The effect of group composition and mineral supplementation during rearing on the behavior and welfare of replacement gilts

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ABSTRACT: Sow longevity supported by good health and reproductive performance is necessary to optimize sow lifetime performance. In some countries, replacement gilts are reared with finisher pigs destined for slaughter, so they are exposed to sexual and aggressive behaviors performed by males. This is associated with stress and injury. Moreover, diets formulated for finishers are not designed to meet the needs of replacement gilts and may not supply the necessary minerals to promote limb health, optimal reproduction, and, thus, sow longevity. In this 2 × 2 factorial design experiment with 384 animals (32 pens [12 animals per pen]), we investigated the effect of female-only (FEM) or mixed-sex (MIX) rearing, with (SUPP) or without (CON) supplementary minerals (copper, zinc, and manganese) on locomotion, salivary cortisol levels, behavior, body lesions (BL), and hoof health of gilts. The experimental period began at transfer to the finisher stage (day 81.3 ± 0.5 of age; day 0) until breeding age (day 196 \pm 0.5 of age; day 115). Locomotion was scored (0–5) biweekly from day 0 until slaughter day 67 or breeding age day 115 for the remaining gilts. Saliva samples were taken monthly from four focal gilts per pen. All counts of aggressive, harmful, sexual, and play behavior were recorded by direct observation 1 d biweekly (5- ×

5-min observations/pen/d). BL scores were recorded on focal pigs biweekly from day 1 until day 99 on the back, neck, shoulder, flank, and hind quarter on each side of the body. Hind hooves were scored for eight disorders (heel erosion [HE], heel sole separation [HSS], and white line separation [WLS], dew claw length and dew claw cracks, toe length and both vertical and horizontal toe cracks) by severity, and a total hoof lesion score was calculated by summing individual scores. General linear mixed models were used to analyze cortisol, behavior, BL, and total hoof scores. Generalized linear mixed models were used for locomotion, bursitis and individual hoof disorders. There was less aggression (P < 0.05) and sexual behavior in the FEM compared to the MIX groups with more play behavior in MIX compared to FEM groups (P < 0.01). Gilts in the MIX groups had higher BL scores than gilts in the FEM groups (P < 0.001). Total hoof scores were higher in MIX (8.01 ± 0.15) than FEM $(7.70 \pm 0.12; P < 0.02)$ gilts. CON diet gilts had higher HE scores than SUPP gilts (P < 0.05). HSS (P < 0.05) and WLS (P < 0.05) scores were higher in MIX than FEM gilts. Rearing gilts in FEM groups had benefits for hoof health likely mediated through lower levels of activity due to male absence, and minerals helped reduce HE.

Key words: gilt development, hoof disorders, hoof health, lameness, pig welfare, trace mineral nutrition

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INTRODUCTION

In many countries, replacement gilts are reared with pigs destined for slaughter, including entire males (Quinn, 2014). This means that they are exposed to high levels of aggressive and sexual (i.e., mounting) behavior (Boyle and Bjorklund, 2007; O'Driscoll et al., 2013; Teixeira and Boyle, 2014). These behaviors increase the risk of injuries such as limb and cartilage damage (Andersson et al., 2005), which are associated with lameness, a major cause of culling in early parities (Dewey et al., 1993; Jensen et al., 2007; Anil et al., 2009; Quinn, 2014). Old data on Irish culling patterns showed that roughly 32% of animals culled for lameness had only produced a single litter (Boyle et al., 1998). It is unlikely that this has changed greatly considering that the sow replacement rate in Ireland was 54.5% in 2017, the second highest of the InterPIG countries (InterPIG, 2017; InterPIG is an independent economic comparison of pig production in 17 countries, mostly in Europe, and the United States, Canada, and Brazil).

Finisher pig diets are not designed to meet the needs of a growing replacement gilt and may not supply appropriate trace minerals to promote longevity (Quinn et al., 2015). The addition of zinc (Zn), copper (Cu), and manganese (Mn) to the diet of breeding sows reduces the incidences of heel erosion (HE), heel overgrowth, white line lesions (Nair and Anil, 2011), lameness, and leg abnormality prevalence (Ferket et al., 2009). Thus, these minerals could also be beneficial to growing gilts. Zn and Mn are particularly important for hoof horn production. Zn has a vital role in cell repair and replacement, and Mn in the formation and maintenance of cartilage and bone in cattle; deficiency is associated with short/weak bones (Mohamadnia, 2008; van Riet et al., 2013). Cu is essential for antibody development and lymphocyte replication (Tomlinson et al., 2004).

Previous work found improved locomotion scores, reduced weight gain, average daily gain (ADG), and average daily feed intakes (ADFI) in gilts fed a restricted diet formulated for fat rather than lean deposition and supplemented with minerals (Quinn et al., 2015). However, that study used terminal line gilts and, furthermore, restricted feeding is difficult to conduct in practice as most maternal line gilts are reared in finisher pig accommodation (Quinn et al., 2015). We hypothesized that simple addition of trace levels of Cu, Zn, and Mn to a standard finisher diet would improve limb health, relative to gilts without supplementation,

and could ameliorate any negative effects on limb and hoof health, and stress levels, of rearing replacement gilts with males. Ultimately, we expected that the combination of mineral supplementation and single-sex rearing would help to optimize the growth, physical development, and welfare of young female pigs destined to enter the breeding herd.

