Doctoraatsproefschrift nr. 1023 aan de faculteit Bio-ingenieurswetenschappen van de K.U.Leuven

# Reducing/adjusting painful management practices in piglets

An attempt to improve animal welfare and zootechnical results

Sanne Van Beirendonck

Promotor: Prof. R. Geers, K.U.Leuven

Leden van de examencommissie: Prof. M. De Proft, K.U.Leuven, voorzitter Prof. B. Goddeeris, K.U.Leuven Prof. D. Lips, K.U.Leuven Prof. J. Relaes, K.U.Leuven Dr. B. Hoet, FOD Ing. J. Van Thielen, KHK

Proefschrift voorgedragen tot het behalen van de graad van Doctor in de Bio-Ingenieurswetenschappen

© 2012 Katholieke Universiteit Leuven, Groep Wetenschap & Technologie, Arenberg Doctoraatsschool, W. de Croylaan 6, 3001 Heverlee, België

Alle rechten voorbehouden. Niets uit deze uitgave mag worden vermenigvuldigd en/of openbaar gemaakt worden door middel van druk, fotokopie, microfilm, elektronisch of op welke andere wijze ook zonder voorafgaandelijke schriftelijke toestemming van de uitgever.

All rights reserved. No part of the publication may be reproduced in any form by print, photoprint, microfilm, electronic or any other means without written permission from the publisher.

ISBN 978-90-8826-238-8 D/2012/11.109/16

## Voorwoord

Een doctoraat is een werk van lange adem, en dit doctoraat kon slechts tot stand komen dankzij de hulp van velen. De vele mensen die bijgedragen hebben tot dit werk (direct en indirect) wil ik hier daarom graag bedanken.

Eerst wil ik mijn promotor, prof. Rony Geers bedanken voor de samenwerking de voorbije 4 jaar en de vele nuttige besprekingen/opmerkingen die er steeds voor zorgden dat het werk er in kwaliteit op vooruitging.

Ook de juryleden prof. M. De Proft, prof. B. Goddeeris, prof. D. Lips, prof D. Maes, prof. J. Relaes, dr. B. Hoet en ing. J. Van Thielen wil ik uitdrukkelijk bedanken. Dankzij hun opmerkingen en suggesties is de finale versie van dit werk er enkel beter op geworden.

Prof. B. Goddeeris, prof. D. Lips, prof. D. Maes, dr. B. Hoet en ing. J. Van Thielen wil ik bedanken als assessoren van dit werk.

KHK, en in het bijzonder Marc Geerinckx en Wim Van Malcot, voor de financiering van het werk.

Jos Van Thielen en Bert Driessen als projectverantwoordelijken van de 2 projecten waarbinnen dit werk heeft plaatsgevonden.

De firma Schippers wil ik bedanken voor het leveren en aanpassen van het CO<sub>2</sub>-verdovingstoestel.

Johan Claes (Lab4Food, KHK) voor het uitlenen van de CO<sub>2</sub>-meter.

Een speciale vermelding voor Bert Driessen. Bert, jij gaf me 4 jaar geleden de kans om mijn tanden in dit project te zetten, en hebt me steeds gemotiveerd als het wat moeilijker ging. Je bent de voorbije jaren een soort van niet-officiële co-promotor geweest, en hebt me mee door de laatste zware loodjes geholpen. Bedankt!

Ook wil ik graag alle (oud-)collega's bedanken, van KHK, KULeuven en het ZTC. Bij KULeuven voornamelijk de collega's van het Labo Kwaliteitszorg in de Dierproductie, bij KHK de collega's van de departementen ABT en IBW, en in het bijzonder de collega's van de onderzoeksgroep Dier&Welzijn. Bedankt voor de fijne samenwerking en de gewaardeerde hulp waar nodig!

Prof. Geert Verbeke wil ik graag bedanken voor de hulp bij de statistische verwerking.

Ook zijn er verschillende thesisstudenten de revue gepasseerd gedurende mijn doctoraatsperiode: Sara Kerselaers, Ellen De Maeyer en Joyce Van Dijck wil ik graag bedanken voor hun nuttige bijdrage.

Een welgemeende dankjewel gaat ook uit naar mijn vrienden. De meiden en hun aanhang, en de vrienden vanuit de bachelor: bedankt om er in te slagen om keer op keer geïnteresseerd te blijven in de stand van zaken en nog belangrijker, om op tijd voor de nodige ontspanning te zorgen!

Uiteraard mag ook de familie niet ontbreken in dit lijstje. Mijn ouders wil ik graag bedanken om me de kans te geven om verder te studeren en om altijd klaar te staan als er iets was. Het was jullie misschien niet altijd even duidelijk waar ik mee bezig was, maar jullie probeerden te helpen waar het kon. Ook mijn broer en zus verdienen een woordje van dank. Thomas voor de nodige hulp wanneer mijn informatica-kennis het liet afweten, Lotte om me nu en dan te verplichten de nodige ontspanning te nemen. Niet op z'n minst de vele avondjes babysitten op Elise en Lars. Ook de rest van de (schoon)familie leefde steeds mee met het hele verhaal, bedankt.

Tot slot kom ik bij Koen. Je bent dit hele verhaal mee ingerold, zonder dat je wist wat het allemaal juist inhield. Toch ben je er steeds zonder problemen in meegegaan, en heb je alle problemen tot vervelens toe aangehoord. Bedankt om steeds te luisteren, raad te geven en te relativeren (en de voorbije maanden zowat het hele huishouden op jou te nemen)! De afronding van dit doctoraat is de afsluiting van een hoofdstuk, ik zie vol enthousiasme uit naar wat komen gaat, samen met jou!

