Animal Welfare

www.cambridge.org/awf

Quantifying chronic stress in group- and stall-housed sows during gestation using cortisol in hair

Talia Everding[1], Yolande Seddon[2], Jorge Perez Palencia[3] and Crystal Levesque[3]

[1]Department of Animal Science, University of Nebraska-Lincoln, United States; [2]Department of Large Animal Clinical Science, Western College of Veterinary Medicine, University of Saskatchewan, Saskatoon, SK, Canada and [3]Department of Animal Science, South Dakota State University, United States

Research Article

Cite this article: Everding T, Seddon Y, Perez Palencia J and Levesque C (2026). Quantifying chronic stress in group- and stall-housed sows during gestation using cortisol in hair. *Animal Welfare*, **35**, e33, 1–8
https://doi.org/10.1017/awf.2026.10059

Received: 05 August 2025
Revised: 24 November 2025
Accepted: 03 December 2025

Keywords:
Animal welfare; gestation housing; gilt; hair cortisol; pig; stress

Corresponding author:
Crystal Levesque;
Email: crystal.levesque@sdstate.edu

Author contributions:
Project administration: T.E.; Conceptualisation: Y.S., C.L.; Data curation: J.P.P.

© The Author(s), 2026. Published by Cambridge University Press on behalf of The Universities Federation for Animal Welfare. This is an Open Access article, distributed under the terms of the Creative Commons Attribution-NonCommercial licence (http://creativecommons.org/licenses/by-nc/4.0), which permits non-commercial re-use, distribution, and reproduction in any medium, provided the original article is properly cited. The written permission of Cambridge University Press or the rights holder(s) must be obtained prior to any commercial use.

LinkedIn: https://www.linkedin.com/showcase/animal-welfare-journal/
webpage: https://www.ufaw.org.uk/

Abstract

The objective of this study was to characterise chronic stress in gestating sows (n = 66; parity 0–6) housed within the same facility, assigned to gestation stalls or group pens (25 sows per pen) within two days of breeding until day 111 (± 1.4) of gestation using hair cortisol concentrations (HCC) as a biological marker of longer-term stress. Hair was shaved from the right hip at day 6.0 (± 3.8), day 37, day 74, and day 111 of gestation; day 37 and day 111 samples were analysed for cortisol. Housing system, parity class (0–1 [n = 23], 2–3 [n = 17], and 4–6 [n = 26]), and their interactions were explored. Stall-housed parity 0–1 females had higher HCC than all other females regardless of housing system, suggesting that gilts and parity 1 females in stall housing may experience greater chronic stress during gestation.

Introduction

In the US, sows are typically housed in stalls during gestation. The favourability of stalls includes that individual housing provides a safe environment for both the sows and employees, protecting sows from injury and physical aggression from other sows, and enabling each female to receive an individual feed ration without competition (Salak-Johnson 2017). Additionally, stall housing is economically efficient, allowing more sows to be housed in fewer square feet of space, resulting in improved economies of scale compared to group-housing facilities (Buhr 2010).

However, stall housing has been criticised (Ryan *et al.* 2015) for limiting sows' movement and normal behaviours, thereby reducing sow welfare in comparison to group-housed females. The ability of the animal to perform normal behaviours that they are motivated to perform is important and will impact their affective state, and definitions of welfare now consider this (Englund & Cronin 2023). The ability of an animal to perform behaviours and have choice in their environment is linked to a higher welfare status in many species (Englund & Cronin 2023). A sow's choice regarding where to walk or lie, or whether to interact with other pigs (*Sus scrofa*) is restricted in stall housing, but group housing provides broader options. Pork consumers have demanded higher welfare standards for gestating sows and see group housing as promoting improved sow welfare, resulting in an international trend for banning gestation stalls (European Commission 2008; Canadian Pork Council and National Farm Animal Care Council 2014; Schulz & Tonso 2015). This has also been seen in the US, most recently with the passing of Proposition 12 in California, which requires 2.23 m^2 of floor space per sow in gestation (Proposition 12 – Prevention of Cruelty to Farm Animals Act 2018). However, the research conclusions are conflicting as to whether sows housed in stalls experience poorer welfare than sows in group housing, as measures of physical health and affective state must both be considered. Measures of reproductive performance such as farrowing rate and litter size have been reported to be both negatively and positively influenced or not impacted by stall housing, as reviewed by Koketsu and Iida (2017). Stereotypical behaviours are considered indicators of chronically poor welfare because they develop as symptoms of frustration and boredom and impoverished housing conditions, and they have been observed with greater frequency in stall-housed sows (Chapinal *et al.* 2010). However, other measures of sow welfare like frequency and severity of lameness have been shown to be worse in group-housed sows (Koketsu & Iida 2017). Biological metrics such as cortisol in blood and saliva have been used to assess stress related to housing systems (Barnett *et al.* 1985; Anil *et al.* 2006), however, blood and salivary cortisol are measures of acute stress, as cortisol is secreted as a response of the hypothalamic-pituitary-adrenal axis to a stressor, following the secretion of adrenocorticotropic hormone (Heimbürge *et al.* 2020b). Cortisol is secreted into the bloodstream and incorporated into saliva within minutes after a stressor, such as restraint or handling during a blood draw (Bozovic *et al.* 2013); blood cortisol has been observed in swine to return to pre-stressor levels within an hour of an acute stress event (Guzik *et al.* 2006). Cortisol in blood and saliva is therefore impractical as a measure of chronic

stress, and measures that reflect long-term cortisol secretion may be more accurate in characterising chronic stress and as an indicator of sow welfare. Recently, there has been increasing interest in cortisol deposited in hair as a measure of long-term stress; with one theory being that cortisol circulating in the bloodstream is incorporated into the hair slowly as it grows capturing the longer-term activity of the HPA axis (Heimbürge *et al.* 2020b). Studies in multiple species have examined hair cortisol concentration (HCC) as an indicator of chronic activation of the hypothalamic-pituitary-adrenal axis and have observed elevated HCC in hypercortisolaemic subjects (Russell *et al.* 2012), infants on ventilators compared to healthy infants (Yamada *et al.* 2007), and animals such as dairy cattle (*Bos taurus*; González-de-la-Vara et al. 2011), goats (*Capra hircus*; Endo *et al.* 2018), and lynx (*Lynx canadensis*; Terwissen *et al.* 2013) undergoing simulated stress conditions via injection of exogenous adrenocorticotropic hormone. Elevated hair cortisol has also been observed in swine experiencing chronic stress conditions (Prims *et al.* 2019; Morgan *et al.* 2021). Cortisol in hair may be used to identify chronic stress and hence provide information on sow welfare by acting as a record of elevated cortisol over a prolonged period.

