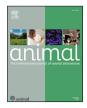
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Early human contact and housing for pigs – part 3: ability to cope with the environment



M.E. Lucas^{a,*}, L.M. Hemsworth^a, K.L. Butler^a, R.S. Morrison^b, A.J. Tilbrook^{c,d}, J.N. Marchant^{e,f}, J.-L. Rault^g, R.Y. Galea^a, P.H. Hemsworth^a

^a The Animal Welfare Science Centre, Faculty of Science, The University of Melbourne, Parkville, Victoria 3010, Australia

^b Rivalea Australia Pty Ltd, Corowa, Victoria 2464, Australia

^c Centre for Animal Science, Queensland Alliance for Agriculture and Food Innovation, The University of Queensland, St Lucia 4072, Australia

^d School of Veterinary Science, The University of Queensland, Gatton Campus, Gatton, Queensland 4343, Australia

^e Organic Plus Trust, Alexandria, VA 22302, USA

^fA World of Good Initiative Inc., Dover, DE 19901, USA

^g Institute of Animal Welfare Science, University of Veterinary Medicine, Vienna A-1210, Austria

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ABSTRACT

Early experiences can have long-term impacts on stress adaptability. This paper is the last of three in a series on early experiences and stress in pigs, and reports on the effects of early human contact and housing on the ability of pigs to cope with their general environment. Using a 2×2 factorial design, 48 litters of pigs were reared in either a farrowing crate (FC) or a loose farrowing pen (LP; PigSAFE pen) which was larger, more physically complex and allowed the sow to move freely. Piglets were provided with either routine contact from stockpeople (\mathbf{C}), or routine contact plus regular opportunities for positive human contact (+HC) involving 5 min of scratching, patting and stroking imposed to the litter 5 days/week from 0 to 4 weeks of age. At 4 weeks of age (preweaning), C piglets that were reared in FC had considerably lower concentrations of serum brain-derived neurotrophic factor (BDNF) than piglets from the other treatment combinations. Compared to C pigs, +HC pigs had fewer injuries at 4 weeks of age. There were no clear effects of human contact on BDNF concentrations or injuries after weaning, or on basal cortisol or immunoglobulin-A concentrations, behavioural time budgets, tear staining, growth, or piglet survival. Compared to FC piglets, LP piglets showed more play behaviour and interactions with the dam and less repetitive nosing towards pen mates during lactation. There was no evidence that early housing affected pigs' behavioural time budgets or physiology after weaning. Tear staining severity was greater in LP piglets at 4 weeks of age, but this may have been associated with the higher growth rates of LP piglets preweaning. There was no effect of lactation housing on growth after weaning. Preweaning piglet mortality was higher in the loose system. The findings on BDNF concentrations, injuries and play behaviour suggest improved welfare during the treatment period in +HC and LP piglets compared to C and FC piglets, respectively. These results together with those from the other papers in this series indicate that positive human interaction early in life promotes stress adaptability in pigs. Furthermore, while the farrowing crate environment deprives piglets of opportunities for play behaviour and sow interaction, there was no evidence that rearing in crates negatively affected pig welfare or stress resilience after weaning. Whether these findings are specific to the two housing systems studied here, or can be generalised to other housing designs, warrants further research.

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Implications

This research provides evidence that positive human interaction can improve the welfare of piglets, based on fewer injuries and higher brain-derived neurotrophic factor concentrations. However, positive human interaction early in life appears to have a stronger and more sustained effect on the responses of pigs to acute stressors than on their ability to cope with the general environment. This research also showed that pigs reared in farrowing crates may be deprived of some opportunities for positive affective experiences early in life, such as play and sow interaction, but we found

E-mail address: megan.lucas@unimelb.edu.au (M.E. Lucas).

* Corresponding author.

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no evidence that rearing in crates impaired pigs' abilities to cope with their environment after weaning.

Introduction

Early life experiences can have both immediate and longer-term effects on farm animal welfare and productivity. Pigs reared in an enriched environment, based on features such as access to substrates and greater opportunity for interaction with the sow and conspecifics during the lactation period, have been reported to show less aggression (De Jonge et al., 1996; Munsterhjelm et al., 2009) and manipulative behaviour directed to pen-mates (Simonsen, 1995) later in life. In contrast, rearing in a more barren environment, for example with less space and no substrates or toys, has been associated with reduced growth (Luo et al., 2020) and long-term effects such as a blunted cortisol rhythm (Munsterhjelm et al., 2010). Given that early life experiences play an important role in shaping the development of stress resilience in rodents and non-human primates (Lyons et al., 2010; Parker and Maestripieri, 2011), the effects on pig behaviour, physiology and productivity in the studies mentioned may represent differences in the ability of pigs to cope with their environment (stress resilience), mediated by differing early experiences.

There are difficulties in measuring stress in animals, and by association, stress resilience, and currently the most useful approach is to interpret a suite of different measurements together. This includes behaviour measurements in-situ, physiological measurements such as basal glucocorticoid concentrations and immune activity, and the cost of coping with stress on animal productivity. Questions around the usefulness of some measurements of stress have been raised (e.g., glucocorticoids: Ralph & Tilbrook, 2016) which has led to the inclusion of more novel indicators in assessments of stress resilience.

Brain-derived neurotrophic factor (BDNF) is a neurotrophin that supports neuronal development and survival, and has been shown to play an important role in learning, memory and stress resilience in rodents and humans (Huang and Reichardt, 2001; Cunha et al., 2010; Mosaferi et al., 2015). Serum BDNF concentrations increased in pigs provided foraging blocks as environmental enrichment (Rault et al., 2018), and lower concentrations of hippocampal BDNF have been reported in pigs exposed to high-stress transport conditions compared to low-stress conditions (Arroyo et al., 2019). Another indicator receiving increasing attention for its use in assessing stress in pigs is tear staining, which refers to the red/ brown accumulation under the inner corner of the eye. Increased levels of tear staining in pigs have been associated with social isolation, lack of environment enrichment, increased tail damage and increased latency to approach a novel object (DeBoer et al., 2015; Telkänranta et al., 2016; Larsen et al., 2019). The mechanisms and function of stress-induced tear staining are not well understood, although it may be part of the immune response to stress (Payne, 1994).

This paper is the final in a series of papers on early experiences and stress resilience in pigs. In Parts 1 and 2 of this series, we showed that human contact and housing experiences during the lactation period affected the fearfulness of pigs and their behavioural and physiological responses towards routine husbandry stressors, during the lactation period but also later in life (Lucas et al., 2024a and 2024b). Compared to rearing with routine human contact and rearing in loose farrowing and lactation pens respectively, rearing pigs with opportunities for positive human interaction and in farrowing crates improved stress resilience on the basis of lower biological responses to stressors such as humans, novelty, isolation, and routine husbandry practices including weaning. Based on improved adaptability to these acute stressors, pigs reared with opportunities for positive human interaction and in farrowing crates may also cope better with their general environment. In the present paper, we report on the effects of these early human and housing experiences on the ability of pigs to cope with their general environment, through measurements of basal physiology, behavioural time budgets, injuries, tear staining, growth and piglet survival.

Material and methods

Experimental design

The experimental design (allocation of treatments to experimental material) of this multiphase experiment had more elements, and thus complexity, as the animals in the experiment grew from piglets to finisher pigs. The methodology of the first phase of the experiment was very similar, but not identical, to a separate previous study (Hayes et al., 2021). The present research was conducted at the same research and innovation unit of a large commercial piggery in Corowa, NSW, Australia, as Hayes et al. (2021).

