for 4 and 11 days postweaning on intestinal parameters of weaned piglets and growth to slaughter

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ABSTRACT

Reduced piglet feed intake immediately postweaning (**pw**) leads to disruption of small intestine structure and function and reduced growth. Our objective was to evaluate the effect of providing supplemental milk or liquid starter diet for either 4 or 11 days pw, on intestinal parameters of newly weaned piglets and growth to slaughter. At weaning (28 ± 0.6 days old), five hundred and eighty−seven piglets ((Large White × Landrace) × Duroc) were divided into 59 pen groups, each containing 9–10 same sex (entire male or female) piglets. The pen groups were blocked by sex and weaning weight and provided with ad-libitum access to one of five dietary treatments: (1) Dry pelleted starter diet (control; **CON**); (2) CON +liquid milk replacer for 4 days pw (**M4**); (3) CON+liquid milk replacer for 11 days pw (**M11**); (4) CON +liquid starter diet for 4 days pw (**S4**) and (5) CON+liquid starter diet for 11 days pw (**S11**). Pen groups were weighed at weaning, days 11, 20, 28, and 47 pw and at target sale weight. Feed disappearance (on a DM basis) was recorded on each weighing day. On day 7 pw, 10 piglets per treatment were euthanised to collect small intestine tissue samples for determination of villus height (**VH**), crypt depth and brush-border membrane enzyme activity. Data were analysed using SAS-version 9.4. Between days 0 and 11 pw, M11 increased average daily feed intake by 48% and average daily gain (**ADG**) by 57% compared to CON ($P < 0.05$), and increased ADG by 54% ($P < 0.05$) compared to S4. Piglets on M11 also had improved feed conversion efficiency compared with CON piglets between days 0 and 11 pw. Treatment did not affect growth performance after day 28 pw, or carcass parameters at slaughter. At day 7 pw, M11 piglets had 37% higher jejunal VH than CON piglets ($P < 0.05$) and S11 piglets had 28% higher ileal VH than S4 piglets ($P < 0.05$). M11 piglets had up to 150% higher ileal sucrase activity than M4, S4 and S11 piglets ($P < 0.05$) and 180% higher ileal maltase activity than S4 piglets ($P < 0.05$). In conclusion, M11 reduced the immediate negative effects of weaning, as it was associated with increased feed intake, growth, brush-border membrane enzyme activity and improved intestinal structure early pw. However, there were no carryover effects of any of the liquid supplements on growth or feed efficiency or carcass weight at slaughter.

Implications

Typically, commercially reared piglets are abruptly separated from their sows at ∼3–4 weeks of age. Newly weaned piglets find it difficult to adjust to solid feed postweaning, leading to reduced feed intake and growth, and impairment of intestinal structure and function. Providing piglets with milk in addition to a dry pelleted diet for 11 days postweaning increased feed intake and growth, but only up to 28 days postweaning. Supplementing milk for 11 days postweaning also improved intestinal structure and function. This strategy can be beneficial in reducing the check in growth normally observed during the first week postweaning.

Introduction

Conventional swine production systems wean piglets from their sows at ∼3–4 weeks of age, then provide them with a dry vegetable-based postweaning (**pw**) diet. The abrupt change from the highly digestible liquid sow's milk to this vegetable-based solid

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diet results in severe nutritional and intestinal stress in newly weaned piglets (Lawlor et al., 2020). In the early pw period, reduced feed intake and growth, disruption of small intestinal structure, and low digestive enzyme activity are commonly observed (Dong and Pluske, 2007).

Many studies have associated reduced feed and nutrient intake at weaning with adverse morphological changes in the small intestine, such as the shortening of villi and deepening of crypts (Hampson, 1986; Pluske et al., 1997). Such impairment of intestinal structure can lead to disruption of barrier function, which can lead to translocation of pathogens, toxins and allergic compounds across the intestinal barrier, thereby causing enteric health issues, such as pw diarrhoea (Vente-Spreeuwenberg and Beynen, 2003). Moreover, digestive enzymes secreted from the brush-border membrane of the villi are affected by villus atrophy (Kelly et al., 1991) and their activities are generally reduced by weaning due to the rapid change in diet (Marion et al., 2005). Therefore, it is important to increase early pw feed intake.

There have been some previous studies investigating the provision of supplemental milk and liquid starter diet as a means of increasing feed intake and preventing damage to intestinal structure and function. Zijlstra et al. (1996) supplemented weaned piglets with milk along with a dry pelleted diet and reported increased feed intake and growth compared to piglets weaned onto only a dry pelleted diet. Similarly, providing liquid starter diet after weaning also improved feed intake and growth (Lawlor et al., 2002; Han et al., 2006). Although such pw supplementation strategies generally only provide transitory benefits to pig growth (Lawlor et al., 2002; Rault et al., 2015), Kim et al. (2001) found that pw liquid diet-fed pigs reached target slaughter weight earlier than dry-fed pigs. Moreover, pw supplementation of a mixture of milk and pelleted starter is suggested to have a benefit at slaughter based on growth estimations (Dunshea et al., 1999). However, to date, studies comparing liquid milk replacer and liquid starter diet as pw strategies to increase feed intake and growth of piglets have not been conducted. Moreover, guidelines for the optimal duration of pw supplementation of liquid milk replacer and liquid starter diet are not available. During the acute pw phase (3–5 days pw), major changes in gut structure and function occur (Montagne et al., 2007) so it is likely that liquid milk replacer/liquid starter diet may be of benefit then. However, continuing liquid supplementation into the pw maturation phase (>5 days pw) may also benefit pig growth. Therefore, in this experiment, we supplemented liquid milk replacer and liquid starter diet for 4 and 11 days pw to determine the optimum duration of supplementation. This information is crucial as milk replacer in particular is expensive (~3 times the price of the starter diet). Therefore, the period of supplementation should be as short as possible to ensure that the practice is economically viable.

Based on findings from previous studies, we hypothesise that supplementing a liquid milk replacer or a liquid starter diet to newly weaned piglets provided with a dry pelleted starter diet will increase early pw feed intake and growth. We further hypothesise that disruption to intestinal integrity, normally associated with weaning, will be reduced in response to the increased pw feed intake, as a result of supplementing liquid milk replacer or a liquid starter diet to newly weaned pigs. This should facilitate the transition of weaned pigs to a dry pelleted pw diet. The overall aim of this study was to compare the effect of supplementing liquid milk replacer and liquid starter diet, in addition to a dry pelleted starter diet, for 4 and 11 days pw to newly weaned piglets on intestinal structure and function, feeding behaviour, growth from weaning to target sale weight and carcass parameters. Currently, weaning at 28 days of age is typically practised in European pig production and so was chosen as weaning age in the current study. Preliminary results have been published in limited form as a conference abstract (Vasa et al., 2023).

Material and methods

Experimental design and diets

The study involved a total of 587 weaned (28 ± 0.6 days of age) piglets ((Large White × Landrace) × Duroc), housed in 59 pen groups, each containing 9–10 piglets of the same sex (entire male or female). The experiment was conducted over two batches with 29 pens in the first batch and 30 pens in the second batch. The pen groups were blocked by sex and weaning weight and provided with ad-libitum access to one of five dietary treatments: (1) Dry pelleted starter diet (control; **CON**; n = 12 pens); (2) CON+liquid milk replacer for 4 days pw (**M4**; n = 11 pens); (3) CON+liquid milk replacer for 11 days pw (**M11**; n = 12 pens); (4) CON+liquid starter diet for 4 days pw (**S4**; n = 12 pens) and (5) CON+liquid starter diet for 11 days pw (**S11**; n = 12 pens). Milk replacer and liquid starter diet were provided by an automated delivery system (Babyfeed; Schauer Agrotronic GmbH, Prambachkirchen, Austria), to a single feed trough (54 × 11 × 5 cm) in each pen. Liquid supplementation with the milk and starter diet commenced on the day of weaning, with M4 and S4 supplementation ending on the morning of day 4 pw, while M11 and S11 supplementation ceased on the morning of day 11 pw. The pens with CON piglets were also equipped with the liquid feeder troughs, although no liquid supplementation was provided.

