

Behavioral Responses of Sows Exposed to Conventional Methods or Precision-Technology to Mitigate Piglet-Crushing

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Abstract

A Precision Animal Management (PAM) toolset (SmartGuard; SwineTech Inc., Cedar Rapids, IA, USA) was developed to intervene piglet-crushing events using a vibration followed by Electrical Impulse (VIB+EI). The objective was to evaluate sow startle, coping, and nursing responses to three crushing-mitigation stimuli: Vibration-only (VIB; n=16), VIB+EI (n=18), or Conventional-methods (CONV; 3 hand slaps; n=18). Sows were exposed to a piglet distress call and the ensuing impulse for 6 sessions on d 1-4, relative to farrowing. Startle-response measures included Heart Rate (HR), cortisol secretion, and behaviors from live observation. Sows were fitted with HR-monitors before each session on days 1-4. Cortisol from ear-vein blood (100 µL) was measured before sessions-1 and-6, and after sessions-2 and-6. A novel startle-index was calculated from live observations during sessions (0=silent, lie; 100=jump, bite sow) and expressed as a percent. Coping and nursing behaviors were quantified from video collected after each session, and after ear-vein blood was collected on d 5, 7, and 9, relative to farrowing. Circadian cortisol was measured using AM and PM ear-vein blood samples for d 0-4, 5, 7, and 9, relative to farrowing. A large proportion of live observations indicated that CONV-sows only sat upright after stimuli. In contrast, most VIB+EI-sows stood-up completely ($\chi^2=207.14$; N=312; $p<0.01$), although many jumped to the upright position ($\chi^2=44.9$; N=216; $p < 0.01$). Both CONV- and VIB+EI sows vocalized ($\chi^2=199.19$; N=312; $p<0.01$), but biting was a rare occurrence. The VIB-sows had the lowest startle-index, with minimal disturbance during sessions. The CONV- and VIB+EI-sows displayed a 31 and 50% startle index, respectively (± 2.1 SEM; $p<0.01$). There were minimal differences in HR or cortisol measures among treatments ($p>0.10$). After sessions, VIB+EI-sows had greater oral behaviors and standing durations, than CONV- and VIB-sows ($p<0.05$). The CONV- and VIB+EI-sows had similar nursing and standing behaviors, which were less than VIB-sows ($p<0.05$). Cortisol measures and coping- and nursing-behaviour differences were not observed on d 5, 7, or 9 ($p>0.10$). These results indicated that if PAM-technology should replace conventional methods, producers are not likely to observe long-term effects on sow behaviors. The results from this experiment were used to adjust the stimuli settings for the PAM-technology on commercial sow operations to reduce jumpings.

Keywords: Machine-learning; Lay-on, Porcine; Welfare

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Introduction

Despite the common use of farrowing stalls in North America, 1 in 10 piglet deaths result from crushing by the sow, and nearly

half of those deaths occur during the first 3 d after birth [1,2]. The farrowing stall is the most common technology used by North American producers to prevent death loss from crushing. There are multiple animal welfare tradeoffs with the use of farrowing

stalls: less piglets suffer from crushing, but the sow's behavior repertoire is restricted during lactation [3,4]. Marchant et al. [5], indicated that increased space-allowance and non-nutritive substrates alleviated sow distress and discomfort during farrowing and lactation-period [3,5]. However, in pens with substrate, the crushing-rate was 12.1%, compared to a 5.6% crushing rate in stalls [3]. Marchant Forde [5] reported 17% and 14% crushing in open-barn systems compared to 8% crushing in stalls. In the U.S., over 80% of producers choose to use the standard farrowing stall as their primary toolset to prevent crushing while still managing sows at the individual level [6].

New technologies and methods were introduced to mitigate crushing by drawing the piglets away from the sow. Methods included sloped floors, solid sloped walls, and supplemental heating [7-11]. To mitigate mortality around the time of parturition, some producers use round-the-clock specialists to observe peri-parturient challenges [12]. This method of increased human intervention decreased stillborn and mortality rates [12]. Nonetheless, many U.S. producers have a 1:250 or 1:500 human:sow ratio, which limits efficient individual care at farrowing. In addition, intense operations face high turn-over rates for labor, which influences variation in animal-caretaker's experience and temperament [13]. In the coming years US pork production should prepare for mandates and production practice requirements that are driven by legislation and animal welfare similar to other nations. The US pork production sector needs alternative solutions for mitigating crushing, with or without the use of restricted housing [14].

Preweaning mortality may be further reduced through Precision Animal Management (PAM) technologies that incorporate machine-learning and are computer sensory-derived [15]. A PAM-technology (SmartGuard, SwineTech Inc., Cedar Rapids, IA, USA) was developed to identify a piglet distress call and stimulate the sow to stand. The stimuli were modelled from medical devices (Transcutaneous Electrical Nerve Stimulation, TENS). Crushing risk is greatest during the first 4 days relative to farrowing. Therefore the device only is attached to the sow for this period [1,16]. The technology registers a distress call from a piglet compared to a recorded crushing event, and sensors determine the location and sow's structural position using deep frame learning. If the sow is lying, the technology provides a vibration (VIB) signal followed by an electrical impulse (VIB+EI; maximum values 1000 v, 1 s). The Electrical Impulse (EI) stimuli used in this system is an additional animal welfare tradeoff to the farrowing stall. Utilizing electrical impulses on animals is a well-documented issue in both companion and production animals [17,18]. Weary et al. [1] reported that if crushing occurred in 60 seconds or less, piglets survived, but the risk of mortality was greatest if they were trapped under the sow for 4 or more minutes. Nonetheless, the VIB+EI presents additional ethical concerns because best management practices indicate that the electric-prod should be used as a last resort [17,19]. Producers that follow the "no electric-prod" guidelines are more likely to intervene piglet crushing with hands-slaps [20]. Both hand-slaps and the manual electric prod application are dependent on the

human's sensory and decision-making response. The human response is confounded by emotion (i.e., panic, frustration), impulse responses, and previous experience [21]. Therefore, a response to a sow crushing a pig has inherent subjectivity from person-to-person, whereas responses from PAM technology are more objective and efficient, and present less variability.

The Conventional method (CONV; hand-slaps) may cause sows to associate aversive stimuli with humans rather than the distress call of a piglet [22]. Sows are cognitively capable of associating actions with consequences [23]. But, the constant noise in a farrowing house may cause sows to easily habituate to distress calls. Chaple et al. [24] in a similar production atmosphere, found that increasing noise and activity along with sow age impacts responsiveness to piglets. An additional stimuli that is sensed over all other inputs is inevitably needed. Thus, for the current study, the first objective was to evaluate the sow's startle-response to the PAM-stimuli and compare it to the conventional methods (CONV; 3 hand slaps) and a control (VIB-vibration only) during play-back of a piglet distress call. The second objective was to determine if the stimuli influenced the normal behaviors of the sow within the 20 minutes after treatment and, in the 9 days after the stimuli were applied.

Materials and Methods

Animals and housing

The experiment was conducted in October and November 2017 at Kansas State University Swine Teaching and Research Center (Manhattan, KS). Animals were housed and managed in accordance to the 'Guide for the Care and Use of Agriculture Animals in Research and Teaching' [25]. All procedures were approved by the Kansas State University Institutional Animal Care and Use Committee (IACUC; Protocol #3913). Fifty-eight sows (DNA line 241; primiparous and multiparous; pre-farrow body weight, 246.54 \pm 56.34 SD kg) were weighed and enrolled within in two blocks (**Figure 1**).

Sows were housed in standard farrowing stalls (length 2.3 m; width 0.43 m; height 1.5 m). No substrate was provided, and it was ensured that the mild impulse was not propagated by the metal crates by a researcher. Sows were provided ad libitum feed with automated feeders (GESTAL, Jyga Technologies, St. Lambert De Lauzon, Qc, Canada) and waterers (Aqua Series, Hog Slat, Columbus, NE). After farrowing (\pm 0.84 d SD), sows were weighed. Feed intake was measured daily. Piglets were offered alternative heat sources via lamps, at the rear of the stall. Piglets were processed at age 1 d, on a per farrowing day basis (day 0 farrowing \pm 0.74 d SD block 1; \pm 0.93 d SD block 2). The sows' pre-, post-partum, and weaning weights were collected, piglets were weighed at birth, age 7 d and at weaning (age 21 \pm 0.93 d SD; **Table 1**). The cull-rate and sow return-to-estrus day (full standing-estrus) was measured after weaning.