Preweaning phase: experimental set-up

Preweaning, the experiment consisted of 24 litters from a farrowing crate housing system (FC), where the sow was confined throughout the farrowing and lactation period, and 24 litters from a larger and more structurally complex loose pen (LP), where the sow was free to move throughout the farrowing and lactation period. All litters were born from second parity Landrace \times Large White sows that previously farrowed in a similar housing system, either farrowing crates or loose housing, at their first parity. Twelve FC litters and 12 LP litters were assigned to a routine human contact (C) treatment, and the remaining 12 FC litters and 12 LP litters were assigned to a positive human contact (+HC) treatment. This resulted in a 2 housing system \times 2 human contact treatment factorial design during the lactation period. There was a 7-day farrowing spread across all treatments. Some cross-fostering occurred in the first 24 h of life, within the same housing system and before handling treatments began, when it was necessary to match litter size with the sow's ability to nurse. The housing systems and human contact treatments are described in subsequent sections, with further detail reported in Hayes et al. (2021). Additionally, further detail related to experimental design, housing and management is reported in Part 1 of this series of papers (Lucas et al., 2024a). This additional detail includes information and a visual representation of the position of handling treatments in each housing system.

Preweaning phase: housing treatment

Detailed diagrams of the housing treatments are available in Part 1 of this series (Lucas et al., 2024a). The farrowing crate and loose pen housing treatments had similar overhead lighting and ambient temperatures, and no bedding or enrichment was provided. Each farrowing crate contained a 2.3 \times 1.7 m area for the piglets with slatted steel flooring, and a 1.1 \times 0.41 m solid creep mat heated by an overhead lamp. The surrounding walls of each farrowing crate allowed sows and piglets to have visual contact with people in the aisles. Each loose pen (PigSAFE design; Baxter et al., 2015) contained a 3.6 \times 2.4 m area for piglets and sows. The pens had combination of solid and slatted plastic flooring and contained a covered piglet-only triangular creep area heated by a lamp. Although rarely used, there was a stalled area within each pen where the sow could be confined briefly to allow safe entry for stockpeople. The walls in the central and back areas of the pen contained sloped sides to reduce the risk of overlay by

the sow. There were windows between pens which allowed limited interaction between adjacent pigs, and piglets' visual contact with people in the room was also minimal. Due to recent design modifications, at the back of each pen, there was a 2.4×0.4 m area of space which was not accessible to sows or piglets.

Preweaning phase: human contact treatment

The routine contact treatment involved human contact with stockpeople through the imposition of routine husbandry and management that was typical of a commercial environment. This included visual contact with stockpeople during health and welfare checks and sow feeding twice per day, and when piglet creep food was provided once per day after 14 days of age. In the positive human contact treatment, piglets received regular opportunities to interact with an experimenter in addition to routine contact with stockpeople. Five days per week from 1 day of age until weaning, the +HC treatment involved an experimenter gently patting, stroking and scratching piglets. Two experimenters, one male and one female, were responsible for delivering the treatment, but only one of these experimenters imposed it each day. The treatment was delivered to any piglets that approached the experimenter or were sleeping in the creep area, and the experimenter attempted to interact with as many different piglets in the litter as possible. The experimenter remained silent during imposition of the treatment. The +HC treatment was delivered to the litter for a duration of 5 min in the morning, between 0700 and 1000 h. In farrowing crates, the experimenter crouched behind the sow's crate and next to the creep area to deliver the +HC treatment from inside the pen, and in loose pens, the experimenter crouched outside the pen and interacted with piglets over the pen wall by removing the creep roof.

Weaner-finisher phase

Weaning occurred at 4 weeks of age (mean age = 27 days; SD = 1.5; no difference between treatments). Before weaning, 2 litters of the same lactation housing system and human contact treatment were selected to be paired after weaning. Litters were selected to be paired on the basis of there being enough pigs from each sex to make up one pen of eight males and one pen of eight females after weaning.

Thus, in the weaner facility, there were 24 pairs of pens each made up of a pen of eight male pigs and a pen of eight female pigs, with all 16 pigs in a pair of pens being from 2 litters of the same farrowing and lactation housing system and human contact treatment. At 10 weeks of age, pigs remained in the same groups but were moved to the grower/finisher facility where they stayed until the conclusion of the experiment.

The weaner and grower/finisher pens were spatially arranged in a six–block split–plot design with housing system associated with main plots, human contact treatment associated with subplots and each subplot being two adjacent pens (one pen containing eight males, one pen containing eight females). The two pens in a subplot contained only pigs from 2 litters that were being paired. The pens in the weaner facility were 3.0×1.5 m, and the pens in the grower/finisher facility were 3.7×2.6 m. All pens contained ³/₄ slatted steel flooring and ¹/₄ solid concrete flooring.

Measurements

Measurements reported in this paper include piglet survival to weaning, and behavioural time budgets, basal serum cortisol, BDNF and IgA concentrations, tear staining severity, injuries and growth, during both the preweaning and weaner-finisher phases of the experiment (Fig. 1).

Behavioural time budgets at 1, 2, 3, 8 and 12 weeks of age

Behavioural time budgets of all pigs were recorded at 1, 2 and 3 weeks of age during the preweaning phase of the experiment and at 8 and 12 weeks of age during the weaner-finisher phase. Observations were conducted from video footage (Uniview turret IP cameras) obtained on 1 day from approximately 1000 – 1700 h in weeks 1, 2 and 3, from 1200 – 1600 h in week 8 and from 0800 – 1200 h in week 12. These times were chosen as they were periods of the day outside of routine feeding and management by stockpeople. Instantaneous scan sampling at 120 s intervals was used to record the number of pigs performing behaviours described in the ethogram in Table 1.

One observer conducted observations for weeks 1, 2 and 3, while another observer conducted observations for weeks 8 and 12 (see subsequent section "Observer Reliability" for further detail). The video cameras covered between 90 to 100% of the pen and behaviours were only recorded from pigs that had at least half of the front of the body visible in the camera's field of view. Behaviours were expressed as the percentage of pigs in the field of view displaying the behaviour at each sampling point. These values were averaged over all sampling points, to obtain a pen value for an observation period.

Physiology at 4, 8, 12 and 17 weeks of age

Blood samples were collected from four pigs from each pen for subsequent analysis of serum cortisol, immunoglobulin-A (**IgA**) and BDNF at 4 weeks of age during the preweaning phase of the experiment, and at 8, 12 and 17 weeks of age during the weanerfinisher phase. The same four pigs were sampled at each timepoint, and these pigs were initially selected for sampling using a number generator (see subsequent section "Blood Sampling Procedure and Assay Characteristics" for further detail).

Tear staining assessments at 1, 4, 7, 12 and 17 weeks of age

Tear staining assessments were conducted on photographs of pigs' eyes obtained at 1 and 4 weeks of age during the preweaning phase of the experiment, and at 7, 12 and 17 weeks of age during the weaner-finisher phase. Left-eye staining may be a more sensitive indicator than right-eye staining (Marchant-Forde and Marchant-Forde, 2014), and thus, only photographs of the left eyes were examined. In weeks 1 and 4 respectively, photographs of all pigs were obtained at 4 days of age after piglet processing and at 22 days of age after weighing. In weeks 7, 12 and 17, photographs of the four pigs from each pen that were blood sampled were obtained after sampling.

The severity of tear staining under the eye was scored by one experienced observer using an adapted version of the DeBoer-Marchant-Forde scale (DeBoer et al., 2015): 0 – no signs of staining or area stained is < 1% of total eye area; 1 – staining is barely detectable and area stained does not extend below the eyelid, area stained is approximately 1–10% of total eye area; 2 – staining is obvious and area stained is approximately 10-50% of total eye area; 3 - staining is obvious and area stained is approximately 50-100% of total eye area; 4 - staining is severe, area stained does not extend below the mouth line, area stained is approximately 100-250% of total eye area, and; 5 - staining is severe, area stained extends below the mouth line, area stained is > 250% of total eye area. In addition to the descriptive score, the total area and perimeter of tear staining were calculated using the image processing software ImageI (Schneider et al., 2012) by using the pigs' ear tag as a known reference distance.

