The Old School, Brewhouse Hill, Wheathampstead,
Hertfordshire AL4 8AN, UK
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Use of meloxicam, buprenorphine, and Maxilene® to assess a multimodal approach for piglet pain management, part 2: tail-docking

AV Viscardi*† and PV Turner†‡

† Department of Pathobiology, University of Guelph, Guelph, ON, N1G 2W1, Canada
‡ Charles River Laboratories Inc, 251 Ballardvale St, Wilmington, MA 01887, USA
* Contact for correspondence: aviscardi@vet.k-state.edu

Abstract

Piglets on commercial pig farms are often tail-docked to reduce the incidence of tail-biting. While this is a painful procedure, piglets are often not provided analgesia or anaesthesia for pain relief. The objectives of this study were to assess a multimodal approach to managing tail-docking pain in piglets, using 0.4 mg kg^{-1} meloxicam (MEL), 0.04 mg kg^{-1} buprenorphine (BUP), and Maxilene® (MAX), a topical anaesthetic. The effectiveness of each drug and drug combination was evaluated using behavioural indicators, vocalisation, and facial grimace analysis. This study also assessed whether male and female piglets responded differently to pain or pain treatments. Piglets were randomly assigned to one of six possible treatments: MEL, BUP, MEL + BUP, MEL + BUP + MAX, no treatment (tail-docked control), or sham (non-tail-docked control). Vocalisations were recorded at initial handling, injection, and tail-docking. Piglets administered MEL + BUP and BUP demonstrated significantly fewer pain behaviours than piglets in the MEL and no treatment group. MEL + BUP + MAX and BUP piglets displayed significantly lower facial grimace scores than piglets in the no treatment group. There were no significant differences in emitted vocalisations between the analgesia-treated piglets and the no treatment group and both injection and tail-docking elicited piglet vocalisations of similar frequency, power, and energy. There were no significant differences in behaviour, facial grimacing or emitted vocalisations between male and female piglets. All treatment groups with buprenorphine were able to alleviate tail-docking-associated pain, suggesting that opioid administration is highly effective for managing piglet pain.

Keywords: *analgesia, animal welfare, multimodal, pain assessment, piglet, tail-docking*

Introduction

Piglets are commonly tail-docked on commercial farms in North America and the EU to minimise tail-biting (Sutherland *et al* 2008). This procedure is known to cause pain, based on behavioural changes and physiologic measures, including an increase in tail wagging, tail jamming (tucking the tail stump between the hind legs), increased blood cortisol levels, and high-frequency vocalisations (Noonan *et al* 1994; Sutherland *et al* 2008; Torrey *et al* 2009). Analgesia is not given routinely to alleviate pain; however, countries, such as Canada, are increasingly requiring analgesia administration to piglets prior to tail-docking in their animal care guidelines (National Farm Animal Care Council [NFACC] 2014). There is limited research regarding effective pain mitigation strategies for piglets, post-procedure (Sutherland 2015). Non-steroidal anti-inflammatory drugs (NSAIDs), such as meloxicam, when administered alone, have been unsuccessful in reducing post-surgical pain behaviours caused by tail-docking (Herskin *et al* 2016). Injecting a local anaesthetic into the base of the tail or applying a topical anaesthetic to the tail-docked wound were also insufficient in alleviating piglet pain post-procedure (Sutherland *et al* 2011). Buprenorphine was found to be effective at reducing surgical castration pain in piglets without causing any obvious side-effects (Viscardi & Turner 2018a). The analgesic capacity of buprenorphine to mitigate tail-docking pain alone, or in combination with an NSAID, has not been assessed. Multimodal analgesia is commonly used to alleviate post-operative pain in veterinary clinical practice, when tail-docking is carried out on dogs to maintain specific breed standards (Hewson *et al* 2006).

Sex-related differences in pain and analgesia sensitivity have been reported in mice, rats and humans (Mogil *et al* 2000; Craft 2003; Fillingim *et al* 2009). Females have largely been found to have greater sensitivity to procedural and post-operative pain (Fillingim *et al* 2009). Studies examining differences in pain and analgesia sensitivity between male and female piglets after tail-docking, beyond the immediate pain response, have been limited (Rutherford *et al* 2009). Understanding potential sex-related differences is important for proper administration of pain treatments and maintenance of good animal welfare on-farm.

The objectives of this study were to assess the efficacy of 0.4 mg kg^{-1} meloxicam, 0.04 mg kg^{-1} buprenorphine, and topical Maxilene® to manage tail-docking pain in piglets. The effectiveness of each drug and drug combination was evaluated using behavioural indicators, vocalisation, and facial grimace analysis (using the piglet grimace scale). This study also assessed whether male and female piglets responded differently to pain and analgesic therapies. Based on the results from a previous study by this research group (Viscardi & Turner 2018a), we hypothesised that piglets receiving buprenorphine alone, or as part of a drug combination, would have a significant reduction in pain behaviour and facial grimacing after tail-docking and would emit lower frequency vocalisations at the time of tail-docking compared to other non-opioid treatments. Finally, we hypothesised that female piglets would demonstrate more pain behaviours and facial grimacing when in pain compared to male piglets (Traub & Ji 2013).

Materials and methods

Ethics statement

All animal use and procedures were approved by the University of Guelph Animal Care Committee (Animal Utilization Protocol #3350). The institution is registered under the Animals for Research Act of Ontario and holds a Good Animal Practice certificate issued by the Canadian Council on Animal Care.

Study animals and treatments

This study was conducted at Arkell Swine Research Station (Arkell, ON, Canada), an active research facility supported by the University of Guelph and the Ontario Ministry of Agriculture, Food and Rural Affairs (OMAFRA). Yorkshire-Landrace × Duroc male and female piglets (n = 165, four days old, mean [± SEM] BW = 1.87 [± 0.03] kg) from 14 litters were used. The floor space available to the piglets was 1.8 × 2.4 m (length × width) and the farrowing crate holding the sow was 0.8 × 2.3 m. Farrowing rooms were maintained at ambient air temperature (23 [± 0.5]ºC) with lights on/off at 0700/2100h, and natural light was provided by windows in each room. Sows were fed *ad libitum* beginning four days after farrowing. The creep areas for piglets were heated to approximately 30–35ºC by means of a heat pad or lamp. Cross-fostering of piglets did occur on-farm when necessary; however, only litters of piglets remaining with their biological sow were selected for this study.