SUMMARY	I
SAMENVATTING	VII
LIST OF ABBREVIATIONS	XIII
CHAPTER 1: GENERAL INTRODUCTION	
SOCIETAL SITUATION OF THE PROBLEM	
PAIN: SCIENTIFIC FOUNDATION THROUGHOUT SPECIES	
Definition of pain	
Consequences of pain	5
PAINFUL PROCEDURES IN PIGLETS: REASON, METHOD AND EFFECTS	6
Castration	6
Tail docking	11
Teeth clipping/grinding	13
Iron injection	16
Vaccination	17
Ear tagging (identification)	17
ETHICAL REFLECTIONS ON PAINFUL PROCEDURES	
AVOIDING PAIN VIA ANESTHETICS: UNDERLYING MECHANISMS AND ALTERNATIVES	19
Underlying mechanisms of pain	19
Anesthetics	21
Local anesthesia	22
General anesthesia	23
Underlying mechanisms of anesthetic techniques	25
ANALGESIA	27
IMMUNOCASTRATION	27
RAISING ENTIRE MALES	
MEASURING PAIN: GOLD STANDARD, HOW TO DETECT PAIN IN FARMED ANIMALS?	
Behavior	
Vocalization	32
Zootechnical results (feed conversion, growth rate, sickness sensitivity)	
Physiological and biochemical parameters	
ANIMAL INTEGRITY	
AIM AND OUTLINE OF THE THESIS	
CHAPTER 2: BELGIAN CONSUMERS' OPINION ON PORK CONSUMPTION CONCERNING ALTERNATION	IVES FOR
UNANESTHETIZED PIGLET CASTRATION	
Abstract	
INTRODUCTION	
MATERIALS AND METHODS	40
Research approach and sampling	40
Questionnaire	

## **Table of Contents**

Statistical analysis	44
Results	44
Awareness	44
Need for alternatives	45
Acceptability of alternatives	46
Preference	48
Willingness to pay extra	49
DISCUSSION	50
Awareness	51
Acceptability of alternatives and preference	52
Willingness to pay extra	53
CONCLUSION	53
CHAPTER 3: BEHAVIOR OF PIGLETS AFTER CASTRATION WITH AND WITHOUT $CO_2$ -ANESTHESIA	55
Abstract	57
INTRODUCTION	57
MATERIALS AND METHODS	59
Animals and Housing	59
Experimental procedure	60
Behavioral Observations	61
Statistical Analysis	63
RESULTS	64
Preliminary experiment	64
Main experiment	65
DISCUSSION	70
Preliminary experiment	70
Main experiment	71
Conclusion	73
CHAPTER 4: IMPROVING SURVIVAL, GROWTH RATE AND ANIMAL WELFARE IN PIGLETS BY AVOIDING	G TEETH
SHORTENING AND TAIL DOCKING?	75
Abstract	77
INTRODUCTION	77
MATERIALS AND METHODS	78
Animals and Housing	78
Experimental procedure	78
Behavioral Observations	80
Statistical Analysis	80
Results	
Behavior from birth to weaning (4 weeks)	81
Growth rate	81
Comparison of weight at weaning	83

Mortality rate	83
DISCUSSION	84
Behavior from birth to weaning (4 weeks)	84
Growth rate	85
Comparison of weight at weaning	85
Survival rate	85
Conclusion	86
CHAPTER 5: PAINFUL STANDARD MANAGEMENT PRACTICES WITH PIGLETS: DOES BUNDLING AND/O	R
ANESTHESIA IMPROVE ANIMAL WELFARE?	87
Abstract	89
INTRODUCTION	89
MATERIAL AND METHODS	90
Animals and Housing	90
Experimental design	90
Behavioral observations	91
Statistical Analysis	91
Results	93
Experiment 1	93
Experiment 2	95
DISCUSSION	96
Experiment 1	96
Experiment 2	98
Conclusion	100
CHAPTER 6: GENERAL DISCUSSION, CONCLUSIONS AND PERSPECTIVES FOR FURTHER RESEARCH	101
APPENDIX	111
CITED REFERENCES	117
LIST OF PUBLICATIONS	133

Summary

In the pig industry, farmers always strive for optimal productions results. In order to obtain these results, they carry out several management practices on piglets like tail docking (to avoid tail biting; routinely prohibited), teeth shortening (to avoid wounding to the udder of the sow or to litter mates; routinely prohibited), castration of male piglets (to avoid boar taint),... All these procedures are however considered to be painful and therefore can also influence animal welfare and zootechnical results of the piglets. The aim of this thesis was to study the effects of reducing or adjusting these painful management practices on piglet welfare and zootechnical results.

In the society of today, animal welfare is gaining consumers' interest and concern. Piglet castration is a sensitive issue that has drawn the attention of the public as a result of animal welfare organizations' campaigns. Therefore it is important to know consumers' opinion on this topic. For that reason the opinion of Flemish consumers on unanesthetized piglet castration, and three possible alternatives is reported in the second chapter. A total of 2018 people, spread over the 5 provinces, were questioned. The results showed that in spite of several media campaigns of animal welfare organizations over the past few years, still about half of the Flemish respondents were not aware of the problem of unanesthetized piglet castration. However, after being informed, the majority wanted unanesthetized castration to be banned. Although the concern about animal welfare implications was very high, the willingness to pay extra for alternatives was low, which might prevent the return of investment for the farmers since production costs will increase.