In previous research, hair cortisol concentration in group-housed sows was not influenced by parity, but litter size and HCC were positively correlated (Roelofs *et al.* 2019); it has also been noted that HCC fluctuates during the reproductive cycle of sows housed both individually and in groups (Bacci *et al.* 2014). However, little research to date has directly compared HCC of gestating sows kept in individual housing versus those housed in groups. The purpose of this study was to determine the HCC of sows in stall- and group-gestation housing systems within the same facility and to compare whether they differed. Results are to contribute knowledge on HCC as a metric of chronic stress influenced by housing system during gestation and to assess the influence of housing system on sow welfare.

Materials and methods

Ethical approval

The experimental protocols used in this study were approved by The South Dakota State University Institutional Animal Care and Use Committee (IACUC #17-072A and 18-064A).

Study animals and experimental design

The study was conducted in the sow barn at the South Dakota State University Swine Education and Research Facility from November 2018 until June 2019. A total of 123 multiparous and primiparous females (PIC 1050) across four breeding groups and housed in gestation stalls (two breeding groups) or group pens (two breeding groups) were used in this study. The sow herd was managed in a batch farrowing system where 20–30 sows per breeding group were bred, farrowed, and/or weaned every four weeks. The east half of the gestation facility contained 76 individual gestation stalls and the west half three group pens. The gestation facility was artificially lit between 0600 and 1400h and mechanically ventilated, and there was no wall separating the group pens from the stalls. The group pens (8.53 × 8.20 m; length × width) were slatted concrete, with a solid concrete portion along the west wall (8.20 × 3.18 m; length × width). The solid portion of each group pen had two partitions, dividing the solid portion into three resting areas (3.18 × 2.73 m; length × width). There were two water nipples along the south side of each pen, and a Mannabek electronic sow feeder (PigTek, Milford, IN, USA) on the east side of each pen. The stall-housed sows were housed in fully slatted gestation stalls (1.98 × 0.61 m; length × width) from breeding until day 111 (± 1.4) of gestation. Each stall had an individual feeder and a water nipple. Rations were delivered once per day and water was accessible *ad libitum*. The first stall-housed breeding group was bred in November 2018, and the second in December 2018. The first pen-housed breeding group was bred in February 2019, and the second in March 2019. The group-housed sows were divided among three group pens approximately 48 h after breeding. The group housing was managed as a dynamic system with females being added at approximate 28-day intervals depending upon housing needs, illustrated in Figure 1. Over the course of this work, gilts and small parity 1 sows from both pen-housed breeding groups were housed in one group pen (e.g. when gilts and small parity 1 sows from the first group reached approximately day 28 of gestation, the second breeding group of gilts and small parity 1 sows was added to the same pen). Multiparous sows were housed in the other two group pens. Each group pen housed up to 25 sows, providing at least 2.79 m^2 per female, and as noted above, over the course of this work the group pens were dynamic due to the introduction or removal of non-experimental herd sows as outlined in Figure 1. During the gilt development period, all gilts were housed in groups of eight females per pen until undergoing acclimation to stalls four weeks prior to first breeding. All multiparous females were housed in stalls at least five days prior to breeding. Sows were moved from their assigned housing system to farrowing crates at day 111 (± 1.1) of gestation.

All females were included in one of two nutrition trials being conducted during this experiment. All stall-housed sows were in a trial examining dietary antioxidants fed in gestation and lactation (Hernandez *et al.* 2021). There were four treatments, receiving a standard gestation diet plus a carrier top dress including either peppermint oil, whole yeast cell, gamma-tocopherol, or nothing (control). All group-housed sows were in a trial examining varying dietary lysine:energy ratios in gestation (Bruhn 2020). There were three treatments: constant lysine:energy ratio throughout gestation; higher lysine:energy ratio from day 90–110 of gestation (phase-feed 1) or lower lysine:energy ratio from day 2–89; and higher:energy ratio from day 90–110 of gestation (phase-feed 2). In both nutrition trials, gestation diets were formulated to meet or exceed gestation nutrient requirement estimates according to National Research Council (2012). Daily feed allotment was managed similarly across both housing systems (i.e. daily feed allotment adjusted at 30-day intervals throughout gestation to maintain body condition score of 3); thus, differences in diets between the trials were not expected to impact sow response to housing. Further details regarding the nutritional studies can be found in Hernandez *et al.* (2021) and Bruhn (2020).

Hair sample collection

If not already housed in a stall, animals were moved one-by-one to an individual stall to facilitate collection of hair; animals were replaced into their original group pen immediately following hair collection, which was completed within 5 min for each animal. An electric trimmer (Remington HC5855B, Middleton, USA) was fitted with a flexible plastic shield to better facilitate the direct movement of trimmed hair into the paper envelope, and hair was shaved as close to the skin as possible. The trimmer was wiped following each individual procedure to remove any hair fragments. Approximately 400 cm^2 on the right hip was shaved at 6.0 (± 3.8)

https://doi.org/10.1017/awf.2026.10059 Published online by Cambridge University Press

Month	Group pen #1 (sows)			Group pen #2 (sows)			Group pen #3 (gilts and small parity 1 sows)			
February	Jan breed group in[1]	*Feb breed group in*[2]		Jan breed group in			Jan gilts in	Feb gilts in[2]		
March					*Mar breed group in*[2]				Mar gilts in[2]	
April										
May	Jan breed group out			Jan breed group out			Jan gilts out			
June		*Feb breed group out*	May breed group in[1]			May breed group in		Feb gilts out		May gilts in
July					*Mar breed group out*				March gilts out	

Figure 1. Entry and exit of breeding groups through the pen housing system in gestation.
[1] Non-experimental breed groups entered pen after pregnancy check at approximately day 28 of gestation; [2] Experimental breed groups entered pen at day 2 after breeding.

days post-breeding and hair was discarded. Hair regrowth samples (150 mg) from the hip at day 37, day 74, and day 111 of gestation were collected. All samples were collected and stored in individual paper envelopes and kept in a dark drawer in the laboratory at 20°C until analysis (not more than seven months).