Within each litter, piglets were randomly assigned to one of six possible treatments: 0.4 mg kg^{-1} meloxicam, 0.04 mg kg^{-1} buprenorphine, 0.4 mg kg^{-1} meloxicam + 0.04 mg kg^{-1} buprenorphine, 0.4 mg kg^{-1} meloxicam + 0.04 mg kg^{-1} buprenorphine + Maxilene®, no treatment (tail-docked control), or sham (non-tail-docked control) (n = 15 male piglets and n = 15 female piglets per treatment group, except the sham group, which contained eight male piglets and seven female piglets). Group size was based on a sample size estimate, using $\alpha = 0.05$, population $\sigma = 0.1$ (determined from a pilot study) and 5% precision (Suresh & Chandrashekara 2012; Viscardi *et al* 2017). Meloxicam (MEL) (Metacam 20 mg ml^{-1}; Boehringer Ingelheim Ltd, Burlington, ON, Canada) was administered as an intramuscular (IM) injection at the label dose of 0.4 mg kg^{-1}. Buprenorphine (BUP) (Vetergesic 0.3 mg ml^{-1}; Champion Alstoe Animal Health Inc, Whitby, ON, Canada; extra-label use) was also administered IM at 0.04 mg kg^{-1} (Flecknell 2015). 1.0 g Maxilene® (MAX) (Maxilene® 4% lidocaine; RGR Pharma Ltd, Windsor, ON, Canada; extra-label use) was applied topically to the entire tail before docking using a swab. The no treatment group was tail-docked without the administration of an analgesic or topical anaesthetic. The sham treatment group underwent a simulated tail-docking without topical application or injection. The treatment groups were identified by a unique symbol ('H', 'V', 'X', square, triangle or circle) marked on the piglet's forehead and back with a black permanent marker for males and a red permanent marker for females prior to tail-docking. This was to ensure that individuals scoring post-procedure behaviours and facial grimacing were blinded as to animal treatment. Numbers were also written on the back leg of piglets for individual animal identification.

Processing procedures

Twenty-four hours prior to the trial, piglets were weighed and marked with the symbol that corresponded to their treatment group. On the day of tail-docking, all piglets were removed from their pen and placed in a transport cart. Treatments were administered and, 20 min later, piglets were tail-docked using side-pliers before being returned to their home pen. Approximately half of the piglet's tail was removed. All procedures occurred between 0800 and 1000h and were done by one individual (AVV). Handling and technical procedures were conducted by female researchers only, to eliminate the potential confound of increased stress and an altered pain response in piglets exposed to male researchers, as has been reported in mice (Sorge *et al* 2014). Piglets in this study were not ear notched, teeth clipped, given an iron injection or castrated previously and were therefore, naïve to painful procedures.

Behaviour recording and scoring

High definition video cameras (JVC GZ-E200 full HD Everio Camcorder, Yokohama, Japan) were placed on tripods outside each farrowing pen. Piglets were video-recorded pre-procedure for 1 h, immediately post-tail docking for 8 h, and then again for 1 h at 24 h post-procedure (ie, a total of 10 h of video data were collected from each litter of pigs). Videos were randomised across litters and time-points using a random number generator (random.org). The behaviour of each piglet was scored continuously for the first 15 min of each hour of data collected by six trained individuals using Observer XT (Version 12.0, Noldus Information Technology, Wageningen, The Netherlands) and a detailed ethogram adapted from Hay *et al* (2003) (Table 1). Observers were blinded as to time-point, pen, and piglet treatment in this study; however, they could observe which piglets had

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been tail-docked as well as differentiate piglet sex. Four observers scored one and a half pens, one observer scored one pen, and one observer scored seven pens. Inter-observer reliability was assessed three times during the behaviour scoring period (once monthly) by having all participants score the same piglet in a video and calculating the intra-class correlation coefficient (ICC). All inter-observer reliability tests produced an ICC above 0.9, indicating excellent correlation between scorers. A total of 24,750 min (412.5 h) of behaviour recordings were scored and analysed.

Piglet behaviours were assessed individually and then grouped into active, inactive and pain categories, to analyse the activity level of piglets across the observation period and the total proportion of pain behaviours displayed. Active behaviours and postures included running, walking, playing, nosing, suckling, chewing, sitting, and standing. Inactive behaviours and postures included sleeping, awake inactive, and lying. Sitting was placed in the active category, as most piglets assumed this posture when suckling or scratching the rump (both considered active behaviours). Pain behaviours included stiffness, spasms, trembling, tail wagging, and rump scratching (Hay *et al* 2003).

Piglet grimace scale scoring

Still images of piglet faces were captured from the first 30 min of every hour of video data collected by an individual blinded as to piglet litter, treatment, and time-point. Videos were uploaded to the Everio MediaBrowser 4 program (Pixela Corporation, Osaka, Japan) and whenever a piglet face was in view, the video was paused, and the still image was collected (excluding times when piglets were lying with their head down or sleeping). An attempt was made to take one facial image of each piglet per time-point. A total of 674 images were captured (Table 2). Prior to scoring, facial images were uploaded to Photoshop (Adobe Systems Incorporated, San Jose, CA, USA) and the symbol marked on each piglet's forehead was blurred to ensure volunteer scorers were not biased by these markings. Faces were then randomised by treatment, litter, and time-point prior to scoring using a random number generator (random.org).

Three individuals were trained to use the piglet grimace scale (PGS) (Viscardi *et al* 2017) prior to scoring in an interactive 30-min session. The PGS score was calculated for each image by summing the scores given to each of the facial action units (ear position, cheek tightening/nose bulge, and orbital tightening). The final PGS score of each piglet per time-point was calculated as a mean of the scores from the three individuals. If more than one image had been pulled from the same piglet at the same time-point, the PGS scores were averaged across images prior to analysis to produce one score per piglet per time-point and avoid pseudo-replication.