Treatments

At farrowing, (0 \pm 0.84 days SD), sows were randomly assigned to 1 of 3 stimuli treatments (VIB, n=16; CONV, n=18; VIB+EI

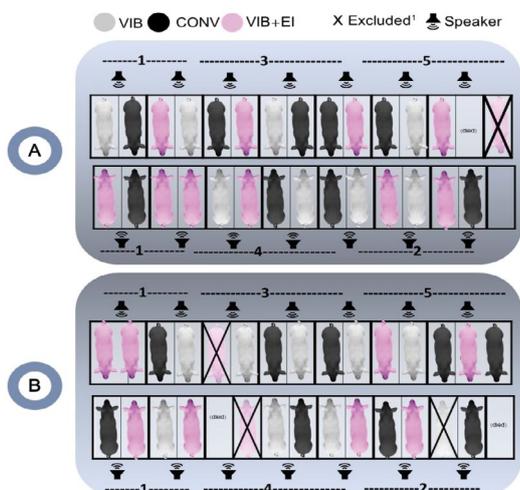


Figure 1 Experimental treatment layout. A total of 56 sows were enrolled over 2 blocks (A, October 2017; B November 2017). Three sows died due to farrowing complications. At farrowing, sows were randomly assigned 3 treatments: Vibration-only (VIB n=16); Conventional (CONV n=18; 3 hand slaps); Vibration+Electrical Impulse (VIB+EI n=18). Sow data sets were excluded (X) for one VIB-sow and 3 VIB+EI due to technical issues with treatments. Every other farrowing stall had a speaker fixed on the back and a camera (90° angle) above two sows. Six sessions were conducted on d 1-4 relative to farrowing in groups of 6 (numbers indicate group treatment order). A distress call was played during each session on days 1-4 relative to farrowing (± 0.84 SD). Video recording was done via one camera fixed to the ceiling above every other stall.

n=18; **Figure 1**). On d 1 relative to farrowing, all sows had a pocket (SmartGuard wearable patches, SwineTech) fastened to the flank region below the hair line. Sows were exposed to the stimuli-treatments in 6 sessions over 4 days in the afternoon and evening (**Figure 2**). At the back of every other stall, a speaker was fixed (**Figure 1**). Sessions were applied in groups of 5-6 sows (**Figure 1**). Before a group-session began, the sows were in the sternal-recumbent position. The group-session began when a 16 s piglet distress call from a crush event was played over speakers (**Figure 1**) in loop for up to a min (4400 Hz: **Figure 2**). Therefore, the entire barn was treated with 5 distress calls per session. Vibration (VIB) sows had devices in their pockets that only produced one vibration stimuli to bare skin (VIB; SmartGuard vibration max 0.4 J for 1 s) synonymous to the vibration indication in any modern handheld device. The same handler applied three open hand-slaps (2 on the back and 1 on the belly, 2-3 s) during each session to the Conventionally-treated (CONV) sows (**Figure 2**). Care was taken for the handler applying the CONV treatment to only be seen by the CONV sows, to avoid confounding human interaction with the PAM-stimuli. Sows treated with PAM-stimuli had the same vibration applied to the bare skin as described above for 1 s, followed by a 1-2 second pause to allow for a response, then an electrical impulse for an additional second (VIB+EI; maximum values, 500 v, 1 s; **Figure 2**) to simulate action of the ‘Smartguard’ technology provided by the funding source.

Startle-response measures

On session-days, sows were fitted with heartrate monitors 1 hour before the first session began and worn one hour after the last session ended (PolarH10 heart monitors; POLAR USA, Warminster,

Table 1: Summary of measures for sows treated with Vibration-only (VIB, n=16).

Behavior or physiological variable	Time frame				Note
	Farrowing ¹	Startle-Response ²	Coping-Response ³	Long-term changes ⁴	
Heart Rate		✓	✓		Belts fastened 1 h before the first session to 1 h after the last session
Cortisol Stress Response	✓*	✓			Sampling limited to d 1 and d 4
Circadian Cortisol	✓*			✓	Sampled at morning and evening for d 1,4,5,7,9
Live Observation		✓			
Video Observation			✓	✓	20 minutes after each session and after blood collection on d 5,7,9
Piglet Total Serum Protein	✓			✓	Sampled 3 per sow on d 7
Piglet Body Weight	✓			✓	In addition, bodyweight at weaning
Sow Feed Intake	✓	✓	✓	✓	At weaning ADFI
Sow Body Weight	✓✓			✓	Before and after farrowing and at weaning
Sow Cull or Rstrus Day				✓	Culled or return to estrus after weaning

conventional hand-slaps (CONV, n=18), or Vibration and Electrical Impulse (VIB+EI, n=18)

*Used as covariate in model

¹Farrowing includes the time from the sow expelling the first piglet to the last

²Startle-Response indicates the immediate response of sows following treatment by VIB, CONV or VIB+EI during sessions 1-6 on days 1-4 post farrowing

³Coping-Response indicates the time period in the 20 minutes immediately post treatment of VIB, CONV or VIB+EI during sessions 1-6 on days 1-4 post farrowing

⁴Longterm changes indicate behaviors or physiological parameters taken on days 5,7 and 9 relative to farrowing, or at conclusion of lactation period

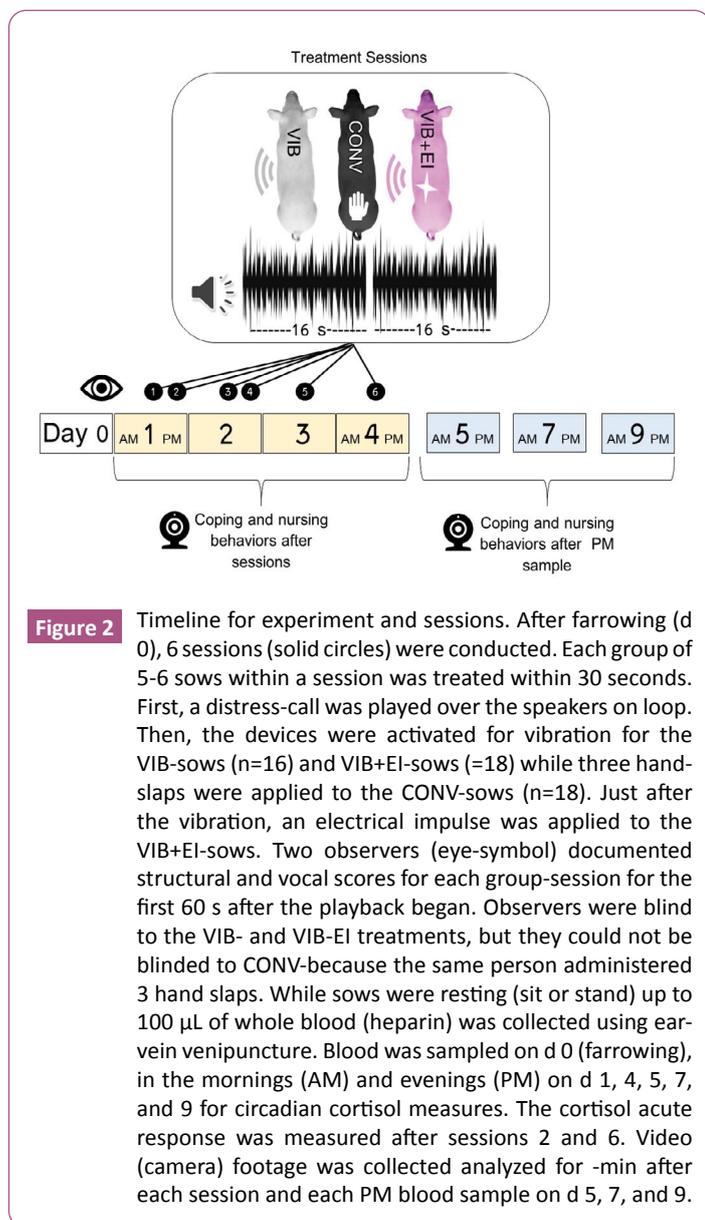
✓ Indicates at what timeframe (top) each measure (left) was collected

PA; **Table 1**). In addition to the heart rate measures (HR; max, min, mean), the latency for each sow's heart rate to return to

that of resting was measured (RR, return to resting HR). If the sow never changed from the lie position to an upright position (sit, stand, jump) during a session, RR could not be measured or included in the data set.