Laboratory analysis

Samples were processed in accordance with a modified version of the MacBeth *et al.* (2010) protocol. Samples were examined, hairs with disturbed cuticles or broken hairs were discarded, split ends were trimmed off and the remaining intact portion of the hair was retained, debris removed, and samples were weighed. Samples ≤ 50 mg were placed into 2-mL cryogenic vials (Corning #430659, NY, USA) and > 50 mg were placed into 5-mL disposable glass culture tubes (Fisher Scientific #14-961-26, MA, USA). The samples were washed three times in methanol (40 µL mg^{-1} sample), blotted dry between rinses, then left to dry overnight. Samples were ground into a fine powder with a Retsch MM310 mixer mill (Retsch, Haan, Germany) at 30 Hz; 25 mg of powdered hair were weighed into 0.6-mL tubes. The analyte was extracted using 0.5 mL methanol per 25 mg of powdered hair, and the samples incubated for 16–24 h on a rotator. Tubes were centrifuged at 20°C for 15 min at 2,150 × *g*, and the supernatant collected and transferred to a 12-mm glass culture tube. To ensure all extracted steroids were recovered, the powdered hair sample was rinsed twice by adding 0.5 mL of fresh methanol, then gently vortexing (40 s), centrifuging, and pooling supernatants. The pooled supernatant was dried at 38°C under a gentle stream of nitrogen gas and reconstituted with 200 µL of extraction buffer from the cortisol assay kit, and samples were frozen at –80°C until analysis with enzyme immunoassay (EA65, Oxford Biomedical, Rochester Hills, MI, USA). The optical density value was read at 450 nm after 30 min on a SpectraMax 190 absorbance plate reader (Molecular Devices, Sunnyvale, CA, USA). A standard curve of optical density value versus cortisol concentration was generated, and hair cortisol concentration was then determined according to the standard curve and expressed in pg cortisol mg^{-1} powdered hair. According to the manufacturer, cross-reactivity of the antibody used for the cortisol kit is as follows: cortisol (100.00%), prednisolone (66.9%), 11-deoxycortisol (58.1%), cortisone (15.9%), prednisone (13.7%), 17-hydroxyprogesterone (5.4%), dexamethasone (4.6%), oestriol 4.5%, oestrone (4.1%), d-aldosterone (3.6%), progesterone (3.5%) 6-β-hydroxycortisol (3.4%), trans dehydroandrosterone (1.9%), testosterone (1.7%), corticosterone (1.4%), and pregnenolone (1.3%).

Sample selection for analysis

All available stall- (n = 64) and group-housed sows (n = 59) were shaved and samples collected 6.0 (± 3.8) days post-breeding. At day 111, 50 stall- and 39 group-housed sows remained, hair was shaved, and samples collected. Overall, 14 stall- and 20 group-housed females were removed from the trial due to failure to become or remain pregnant or removed from group pen due to injury or illness, or insufficient hair growth. During the laboratory selection and analysis, a further 16 samples from stall-housed sows and seven from group-housed females were removed due to inadequate sample size or poor sample quality (badly damaged or soiled). In total, samples from 34 stall-housed sows and 32 group-housed sows were analysed for HCC and included in the statistical analysis. Hair regrowth samples from the hip at day 37, day 74, and day 111 of gestation were collected with only samples from days 37 and 111 analysed for cortisol concentration. These time-points were selected because day 37 captured the period when sows were introduced to groups, and day 111 captured the final stage of gestation. Unless otherwise noted, results are expressed as an average of day 37 and day 111 cortisol concentrations. Samples from day 74 were not analysed in this experiment due to budget constraints. The final analysis contains the following number of samples from stall-housed sows from each dietary treatment: peppermint oil (n = 1); whole yeast cell (n = 11); gamma-tocopherol (n = 9); and control (n = 13). The final analysis contains the following number of samples from group-housed sows from each treatment: control (n = 12); phase-feed 1 (n = 10); and phase-feed 2 (n = 10).

https://doi.org/10.1017/awf.2026.10059 Published online by Cambridge University Press

Statistical analysis

The UNIVARIATE procedure of SAS (Version 9.4, SAS Institute Inc, Cary, NC, USA) was used to review distribution of the data, test for normality, and identify outliers. Data were analysed using the MIXED procedure of SAS with 'sow' as the experimental unit and 'HCC' as the dependent variable. Sows were assigned to one of three parity classes, 0–1 (gilts and sows having completed one successful parity) (n = 23), 2–3 (n = 17), and ≥ 4 (n = 26) and to one of three litter size classes based on total born, ≤ 14 (n = 22), 15–17 (n = 20), and ≥ 18 (n = 24). There were potential confounding effects of dietary treatment, breed group, parity class, and litter size; therefore, the main effect of each was tested. Main effect of dietary treatment and breed group were analysed for each housing system separately. Where the effect of a confounding variable was significant, the interaction with housing system was tested. Non-significant interactions were removed from the model. The final model for housing system and day of gestation included the main effects of housing system, day of gestation, and their interactions were tested with parity class as a random effect. 'Pen' was not included in the model either as confounding or random variable because of the difference in absolute number of pens between housing system (i.e. each sow equalled a 'pen' for stall-housed females) and because pen was synonymous with group as outlined in Figure 1. Data are presented as least significant means and standard error. Tukey's adjusted means test was used to detect differences where statistical significance and tendencies were set at $P \leq 0.05$ and $P < 0.10$, respectively, for all statistical tests.

Results

Dietary treatment did not influence HCC in that the HCC of control-fed females did not differ from the HCC of experimentally fed females in either the stall-housed or group-housed females ($P > 0.1$). Summary statistics of the sow groups used in this study are shown in Table 1.

There was no difference in HCC between breed groups in stall-housed females (66.5 pg mg^{-1} for the first breed group and 46.4 pg mg^{-1} for the second breed group; SEM = 13.3) and a tendency for group-housed females in the second breed group to have higher HCC (18.4 pg mg^{-1} for the first breed group and 23.7 pg mg^{-1} for the second breed group; SEM = 2.9; $P = 0.0752$).

There was a significant interaction between housing system and parity class on HCC; parity 0–1 females housed in stalls had higher HCC than older sows housed in stalls (parity ≥ 2) and there was no difference among parity class for females housed in group pens (Figure 2).

There was a tendency for day of gestation to influence HCC, with an increase in HCC at day 111 of gestation (Table 2). The HCC of stall-housed females increased 39% from day 37 to day 111 of gestation (41.3 pg mg^{-1} to 57.5 pg mg^{-1}; SEM = 8.8) and 26% in group-housed sows (17.5 pg mg^{-1} to 22.1 pg mg^{-1}; SEM = 9.0), but there was no interaction between day of gestation and housing system.