Results of the second chapter demonstrated that castration under anesthesia was the most accepted alternative by Flemish consumers. Therefore castration under carbon dioxide (CO<sub>2</sub>) anesthesia was compared to unanesthetized castration in the third chapter. No behavioral differences were found in piglets treated with CO<sub>2</sub>-anesthesia or zolazepam, tiletamine (Zoletil<sup>®</sup>) and xylazine (Xyl-M<sup>®</sup>), which is an indication that CO<sub>2</sub> matches the anesthetic properties of the combination of Zoletil<sup>®</sup> and Xyl-M<sup>®</sup>. In the main experiment, observed differences in behavior were not conclusive. However, a difference in interactive behavior indicated a better state of welfare for the CO<sub>2</sub>-anesthetized castrated piglets compared to the unanesthetized castrated piglets. On the other hand, all barrows, including the anesthetized group, displayed behaviors indicative of pain or discomfort. Therefore, piglets may need to be provided with additional analgesia to eliminate the pain caused by castration even if they are anesthetized prior to castration.

Castration is not the only event that may threaten piglets' welfare. As mentioned before, piglets are subjected to several painful management practices, especially during their first week of life. In order to improve overall welfare, not only castration has to be dealt with, but painful management procedures in general. In the fourth chapter it was therefore investigated if reducing painful interventions during the first week of life resulted in better zootechnical performance of the piglets, reduced piglet mortality and if the overall welfare, indicated by behavioral criteria, was improved. In 22 litters, all piglets were weighed after birth. The four lightest piglets of each litter of the experimental group were not subjected to tail docking or teeth shortening, the other procedures (castration for the male piglets, iron injection, vaccination, ear tagging) were carried out as normal. The four lightest piglets of each litter of their tails docked and teeth shortened, next to the other management procedures. All procedures were applied on the heavier piglets of both the control and the experimental group.

The lightest piglets seemed to show less pain related behavior when their teeth and tail were left intact. Moreover, mortality rate tended to be lower when compared with the lightest piglets of the control group but further research specified on neonatal mortality would be useful.

The objectives of the previous two chapters, reducing painful procedures and using anesthesia during these procedures, were combined in the fifth chapter. Two experiments, using 41 litters, were carried out. In the first experiment, all procedures were performed on one moment in time in the experimental group while in the control group procedures were carried out as normal (spread over the first week of life). In the second experiment, bundling of the procedures without anesthesia was compared to bundling the procedures after the animals were anesthetized with CO<sub>2</sub>. Piglets seemed to cope better with pain if painful interventions were not combined. Moreover, the applied CO<sub>2</sub>-anesthesia had facilitated the pain experience after treatment, since lying, interactive and walking behavior indicated more discomfort for the anesthetized piglets. Anesthetized piglets had only an advantage when considering nursing behavior. Although the beneficial effect of anesthesia during painful procedures is not really confirmed by the results, these results should be interpreted as a 'delayed' pain experience for anesthetized piglets rather than an additional pain experience. As already found in chapter one CO<sub>2</sub>-anesthesia relieves the pain during the procedure, but not after the anesthesia has worn of. The post-operative pain may have been present in both treatments, but the absent pain

experience during the procedures for piglets of the anesthetized group can still be interpreted as advantageous to piglet welfare.

The results of this thesis showed that public awareness on piglet castration was still low. On the other hand, when respondents were informed, they felt the need for alternatives was high. Castration under anesthesia was the most accepted alternative but willingness to pay an extra price to improve piglet welfare was low. Reducing painful procedures (tail docking and teeth clipping) improved animal welfare and survival rate to a certain extent. Adjusting the painful procedures by using CO<sub>2</sub>-anesthesia and/or bundling all procedures on one moment in time did not give clear results. Bundling of the procedures without anesthesia did not seem to benefit the piglets. Anesthesia with  $CO_2$  provides anesthesia and analgesia on the moment of the procedure, which improves animal welfare, but it wears off quickly resulting in postoperative pain. Further research could be useful to optimize the procedure, e.g. by providing additional analgesia for the postoperative pain.

Samenvatting

Varkenshouders streven steeds naar optimale productieresultaten. Om deze resultaten te behalen, voeren ze verschillende procedures uit bij biggen zoals staartjes couperen (om staartbijten te vermijden; routinematig verboden), tandjes inkorten (om wonden aan de uier van de zeug of aan hokgenootjes te vermijden; routinematig verboden), castratie van mannelijke biggen (om berengeur te vermijden)... Al deze procedures worden echter verondersteld pijnlijk te zijn en kunnen daarom het welzijn en de zoötechnische resultaten van biggen beïnvloeden. Het doel van deze thesis was om de effecten van het reduceren of aanpassen van deze pijnlijke ingrepen op het welzijn en de zoötechnische resultaten van de biggen te bestuderen.