Table 1 Ethogram used to score piglet behaviour, grouped into feeding, locomotion, non-specific behaviours, pain-related behaviours, posture, and social cohesion (adapted from Hay *et al* 2003).

Behaviours	Description
Suckling	Teat in mouth and suckling movements
Nosing udder	Nose in contact with udder, up and down head movements
Playing	Springing, bouncy movements with littermates
Agonistic	Biting or fighting other littermates
Walking	Moving forward at a normal pace
Running	Trot or gallop
Awake inactive	No special activity, but awake
Sleeping	Lying down, eyes closed
Nosing	Snout in contact with a substrate
Chewing	Nibbling at littermates or substrates
Trembling	Shivering, as with cold
Spasms	Quick and involuntary contractions of the muscles
Scratching	Rubbing the rump against the floor, pen walls, or littermates
Tail wagging	Tail's movements from side-to-side (or up and down)
Stiffness	Lying with extended and tensed legs
Lying	Bodyweight supported by side or belly
Sitting	Bodyweight supported by hindquarters and front legs
Standing	Bodyweight supported by four legs
Kneeling	Bodyweight supported by front carpal joints and hind legs
Isolated	Alone or with one littermate at most, distance of 40 cm separates the animal(s) from the closest group of littermates
Desynchronised	Activity different from that of most littermates (at least 75%)

Vocalisations

Vocalisations of each piglet were measured at three points during the study: at initial handling when they were marked with a symbol (marking); when they received their treatment injection (injection); and when they were tail-docked (tail-docking). A video camera on a tripod was placed as close to the focal piglet's face as possible to record each procedure-induced vocalisation. Audio clips from the recorded videos were analysed using Raven Pro 1.5 (Cornell Lab of Ornithology, Ithaca, NY, USA) by three individuals blinded as to procedure and piglet treatment. From the spectrograms, maximum frequency (Hz), maximum amplitude (μ), maximum power (dB) and energy (dB) of each call was determined (Marx *et al* 2003).

Table 2 Total number of piglet faces captured for piglet grimace scale scoring.

	Treatment												
Time-point (h)	1		2		3		4		5		6		Total
	M	F	M	F	M	F	M	F	M	F	M	F	
pre	3	9	5	6	6	3	7	1	3	4	1	2	**50**
0	10	10	14	8	10	5	12	9	12	11	4	2	**107**
1	2	5	12	11	11	3	10	7	3	4	2	0	**70**
2	3	5	11	6	9	8	7	12	5	1	2	1	**70**
3	6	6	9	7	7	5	10	8	7	5	2	1	**73**
4	5	4	9	4	10	4	5	10	2	8	1	1	**63**
5	8	4	12	4	6	3	8	5	5	5	2	1	**63**
6	4	2	7	3	4	7	10	6	0	6	2	1	**52**
7	1	1	11	3	4	3	4	7	2	2	3	1	**42**
24	8	4	13	9	8	10	12	9	4	5	1	1	**84**
	50	**50**	**103**	**61**	**75**	**51**	**85**	**74**	**43**	**51**	**20**	**11**	**674**

M: male piglets, F: female piglets;
[1] Meloxicam;
[2] Buprenorphine;
[3] Meloxicam + Buprenorphine;
[4] Meloxicam + Buprenorphine + Maxilene® ;
[5] No Treatment;
[6] Sham.

Statistical analysis

The total duration of behaviours was converted into proportion of time piglets engaged in each behaviour prior to analysis (to account for periods of time when the piglet was out of view and could not be scored). Normality was evaluated using the univariate procedure in SAS (Statistical Analysis System 9.4, SAS Institute Inc, NC, USA). Data were analysed using a GLIMMIX procedure with a beta distribution, including treatment, time, litter, sex, treatment × time, time × sex, and treatment × sex interactions. Litter was included as a random effect and time was a repeated measure with piglet as the experimental unit. *Post hoc* tests were conducted using the Tukey-Kramer adjustment. Statistical significance was set at $P < 0.05$.

The grimace scale scores were analysed using a mixed procedure model, including litter, time, treatment, sex, time × treatment, and the treatment × sex interaction. Litter was included as a random effect and time was a repeated measure with piglet as the experimental unit. A *post hoc* Tukey's test was conducted for significant outcomes.

Vocalisation data were analysed using a mixed procedure, including litter, treatment, and procedure in the model. Litter was included as a random effect and piglet was the experimental unit. Significant outcomes were further analysed using a *post hoc* Tukey's test. Behaviour, PGS, and vocalisation data were used to assess each treatment's effectiveness in reducing tail-docking pain and to determine if male and female piglets responded differently to pain and pain treatments.

Results

Behavioural observations

Prior to tail-docking, male piglets spent significantly more time sitting than female piglets and female piglets engaged in more agonistic behaviours ($P = 0.003$ and $P = 0.04$, respectively). After tail-docking, there was no significant difference in behaviour between male and female piglets. Due to this, treatment groups were combined across sexes post-procedure.

There were no significant behavioural differences between any of the treatment groups pre-tail-docking ($P > 0.05$) (Table 3). Four individual behaviours and two grouped behaviours had significant treatment effects after tail-docking: lying ($P < 0.0001$), standing ($P < 0.0001$), tail wagging ($P < 0.0001$), walking ($P < 0.0001$), active ($P < 0.0001$), and pain ($P = 0.0002$) (Table 4). MEL and no treatment piglets displayed significantly more pain behaviours than piglets in the MEL + BUP, BUP, and sham treatment groups ($P < 0.05$) (Figure 1). Female piglets administered BUP demonstrated significantly fewer pain behaviours across the observation period than

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Table 3 Proportion of time piglets were engaged in specific behaviours (n = 165) pre- and post-treatment across all litters and time-points. Values represent the proportional means (± SEM).