MATERIALS AND METHODS

Care and Use of Animals

This study was carried out in the 200 sow unit at the Pig Development Department in Moorepark, Fermoy, Co., Cork, Ireland between December 2016 and August 2017. The experimental work was authorized by the Teagasc animal ethics committee (Approval no.: TAEC136-2016) and licensed by the Health Products Regulatory Authority (License no.: AE19132-P057).

Experimental Design and Treatments

In August 2016, 52 sows in the Moorepark herd were served using maternal line semen from Landrace sires (0153H Longo and 0096H Grande from Hermitage Pedigree Pigs Ltd., The Hermitage, Sion Road, Kilkenny, Ireland). Four batches of sows were served to create four replicates, each 3 wk apart (average 13 sows per replicate). At farrowing, these sows provided a pool of 677 piglets for use in the experiment. The experimental period was from February 2017 (transfer to finisher house of the first replicate at 81.3 ± 0.5 d of age; experimental day 0) to August 2017 (breeding age of the last replicate, 196.3 ± 0.5 d of age; experimental day 115). From this point forward, in the manuscript, all experimental time points will be referred to as experimental days instead of days of age.

All piglets were weighed and tagged at birth, then back-tested at 21.6 ± 2.38 d of age. For this, each piglet was lifted out of the pen and placed on its back on a V board, then held in place by the experimenter. The right hand was placed on the piglet's thorax with the forefinger between the front legs, and the hind legs were held using the other hand. The test started when the piglet remained still for 3 s and lasted for exactly 60 s, determined using a stopwatch. The number of resistance attempts (struggles) was counted. At weaning $(27.4 \pm 0.4 \text{ d of age})$, pigs were weighed, then blocked on the basis of sow, sire, sex, back-test score, and wean weight and assigned to 1 of 32 (eight pens per treatment)

separate groups of 12 pigs (96 ± 16.9 pigs per replicate). Each pen had an even distribution of sire (Longo or Grande) and back-test score (av. 2, range 0–5), and a wean weight average of 7.8 ± 1.5 kg. Half of the groups contained only females (FEM) and half of the groups were mixed (MIX, six males and six females). Four focal gilts were selected from each pen according to their back-test scores; one was a low responder (score 0–1), two were picked as medium responders (score 2–3), and one was a high responder (4–5). Any piglet with a score of 5+ was not included in the trial.

Pigs were assigned to their treatment groups in a 2×2 factorial design with the first factor being group composition (FEM or MIX as described above) and the second factor being mineral supplementation. The control diet (CON) represented a standard finisher diet used on commercial pig farms. This was fed to all pigs from entry to the finisher stage (day 0) until 5 wk into the finisher stage (day 36) when pigs in half of the groups were switched to a similar diet but supplemented (SUPP) with Availa Sow minerals (Zinpro Corp, Eden Prairie, MN, USA; Table 1). This provided additional Cu, Zn, and Mn (Table 2). Levels of Cu, Zn, and Mn exceeded NRC (2012) recommendations for a finisher diet. The supplemented diet was formulated by taking the gestating and lactating sow NRC recommendations into account. All feed was in dry-pelleted form (3 mm).

The pigs were in the weaner stage for 6 wk postweaning (i.e., until day 0), at which time they were moved to the finisher stage accommodation. They remained in the finisher accommodation in the same groups for 10 wk (i.e., day 67); at this time, half of their pen mates (all of the male pigs and 50% of the females) were sent for slaughter. The remaining 182 gilts stayed in the same pens (six females per pen) until day 115 when 102 of these gilts were sent for slaughter (weighing 143.6 \pm 12.8 kg). The remaining 80 gilts were kept for breeding as replacements.

In both the weaner and finisher accommodation, pigs had access to freshwater ad libitum from a drinker and to feed ad libitum from a single space trough feeder. Weaners were fed a starter feed for the first week, a link feed for the following 2 wk, and, thereafter, a standard weaner diet. The weaner pens $(2.4 \times 2.6 \text{ m})$ had plastic slatted flooring, and the finisher pens $(4 \times 2.4 \text{ m})$ had fully slatted concrete floors. The rooms used for the trial contained either 10 or 30 pens. All rooms were mechanically ventilated. The temperature of the weaner rooms was kept at 27–28 °C for 1 wk postweaning and

Table 1. Details of the finisher (CON; fed to all pigs from day 0 to day 115) and supplemented (SUPP; for half of the pigs from day 36) diets which were used in the study

======================================		
Ingredients	CON	$SUPP^a$
Barley	50	50
Wheat	33.50	33.38
Soybean (47% CP)	12	12
Soya oil	1	1
Lysine HCl	0.4	0.4
dl-Methionine	0.1	0.1
1-Threonine	0.12	0.12
Premix ^b	0.1	0.1
Availa Sow ^c	0	0.1
Phytase	0	0
Salt feed grade	0.5	0.5
Di-calcium phosphate	1.3	1.3
Limestone flour	1	1
	100.02	100.00
Chemical composition		
Dry matter	89.8	89.8
Crude protein	15.56	15.56
Crude fiber	3.74	3.77
Total oil	5.06	5.06
Ash	4.48	4.48
Lysine	0.969	0.969
Threonine	0.639	0.639
Methionine	0.337	0.337
Methionine and cysteine	0.639	0.639
Tryptophan	0.182	0.182
Calcium	0.779	0.779
Phosphorous	0.609	0.608
Digestible phosphorus	0.280	0.280
Digestible energy, MJ of DE/kg	13.50	13.49

^aSupplemented with Availa Sow (Zinpro Corp).

reduced by 2 °C every 2 wk. Heating was generated by hot water pipes (controlled via computer). Finisher rooms were maintained at 20 °C, with no heating, and mechanically ventilated. Artificial lighting was on from 0700 to 1700 hours. There was also natural lighting through the windows. Light to darkness cycle was approximately 12:12 (h) to allow for normal circadian rhythm.