The supplemental milk was probiotic-free and commercially sourced (Opticare milk; Swinco B.V, Helmond, The Netherlands). The milk powder contained the following, in descending order of inclusion: sweet whey powder, vegetable oils, porcine dried plasma powder, whey powder, digestible starch, dextrose, hyper-immunised egg powder, soya protein concentrate, hydrolysed wheat gluten, premix of amino acids, vitamins and trace minerals. The milk powder contained 11.9 MJ/kg net energy, 21.5% CP, 9% fat, 0.1% crude fibre, 6.5% crude ash, 1.8% lysine, 0.46% methionine, 0.7% calcium, 0.55% phosphorus, and 0.7% sodium. As per the supplier's recommendations, 150 g of the milk powder was mixed with 1 L of warm water (55 ℃) leading to 13.04% DM content in the liquid milk.

The ingredient content and chemical composition of all diets are provided in Table 1. The starter diet was fed dry as a 3 mm pellet to all treatment groups and was also used to prepare liquid starter diet for the S4 and S11 treatments. To achieve the same DM content in the liquid starter diet as for the liquid milk, 165.6 g of the dry starter was mixed with 1 L of warm water (55 ℃). Pelleted link diet was provided from day 11 to 20 pw followed by pelleted weaner diet until transfer to the finisher room at day 47 pw. Pelleted finisher diet was provided from day 47 pw until slaughter. All of the piglets were offered dry pelleted starter diet in the farrowing room from day 12 after birth until weaning. All of the diets were formulated to meet or exceed NRC (2012) recommendations. The feed and water were not medicated and were provided on an ad-libitum basis via single−space stainless-steel feeders (O'Donovan Engineering, Coachford, Ireland) and a single bowl drinker (DRINK-O-MAT, Egebjerg International A/S, Nykøbing Sjælland, Denmark).

Liquid feeding system

Ten feeding cycles (each lasting ~2 h) were programmed between 0930 and 0400 h, with two fresh mixes being prepared at 0900 and 1700 h. During each cycle, a sensor installed above

Table 1
Ingredient and chemical composition of experimental diets fed to pigs in this study (on an air-dry basis; g/kg unless otherwise stated).

	Diet specifications			
Item	Starter[1]	Link[1]	Weaner[1]	Finisher[1]
Ingredient composition				
Barley	50.00	68.43	495.88	410.51
Wheat	0	100.00	216.86	390.00
Maize	231	300	0	0
Soybean meal	143.42	186.91	163.16	165.00
Soya full fat	130.81	70.00	50.00	0
Skim milk powder	125	50	0	0
Whey permeate	200	150	0	0
Soya oil	85.00	38.21	40.00	11.00
Lysine HCl	6.22	6.72	5.93	4.27
DL-Methionine	3.62	3.18	2.17	1.00
L-Threonine	3.64	3.42	2.71	1.90
L-Tryptophan	1.40	1.27	0.57	0.22
L-Valine	1.29	1.26	0.62	0
Vitamin and mineral premix[2]	3	3	3	1
Ronozyme HiPhos GT[3]	0.1	0.1	0.1	0.1
Salt	3	3	3	3
Mono di-calcium phosphate	5.5	7.0	5.5	1.0
Limestone flour	7.0	7.5	10.5	11.0
Chemical composition				
DM[4]	914.3	904.0	890.5	880.0
CP[4]	191.3	174.0	162.5	163.0
Ash[4]	55.5	51.0	44.0	46.5
Fat[4]	129.15	83.40	74.80	36.60
Crude fibre[4]	16.25	19.50	31.00	33.50
NDF[5]	60.54	80.78	139.95	140.16
Lysine[5]	16.20	15.00	12.99	10.85
Methionine[5]	7.04	6.12	4.70	3.44
Methionine + Cysteine[5]	9.80	9.05	7.86	6.58
Threonine[5]	10.87	10.05	8.78	7.57
Tryptophan[5]	3.66	3.36	2.66	2.22
Standardised ileal digestible lysine[5]	15.28	14.14	12.00	10.00
Ca[5]	8.19	7.54	7.37	6.52
Digestible P[5]	4.62	4.23	3.32	2.46
Net energy[5] (MJ/kg)	12.06	10.94	10.30	9.80

[1] The starter diet was fed dry as a 3 mm pellet to all treatment groups and was also used to prepare liquid starter diet. Pelleted link diet was provided from day 11 to day 20 postweaning (pw) followed by pelleted weaner diet until transfer to the finisher room at day 47 pw. Pelleted finisher diet was provided from day 47 pw until slaughter.
[2] Premix provided per kilogram of complete diet (starter, link and weaner): Cu from copper sulphate, 85 mg; Fe from ferrous sulphate monohydrate, 90 mg; Mn from manganese oxide, 47 mg; Zn from zinc oxide, 120 mg; I from potassium iodate, 0.6 mg; Se from sodium selenite, 0.3 mg; vitamin A as retinyl acetate, 2.1 mg; vitamin D3 as cholecalciferol, 25 μg; vitamin E as DL-alpha-tocopheryl acetate, 100 mg; vitamin K, 4 mg; vitamin B12, 15 μg; riboflavin, 2 mg; nicotinic acid, 12 mg; pantothenic acid, 10 mg; choline chloride, 250 mg; vitamin B1, 2 mg; and vitamin B6, 3 mg. Premix provided per kilogram of complete diet (finisher): Cu from copper sulphate, 15 mg; Fe from ferrous sulphate monohydrate, 24 mg; Mn from manganese oxide, 31 mg; Zn from zinc oxide, 80 mg; I from potassium iodate, 0.3 mg; Se from sodium selenite, 0.2 mg; vitamin A as retinyl acetate, 0.7 mg; vitamin D3 as cholecalciferol, 12.5 μg; vitamin E as DL-alpha-tocopheryl acetate, 40 mg; vitamin K, 4 mg; vitamin B12, 15 μg; riboflavin, 2 mg; nicotinic acid, 12 mg; pantothenic acid, 10 mg; vitamin B1, 2 mg; vitamin B6, 3 mg.
[3] Ronozyme HiPhos GT (Inform Nutrition, Whites Cross, Ireland) was included to provide 1000 g phytase units (FYT) in each diet.
[4] Analysed composition.
[5] Calculated composition.

the liquid feeder troughs checked the amount of liquid feed present in the feeder trough 5 times. Whenever the feed level was below the level of the sensor, the trough was detected as empty, and milk or liquid starter diet was delivered to the trough. Therefore, each pen was potentially supplied with milk or liquid feed up to 50 times each day. Each day after the last feeding, the system was cleaned in a closed circuit, which included all of the pipelines, with a 1% acid solution (Deosan Acidbrite AG313, Diversey Europe Operations BV, Utrecht, The Netherlands). In addition, the pipelines were cleaned once a week with a 0.5% solution of an alkaline detergent (AvalKsan Gold Standard CF, Carbon Group, Ringaskiddy, Ireland) to remove lime scale accumulating in the circuit. The mixing tanks were cleaned by rinsing with water once a day, and the feed troughs were cleaned with air pressure and rinsed with water once a day.

Housing

The weaner pens (2.1 × 3 m) had fully-slatted plastic floors, and the finisher pens (4 × 2.4 m) had slatted concrete floors. Temperature was automatically controlled in both of the rooms. In the weaner room, temperature was maintained at 28–30 °C in the first week and reduced by 2 °C per week to 22 °C in the 4th week and remained at that temperature until 47 days pw. In the finisher rooms, temperature was maintained at 20 °C. Ventilation was provided by punched ceiling ventilation with air exhausted by a variable speed fan linked to a thermostat and was automatically controlled (Big Dutchman 135, Vetcha, Germany). Lighting was provided by tubular fluorescent lights from 0830 to 1630 h. Environmental enrichment included a chain mounted on the side of each pen in both the weaner and finisher rooms, two floor-based star-shaped rubber toys in the weaner pens and a 20 × 2.5 cm timber post in the finisher pens.

Data recording and sampling

Growth performance, faecal scoring, medication usage and carcass data

Pigs were individually weighed at days 0, 11, 20, 28 and 47 pw and on the day of sale (~day 131 pw) using an electronic scale (Ezi-Weigh 7i, O'Donovan Engineering). Pigs were fasted for 15–18 h before weighing prior to slaughter. Using the individual pig BW,

the average daily gain (**ADG**) of each pen was calculated. The CV of within−pen pig weight was calculated as an indicator of within-pen pig weight uniformity. Feed disappearance in each pen was monitored at intervals (between each weighing day) from weaning until sale to calculate average daily feed intake (**ADFI**) per pig on a DM basis, including both solid and liquid feed. The ADFI per pig from solid feed and from liquid feed was calculated on a DM basis. The total feed intake per pig in the first 11 days pw from solid feed and from liquid feed was calculated on a DM basis. Feed conversion ratio was calculated as ADFI / ADG.