Litter size was affected by parity class, where parity ≥ 4 females had more total piglets born than parity 0–1, and parity 2–3 did not differ significantly from parity ≥ 4 (Table 2). However, litter size did not affect overall HCC ($P = 0.212$).

Discussion

Elevated HCC has been observed in animals undergoing simulated chronic stress conditions via injection of exogenous adrenocorticotropic hormone (González-de-la-Vara *et al.* 2011; Terwissen *et al.* 2013; Endo *et al.* 2018). Further, elevated HCC has been detected in multiple species (humans [Iob & Steptoe 2019], non-human primates [Davenport *et al.* 2008; Fairbanks *et al.* 2011; Dettmer *et al.* 2012; Carlitz *et al.* 2014], wolves [*Canis lupus*; Bryan *et al.* 2015], grizzly bears [*Ursus arctos horribilis*; Bryan *et al.* 2013], sheep [*Ovis aries*; Nejad *et al.* 2014], and pigs [Prims *et al.* 2019; Heimbürge *et al.* 2020b; Morgan *et al.* 2021]) experiencing chronic stress conditions, either induced experimentally or naturally occurring. This suggests that in the present study, the parity 0–1 stall-housed females may have experienced greater chronic stress than all other group-housed and stall-housed females, suggesting stall housing reduced welfare of gilts and parity 1 sows, whereas group-housing did not negatively affect the welfare of pregnant females regardless of parity. However, because older sows in stall housing had indistinguishable HCC compared to older group-housed sows, acclimation to stall housing may occur over the course of multiple parities. Stall-housed females are observed as exhibiting more frequent stress behaviours, such as sham chewing and oronasofacial stereotypies, compared to females in group pens (Chapinal *et al.* 2010). Sows in group pens also can experience stress from exposure to fighting for social hierarchy, aggression, and injury from other sows (Salak-Johnson 2017). Other research has shown that frequent, repeated mixing of unfamiliar piglets over the course of three weeks does increase HCC (Prims *et al.* 2019). In this trial, behavioural measures and injury scores were not recorded, so it is not possible to evaluate the acute stress experienced by the group-housed sows during mixing. Group-housed females were kept in dynamic groups, and it is probable therefore they experienced acute stress during mixing and for assembling order on the electronic sow feeder. Nevertheless, this period of acute stress at mixing was not sufficiently stressful or prolonged to raise the HCC of group-housed females above the HCC of their stall-housed counterparts. The HCC of group-housed sows was lower than stall-housed females younger than parity 2 despite the presumption acute stress

Table 1. Summary statistics regarding the specific groups used in this study into hair cortisol in gestating sows.

	Bruhn (2020)				Hernandez *et al.* (2021)				
Sows	Control	Phase fed 1	Phase fed 2	SD	Control	Yeast cell wall	Mint oil	ɣ-tocopherol	SD
Sow weight gain, breeding to day 110 of gestation (kg)	54.6	57.5	56.1	32.8	37.5	36.6	37.9	34.8	20.3
Live born (n)	14.8	14.0	14.3	2.8	15.0	13.8	14.8	14.4	3.3
Stillborns (n)	1.8	1.8	1.5	1.8	1.0	1.0	1.6	1.1	1.9
Birth weight (kg)	1.33	1.45	1.32	0.09	1.34	1.34	1.35	1.36	0.11

SD: Standard deviation.

https://doi.org/10.1017/awf.2026.10059 Published online by Cambridge University Press

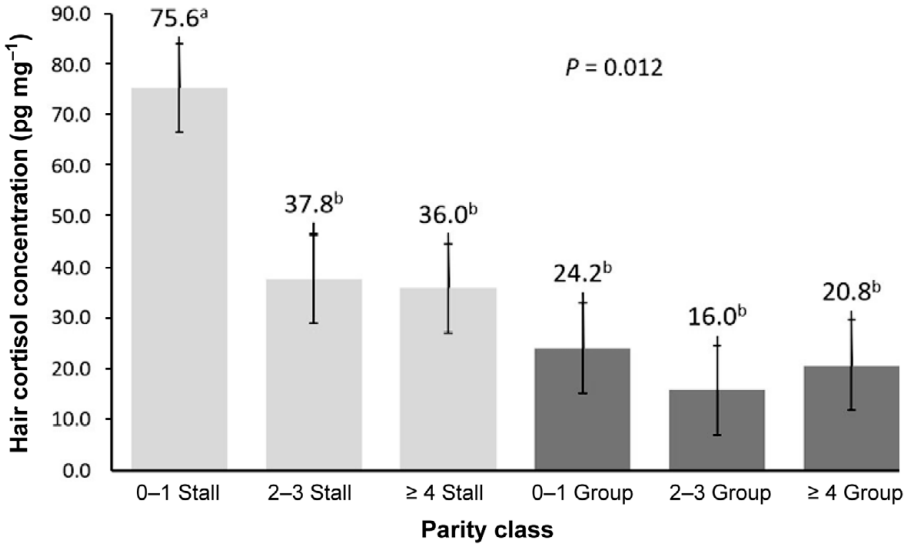


Figure 2. Interaction between parity class[1] and housing system (stall- and group-housed females) as regards hair cortisol concentration[2,3]. [1]Parity 0–1 refers to gilts and sows that have successfully completed one parity, 2–3 refers to sows successfully completing 2 or 3 parities, and ≥ 4 refers to sows successfully completing 4 or more parities. [2] Bars with different superscript a, b, c denote significant differences where $P \leq 0.05$. [3] 0–1 Stall (n = 11), 2–3 Stall (n = 10), ≥ 4 Stall (n = 13); 0–1 Group (n = 12), 2–3 Group (n = 7), ≥ 4 Group (n = 13). Light grey represents stall-housed females and dark grey represents group-housed females.

Table 2. Hair cortisol concentration in gestating sows by day of gestation[1] and parity class[2]; and average litter size[3] by parity class

	Day of gestation		Parity class				*P*-value		
Item	Day 37	Day 111	0–1	2–3	≥ 4	SEM	Day of gestation	Parity class	Interaction
Cortisol (pg mg^{-1})	29.4	39.8	48.8	28.5	28.4	8.9	0.095	0.008	0.601
Litter size (n)	–	–	15.1	16.8	16.9	0.51	–	0.0204	–

[1]Hair cortisol concentration was measured in hair growth between day 0 (breeding) and day 37 of gestation, and growth between day 74 and day 111 of gestation, representing the first and last third of gestation, respectively
[2]Parity 0–1 refers to gilts and sows that have successfully completed one parity, 2–3 refers to sows successfully completing 2 or 3 parities, and ≥ 4 refers to sows successfully completing 4 or more parities
[3]Litter size refers to total piglets born per litter
SEM: Standard error of the mean.

at mixing in group-housing systems increases overall sow stress. This suggests that the stress of confinement parity 0–1 stall-housed females experienced over the course of gestation was greater and persistent and led to chronically elevated cortisol. Adaptation, defined as lower prolonged stress, to stall housing may not occur until the third or fourth pregnancy.