Weaner pigs were provided with an EASYFIX LUNA 117 (Perssepark, Ballinasloe, Co., Galway, Ireland) floor toy, and finisher pigs were provided with two sources of enrichment—a wooden post attached to the side of the pen by a plastic holder and a rubber chew toy (EASYFIX ASTRO 200 Perssepark, Ballinasloe, Co., Galway, Ireland) hanging overhead on chains. Pigs were inspected

 $^{^{\}it b}$ Premix provided per kilogram of diet: Cu, 15 mg; Zn, 30 mg; Mn, 0 mg; Fe, 24 mg; I, 0.15 mg; Se, 0.4 mg.

 $^{^{\}circ}$ Availa Sow provided per kilogram of complete diet: Cu, 10 mg; Zn, 50 mg; Mn, 20 mg.

twice daily and sick/injured animals were treated immediately. Gilts were continually monitored from weaning to the end of the experiment and all instances of disease, illness, and death were recorded.

Live Weight

All animals were weighed individually at the entry to the finisher stage accommodation (day 0), when the experimental diet was applied (day 36), at slaughter (day 67), and, for the remaining gilts, at breeding age (day 115).

Locomotory Ability

Gilts were locomotion scored every 2 wk $(14.1 \pm 3.6 \text{ d})$ from day 0 until they left the experiment (i.e., either at slaughter day 67 or at breeding age day 115). Thus, most gilts had at least seven inspections, with those retained until breeding age having two additional inspections. Locomotory ability was scored while the gilts walked on solid concrete for a distance of approximately 30 m from the front, rear, and side of the animal. All observations were carried out by one trained observer. Locomotion was assessed using an adapted version of Hartnett et al. (2019) ranging from 0 to 5 (Table 3).

Salivary Cortisol

One saliva sample was collected from each of the focal gilts once per month between 0930 and 1000 hours during the finisher stage (i.e., four samples per gilt). Gilts remained in their home pens during saliva collection and chewed on a large cotton bud (Salivette, Sarstedt, Wexford, Ireland) until it was thoroughly moistened for approximately 30-60 s. The samples were placed in tubes and centrifuged for 15 min at $2500 \times g$, then stored at -20 °C until analysis.

Saliva samples were analyzed using a commercially available cortisol assay kit (Salivary Cortisol kit, Salimetrics Europe Ltd., Suffolk, UK). Cortisol was detected at a minimum concentration of $<0.003 \mu g/dL$. The inter-assay CV (n = 24) was 4.69%, and intra-assay CV (n = 794) was 7.68%.

Animal Behavior

Animal behavior was recorded by direct observation for 1 d every 2 wk (every 14 ± 0.5 d) from day 10 of the experiment. All pens were observed for 5 min five times (i.e., 25 min in total per pen) between 0945 and 1708 hours. All occurrences of aggressive, sexual, and play behavior were recorded (Table 4). Observations were equally distributed across pens for each recording period.

BL Scores

Focal gilts were scored for BL caused by aggression every 2 wk from experimental day 1 until day 99 (i.e., seven recordings per gilt). BL were scored on the back, neck, shoulder, flank, and hind quarter on each side of the body and, then, summed so

Table 2. Mineral inclusion rates in the diets

	NRC, mg/kg ^a	CON, mg/kg	SUPP, mg/kg	CON %b	SUPP %b
Mn	25	25.1	51.45	101	206
Zn	100	55.6	122.29	56	122
Cu	10	4.5	17.89	45	179

Values are expressed as mg/kg, i.e., parts per million and as percentage of NRC recommendations for gestating and lactating sows.

Table 3. Scoring system for locomotory ability (Hartnett et al., 2019)

Score	Description
0	Even strides. Pig is able to accelerate and change direction rapidly
1	Pig appears stiff. Abnormal stride, which isn't easily identified. Movements no longer fluid but pig still able to accelerate and change direction rapidly. Caudal swagger evident
2	Uneven stride. Sensitivity while walking detected on at least one limb. Pig able to accelerate and change direction. Caudal swagger evident
3	Uneven stride, with a stagger. Minimum-weight bearing on affected limb. Slow to move. Obviously lame even to the untrained observer
4	Pig may not place affected limb on floor
5	Does not move

^aNRC gestating and lactating sow requirements.

^bValues in the CON (control) diet and SUPP (mineral supplemented) diet as percentage of the NRC recommendations.