For each session, 2 observers evaluated the behaviors included in the startle response (**Table 2**). The 2 observers stood behind each group of sows and used binary scoring to record the structural-position, vocalization-type, and if any bites occurred (**Table 2**).

The observers had greater than >95% inter-observer agreement (kappa statistic>0.95) for all sessions and groups. The structural behaviors were prioritized by researchers with over 50 years of combined production swine experience, from least active to most active (lie, sit, stand, jump). The vocalization type (as noted by the trained observers) and bite were prioritized from least to most egregious (silent, grunt, bark, squeal, bite). A startle-index was formulated (**Figure 3**) so that the least active, silent sow would score a 0, and the most active biting sow would score a 100.



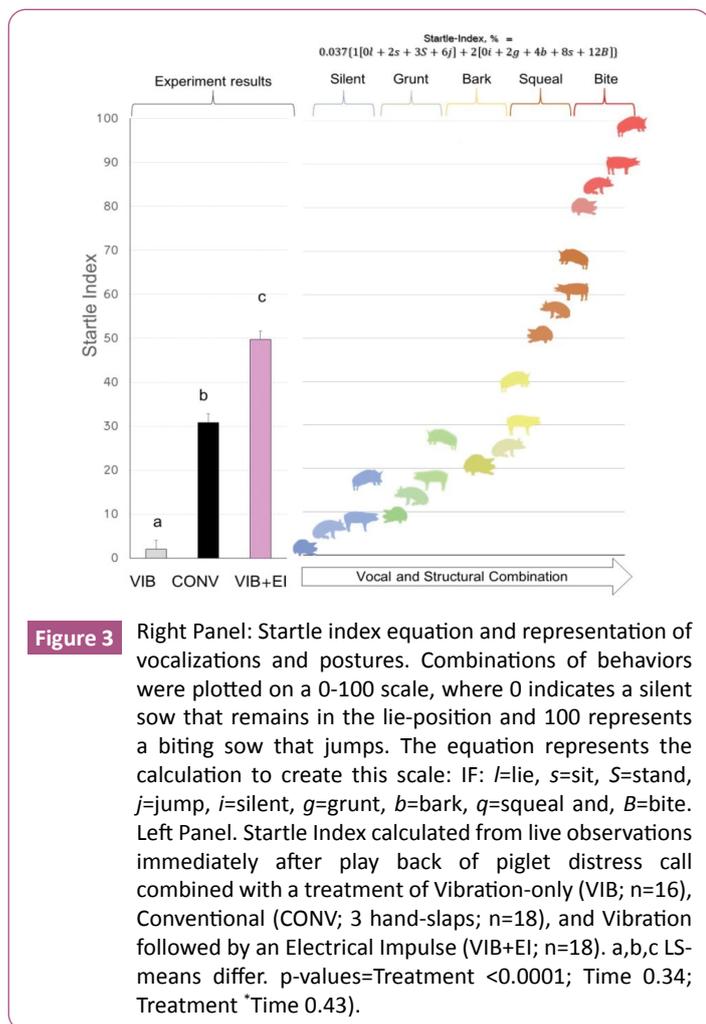
Cortisol analyses

All blood samples were collected from the ear vein (100 µL; 26-gauge, 1-cc syringes with heparin) while sows were in a resting-position (lateral or sternal recumbency). Plasma was harvested after centrifugation and frozen at -20°C until analysis. Samples analyzed for circadian cortisol included collection times 0600h and 1700h on days 0, 1, 4, 5, 7 and 9, relative to farrowing (**Figure 2 and Table 1**). Samples collected just before session 1, after session 2, and before and after session 6 were analyzed for startle-responses to stimuli. Cortisol analysis was performed using a commercially available ELISA (DetectX; Arbor Assays, Anne Arbor, MI). The intra- and inter-assay coefficients of variation were 10.4 and 12.1%, respectively. For circadian cortisol, area under the curve (AUC) was calculated in SigmaPlot (v 13.0) using cortisol samples for farrowing (d 0) and the morning and evening (0600 h, 1700 h) on d 1, 5, 7, 9 (**Table 1**). Farrowing cortisol was collected after the third pig in each litter was born and used as a covariate for all other cortisol models. Cortisol for pre- and post-treatment was expressed as a percent change by subtracting the post treatment sample from the pre-treatment sample and converting to a percent scale. Similarly, for circadian cortisol, the

Table 2: Live behavioral observations included in the novel startle index. During a piglet distress call play back, sows were stimulated with Vibration-only (VIB, n=16), Conventional hand-slaps (CONV, n=18), or Vibration and Electrical Impulse (VIB+EI, n=18). Behaviors were analyzed during 6 sessions (live observation).

Behaviors	Description
Body-structure	
Sit	Only front legs in upright position
Stand	All legs supporting sow
Jump	Sudden propulsive action to standing
Vocalizations	
Grunt	Short-duration, low frequency (<1 second) burst of noise
Squeal	High pitched scream, >1 second
Bark	A short sudden loud burst of noise at a lower pitch than squeal
Bite	A sudden snap of mouth, without audible sound

Live observers recorded frequency just after a group of 5-6 sows were exposed to piglet distress call and their respective treatment and used in calculation of the startle index



AM sample was subtracted from the PM sample and converted to a percent.

Coping and nursing behaviors

Prior to sows entering farrowing, one camera (Points North Surveillance Inc., Auburn, ME, USA) was installed on the ceiling above every 2 crates and continuous video was collected. Twenty-minutes of video footage was sampled after each session and after the additional ear vein blood collection on days 5, 7, and 9 relative to farrowing (Figure 2 and Table 1). One trained observer timestamped the 20-min videos for each sow and her litter (Table 2) using specialized software (Observer XT 11 Noldus, Leesburg VA). Additional continuous behaviors were analyzed using wearable devices (see supplementary wearable device methods).

Piglet Total Plasma Protein (TPP) and weights

Piglet total plasma protein (Reichert-Jung 0 50° Brix hand-held refractometer) was used as a nursing-quality measure in addition to duration of sow lie-lateral and nursing behaviors (Table 3). At birth, each pig was weighed, and 500 µL of umbilicus blood was stripped into a heparinized tube. All pigs were weighed on day 7 relative to farrowing and 500 uL of whole blood was collected from every other gilt via jugular-venipuncture (Table 1). Plasma was harvested after centrifugation and stored at -20°C until refractometer analysis. Each subsampled gilt's age d 0 TPP measure was subtracted from age d 7 and the percent change in TPP was calculated.

Table 3: Ethogram. During a piglet distress call play back, sows were stimulated with Vibration-only (VIB, n=16), Conventional hand-slaps (CONV, n=18), or Vibration and Electrical Impulse (VIB+EI, n=18). Behaviors were analyzed during sessions (live observation), 20 min after sessions (video), and 20 min after ear vein blood was collected on d 5,7, and 9 relative to farrowing (0).