In de hedendaagse maatschappij wekt dierenwelzijn meer en meer de interesse en bezorgdheid van de consument. Daarom is het belangrijk om te onderzoeken of er, wat de consument betreft, een draagvlak is voor het verbeteren van dierenwelzijn en productieresultaten. Biggencastratie is een gevoelig onderwerp dat de aandacht van het publiek getrokken heeft als gevolg van campagnes van dierenrechtenorganisaties. De opinie van de Vlaamse consument over onverdoofde biggencastratie en drie mogelijke alternatieven werd daarom gerapporteerd in het tweede hoofdstuk. Een totaal van 2018 mensen, verspreid over de 5 Vlaamse provincies werd ondervraagd. De resultaten toonden aan dat ondanks verschillende mediacampagnes van dierenwelzijnsorganisaties de voorbije jaren, nog steeds ongeveer de helft van de ondervraagden niet op de hoogte was van de problematiek rond onverdoofde biggencastratie. Toch pleitte de meerderheid, na geïnformeerd te zijn, voor een verbod op onverdoofde castratie. Hoewel de bezorgdheid over de implicaties voor dierenwelzijn groot was, was de bereidheid tot het betalen van een meerprijs voor alternatieven laag, wat ervoor kan zorgen dat varkenshouders geen rendement voor hun investeringen zullen krijgen aangezien de productiekosten zullen toenemen.

Aangezien in het tweede hoofdstuk bleek dat de consument castratie onder verdoving het meest acceptabele alternatief vond, werd in het derde hoofdstuk castratie onder CO<sub>2</sub>-verdoving vergeleken met onverdoofde castratie. Er werden geen verschillen in gedrag gevonden bij biggen die met CO<sub>2</sub> verdoofd werden of biggen die met zolazepam, tiletamine (Zoletil<sup>®</sup>) en xylazine (Xyl-M<sup>®</sup>) verdoofd werden voor castratie, wat een indicatie is dat CO<sub>2</sub> dezelfde anesthetische eigenschappen heeft als de combinatie van Zoletil<sup>®</sup> en Xyl-M<sup>®</sup>. In het hoofdexperiment gaven de geobserveerde gedragsverschillen geen uitsluitsel, hoewel een verschil in interactief gedrag een indicatie gaf van een beter welzijn voor CO<sub>2</sub>-verdoofde

biggen vergeleken met onverdoofd gecastreerde biggen. Langs de andere kant werd bij alle bargjes, ook bij de verdoofde groep, gedragingen die pijn of discomfort aantonen geobserveerd. Daarom kan het nodig zijn om biggen te voorzien van bijkomende analgesie om de castratiepijn volledig weg te nemen, zelfs al werden ze verdoofd met  $CO_2$  voor castratie.

Castratie is niet de enige ingreep die het welzijn van de biggen bedreigt. Zoals reeds vermeld worden verschillende pijnlijke ingrepen uitgevoerd bij biggen, voornamelijk in hun eerste levensweek. Om het algemeen welzijn te verbeteren moet er niet enkel rekening gehouden worden met castratie, maar met pijnlijke ingrepen in het algemeen. In het vierde hoofdstuk werd daarom onderzocht of een reductie van pijnlijke ingrepen in de eerste levensweek resulteerde in betere zoötechnische resultaten, verlaagde sterfte en of het algemeen welzijn, aangetoond door gedragscriteria, beter was. In 22 nesten werden alle biggen gewogen na de geboorte. Bij de vier lichtste biggen van elk nest van de experimentele groep werden geen staartjes gecoupeerd of tandjes ingekort; de andere ingrepen (castratie bij de vier lichtste biggen, ijzerinjectie, vaccinatie, oormerken) werden normaal uitgevoerd. Bij de vier lichtste biggen van elk nest van de controlegroep werden wel staartjes gecoupeerd en tandjes ingekort, naast de andere ingrepen. Bij de zwaarste biggen van zowel de controle- als de experimentele groep werden alle ingrepen uitgevoerd.

De lichtste biggen leken minder pijngerelateerd gedrag te vertonen wanneer hun tanden en staart intact werden gelaten. Verder was er een tendens van verlaagde mortaliteit vergeleken met de lichtste biggen van de controlegroep, maar verder onderzoek naar neonatale mortaliteit zou nuttig zijn.

De doelstellingen van de twee vorige hoofdstukken, reductie van pijnlijke ingrepen en verdoving tijdens de ingrepen, werden gecombineerd in het vijfde hoofdstuk. Twee experimenten met in totaal 41 nesten werden uitgevoerd. In het eerste experiment werden alle ingrepen op hetzelfde moment uitgevoerd in de experimentele groep terwijl in de controlegroep alle ingrepen op de normale manier werden uitgevoerd (verspreid over de eerste levensweek). In het tweede experiment werd het bundelen van de ingrepen zonder verdoving vergeleken met bundelen van de ingrepen nadat de biggen verdoofd werden met CO<sub>2</sub>. Biggen leken beter te kunnen omgaan met pijn wanneer pijnlijke ingrepen niet gecombineerd werden. Meer zelfs, de toegepaste verdoving leek de pijnervaring na de ingreep te versterken aangezien liggen, interactief gedrag en wandelen meer discomfort aantoonde

voor de verdoofde biggen. Verdoofde biggen hadden enkel een voordeel wanneer er naar zuiggedrag gekeken werd. Hoewel het voordelige effect van verdoving tijdens pijnlijke ingrepen niet bevestigd werd door deze resultaten, moeten deze resultaten eerder geïnterpreteerd worden als een 'uitgestelde' pijnervaring dan als een bijkomende pijnervaring. Zoals reeds gevonden in het eerste hoofdstuk neemt CO<sub>2</sub>-verdoving de pijn tijdens de ingreep weg, maar niet nadat de verdoving is uitgewerkt na de ingreep. De postoperatieve pijn mag dan aanwezig geweest zijn in beide behandelingsgroepen, de afwezigheid van pijn tijdens de ingrepen voor biggen van de verdoofde groep kan nog steeds geïnterpreteerd worden als voordelig voor het welzijn van de biggen.