Injuries at 4, 5 and 17 weeks of age

Injuries were assessed in four pigs from each pen at 4 weeks of age during the preweaning phase of the experiment, and at 5 and 17 weeks of age during the weaner-finisher phase. The selected

	Pre-	wean	ing pl	hase			Weaner-finisher phase										
	Tre	atme	nt pei	riod	Weaner period					Grower/finisher period							
Week	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
	TB TS W	ТВ	ТВ	TS BS I W	I		TS	TB BS				TB TS BS					TS BS I W
	TS - 7		tainir	ets of ng ass ples				- Wei <u>c</u> Injury		es							

Fig. 1. Timeline of measurements collected in the experiment examining early human contact and housing for pigs.

pigs were the same four pigs that were selected for blood sampling and tear staining assessments. In weeks 4 and 5, respectively, injury assessments took place 2 days prior and 1 week postweaning. Each pig received two injury scores; one for scratches and abrasions on the head and one for scratches and abrasions on the rest of the body. Injuries were scored by one experimenter using the following scale from Widowski and colleagues (2003): 0 – no scratches or skin loss were evident; 1 – one to three small (2 cm) scratches or areas of abraded skin were evident; 2 – one to three larger (>2 cm) scratches or areas of abraded skin were observed; 3 – more than three scratches (usually > 2 cm) or larger areas of superficial skin loss.

Piglet survival until weaning at 4 weeks of age

Piglet survival was determined through records of litter counts at 1 day of age and at weaning at 4 weeks of age.

Growth at 4 days, 22 days and 17 weeks of age

Individual weights of all pigs were obtained at 4 and 22 days of age during the preweaning phase and at 17 weeks of age during weaner-finisher phase of the experiment.

Validation and quality assurance

Observer reliability

Observers conducting video observations, tear staining assessments and injury assessments were blind to treatment, with the exception of measurements collected in the home pens during the lactation period where it was impossible to be blind to housing treatment. The observer repeated behavioural time budgets observations for 2 litters of pigs for observations at 2 weeks of age, and three pens of pigs for observations at 8 weeks of age. Interobserver reliability was assessed for all video observations using intraclass correlation coefficient estimates based on single measure, absolute agreement, two-way mixed effects models analysed in SPSS (IBM Corp, 2020). Intraclass correlation coefficient estimates were all above 0.89, with 95% confidence intervals for the estimates between 0.83 and 1, indicating good to excellent reliability. Blood sampling procedure and assay Characteristics

Two teams of experienced technicians collected blood samples from pigs in neighbouring pens simultaneously. In each team, one technician secured the pig and the other collected the sample via jugular venepuncture within 2 min of the pig being secured. Samples were collected in the aisle in front of the home pen of the sample pig. During the lactation and weaner periods (samples at 4 and 8 weeks), pigs were held inverted on the lap for sample collection and during the grower/finisher period (samples at 12 and 17 weeks), pigs were secured with a snout snare.

Samples were collected into serum tubes (BD Vacutainer, New South Wales, Australia), inverted 5–6 times and left to clot for at least 1 h before being centrifuged for 10 min at 1 300 \times g. After centrifugation, serum was transferred to polypropylene tubes and stored in a -20 °C freezer before being moved to a -80 °C freezer. All samples were assayed in duplicate. Serum concentrations of cortisol were determined using a commercial radioimmunoassay kit (Cortisol Coated Tube RIA Kit, MP Biomedicals Australia Pty Ltd, Seven Hills, New South Wales, Australia). The intra-assay coefficients of variation for samples containing 20.2 and 53.2 ng/ L were 6.9 and 8.0%, and the inter-assay coefficients of variation were 8.7 and 9.2%, respectively. Serum concentrations of IgA were determined using a pig immunoglobulin A ELISA kit (#CSB-E13234p, Cusabio, Houston, Texas, USA). The samples were diluted 1:1 500 or 1:1 000 in serum diluent as recommended by the manufacturer. The intra-assay CV was 6.4%, and the inter-assay CV was 8.5%. Serum concentrations of BDNF were determined using a BDNF ELISA kit (#BEK-2211, Biosensis, Thebarton, South Australia, Australia). The samples were diluted 1:10 (week 4 samples) or 1:5 (week 8, 12 and 17 samples) in serum diluent. The intra-assay CV was 3.3%, and the inter-assay CV was 13.5%.

Statistical analysis

When measurements were assessed on individual pigs, an average value of the pigs for the litter (preweaning phase) or pair of adjacent pens (weaner-finisher phase; one pen of males and one pen of females) was calculated. Each measurement was analysed using ANOVA with one of the structures presented in Table 2. The split–plot ANOVA in the weaner and grower/finisher facilities was used to account for the spatial distribution of the pens in the

Table 1

Ethogram of pig behaviours.

Behaviour	Description
Play	Pig performs one of more of the following behaviours using bouncy, jerky movements (descriptions adapted from Martin and colleagues (2015)):
	Scampering – Sequence of at least two forward hops in rapid succession
	Gamboling – Running forward energetically
	Pivoting – Turning on the spot on a horizontal plane
	Tossing head – Circular, vertical or horizontal movement of the head
	Flopping – Rapid drop of the body from the upright position to recumbency
	Hopping – Two or all four feet off the floor in an energetic upwards movement
	Rolling – Lying on back while swaying entire body left to right
Aggression	Pig performs one or more of the behaviours below using fast, rigid movements, resulting in avoidance or retaliation by the receiver.
	During the lactation period, only aggressive behaviour outside of suckling bouts was recorded.
	Knocking – Vigorously thrusting the head against another pig
	Biting – Rapid opening and closing of the mouth on the body of another pig
Nesterness	Pushing – Using the head or shoulders to press against the body of another pig
Nosing pen mate	Repetitive movement of the snout up and down on the body of another pig
Investigating pen mate	Gentle tactile contact using the snout, directed to any part of another pig's body
Tail biting	Mouthing or chewing the tail of another pig
Ear biting	Mouthing or chewing the ear of another pig
Climbing on sow	Piglet uses feet to elevate itself onto the body or head of the sow
Investigating sow Interacting with pen	Gentle contact with the sow using the snout, excluding nosing around the mammary area Sniffing, nosing or chewing physical components of the pen including the floor
Walking	Slow locomotion around the pen, not engaged in another activity
Standing	Standing stationary on all four legs, not engaged in another activity
Sitting/lying	Sitting or lying, not engaged in another activity. During the lactation period, piglets were recorded as either lying/sitting with the
Sitting/lying	sow (lying or sitting next to or on top of the sow, including lying at the mammary area when no suckling is occurring) or lying/
	sitting away from the sow (lying or sitting with no tactile contact between piglet and sow)

Table 2

ANOVA structures for statistical analysis of measurements collected during different phases of the experiment examining early human contact and housing for pigs.

Source of variation	df
Measurements collected during the preweaning phase ¹	
Housing system	1
Human contact treatment	1
Housing system by human contact treatment interaction	1
Residual	44
Measurements collected during the weaner-finisher phase ²	
Block stratum	5
Row within block stratum	
Housing system	1
Residual	5
Pair within row stratum	
Human contact treatment	1
Housing system by human contact treatment interaction	1
Residual	10

¹ Unit of analysis is litter/lactation pen.