Behaviour[1]	Pre-treatment		Post-treatment					
	Treatment *P*-value	Pre-treatment	Treatment *P*-value	Time *P*-value	Sex *P*-value	Time × Treatment *P*-value	Sex × Time *P*-value	Sex × Treatment *P*-value
Awake inactive	0.8612	0.46 (± 0.05)	0.2625	< 0.0001	0.4065	0.2298	0.8384	0.8938
Lying	0.9294	0.69 (± 0.07)	< 0.0001	< 0.0001	0.5485	0.2050	0.1331	0.3647
Nosing udder	0.3624	0.18 (± 0.05)	0.9400	0.0315	0.6081	0.6259	0.9664	0.4857
Sleeping	0.5793	0.50 (± 0.05)	0.0662	0.0004	0.2116	0.5548	0.5676	0.6074
Standing	0.5319	0.25 (± 0.05)	< 0.0001	< 0.0001	0.4255	0.3460	0.2503	0.4999
Tail wagging	0.0928	0.01 (± 0.00)	< 0.0001	0.0007	0.2536	0.8309	0.0364	0.8377
Walking	0.2142	0.07 (± 0.02)	< 0.0001	< 0.0001	0.5127	0.0626	0.0988	0.3775
Sitting	0.1828	0.04 (± 0.00)	0.1008	0.0607	0.3603	0.6446	0.0384	0.4388
Spasms	0.9306	0.00 (± 0.00)	0.9339	0.3460	0.2684	0.2076	0.3769	0.0121
Playing	0.2518	0.02 (± 0.00)	0.8993	0.0441	0.6888	0.6958	0.8662	0.6207
Active[2]	0.3767	0.29 (± 0.06)	< 0.0001	< 0.0001	0.5573	0.2949	0.1697	0.3522
Pain[3]	0.0832	0.01 (± 0.00)	0.0050	0.9547	0.2956	0.0239	0.0693	0.0413

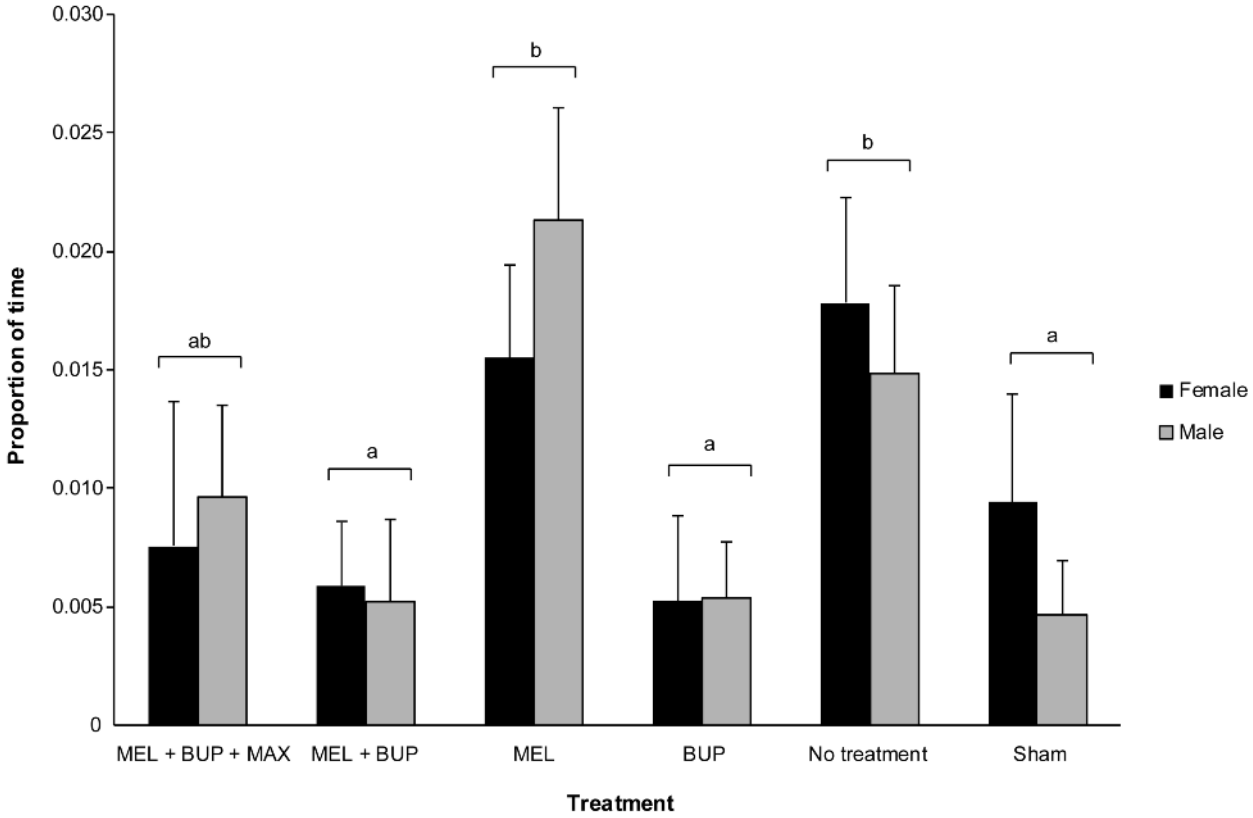
[1] Only significant behaviour variables are presented;
[2] Active behaviours include: nosing, suckling, walking, chewing, playing, running;
[3] Pain behaviours include: stiffness, trembling, spasms, tail wagging and rump scratching.

Table 4 Proportion of time piglets were engaged in specific behaviours (n = 165) across all litters and time-points. Values presented represent the proportional means (± SEM).