Behaviors	Description
Oral behaviors	
Headstill	Sow's head remains immobile
¹ Non-nutritive	
Floor	Directed at floor
Stall	Directed at stall
Feeder	Directed at Feeder, not eating
Piglets	Directed at piglets
¹ Nutritive	
Eat	Sow's head in the feeder with locomotion
Drink	Sow's snout and mouth on water nipple
Body-Structure	
Sit	Only front legs in upright position
Stand	All legs supporting sow
Jump	Sudden propulsive action to standing
Lie Sternal	Sow lying on her stomach
Lie Lateral	Sow lying on her side
Nursing	
1	≥ 1 ≤ 4 piglets manipulate udder
5+	≥ 5 manipulate the udder

¹Behaviors timestamped from video footage for 20 minutes after each session or blood sample collection on d 5,7, and 9 relative to farrowing. Definitions adapted from Hurnik et al., 1995 and Hulbert and McGlone 2006. Sham chewing was considered, but not observed in this project

Statistical Analysis

For cortisol and nominal behavior data, a general linear mixed model was fit using proc GLIMMIX of SAS (v. 9.4, SAS Inst. Inc., Cary, NC, USA) with the fixed effects of time, treatment, and the interactions of treatment × time. Sow nested within treatment was included as the random effect. Production parameters of number weaned, daily feed intake, total litter weight, sow weight at weaning were analyzed using the GLIMMIX procedure of SAS with sow body weight as a covariate. Fixed effects were sow ID and parity, with treatment included as a random effect. All data was checked for normality using the Kolmogorov-Smirnov test in the UNIVARIATE procedure of SAS (v. 9.4, SAS Inst. Inc., Cary, NC, USA) and transformation of Log (10) or SquareRoot were made when necessary. Tukey-Kramer adjustment was made to account for type-1 error in detailed pairwise comparisons within a subset of data. Categorical data were subjected to chi-square analyses and results are presented as observed, expected and residual, with significance levels set at $p < 0.05$ and tendencies at $p < 0.10$.

Results and Discussion

Precision animal management systems may help monitor and mitigate treatments at the individual level, potentially further reducing preweaning mortality [11,15,26]. Due to these advances in technology, there now exists the ability to monitor individual sows and incite a standing response when piglets vocalize at distress levels. Besides the farrowing stall, the other popular crushing mitigation strategy available to animal caretakers in the US is round-the-clock monitoring. This method can detect piglet crushing but uses hand-slaps or other means to incite standing in sows. This approach is not sustainable and is subject to human-error and egregious handling when sows refuse to respond to initial hand slaps. The trade-off between inciting a standing response by applying electrical impulse to the sow during crushing is that human-error is eliminated and piglets are saved, but the distress from the electrical impulse may cause long-term behavioral changes that are detrimental to sow welfare. Therefore, behavioral and physiological implications are discussed herein. The use of highly prolific genetic lines cannot be overlooked as a potential for increased piglet crushing based simply on the fact of more piglets offer more opportunity for crushing [27]. For the present study, 2 sows were euthanized at farrowing, for dystocia-related reasons (Figure 1). Four sows

were removed (Figure 1), due to technical difficulties (incorrect treatment). Thus, 52 sows were analyzed (VIB, $n=16$; CONV, $n=18$; VIB+EI, $n=18$).

Startle-response

Heart rate monitors potentially collect data at a high sampling rate and can provide the sympathetic nervous system response to stimuli [15,28,29]. After the sessions in this experiment, there were no treatment, or treatment × time significant effects for most of the Heart Rate (HR) measures ($p > 0.10$; Table 4).

Maximum HR irrespective of treatment was 118 ± 4.33 bpm (Table 4). These maximum HR values are comparable to maximum HR (116 to 129 ± 5 bpm) for gestating and farrowing sows in stalls while they are in the stand-position [28]. There was a tendency for treatment by time interaction for the minutes to return to resting heart rate ($p=0.07$; Supplementary Figure 1). After session 5 and 6 CONV or VIB+EI sows, respectively, had a greater return to resting HR than the other sows ($p < 0.05$; Supplementary Figure 1).

Cortisol is a common biomarker to measure stress responses [30]. Therefore, a blood sample was collected before the first and sixth session, and after the second and sixth session. Collection was limited to just 100 μ L from the ear vein to prevent disturbing the sows while they remained in the rest-position (sit or lie). From eustress or distress, the stress axis is activated within 5-20 minutes after stimuli [30,31]. For this experiment, there were few treatment × time significant effects for acute cortisol responses to the treatments ($p > 0.05$; Table 5). Farrowing blood samples had the greatest cortisol concentrations. Therefore, this sample was used as a covariate for the acute cortisol response. Parturition causes a significant increase in cortisol, which is thought to help regulate inflammation [32]. However, this makes cortisol a challenging biomarker for acute stress in the perinatal period. Nonetheless, when the percent change was considered, more CONV-sows had a negative percent change than VIB+EI sows ($p < 0.01$; Table 5). The authors suspect that the CONV-sows may have mounted more of a stress-response to humans than VIB+EI sows, so by the time the blood was collected after the sessions, a negative-feedback had already occurred. VIB+EI sows had similar percent change to the VIB-only sows ($p > 0.10$; Table 5). Hemsworth's review [22] of stockperson attitude and handling methods indicates that gentler handling is more neutral

Table 4: Session Heart Rate Measures. Sows were treated with Vibration-only (VIB, $n=16$), Conventional (CONV, $n=18$; 3 hand slaps), or Vibration and Electrical Impulse (VIB+EI, $n=18$) during a play back of a distress piglet call (starting point, 20 minutes after sessions).

	Treatment			SEM	p-values		
	VIB	CONV	VIB+EI		TRT	Time	TRT × Time
Heart Rate during sessions, bpm							
Mean	108.3	109.8	108.6	3.87	0.96	0.47	0.51
Max	115.1	118.2	111.6	4.33	0.54	0.75	0.34
Min	99.6	102.8	102.5	3.80	0.82	0.12	0.32
Resting	92.6	94.3	94.0	2.63	0.89	0.01	0.22
HR Return to Resting, min¹	15.9	10.8	13.8	2.63	0.72	0.20	0.07

¹p-values obtained from log(10) transformed data, LS-means derived from untransformed data

Table 5: Session and d 5, 7, 9 Cortisol Measures. Sows were treated with Vibration-only (VIB, n=16), Conventional (CONV, n=18; 3 hand slaps), or Vibration and Electrical Impulse (VIB+EI, n=18) during a play back of a distress piglet call. Cortisol was measured at farrowing, before session-1 and-6, and after session-2 and-6. Cortisol was also measured at 0600h and 1800h on d 1, 4, 5, 7, and 9. Farrowing-measures were used as a covariate in all models.

	Treatment			SEM	p-values		
	VIB	CONV	VIB+EI		TRT	Time	TRT x Time
Farrowing ¹ , ng/mL	17.7	17.6	19.0	1.13	0.59	--	--
Session ² Response							
Before, ng/mL	20.1	17.9	17.7	1.60	0.51	0.87	0.11
After, ng/mL	20.0	18.0	20.4	2.03	0.68	0.44	0.24
Difference ³ , % Δ	-22.14 ^a	-88.29 ^{a,b}	-5.50 ^a	22.23	0.03	0.92	0.49
Days 4 5, 7, 9							
Morning, ng/mL	15.4	19.6	19.9	1.96	0.21	<0.01	0.20
Evening, ng/mL	16.2	17.5	18.3	2.27	0.80	0.74	0.83
Difference ⁴ , % Δ	10.8	11.9	11.2	1.7	0.89	0.03	0.50
Circadian Cortisol ⁵ , ng/mL	16.6	18.0	18.4	1.31	0.59	0.24	0.04
Mornings, ng/mL	17.4	20.5	20.2	1.60	0.34	<0.01	0.41
Evenings, ng/mL	17.7	17.9	17.5	1.83	0.98	0.81	0.32
Difference ⁴ , % Δ	-47.5	-32.7	-38.5	21.37	0.88	0.73	0.89
Area Under the Curve ⁶	221.1	217.6	224.7	13.87	0.94	--	--

^{a,b}LS means differ p<0.05; LS-means are in seconds, untransformed

¹Covariate for all other cortisol models; sample was collected after the third pig was born for each sow

²On days 1 and 4 ± 0.84 SD relative to farrowing; before sessions 1 and 10 min after sessions 2 and 6

³Δ After-sample subtracted by the before-sample then, divided by the after-sample and finally converted to percent

⁴Δ PM-sample subtracted by the AM-sample then, divided by the AM-sample and finally converted to percent