² Unit of analysis is pair of pens; one pen containing females; one pen containing males.

sheds. The only exception was the number of piglets weaned and the proportion of mortalities from 1 day of age until weaning, in which both the means and residual variation differed between housing systems. With these measurements, a restricted maximum likelihood (**REML**) analysis that included a separate residual variance for the two housing systems was used. Main effect means and SE were estimated using a model that excluded the interaction of housing and human contact, but utilised the estimates of residual variance obtained from analyses that included the interaction.

Prior to analyses of variance, all behavioural time budget measurements were angularly transformed and all physiological measurements were logarithmically transformed to ensure the distribution of the residuals was not markedly skewed and/or that the amount of residual variation did not increase as the mean increased. Transformations were determined by examining graphs values, were used to calculate *P*-values for the proportion of pigs playing at 1, 2, 3 and 12 weeks of age, climbing on the sow at

of fitted values versus residuals, both before and after potential

Non-parametric permutation tests, based on the usual ANOVA F

1 week of age and nosing pen mates at 12 weeks of age. In all these cases, there were many pens with no pigs partaking in the behaviour, or there was discreteness in the data, and thus, the usual parametric *P* values could not be considered as reliable.

Due to the poor quality of the photographs, there were no tear staining measurements for one litter in week 1 and 2 litters in week 4. Due to blood samples being haemolysed or returning results above or below the normal detectable range of the assay, there were no BDNF concentration measurements for one pen in week 4. Analyses for the number of piglets weaned and the proportion of mortalities during the lactation period included foster piglets.

Analyses were carried out using the ANOVA directive, the REML directive and the APERMTEST procedure of Genstat for Windows 19th edition (VSN International, 2018).

Results

transformation.

A large number of *P*-values (177 in total) are presented for behavioural time budget measurements in Table 3. To partially avoid multiple testing issues, for time budget measurements, it was decided to only consider *P*-values less than 0.01. The pattern of results for BDNF is somewhat different to other measurements, and thus BDNF is being presented separately to other measurements (see below).

With the exception of BDNF, using the criteria of P < 0.01 for behavioural time budget measurements and P < 0.05 for other measurements, there was no evidence of an interaction between housing system and human contact for any measurement. Thus, the main effects of human contact and housing system on all measurements excluding BDNF are reported separately (Tables 3–7).

Table 3

Effects of early housing and human contact on the behavioural time budgets of pigs at 1, 2 and 3 weeks of age (preweaning) and at 8 and 12 weeks of age. All data were angularly transformed prior to analysis; back-transformed means are presented in parentheses.

	Housing Sys	tem		Human Cont	act		P-value		
	FC	LP	SED	С	+HC	SED	Housing System	Human Contact	Housing \times Conta
Behaviour 1 week, lactation period	(% pigs in vie	w)							
Playing	1.6 (0.080)	1.8 (0.098)	0.49	1.6 (0.082)	1.8 (0.096)	0.49	0.73 ¹	0.77 ¹	0.75 ¹
Aggression	2.4 (0.17)	2.9 (0.25)	0.47	2.3 (0.17)	2.9 (0.26)	0.47	0.30	0.23	0.37
Nosing pen mate	2.7 (0.22)	1.5 (0.069)	0.33	2.0 (0.12)	2.2 (0.15)	0.33	0.00094	0.54	0.56
Investigating pen mate	3.5 (0.37)	3.6 (0.39)	0.39	3.2 (0.31)	3.9 (0.46)	0.39	0.85	0.077	0.83
Tail biting	0(0)	0(0)	-	0(0)	0(0)	-	1.0 ¹	1.0 ¹	1.0 ¹
Ear biting	0(0)	0(0)	-	0 (0)	0(0)	_	1.0 ¹	1.0 ¹	1.0 ¹
							0.073 ¹	0.53 1	0.052 1
Climbing on sow	1.4 (0.058)	0.83(0.021)	0.30	1.0 (0.031)	1.2 (0.044)	0.30			
Investigating sow	4.0 (0.48)	4.5 (0.62)	0.42	4.5 (0.61)	4.0 (0.49)	0.42	0.19	0.31	0.65
Interacting with pen	9.0 (2.5)	8.3 (2.1)	0.58	8.6 (2.2)	8.8 (2.3)	0.58	0.20	0.72	0.78
Walking	8.3 (2.1)	9.1 (2.5)	0.50	8.7 (2.3)	8.8 (2.4)	0.50	0.12	0.72	0.67
Standing	9.3 (2.6)	10.0 (3.0)	0.52	9.7 (2.9)	9.5 (2.8)	0.52	0.20	0.70	0.96
Sitting/lying, away from sow	47 (53)	47 (54)	2.0	47 (54)	47 (53)	2.0	0.85	0.81	0.82
Sitting/lying, with sow	28 (23)	27 (21)	2.3	28 (21)	28 (22)	2.3	0.58	0.84	0.69
Sehaviour 2 weeks, lactation perio	d (% pigs in vi	ew)							
Playing	0.7 (0.015)	2.1 (0.13)	0.48	1.3 (0.049)	1.5 (0.069)	0.48	0.0061 ¹	0.64 ¹	0.32 ¹
Aggression	2.4 (0.18)	3.1 (0.42)	0.60	2.8 (0.24)	3.3 (0.34)	0.60	0.034	0.35	0.78
Nosing pen mate	2.5 (0.18)	2.0 (0.12)	0.35	2.1 (0.13)	2.4 (0.18)	0.35	0.20	0.35	0.67
Investigating pen mate	3.7 (0.41)	4.3 (0.55)	0.42	3.9 (0.47)	4.0 (0.49)	0.43	0.19	0.89	0.80
Tail biting	0 (0)	4.3(0.55) 0(0)	-	0 (0)	4.0 (0.49) 0 (0)	-	1.0 ¹	1.0 ¹	1.0 ¹
						_			
Ear biting	0(0)	0(0)	-	0(0)	0(0)		1.0 ¹	1.0 ¹	1.0 ¹
Climbing on sow	2.5 (0.19)	1.2 (0.045)	0.38	1.9 (0.11)	1.8 (0.10)	0.38	0.0017	0.90	0.33
Investigating sow	3.8 (0.423)	5.2 (0.82)	0.51	4.5 (0.62)	4.4 (0.60)	0.51	0.0063	0.86	0.69
Interacting with pen	9.2 (2.6)	9.7 (2.9)	0.69	9.7 (2.9)	9.2 (2.6)	0.69	0.44	0.43	0.12
Walking	6.4 (1.2)	9.0 (2.4)	0.65	7.3 (1.6)	8.0 (2.0)	0.65	0.00023	0.27	0.14
Standing	6.0 (1.1)	8.6 (2.3)	0.70	7.2 (1.6)	7.4 (1.7)	0.70	0.00048	0.76	0.58
Sitting/lying, away from sow	46 (51)	46 (52)	2.3	47 (54)	44 (49)	2.3	0.98	0.23	0.44
Sitting/lying, with sow	32 (28)	28 (22)	2.1	29 (24)	31 (26)	2.1	0.053	0.53	0.33
ehaviour 3 weeks, lactation perio	d (% nigs in vi	ew)							
Playing	1.5 (0.071)	2.4 (0.18)	0.49	1.8 (0.099)	2.2 (0.14)	0.49	0.079 ¹	0.47 ¹	0.54 ¹
Aggression	4.0 (0.49)	4.3 (0.55)	0.61	3.8 (0.43)	4.5 (0.63)	0.61	0.69	0.21	0.57
		, ,					2.4×10^{-8}	0.031	
Nosing pen mate	4.0 (0.48)	1.8 (0.094)	0.32	2.5 (0.19)	3.2 (0.31)	0.32			0.57
Investigating pen mate	5.1 (0.78)	4.6 (0.65)	0.43	4.1 (0.62)	5.2 (0.82)	0.43	0.32	0.11	0.47
Tail biting	0 (0)	0(0)	-	0 (0)	0(0)	-	1.0 ¹	1.0 ¹	1.0 ¹
Ear biting	0 (0)	0(0)	-	0 (0)	0(0)	-	1.0 ¹	1.0 ¹	1.0 ¹
Climbing on sow	2.8 (0.23)	1.6 (0.079)	0.35	1.9 (0.11)	2.5 (0.18)	0.35	0.0022	0.13	0.88
Investigating sow	4.6 (0.65)	5.5 (0.92)	0.42	4.9 (0.73)	5.2 (0.83)	0.42	0.046	0.46	0.71
Interacting with pen	11 (3.4)	11 (3.6)	0.8	10 (3.3)	11 (3.7)	0.8	0.54	0.28	0.89
Walking	8.4 (2.1)	9.0 (2.4)	0.60	8.6 (2.2)	8.8 (2.3)	0.60	0.34	0.75	0.53
Standing	9.6 (2.8)	9.4 (2.7)	0.67	9.6 (2.8)	9.4 (2.7)	0.67	0.81	0.68	0.35
Sitting/lying, away from sow	43 (47)	48 (55)	1.9	46 (52)	45 (49)	1.9	0.025	0.39	0.53
Sitting/lying, away from sow	30 (25)	26 (20)	1.9	40 (32) 27 (21)	29 (23)	1.9	0.025	0.50	0.61
			1.3	21 (21)	23 (23)	1.5	0.005	0.00	0.01
ehaviour 8 weeks, weaner period			0.27	26(0.20)	25 (0.26)	0 5 2	0.071	0.78	0.46
Playing	3.1 (0.29)	3.9 (0.47)	0.37	3.6 (0.39)	3.5 (0.36)	0.53	0.071		0.46
Aggression	11 (3.6)	12 (4.4)	0.9	12 (4.3)	11 (3.8)	1.0	0.30	0.55	0.87
Nosing pen mate	3.3 (0.34)	4.5 (0.62)	0.60	4.3 (0.56)	3.5 (0.38)	0.82	0.11	0.38	0.23
Investigating pen mate	8.3 (2.1)	8.0 (2.0)	0.89	8.1 (2.0)	8.3 (2.1)	0.35	0.75	0.47	0.042
Tail biting	3.6 (0.39)	4.1 (0.52)	0.23	4.1 (0.50)	3.6 (0.40)	0.66	0.061	0.54	0.43
Ear biting	6.7 (1.4)	7.0 (1.5)	0.48	7.2 (1.6)	6.5 (1.3)	0.86	0.63	0.49	0.92
Interacting with pen	27 (20)	30 (25)	1.2	27 (21)	29 (24)	2.3	0.039	0.44	0.81
Walking	11 (3.6)	12 (4.4)	0.4	12 (4.3)	11 (3.8)	0.6	0.11	0.90	0.79
Standing	8.5 (2.2)	9.9 (2.9)	0.4	9.2 (2.6)	9.2 (2.6)	0.62	0.15	0.99	0.73
Sitting/lying	8.5 (2.2) 48 (55)	9.9 (2.9) 45 (49)	1.0	9.2 (2.6) 47 (53)	9.2 (2.6) 46 (51)	2.7	0.034	0.73	0.73
ehaviour 12 weeks, grower/finish					(- •)				
Playing	1.0 (0.028)	0.7 (0.016)	0.38	0.9 (0.026)	0.8 (0.017)	0.61	0.56 ¹	0.78 ¹	0.79 ¹
Aggression	8.0 (1.9)	7.6 (1.8)	0.37	8.0 (1.9)	7.6 (1.8)	0.39	0.41	0.38	0.33
		. ,		. ,	. ,				
Nosing pen mate	1.0 (0.031)	1.4 (0.058)	0.52	0.9 (0.023)	1.5 (0.069)	0.56	0.57 ¹	0.29 1	0.56 1
Investigating pen mate	6.7 (1.4)	7.5 (1.7)	0.47	7.3 (1.6)	6.9 (1.4)	0.61	0.16	0.53	0.45
Tail biting	2.5 (0.19)	1.9 (0.11)	0.39	2.3 (0.16)	2.1 (0.13)	0.45	0.16	0.68	0.56
Ear biting	3.7 (0.42)	4.1 (0.50)	0.44	4.2 (0.53)	3.6 (0.39)	0.45	0.44	0.23	0.60
Interacting with pen	27 (21)	29 (23)	1.0	29 (23)	27 (20)	1.3	0.18	0.20	0.47
Walking	9.3 (2.6)	9.5 (2.7)	0.38	9.3 (2.6)	9.5 (2.7)	0.30	0.68	0.69	0.64
Standing	14 (5.6)	13 (4.9)	1.2	14 (5.5)	15 (6.6)	0.7	0.45	0.42	0.99