Table 4. Ethogram for the recording of behavior in mixed-sex (MIX) and female-only (FEM) groups of pigs on two different diets (adapted from O'Driscoll et al., 2013)

	Behavior	Description
Aggression	Inverse pressing	Inverse parallel pressing, the pigs' face front to front pushing their shoulders hard against one another throwing head against neck/flanks of other. Bites can be directed towards the head, neck, flanks of other.
	Parallel pressing	Parallel pressing, the pigs stand side by side and push hard with shoulders against each other throwing the head against the neck/head of other pig. Bites can be directed toward the head, neck, and flanks of other.
	Attack	One pig vigorously bites another and the recipient attempts to get away without trying to defend themself/retaliate.
	Bite	Pig bites another pig with a vigorous movement of the head: mouth open, contact made with body
	Head knock	As for threat but makes contact with head against recipient pigs body
	Threat	Pig swipes head vigorously in the direction of another pig
	Displaced	Pig with head in feeder displaced by another pig due to a threat, bite, climb etc.
Harmful	Tail bite	Pig forcefully bites down on another pigs tail—often reflected in reaction (vocalization, fast movement away) by the recipient
	Ear bite	Pig takes another pigs ear in its mouth and closes jaws around it—often associated with vocalization or swift head movement to extract its ear by recipient pig
	Flank/shoulder bite	Open mouthed bite directed to pigs flank or shoulder
	Bite other	Biting aimed at another part of the body, e.g., leg. Not to be confused with aggression
	Belly nosing	Vigorous nosing of another pigs belly
	Climb	Pig places front legs on the back of another either to displace from feeder
	Vocalization	Loud, shrill "scream" made by pig in response to aggression or being mounted
Sexual	Sexual mount	Pig mounts another by placing both forelimbs on either side of the recipients flanks, sternum rests on mounted pigs loins, may be accompanied by thrusting movements and/or vocalizations by the mounted pig
	Sexual nudging	The boar noses the flanks and vulva of female prior to mounting.
Play	Head jostling	Two pigs engage/toss their heads together as they move around, mouths often open but not biting
	Individual play	Pig prances or jumps up in the air and often changes orientation of the body in mid air
	Locomotory play	Pig canters from one side of the pen to another often associated with head tossing and prancing

Table 5. Scoring system for body lesions (O'Driscoll et al., 2013)

Score	Description
0	No lesion
1	1 small lesion
2	>1 small lesion ^a or 1 red lesion
3	>1 red lesion
4	1 deep red lesion. Skin has been broken and either fresh blood or scabbing present
5	>1 deep red or 1 big lesion. Big lesions are as per deep red but cover a greater area than a linear lesion
6	>1 big lesion

 $^{o}\text{Small}$ lesion refers to any lesion that is light in color and less than 5 cm in length.

that there was one score per gilt at each inspection. The scoring system is described in Table 5.

Hoof Lesion and Bursa Scores

Hoof scoring was carried out by visual inspection on three occasions on days 36, 70, and 113. The gilts were raised 0.75 m above the ground using a hydraulic chute (FeetFirst Sow Chute; Zinpro Performance Minerals, Eden Prairie, MN, USA).

The medial and lateral toes, medial and lateral dew claws, sole, and heels of both hind feet were inspected and the severity of the following lesions was scored: HE, heel-sole (HS) separation, white line separation (WLS), dew claw length, dew claw cracks, toe length, and vertical and horizontal toe cracks. The scoring system was a modified version of the FeetFirst claw lesion scoring guide from Zinpro Corporation. Details of the scoring system are described in Table 6.

While the animals were raised in the crate, the hind legs were palpated and bursas were scored as follows: 0 = none, 1 = hazelnut, 2 = walnut, 3 = hens egg (Mouttotou et al., 1998). If more than one bursa were present, each was scored separately and was summed together for analysis.

Statistical Analysis

Data were analyzed using SAS v 9.4. The individual gilt was used as the experimental unit for Live weight (LW), locomotory ability, salivary cortisol, and body and hoof lesion and bursa scores. The group was the experimental unit for behavior analysis. Results were deemed statistically significant

 Table 6. Scoring system for hoof lesions (adapted from Zinpro Feet First Lesion Scoring System)

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Scores	Toe length	DCL	DCC	HE	HSS	WLS	HORIZ crack	VERT crack
0	Normal	Normal	Normal	Normal	Normal	Normal	Normal	Normal
-	One or more toes slightly longer than normal: difference detected only when sole is pressed flat	Slightly longer than normal	Slightly longer than Short crack or cracks normal	Flaking of the skin of the heel	Separation at the junction has just begun; length of separation <0.5 cm	Shallow and/or short separation along white line	Hemorrhage evident, short/ shallow hori- zontal crack in toe wall	Short/shallow vertical crack in wall
7	One or more toes moder- Significantly lo ately longer than normal: than normal difference obvious without flattening sole	Significantly longer than normal	Long but shallow crack or cracks in dew claw wall	Slight overgrowth and/or erosion in soft heel tissue	Separation slightly longer Long separation along than score 1 but still white line shallow	rLong separation along white line	ĭ	ong but shallow Long but shallow horizontal crack vertical crack in toe wall in wall
т	One or more claws much longer than normal and/ or the toes are torn and/ or partially or completely missing	One or more claws much longer than normal and/or the claws are torn	Multiple or deep crack or cracks in dew claw and or/partially or completely missing	Numerous cracks with obvious overgrowth and erosion	Long separation at the juncture, which is deeper than score 2	Long and deep separation along white line	Multiple or deep Multiple or deep horizontal crack vertical crack or cracks in toe or cracks in the wall wall	Multiple or deep vertical crack or cracks in the wall
4	One or more toes signifi- cantly longer than normal			Large amount of erosion and overgrowth with cracks	Long and deep separation at the juncture			

when α level was below 0.05, and a significant tendency was considered when α level was between 0.05 and 0.1. The Tukey–Kramer adjustment was used for multiple comparisons where least squares means (LS means) were determined and *P*-values were adjusted. Degrees of freedom were estimated using Kenwood–Rogers adjustment. Data are presented as LS means and standard errors. PROC UNIVARIATE was used initially for evaluating data distribution.

