Title: Translactational analgesia to reduce pain during piglet castration - NPB #:12-063

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Institution: Prairie Swine Centre

Date Submitted: July 19, 2013

Industry Summary: Commercially reared piglets receive a number of routine management procedures including castration, tail docking, teeth clipping, and vaccination. Castration of pigs is done primarily to avoid boar taint, an objectionable flavor found in the meat of mature males. However, due to the pain caused by this procedure and the fact that analgesics are not commonly used, surgical castration of piglets is a welfare concern. The pork industry is responding to this concern by seeking effective ways of providing pain relief at castration, or alternative methods for reducing boar taint. This study investigated a novel approach for providing analgesia to piglets, in which the analgesic drug, Metacam®, was administered to sows, and obtained indirectly by piglets through the sow’s milk. Our main objectives were to determine how much of the drug was transferred to piglets, the time course of drug absorption and elimination, and whether or not this treatment would reduce pain responses after castration. Administration of the drug to the piglets through the sow’s milk could eliminate the need to handle the piglets twice at castration and decrease the labor requirements of injecting each piglet. When sows were injected with 0.5, 0.75 or 1.0 mg/kg Metacam®, drug levels in milk were found to increase over time, peaking at approximately 215 ng/ml at 3 h following injection. Unfortunately only a small amount of drug was transferred to piglets via the milk, with maximum levels of less than 3 ng/ml observed over a 5 h period. This drug concentration is only a fraction (less than 1/100th) of the amount achieved typically by administration through intramuscular injection, and is not believed to be sufficient to provide effective pain relief. However, use of an alternative drug or higher drug dosages and longer wait times may provide a better result. Increasing the dose of drug administered to the sow would increase the amount of drug available to the piglets in the sow’s milk, but this approach is limited due to potential effects on sow health and cost of the drug. Overall, this study showed limited success in transferring the analgesic, Metacam®, through the sow’s milk to piglets: the drug was successfully transferred to piglets, but the rate of transfer was slow and the quantity transferred was low. Other analgesic drugs such as Ketoprofen® could be tried using a similar approach, as this method has the potential to significantly reduce labor requirements for handling and injecting piglets, as well as reducing stress in piglets by minimizing handling.

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Keywords: piglets, castration, pain, translactational analgesia, Metacam
Scientific Abstract:
The cost of drugs and the additional labor required to administer analgesics to individual piglets reduces the likelihood of their adoption by producers when conducting painful procedures. This study examined the feasibility of a novel approach for providing analgesia at castration, using the sow as the vehicle to deliver an analgesic to the entire litter. The work was completed in two parts. The first studied the kinetics of drug absorption and distribution. Twelve sows were injected with 0.5, 0.75 or 1.0 mg/kg of Metacam® at 7 days after parturition and serial blood and milk samples were collected over a 5 hour period. Study sows were high producing animals (12 or more live piglets per litter) in their 3rd or 4th parity. Drug levels in plasma and milk were measured by LC-MS. Metacam® concentrations in sow plasma peaked at 1 h post injection, averaging 468 ±136, 438 ±143 and 501 ±338 ng/ml for the 0.5, 0.75 and 1.0 mg/kg treatments, respectively. The average drug concentration in milk across treatments peaked at 215 ng/ml at approximately 3 h post injection, with concentrations of 105 ±18, 352 ±467 and 184 ±92 ng/ml, respectively, for the three treatments. A second study was performed to determine the amount of drug absorbed by piglets at nursing. Twelve sows were injected with 1.0 mg/kg of Metacam®. Over 4.5 hours, serial milk samples were collected from each sow and serial blood samples were collected from 2 male piglets per litter. Analgesic concentrations in piglet serum were found to increase over time, from 0.44 ng/ml at 45 minutes, to 2.65 ng/ml at 295 minutes. Piglet serum levels were thus less that 1/100th of the levels in sow serum. Based on these results it was determined that translactational transfer of Metacam® is unlikely to be sufficient to provide therapeutic pain relief following castration. However, a modified form of the drug, or alternative drugs such as Ketoprofen®, may be more readily transferred via the milk. While this method has the potential to significantly reduce labor requirements for handling and injecting piglets, the large amount of drug required in sows to provide adequate transfer to piglets remains problematic due to toxicity and cost concerns. It is possible that, with the use of alternative drugs, this technique could become a viable approach for the hog industry.