Abbreviations: FC = farrowing crate; LP = loose pen; C = routine human contact; +HC = positive human contact. ¹ P-values calculated using permutation tests.

Table 4

Effects of early housing and human contact on basal concentrations of serum cortisol and immunoglobulin A in pigs at 4 weeks of age (preweaning) and at 8, 12 and 17 weeks of age. All measurements were logarithmically transformed prior to analysis; back-transformed means are presented in parentheses.

	Housing Syste	em		Human Conta	ict		<i>P</i> -value			
	FC	LP	SED	С	+HC	SED	Housing System	Human Contact	Housing \times Contact	
Cortisol (ng/ml)										
4 weeks	1.5 (33)	1.5 (30)	0.06	1.5 (32)	1.5 (31)	0.06	0.44	0.77	0.36	
8 weeks	1.6 (41)	1.7 (52)	0.05	1.7 (48)	1.6 (44)	0.03	0.088	0.19	0.81	
12 weeks	1.5 (34)	1.5 (31)	0.03	1.6 (36)	1.5 (29)	0.04	0.31	0.040	0.29	
17 weeks	1.6 (36)	1.5 (34)	0.02	1.6 (39)	1.5 (34)	0.03	0.35	0.19	0.35	
IgA (µg/ml)										
4 weeks	2.9 (720)	3.0 (910)	0.07	2.9 (790)	2.9 (830)	0.07	0.13	0.74	0.76	
8 weeks	3.6 (4 100)	3.6 (4 000)	0.04	3.7 (4 500)	3.6 (3 700)	0.05	0.73	0.17	0.36	
12 weeks	3.2 (1 600)	3.2 (1 600)	0.16	3.1 (1 400)	3.3 (1 800)	0.08	0.95	0.19	0.30	
17 weeks	3.3 (2 000)	3.1 (1 100)	0.07	3.2 (1 600)	3.2 (1 400)	0.09	0.021	0.54	0.32	

Abbreviations: FC = farrowing crate; LP = loose pen; C = routine human contact; +HC = positive human contact; IgA = immunoglobulin A.

Table 5

Effects of early housing and human contact on the severity of tear staining in pigs at 1 and 4 weeks of age (preweaning) and at 7, 12 and 17 weeks of age.