For all models, fixed effects included group composition (MIX vs. FEM), dietary treatment (CON vs. SUPP), where diets were applied (day 36), and the interaction, as well as replicate. Inspection day was included as a fixed effect for LW, behavior, cortisol, and hoof score analysis. For all analyses, only female pigs were included and, for cortisol and BL, only focal gilts were scored. Pen was included as a random effect.

In all analyses where the mixed procedure was used, residuals were examined to verify normality and homogeneity of variances. General linear mixed models (PROC MIXED) were used to analyze live weight, cortisol, behavior, BL, and total hoof scores. Generalized linear mixed model (PROC GLIMMIX) was used for locomotion and bursitis. And (GENMOD) was used for individual hoof disorders.

Locomotion scores were analyzed using a generalized linear mixed model (PROC GLIMMIX) as the data were ordinal in nature. As there were extremely low numbers of gilts with a score of greater than 2 (n=16), these were recategorized as greater than or equal to 2. A multinomial distribution was specified. The model could not converge when data from all weeks were included as repeated measures and, as such, each week was analyzed separately and P-values adjusted post hoc using a bonferroni adjustment. Fixed effects on all inspection dates included group composition (MIX vs. FEM) and replicate. Dietary treatment (CON vs. SUPP) and the interaction were included for inspection dates after the minerals were added to the diet.

Salivary cortisol was analyzed using a general linear model (PROC MIXED). For this analysis, fixed effects were as above, with the addition of plate as random effects. We used a log transformation to normalize the data. Inspection day was included in the analysis as a repeated measure.

Behavior data were also analyzed using a general linear model (PROC MIXED). Behaviors were summed into four categories: 1) aggressive, 2) harmful, 3) sexual and 4) play (categorized in Ethogram; Table 4) Statistical analysis for "aggressive, harmful,

and play" behaviors were analyzed individually. Behavior data were summarized per 5-min recordings and divided by the number of pigs present in the pen. Fixed effects included group composition, diet, and replicate. Pen was included as a random measure and week as a repeated measure. Due to its low occurrence in FEM groups, sexual behavior could not be statistically analyzed, so the average performance of behavior was calculated instead.

BL score analysis was carried out using a general linear model (PROC MIXED). Fixed effects included group composition diet, interactions, and rep. Inspection day was included as a repeated measure.

A total hoof score for each female pig was generated by summing the score for all hoof disorders identified. These were analyzed using linear mixed models (PROC MIXED). The individual hoof disorders were also analyzed individually using generalized linear models (PROC GENMOD) as the data were ordinal, using a similar model to that used for locomotion scores. Gilts were categorized based on their maximum scores for each disorder. Fixed effects included group composition, diet, replicate, and their interactions, with inspection day being a repeated measure.

Bursitis scores were analyzed using a generalized mixed model (PROC GLIMMIX) as the data were ordinal. Fixed effects included group composition, diet, rep, and interactions, with a random effect of inspection day.

RESULTS

Live Weight

There was no effect of either diet or group composition on live weight. However, there was an interaction between group composition and

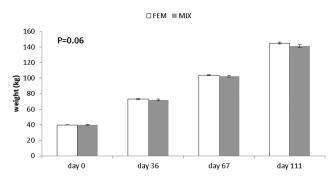


Figure 1. The live weight (kilograms) of gilts on the FEM (female only) or MIX (mixed sex) treatments on each recording day. There tended to be an interaction between group composition and inspection day (P < 0.06). *Experimental day 0 was at 81 d of age.

experimental day; gilts in the FEM groups tended to gain more weight than gilts in the MIX groups by the end of the experiment (P < 0.1; Figure 1).

Locomotory Ability

There was no effect of either group composition or mineral supplementation on locomotion score on any observation day. The median and interquartile ranges during the experiment were all 1.

Salivary Cortisol

There was no effect of group composition or mineral supplementation on cortisol levels, although there tended to be an interactive effect between group composition and sample point (P < 0.1; Figure 2). There was an effect of inspection day on cortisol levels, but there was no discernible pattern over time (P < 0.001).

Animal Behavior

The amount of aggression observed was greater in the MIX than FEM groups (Table 7). There was an effect of week (P < 0.01) and, in general, aggressive behaviors increased over time (weeks 2–6); however, they declined in weeks 8–10. There was no effect of group composition or diet on the performance of harmful behavior, although there was an effect of week with week 4 being the highest. Play behavior was observed more often in the MIX pens (P < 0.01; Table 7). There was an effect of week on play behavior with play behavior peaking in week 6 and declining in weeks 8 and 10. The mean number of sexual behaviors performed in FEM was $0.000 \pm 0.005/5$ -min/pig compared to $0.036 \pm 0.104/5$ min/pig in MIX groups.