The prevalence of pw diarrhoea was determined at pen level by daily visual scoring of faecal consistency in each pen between days 1 and 12 pw using a 4-point scoring system (Casey et al., 2007) as follows: 0 for dry pelleted faeces; 1 for soft faeces with shape; 2 for mild diarrhoea (very soft without shape or viscous liquid faeces) and 3 for severe diarrhoea (watery or with blood). The average score from five pigs was determined as the average score for each pen. The average number of days with diarrhoea in each pen between days 1 and 12 pw was calculated by adding all of the days when a faecal consistency score of 2 or 3 was assigned to that pen. All antibiotic and anti-inflammatory usage, in terms of number of injections and ml of doses used on average per pig per pen, and number of clinical cases of disease (number of pigs that were treated on one or more occasion) per pen were recorded. The only antibiotic used was Unicillin (Procaine Benzylpenicillin, 300 mg/ml injection, Univet, Cootehill, Ireland) and the only anti-inflammatory used was Loxicom® (5 mg/ml injection, Norbrook, Monaghan, Ireland). All mortalities and removals from the experiment were recorded. Pigs were slaughtered at ∼127.6 (± 12.9) kg live weight by CO_2 stunning followed by exsanguination. Carcass weight, kill-out percentage, muscle and back fat depth and lean meat percentage were calculated as described previously by Crespo-Piazuelo et al. (2022). A full description of the methods used for the calculation of carcass parameters is provided in Supplementary Material S1.

Live observations of feeding behaviour

At weaning, all of the piglets were individually marked with numbers on their backs using dark hair dye (Pro Colour Plus 0.1 black, Wolverhampton, United Kingdom) to facilitate the identification of individual animals. Feeding behaviour of individual piglets was observed using 3-minute instantaneous scan sampling on days 2, 4, 6 and 8 pw. During each 1-h session of live observations, each pen was scanned every 3 min, leading to 21 scans of a pen per session. On days 2 and 4 pw, six 1-h sessions were conducted between 0900 and 1600 h. On days 6 and 8 pw, five sessions were conducted (the last session was excluded due to time constraints).

A simple ethogram was used for recording feeding behaviour at the individual pig level. At every scan, feeding activity for solid feed was recorded when a piglet was seen engaging in solid feeder trough-directed activity. This was defined as when a piglet snout was positioned over the rim of the feeder so that it was inserted into the solid feed trough. Liquid (milk or liquid starter) feeding activity was recorded when a piglet had their snout immersed in the liquid for at least 2 s. Observations of piglets sleeping with their snout in the feed trough were excluded as the piglets were not actively engaging with the feeder troughs. The percentage of non-eaters of pelleted solid feed in each pen on day 2 pw was determined by calculating the number of piglets in each pen that were not observed to engage in solid feeder-directed activity even once during all of the sessions on that day. The percentage of observations during which piglets engaged in solid and liquid feeder-directed activities was calculated separately on an individual piglet basis on days 2, 4, 6 and 8 pw. This was carried out by dividing the total number of scans where an individual piglet was observed engaging with either the liquid feeder or solid feeder on each day by the total number of scans on the day, then multiplying the result by 100 to express as a percentage.

Euthanasia and tissue sampling

On day 7 pw, 50 piglets (10 piglets per treatment) were euthanised using a captive bolt followed by immediate exsanguination. Female (5 per treatment) and male (5 per treatment) piglets that had been observed to be interacting with both solid and liquid feeders at least once during observation days 2 and 4, and were of median pen weight on day 5 pw, were selected for euthanasia. After euthanasia, the intestinal tract was removed and whole tissue samples (∼2 cm) and mucosal scrapings were collected from the duodenum (15 cm distal to the pyloric junction), jejunum (1.5 m distal to the pyloric junction) and ileum (15 cm proximal to the ileo-caecal junction). The whole tissue samples were carefully immersed in a tube containing NOTOXhisto™ fixative (Scientific Device Laboratory, Des Plaines, Illinois, USA) and the tubes were placed on a shaker for 48 h after collection. Mucosal scrapings were collected by longitudinally opening the tissue samples, rinsing with cold phosphate−buffered saline to remove the digesta, and scraping the mucosal layer with a glass slide into 2 ml tubes. The tubes were snap-frozen in liquid nitrogen and stored at −80 °C for enzyme activity analysis.

Laboratory analysis

Compositional analysis of liquid milk and diets

Representative samples were collected from dry pelleted diets and liquid starter diet. The liquid starter diet samples were oven−-dried for 3 days at 55 °C to determine the DM%. The oven-dried liquid starter and dry pelleted diet samples were analysed for DM (oven drying), ash (gravimetry), CP (Dumas method), total fat (Wiebul acid hydrolysis) and crude fibre (Ankom 200 fibre analyser, Macedon, New York, USA) by Sciantec Analytical services Ltd, Selby, United Kingdom. Liquid milk samples were collected and analysed for percentage of solids, total protein, and fat, using a Bentley Dairyspec FT (Bentley Instruments Inc., Chaska, Minnesota, USA).

Small intestinal histology

Slides for histological analysis were prepared using whole tissue samples from the duodenum, jejunum and ileum by Nationwide Laboratories, Devon, United Kingdom. Haemotoxylin and eosin staining was performed to study gross morphological parameters of the intestinal structure. For each sample, the villus height (**VH**) and crypt depth (**CD**) were measured as described previously in Crespo-Piazuelo et al. (2022) and the ratio of VH to CD was calculated. A full description of the methods used for small intestinal histological analysis is provided in Supplementary Material S1.

Enzyme activity analysis

Mucosal scrapings were weighed and homogenised with a 1% Triton-X buffer to extract the membrane−bound brush-border membrane enzymes. After centrifugation (2 000 x G for 10 min at 4 °C), the supernatant was used to determine the enzyme activities of three disaccharidases and three peptidases. The disaccharidases measured were lactase, maltase and sucrase, which were analysed using a spectrophotometric assay, as previously described (Sangild et al., 1995). Briefly, the samples were mixed with their specific substrates (lactose, maltose and sucrose) and incubated for 30 min at 37 °C. Postincubation, 250 μL of Peroxidase − Glucose oxidase colour solution was added and the samples were spectrophotometrically analysed at 450 nm and 37 °C using a plate reader (SpectraMax® ABS, VWR International, Radnor, Pennsylvania, USA). All reagents and substrates used for homogenisation of

tissue and disaccharidase enzyme activity assays were supplied by Merck Life Sciences Limited, Arklow, Ireland. The peptidases measured were aminopeptidase-A, aminopeptidase-N and dipeptidylpeptidase IV. These were also analysed using a spectrophotometric assay, as previously described (Sangild et al., 1995). The substrate used to determine the activity of aminopeptidase-A was L-Alanyl-p-nitroanilide-hydrochloride (Merck Life Sciences Limited), for aminopeptidase-N, it was Glu-p-nitroaniline (PeptaNova GmbH, Sandhausen, Germany) and for dipeptidylpeptidase IV, it was Glycyl-Prolyl p-nitroanilide p-toluenesulphonate salt (Bachem, Bubendorf, Switzerland). The samples with specific substrates were kinetically measured at 405 nm at 37 °C using the same plate reader as mentioned above. All enzyme activities were expressed as enzyme units per gram of tissue (U/g). A full description of the methods used for enzyme activity analysis is provided in Supplementary Material S1.

Statistical analysis

Statistical analyses were conducted using SAS version 9.4 (SAS Institute Inc.). Growth performance indicators i.e. BW, ADFI, ADG, feed conversion ratio and CV of individual piglet BW within pen which were recorded at multiple time-points were analysed by considering repeated measures, using the MIXED procedure. Treatment was included in the model as a fixed effect, block as a random effect, days pw as the repeated measure, and pen group was considered the experimental unit. Weaning weight, if significant in the model, was used as a co-variate, and the most appropriate co-variance structure as indicated by the model fit statistics was applied. The 'slice' option in SAS was used to obtain the simple main effects at each day pw. The percentage of observations that individual piglets engaged in solid and liquid feed-directed activity over the four observation days were also analysed by considering repeated measures, and using the MIXED procedure, with the same conditions as outlined above, other than the individual pig nested within pen was considered the experiment unit.