Behaviour[†]	Treatment (post-tail docking)						*F*-value	*P*-value
	1	2	3	4	5	6		
Lying	0.64 (± 0.03)[b]	0.51 (± 0.03)[a]	0.51 (± 0.03)[a]	0.53 (± 0.03)[a]	0.65 (± 0.03)[b]	0.69 (± 0.04)[b]	12.68	< 0.0001
Standing	0.33 (± 0.02)[b]	0.48 (± 0.03)[a]	0.46 (± 0.03)[a]	0.47 (± 0.03)[a]	0.32 (± 0.02)[b]	0.30 (± 0.04)[b]	14.36	< 0.0001
Tail wagging	0.02 (± 0.00)[b]	0.00 (± 0.00)[a]	0.00 (± 0.00)[a]	0.00 (± 0.00)[ab]	0.01 (± 0.00)[ab]	0.00 (± 0.00)[a]	5.93	< 0.0001
Walking	0.08 (± 0.01)[b]	0.14 (± 0.02)[a]	0.13 (± 0.02)[a]	0.14 (± 0.02)[a]	0.07 (± 0.01)[b]	0.06 (± 0.01)[b]	20.45	< 0.0001
Active[‡]	0.36 (± 0.03)[b]	0.49 (± 0.03)[a]	0.49 (± 0.03)[a]	0.47 (± 0.03)[a]	0.35 (± 0.03)[b]	0.30 (± 0.04)[b]	12.43	< 0.0001
Pain[§]	0.02 (± 0.00)[b]	0.00 (± 0.00)[a]	0.01 (± 0.00)[a]	0.00 (± 0.00)[ab]	0.02 (± 0.00)[b]	0.00 (± 0.00)[a]	4.88	0.0002

[1] Meloxicam;
[2] Buprenorphine;
[3] Meloxicam + Buprenorphine;
[4] Meloxicam + Buprenorphine + Maxilene® ;
[5] No Treatment;
[6] Sham;
[†] Only significant behaviour variables are presented;
[‡] Active behaviours include: nosing, suckling, walking, chewing, playing, running;
[§] Pain behaviours include: stiffness, trembling, spasms, tail wagging and rump scratching;
[a,b] Values within a row with different superscripts differ significantly at $P < 0.05$.

Figure 1



Mean (± SEM) proportion of time piglets demonstrated pain-related behaviours (spasms, rump scratching, tail wagging, trembling, stiffness) after tail-docking across treatment group. MEL = 0.4 mg kg^{-1} meloxicam, BUP = 0.04 mg kg^{-1} buprenorphine, and MAX = Maxilene® (n = 15 piglets per sex per treatment group, except sham: n = 8 male and n = 7 female piglets). Observers (n = 6) were unaware of piglet treatment, litter, and time-point when scoring. Different superscripts indicate significant differences between treatment groups ($P < 0.05$).

both male and female piglets in the MEL and tail-docked control groups ($P < 0.05$). There were no other treatment × sex differences found. There was also no effect of time on the amount of pain behaviours displayed. Piglets in the MEL, no treatment, and sham groups spent significantly more time lying and less time standing, walking, and engaged in fewer active behaviours than piglets in all other treatment groups ($P < 0.05$) (Figure 2). There were no sex differences in activity level ($P > 0.05$). MEL piglets wagged their tails significantly more than MEL + BUP, BUP, and sham piglets ($P < 0.05$).

Regardless of treatment, all piglets spent significantly less time lying and more time standing, walking and engaged in active behaviours at 0 h compared to all other time-points ($P < 0.05$). All piglets also spent significantly less time lying and sleeping and more time standing at 1 vs 2 h, 4 to 24 h ($P < 0.05$).

Piglet grimace scale

There were no significant time or time × treatment interactions found for PGS score ($P = 0.30$ and $P = 0.57$, respectively). There was a significant treatment effect ($P = 0.002$) (Figure 3). Male and female piglets in the no treatment group grimaced significantly more than MEL + BUP + MAX, BUP, and sham piglets ($P = 0.002$, 0.02 and 0.02, respectively). There was a trend for MEL + BUP piglets of both sexes to grimace less than no treatment piglets ($P = 0.08$). Similarly, there was a trend for male piglets to grimace more than female piglets, irrespective of treatment ($P = 0.064$).

Vocalisation

There were significant procedure × treatment effects on the frequency, power, and energy of piglet vocalisations ($P < 0.0001$, $P = 0.0009$ and $P < 0.0001$, respectively) (Figure 4). All tail-docked piglets, regardless of treatment group produced significantly higher vocalisations than the sham treatment group during tail-docking ($P < 0.05$). There were no treatment differences in vocalisations during marking and injection ($P > 0.05$). Tail-docking and injection produced vocalisations of significantly higher frequency, energy, and power than marking ($P < 0.0001$). Injection and tail-docking produced similar vocalisations. There were no sex differences in emitted vocalisations ($P > 0.05$).