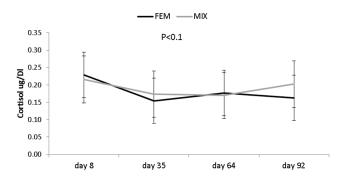


Figure 2. Salivary cortisol levels in gilts reared either in FEM (female only) or MIX (mixed sex) groups from weaning. There tended to be an interaction between diet and inspection day (P < 0.1). *Experimental day 8 was at 89 d of age.

Table 7. Average number of occurrences of aggressive, harmful, play and sexual behaviors performed per 5-min observation of pens of pigs in either mixed-sex (MIX) or female-only (FEM) groups and on a standard finisher diet (CON) or on a CON diet supplemented with minerals (SUPP)

	Diet		'	Group composition		
	CON	SUPP	P-value	MIX^a	FEM	P-value
Aggression ^b	0.495 ± 0.031	0.566 ± 0.031	0.119	0.589 ± 0.031	0.472 ± 0.031	0.013
$Harmful^b$	0.436 ± 0.029	0.482 ± 0.029	0.252	0.485 ± 0.029	0.433 ± 0.029	0.193
$Play^b$	0.038 ± 0.006	0.030 ± 0.006	0.191	0.044 ± 0.006	0.024 ± 0.006	0.002
Sexual ^c	_	_	N/A	0.036 ± 0.104	0.000 ± 0.005	N/A

 $^{^{}a}GC = Group composition.$

^cThe mean number of sexual behaviors per 5 min/pig with standard deviation.

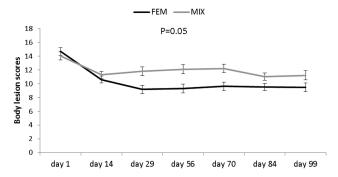


Figure 3. Total body lesion scores of gilts reared in female-only (FEM) or mixed-sex (MIX) groups at seven inspection days during the finisher period (days 82–196 of age). *Experimental day 1 was at 82 d of life.

BL Scores

Gilts in the MIX groups had higher BL scores than gilts in the FEM groups (P < 0.001). However, there was no effect of diet on BL scores. There was an effect of inspection day on lesion scores (P < 0.001), although no clear pattern over time. There was an interaction between group composition and inspection day (P = 0.05; Figure 3) and a tendency for an interaction between diet and inspection day (P = 0.1).

Hoof Lesion Scores

Total hoof lesion scores were higher (i.e., worse) in the MIX (8.01 \pm 0.15) gilts than in the FEM (7.70 \pm 0.12) gilts (P = 0.12). There was also an effect of inspection day, with hoof lesion scores increasing over time (inspection day 1, 6.39 \pm 0.1; day 2, 7.69 \pm 0.13; day 3, 9.48 \pm 0.20; P < 0.001)

Gilts on the CON diet had higher HE scores than SUPP gilts (P < 0.05; Table 8). There was also an interaction (P = 0.05) between diet and inspection day with regard to HE scores, where scores increase over time with CON gilts scoring higher than SUPP gilts at each inspection day. There were no other effects of mineral supplementation.

HS separation and WLS scores were higher in gilts reared in MIX groups compared with FEM reared gilts (P < 0.05 for both). For both horizontal cracks (P < 0.02) and dew claw cracks (P < 0.03), there was an interaction between group composition and inspection day where the percentage of animals with a score of 2 increased over time (Figure 4). There was also an interaction between diet and inspection day for dew claw length (P < 0.04) with no discernible pattern over time (Figure 5) and a tendency for an interaction between diet and inspection day on WLS (P = 0.1), where the percentage of animals with scores of 1 and 2 are increasing over time. HE, WLS, dew claw cracks, vertical toe cracks, and toe length all increased with time (P < 0.001); however, horizontal toe cracks fluctuated over time (P < 0.001).

Bursa Scores

There was no effect of group composition or mineral supplementation on limb lesion scores. However, there was an effect of inspection day on bursa scores (P < 0.001), with bursa size increasing over time.

DISCUSSION

Improvements in the management of replacement gilts during rearing could help to improve sow longevity (Quinn, 2014), primarily through reduced culling for lameness with associated benefits to animal health and welfare. This study investigated the effect of supplementation with additional Cu, Zn, and Mn and the effect of rearing in female-only or mixed-sex groups on behavior and welfare of replacement gilts. In general, few effects of mineral supplementation were found, while rearing gilts in single-sex groups reduced their exposure to aggression and sexual behavior with associated benefits to their health (reduced injuries to the body and hooves).