The lack of difference in HCC between parity classes in group housing demonstrates that parity 0–1 females in group housing do not experience significantly higher stress than their older and more experienced counterparts. The experience of being bred for the first time and carrying a litter does not appear to cause significantly greater stress to a naïve female compared to multiparous females when housed in group pens. This result is consistent with Roelofs *et al.* (2019) who, similarly, did not detect a difference in HCC between multiparous and primiparous females in group housing.

It is generally recommended to acclimate gilts to stall housing for two to three weeks prior to boar exposure (Kraeling & Webel 2015), as pigs are known to be social animals (Barnett *et al.* 1985) and individual housing can induce stress, which can disrupt oestrus, prolong the oestradiol peak, and decrease early embryo survival (Roongsitthichai *et al.* 2011). Gilts in this study were acclimated to stall housing four weeks prior to first breeding. Despite this acclimation period, the results of this study indicate that gilts and parity 1 sows housed in stalls throughout gestation experienced higher stress than group-housed gilts and parity 1 sows and therefore were not adequately acclimated to stall confinement. This may be because of the restricted ability to choose whether to interact with other pigs and where to lie or walk: this freedom of choice is associated with improved welfare status (Englund & Cronin 2023).

An increase in HCC with progressing pregnancy is expected, as both fetal and maternal cortisol rise as parturition approaches. Fetal cortisol increases as gestation progresses (Silver & Fowden 1989) and in many mammals plays a role in inducing parturition (Decaluwe *et al.* 2012; Wood 2013). Cortisol is critical for late fetal development, assisting in lung and kidney maturation (Challis *et al.* 2000), skeletal growth in sheep (Fowden *et al.* 1995), gastrointestinal development, and the transition from trans-placental glucose supply to liver glycogen and gluconeogenesis (Fowden *et al.* 1995; Fishman *et al.* 2018). Maternal cortisol also contributes to fetal growth by promoting maternal insulin resistance (Herrera 2000), which reduces the female's ability to utilise glucose from the bloodstream, thereby increasing glucose availability for the litter. As glucose is the primary nutrient crossing the placenta, this promotes fetal growth in late gestation (Herrera 2000). The rise in cortisol in late gestation in stall-housed females may also be related to the increase in discomfort due to progressing pregnancy and space restriction as reported by Anil *et al.* (2006); however, there are no

https://doi.org/10.1017/awf.2026.10059 Published online by Cambridge University Press

data to our knowledge similarly associating late gestation HCC increase to discomfort in group-housed females.

There may be consequences regarding chronically heightened cortisol in all stages of gestation for litter productivity and piglet welfare. Elevated maternal glucocorticoids during gestation have been linked with lower offspring birth weights and weaning weights (Kranendonk *et al.* 2006), and lower lactation and nursery performance (Jarvis *et al.* 2006; Otten *et al.* 2015). Piglets from sows experiencing elevated glucocorticoids during gestation may be more likely to have poorer welfare, demonstrating more anxiety-related behaviour or aggression in novel situations and higher pain score during tail docking (Otten *et al.* 2015). In addition, gilts raised from mothers who had experienced stress during gestation appeared more restless and uncomfortable in the periparturient period (Jarvis *et al.* 2006; Rutherford *et al.* 2014), and in one trial tended to be more aggressive towards piglets (Jarvis *et al.* 2006); both frequent posture changes and sow aggression can result in newborn piglets being injured or killed, contributing to pre-weaning mortality. Therefore, chronic stress in sows can detrimentally affect the productivity and welfare of their offspring. Our results suggest that parity 0–1 sows housed in stalls are more likely to experience chronic stress than group-housed females and may therefore produce less economically productive offspring with lower welfare status. In this trial, piglet birth weights and mortality were not evaluated, so there is a need for future research to consider the productivity and welfare of piglets born from stall- and group-housed sows.

Study limitations

Research into sow housing can be challenging due to the statistical limitations of comparing individual sows (such as those in stalls) to groups of sows. The choice was made in this research to use individual 'sow' as experimental unit rather than 'pen': with respect to the primary variable of interest (hair cortisol), the individual animal is the most appropriate experimental unit. The 'stress' associated with a given group-housing pen is a dynamic interaction between individual animal characteristics (i.e. age, size, dominance) (Li *et al.* 2012; Wang & Li 2016) and cannot be suitably represented with an average value for all females within a pen. There is a precedent for this choice of statistical analysis in sow housing research, when using 'pen' as experimental unit is impractical (Chapinal *et al.* 2010; Bacci *et al.* 2014; Zhou *et al.* 2014; Min *et al.* 2020; Liu *et al.* 2021; Tokareva *et al.* 2022).

In this study, the floor space allowance and group size in the group pens is not analogous to what is commonly seen in modern commercial sow facilities. Proposition 12 – Prevention of Cruelty to Farm Animals Act (2018) requires 2.23 m^2 per sow, the EU requires 2.25 m^2, and Canada requires at least 1.8 m^2; and group sizes can be upwards of 50 sows per pen in group housing systems currently in use in the US (Li *et al.* 2018). In this study, floor space per sow was, on average, 2.79 m^2 and group pens held up to 25 sows. Therefore, the apparent lower stress of parity 0–1 sows in group housing compared to their stall-housed counterparts may, here, be related to large space allowance. Further investigation into the influence of stocking density and large group sizes on HCC in group-housed gestating sows is needed.

Despite differences in diets between the group- and stall-housed females, the lack of diet effect indicates gestation diet had limited impact on HCC. Further, total macro- and micro-nutrient supply for all females met or exceeded National Research Council (2012) requirement estimates and daily diet allocation was the same between all groups (i.e. allocated to maintain body condition score of 3) which means nutrient supply for fetal growth and metabolism of the pregnant females was similar across both groups.