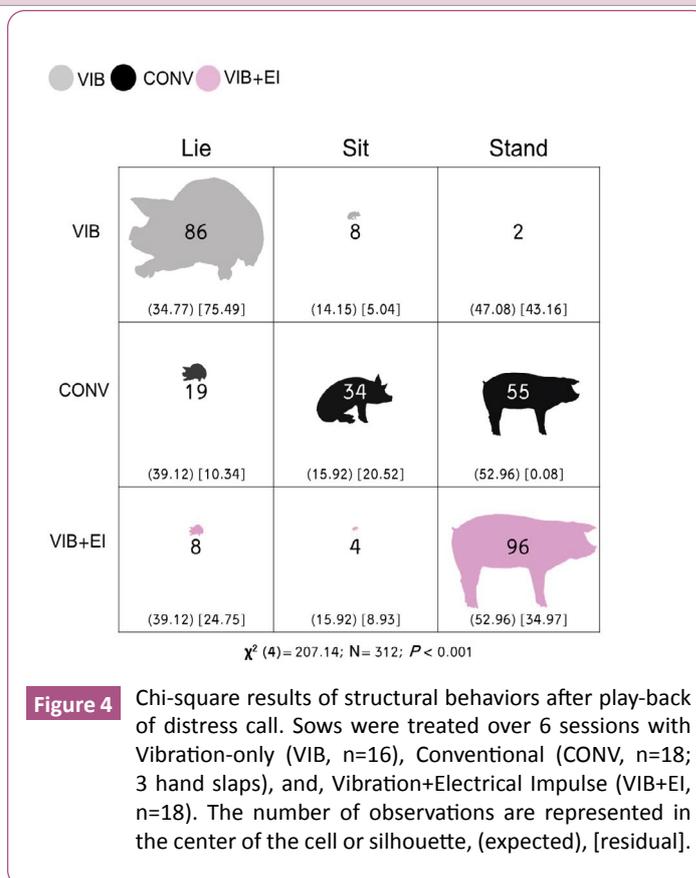
⁵All samples excluding the after-samples and farrowing-sample

⁶Area Under the curve was calculated with all the samples for each sow

to the sow, but erroneous handling increases generalized fear of humans.

Behavioral measures are more precise and accurate at evaluating stress responses than cortisol measures in the perinatal period. For the current project, the startle-index (**Figure 3**) quantified the severity of responses to the stimuli on a 0-100 scale. A startle-index of 0 indicated that the sow stayed in the lying position and remained silent during the session, whereas a startle-index of 100 represents a sow that jumped, grunted, barked, squealed, and bit during a session. There were no time or treatment by time interactions for startle index ($p>0.10$). Thus, the number of observations for single behaviors were also examined. Following the distress call playback, most VIB-sows remained in the lie-position (**Figure 4**) and did not vocalize (**Figure 5 and Table 6**), resulting in a low startle-index (**Figure 3**; $p<0.01$). This finding also confirmed research that sows are not responsive to piglet distress calls [33]. In addition, this indicates that the PAM-technology will not likely disturb neighboring sows versus those that are treated for piglet crushing. However, Chaple et al. [24] outlines a changing spectrum of sow response based on environmental noise and sow age, so more sow numbers are likely needed for definitive results.

Conventional methods included 3-hand slaps to the hind quarters of the sow. A significant portion of the CONV-sows only sat up after treatment (**Figure 4**), although most of them vocalized (**Figure 5 and Table 6**). Sitting is not a desired outcome because if the piglet is crushed by the hindquarters, it would not be freed if the sow simply sat up. A large proportion of CONV-sows barked



after the hand slaps (**Figure 5 and Table 6**). The sitting and bark response attributed a startle index that was 30% greater than VIB-

Table 6: Startle-response Vocalization and Bite. During play-back of a piglet distress, sows were treated with Vibration (VIB n=16), Conventional (CONV n=18), Vibration+Electrical Impulse (VIB+EI n=18) over 6 sessions. The number of observations are represented in the center, (expected), [residual].

	Silent		Grunt		Bark		Squeal		Bite	
VIB	89		4		1		2		0	
	(34.5)	[86.31]	(10.2)	[3.7]	(28.9)	[27.0]	(22.5)	[18.6]	(--)	(-)
CONV	9		21		60		17		1	
	(38.8)	[22.9]	(11.4)	[8.0]	(32.5)	[23.2]	(25.3)	[2.1]	(--)	(-)
VIB+EI	14		8		33		51		2	
	(38.8)	[15.8]	(11.4)	[1.0]	(32.5)	[0.0]	(25.3)	[30.4]	(--)	(-)

$\chi^2(6)=239.08, N=312, p \leq 0.05$

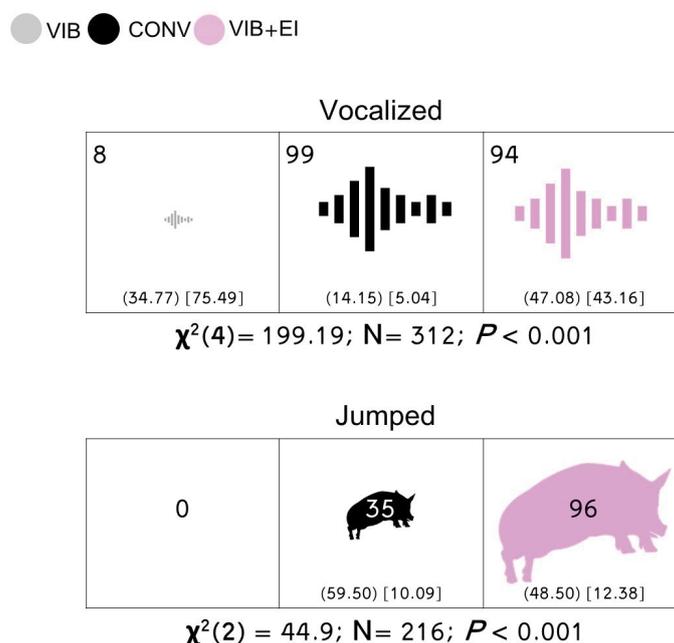


Figure 5 Chi-square of all vocalizations (top) and jumping (bottom) after play-back of distress call. Sows were treated with Vibration-only (VIB, n=16), Conventional (CONV, n=18; 3 hand slaps), and, Vibration+Electrical Impulse (VIB+EI, n=18). No VIB-sows jumped. Therefore the analysis was conducted for just CONV-vs. VIB+EI-sows. The number of observations are represented in the center of the cell or silhouette, (expected), [residual].

sows. A sitting response poses a challenge for conventional sows because the manager would need to use more force to make the sow stand completely, or risk piglet welfare, in the event a piglet crushed by a sow's hindquarters.

The PAM-technology's stimuli was more effective than CONV-methods; most VIB+EI sows were in the upright, standing position after each session (Figure 4). Nonetheless, VIB+EI-sows had a 1.5-fold greater startle index than CONV-sows ($p < 0.01$; Figure 3). The VIB+EI sow startle-indexes were in the stand-jump range (Figure 3), whereas, CONV-sows were in the sit-stand range. Biting was measured over concern that sows might retaliate against piglets after the electrical impulse. Biting was a rare occurrence for this experiment (Table 6).

Jumping is also undesired because the sow may injure herself or her piglets. This challenge may be overcome because PAM-

technologies can gather data from sensors and process the information for an individual animal [15,26]. Thus, each sow's primary response data could be used by the machine-learning software to adjust the electrical impulse during a subsequent crush-event.

For the current project, the authors noted that two multiparous, VIB-EI sows stood up before the stimulus was applied. This may indicate that these sows associate the distress call or vibration with the previous electrical impulse treatment. Future research is needed to determine the learning curve of each sow and chance-response percentage associated with PAM-technologies. On the other hand, 1 sow had a vocal response (squeal) but not a postural change during any of the VIB+EI sessions. Lameness can be found in up to 16% of sows [34], but the veterinarian did not observe clinical signs of sickness or injury. She was, however,

in the top 10 percentile body weight for all the sows in the project (mean BW 246.5 kg), weighing 266.4 kg. Sow size relative to the stall may have influenced motivation to respond with postural changes to the PAM-stimuli. Therefore, body weight was considered as a covariate in all models but removed due to lack of significance ($p>0.10$). A postural change to aversive stimuli indicates that the sow is using coping mechanisms to control her environment [33,35]. The non-standing response to a primary VIB+EI-stimuli may be used as a method to identify sow health and compromised psychological welfare.