^bResults are the LS means and standard errors of the number of occurrences over a 5-min period divided by the number of pigs in the pen

Table 8. Individual hoof disorders of gilts reared in either mixed-sex (MIX) or female-only (FEM) groups and on a standard finisher diet (CON) or on a CON diet supplemented with minerals (SUPP)

	D	iet	Group composition		mposition	
	CON	SUPP	P-value	MIX^a	FEM	P-value
Number of animals, n ^b	352	343	_	250	445	
Heel erosion ^c	1 (1–2)	1 (1–1)	0.02	1 (1–1)	1 (1–1)	0.70
Heel-sole separation ^c	0 (0-1)	0 (0–1)	0.94	0 (0-1)	0 (0–1)	0.04
White line separartion ^c	0 (0-1)	0 (0–1)	0.51	0 (0-1)	0 (0–1)	0.03
Vertical cracks ^c	0 (0-0)	0 (0-0)	0.16	0 (0-0)	0 (0-0)	0.76
Horizontal cracks ^c	0 (0-0)	0 (0-0)	0.68	0 (0-0)	0 (0-0)	0.95
Dew claw length ^c	0 (0-0)	0 (0-0)	NS	0 (0-0)	0 (0-0)	NS
Dew claw cracks ^c	0 (0-0)	0 (0-0)	NS	0 (0-0)	0 (0-0)	NS
Toe length ^c	1 (1–1)	1 (1–1)	0.38	1 (1–1)	1 (1–1)	0.82

^a6 males and 6 females per group.

^cData are provided as median and interquartile ranges.

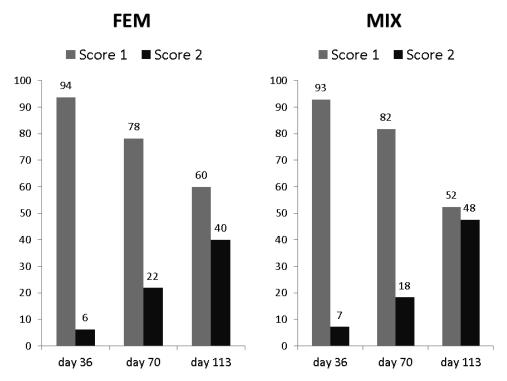


Figure 4. Details of the percentage of animals with either a heel erosion score of 1 or a score of 2 in FEM (female only) groups and in MIX (mixed-sex groups). *Experimental day 36 was at 117 d of age (the day the animals received their experimental diets).

As expected, the amount of aggression and sexual behavior observed was greater in the mixed-sex groups than in the female-only groups, which is in line with previous studies on pigs (Andersson et al., 2005; Björklund and Boyle, 2006; Rydhmer et al., 2006; Boyle and Bjorklund, 2007; O'Driscoll et al., 2013; Teixeira and Boyle, 2014). Indeed, there was generally more activity in the mixed-sex pens, with numerically more instances of harmful behavior and significantly more play behavior also observed. As male pigs mature, their levels of testosterone rise and, therefore, it is not surprising that levels

of mounting and aggression rose as these animals aged (Giersing et al., 2000); indeed, immune castration has been demonstrated to induce calmness and reduce aggression and general activity in male pigs (Aluwé et al., 2016; Heyrman et al., 2019). However, aggression decreased in week 10, which may correlate with studies that suggest that aggression reduces as the animals get older due to a greater focus on mounting (Clark and D'Eath, 2013). An alternative hypothesis is that this may be due to space restrictions, limiting space for parallel pressing, and, therefore, interfering with aggressive motor patterns (Straw, 2013).

^bTotal number of assessed animals per group.

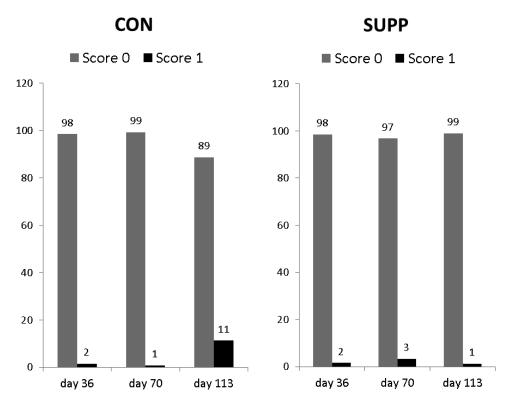


Figure 5. Details of the percentage of animals with either a dew claw crack score of 0 or a score of 1 in CON (contol diet) groups and in SUPP (supplemented diet) groups. *Experimental day 36 was at 117 d of age (the day the animals received their experimental diets).

We hypothesized that gilts reared with males would have higher levels of cortisol either due to: first, the stress of being mounted and exposure to aggression or, second, due to underlying pain associated with hoof and leg injuries resulting from these behaviors. We also expected that these effects could be modulated by the dietary treatments as any benefits to limb health through mineral supplementation could have reduced the impact of rearing with males. However, although the behavior observations confirmed that gilts were exposed to higher levels of mounting and aggression when reared with male pigs than when in female-only groups, we did not observe any corresponding increase in cortisol, although there was a tendency for an interaction between group composition and inspection day. This was due to an increase in cortisol in gilts from the mixed-sex groups at breeding age, relative to gilts from the single-sex groups. Even though this was after the males had been removed from the groups for slaughter, there may have yet been on-going physical effects on the gilts. Indeed, once the gilts were slaughtered at day 115 (196 d of age), we found more damage to the cartilage on the surface of the humeral condyle joint and trochlear notch, as well as a higher incidence of Osteochondrosis (OCD) in gilts reared in the mixed-sex groups (Hartnett et al., 2019). It is known that pain is associated with OCD (Frantz, 2006) and, thus, it can be assumed that, with overlying biomechanical stress or trauma, this type of damage to the joints could progress to pain and lameness. OCD lesions do not always produce clinical signs of pain and discomfort as seen in horses even with severe pathological damage (Jeffcott, 1996). This may be similar to our gilts as we saw no difference in locomotory ability in gilts from which we identified OCD lesions and cartilage damage postmortem. In line with this, in a recent study by Uilenreef et al. (2019), there was a low correlation between joint damage and the degree of lameness scored and pigs with induced osteoarthritis were even considered "sound" during visual assessment by trained observers. Damage of this nature could have contributed to the slight rise in cortisol in gilts reared with males as they aged.