De resultaten van deze thesis toonden aan dat het maatschappelijk bewustzijn rond biggencastratie nog steeds laag is. Langs de andere kant vonden de ondervraagden, na informatie over de problematiek, dat de nood aan alternatieven hoog was. De bereidheid om een meerprijs te betalen om het welzijn van biggen te verbeteren was echter laag.

Verder bleek dat het reduceren van pijnlijke ingrepen (staartjes couperen en tandjes inkorten) biggenwelzijn en overlevingsgraad tot op een bepaalde hoogte verbeterde. Het aanpassen van de ingrepen door het gebruik van CO<sub>2</sub>-verdoving en/of het bundelen van de ingrepen gaf geen duidelijke resultaten. Bundelen van de ingrepen zonder verdoving leek geen voordeel te geven voor de biggen. Verdoving met CO<sub>2</sub> voorziet de biggen van anesthesie en analgesie op het moment van de ingreep, wat het welzijn verbetert, maar de verdoving werkt snel uit waardoor postoperatieve pijn aanwezig blijft. Verder onderzoek zou nuttig zijn om de procedure te optimaliseren, bv door bijkomende analgesie te voorzien voor de postoperatieve pijn.

List of abbreviations

ABE	acid-base equilibrium
ACTH	adrenocorticotropic hormone
CNS	central nervous system
$CO_2$	carbon dioxide
CV	coefficient of variance
ECG	electrocardiogram
EEG	electro-encephalography
e.g.	exempli gratia
EU	European Union
FSH	follicle stimulating hormone
GnRH	gonadotrophic releasing hormone
i.e.	id est
LH	luteinizing hormone
MRL	maximum residue limits
NSAID	non-steroidal anti-inflammatory drug
O <sub>2</sub>	oxygen
Р	probability
pCO2	carbon dioxide partial pressure
s.a.	sine anno
SAS	statistical analysis system
SEM	standard error of the means

**Chapter 1: General introduction** 

## Societal situation of the problem

Piglets are subjected to several painful management practices, especially during their first week of life. Tail docking, teeth clipping or grinding, ear tagging, injections and castration of male piglets are the most common procedures and they are all regulated by law. European directive 2001/93/EG describes these regulations as follows. Tail docking and teeth shortening may not be carried out routinely, but only when injuries on sow's udder or on ears and tails of other piglets have been observed. Before it is decided to carry out these procedures, arrangements should be made to prevent tail biting and other behavioral disorders. Therefore, inadequate environmental factors or management systems should be adjusted, e.g. stocking density, ventilation requirements and pen enrichment. The directive also states that surgical castration without anesthesia must be carried out within the first 7 days of life. When castration is performed on piglets aged older than 7 days it should be done by a veterinarian applying anesthesia, and prolonged analgesia should be used.

In 2010 different actors of the European pig chain, European retailers and ngo's published a declaration which states that they want to stop unanesthetized piglet castration. As a first step, castration may only be carried out with local or general anesthesia or prolonged analgesia according to established methods, starting from 01/01/2012. A second step is to abolish surgical castration of piglets ultimately on 01/01/2018.

In the society of today, animal welfare is gaining consumers' interest and concern. This was demonstrated by several media campaigns of animal welfare organizations over the last few years, which are getting more and more attention of the public. Consumer acceptance on food products resulting from new technologies depends on both processing considerations and quality characteristics associated with the product itself. The concern of the public on new technologies may have as much as an influence in terms of impact on consumer decisions as the quality considerations associated with the product itself (Frewer et al., 1997). It is acknowledged nowadays that consumer attitudes towards food production methods are not only dependent on an analytical assessment of risk, benefit, economics and nutrition alone. Ethical and moral considerations may also have an influence on establishing the societal acceptability of a particular production process (Frewer et al., 2005).

Because of growing public concern, the pig industry and the government are pressured to improve animal welfare and abandon the tradition of routinely applied painful management procedures. Alternatives to reduce painful experiences are therefore urgently needed and the present study tries to contribute to this challenge. In 2009 a field study on alternatives for

unanesthetized piglet castration was started on the request of the Flemish government (CASPRAK). In that study four alternatives for unanesthetized piglet castration were tested on 20 farms throughout Flanders: surgical castration with CO<sub>2</sub>-anesthesia, surgical castration with the administration of Metacam<sup>®</sup>, immunocastration, and raising entire males. The goal of the CASPRAK-project was to investigate the economic, social and ecological impact of each alternative. The added value of the present study is that the emphasis is more on the effect on animal welfare, as measured by behavioral observations, and that painful management procedures other than castration are also looked at. This way, the present study can be seen as complementary to the CASPRAK-project.