		Housing System		Human	Contact		<i>P</i> -value		
	FC	LP	SED	C +HC		SED	Housing System	Human Contact	$\textbf{Housing} \times \textbf{Contact}$
Tear staining 1 week									
Area (mm ²)	4.6	5.5	0.69	5.4	4.7	0.69	0.20	0.28	0.74
Perimeter (mm)	9.1	11	1.25	11	9.5	1.25	0.11	0.28	0.77
Score	0.9	1.0	0.08	1.0	0.9	0.08	0.060	0.25	0.95
Tear staining 4 weeks									
Area (mm ²)	8.4	13.6	1.40	10.9	11.1	1.40	0.00055	0.90	0.89
Perimeter (mm)	14	21	1.8	18	17	1.8	0.00036	0.71	0.36
Score	1.0	1.4	0.10	1.3	1.2	0.10	0.000078	0.22	0.65
Tear staining 7 weeks									
Area (mm ²)	13	22	4.1	17	17	3.2	0.082	0.99	0.52
Perimeter (mm)	17	20	3.5	19	19	2.2	0.45	0.93	0.62
Score	1.0	1.3	0.13	1.2	1.1	0.13	0.14	0.65	0.32
Tear staining 12 weeks									
Area (mm ²)	98	110	22.0	110	90	18.5	0.70	0.32	0.19
Perimeter (mm)	39	41	5.1	44	36	3.7	0.61	0.082	0.19
Score	1.9	1.8	0.24	2.0	1.7	0.13	0.88	0.057	0.43
Tear staining 17 weeks									
Area (mm ²)	160	210	39.6	210	160	26.2	0.26	0.12	0.097
Perimeter (mm)	58	61	7.9	64	54	5.6	0.68	0.10	0.14
Score	2.3	2.4	0.24	2.5	2.2	0.18	0.52	0.12	0.10

Abbreviations: FC = farrowing crate; LP = loose pen; C = routine human contact; +HC = positive human contact.

Table 6

Effects of early housing and human contact on injury scores of pigs at 4 weeks of age before weaning, at 5 weeks of age 1 week after weaning and mixing, and at 17 weeks of age. Injury scores were recorded using the scale described by Widowski and colleagues (2003).

	Housing System			Humar	n Contact		P-value			
	FC	LP	SED	С	C +HC		Housing System	Human Contact	Housing \times Contact	
Injuries 4 weeks										
Head	1.4	1.5	0.18	1.7	1.3	0.18	0.60	0.030	0.98	
Rest of body	0.8	0.8	0.14	0.9	0.6	0.14	0.97	0.045	0.44	
Injuries 5 weeks										
Head	2.0	1.6	0.17	2.0	1.7	0.17	0.055	0.12	0.42	
Rest of body	1.3	1.5	0.12	1.6	1.3	0.17	0.19	0.058	0.68	
Injuries 17 weeks										
Head	1.6	1.3	0.16	1.6	1.3	0.13	0.25	0.042	0.76	
Rest of body	1.7	1.5	0.09	1.6	1.6	0.20	0.15	0.80	0.73	

Abbreviations: FC = farrowing crate; LP = loose pen; C = routine human contact; +HC = positive human contact.

Effect of human contact treatment on all measurements excluding brain-derived neurotrophic factor

There was no evidence of any effect of human contact treatment on any behavioural time budget (using P < 0.01 cutoff), IgA (using P < 0.05 cutoff), tear staining (using P < 0.05 cutoff), growth (using P < 0.05 cutoff) or survival (using P < 0.05 cutoff) measurement (Tables 3–5 and 7). The effect of human contact on cortisol concentrations at 12 weeks of age was statistically significant at P = 0.04, but there was no statistically significant (P > 0.1) effect found at 4,

M.E. Lucas, L.M. Hemsworth, K.L. Butler et al.

Table 7

Effects of early housing and human contact on piglet survival until weaning at 4 weeks of age, and growth from 4 days to 17 weeks of age.

	Housing System			Human Contact			P-value			
	FC	LP	SED	С	+HC	SED	Housing System	Human Contact	$\text{Housing} \times \text{Contact}$	
No. pigs 1 day of age	13	12	0.8	12	12	0.8	0.042	0.91	0.83	
No. pigs at weaning	11	8.4	0.51	9.9	9.6	0.46	4.6×10^{-6}	0.58	0.57	
Mortality 1 day of age until weaning (%)	14	26	4.6	20	20	3.7	0.019	0.93	0.50	
Weight 4 days (kg/pig)	2.3	2.2	0.13	2.3	2.2	0.13	0.62	0.52	0.63	
Weight 22 days (kg/pig)	5.8	6.4	0.26	6.2	6.0	0.26	0.029	0.56	0.79	
Gain 4 – 22 days (kg/day)	0.19	0.23	0.012	0.21	0.21	0.012	0.0051	0.75	0.55	
Weight 17 weeks (kg/pig)	76	76	1.8	75	77	1.7	0.87	0.37	0.64	
Gain 22 days – 17 weeks (kg/week)	5.3	5.3	0.13	5.3	5.4	0.12	0.81	0.34	0.66	

Abbreviations: FC = farrowing crate; LP = loose pen; C = routine human contact; +HC = positive human contact.

8, or 17 weeks (Table 4). Neither of the cortisol values at 12 weeks of age (C: 36 ng/ml, +HC: 29 ng/ml) were elevated above the corresponding cortisol values in other weeks, and thus, there is a strong possibility that the statistical significance at 12 weeks was a chance effect.

Compared to C pigs, +HC pigs had lower injury scores on both the head (injuries per/pig, C: 1.7 and +HC: 1.3, P = 0.030) and the rest of the body (C: 0.9 and +HC: 0.6, P = 0.045) at 4 weeks of age during the preweaning phase (Table 6). The effect was statistically significant for injuries on the head at 17 weeks (P = 0.042), but this was associated with a considerably lower standard error of difference (SED) than that observed at 4 and 5 weeks. This lower SED, together with no evidence of a human contact effect on body injury score at 17 weeks, indicates a strong possibility that the effect on the head injury score at 17 weeks of age was a chance effect.

Effect of housing system on all measurements excluding brain-derived neurotrophic factor

There was evidence (P < 0.01) that some behaviours were affected by the housing system during the lactation period. The FC piglets investigated the sow less than LP piglets at 2 weeks of age (Piglets in view, FC: 0.4% and LP: 0.8%, P = 0.006, Table 3). In contrast, FC piglets climbed on the sow more than LP piglets at 2 and 3 weeks of age (Week 2, FC: 0.2% and LP: 0.05%, P = 0.002; Week 3, FC: 0.2% and LP: 0.08%, P = 0.002).

During lactation, there were more FC piglets than LP piglets repetitively nosing pen mates (Week 1, FC: 0.2% and LP: 0.07%, P = 0.0009; Week 2, FC: 0.2% and LP: 0.1%, P = 0.2; Week 3, FC: 0.5% and LP: 0.1%, P < 0.0001; Table 3). At 2 weeks of age, there were more LP piglets than FC piglets walking (FC: 1% and LP: 2%, P = 0.0002), and standing (FC: 1% and LP: 2%, P = 0.0005). There were also more LP piglets than FC piglets engaging in play behaviour at 2 weeks of age (FC: 0.02% and LP: 0.1%, P = 0.0006), and perhaps at 3 weeks of age (FC: 0.07% and LP: 0.2%, P = 0.08).

There was no evidence (P > 0.01) for effects of the early housing system on behavioural time budget measurements at 8 and 12 weeks of age after the treatment period (Table 3).

At 4 weeks of age preweaning, there was strong evidence for greater severity of tear staining in LP piglets than FC piglets (Staining area, FC: 8 mm² and LP: 14 mm², P = 0.0006; Perimeter, FC: 14 mm and LP: 21 mm, P = 0.0004; Score, FC: 1.0 and LP: 1.4, P = 0.00008; Table 5). Tear staining at 1 and 7 weeks of age was in accord with the results at 4 weeks, although no effects were significant at the 5% level (Week 1 score, FC: 0.9 and LP: 1.0, P = 0.06; Week 7 score, FC: 1.0 and LP: 1.3, P = 0.14). There was no indication (P > 0.1) of a housing system effect on tear staining at 12 or 17 weeks of age.