Carcass parameters, medication usage, average number of days with diarrhoea, the percentage of non-eaters on day 2 pw, and the ADFI and total feed intake of solid and liquid feed (days 0–11 pw) were analysed using PROC MIXED with treatment as a fixed effect, block as a random effect, weaning weight as a co-variate (if significant in the model) and pen group was considered the experimental unit. Histology and enzyme activity parameters were analysed using PROC MIXED with treatment as a fixed effect, block as a random effect and individual pig was considered the experimental unit. In all cases, the Tukey-Kramer adjustment was applied to account for multiple comparison of means. Significant differences between treatments were considered to occur when $P < 0.05$.

In addition, five specific hypotheses were investigated using contrast and estimate statements. Each time-point/period was analysed separately (same conditions as described above) for the growth performance indicators (BW, ADFI, ADG, feed conversion ratio and CV of individual piglet BW within pen), carcass and intestinal parameters. The five comparisons were 1) Milk treatments (M4 + M11) vs CON; 2) Liquid starter diet treatments (S4 + S11) vs CON; 3) 11-day duration treatments (S11 + M11) vs 4-day duration treatments (S4 + M4) 4) 4-day duration treatments (S4 + M4) vs CON; 5) 11-day duration treatments (S11 + M11) vs CON. Comparisons 1 and 2 were performed to evaluate the effectiveness of milk and liquid starter supplementation regardless of the duration of supplementation. Comparisons 3, 4 and 5 were performed to gain insight into the optimum duration of supplementation. Results were subsequently adjusted using posthoc Bonferroni adjustment.

Results

Mortality and removals

Mortality and pig removals from the experiment were considered as a single metric, which between weaning and slaughter was ~3.9%. Eight pigs died/were removed from the CON group, six from M4, three from S11 and six from M11. In the CON group, two pigs were removed due to tail biting, four pigs were euthanised due to joint inflammation, and two pigs had sudden deaths. In the M4 group, four pigs experienced sudden deaths and two pigs were euthanised due to leg inflammation. Three pigs were removed due to tailbiting in the S11 group. In the M11 group, two pigs were removed because of tail biting, two pigs died of sudden deaths and two pigs were euthanised because of joint inflammation.

Compositional analysis of liquid milk and diets

The analysed chemical composition of the dry pelleted starter, link, weaner and finisher diets is presented in Table 1. The average DM of the liquid starter diet samples was 12.10 ± 1.10%. The liquid milk samples contained 12.55 ± 1.02% DM, 3.11 ± 0.26% total protein and 0.90 ± 0.15% fat.

Growth performance, faecal scoring, medication usage and carcass data

The effect of postweaning treatment on pig growth performance to target slaughter weight is presented in Table 2. Pig BW at days 11, 20, 28, 47, at slaughter and overall was not affected by treatment ($P > 0.05$). There was an overall effect of treatment on ADFI ($P < 0.05$), with M11 piglets having higher ADFI than CON and S4 piglets. Between days 0–11 pw, M11 piglets had 48% higher ADFI than CON piglets ($P < 0.01$). However, the ADFI of M4, S4 and S11 piglets did not differ from that of piglets in the CON or M11 treatment groups ($P > 0.05$) during the same period. There was no effect of treatment on ADFI during any other period up to target slaughter weight ($P > 0.05$).

There were treatment differences with regard to the ADFI and total feed intake from solid feed and from liquid feed between days 0 and 11 pw. The feed intake from solid feed (both total feed intake and ADFI from day 0 to 11 pw) of M11 and S11 piglets did not differ ($P > 0.05$) with both being lower than that for CON, S4 and M4 piglets ($P < 0.001$). The ADFI and total feed intake from solid feed was similar for the CON, S4 and M4 treatment groups between days 0 and 11 pw ($P > 0.05$). The ADFI from liquid feed was 55% higher in M11 than S11 piglets ($P < 0.01$) between days 0 and 11 pw. The total feed intake from liquid feed between days 0 and 11 pw increased in the order S4, M4, S11, M11 ($P < 0.001$). The ADFI from liquid feed of M4 and M11 piglets was almost twice that of S4 and S11 piglets between days 0 and 4 pw ($P < 0.001$). During this period, the ADFI from liquid feed of piglets on both milk treatments was similar, as it was for piglets on both liquid starter treatments ($P > 0.05$). The M11 piglets had higher total net energy, total CP and total lysine intake between days 0 and 11 pw compared to CON and S4 piglets ($P < 0.05$). However, the total net energy, total CP and total lysine intake of M4 and S11 piglets were not different to that of any other treatment during the same period ($P > 0.05$).

There was an overall treatment effect on ADG from weaning to slaughter ($P < 0.001$), with M11 piglets having a higher ADG than CON piglets. The M11 piglets had a 57 and 54% higher ADG compared to CON and S4 piglets, respectively, between days 0 and 11 pw ($P < 0.001$); however, the ADG of M4 and S11 was not different to that of any other treatment during the same period ($P > 0.05$).

Table 2
Effect of supplemental milk and liquid starter diet for 4 and 11 days postweaning on growth performance of pigs from weaning to slaughter (Least square means ± SEM).

Item	CON	S4	M4	S11	M11	SEM	*P*-value
Number of pens	12	12	11	12	12		
BW (kg)[1]							
Day 0 (weaning)	8.9	8.9	8.6	8.8	8.6	0.56	0.991
Day 11	11.5	11.5	12.0	11.9	12.7	0.16	0.236
Day 20	16.0	16.0	16.3	16.5	17.1	0.30	0.457
Day 28	20.3	20.5	20.8	21.2	21.9	0.41	0.439
Day 47	34.4	35.4	35.6	36.1	37.2	0.83	0.543
Day of slaughter (~day 131 pw)	125.8	126.6	127.7	128.2	129.2	2.60	0.873
Overall	41.6	42.0	42.5	42.8	43.6	0.70	0.718
ADFI (DM basis; g/day/pig)[2]							
Days 0–11	256^{a}	273^{ab}	326^{ab}	326^{ab}	378^{b}	20.0	0.001
Days 11–20	494	493	496	509	490	13.8	0.889
Days 20–28	610	656	647	648	663	15.0	0.123
Days 28–47	965	989	993	1 026	1 051	28.2	0.243
Day 47-slaughter	2 020	2 010	1 969	1 996	2 079	40.9	0.421
Overall	869^{a}	884^{a}	886^{ab}	901^{ab}	932^{b}	13.6	0.020
ADFI from solid and liquid feed separately (DM basis; g/day/pig)							
Days 0–11 solid only	258^{a}	238^{a}	228^{a}	164^{b}	132^{b}	14.1	<0.001
Days 0–4 liquid only	–	103^{a}	221^{b}	115^{a}	192^{b}	14.8	<0.001
Days 0–11 liquid only[3]	–	–	–	158^{a}	246^{b}	17.6	0.002
Total feed intake from day 0 to 11 pw (DM basis, g/pig)							
Solid feed only	2 843^{a}	2 580^{a}	2 596^{a}	1 855^{b}	1 381^{b}	158	<0.001
Liquid feed only	–	411^{a}	952^{b}	1 727^{c}	2 703^{d}	138	<0.001
Total net energy intake from days 0 to 11 pw (MJ/pig)	34.6^{a}	36.2^{a}	42.4^{ab}	43.6^{ab}	48.2^{b}	2.68	0.004
Total CP intake from days 0 to 11 pw (g/pig)	549^{a}	575^{a}	675^{ab}	692^{ab}	771^{b}	43	0.003
Total lysine intake from days 0 to 11 pw (g/pig)	46.5^{a}	48.7^{a}	58.8^{ab}	58.6^{ab}	70.1^{b}	3.72	<0.001
ADG (g/day/pig)							
Days 0–11	224^{a}	228^{a}	287^{ab}	283^{ab}	353^{b}	15.7	<0.001
Days 11–20	494	504	488	508	495	16.1	0.911
Days 20–28	526^{a}	562^{ab}	565^{ab}	583^{b}	607^{b}	18.3	0.039
Days 28–47	739	779	780	777	808	17.0	0.094
Day 47-slaughter	1 102	1 102	1 098	1 125	1 126	23.5	0.854
Overall	617^{a}	635^{ab}	644^{ab}	655^{ab}	678^{b}	8.62	<0.001
Feed conversion ratio (DM basis; g/g)							
Days 0–11	1.15^{a}	1.16^{a}	1.12^{ab}	1.14^{ab}	1.03^{b}	0.033	0.026
Days 11–20	1.01	0.98	1.02	1.01	0.98	0.033	0.914
Days 20–28	1.18	1.17	1.15	1.11	1.09	0.033	0.212
Days 28–47	1.30	1.27	1.27	1.32	1.29	0.034	0.793
Day 47-slaughter	1.83	1.83	1.79	1.80	1.92	0.034	0.054
Overall	1.29	1.28	1.27	1.27	1.28	0.017	0.654