In this study, possible seasonal influence on HCC was confounded with group. Due to farm management and research project requirements, stall-housed sows had to be bred in late fall and the group-housed sows bred in late winter; thus, having both housing types in each group was not possible. However, when the two stall-housed breed groups were compared to each other there was no difference in HCC; when group-housed breed groups were compared, there was a tendency for the second group to have a higher HCC ($P = 0.0752$). For the stall-housed sows, the average outside temperature ranged from 4–16°C, and the ambient barn temperature was maintained in the sows' thermoneutral zone. During the last part of gestation for the group-housed females (May and June) the average outside temperature ranged from 22–27°C, so it is likely that the ambient barn temperature was at times above the sows' thermoneutral zone. Previous research is inconclusive regarding the influence of season on HCC in swine. Heimbürge *et al.* (2020a) failed to detect a difference in HCC between samples collected in July versus January, and Wiechers *et al.* (2021) also failed to associate HCC with barn temperature. Bacci *et al.* (2014) found that HCC of sows was lower during summer months and higher barn temperatures. However, Escribano *et al.* (2023) observed elevated cortisone in the hair of pigs under heat stress conditions, and López-Arjona *et al.* (2020) found higher cortisone:cortisol ratios in the hair of gestating sows during summer months. In this trial, the group-housed sows in the second breed group may have experienced periods of heat stress, elevating cortisone and increasing HCC due to cross-reactivity in the assay. If hair grown in the summer has higher HCC, we expect in this trial that the group-housed females would have had higher HCC; however, that was not the case. There was little difference between HCC of stall-housed and group-housed females. Further, HCC was greatest in stall-housed gilts collected during cooler months, thus it is unlikely seasonality influenced HCC in this trial. In future research, consideration for and investigation into the possibility of seasonal effect on HCC is recommended, along with making use of highly precise analytical techniques, such as liquid chromatography mass spectometry, to distinguish and quantify cortisol and cortisone where heat stress could be a factor.

Animal welfare implications

Housing parity 0–1 sows in stalls during gestation promoted more severe and/or prolonged stress than the stress experienced by their group-housed counterparts. These results add to the body of data that demonstrate that group-housing gestating sows has the potential to improve their welfare by providing greater freedom of movement, freedom of choice, and opportunities for social interaction.

Conclusion

This study demonstrated that parity 0–1 females housed in stalls during gestation have higher HCC compared to group-housed sows, suggesting greater chronic stress. Further, stress associated with group sow housing may be more characteristic of acute stress around mixing rather than chronic. Finally, litter size did not influence HCC, and the impact of greater HCC on postnatal litter performance is unknown and should be explored in further work.

https://doi.org/10.1017/awf.2026.10059 Published online by Cambridge University Press

Acknowledgements. Financial support for this project was provided by USDA National Institute of Food and Agriculture (Hatch Project: SD00H666-18). We thank the University of Saskatchewan Western College of Veterinary Medicine for the training and resources involved in the laboratory processing of hair samples.

Competing interests. None.

References

Anil L, Anil S, Deen J and Baidoo SK 2006 Cortisol, behavioral responses, and injury scores of sows housed in gestation stalls. *Journal of Swine Health and Production* **14**(4): 196–201.

Bacci ML, Nannoni E, Govoni N, Scorrano F, Zannoni A, Forni M, Martelli G and Sardi L 2014 Hair cortisol determination in sows in two consecutive reproductive cycles. *Reproductive Biology* **14**: 218–223. https://doi.org/10.1016/j.repbio.2014.06.001

Barnett JL, Winfield CG, Cronin GM, Hemsworth PH and Dewar AM 1985 The effect of individual and group housing on behavioural and physiological responses related to the welfare of pregnant pigs. *Applied Animal Behaviour Science* **14**: 149–161. https://doi.org/10.1016/0168-1591(85)90026-7

Bozovic D, Racic M and Ivkovic N 2013 Salivary cortisol levels as a biological marker of stress reaction. *Medical Archives* **67**: 374. https://doi.org/10.5455/medarh.2013.67.374-377

Bruhn T 2020 *Impact of altering lys:energy ratio during gestation on sow productivity, piglet robustness, and piglet post-wean growth performance*. Master's thesis, South Dakota State University, Brookings, South Dakota, USA.

Bryan HM, Darimont CT, Paquet PC, Wynne-Edwards KE and Smits JEG 2013 Stress and reproductive hormones in grizzly bears reflect nutritional benefits and social consequences of a salmon foraging niche. *PLoS ONE* **8**: e80537. https://doi.org/10.1371/journal.pone.0080537

Bryan HM, Smits JEG, Koren L, Paquet PC, Wynne-Edwards KE and Musiani M 2015 Heavily hunted wolves have higher stress and reproductive steroids than wolves with lower hunting pressure. *Functional Ecology* **29**: 347–356. https://doi.org/10.1111/1365-2435.12354

Buhr BL 2010 *Economic impact of transitioning from gestation stalls to group pen housing in the US pork industry (Staff paper P10-4)*. University of Minnesota, Department of Applied Economics, USA.

Canadian Pork Council and National Farm Animal Care Council 2014 *Code of practice for the care and handling of pigs*. National Farm Animal Care Council: Canada.

Carlitz EHD, Kirschbaum C, Stalder T and Van Schaik CP 2014 Hair as a long-term retrospective cortisol calendar in orang-utans (*Pongo* spp): New perspectives for stress monitoring in captive management and conservation. *General and Comparative Endocrinology* **195**: 151–156. https://doi.org/10.1016/j.ygcen.2013.11.002

Challis JRG, Matthews SG, Gibb W and Lye SJ 2000 Endocrine and paracrine regulation of birth at term and preterm. *Endocrine Reviews* **21**: 514–550. https://doi.org/10.1210/edrv.21.5.0407

Chapinal N, Ruiz De La Torre JL, Cerisuelo A, Gasa J, Baucells MD, Coma J, Vidal A and Manteca X 2010 Evaluation of welfare and productivity in pregnant sows kept in stalls or in 2 different group housing systems. *Journal of Veterinary Behavior* **5**: 82–93. https://doi.org/10.1016/j.jveb.2009.09.046

Davenport MD, Lutz CK, Tiefenbacher S, Novak MA and Meyer JS 2008 A rhesus monkey model of self-injury: effects of relocation stress on behavior and neuroendocrine function. *Biological Psychiatry* **63**: 990–996. https://doi.org/10.1016/j.biopsych.2007.10.025

Decaluwe R, Janssens GPJ, Declerck I, De Kruif A and Maes D 2012 Induction of parturition in the sow. *Vlaams Diergeneeskundig Tijdschrift* **81**(3): 158–165. https://doi.org/10.21825/vdt.v81i3.18353