Daily liveweight gain from 4 until 22 days of age was higher in LP piglets compared to FC piglets (FC: 0.19 kg/day and LP: 0.23 kg/day, P = 0.005; Table 7). Consequently, whilst piglet weights at 4 days of age were similar in both housing systems, at 22 days, the LP piglets were about 10% heavier. There was no evidence (P > 0.1) of a housing system effect on weight at 17 weeks of age or on weight gain from 22 days to 17 weeks of age. Piglet survival was lower in LP compared to FC, with smaller litter sizes in LP at 1 day of age (FC: 13 and LP: 12, P = 0.04) and at weaning (FC: 11 and LP: 8, P < 0.0001), and a higher mortality rate during this period (FC: 14% and LP: 26%, P = 0.02).

Of the 14 tests for housing system effects on cortisol, IgA and injuries, only one was significant at the 5% level (Serum IgA concentrations at 17 weeks, P = 0.02, Table 4). Given the principle that, on average, one in every 20 tests will be significant by chance when no effect exists, there is every possibility that the apparent IgA effect at 17 weeks is a chance effect.

Effect of human contact treatment and housing system on brainderived neurotrophic factor

At 4 weeks of age, the *P*-values of the three treatment components of the treatment effects (housing system main effect, human contact main effect, interaction of housing system and human con-

Table 8

Effects of early housing and human contact on concentrations of serum brain-derived neurotrophic factor in pigs at 4 weeks of age (preweaning) and at 8, 12 and 17 weeks of age. All data were logarithmically transformed prior to analysis; back-transformed means are presented in parentheses. Note that this table contains means of each of the four treatment combinations, rather than the main effect means.

	C/FC	+HC/FC	C/LP	+HC/LP	SED		<i>P-v</i> alue			
					Same Housing	Other	Housing System	Human Contact	Housing \times Contact	
BDNF (pg/ml)										
4 weeks	2.4 (235)	3.1 (1 260)	3.1 (1 340)	3.3 (1 760)	0.18	0.18	0.014	0.020	0.089	
8 weeks	2.6 (372)	2.8 (660)	2.7 (468)	2.6 (407)	0.33	0.29	0.78	0.68	0.52	
12 weeks	1.9 (71)	2.1 (117)	1.8 (66)	1.8 (66)	0.13	0.14	0.25	0.27	0.27	
17 weeks	1.7 (46)	1.7 (50)	1.6 (38)	1.8 (59)	0.07	0.10	0.98	0.036	0.14	

Abbreviations: FC = farrowing crate; LP = loose pen; C = routine human contact; +HC = positive human contact; BDNF = brain-derived neurotrophic factor.

tact) for BDNF concentrations were all between 0.01 and 0.1 (Table 8). This suggested that we carry out an additional test for comparing the four treatments as four unstructured treatments (that is a four-treatment, one-way ANOVA). This additional test resulted in a *P*-value of 0.0041 (F = 5.11 on 3, 43 df), which is much stronger evidence of a response than obtained from any of the three individual treatment components. Thus, it was decided to report all the BDNF results as four separate treatment combinations, rather than as the main effect means of housing system and human contact.

At 4 weeks of age during the preweaning phase, serum BDNF concentrations from piglets in farrowing crates with routine human contact (C/FC) were at least 80% lower than the concentrations in any other treatment combination, at about 200 pg/ml, while concentrations for the three other treatment combinations were similar at around 1 500 pg/ml (Table 8).

The *P*-value for the human contact effect on serum BDNF concentrations at 17 weeks of age was 0.04 (Table 8). However, at 17 weeks of age, SE of difference were lower than at other times, and in the context of no effects (P > 0.1) on BDNF at 8 or 12 weeks of age, this finding should be interpreted with caution.

Discussion

Overall, there were few longer-term effects of the early experience treatments on the ability of pigs to cope with their general environment, as measured by pig behaviour and physiology at rest during the weaner-finisher period. However, during the lactation period, piglets reared in farrowing crates showed less play behaviour and sow interaction and more pen-mate manipulation compared to piglets reared in pens, and piglets reared with positive human contact had fewer injuries. Notably, BDNF concentrations were much lower in piglets reared in farrowing crates with routine exposure to humans.

Brain-derived neurotrophic factor has been linked with improved stress resilience in various species (rat: Taliaz et al., 2011: mouse: Leschik et al., 2022; chicken: Yan et al., 2020; human: Sen et al., 2008). In the present experiment, serum BDNF concentrations at 4 weeks of age during the treatment period were considerably lower in piglets reared in farrowing crates with routine human contact, compared to piglets from the other treatment combinations. Other research on pigs has reported that BDNF concentrations were higher with the provision of a foraging block as enrichment (Rault et al., 2018), and in the present experiment, the +HC and LP treatments may have been enriching for pigs. In particular, the lack of enrichment from either +HC or LP in the C/ FC treatment may have led to the lower BDNF concentrations in these piglets. Furthermore, research on rodents has shown that BDNF is associated with social enrichment (Branchi et al., 2006), increased maternal care (Liu et al., 2000; Branchi et al., 2006) and improved spatial learning and memory (Mizuno et al., 2000; Mizuno et al., 2003). Physical exercise is also known to increase BDNF (Rasmussen et al., 2009). Therefore, the loose housing treatment may be linked to higher BDNF concentrations due to piglets having greater opportunities for social play, maternal care, and spatial learning, facilitated through more space, physical complexity of the pen, and non-restricted interactions with the sow.

Despite early human contact and housing having a considerable impact on BDNF concentrations during the time when the treatments were imposed, there was little or no evidence of sustained higher BDNF concentrations as a result of these early experiences. There was also no clear indication of effects of the treatments on cortisol or IgA during the weaner-finisher period. Concentrations of BDNF decreased in all groups over time; a finding that was expected due to BDNF's central role in brain plasticity, which is known to be greatest early in life and reduce with age (Kolb and Gibb, 2011), and an age-related decline is also known in pigs (Rault et al., 2018).

In addition to BDNF concentrations being lower in FC piglets (with routine human contact), during the lactation period, there was evidence of less play behaviour and investigation of the sow in FC piglets compared to LP piglets. The FC piglets also showed a higher frequency of nosing behaviour directed towards littermates, which involved repetitive vertical movements of the snout on any part of another pig's body. These findings align with past research showing more frequent sow-piglet interactions in pens compared to farrowing crates (Chidgey et al., 2016; Singh et al., 2017), more play behaviour in PigSAFE pens (the same loose system studied in the present research) than in crates (Martin et al., 2015), and less manipulation of littermates in pens than in crates (Singh et al., 2017). Play behaviour in the PigSAFE system may be enabled by greater space, more comfortable flooring, nonrestricted interactions with the sow and/or greater complexity of the physical environment. Similarly, the higher frequency of sow interaction by LP piglets may have been a consequence of more opportunity based on more sow-initiated contact in the loose system. Although sow interaction through nasal contact was more frequent in loose pens, there were more piglets climbing on the sow in the farrowing crate system particularly at 2 and 3 weeks of age. This may be due to limited space in the FC system leaving piglets with fewer areas to rest and/or an inability of the sow to move awav.

Effects of the early housing system on preweaning behaviours were not always consistent over the different time periods examined (for instance, the effect on manipulation of pen mates was evident at 1 and 3 weeks of age but not 2 weeks), and additionally, many behaviours pre- and postweaning were not affected by the housing treatment at all. This may have been a consequence of the procedure of instantaneous scan sampling resulting in many events of pig behaviour being missed, or in certain instances, it may just reflect rapid changes that occur during the early life behavioural development of the pig.