Abbreviations: CON = dry pelleted starter diet; S4 = CON+liquid starter diet for 4 days; M4 = CON+liquid milk replacer for 4 days; S11 = CON+liquid starter diet for 11 days; M11 = CON+liquid milk replacer for 11 days; ADFI = Average daily feed intake; ADG = Average daily gain; pw = postweaning.
$^{a\text{-}d}$Values within a row that do not share a common superscript differ significantly at $P < 0.05$.
[1] Day indicates day postweaning.
[2] ADFI from both solid and liquid feed combined.
[3] Only S11 and M11 groups were compared as only these groups had liquid supplementation throughout days 0 to 11 pw.

From days 11 and 20 pw, ADG was not affected by any treatment ($P > 0.05$). Between days 20 and 28 pw, the M11 piglets had a 15% higher ADG compared to CON piglets ($P < 0.05$), while the ADG of all other treatments was similar to that of both CON and M11 ($P > 0.05$). There was no treatment effect on ADG during the periods from days 28–47 pw or day 47 pw-slaughter ($P > 0.05$).

The M11 piglets had a better feed conversion ratio than CON and S4 piglets between days 0 and 11 pw ($P > 0.05$); however, the feed conversion ratio of S4, M4 and S11 piglets was not different to that of CON and M11 piglets ($P < 0.05$) during the same period. There was no effect of treatment on feed conversion ratio during any other period up to target slaughter weight ($P > 0.05$).

The CV of within-pen pig BW and carcass parameters, including cold carcass weight, muscle depth, fat depth, lean meat% and kill-out% are presented in Supplementary Table S1. None of these were affected by dietary treatment ($P > 0.05$).

The results of the specific hypothesis statements are presented in Supplementary Table S2. There were differences in pig BW at days 11, 20, 28 and 47 pw for the comparisons M4 + M11 vs CON and S11 + M11 vs CON ($P < 0.05$), where the CON group had a lower estimated mean compared to the other groups. Moreover, S4 + M4 had lower pig BW at day 11 pw compared to S11 + M11 ($P < 0.05$). Between days 0 and 11 and 28 and 47 pw, ADFI for CON was lower than M4 + M11 ($P < 0.05$). Between days 0 and 11 pw, the ADFI of CON was lower than S11 + M11 and the ADFI of S4 + M4 was lower than S11 + M11 ($P < 0.05$). Between days 20 and 28 pw, ADG was different between comparisons M4 + M11 vs CON; S4 + S11 vs CON; S4 + M4 vs CON and S11 + M11 vs CON ($P < 0.05$), where the CON group had a lower estimated mean compared to the other groups. Between days 0 and 11 pw, feed conversion ratio was different in the contrast statements M4 + M11 vs CON and S11 + M11 vs CON ($P < 0.05$), where the CON group had a higher estimated mean compared to the other groups. There were no other differences observed between the specific comparison groups for growth performance or carcass parameters ($P > 0.05$).

The average number of days that diarrhoea was observed in pens between days 1 and 12 pw was not affected by treatment, and is presented in Supplementary Table S3 ($P > 0.05$). Treatment did not affect the medication (antibiotic and anti-inflammatory)

usage per pig per pen, number of injections and clinical cases of disease from weaning until sale and these data are presented in Supplementary Table S4 ($P > 0.05$).

Feeding behaviour

Results relating to feeding behaviour are presented in Table 3. The CON piglets directed more feeding behaviour towards the solid feeder than any other treatment on day 2 pw ($P < 0.001$). The solid feeder-directed activity of S4, M4, S11 and M11 piglets was similar on day 2 pw ($P > 0.05$). On day 4 pw, M11 piglets had lower solid feeder-directed activity than CON, S4 and M4 piglets ($P < 0.001$) and S11 piglets had lower solid feeder-directed activity than CON and M4 piglets ($P < 0.001$). The solid feeder-directed activity of S11 and M11 piglets was similar, while that of CON, S4 and M4 piglets also did not differ on day 4 pw ($P > 0.05$). On both day 6 and 8 pw, M11 and S11 piglets had lower solid feeder-directed activity than CON, S4 and M4 piglets ($P < 0.001$). On both days, the solid feeder-directed activity of CON, S4 and M4 groups was similar and the solid feeder-directed activity of S11 and M11 groups also did not differ ($P > 0.05$). The liquid feeder-directed activity on day 2 pw was similar in the four liquid-fed (M4, S4, S11 and M11) treatment groups ($P > 0.05$) and liquid feeder-directed activity of S11 and M11 on days 4, 6 and 8 pw also did not differ ($P > 0.05$). On day 2 pw, M11 and M4 treatments had a higher percentage of non-eaters of solid pelleted feed than CON pens ($P < 0.01$). On the same day, the percentage of non-eaters of solid feed was similar in M11 and M4 pens, while that of S4 and S11 treatments were not different compared to the CON, M4 and M11 groups ($P > 0.05$). The percentage of non-eaters of liquid feed for all days and of solid feed for the days 4, 6 and 8 pw observation days could not be analysed statistically as the occurrence of more than one non-eater in a pen was rare.

Small intestinal histology and brush-border membrane enzyme activity

The results regarding morphology of the three regions of the small intestine at day 7 pw are presented in Fig. 1 with VH in Fig. 1a, CD in Fig. 1b, VH:CD ratio in Fig. 1c and a representative image of the jejunal VH and CD for CON and M11 in Fig. 1d. The jejunal VH of M11 piglets was 37% greater than that of CON piglets, and ileal VH of S11 piglets was 28% greater than that of S4 piglets ($P < 0.05$). The jejunal VH:CD ratio was higher for M11 than CON ($P < 0.05$). The jejunal VH and VH:CD ratio of S4, M4 and S11 was not different from that of CON and M11 piglets, and the ileal VH of CON, M4 and M11 was not different from that of S11 and S4 piglets ($P > 0.05$). Dietary treatment did not affect VH or VH:CD ratio in the duodenum, VH:CD ratio in the ileum or CD in the three regions of the small intestine ($P > 0.05$). The VH and VH:CD ratio in the jejunum at day 7 pw were different in the comparison CON vs M4 + M11 ($P < 0.05$), where the CON group had lower estimated means compared to the M4 + M11 group.

Treatment effects on brush-border membrane enzyme activity in all three regions of the small intestine on day 7 pw are presented in Fig. 2. (disaccharidases; sucrase = 2a, maltase = 2b, lactase = 2c) and Supplementary Table S5 (peptidases). The M11 piglets had up to 150% more ileal sucrase activity than M4, S4 and S11 piglets ($P < 0.05$). The ileal sucrase activity of CON piglets did not differ from that of any other treatment group ($P > 0.05$). The M11 piglets also had 180% more ileal maltase activity than S4 piglets ($P < 0.05$). The ileal maltase activity of CON, M4 and S11 piglets did not differ from that of the M11 and S4 piglets ($P > 0.05$). The VH and sucrase activity in the ileum at day 7 pw were different in the comparison S4 + M4 vs S11 + M11 ($P < 0.05$), where the S4 + M4 group had lower estimated means compared to S11 + M11 group. Dietary treatment did not affect disaccharidase (sucrase, maltase and lactase) activity in the duodenal and jejunal mucosa, lactase activity in the ileum or peptidase activities in the three regions of the small intestine ($P > 0.05$).