Introduction:
Surgical castration of neonatal piglets in North America is performed to eliminate boar taint as well as reduce aggression and undesirable sexual behavior. This procedure, however, is typically performed without the use of pain relief and thus raises concerns regarding animal well being. The provision of analgesics to reduce pain following castration is costly in terms of drug requirements and added labor time to handle and inject each piglet. Administering analgesics can provide pain relief but has no benefit to piglet growth (ADG) or mortality. Due to the added costs and limited benefits of this practice, analgesic use has not been widely adopted by producers. It is important that the swine industry address this problem by identifying convenient and cost-effective methods of providing analgesia at castration, or alternative ways for reducing boar taint. This study focused on determining whether translactational delivery of Metacam® to piglets via the sow’s milk can provide adequate analgesia to mitigate pain responses following castration.

Behavioral measures have shown that the pain from castration lasts up to four days after the procedure in piglets (Hay et al, 2003). This has lead to the need for post-operative analgesia to reduce the pain associated with castration. Lidocaine local anesthetic has shown efficacy in reducing pain-related behavior and the physiological response to castration (McGlone and Hellman, 1988; Haga and Ranheim, 2005; and Kluivers-Poodt et al, 2012) but requires the piglet to be handled twice because lidocaine takes approximately three minutes to reach its highest concentration in the spermatic cord (Ranheim et al, 2005). The added labour of individually treating each piglet decreases the feasibility of this method in a commercial operation. General anesthesia methods have been studied, most notably with carbon dioxide, but do not provide post-operative analgesia (Van Beriendonck et al, 2011) and the stress during recovery lessens the benefits of its use (Sutherland et al, 2012). The topical anesthetics Cetacaine and Tri-Solfen and the NSAIDs Aspirin and Butorphanol have proven ineffective at reducing pain-related behaviors (Sutherland et al, 2010; McGlone et al, 1993) from piglet castration and are therefore not a viable option. The NSAID Metacam, however, has had much success in reducing both pain-related behaviours and physiological changes due to castration in piglets when injected intramuscularly. Specifically, Metacam® has been shown to reduce cortisol and ACTH concentration spikes (Keita et al, 2010), decrease the amount of pain-related behavior post-operatively such as huddling, spasms, prostration, and scratching of the rump and decrease levels of serum Amyloid A (indicative of inflammation, trauma or stress; Hansson et al, 2011) following castration. Also, piglets treated with Metacam® spent more time suckling post-operatively and exhibit less pain-related behaviors in the six hours post-castration (Schmidt et al, 2012). Intramuscular injection of Metacam, however, presents the same laborious issue as the local anesthetic Lidocaine. To mitigate this problem, this study investigated the
feasibility of translactational delivery of Metacam.

Using the sow as a delivery vehicle for analgesia eliminates the need to handle piglets twice during castration - i.e. once to administer the analgesic, and once to perform the procedure. The transfer of drugs through the milk has been the subject of many studies in humans, for obvious health reasons, and has shown that the process is complicated. Factors that are relevant to the absorption and distribution of these compounds include the nature of the milk, the nature of the drug, the time in lactation and the maturity of the neonate. Factors affecting drug deposition in the milk include pH, fat content and protein content of the milk. Drug characteristics influencing excretion in milk include ionization, protein binding, molecular weight and lipophilicity. Studies have suggested that factors such as low protein binding, low molecular weight and high lipophilicity may increase the concentration of drugs in milk, but there are many other factors involved (Ito and Lee, 2003). At this time only limited information is available on the constituents of sow milk compared to the information available for humans and cows. Metacam has been detected in bovine milk (Boeringher Ingelheim Datasheet, 2005) so it is likely that it will pass through sow’s milk to piglets.

**Objectives:**
The primary objective of this project was to investigate a technique for providing an analgesic simultaneously to an entire litter of piglets via the sow’s milk. Specifically, we injected sows (after parturition) with Metacam to determine if the analgesic is transferred via the sow’s milk into the piglets. Providing practical methods to reduce pain and suffering of animals will aid the swine industry to meet increasing demands for assuring animal well-being at all stages of production.