Dettmer AM, Novak MA, Suomi SJ and Meyer JS 2012 Physiological and behavioral adaptation to relocation stress in differentially reared rhesus monkeys: Hair cortisol as a biomarker for anxiety-related responses. *Psychoneuroendocrinology* **37**: 191–199. https://doi.org/10.1016/j.psyneuen.2011.06.003

Endo N, Yamane H, Rahayu LP and Tanaka T 2018 Effect of repeated adrenocorticotropic hormone administration on reproductive function and hair cortisol concentration during the estrous cycle in goats. *General and Comparative Endocrinology* **259**: 207–212. https://doi.org/10.1016/j.ygcen.2017.11.027

Englund MD and Cronin KA 2023 Choice, control, and animal welfare: definitions and essential inquiries to advance animal welfare science. *Frontiers in Veterinary Science* **10**: 1250251. https://doi.org/10.3389/fvets.2023.1250251

Escribano D, Contreras-Jodar A, López-Arjona M, Cerón JJ, Fàbrega E, Aymerich P and Dalmau A 2023 Changes in cortisol and cortisone in hair of pigs reared under heat stress conditions. *Frontiers in Veterinary Science* **10**: 1156480. https://doi.org/10.3389/fvets.2023.1156480

European Commission 2008 Council Directive 2008/120/EC of 18 December 2008 laying down minimum standards for the protection of pigs. *OJ L* **47**: 5–13.

Fairbanks LA, Jorgensen MJ, Bailey JN, Breidenthal SE, Grzywa R and Laudenslager ML 2011 Heritability and genetic correlation of hair cortisol in vervet monkeys in low and higher stress environments. *Psychoneuroendocrinology* **36**: 1201–1208. https://doi.org/10.1016/j.psyneuen.2011.02.013

Fishman R, Vortman Y, Shanas U and Koren L 2018 Cortisol advantage of neighbouring the opposite sex *in utero*. *Royal Society Open Science* **5**: 171636. https://doi.org/10.1098/rsos.171636

Fowden A, Apatu R and Silver M 1995 The glucogenic capacity of the fetal pig: developmental regulation by cortisol. *Experimental Physiology* **80**: 457–467. https://doi.org/10.1113/expphysiol.1995.sp003860

del González-de-la-Vara MR, Valdez RA, Lemus-Ramirez V, Vázquez-Chagoyán JC, Villa-Godoy A and Romano MC 2011 Effects of adrenocorticotropic hormone challenge and age on hair cortisol concentrations in dairy cattle. *Canadian Journal of Veterinary Research* **75**: 216–221.

Guzik AC, Matthews JO, Kerr BJ, Bidner TD and Southern LL 2006 Dietary tryptophan effects on plasma and salivary cortisol and meat quality in pigs. *Journal of Animal Science* **84**: 2251–2259. https://doi.org/10.2527/jas.2005-292

Heimbürge S, Kanitz E, Tuchscherer A and Otten W 2020a Within a hair's breadth – Factors influencing hair cortisol levels in pigs and cattle. *General and Comparative Endocrinology* **288**: 113359. https://doi.org/10.1016/j.ygcen.2019.113359

Heimbürge S, Kanitz E, Tuchscherer A and Otten W 2020b Is it getting in the hair? – Cortisol concentrations in native, regrown and segmented hairs of cattle and pigs after repeated ACTH administrations. *General and Comparative Endocrinology* **295**: 113534. https://doi.org/10.1016/j.ygcen.2020.113534

Hernandez L, Dunn J, Wenninghoff J, Hesse A and Levesque CL 2021 Mint oil and whole yeast cell in sow diets enhance offspring performance in the post-weaning period. *Front Vet Sci* **8**: 658956. https://doi.org/10.3389/fvets.2021.658956

Herrera E 2000 Metabolic adaptations in pregnancy and their implications for the availability of substrates to the fetus. *European Journal of Clinical Nutrition* **54**: S47–S51. https://doi.org/10.1038/sj.ejcn.1600984

Iob E and Steptoe A 2019 Cardiovascular disease and hair cortisol: a novel biomarker of chronic stress. *Current Cardiology Reports* **21**: 116. https://doi.org/10.1007/s11886-019-1208-7

Jarvis S, Moinard C, Robson SK, Baxter E, Ormandy E, Douglas AJ, Seckl JR, Russell JA and Lawrence AB 2006 Programming the offspring of the pig by prenatal social stress: Neuroendocrine activity and behaviour. *Hormones and Behavior* **49**: 68–80. https://doi.org/10.1016/j.yhbeh.2005.05.004

Koketsu Y and Iida R 2017 Sow housing associated with reproductive performance in breeding herds. *Molecular Reproduction and Development* **84**: 979–986. https://doi.org/10.1002/mrd.22825

Kraeling RR and Webel SK 2015 Current strategies for reproductive management of gilts and sows in North America. *Journal of Animal Science and Biotechnology* **6**: 3. https://doi.org/10.1186/2049-1891-6-3

Kranendonk G, Hopster H, Fillerup M, Ekkel ED, Mulder EJH, Wiegant VM and Taverne MAM 2006 Lower birth weight and attenuated adrenocortical response to ACTH in offspring from sows that orally received cortisol during gestation. *Domestic Animal Endocrinology* **30**: 218–238. https://doi.org/10.1016/j.domaniend.2005.07.001

Li YZ, Wang LH and Johnston LJ 2012 Sorting by parity to reduce aggression toward first-parity sows in group-gestation housing systems. *Journal of Animal Science* **90**: 4514–4522. https://doi.org/10.2527/jas.2011-4869

Li YZ, Cui SQ, Yang XJ, Johnston LJ and Baidoo SK 2018 Minimal floor space allowance for gestating sows kept in pens with electronic sow feeders on fully

slatted floors. *Journal of Animal Science* **96**: 4195–4208. https://doi.org/10.1093/jas/sky282

Liu X, Song P, Yan H, Zhang L, Wang L, Zhao F, Gao H, Hou X, Shi L, Li B and Wang L 2021 A comparison of the behavior, physiology, and offspring resilience of gestating sows when raised in a group housing system and individual stalls. *Animals* **11**(7): 2076. https://doi.org/10.3390/ani11072076

López-Arjona M, Tecles F, Mateo SV, Contreras-Aguilar MD, Martínez-Miró S, Cerón JJ and Martínez-Subiela S 2020 Measurement of cortisol, cortisone and 11β-hydroxysteroid dehydrogenase type 2 activity in hair of sows during different phases of the reproductive cycle. *The Veterinary Journal* 259–260: 105458. https://doi.org/10.1016/j.tvjl.2020.105458