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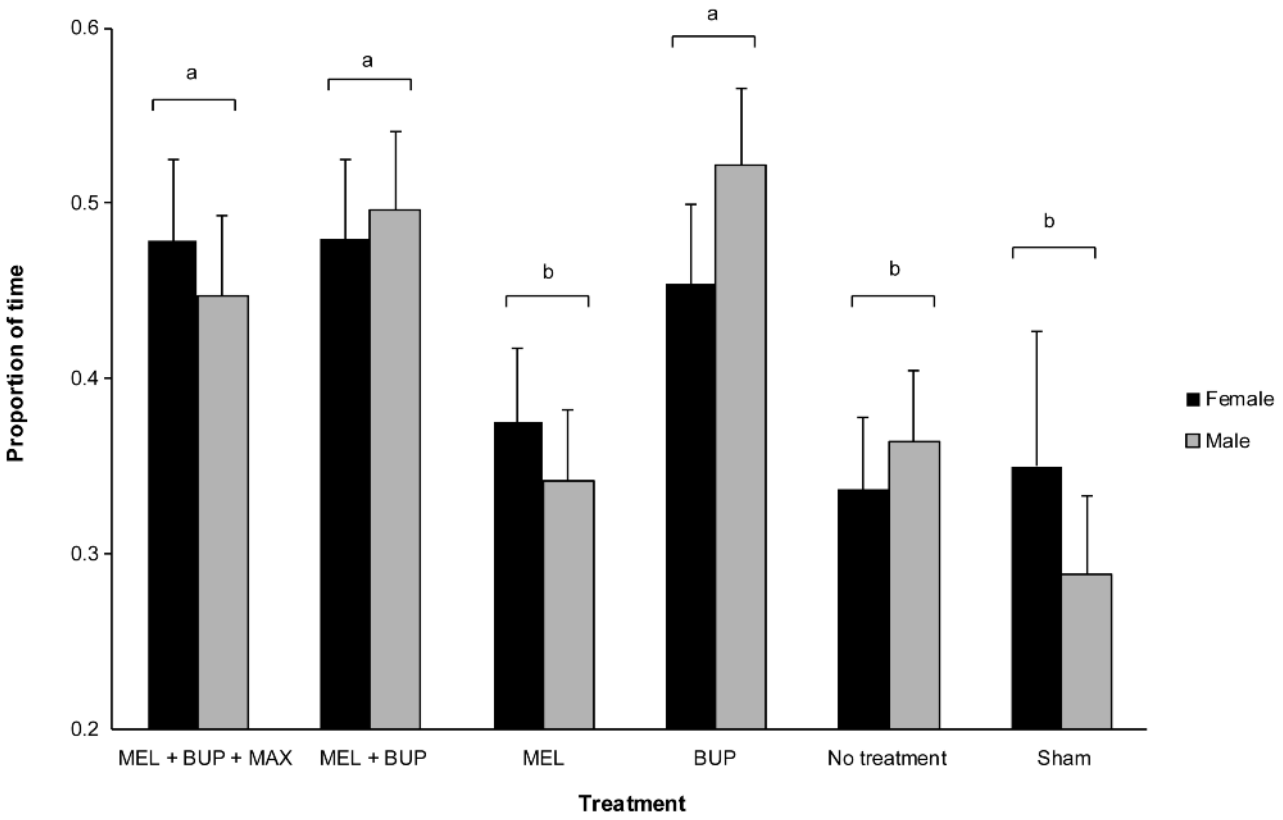
Figure 2



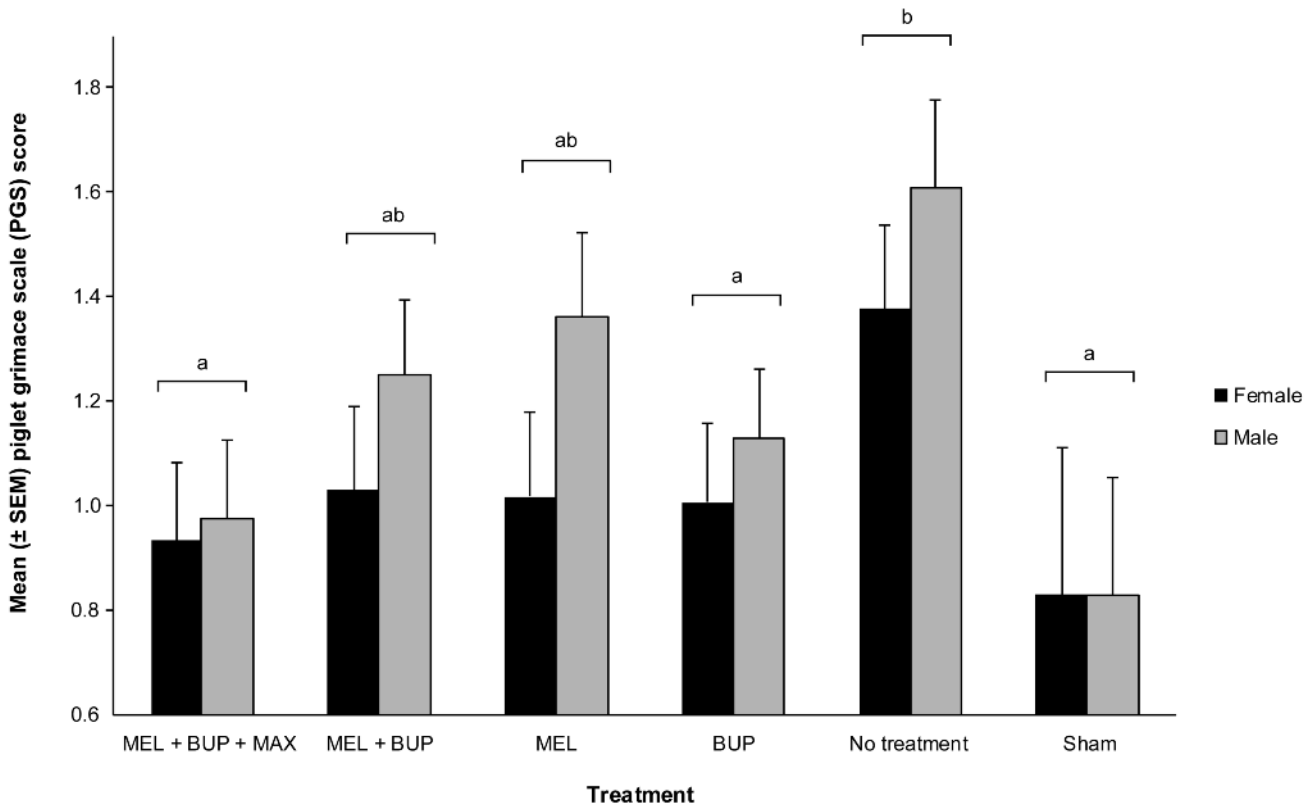
Mean (± SEM) proportion of time piglets engaged in active behaviours (playing, walking, suckling, nosing, chewing and running) after tail-docking across treatment groups. MEL = 0.4 mg kg^{-1} meloxicam, BUP = 0.04 mg kg^{-1} buprenorphine, and MAX = Maxilene® (n = 15 piglets per sex per treatment group, except sham: n = 8 male and n = 7 female piglets). Observers (n = 6) were unaware of piglet treatment, litter, and time-point when scoring. Different superscripts indicate significant differences between treatment groups ($P < 0.05$). There were no significant differences found between male and female piglets within the same treatment group ($P > 0.05$).