The aim of this research was to conduct a pilot study investigating the feasibility of using the sow as a vehicle to deliver painkillers to the entire litter prior to processing (e.g. castration). The work would be carried out in two parts. Specifically, we will;

1. Inject sows with Metacam at 3 and 7 days following parturition, and by serial analysis of blood and milk samples determine if the analgesic is transferred via the sow’s milk to piglets via oral route.

2. If the drug is successfully transferred, we will measure the amount of drug received and determine whether beneficial effects are observed, based on behavioural and physiological measures in piglets following castration.

In addition to these objectives, we performed initial validation studies for a novel behavioural method for assessing piglet pain following castration using a handling chute. The test was developed to aid in exploring objective 2, however studies were not completed due to the limited drug transfer. It is hoped that the chute can be used in future studies as a means to evaluate and compare the effectiveness of different analgesic treatments following castration.

**Materials & Methods:**

**Animals**

*Pharmacokinetics and translactational transfer of Metacam (Studies 1 & 2):* A total of 15 sows and 6 piglets were studied over 2 experiments. All animals were of Yorkshire x Landrace genetics (Camborough Cross, PIC Canada, Winnipeg, MB). The sows and their piglets were housed at the Prairie Swine Centre’s Floral research facility, in Saskatoon, Saskatchewan.

*Pain assessment following castration (Study 3):* A total of 32 piglets from six litters were selected. Piglets were randomly allocated to one of two treatment groups at four days of age, prior to training: sham castration (“S”, n=16) or castration (“C”, n=16). Piglets were ear notched within 24 hours of birth. At four days of age piglets were randomly assigned to treatment groups, with each treatment represented by at least one piglet per litter. Each piglet was individually weighed at four and five days of age and marked with a unique series of lines along their back and sides using sharpie for individual identification. Piglets were housed with their dams and littermates in standard farrowing crates (dimensions: 177 cm wide x 250 cm long).

All trials were approved by the University of Saskatchewan’s Animal Research and Ethics Board, and adhered to the Canadian Council on Animal Care guidelines for humane animal use.

*Study 1: Pharmacokinetics of Metacam*

In the first trial, 12 sows were allocated to three treatments based on drug dosage (0.5, 0.75 and 1.0 mg/kg)
at seven days following parturition. Metacam® (Boeringher Ingelheim, Burlington, Ontario) was administered as a single injection in the neck region of each sow. Eight blood samples (5 ml per sample) were collected from each sow using an ear vein jugular catheter at 0, 20, 40 and 60 minutes post injection, and continued then every 60 minutes for a total of 5 hours to determine the absorption and disposition characteristics of Metacam. Three milk samples (2 ml each) were collected from each sow following oxytocin injection at approximately 1, 3 and 5 hours following Metacam injection.

Blood samples were centrifuged at 1400 xg for 10 minutes and the serum portion was transferred to labeled vials for storage at -20°C awaiting analysis. Milk samples were transferred to labeled vials and frozen at -20°C awaiting analysis.

**LC-MS analysis**

Metacam analysis in blood serum and the milk were completed by high-performance liquid chromatography (HPLC)- mass spectrometry, with development and validation of the assays directed by Dr. Jane Alcorn, Associate Professor of Pharmacy in the College of Pharmacy and Nutrition, University of Saskatchewan. Instrumentation used and sample preparation are described below.

**Instrumentation:** Samples were analyzed on an Agilent 1200 series HPLC coupled with a QTrap 4000 Mass spectrometer (AB Sciex). The mobile phase consisted of 0.1% formic acid (mobile phase A) with a flow rate of 0.4 mL/min. Mobile phase B consisted of acetonitrile with a flow rate of 0.4mL/min. A mobile phase gradient was used starting at 100% B from 0 to 1 min, 50% B from 1 to 2 min and maintained until 12 min, 100% B from 12 to 13 min and held at 100% B until 15 minutes. The column was a Thermo Scientific 150 x 3mm, 5µm coupled with a Hypersil Gold Phenyl 5 µm 10 x 2.1 mm Drop-In Guard. The qualifying ion for Metacam (Meloxicam) was 352.1 m/z and the quantifying ion was 114.9 m/z, while the qualifying ion for Piroxicam (internal standard) was 332.1 m/z and the quantifying ion was 95.1 m/z. The retention time for Metacam was 9.6 min while Piroxicam was 8.7 minutes. The mass spec setting for Metacam had a declustering potential of 61V, collision energy of 27 V and exit potential of 20V. Piroxicam mass spec setting had a declustering potential of 61V, collision energy of 61V and exit potential of 16V. The source temperature was 350°C and the ionization spray energy was 5500V. The curtain gas, gas 1 and gas 2 flow rates were all set to 40 arbitrary units.