## Pain: Scientific foundation throughout species

## **Definition of pain**

Pain is defined by the International Association for the Study of Pain as an "unpleasant sensory and emotional experience associated with actual or potential tissue damage" (IASP, 1979). Pain is always a subjective experience which is partly determined by 'experiences' related to wounding at a young age (IASP, 1979). The neurophysiological processes involved in detection, transduction and transmission of noxious information by nerves and their relay to the central nervous system appears essentially similar in all mammals (Vinuela-Fernandez et al., 2007). According to the principle of analogy it can be assumed on grounds of similarities in anatomy (pain system), physiology (pain sensation) and behavior (pain expression) between humans and 'higher' animals that pain in animals is analogue to pain in humans (Hendriksen and Boumans, 2006). Molony and Kent (1997) described pain in animals as an aversive sensory and emotional experience that represents the notion of an animal of damage or threat to the integrity of its tissues. It influences the physiology of the animal and the behavior to reduce or avoid damage, to avoid the chance on reoccurrence and improve recovery. Jourdan et al. (2001) defined pain in animals as an aversive sensory experience caused by real or potential damage that provokes motor and vegetative protective reactions, avoidance behavior and can modify the species specific behavior of the individual including its social behavior. Anand and Craig (1996) suggested adjusting the traditional vision, which emphasizes mostly self-assessment as a measure for pain. These authors stated that behavioral changes caused by pain are a form of self-assessment for non-verbal individuals, and that behavior cannot be perceived as a 'surrogate measurement' of pain. They accentuated this in the first place for neonates, but also for other individuals not capable of expressing their pain, and by extension for animals. Although nociception (the physiological side of pain) pathways exist in animals, it has been a widespread discussion whether animals also experience the aversive emotional component of pain (Rutherford, 2002). Gentle (2001) noted that human pain experience can be mitigated by redirecting the patient's attention, and based a pain study in chickens on this. He reasoned that if a chicken's reaction to a harmful event was simply an unconscious, automatic reaction, then shifting the chicken's attention should not influence the response. If however, the chicken experienced the pain as an unpleasant feeling, then redirecting the bird's attention might reduce the signs of pain, as it does in the case of humans. He found that chickens that received an injection of sodium urate crystals into a leg joint displayed less pain related behavior when they were placed in a pen with a novel feature (wood shavings) as compared to their barren home pen. The presence of a second, unfamiliar, chicken to distract the injected bird's attention reduced the pain related behavior even more. Gentle concluded that because the bird's reaction to the injection was modified by redirecting its attention, the reaction can not have been an unconscious adjustment of behavior that happened automatically. Instead, it must have been mediated by conscious awareness of the pain (Weary et al., 2006).

Pain experience depends on several factors. Depending on the nature of the pain, location, duration and intensity, this experience can vary from a negligible discomfort to a completely debilitating condition (Magalhaes-Sant'Ana et al., 2009). The response of an animal to a painful stimulus can vary according to species, age, sex, previous experience, and the response can be altered by individual experience (Martini et al., 2000).

## Consequences of pain

There are indications that exposure to pain at a young age has an influence on pain experience and the expression of it at a later age. Taddio et al. (1997) investigated the behavior of children when they were vaccinated at 4-6 months of age. One group had endured unanesthetized circumcision as a neonate, a second group had received a pretreatment with lidocaine-prilocaine before circumcision, and a third group was not circumcised. The authors noticed increasing pain scores at vaccination: non-circumcised children had the lowest pain score, children circumcised after lidocaine-prilocaine treatment had a higher pain score, and children who were circumcised unanesthetized had the highest pain score. However, they made the remark that it is possible that the highest vaccination response of unanesthetized circumcised children can be partially caused by an analogue of posttraumatic stress syndrome, which is triggered by a traumatic and painful experience that is relived under similar circumstances of pain during vaccination.

Anand et al. (1998) investigated the long term effects of exposure to repetitive pain stimuli during the neonatal period in rats. They found that these rats developed a lowered pain threshold and that they showed more fear and avoidance behavior. The authors concluded that repetitive neonatal pain leads to increased sensitivity to stress and anxiety-mediated adult behavior.

Pain does not only have consequences on the behavior of animals, but also on other aspects. Pain causes a slower surgical recovery, as a consequence of an increased basal metabolism, a decreased feed and water intake and a slower recovery of homeostasis (McMeekan et al., 1999; Faulkner and Weary, 2000; Short, 1998). Pain can also lead to a diminished production of meat and milk or reduced reproduction capacity (Bath, 1998). Because of a change in fluid intake and elimination, there can be an imbalance in the fluid and electrolyte equilibrium. In addition, weight loss can occur because of reduced nutrient intake (Short, 1998). Chronic stress, caused by pain, also has a negative influence on the functioning of the immune system of the animal (Salak-Johnson and McGlone, 2007). Pain may prevent newborn animals from feeding and can lead to starvation-induced hypothermia, an associated dulling of consciousness and sleep, or it may cause the newborn to remain alert, restless and continuously disstressed. Preventing or treating the causes of pain or treating the pain itself are important strategies for minimizing its harmful effects on the newborn as such, but also later in life (Mellor and Stafford, 2004).