The higher frequency of play behaviour and sow interaction and reduced manipulation of pen mates in the loose system, coupled with the BDNF findings, collectively suggest improved welfare in LP piglets compared to FC piglets during the treatment period. Interactions with the dam are known to have a significant influence on the development of stress-coping mechanisms in offspring (Liu et al., 1997), and as previously discussed, BDNF has been linked to stress resilience. Furthermore, play behaviour can serve as both a contributor to and an indicator of stress resilience, as it is typically associated with a positive emotional state and has been proposed to enhance stress-coping skills by enabling animals to recover from unexpected events (Spinka et al., 2001). It would therefore be reasonable to anticipate that the LP piglets would demonstrate improved resilience when exposed to challenges in their environment. In fact, the findings of Parts 1 and 2 of this series of papers indicated the opposite (Lucas et al., 2024a and 2024b).

In Parts 1 and 2, we reported that the loose housing treatment increased pigs' fear and stress responses to humans, novelty, social isolation, and husbandry practices (Lucas et al., 2024a and 2024b). Interpreted together, the results of all parts in this series suggest the welfare of piglets kept in loose pens was superior when they were left undisturbed in the home pens, but that they lacked the flexibility to cope with additional stressors in their environment. While the farrowing crate environment clearly deprives piglets of opportunities for some positive affective experiences early in life, including play behaviour and sow interaction, we did not find any evidence that rearing in crates had negative effects on pig welfare or stress resilience after weaning. Actually, Parts 1 and 2 provided evidence that rearing in farrowing crates led to more flexibility in coping with stressors after weaning, as well as preweaning. Whether these results are linked to the specific type of farrowing crate and loose pen used in the present experiment, or can be generalised to other housing designs, warrants further research.

The early handling treatment had no impact on any behaviours measured at rest, but +HC piglets had fewer injuries on the head and the rest of the body at 4 weeks of age during the treatment period. Hayes and colleagues (2021) found a similar effect on injuries 2 days postweaning. Injuries may be obtained during bouts of aggressive or play behaviour, but there was no evidence of the +HC treatment affecting these behaviours. It may be that +HC pigs sustained fewer injuries due to less fear and avoidance of people, and therefore fewer collisions with pen fittings, the sow and/or pen mates.

The findings of the present research on BDNF concentrations and injuries provide further support to the findings reported in Parts 1 and 2 of this series, which showed that early positive human interaction fosters stress resilience in pigs, based on lower fear and stress responses to humans, novelty, social isolation and husbandry practices (Lucas et al., 2024a and 2024b). However, other than the findings on BDNF and injuries, the present research found limited evidence of effects of early positive human contact on the ability of pigs to cope with their general environment. The handling treatment did not appear to impact basal cortisol or IgA concentrations, tear staining or behavioural time budgets, during the lactation period or subsequently. The +HC treatment also had no effect on piglet survival during the lactation period or growth throughout the experiment. Thus, early handling appears to have a stronger and sustained effect on the responses of pigs to acute stressors than on the welfare of pigs when they have not been recently challenged.

There were more piglet mortalities in the loose system from 1 day of age until weaning, likely due to an increased prevalence of crushing although specific causes of mortality were not recorded. Litter sizes were lower in the loose system even at only 1 day after farrowing, which highlights the mortality risk soon after parturition to piglets in loose farrowing and lactation systems. Most likely because of smaller litter sizes in loose pens, weight gain during the lactation period was higher in the loose system. There was no effect of early housing on injury scores at 4, 5 and 17 weeks of age. In contrast, our previous work showed more injuries in LP than FC piglets at 2 weeks of age (Hayes et al., 2021) which suggests the timing of injury assessments is important. Further detail on the specific causes of piglet injuries and mortalities would be valuable.

The severity of tear staining was higher in LP piglets than FC piglets at 4 weeks of age during the lactation period, with some evidence also suggesting greater tear staining at 1 and 7 weeks of age. Tear staining measurements at 4 weeks of age were obtained 1-3 days after piglets had undergone vaccinations and behaviour testing. Tear staining can increase after 1 day of pigs being exposed to a stressor (e.g. new housing: DeBoer et al., 2015), and it is possible that the increased staining in LP pigs at 4 weeks of age reflected a greater response to the stressors of vaccination and behaviour testing that had occurred in the days prior (reported in Lucas et al., 2024a and 2024b). However, there may have been confounding factors such as air quality, humidity and ammonia levels that affected tear staining and differed between the housing systems located in different rooms. Additionally, tear staining has been positively correlated with growth in pigs (Larsen et al., 2019), and as mentioned earlier, weight gain during the lactation period was higher in the loose system. It is therefore unclear if the higher levels of staining early in life in LP pigs were stress-induced or simply a result of differences in growth or environmental conditions.

Overall, the results from the three papers in this series indicate that positive human interaction early in life promotes stress adaptability in pigs. In Parts 1 and 2, we showed that early positive human interaction reduced stress responses to humans, novelty, social isolation, and husbandry practices, and here in Part 3, it reduced piglets' injuries and increased levels of BDNF. However, the effects on BDNF and injuries were not prolonged and there was little evidence for any other benefits of early handling on pig behaviour and physiology at rest. Therefore, when considering different aspects of stress resilience, early positive human interaction was more powerful at improving resilience to acute challenges than improving the ability of pigs to cope with their general environment. Regarding early housing experiences, the farrowing crate environment reduced BDNF concentrations and limited opportunities for play behaviour and sow interaction during the lactation period. However, there was no evidence in the present research that rearing in crates negatively affected pig welfare or general stress-coping ability after weaning. In fact, as reported in Parts 1 and 2, it was piglets from the loose housing system that showed less flexibility in response to some specific stressors before and after weaning. Collectively, the findings of this series suggest the welfare of piglets in loose pens was superior when undisturbed in their home environments, but compared to crate-reared piglets, pen-reared piglets were not as well-equipped to cope with additional challenges in their environment.

Ethics approval

All animal procedures were conducted with prior institutional ethical approval under the requirements of the New South Wales Prevention of Cruelty to Animals Act 1985 in accordance with the National Health and Medical Research Council/Commonwealth Scientific and Industrial Research Organization/Australian Animal Commission Code of Practice for the Care and Use of Animals for Scientific Purposes (Rivalea Animal Ethics Committee #19B018C).

Data and model availability statement

The data were not deposited in an official repository but are available from the corresponding author 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.

Author ORCIDs

Megan E. Lucas: https://orcid.org/0000-0002-5730-7535. Lauren M. Hemsworth: https://orcid.org/0000-0002-7752-8917.

Kym L. Butler: https://orcid.org/0000-0002-1958-3942. Rebecca S. Morrison: https://orcid.org/0000-0002-8412-4945. Alan J. Tilbrook: https://orcid.org/0000-0002-1116-1470. Jeremy N. Marchant: https://orcid.org/0000-0002-5287-2914. Jean-Loup Rault: https://orcid.org/0000-0001-6015-8318. Rutu Y. Galea: https://orcid.org/0000-0002-8740-0589. Paul H. Hemsworth: https://orcid.org/0000-0002-0211-0751.

CRediT authorship contribution statement

M.E. Lucas: Writing – original draft, Visualization, Project administration, Methodology, Investigation, Formal analysis. **L.M. Hemsworth:** Writing – review & editing, Supervision, Project

administration, Methodology, Funding acquisition, Conceptualization. **K.L. Butler:** Writing – review & editing, Methodology, Formal analysis. **R.S. Morrison:** Writing – review & editing, Supervision, Methodology. **A.J. Tilbrook:** Writing – review & editing, Supervision, Methodology, Funding acquisition. **J.N. Marchant:** Writing – review & editing, Methodology, Investigation. **J.-L. Rault:** Writing – review & editing, Methodology. **R.Y. Galea:** Project administration, Investigation. **P.H. Hemsworth:** Writing – review & editing, Supervision, Project administration, Methodology, Funding acquisition, Conceptualization.

Declaration of interest

None.

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