**Serum Sample Preparation:** A 100µL sample of pig serum was added to 400µL of 2 ng/mL Piroxicam, dissolved in methanol containing 0.1% formic acid, to precipitate the proteins. Samples were vortex-mixed for 5 seconds and then centrifuged for 10 minutes at 15000 xg. The supernatant was transferred to an injection vial and 20µL was injected onto the column. The lowest limit of detection for Metacam in serum was 0.04 ng/mL. Metacam was linear over the range of 0.04 ng/mL to 100 ng/mL.

**Milk Sample Preparation:** A 200µL sample of pig milk was added to 100µL of 4.5ng/mL Piroxicam, dissolved in methanol containing 0.1% formic acid. Trichloroacetic acid (200µL) was added to each sample, vortexed for 5 seconds and centrifuged for 5 minutes at 15000 xg. The analytes were extracted by using SOLA solid phase extraction (SPE) columns from Thermo Scientific. The SPE was conditioned by preconditioning with 2 columns of methanol followed by 4 columns of water. 350µL of supernatant was added to the column and washed with 1 column of water and eluted with 1mL methanol. The eluate was evaporated under a stream of air. Residue was reconstituted in 200µL of 50% methanol. The sample was transferred to an injection vial and 20µL was injected onto the column. The lowest limit of detection for Metacam in milk was 0.35 ng/mL. Metacam was linear over the range of 0.35 ng/mL to 175 ng/mL.

**Study 2: Translactational transfer of Metacam to piglets**

Three sows were selected at 7 to 9 days post partum that were either parity three or four. High producing sows were selected, with each sow weaning 11 or more piglets in her previous litter and having at least 12 piglets in the current litter. Each sow was injected with 1 mg/kg Metacam (20mg/ml, Standard dosage: 0.5 mg/kg or 2.0 ml/ 100 kg body wt) using a slaphot injection device with a 16G ½” needle. Video cameras were mounted over each sow to record nursing bouts and the teat position and suckling duration of the two focal piglets per litter. Video cameras were run for the duration of the sampling period (4.5 hours).

To determine analgesic concentrations in milk, three milk samples were collected per sow at set intervals.
(1, 3 and 5 hours) following analgesic administration (three samples per sow). Injections of oxytocin were used to promote milk let-down for sampling. An initial dose of 0.5 ml was injected into the vulva of the sows. Teats were then checked periodically for milk let down. If no milk was present after 10 to 15 minutes, another injection of 0.5 ml was given, without exceeding 2.0 ml at each collection interval. Milk collected was transferred into micro vial sample containers and placed in the freezer.

Blood samples were collected from two male piglets per litter (six piglets in total over three sows) in intervals ending at 4.5 hours after first nursing (Figure 1). Only healthy male piglets that suckled at anterior teats were selected for sampling. Blood sampling was staggered over time, so that each of the two focal piglets was only sampled twice, producing a total of 12 blood collections from piglets over the three study litters. A 20G, ½” needle and 6 ml syringe were used to collect 4 ml blood per sample by jugular vein puncture. Blood samples were centrifuged at 2000g for 10 minutes, serum was separated and frozen at -20ºC. Samples were analyzed as described for Study 1.

A further study was initially proposed to examine the effectiveness of translacational drug transfer on reducing pain responses at castration. However, due to the low levels of drug found in piglet serum, the likelihood of achieving anesthesia translacationally was deemed to be very low and this part of the study was cancelled.

**Piglet Blood Sampling Schedule**

![Piglet Blood Sampling Schedule](image)

**Figure 1.** Piglet blood sampling schedule. Each piglet was sampled twice: within a litter, piglet 1 was sampled at 15 min and 150 min, and piglet 2 at 60 and 270 min after the first nursing.