Gilts in the mixed-sex groups had higher BL scores than gilts reared in female-only groups. This is to be expected as lesions are associated with higher levels of aggression, sexual behavior, and activity (Turner et al., 2006; Teixeira and Boyle, 2014). There was an interaction between group composition and inspection day on BL scores. Both treatments had similar levels at the first inspection day and, then, lesion scores in gilts reared in mixed-sex groups remaining higher than gilts reared in female-only groups for the rest of the experiment. This is likely due to on-going skin damage due not only to aggression, but due to mounting and play.

Hoof scores were higher in mixed-sex groups, which is likely due to the higher levels of activity (mounting, aggression, and play) in pigs in these groups on the concrete slatted flooring that is known to be injurious to the hooves (Gjein and Larssen, 1995; Pluym et al., 2011; Quinn et al., 2015). Hoof scores also increased over time, which is likely due to general wear and tear with age, as the hoofs must support more weight as pigs increase in size. Hoof lesions are associated with pain and irritation as the lesions on the surface of the hoof can affect sensitive tissues and secondary infections can occur as a result (Zoric et al., 2004). Nevertheless, none of the hoof disorders we observed were severe, and this could be why we did not observe differences in locomotion score.

Nair and Anil (2011) found that the addition of trace minerals Zn, Cu, and Mn to the diet of the breeding sow herd resulted in the reduction of HE and white line lesions. Similar to this, we found that gilts reared without the addition of minerals had higher HE scores than supplemented gilts.

In this study, there tended to be an interactive effect of group composition and experimental day on live weight, with gilts in the single-sex groups tending to gain more weight than gilts reared in the mixed-sex groups by the end of the experiment. We previously showed that gilts coming from the same mixed-sex groups had more mechanical damage to the cartilage on the elbow joint than gilts reared in the single-sex groups (Hartnett et al., 2019), so it is possible that this type of injury could have contributed to a lower growth rate. However, Fabà et al. (2018) found that the incidence of lameness increased as body weight increased, and lame gilts had lower growth rates.

Surprisingly, there was no effect of any treatment on locomotion score. This may be due to hoof and bone disorders not having yet developed to a stage where impaired locomotion is evident, although several previous studies did find improvements in locomotory ability in gilts which were supplemented with similar levels of Cu, Zn, and Mn as employed in this trial (Quinn, 2014; Quinn et al., 2015; Fabà et al., 2018). Quinn et al. (2015) included the mineral supplement in a diet formulated for fat rather than lean meat deposition (i.e., a developer diet), which had a higher energy to lysine ratio than a standard finisher diet. The diet was restricted fed, with the aim of slowing growth rates, which necessitated housing the gilts individually; individual housing means that gilts were likely less active than animals in our study. This could explain why the previous authors found an effect of diet.

Similar to the current study, (Quinn, 2014) found that gilts reared on a "developer" diet (formulated for slow growth and supplemented with Cu, Zn, and Mn) had the lowest risk of abnormal locomotion compared with gilts fed a finisher or gestating sow diet during rearing. Fabà et al. (2018) carried out similar work, rearing gilts on one of four diets: a standard finisher diet, a finisher diet supplemented with either minerals (Cu. Zn, and Mn), methionine, or both minerals and methionine. These authors found that there was a higher percentage of lame gilts when fed the control diet compared to all other treatments. These results are, thus, quite different to ours, where no beneficial effect of mineral supplementation on locomotion was observed. However, this could be because, in general, locomotion score was good across both dietary treatments; this was the case even though half of all gilts were reared in mixedsex groups and were, thus, exposed to the additional activity of male pigs, which was not the case in the studies by Faba and Quinn. However, Quinn used terminal line pigs, which may have been more susceptible to lameness than maternal lines. Fabà et al. (2018) used a different scoring system for locomotory ability and reported only lameness prevalence and, thus, no data were provided regarding the distribution of locomotory ability across the scoring scale.

Clearly, our findings indicate that supplementation of a finisher diet with minerals associated with the promotion of hoof health is not enough to influence locomotory ability. This was the case even though aspects of hoof and limb health were improved by the treatments as gilts had a higher areal bone mineral density and reduced number of OCD lesions when fed the supplemented diet (Hartnett et al., 2019). In fact, locomotory ability was generally very good across all treatments and no lameness was detected in any of the trial animals.

CONCLUSION

Gilts reared with entire male pigs were exposed to higher levels of activity and, importantly, more aggression and sexual mounting compared to gilts reared in female-only pens. As a consequence, they had higher BL scores and more hoof damage. Supplementation with the minerals Cu, Zn, and Mn had only a minor albeit positive effect on HE. Rearing gilts in female-only pens reduces their exposure to behaviors that can injure the hooves and, therefore, increase the likelihood of culling for lameness in later life.

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