The use of electrical impulse is a hotly debated topic and offers immediate and large pushback from the public in both companion and production animals [17,18]. By measuring the responses of sows treated with all 3 levels, direct comparisons can be made between treatments with the goal of eliminating the distress of a piglet which is being crushed. This does not excuse the fact that the sow is subject to the aversiveness of electric impulse. This impulse driven by the technology should be minimized and controlled per animal welfare councils worldwide [18]. In the instance of the current technology, the PAM-settings can be adjusted to stop any electrical impulses after three unsuccessful applications and

provide an alert for the animal caretaker. The ability of sows to learn from the vibration stimulus in a Pavlovian manner is not out of the questions [23]. Thus, mitigating the impulse in subsequent farrowing's is possible with the vibration of the device alone. However, affecting sow position with vibration alone may not be learned until subsequent lactations. Lactation is arguably the most important phase of swine production as numbers such as 13.9 pigs born per litter and mortality of 17.5% (Stalder, 2017), [36] have direct impact on the rest of the production system. Assuming sows can be trained to respect the vibration of PAM-technology and relieve a pig of crushing, mortality may decrease, but opportunity for training is somewhat sparse in production systems. At 2.3 litters per sow per year of 13.9 piglets [36], and an estimated mortality due to crushing alone of 20% (estimated by 80 % crushing of the 25% total mortality [2,9]), 2.78 piglets per litter are crushed. At 2.3 litters per sow per year, this gives each sow 6.4 estimated incidences of crushing per year which she would be subject to the PAM-technology. For the scope of this project, the impulse was under complete control by a remote in the hands of a researcher. Not identical to the product that will be marketed and available for producers. Future work is needed to

Table 7: Session-duration of coping and nursing behaviors (s per 20 min observation). Over six sessions, sows were treated with Vibration-only (VIB, n=16), Conventional (CONV, n=18; 3 hand slaps), or Vibration and Electrical Impulse (VIB+EI, n=18) during a play back of a distress piglet call (starting point).

	Treatment			SEM	TRT	p-values	
	VIB	CONV	VIB+EI			Time	TRT × Time
n	16	18	18	--	--	--	--
Sow weight							
Enrollment, kg	248.1	243.3	245.3	6.50	0.87	-	-
Day 21, kg	221.9	222.5	221.8	5.70	1.00	-	-
Difference¹, % Δ	10.4	9.2	9.4	0.87	0.58	-	-
ADI², kg	5.7	5.5	5.6	0.21	0.78	-	-
Sessions³, kg	4.0	4.1	4.3	0.21	0.57	<0.01	0.75
Days⁴ 5-9,kg	5.7	5.5	5.8	0.24	0.51	<0.01	0.47
Piglets							
Born⁵, no.	15	13	15	0.7	-	-	-
Weaned, no.	14	12	13	0.4	0.16	-	-
Piglet weight							
Birth⁶, kg	1.3	1.4	1.3	0.05	-	-	-
Day 7, kg	2.5	2.8	2.7	0.10	0.26	-	-
Difference, % Δ	48.9	49.4	49.6	1.43	0.95		
Day 21	5.3 ^c	5.8 ^d	5.8 ^d	0.19	0.10	-	-
Difference⁷, % Δ	52.5	52.3	53.9	0.90	0.39	-	-
Overall Difference, % Δ	75.8	75.9	76.9	0.65	0.46	-	-
Total litter weaning weight	71.2	72.3	71.6	3.11	0.97	-	-
Total Plasma Protein⁸, % Δ	71.7	71.7	78.0	15.14	0.84	-	-

^{c,d}tend to differ $p=0.10$

Δ Later value, subtracted by the initial value, then divided by the initial value and finally converted to percent

¹Enrollment to d 21 (weaning)

²Average Daily Feed Intake (ADFI) from enrollment until day 21

³ADFI from enrollment until the six session on d 4 relative to farrowing (d 0 ± 0.84 SD)

⁴ADFI from d 5 to 9 relative to farrowing (d 0 ± 0.84 SD)

⁵Piglets directly born from dam; Some cross-fostering caused sows to gain piglet(s)

⁶Birth weight was used as a covariate for all piglet weight models and the Total Plasma Protein model

⁷d 7 to 21 relative to farrowing (d 0 ± 0.84 SD)

⁸Percent change in total plasma protein (TPP) was calculated by subtracting day 0 TPP (total plasma protein) from day 7 TPP

analyze responses of sows to the commercially available product in large production systems.

Coping responses

In addition to evaluating the acute stress response, the authors examined cortisol circadian function from the morning and evening samples throughout the experiment [30,31]. Circadian cortisol revealed few differences between the cortisol response of sows treated with VIB-, CONV-, or VIB+EI-stimulus over the total experimental timeline (**Table 5**). A treatment by time interaction was observed for circadian cortisol measures (**Supplementary Figure 2**; $p < 0.05$). Sows among each treatment had varied cortisol concentrations. But there was no indication of significant differences for treatment within day (Tukey's adjustment LS-means $p > 0.10$). The performance axiom was also considered because deviations in feed intake and bodyweight maintenance may be indicators of chronic stress [37]. For this experiment, no treatment or treatment by time interactions were observed for feed intake ($p > 0.10$; **Table 7**). In addition, sow body weight and return-to-estrus rates did not detect differences between treatments ($p > 0.10$; **Table 7**).

Sows in farrowing stalls are least limited in oral behaviors. Non-Nutritive Oral Behaviors (NNOB) can be viewed as exploratory and coping behaviors that are stereotypically observed in sows in many housing environments [35]. Coping behaviors can become abnormally expressed (too much or too little). Therefore NNOB provide a direct measurement of animal welfare. For this project, desired behaviors included NNOB directed at the floor, stall and feeder because these are precursors to nutritive behaviors such as eat and drink [38]. Undesired behaviors were those NNOB directed at the piglets, over concern that this potentially leads to savaging [39]. The observation time in the present study was not long enough to determine if NNOB behaviors should be defined as stereotypic. Thus, the only negative NNOB behavior was piglet directed. In addition to duration of these behaviors, latency can provide insight into desired behaviors. The authors considered a short latency to perform NNOB desired because the opposite of this behavior indicates a freezing or fear response [40]. Sows spent more time performing NNOB behaviors directed at floor and stall, as well as a difference in the total amount of NNOB behavior ($p < 0.05$, **Figure 6 and Table 8**).

These findings in conjunction with the startle-response confirm that the stimuli cause an acute behavioral response which garners more activity of the sow immediately after treatment. This activity, however, should be taken cautiously as its benefit or detriment to sow welfare is yet to be defined. The same observation protocol was applied on d 5, 7, and 9 relative to farrowing to determine if treatment differences existed after sessions. There were not any behavioral differences among treatments on d 5, 7, and 9 ($p > 0.10$; **Supplementary Tables 1 and 2**). The authors also considered the behaviors throughout the day, therefore, a wearable device that tracks any head movement (correlated with NNOB) and an accelerometer that detects sow standing was applied (Supplementary materials). The continuous data were analyzed for 20 min, 60 min and 20 h after sessions

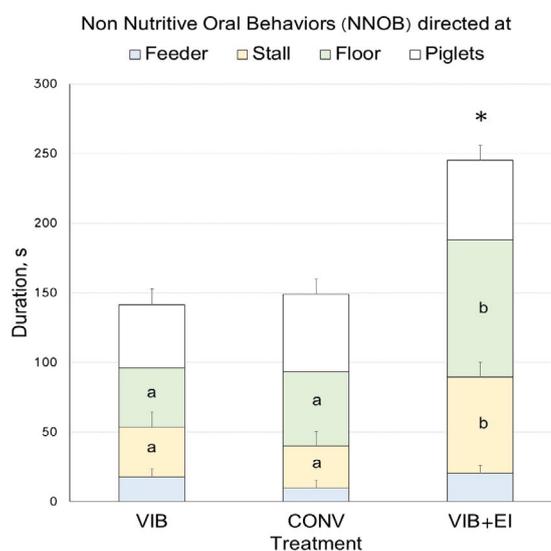


Figure 6 Coping behavior duration after sessions. After play-back of a piglet distress call, sows were treated with Vibration-only (VIB, n=16), Conventional (CONV, n=18; 3 hand slaps), and, vibration +electrical Impulse (VIB+EI, n=18). Twenty-minute videos were analyzed after each session. a,b LS means within behavior differ ($p < 0.05$). *LS means for total non-nutritive oral behaviors differ (sum of stack bars; $p < 0.05$).