**Study 3: Pain assessment following castration**

A total of 31 male piglets were used from 6 litters in a study to validate a novel behavioral method for assessing pain following castration. A moveable handling chute was constructed for the purpose of this study (Figure 2). The chute is designed to fit at the back of a 177 cm wide farrowing crate, replacing the back gate. It is 177.8 cm long and 17.8 cm wide, with surrounding walls at a height of 30.5 cm, and an open top to allow for observer viewing. Two slots for adjustable hurdles approximately 37 cm and 87 cm from the start position allowed for removal and replacement of hurdles of various heights. For this study, 0cm hurdles (slot empty, no hurdle), 5.1 cm hurdles, and 10.2 cm hurdles were used. At the finish position, the chute opens into the farrowing crate, allowing the piglet to return to his dam.

For each trial, the chute system was fitted to the back of the farrowing crate for the litter being tested. Piglets were caught and kept in a large plastic bin prior to each trial.
Selection and Training
Selection and training of piglets was carried out for each litter at 4 days of age. Each male piglet was weighed and paired study piglets per litter were selected and randomly assigned to sham or regular castration treatments. Body weights ranged from 1.25-2.85 kg (mean: 1.87 kg, SD: 0.30). Piglets were marked individually with broad tipped felt marker (Sharpey) for identification.

All piglets were trained to navigate the chute, in preparation for the treatment trials the following day. Training sessions involved a series of four trials. The first trial was performed at 0 minutes, using no hurdles. The second trial was performed at 45 minutes, also using no hurdles. The third trial was performed at 90 minutes, using two 5.1 cm hurdles. The fourth and final trial was performed at 180 minutes, using two 10.2 cm hurdles. Navigation time (NT) was recorded for all four trials, while stride length (SL) was recorded in the fourth trial only.

Treatments and Testing
Treatment sessions for each litter occurred at five days of age, on the day following their training session. For all trials in the treatment session, two 10.2 cm hurdles were used. A PreTrial (PT) was run 10 minutes prior to treatment. Piglets were then treated, with castration performed by the researcher according to industry standards/SOP, and sham castrates handled in similar manner and for the same length of time, but were not castrated. Following treatment, piglets were run through the chute immediately after treatment (0 min). Trials were repeated at 15 minutes (15 min), 30 minutes (30 min), one hour (1 hr), four hours (4 hr), eight hours (8 hr) and 24 hours (24 hr) following treatment. For all treatment trials, Navigation Time (NT) and Stride Length (SL) measures were recorded.

Navigation Time: For all trials, piglets were placed at the start position (with nose behind the start line) by the researcher. The time taken to move from the start position to finish position (nose touches the finish line), called Navigation Time (NT), was recorded in seconds by a second individual using a stopwatch.

Stride Length: Piglets’ stride length in centimeters was recorded while in the chute. A sheet of paper 20.3 cm by 61 cm was placed on the floor of the chute between the start position and the first hurdle. The piglets’ hind feet were pressed to an inkpad immediately prior to their being placed in the start position. The placement of the hind feet was thus recorded during testing in the chute. If footprints were hard to identify, or if the pattern on paper did not clearly reflect steps taken, marks were made on the paper to clarify steps taken immediately following the trial. Papers were removed and replaced between trials.
Stride Length (SL) was obtained by measuring distance between two footprints. The footprint nearest the hurdle (and farthest from the start position) was chosen. Length of stride was measured between this footprint and the proceeding footprint of the same foot. If a clear stride could not be isolated nearest the hurdle, the clearest stride next closest to the hurdle was used.

Analysis
Navigation time (NT) and stride length (SL) for each piglet and test period were calculated as the percent change from the PreTrial (PT) value. For NT the value was calculated using the formula: \(\frac{(X_{\text{trial NT}} - PT_{\text{NT}})}{PT_{\text{NT}}} \times 100\), and for SL the value was calculated using the formula: \(\frac{(X_{\text{trial NT}} - PT_{\text{NT}})}{PT_{\text{NT}}} \times 100\). Results for each time point (0 min, 15 min, 30 min, 1 hr, 4 hr, 8 hr, 24 hr) were analyzed using Proc Mixed in SAS (SAS 9.2, SAS Institute Inc., Carey, NC) with main effects of litter and treatment, and their interaction, and random effect of piglet nested within litter. NT data were log transformed to obtain normality.