Discussion

This study examined the effect of supplementing milk and liquid starter diet to weaned pigs for 4 and 11 days pw on growth to slaughter and intestinal parameters of newly weaned piglets. To our knowledge, this is the first study to evaluate the response to these dietary strategies in weaned piglets when provided supplementary to a dry pelleted starter diet. Our results indicate that milk supplementation for 11 days in addition to ad-libitum access

Table 3
Effect of supplemental milk and liquid starter diet for 4 and 11 days postweaning on feeding behaviour of piglets on days 2, 4, 6 and 8 postweaning (Least square means ± SEM).

Item	CON	S4	M4	S11	M11	SEM	*P*-value
Number of pens	12	12	11	12	12		
Observations of individual piglets seen engaging in solid feeder trough-directed activity (%)[1]							
Day pw							
2	6.2^{a}	3.3^{b}	2.1^{b}	3.4^{b}	1.6^{b}	0.49	<0.001
4	6.2^{a}	5.3^{ab}	6.4^{a}	3.4^{bc}	2.8^{c}	0.49	<0.001
6	7.0^{a}	7.0^{a}	7.1^{a}	3.5^{b}	2.2^{b}	0.49	<0.001
8	9.0^{a}	8.9^{a}	8.3^{a}	5.0^{b}	3.9^{b}	0.49	<0.001
Observations of individual piglets seen engaging in liquid feeder trough-directed activity (%)[2]							
Day pw							
2	–	2.3	2.4	3.1	2.9	0.45	0.540
4	–	–	–	3.0	3.6	0.44	0.336
6	–	–	–	1.4	2.1	0.44	0.234
8	–	–	–	5.3	4.8	0.44	0.402
Non-eaters of solid feed per pen on day 2 pw (%)[3]	3.3^{a}	13.0^{ab}	22.7^{b}	15.8^{ab}	30.7^{b}	4.99	0.004

Abbreviations: CON = dry pelleted starter diet; S4 = CON+liquid starter diet for 4 days; M4 = CON+liquid milk replacer for 4 days; S11 = CON+liquid starter diet for 11 days; M11 = CON+liquid milk replacer for 11 days; pw = postweaning.
[a-c]Values within a row that do not share a common superscript differ significantly at $P < 0.05$.
[1] Observations of individual pigs seen engaging in solid feeder trough-directed activity as a percentage of total number of observations per day.
[2] Observations of individual pigs seen engaging in liquid feeder trough-directed activity as a percentage of total number of observations per day. Observations of piglets seen engaging in liquid feeder trough-directed activity are not applicable for CON at any time-point and for S4 and M4 for days 4, 6 and 8 pw, as liquid feed was not provided to these piglets on these days.
[3] Non-eaters of pelleted solid feed were defined as having no solid feeder trough-directed activity observations on day 2 pw.

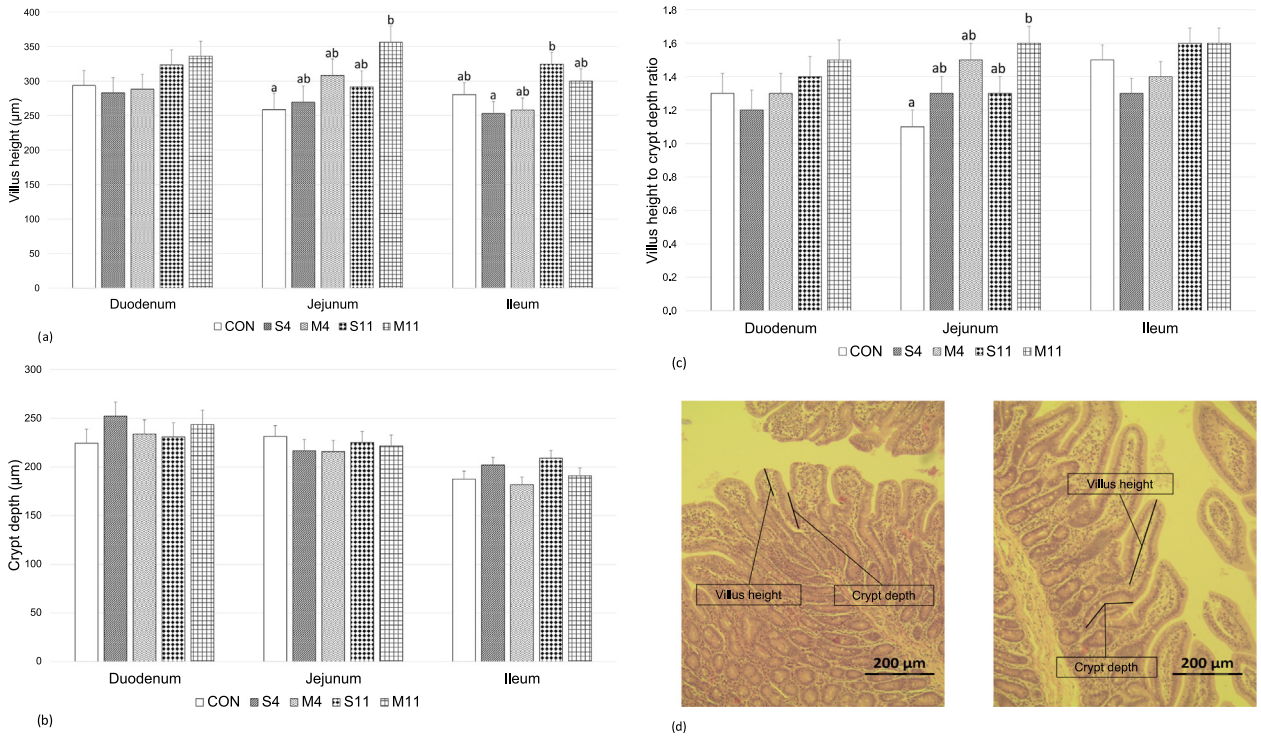

Fig. 1. Small intestinal morphology of piglets on day 7 postweaning with five dietary treatments: CON = dry pelleted starter diet; S4 = CON+liquid starter diet for 4 days; M4 = CON+liquid milk replacer for 4 days; S11 = CON+liquid starter diet for 11 days; M11 = CON+liquid milk replacer for 11 days. The bars portray the effect of the dietary treatments on the (a) Villus height, (b) Crypt depth and (c) Villus height to crypt depth ratio. Fig. 1(d) is a representative image of jejunal villus height and crypt depth for CON (left) and M11 (right). a-b Bars that do not share a common letter differ significantly at $P < 0.05$.

to dry pelleted starter diet, increased early pw feed intake and growth, relative to that of piglets provided with only dry pelleted starter diet. This growth benefit was observed during the period of supplementation and between days 20 and 28 pw. Moreover, when both durations of milk supplementation (4 and 11 days) were considered together and compared to the control group, milk-supplemented piglets had higher feed intake and growth during the period of supplementation, higher feed intake between days 28 and 47 pw and higher growth between days 20 and 28 pw, further supporting the benefits of milk supplementation. The feed intake and growth benefits observed at the end of the supplementation period are in line with previous studies which investigated pw milk supplementation strategies. For example, providing 3-week-old weaned piglets with milk plus pelleted starter diet (Zijlstra et al., 1996) or a mixture of liquid milk and pelleted diet (Dunshea et al., 1999; Rault et al., 2015) for 7 days pw increased feed intake and growth compared to piglets provided only with a dry pelleted diet. Therefore, there is a distinct and consistent benefit of supplementing liquid milk, along with a dry pelleted diet, to newly weaned piglets on feed intake and growth early pw.

In the present study, offering a starter diet in liquid form in addition to a pelleted starter diet pw (irrespective of the duration of feeding) did not result in a statistically significant increase in feed intake or growth compared to providing only dry pelleted feed. However, supplementing liquid starter for 11 days pw resulted in a 27% numerical increase in feed intake and a 28% numerical increase in growth relative to piglets fed dry feed only, which is biologically and commercially relevant. Results from previous studies that supplemented liquid diets to newly weaned piglets are inconsistent. Kim et al. (2001) and Han et al. (2006) observed increased feed intake and growth as a result of liquid feeding. However, Lawlor et al. (2002) reported increased feed intake but reduced growth with liquid feeding of weaned pigs. The authors suggested that feed wastage and uncontrolled fermentation of the liquid feed could be the reason behind this (Lawlor et al., 2002). In the present study, however, the likelihood of feed wastage and undesirable fermentation was reduced because of the automatic delivery system (with sensor checks) and preparation of two fresh mixes each day. Lawlor et al. (2002) did, however, report increased growth with acidified liquid feed. Inclusion of organic acids can reduce gastric pH which has been shown to improve nutrient digestibility and to delay the multiplication of pathogens such as enterotoxigenic *E. coli* (Suiryanrayna and Ramana, 2015). Hence, supplementing an acidified liquid starter diet for 11 days pw using the automatic delivery system used in the current study could have the potential to further increase the feed intake and growth benefits of liquid feeding observed in the current study.