Discussion

This study examined several approaches to mitigate tail-docking pain in male and female piglets. Buprenorphine, when administered alone as a single IM injection, was the only treatment to significantly reduce both facial grimacing and piglet pain behaviours. Buprenorphine has previously been shown to alleviate pain in piglets and growing swine without causing any adverse effects (Rodriguez *et al* 2001; Meijer *et al* 2015; Viscardi & Turner 2018a). All piglets that were tail-docked and administered buprenorphine (ie, MEL + BUP + MAX, MEL + BUP, and BUP alone) were significantly more active than the MEL, no treatment, and sham groups, further supporting its efficacy, as animals will often show a decrease in activity level when in pain (Berger & Eeg 2006) and reinforcing the lack of sedative side-effects at this dose. While there were significant benefits to buprenorphine administration found in this study, its use on-farm is largely impractical at this time. Opioids, such as buprenorphine, are controlled drugs with high human abuse potential. Farmers and farm workers are at greater risk of opioid dependence in the US, and widespread uncontrolled distribution of this drug for use on-farm could potentially have devastating consequences (United States Department of Agriculture [USDA] 2018). Currently, its use in pigs and other food animals is strictly prohibited and piglets administered buprenorphine in this study were not allowed to enter into the food chain (Food and Drug Administration [FDA] 2014). The positive results in this study that were found using buprenorphine to alleviate pain may encourage future work looking at how we can make this a practical option for use on-farm (eg, through novel formulation or drug compounding that limits abuse potential).

Increased piglet activity was observed immediately after tail-docking (at 0 h). This was likely due to stress from piglet handling, injection, short-term separation from the sow or pain from the procedure. In a previous study examining piglet pain behaviour after surgical castration, we found a significant increase in pain behaviour displayed by saline-castrated piglets at 24 h post-procedure (Viscardi & Turner 2018a). In the current tail-docking study, there was no significant time effect on pain behaviour, and by comparison, male piglets tail-docked without analgesia displayed significantly fewer pain behaviours 24 h post-procedure than castrated piglets without analgesia at the

Figure 3



Mean (± SEM) piglet grimace scale (PGS) scores in each treatment group after tail-docking. Higher PGS scores indicate increased pain expression. MEL = 0.4 mg kg^{-1} meloxicam, BUP = 0.04 mg kg^{-1} buprenorphine, and MAX = Maxilene® (n = 15 piglets per sex per treatment group, except sham: n = 8 male and n = 7 female piglets). Observers (n = 3) were unaware of piglet treatment, litter and time-point when scoring. Different superscripts indicate significant differences between treatment groups ($P < 0.05$).

same time-point (0.04 [± 0.01] vs 0.26 [± 0.04]). This suggests that surgical castration is a more painful procedure for piglets to undergo than tail-docking. Tail wagging and tail jamming, key tail-docking-related pain behaviours, were difficult to assess after docking and this may have also contributed to the reduction in observed pain behaviours (Noonan *et al* 1994).

Facial grimace analysis is a recent technique that has been developed to assess pain in animals. Species-specific scales have been developed for many animals, including mice, horses, and piglets (Langford *et al* 2010; Costa *et al* 2014; Viscardi *et al* 2017). Grimace scales involve identifying and quantifying facial features (or facial action units) that change in response to pain. PGS results corresponded well to overall pain behaviour results in this study. This is consistent with previous piglet pain studies where the PGS was used (Viscardi & Turner 2018a,b). However, the PGS results have yet to correspond perfectly with observed pain behaviours of piglets. This is important for validation of the PGS as a tool for pain assessment. The ability of swine producers, technical caregivers or veterinarians working with swine to accurately use the PGS to score piglet facial expressions has not been assessed. This should be evaluated in a future study to determine its true on-farm applicability.

Piglets emit distinct vocalisations associated with tail-docking that have been attributed to pain (Marchant-Forde *et al* 2009; Torrey *et al* 2009). None of the treatments in this study reduced the frequency, amplitude, power or energy of these vocalisations at the time of tail-docking. Piglets that had Maxilene® applied to the tail were expected to vocalise less, but this was not observed. Perhaps waiting the recommended 30 min (instead of 20 min in this study) or wrapping the tail after application of the topical might have enhanced the local numbing effect of Maxilene® (Eichenfield *et al* 2002). However, a topical agent is unlikely to provide sufficient analgesia for tail removal. IM injections elicited a similar vocal response as the tail-docking procedure, suggesting it also caused acute pain, although likely of very short duration. Small, sharp needles (25 G) were used for injections in this study and discarded after one use, to eliminate unnecessary pain caused by blunt-needle injection. As two of the four drug combinations evaluated required two injections (meloxicam and buprenorphine), and the addition of meloxicam and Maxilene® did not provide piglets any significant benefit beyond buprenorphine administered alone, a single IM injection of buprenorphine is recommended to alleviate tail-docking pain. The longer handling time required for