Results:

Study 1: Pharmacokinetics of Metacam
Serum and milk samples from the first phase of the study have been evaluated. The analgesic was present in sows’ blood and milk samples, with an average peak concentration in blood of approximately 500 ng/ml at between 1 and 2 hours post injection (Figure 3). Considerable variation in Metacam concentrations was observed between individual sows. Maximum drug levels in serum observed in individual sows were 672, 768 and 1042 ng/ml for treatments 1, 2 and 3, respectively.

![Average Drug Concentration Over a 5 Hour Period](image)

**Figure 3.** Average concentration of Metacam in sow blood over a 5 hour period following injection. Metacam dosage: Treatment 1: 0.5 mg/kg; Treatment 2: 0.75 mg/kg; Treatment 3: 1.0 mg/kg (n= 12, with 4 sows per treatment).
The average peak concentration in sow milk was approximately 200 ng/ml at 3 hours post injection (Figure 4). Considerable variation in Metacam concentrations was similarly observed between the milk concentrations of individual sows. Maximum drug levels in milk observed in individual sows were 138, 1220, and 406 ng/ml for treatments 1, 2 and 3, respectively, however it should be noted that these are preliminary results as the results for treatments 2 and 3 exceeded the standard curve.

**Figure 4.** Average concentration of Metacam in sow milk over a 5 hour period following injection. Metacam dosage: Treatment 1: 0.5 mg/kg; Treatment 2: 0.75 mg/kg; Treatment 3: 1.0 mg/kg (n= 12, with 4 sows per treatment).

*Translactational transfer of Metacam® to piglets*

Based on the results from the first experiment the sows were given the highest dose of Metacam (1.0 mg/kg) for the second experiment. Analgesic concentration in the sow’s milk peaked at three hours post injection. Average Metacam concentrations in milk at 60, 180 and 300 min following injection are shown in Table 1. The levels are similar to those measured in study 1 at the same drug dosage.

**Table 1.** Average Metacam concentrations in sow milk over a 5 hour period following IM injection of 1.0 mg/kg Metacam®.

<table>
<thead>
<tr>
<th>Time post injection (min)</th>
<th>Mean drug conc. (ng/mL)</th>
<th>Standard deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>60</td>
<td>148.67</td>
<td>11.33</td>
</tr>
<tr>
<td>180</td>
<td>188.33</td>
<td>80.03</td>
</tr>
<tr>
<td>300</td>
<td>139.03</td>
<td>84.11</td>
</tr>
</tbody>
</table>

Analgesic concentrations in piglet serum were found to increase significantly over time (Figure 5). The largest concentration increase was found between 90 to 175 minutes (1.5 hrs. to 3 hrs.) post-injection. A decline in analgesic concentration was not observed in the five hour time period. Therefore, the peak drug concentration is unknown, however, based on the slight increase of 0.4 ng/mL between the 3rd and 4th samples (Table 2), the concentration may have reached its plateau.
Figure 5. Average Metacam concentrations in piglet serum following injection of the sow at 0 min.

**Table 2.** Average Metacam concentrations in piglet serum over time (n = 3 per time point).

<table>
<thead>
<tr>
<th>Time (m)</th>
<th>Calculated conc. mean (ng/mL)</th>
<th>Standard deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>45</td>
<td>0.443</td>
<td>0.247</td>
</tr>
<tr>
<td>90</td>
<td>0.385</td>
<td>0.343</td>
</tr>
<tr>
<td>175</td>
<td>2.240</td>
<td>0.624</td>
</tr>
<tr>
<td>295</td>
<td>2.647</td>
<td>0.547</td>
</tr>
</tbody>
</table>

Pain assessment following castration

Navigation time (NT, % change vs PreTrial) in the chute at 15 minutes after treatment was significantly greater for castrated piglets than for sham castrates (Table 3, Figure 6). As well, there was a tendency for a longer NT in castrates at 0 min, 30 min and 8 h. Stride length did not show any significant differences between castrated and sham castrated piglets, although there was a tendency for shorter stride vs PreTrial in castrated piglets at 15 min following castration (Table 4, Figure 7).