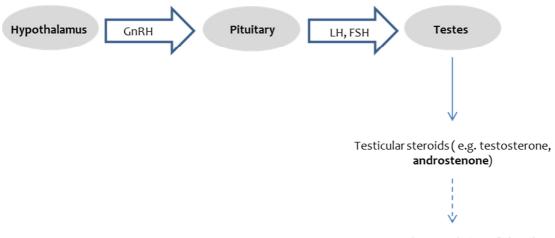
## Painful procedures in piglets: reason, method and effects

## **Castration**

#### Reason

Male piglets for meat production have been castrated for centuries to obtain animals that are easier to handle and less aggressive. But nowadays the main reason for castration is that it eliminates the risk of boar taint. Boar taint is a deviating smell that can be released when meat of entire males is heated and according to Cazaux et al. (2008) 5 to 15% of entire males with a slaughter weight above 85 kg are at risk. Leidig et al. (2009) on the other hand, found that the incidence of boar taint can vary greatly, between 10 and 75%. Because it is not known in advance which boars contain a risk on boar taint, all boars are castrated. The two main

components of boar taint are androstenone and skatole (Figure 1.1). Androstenone is produced in the testicles (Lundstrom and Zamaratskaia, 2006) and depends on the stage of sexual maturity (Bonneau, 1982). Skatole is metabolized from the amino acid tryptophan in the large intestine (Lundstrom and Zamaratskaia, 2006) and also depends on genetics (Lundstrom et al., 1994) in addition to sexual maturity (Zamaratskaia et al., 2005). Because it is derived from tryptophan, skatole is present both in sows and boars. It is however only a potential problem in boars because sows and barrows can metabolize redundant skatole concentrations. In boars, this metabolization is stemmed by the male sex hormones and skatole is -like androstenone- stored in the fatty tissue (EUFIC, 2010). When male piglets are castrated, male sex hormones (e.g. androstenone) are eliminated with the removal of the testicles, which means that skatole can be eliminated completely by metabolization. Castration therefore excludes the risk on boar taint.



Increased accumulation of skatole

Figure 1.1 Physiology of boar taint and influence of the testis function.

Next to elimination of boar taint, castration also leads to more fat deposit in the pig which used to be a desirable quality for taste (Maes et al., 2002). Nowadays, consumers want lean meat. Because of the increased demand of lean meat and lower production costs for non-castrated animals, castration becomes less frequent in some countries. Although there is a pressure to stop castration from an animal welfare point of view, raising non-castrated boars (intact males) is avoided in most countries because of the risk on boar taint (Prunier et al., 2006).

Castration is performed on male piglets before 8 days of age (EU-Directive 2001/93/EC) and mostly without anesthesia. The underlying motivation for this is that it was assumed for a

long time that neonates felt less pain because of the immaturity of their neural development (Anand, 1990; Fitzgerald, 1994; Andrews and Fitzgerald, 1994). Research in human medicine has however demonstrated that perception of pain in neonates and children is comparable to pain perception in adults. It can be assumed on grounds of similarities in anatomy (pain system), physiology (pain sensation) and behavior (pain expression) between man and 'higher' animals that pain in animals is analogue to pain in humans (Hendriksen and Boumans, 2006). There are strong indications that castration causes pain in piglets, not only during the procedure but also in the days following the procedure (Taylor et al., 2001; Hay et al., 2003; Henke and Erhardt, 2004). The innervation of the scrotum and testicles is very complex. Sensory sympathetic nerves can detect pain in the testicles and associated structures and they also innervate the superficial tissues of the scrotum and the blood vessels. All tissues associated with castration are innervated by these nerves. The tissue damage that is inevitable during castration is therefore considered as being painful (EFSA, 2004).

#### Method

Before the actual castration can start, the piglet has to be constrained in order to carry out the procedure without any problems. This can be done by holding the piglet in the hand or by using commercial applications which have the advantage that both hands are free to perform castration (EFSA, 2004). After fixation, one or two incisions are made in the scrotum and the testicles are pushed out through the incision. To improve drainage of wound fluids, the incision has to be made as low as possible on the scrotum (EFSA, 2004). After the testicles have been pushed out, the spermatic cords are cut (Hay et al., 2003). Tearing of the spermatic cord is prohibited since 2003; the spermatic cord has to be cut (EU Council Directive 2001/88/EC). The wound is left open and a disinfectant spray is applied.

It is regulated by law (European Directive 2001/93/EG) that castration on piglets has to be carried out before one week of age. If pigs are castrated after 1 week of age, the procedure has to be carried out by a veterinarian and anesthesia and analgesia must be applied.

### Behavioral effects

Behavior of castrated piglets was studied by Hay et al. (2003). They made a distinction between non-specific behaviors, pain related behaviors, social cohesion and postures. The ethogram of Hay et al., which is also used in other studies, is shown in table 1.1.

Noonan et al. (1994) related spending more time at the udder with painful procedures; they found it is a way of coping with pain for piglets. Noonan et al. (1996) found an explanation

for this in the fact that suckling causes endorphins to be released, which have an analgesic effect. Taylor et al. (2001) confirmed these results when they saw that piglets spent more time at the udder the first 22 hours after castration. Hay et al. (2003), McGlone and Hellman (1988) and McGlone et al. (1993) on the other hand, found that castrated piglets suckled and massaged the udder less frequently the first few hours following treatment.

Castrated piglets spent about 4% of their time trembling the first few hours after castration as being observed by Hay et al. (2003). This behavioral pattern was already seen in lambs the first hours following castration. Lambs that were given a pre-treatment with a local anesthetic trembled significantly less. Based on this fact we can presume that the trembling behavior is caused by pain induced by castration (Molony et al., 1997; Wemelsfelder and Van Putten, 1985).

According to Hay et al. (2003), scratching the rump was nearly absent in uncastrated piglets, in contrast with castrated piglets. Llamas Moya et al. (2008) confirmed the finding that scratching occurs more frequently in castrated piglets. Melzack and Wall (1983) interpreted scratching as a way to relieve pain on a short term. Moreover, it was used to evaluate post-surgical pain in humans (Melzack and Wall, 1983).