Macbeth BJ, Cattet MRL, Stenhouse GB, Gibeau ML and Janz DM 2010 Hair cortisol concentration as a noninvasive measure of long-term stress in free-ranging grizzly bears (*Ursus arctos*): considerations with implications for other wildlife. *Canadian Journal of Zoology* **88**: 935–949. https://doi.org/10.1139/Z10-057

Min Y, Choi Y, Kim J, Kim D, Jeong Y, Kim Y, Song M and Jung H 2020 Comparison of the productivity of primiparous sows housed in individual stalls and group housing systems. *Animals* **10**(11), 1940. https://doi.org/10.3390/ani10111940

Morgan L, Meyer J, Novak S, Younis A, Ahmad WA and Raz T 2021 Shortening sow restraint period during lactation improves production and decreases hair cortisol concentrations in sows and their piglets. *Animal* **15**: 100082. https://doi.org/10.1016/j.animal.2020.100082

National Research Council 2012 *Nutrient Requirements of Swine*, 11th Review Edition. National Academies Press: Washington, DC, USA.

Nejad JG, Lohakare JD, Son JK, Kwon EG, West JW and Sung KI 2014 Wool cortisol is a better indicator of stress than blood cortisol in ewes exposed to heat stress and water restriction. *Animal* **8**: 128–132. https://doi.org/10.1017/S1751731113001870

Otten W, Kanitz E and Tuchscherer M 2015 The impact of pre-natal stress on offspring development in pigs. *The Journal of Agricultural Science* **153**: 907–919. https://doi.org/10.1017/S0021859614001361

Prims S, Vanden Hole C, Van Cruchten S, Van Ginneken C, Van Ostade X and Casteleyn C 2019 Hair or salivary cortisol analysis to identify chronic stress in piglets? *The Veterinary Journal* **252**: 105357. https://doi.org/10.1016/j.tvjl.2019.105357

Proposition 12—Prevention of Cruelty to Farm Animals Act 2018 *Legislative Analyst's Office*. https://lao.ca.gov/ballot/2018/prop12-110618.pdf (accessed 4 December 2025).

Roelofs S, Godding L, De Haan JR, Van Der Staay FJ and Nordquist RE 2019 Effects of parity and litter size on cortisol measures in commercially housed sows and their offspring. *Physiology & Behavior* **201**: 83–90. https://doi.org/10.1016/j.physbeh.2018.12.014

Roongsitthichai A, Suwimonteerabutr J, Kaeoket K, Koonjaenak S and Tummaruk P 2011 Association between serum cortisol and progesterone concentrations and the infiltration of immune cells in the endometrium of gilts with vaginal discharge. *Comparative Clinical Pathology* **20**: 563–571. https://doi.org/10.1007/s00580-010-1033-2

Russell E, Koren G, Rieder M and Van Uum S 2012 Hair cortisol as a biological marker of chronic stress: Current status, future directions and unanswered questions. *Psychoneuroendocrinology* **37**: 589–601. https://doi.org/10.1016/j.psyneuen.2011.09.009

Rutherford KMD, Piastowska-Ciesielska A, Donald RD, Robson SK, Ison SH, Jarvis S, Brunton PJ, Russell JA and Lawrence AB 2014 Prenatal stress produces anxiety prone female offspring and impaired maternal behaviour in the domestic pig. *Physiology & Behavior* **129**: 255–264. https://doi.org/10.1016/j.physbeh.2014.02.052

Ryan EB, Fraser D and Weary DM 2015 Public attitudes to housing systems for pregnant pigs. *PLOS ONE* **10**: e0141878. https://doi.org/10.1371/journal.pone.0141878

Salak-Johnson JL 2017 Social status and housing factors affect reproductive performance of pregnant sows in groups. *Molecular Reproduction and Development* **84**: 905–913. https://doi.org/10.1002/mrd.22846

Schulz LL and Tonsor GT 2015 The US gestation stall debate. *Choices* **30**. https://doi.org/10.22004/ag.econ.196759

Silver M and Fowden AL 1989 Pituitary-adrenocortical activity in the fetal pig in the last third of gestation. *Quarterly Journal of Experimental Physiology* **74**(2): 197–206. https://doi.org/10.1113/expphysiol.1989.sp003255

Terwissen CV, Mastromonaco GF and Murray DL 2013 Influence of adrenocorticotrophin hormone challenge and external factors (age, sex, and body region) on hair cortisol concentration in Canada lynx (*Lynx canadensis*). *General and Comparative Endocrinology* **194**: 162–167. https://doi.org/10.1016/j.ygcen.2013.09.010

Tokareva M, Brown J, MacPhee D, Janz D and Seddon Y 2022 The effect of providing a greater freedom of movement through periodic exercise on the welfare and stress physiology of stall-housed gestating sows and on piglet behaviour. *Animal Welfare* **31**: 293–308. https://doi.org/10.7120/09627286.31.3.002

Wang LH and Li YZ 2016 Effect of continuous access to feeding stalls during mixing on behavior, welfare, and performance of group-housed gestating sows in different social ranks. *Canadian Journal of Animal Science* **96**: 386–396. https://doi.org/10.1139/cjas-2015-0054

Wiechers DH, Brunner S, Herbrandt S, Kemper N and Fels M 2021 analysis of hair cortisol as an indicator of chronic stress in pigs in two different farrowing systems. *Frontiers in Veterinary Science* **8**: 605078. https://doi.org/10.3389/fvets.2021.605078

Wood CE 2013 Development and programming of the hypothalamus-pituitary-adrenal axis. *Clinical Obstetrics and Gynecology* **56**: 610–621. https://doi.org/10.1097/GRF.0b013e31829e5b15

Yamada J, Stevens B, De Silva N, Gibbins S, Beyene J, Taddio A, Newman C and Koren G 2007 Hair cortisol as a potential biologic marker of chronic stress in hospitalized neonates. *Neonatology* **92**: 42–49. https://doi.org/10.1159/000100085

Zhou Q, Sun Q, Wang G, Zhou B, Lu M, Marchant-Forde JN and Zhao R 2014 Group housing during gestation affects the behaviour of sows and the physiological indices of offspring at weaning. *Animal* **8**(7): 1162–1169.

https://doi.org/10.1017/awf.2026.10059 Published online by Cambridge University Press