**Table 3.** Average Navigation Time for castrated and sham castrated piglets as % change vs PreTrial.

<table>
<thead>
<tr>
<th>Test</th>
<th>Treatment</th>
<th>SE</th>
<th>Significance (P)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Castrated</td>
<td>Sham castrated</td>
<td></td>
</tr>
<tr>
<td>0 min</td>
<td>278.8</td>
<td>93.7</td>
<td>60.6</td>
</tr>
<tr>
<td>15 min</td>
<td>665.05</td>
<td>159.9</td>
<td>98.4</td>
</tr>
<tr>
<td>30 min</td>
<td>252.4</td>
<td>69.2</td>
<td>46.0</td>
</tr>
<tr>
<td>1 h</td>
<td>128.7</td>
<td>60.5</td>
<td>23.2</td>
</tr>
<tr>
<td>4 h</td>
<td>117.5</td>
<td>66.8</td>
<td>40.4</td>
</tr>
<tr>
<td>8 h</td>
<td>87.6</td>
<td>16.7</td>
<td>28.3</td>
</tr>
<tr>
<td>24 h</td>
<td>65.5</td>
<td>13.22</td>
<td>32.6</td>
</tr>
</tbody>
</table>

**Table 4.** Average Stride Length for castrated and sham castrated piglets as % change vs PreTrial.

<table>
<thead>
<tr>
<th>Test</th>
<th>Treatment</th>
<th>SE</th>
<th>Significance (P)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Castrated</td>
<td>Sham castrated</td>
<td></td>
</tr>
<tr>
<td>0 min</td>
<td>-11.15</td>
<td>7.19</td>
<td>9.90</td>
</tr>
<tr>
<td>15 min</td>
<td>4.09</td>
<td>4.42</td>
<td>0.13</td>
</tr>
<tr>
<td>30 min</td>
<td>-8.31</td>
<td>4.09</td>
<td>7.54</td>
</tr>
<tr>
<td>1 h</td>
<td>-6.11</td>
<td>-1.34</td>
<td>7.15</td>
</tr>
<tr>
<td>4 h</td>
<td>-0.93</td>
<td>15.49</td>
<td>7.40</td>
</tr>
<tr>
<td>8 h</td>
<td>1.44</td>
<td>13.08</td>
<td>10.51</td>
</tr>
<tr>
<td>24 h</td>
<td>7.63</td>
<td>11.85</td>
<td>8.40</td>
</tr>
</tbody>
</table>
Figure 6: Average Navigation Times as percent change from PreTrial for Castrated and Sham Castrated treatment groups. Navigation Times for each piglet were calculated as the percentage of PreTrial value using the formula: \( \frac{(X_{\text{trial NT}} - PT_{NT})}{PT_{NT}} \times 100 \). One star (*) indicates a statistical significance of \( p < 0.01 \).

Figure 7: Average Stride Length as percent change from PreTrial for Castrated and Sham Castrated treatment groups. Stride Lengths for each piglet were calculated using the following formula: \( \frac{(X_{\text{trial SL}} - PT_{SL})}{PT_{SL}} \times 100 \).
Discussion:

Surgical castration of piglets is generally practiced to avoid boar taint and undesirable sexual behavior, however, performing surgical castration without analgesia is a welfare concern. There is evidence that castration is painful, and may negatively affect productivity of the animal. Castration of piglets causes an increase in blood cortisol concentration (Carroll et al., 2006), an increase in heart rate and an increase in high frequency vocalizations (White, et al., 1995) indicative of pain and distress. Also, castrated piglets spend less time standing and suckling and more time lying than non castrated piglets (McGlone, et al., 1993). Castrated piglets walk less after castration, spend less time sitting than non castrated piglets and display pain-related behaviors like huddling and trembling (Moya, et al., 2008). Given the growing awareness of animal welfare issues within the general public, the swine industry needs to respond proactively to the issue of pain management at castration. Many of the changes in animal husbandry are consumer - and consequently – retailer driven. Identifying solutions to this problem can also provide US producers with a competitive advantage in world markets. Practical analgesia and/or anesthesia methods, or alternative ways of reducing boar taint, are therefore needed to address the animal welfare concerns related to surgical castration.

Methods of providing analgesia or anesthesia through injection increase labor requirements because each piglet must be treated individually. For most medications, the time delay between drug administration and effectiveness that requires that animals to be handled twice, which is stressful for the piglet. Because of this, it is unlikely that the current pain mediation options available to producers will be readily adopted in commercial practice. The concept of providing analgesia through the sow’s milk is a less laborious method of pain mediation that could provide producers with a practical method of improving animal welfare.