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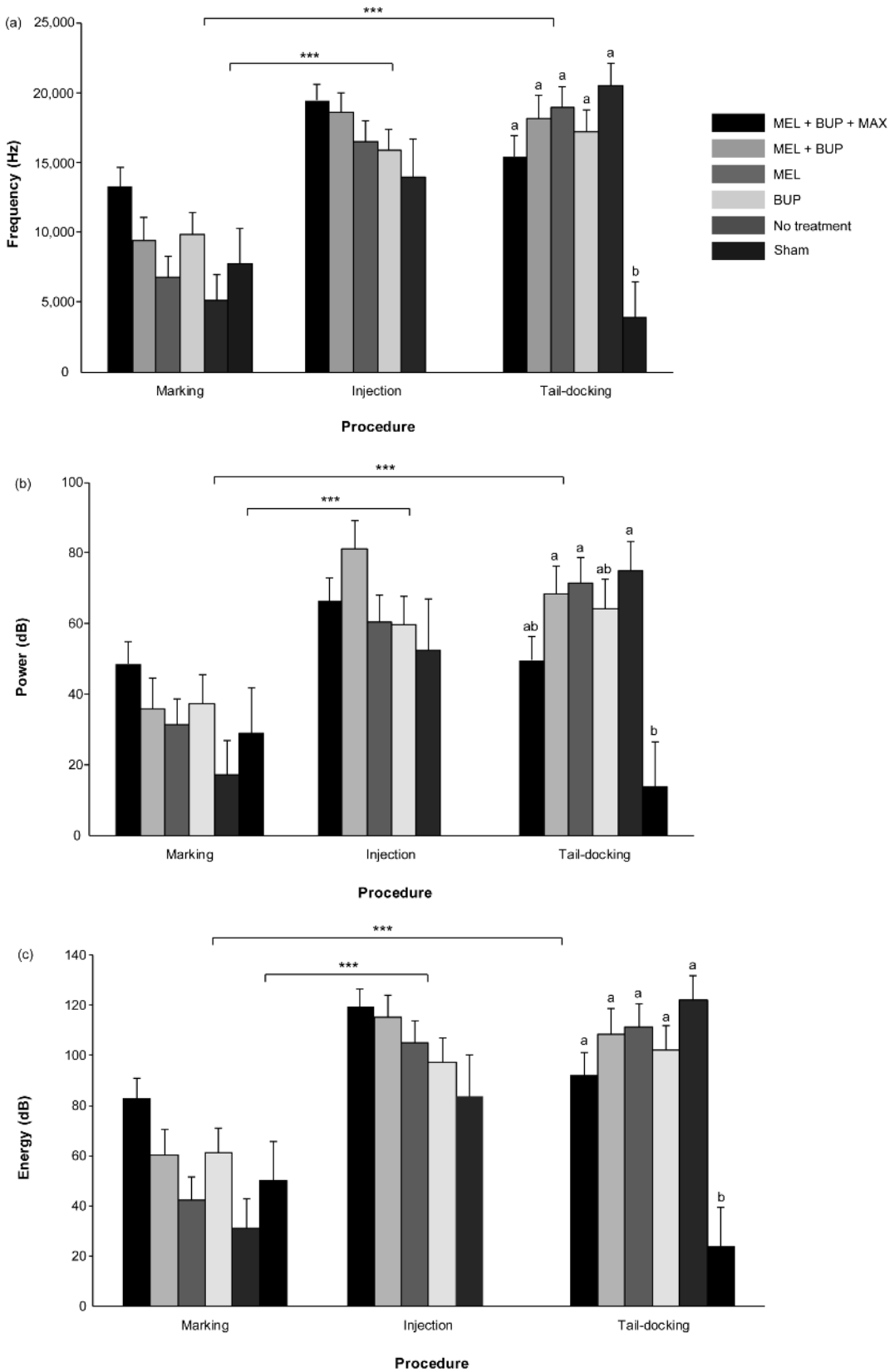
Figure 4



Mean (± SEM) (a) frequency (Hz), (b) power (dB), and (c) energy (dB) of vocalisations emitted by piglets during marking, injection and tail-docking. MEL = 0.4 mg kg^{-1} meloxicam, BUP = 0.04 mg kg^{-1} buprenorphine, and MAX = Maxilene® (n = 30 piglets per treatment group, except sham: n = 15 piglets). Individuals (n = 3) scoring data were unaware of piglet treatment, litter, and procedure when analysing vocalisation measurements. Different superscripts show significance between treatment groups ($P < 0.05$). Asterisks indicate significant differences between procedures ($P < 0.0001$).

multiple drug administration was also likely to have contributed to increased piglet stress (Marchant-Forde *et al* 2009) and is less practical in a production setting.

Sex differences in pain and sensitivity to analgesia have been reported in rodents and humans (Mogil *et al* 2000; Greenspan *et al* 2007; Fillingim *et al* 2009). It is generally accepted that females have a higher sensitivity to pain than males (Goffaux *et al* 2011). Previous work found that sex was not a factor affecting nociceptive thresholds in piglets (Janczak *et al* 2012). No differences were seen in the proportion of pain behaviours displayed, PGS scores or vocalisations between male and female piglets. Gonadal hormones (eg oestradiol, testosterone) and oestrogen in females are functionally important for the differences observed in pain sensitivity and analgesia (Craft *et al* 2004; Sarajari & Oblinger 2010). As the animals in this study were sexually immature, this may explain why no sex differences were observed in pain expression.

Tail-docking, or removal of the distal portion of the tail, is often performed using side-pliers, a scalpel blade, scissors, or electrical cautery iron to decrease tail-biting on-farm (Sutherland *et al* 2008, 2015). Tail-biting is a common behavioural issue observed in commercial swine, likely caused by stress, boredom or frustration. Improving the housing environment by providing enrichment materials for pigs to chew and manipulate, decreasing stocking density, and ensuring both ambient temperature and good ventilation significantly reduces the incidence of tail-biting (Telkänranta *et al* 2014). These adjustments not only provide benefits to the emotional and physical well-being of the pigs, but also eliminate the need to tail-dock, which is an ideal outcome. While tail-docking itself does not stop pigs from tail-biting, it does reduce its prevalence and is the most practical solution to this problem with current North American swine management systems (Hunter *et al* 2001; Sutherland *et al* 2009). This study confirmed that tail-docking causes acute pain in piglets. It has been suggested that tail-docked pigs experience chronic pain (Di Giminiani *et al* 2017), and this is an area of future work.

In conclusion, buprenorphine was effective at alleviating tail-docking pain in piglets. PGS results corresponded well to piglet pain behaviours and may have utility as an adjunctive pain assessment tool. None of the treatments evaluated reduced pain-related vocalisations at the time of tail-docking. Male and female piglets in this study responded to painful procedures and analgesic drugs similarly. Future work should focus on making buprenorphine a practical drug to administer on-farm, as it has proven efficacy in alleviating both tail-docking and castration pain in piglets.

Animal welfare implications

Tail-docking is a routine, and painful, procedure for piglets raised in commercial production systems. Analgesia administration is required in animal care guidelines for countries in the EU and Canada for post-operative pain relief. Some EU countries have banned the tail-docking procedure and raise pigs with intact tails; however, this is unlikely to occur in North America at this time. Identifying an analgesic drug (or drug combination) that is most effective at alleviating tail-docking pain is important for appropriate recommendations to be made to pig producers, and to maximally improve animal welfare on-farm, a topic of increasing societal concern.

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