

RESEARCH PAPER

## Factors affecting mechanical nociceptive thresholds in healthy sows

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### Abstract

**Objective** To describe anatomical and methodological factors influencing mechanical nociceptive thresholds (MNTs) and intra-site variability in healthy sows.

**Study design** Prospective, randomized validation.

**Animals** Eight pregnant, healthy, mixed-parity sows (176–269 kg).

**Methods** Repeated MNT measurements were taken: 1) with a hand-held probe and a limb-mounted actuator connected to a digital algometer; 2) at nine landmarks on the limbs and tail; and 3) at 1 and 3 minute intervals. Data were analysed using linear mixed regression models.

**Results** The MNTs ( $\pm$ SEM) of the limbs were lower with the probe ( $14.7 \pm 1.2$  N) than with the actuator ( $21.3 \pm 1.2$  N;  $p < 0.001$ ), in the pelvic *versus* the thoracic limbs ( $16.7 \pm 1.2$  *versus*  $19.2 \pm 1.2$  N;  $p < 0.001$ ), and in the lateral *versus* the dorsal metatarsi and metacarpi ( $17.6 \pm 1.2$  *versus*  $18.4 \pm 1.2$  N;  $p = 0.002$ ). MNTs were higher in all subsequent measurements compared with the first ( $p < 0.001$ ) and in the morning compared with the afternoon ( $p = 0.04$ ). We found

no evidence of MNT differences based on interval between consecutive measurements (1 *versus* 3 minutes). Variability was lower in the thoracic limbs [mean back-transformed  $\log_{10}$  coefficient of variation (CV)  $\pm$  SE =  $25.5 \pm 1.5\%$  *versus*  $30.6 \pm 1.5\%$  in the pelvic limbs;  $p < 0.001$ ], with the actuator ( $22.7 \pm 1.5\%$  *versus*  $33.4 \pm 1.5\%$  with the probe;  $p < 0.001$ ), and on the left (CV =  $26.9 \pm 1.5\%$  *versus*  $29.3 \pm 1.5\%$  on the right;  $p = 0.01$ ). Tail data (probe only) were analysed separately: mean MNT ( $\pm$  SE) was  $11.7 (\pm 1.8)$ ; MNT increased in days 3–6 of testing compared with day 1 ( $p < 0.001$ ). The mean CV ( $\pm$ SE) was  $38.9\% (\pm 1.1\%)$ .

**Conclusions and clinical relevance** MNTs and intra-site variability in healthy sows were affected by several factors, indicating that this methodology requires considerable attention to detail.

**Keywords** measurement, mechanical nociceptive threshold, pain, pig, variability.

### Introduction

Animals have been used for decades to study nociception – the neural process of encoding noxious stimuli (IASP 2012) – and to derive information about the physiological pathways that generate pain (Le Bars et al. 2001; Mogil 2009,

2012). Inflammatory processes can induce pathological changes in pain perception pathways, such as allodynia, defined as pain resulting from a stimulus that does not normally provoke pain (IASP 2012), and hyperalgesia, defined as increased pain from a stimulus that normally provokes pain (IASP 2012). Both conditions can become debilitating and compromise quality of life in human patients (Laursen et al. 2005; Imamura et al. 2008) and they can also presumably negatively affect animal well-being. For this reason, a number of nociception studies have focused on normal *versus* abnormal processing of noxious stimuli in many species of veterinary interest. The principal aim was to investigate the presence of hyperalgesia – which may develop both during and long after the original inflammation process – and to verify the efficacy of anti-inflammatory and analgesic drugs (Whay et al. 2005; Dixon et al. 2007; Haussler et al. 2007; Caplen et al. 2013; Tapper et al. 2013; Mohling et al. 2014). During the past 15 years, livestock species have been under increasing scrutiny, as they are often affected by painful inflammatory conditions causing hyperalgesia that may be overlooked. Limb disease is one example: lame dairy cows (Whay et al. 1998), sheep (Ley et al. 1995; Colditz et al. 2011) and pigs of different ages and gender had lower mechanical nociceptive thresholds (MNTs) in the affected limbs, whether tested at the site of the lesion or elsewhere on the limb (Sandercock et al. 2009; Fosse et al. 2011a,b; Nalon et al. 2013; Tapper et al. 2013; Mohling et al. 2014).

Recently, MNT testing of the limbs has been described as a potentially objective tool to assess the efficacy of analgesic protocols in sows affected by lameness (Tapper et al. 2013; Mohling et al. 2014; Pairis-Garcia et al. 2014). However, the methodology currently suffers from a number of limitations: MNTs in pigs differ substantially depending on the configuration of the instrument used (Nalon et al. 2013), the size and shape of the probe tip (Fosse et al. 2011b), the age and weight of the animals (Janczak et al. 2012; Di Giminiani et al. 2013), anatomical location (Di Giminiani et al. 2013; Nalon et al. 2013), as well as familiarity with the procedure (Di Giminiani et al. 2015). Furthermore, MNT can be influenced by the time of day (pigs, Nalon et al. 2013; dogs, Coleman et al. 2014), anticipation or sensitization phenomena (humans, Jones et al. 2007; dogs, Coleman et al. 2014) and distraction (donkeys, Ruscheweyh et al. 2011; Grint et al. 2014). The low repeatability of consecutive

MNT measurements at the same anatomical site is also an issue (horses, Haussler et al. 2007; sheep, Stubbsjøen et al. 2010; dogs, Coleman et al. 2014). In conclusion, before MNT testing is proposed as a valid and reliable research tool, 'it should be fully evaluated in normal animals for consistency, repeatability, and the factors influencing threshold responses need to be known' (Coleman et al. 2014).

In our study we investigated methodological and anatomical factors affecting MNT in healthy sows, namely the measuring method (hand-held probe *versus* remotely controlled actuator), the anatomical location (thoracic *versus* pelvic limbs, right *versus* left side of the body, dorsal *versus* lateral metacarpi and metatarsi, and ventral aspect of the tail), and the interval (1 *versus* 3 minutes) between repeated stimuli. We also investigated factors affecting variability when taking repeated measurements at the same site.

## Materials and methods

### Ethical statement

The experiment was approved by the Ethical Commission of the Institute for Agricultural and Fisheries Research (ILVO) (authorization no. 2011/146 and subsequent modifications).

### Animals and housing

The experiment was carried out at the ILVO, Melle, Belgium in March and April 2012. Eight pregnant hybrid sows (RA-SE Genetics, Belgium) from the same experimental herd were studied. Sample size was determined with Win Episcope 2.0 based on a two-tailed, paired-sample test (hand-held probe and remotely controlled actuator) and assuming a population mean of 12.6 and 15.9 N, respectively, and an expected standard deviation (SD) of 6.7 N (Nalon et al. 2013), at a 95% confidence level and 85% power. The sows were chosen from a pool of 20 belonging to the same static mid-gestation group (the only one available at the time of the trial). This enabled us to train and test the animals for 6 weeks consecutively. Average parity was 5.6 (range 2–11), and gestation stage 54 ( $\pm 11$ ) days. The average body weight was 233 kg ( $\pm 33$ ; range 176–269). The selected sows were group-housed in a 10.7  $\times$  3.5 m pen with a solid concrete floor and deep straw bedding, and were fed a restricted diet (2.6 kg of standard commercial gestation feed) in

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