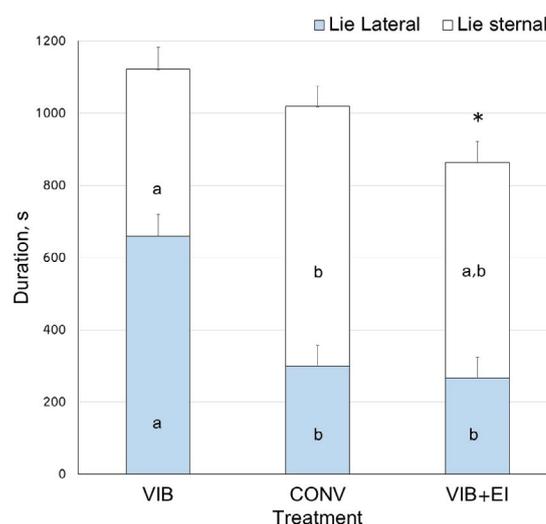


Figure 7 Rest behavior duration after sessions. After play-back of a piglet distress call, sows were treated with Vibration-only (VIB, n=16), Conventional (CONV, n=18; 3 hand slaps), and, Vibration+ Electrical Impulse (VIB+EI, n=18). Twenty-minute videos were analyzed after each session. a,b LS means within behavior differ ($p < 0.05$). *LS means for total lie differ (sum of stack bars; $p < 0.05$).

and days 5, 7, and 9. Only the 20-minute interval for the head-movement was significant ($p < 0.05$; **Supplementary Table 3**), which matched the video observations for NNOB. No additional conclusions could be gathered from these results (**Figure 7**).

Given the limitation of confinement among sows in farrowing stalls, these increased NNOB behaviors may result in sows eating more feed, but more definitive results with larger sow numbers are needed. After the sessions, sows in VIB+EI and CONV treatments had a shorter latency to eat than the VIB treated sows ($p < 0.05$; **Figure 8 and Table 9**). Postpartum, the sow requires more monitoring of oral behaviors because lactation requires high amounts of nutrient intake [41]. Sows in farrowing stalls often display anorexia and lose conditioning if they are not closely monitored [33,42]. Hence, the authors suggest that any oral behaviors related to water and feed intake may benefit the sow during farrowing. Feed intake was not different among treatments in the current experiment (**Table 7**). These authors suspect that the NNOB-coping behaviors may translate into increased feed intake if the technology were used on a larger sample size.

Nursing quality

Stressors during lactation are a known cause of unsuccessful nursing and increased morbidity in piglets [43]. A main concern over the PAM-stimuli is that it may negatively impact nursing behaviors and subsequently influence the piglets. Therefore, nursing behaviors after the treatment sessions were evaluated as well as on d 5, 7 and 9 relative to farrowing. After treatment

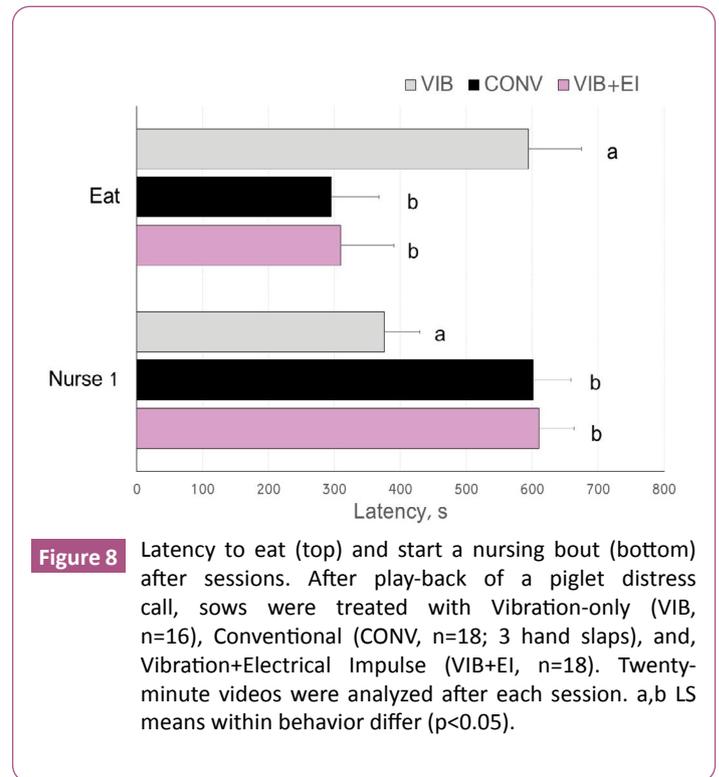


Figure 8 Latency to eat (top) and start a nursing bout (bottom) after sessions. After play-back of a piglet distress call, sows were treated with Vibration-only (VIB, n=16), Conventional (CONV, n=18; 3 hand slaps), and, Vibration+Electrical Impulse (VIB+EI, n=18). Twenty-minute videos were analyzed after each session. a,b LS means within behavior differ ($p < 0.05$).

Table 8: Session-duration of coping and nursing behaviors (s per 20 min observation). Over 6 sessions, sows were treated with Vibration-only (VIB, n=16), Conventional (CONV, n=18; 3 hand slaps), or Vibration and Electrical Impulse (VIB+EI, n=18) during a play back of a distress piglet call (starting point).

	Treatment			SEM	p-values ¹		
	VIB	CONV	VIB+EI		TRT	Time	TRT × Time
n	16	18	18	--	--	--	--
Headstill	982.7 ^a	950.9 ^a	781.2 ^b	29.20	<0.01	0.03	0.64
Oral behaviors ²	217.3 ^a	249.1 ^a	418.8 ^b	29.20	<0.01	0.03	0.64
NNOB ^{3,4}	141.9 ^a	149.4 ^a	247.3 ^b	20.80	<0.01	0.33	0.84
Floor	42.6 ^a	53.3 ^a	98.4 ^b	61.60	0.01	0.06	0.95
Stall	35.6 ^a	30.3 ^a	68.9 ^b	10.68	0.03	0.09	0.26
Feeder	17.8	9.7	20.7	5.60	0.24	0.42	0.58
Piglets	45.3	55.7	57.3	11.04	0.78	0.01	0.99
Nutritive	75.4	100.3	173.1	18.80	0.23	0.13	0.62
Eat	56.0	47.3	64.1	9.08	0.36	0.34	0.19
Drink	18.8	52.8	108.5	15.64	0.81	0.63	0.83
Upright	77.9 ^a	182.0 ^b	335.3 ^c	30.40	0.01	0.01	0.66
Sit	39.8	67.5	76.5	16.52	0.73	0.28	0.14
Stand	38.4	112.6	256.9	32.40	0.12	0.47	1.00
Lie	1122.1 ^a	1017.9 ^b	862.4 ^b	30.40	<0.01	0.06	0.49
Sternal ⁴	463.8 ^a	718.9 ^b	597.2 ^a	58.00	0.01	0.31	0.38
Lateral ⁴	658.8 ^a	299.8 ^b	267.1 ^b	58.80	<0.01	0.77	0.08
Nursing ⁵	462.0	279.4	257.8	53.20	0.09	0.04	0.28
¹ piglet	175.1 ^a	91.7 ^b	75.4 ^b	22.36	<0.01	0.05	0.64
5+piglets	287.1	186.6	174.62	34.80	0.09	0.33	0.22