The positive early pw intake and growth responses to milk supplementation are likely due in part to the liquid form in which the milk was provided (Patridge and Gill, 1993; Brooks and Tsourgiannis, 2003). However, the higher lactose levels in the milk compared to the starter diet may provide a better explanation. In the present study, the milk replacer contained ~41% lactose (Swinco, personal communication), while the starter diet contained ~23% lactose (Sauvant et al., 2004). Therefore, the average amount of lactose ingested by each piglet during the entire 11-day supplementation period is estimated as follows: 654 g for CON; 688 g for S4; 987 g for M4; 824 g for S11 and 1 426 g for M11. The inclusion of lactose-containing highly digestible milk by-products in piglet diets at weaning is important, and the benefits are well-established (Tokach et al., 2003; Mahan et al., 2004). Due to its sweetness, lactose is highly palatable for piglets and consequently leads to increased feed intake (Zhao et al., 2021). Increasing lactose levels in postweaning diets previously resulted in a linear increase in piglet feed intake and growth (Pierce et al., 2005). Moreover, fermentation of lactose in the stomach reduces

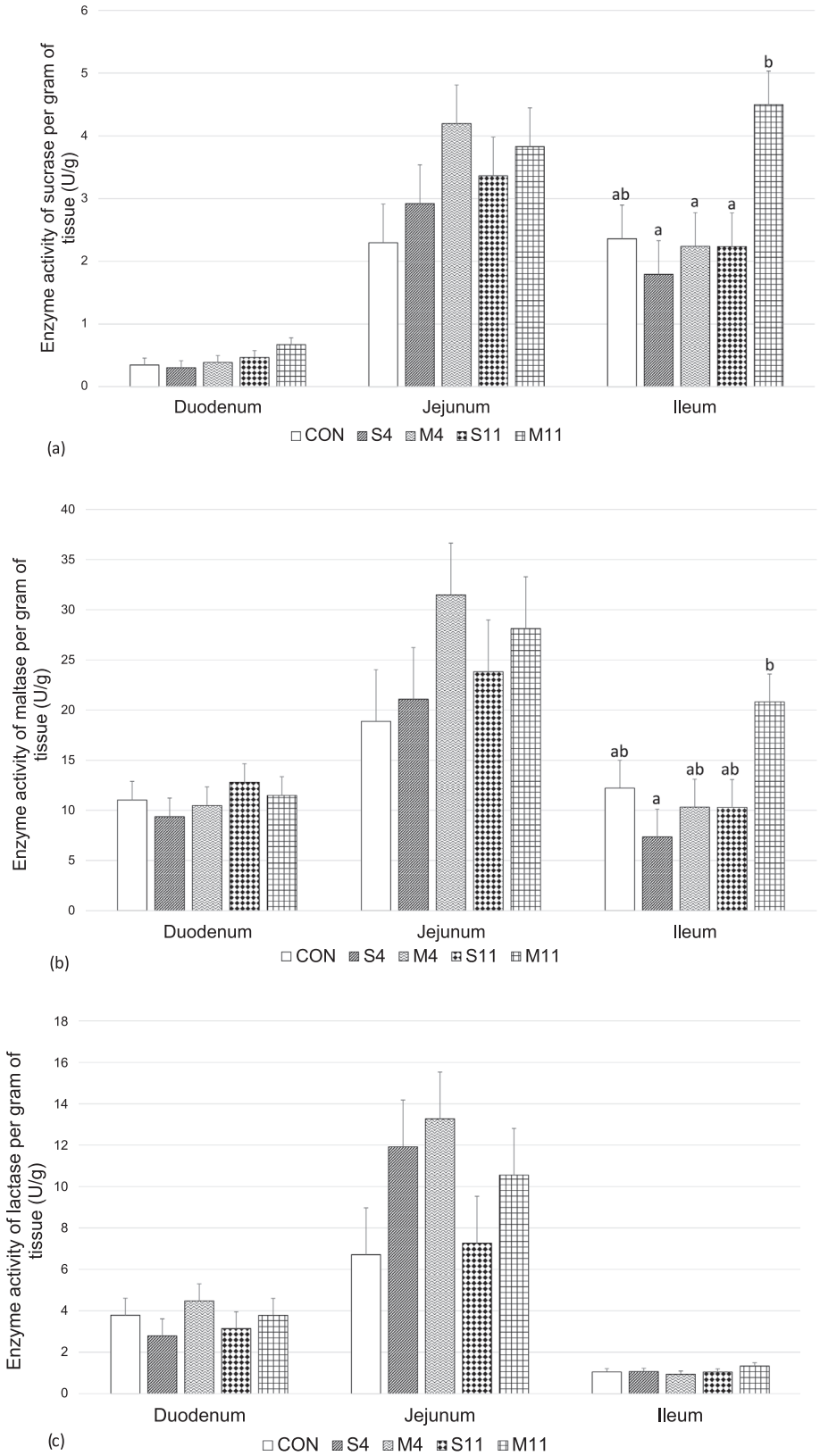

Fig. 2. Brush border membrane enzyme (disaccharidase) activities of intestinal mucosa of piglets on day 7 postweaning with five dietary treatments: CON = dry pelleted starter diet; S4 = CON+liquid starter diet for 4 days; M4 = CON+liquid milk replacer for 4 days; S11 = CON+liquid starter diet for 11 days; M11 = CON+liquid milk replacer for 11 days. The bars portray the effect of the dietary treatments on the enzyme activities of (a) sucrase (b) maltase and (c) lactase. a-b Bars that do not share a common letter differ significantly at $P < 0.05$.

gastric pH which improves protein digestion and inhibits pathogen growth (Beasley et al., 2015) at a time in the pig's life when it has a very limited ability to produce gastric HCl. Therefore, the higher levels of lactose ingested by the 11-day-milk-supplemented piglets likely contributed to increased feed intake and growth relative to the other treatments. Moreover, the milk replacer also contained

porcine spray dried plasma which is also known to increase feed intake in weaned piglets (Van Dijk et al., 2001; Zijlstra et al., 2009).

Disruption to intestinal structure, as evidenced by a reduction in VH and VH:CD ratio, is a key indicator of early pw stress (Hedemann et al., 2003). Low pw feed intake typically leads to a reduction in VH and VH:CD ratio, as the enterocytes along the intestinal mucosa die due to an insufficient supply of energy and nutrients (Dong and Pluske, 2007). In the current study, milk supplementation for 11 days pw prevented the reduction in jejunal VH which was observed in piglets weaned solely onto the dry pelleted starter diet. The increased feed intake from milk supplementation likely increased energy and nutrient supply to the enterocytes, thereby reducing the disruption of intestinal villi caused by weaning (Pluske et al., 1997). This is in line with outcomes from previous studies; for instance, weaned piglets provided with ad-libitum access to cow's or ewe's milk (without pelleted starter diet) for 5 days pw had increased jejunal VH at 5 days pw compared with piglets fed dry pelleted starter diet (Pluske et al., 1996a; b). In the current study, liquid starter diet supplementation for 11 days pw increased ileal VH, but only compared to piglets supplemented with liquid starter diet for 4 days pw. This could be due to a 'second weaning' being experienced by piglets at the end of supplementation where liquid feed was withdrawn after 4 days pw, as suggested by Zijlstra et al. (2009). Liquid starter diet supplementation for 11 days pw resulted in numerically higher ileal VH than in all of the other groups. Deprez et al. (1987) also found increased jejunal and ileal VH in piglets weaned onto liquid feed compared to those provided solely with a dry pelleted diet pw. Higher VH at this time in response to the increased energy, nutrient and water supply to the enterocytes provided by liquid feeding (Pluske et al., 1997), likely improved digestive and absorptive capacity, leading to the increased growth observed.