Metacam was successfully transferred through the sow into her milk in the first experiment. Metacam injected intramuscularly into the sow resulted in Metacam levels of up to 1042 ng/ml in serum. Peak concentrations were found between 1 and 2 hours following injection, and there was considerable variation in Metacam concentrations between individual sows. Looking at milk samples from these sows, the average serum Metacam concentration was approximately 200 ng/ml at 3 hours post injection, and considerable variation in drug concentration between individual sows was similarly observed.

Based on these results, the Metacam concentration in milk at approximately 1 hour post-injection is 200 ng/ml. Thus a 7 day piglet weighing 2 kg and consuming 20 ml of milk will obtain a total of 4000 ng, for a drug dosage of 2000 ng/kg. The standard intramuscular treatment dosage is 0.4 mg/kg (400,000 ng/kg), so the oral dose received by a piglet ingesting 20 ml of milk is only 1/200th of the standard therapeutic dosage. In light of these concentrations, we did not expect piglets to obtain therapeutic amounts of drug or to benefit from pain relief.

The second experiment studied the actual amount of Metacam transferred to piglets through the sow’s milk at feeding. Analgesic concentrations in piglet serum were found to increase significantly over time from an average level of 0.44 ng/ml at 45 minutes, to 2.65 ng/ml at 295 minutes. The largest concentration increase was found between 90 to 175 minutes (1.5 hrs. to 3 hrs.) post- injection. A decline in analgesic concentration was not observed in the five hour time period, and therefore, the peak drug concentration is unknown. However, based on the slight increase of 0.4 ng/ml between the 3rd and 4th samples, the concentration may have reached its plateau.

The low levels of Metacam detected in piglet serum are not promising in terms of the potential for this technique to offer pain relief. For example, sows given the recommended dosage in Study 2 achieved average drug levels in serum up to 470 ng/ml, a level approximately 150 times higher than is found here in piglet serum. This confirms that analgesic effects using translactational transfer are unlikely. It has been suggested that piglets may be more sensitive to the analgesic effects of Metacam than adult sows, but this remains to be demonstrated.

In previous rodent studies using Metacam in rats, it was found that the drug concentration in rat milk increases over time from 1 to 24 hours after administration (Busch, et al., 1998). Five mg/kg of Metacam was administered orally to female rats nursing 9 to 11 day old pups, and concentrations in plasma and milk were studied. At 1 hour after administration, the rat plasma contained 12.6 mg/liter Metacam whereas the milk contained 9.7 mg/liter Metacam. Drug concentrations in plasma and milk at 5 hours after administration were 18.4 mg/liter and 22.3 mg/liter, respectively and at 24 hours 6.0 mg/liter and 9.9 mg/liter, respectively (Busch, et al., 1998). These results demonstrate increasing concentrations of the drug in milk relative to plasma over time which was not exhibited in the present study. The peak drug concentration in the milk occurred at 5 hours post injection and is higher than that observed in the present study, corresponding to the 5x higher drug dosage used in rodent work. It would therefore be of interest to study the pharmacokinetics of Metacam excretion in pig milk over a longer interval, and potentially with a higher dosage, to determine if a similar excretion profile occurs in pigs.
Two alternatives to this protocol which may provide more benefit to piglets include increasing the amount of drug injected into the sow, and modifying the drug to preferentially increase excretion via the milk. The first option is limited by drug cost and health concerns for the sow (eg liver damage) when administering an elevated dosage. The second option, drug modification, will require further research and collaboration with the drug manufacturer, Boehringer Ingelheim. Overall, this study provides initial results regarding a novel method to administer pain relief to piglets; based on these results we conclude that either an alternative drug or method is needed to achieve pain relief following castration of piglets.

Regarding the handling chute assessment and validation, it was found that a piglet handling chute with hurdles was effective for indentifying behavioral differences between castrated and sham castrated piglets at 15 minutes post-surgery. At 15 minutes, piglets that had been castrated took on average 6.6-fold longer to navigate the chute compared to before treatment, whereas sham castrated piglets took 1.6-fold longer to traverse the chute than before treatment. Based on this validation work, the handling chute appears to be a simple and useful tool for assessing the effectiveness of analgesic treatments at castration.

**Publications**

**References**