^{a,b}LS means differ $p < 0.05$; LS-means are in seconds, untransformed

¹Log-transformed P-values unless otherwise noted

²Data fit a normal distribution and were not transformed

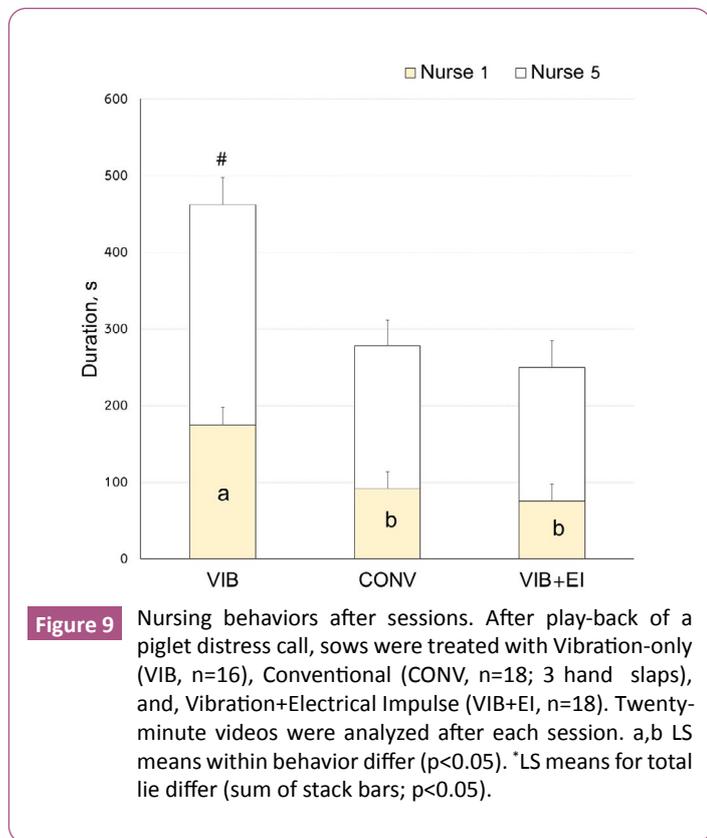
³Non-nutritive behaviors directed at any object

⁴Data were analyzed using the square root transformation to better fit normality

⁵Nursing ¹piglet was scored when ≥ 1 but ≤ 4 piglets suckling. Nursing 5+piglets was noted when the sow had ≥ 5 piglets suckling

session, VIB-sows had the least latency to start nursing, while CONV-and VIB+EI sows had similar latencies to start nursing ($p=0.01$; **Table 8 and Figure 8**). Likewise, duration for nursing at

least 1 piglet was greatest among VIB-sows ($p<0.05$; **Figure 9**). This finding was not surprising because most VIB-sows remained in a resting position during the administration of stimuli. This difference was not observed after blood was sampled from ear veins on in days 5,7,9 relative to farrowing (Treatment x Time and Treatment $p>0.10$; **Supplementary Table 1**). All sows spent more time nursing 5 or more piglets on d 7 compared to d 5 and 9 relative to farrowing ($p<0.05$; **Supplementary Table 1**). The authors suspect that this difference was due either to the increased human-time in the barn for piglet bodyweight and blood collection, or to a common observation that the number of nursing-bouts decrease each day after farrowing [44].



In addition to measuring nursing behaviors, Total Plasma Protein (TPP) and piglet performance measures were assessed. Total plasma protein is an indirect measure of IgG that is acquired from colostrum. In addition, colostrum quality and intake decrease piglet’s risk of mortality and enteritis, and increases weight gain [45-47]. For this experiment, there were no differences in percent change of TPP among treatments, ($p>0.10$; **Table 9**). Piglet performance numbers (**Table 9**) were consistent with the standard numbers for commercial systems in the midwestern United States [48]. No differences in piglet performance, mortality, and morbidity were observed in this experiment ($p>0.10$; **Table 9**). More data are needed at the commercial level to determine if PAM-technology will influence piglet performance. Nonetheless, nursing outcomes on the same days sows were treated with stimuli were the same in CONV-sows as VIB+EI sows.

Table 9: Session-Latency of Coping and Nursing Behaviors (s per 20 min observation). Over 6 sessions, sows were treated with vibration-only (VIB, n=16), conventional (CONV, n=18; 3 hand slaps), or vibration and electrical Impulse (VIB+EI, n=18) during a play back of a distress piglet call (starting point). If the behavior was not observed, latency could not be analyzed.

	Treatment			SEM	TRT	p-values ¹	
	VIB	CONV	VIB+EI			Time	TRT x Time
n	16	18	18	--			
Any oral behavior ²	102.4	70.3	54.0	15.37	0.10	0.02	0.88
Any NNOB ³	113.9	74.1	65.7	84.53	0.47	<0.01	0.70
Floor	215.3	202.7	167.7	27.47	0.88	0.52	0.47
Stall	263.1	146.6	167.4	32.23	0.27	<0.01	0.26
Feeder	316.7	270.9	332.9	53.61	0.40	0.81	0.29
Piglets	245.1	222.0	188.1	30.40	0.12	0.03	0.43
Any nutritive	248.7	319.2	269.9	40.03	0.10	0.01	0.09
Eat	574.0 ^a	295.2 ^b	302.0 ^b	69.50	0.03	0.56	0.47
Drink ²	246.6	351.5	369.3	40.03	0.13	0.84	0.34
Lie after sit or stand							
Sternal ²	150.5	119.0	153.8	46.20	0.80	0.48	0.63
Lateral ²	513.9	774.0	636.7	79.17	0.12	0.41	0.63
Nursing ⁴							
1 piglet	375.7 ^a	601.1 ^b	609.8 ^b	58.63	0.01	0.35	0.53
5+piglets ⁵	574.1	652.9	656.6	68.07	0.61	0.06	0.35

^{a,b}LS-means differ $p<0.05$; LS-means are in seconds, untransformed

¹Log-transformed p -values unless otherwise noted

²Data were analyzed using the natural log transformation to better fit normality

³Non-nutritive behaviors directed at any object

⁴Nursing 1 piglet was scored when ≥ 1 but ≤ 4 piglets suckling. Nursing 5+piglets was noted when the sow had ≥ 5 piglets suckling

Conclusion and Implications

Pre-weaning mortality varies between 8% and 25% in systems using farrowing stalls [2,9,36] with up to 70%-80% of those losses due to crushing. A logical mitigation of these losses beyond current practices can be PAM. The VIB+EI stimulation was the most effective at motivating the sows to stand, although some did so with a more startled response. However, the average response was still below 60% on the startle scale. If accelerometers are used to detect jumping, this added input into the PAM-technology could be used to further adjust impulse levels based on individual sow responses. This is in contrast to conventional methods, where the human-to-sow ratio in a commercial system reduces the likelihood of treating sows on an individual basis. For this experiment, coping and nursing behaviors were influenced just after treatment sessions. The main difference observed between CONV and VIB+EI sows was that the PAM-stimuli increased NNOB after treatment sessions. Producers may observe increased feeding behaviors because NNOB may transgress into significant increased feed intake in the first few days after farrowing, when sows appear least motivated to eat. However, a concern is that NNOB-can be abnormally expressed. For this experiment, the changes in NNOB were observed in the days following the last treatment session.

In the US, over 80% of swine producers currently use the standard farrowing stall [6]. Apart from the farrowing stall only, methods to prevent crushing included sloped floors, solid sloped walls, and supplemental heating to motivate piglets to spend non-nursing time away from the sow. This PAM-technology may greatly decrease the crushing rate in addition to these housing modifications. Using PAM in place of humans to mitigate crushing may be beneficial to long-term wellbeing of sows because they are treated at the individual animal level. Nonetheless, a more

accepted animal welfare improvement for the sow would be a housing system that does not restrict her movement. This PAM-technology has the potential to mitigate piglet crushing in a pen-system, rather than the farrowing stall system. Pen-systems were examined to increase space-allowance and add non-nutritive substrates to promote NNOB during farrowing and lactation-period. However, crushing rate was over 2 times greater among sows in pens with substrate than sows in traditional farrowing stalls.

When open-barn housing was evaluated, crushing rate among open-housed sows was also over 2 times greater than farrowing stalls. The creators of this PAM-technology have seriously considered the technology for sows that are not restricted by movement. They found that the current limitation is that every housing system of the less than 20% of housing systems differs greatly among systems. The PAM-technology would need to be enhanced on a case-by-case basis, which currently is not feasible for one company with limited resources. Therefore, research (and funding for research) investigating the behavioral responses in pen-housed sows needs consideration to create a homogenous, effective system at the pre-competitive level.

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