Brush border membrane enzymes are an integral part of digestive function, involved in carbohydrate and protein digestion. Generally, higher enzyme activity signifies increased intestinal maturity, where pigs have a greater ability to degrade and absorb complex feed ingredients (Marion et al., 2005). In the present study, lactase activity in all three regions of the small intestine examined was not affected by any of the dietary treatments employed. This was the case even for the supplemental milk treatments (M4 and M11), despite the higher levels of lactose ingested by pigs on these treatments. Similar results have been observed in previous studies where pw milk supplementation did not affect intestinal lactase activity (Pluske et al., 1996a; b). Typically, lactase activity gradually decreases as piglets age, with loss in activity accelerated by weaning (Kelly et al., 1991). This could explain the results obtained, as this age-related lactase activity reduction is suggested to be independent of the amount of lactose in the diet (Pluske et al., 2003; Forsgård, 2019). Additionally, piglets supplemented with milk for 11 days pw had higher ileal sucrase and maltase activity compared to those in the other liquid-fed groups. The increased feed and nutrient intake as a result of milk supplementation likely explains the increased enzyme activity, as decreased enzyme activity is associated with disruption in small intestinal structure as a consequence of decreased feed intake (Pluske et al., 1997; Dong and Pluske, 2007). A similar effect was previously observed, where piglets artificially reared from 3 days of age and fed a commercially sourced milk replacer had higher maltase and sucrase activity at 28 days of age compared to conventionally suckled piglets (De Vos et al., 2014). The authors hypothesised that the increased activity of these enzymes was due to the presence of stimulatory plant-based ingredients in the commercially sourced milk powder used (De Vos et al., 2014). The commercially sourced milk powder used in the current study also contained plant-based ingredients, which might be one of the reasons that the same response was observed. However, a similar response in terms of sucrase and maltase activity was not observed in liquid starter-supplemented piglets even though the starter diet contained higher levels of plant-based ingredients than the milk replacer. Therefore, increased feed and nutrient intake as a result of supplementing piglets with milk for 11 days pw, even compared with liquid feeding of a starter diet, provides a better explanation for increased enzyme activity, improved intestinal maturity and pig growth.

Supplementing milk or a liquid starter diet to piglets for 4 days pw was not sufficiently long to affect pw growth or intestinal structure/function. In particular, feeding liquid starter diet for 4 days pw did not benefit pw growth of piglets. It is likely that piglets were still in the vulnerable acute pw phase (3–5 days pw) during which major changes in gut structure and function occur (Montagne et al., 2007) when the liquid milk or starter diet was withdrawn. Continuing liquid supplementation for 11 days pw, into the pw maturation phase [>5–7 days pw, when the pig starts to adapt to the changes imposed by weaning (Montagne et al., 2007)] and beyond was beneficial for growth and intestinal structure and function of weaned pigs. The results from the specific hypotheses comparisons in the current study support this argument as the 11-day-supplemented groups had higher BW and feed intake at the end of the supplementation period and higher ileal villus height and sucrase activity than the 4-day-supplemented groups.

The dietary interventions investigated in the current study did not provide consistent long-term benefits to the growth performance of pigs, as after day 28 pw, feed intake, growth and feed conversion ratio were not affected by any of the postweaning treatments. This outcome is in line with findings from previous studies (Lawlor et al., 2002; Han et al., 2006; Rault et al., 2015), where feeding milk or liquid starter diet pw provided only a transitory benefit to piglet growth, which disappeared soon after supplementation ceased. Interestingly, the carcass weight of the 11-day milk-supplemented pigs was numerically higher (by 3.2 kg) than that of the control pigs which might be commercially relevant but further research is required to confirm this potential benefit. Nevertheless, the lack of long-term benefits reduces the economic viability of milk supplementation as milk powder is ~3 times more expensive than starter diet. The average feed cost per pig during the entire 11-day supplementation period is estimated as follows: €3.27 for CON; €3.44 for S4; €5.63 for M4; €4.12 for S11 and €9.09 for M11. If weight gain from days 0–11 is taken into account, the cost per kg gain is: €1.26 for CON; €1.32 for S4; €1.66 for M4; €1.33 for S11 and €2.22 for M11. Hence, feed costs of supplementing milk for 11 days are 2.8 times higher than the feed costs of unsupplemented pigs which is not economically viable particularly since long-term growth benefits did not result. Therefore, future studies should investigate reducing the duration of supplementation or supplementing a liquid mixture of milk and starter diet, so as to make the practice economically viable.

The pattern of solid and liquid feed consumption during the period of supplementation differed between treatment groups. Although the percentage of observations of piglets engaging in solid- or liquid feed-directed activity does not exclusively imply that piglets were consuming these feeds, it does provide an insight into their feed-directed investigatory behaviour. The treatment differences in behaviour directed towards the solid feeder over the four observation days support the feed intake data from this study; piglets provided with either milk or liquid starter for 11 days pw had lower intakes of dry pelleted starter diet than piglets on the control treatment and those supplemented with liquid starter diet/milk for 4 days pw. When access to liquid feed was removed at 4 days pw, piglets increased their solid feeder-directed activity almost immediately, as expected, and at the same rate as that of piglets on the control treatment. This suggests that when liquid

feed is available, the behaviour of piglets towards solid feed is reduced. Interestingly, supplementing liquid milk or starter diet for 4 days pw only did not reduce the total intake (from days 0 to 11) of solid starter diet compared to control piglets. However, liquid milk/starter diet supplementation for 11 days pw resulted in a reduction in total solid feed intake during the period of supplementation. This was more pronounced with milk than liquid starter supplementation, with the DM intake from milk being twice as high as that from the dry pelleted starter diet during the supplementation period.

Low feed intake at weaning is generally associated with compromised intestinal barrier and function, which can lead to the translocation of pathogens, toxins and allergic compounds across the intestinal barrier, thereby causing enteric infections and diarrhoea (Vente-Spreeuwenberg and Beynen, 2003). In the current study, although feed intake was increased and intestinal structure less disrupted by the 11-day milk supplementation strategy, medication usage and postweaning diarrhoea prevalence was not impacted. However, it should be noted that the occurrence of pw diarrhoea was low as the study was performed in a high−health status facility. Positive effects from pw milk supplementation on diarrhoea prevalence may be more likely in a disease challenge situation (e.g. enterotoxigenic *E. coli)*. The increased lactose intake from pw milk supplementation can play a crucial role in protecting piglets against pathogens, as lactose is fermented into lactic acid which reduces gastrointestinal pH (Pierce et al., 2005, 2006). Reducing gastrointestinal pH is a well-established approach to controlling enteric pathogens; for example, dietary lactic acid supplementation has been reported to reduce enterotoxigenic *E. coli* counts in the intestine of weaned piglets (Tsiloyiannis et al., 2001). Therefore, future studies investigating the effect of pw milk supplementation in a disease challenge study may be warranted.

In conclusion, supplementary milk provision to piglets for 11 days pw increased feed intake and growth during that period and increased brush-border membrane enzyme activity indicating accelerated intestinal maturity. Moreover, supplementing weaned piglets with milk and liquid starter diet for 11 days but not 4 days pw improved intestinal structure, likely increasing absorptive capacity. Thus, offering supplementary milk and liquid starter diet for 11 days pw is likely to be beneficial for growth and intestinal structure/function but only during the period of supplementation. Liquid milk replacer supplementation for 11 days pw greatly increased early pw feed intake and growth. However, as there was no residual effect on growth, feed efficiency and carcass weight, and because milk replacer is expensive relative to starter diet, it is essential that liquid milk replacer is used sparingly so that the practice is economically viable.

Supplementary material

Supplementary material to this article can be found online at https://doi.org/10.1016/j.animal.2024.101271.

Ethics approval

The experiment was conducted in accordance with the legislation for commercial pig production set out in the European Communities (Welfare of Farmed Animals) regulations 2010 and in Irish legislation (SI no. 311/2010). The care and use of the animals were approved by the Teagasc Animal Ethics Committee (Approval No. TAEC2021-306), Waterford Institute of Technology Ethics Committee (Approval No. WIT2021REC033) and the procedures performed on the animals were authorised by the Irish Health Products Regulatory Authority (project authorisation no. AE19132/P142).

Data and model availability statement

The data/models were not deposited in an official repository but are available from the authors upon request.

Declaration of Generative AI and AI-assisted technologies in the writing process

During the preparation of this work the author(s) did not use any AI and AI-assisted technologies.

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Declaration of interest

None.

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