Castrated piglets also wagged their tail more frequently (Hay et al., 2003). Tail wagging in castrated piglets was already described by Wemelsfelder and Van Putten (1985) and could be related to tail stump hyperalgesia. Indeed, tail docking was performed a few days before castration and it is not uncommon that pain that pre-exists before a surgery reappears or increases in intensity during the post-operative period (Hay et al., 2003).

Castrated piglets licked and chewed less than their littermates, which implies reduced oral exploration. This may be interpreted as an indication of pain (Hay et al., 2003).

Hay et al. (2003) also observed increased walking in castrated piglets throughout their study, which can be interpreted as increased restlessness. Increased restlessness has been observed in animals suffering from pain and can be seen as an adaptation designed to stop other animals from inflicting more pain (Molony and Kent, 1997; Mellor et al., 2000). Llamas Moya et al. (2008) on the other hand found in their study that animals in pain avoid behaviors such as walking in order to minimize pain.

Playing behavior is considered as a positive indication of animal welfare (Blackshaw et al., 1997). Based on this, it would be expected that castrated piglets display less playing behavior. The study of Hay et al. (2003) however, found inconsistencies in playing behavior. Castrated piglets played less frequent than their littermates during the day, but more frequent during the night. A possible explanation is that playing in general did not occur frequently in that study.

Piglets were 5-9 days old in the study while playing behavior is displayed the most on day 21-25 (Blackshaw et al., 1997).

Isolated and desynchronized behavior was observed more in castrated piglets on the day of castration compared to their littermates (Hay et al., 2003). A plausible explanation is that piglets try to protect themselves that way in order to avoid more pain.

All described behavioral changes can be brought down to 2 reasons. The first one is that piglets avoid certain behaviors and contacts that can aggravate the pain; the second one is that they display certain behaviors that are thought to create analgesic effects (Llamas Moya et al., 2008).

"Non-specific" behaviors		
Suckling	Teat in the mouth. Vigorous rhythmic suckling movements.	
Udder massage	Nose in contact with the udder, leaning against it. Ample and rhythmic up and	
	down head movements.	
Teat seeking	Attempts to find a teat by walking and pushing other piglets, while most of	
	the others are suckling.	
Nosing	The snout is close to or in contact with a substrate or a pen-mate. Snout	
	movements may be observed.	
Chewing	Nibbling at littermates (ears, tail or foot, etc.) or substrates.	
Licking	Rubbing the tongue over littermates, floor or pen walls.	
Playing	Head shaking, springing (sudden jumping or leaping), running with vertical	
	and horizontal bouncy movements. Can involve partners (gentle nudging or	
	pushing, mounting, chasing).	
Aggression	Forceful fighting, pushing with the head or biting littermates in a violent	
	manner.	
Walking	Slowly moving forward with one leg at a time.	
Running	Trot or gallop without sudden change in direction or speed.	
Sleeping	Lying down, eyes closed.	
Pain-castration re	lated behaviors	
Huddled up	Lying with at least 3 legs tucked under the body.	
Trembling	Shivering as with cold. The animal may be lying, sitting or standing.	
Spasms	Quick and involuntary contractions of the muscles under the skin, of a leg.	
Scratching	Scratching the rump by rubbing it against the floor or the pen walls.	

 Table 1.1 Description of behavior of piglets according to Hay et al.(2003).

Tail wagging Tail's movements from side to side or up and down.

Postures						
Lateral lying	ateral lying Motionless; body weight supported by side. Shoulder in contact with floor.					
Ventral lying	Motionless; body weight supported by belly. Sternum in contact with floor.					
Sitting	Motionless; body weight supported by hind-quarters and front legs.					
Standing	Motionless; body weight supported by the 4 legs.					
Kneeling	neeling Motionless; body weight supported by front carpal joints and hind legs.					
Social cohesion						
Isolated	Apart from other piglets, alone or with one pen-mate at the most. A distance					
	of at least 40 cm (about the width of two piglets) separates the animal from					
	the closest group of littermates.					

# Growth rate and mortality

Hay et al. (2003) studied castrated piglets in comparison with non-castrated ones and weighed the animals 2 times a day during the first 4 days of the study. Although castrated piglets spent less time suckling and massaging the udder the first few hours after castration, there was no significant effect in weight gain during 4 days after castration (5-9 days of age) between castrated and non-castrated piglets.

Other studies (Kielly et al., 1999; McGlone et al., 1993) found that growth of piglets varied according to the age when they were castrated. Piglets that were castrated at 1-3 days of age showed signs of growth depression. This could be due to the fact that at this age teat order was not yet established and therefore castrated piglets were disadvantaged. Piglets that were castrated at a later age did not have such a growth depression and they did not show any difference in weight gain compared to non-castrated piglets. At weaning, any possible difference in weight gain had disappeared between different treatments in those studies.

# Tail docking

### Reason

Tail biting is a problem of welfare in pigs and can lead to negative economic consequences. To prevent tail biting, tails are often docked (Breuer et al., 2005) although it is routinely prohibited by European and Belgian law. Farmers may only dock tails if tail biting cannot be solved or prevented by adapting farm management (Royal Decree of 17/12/2008 concerning the permitted interventions on vertebrates). Although tail biting has a negative influence on

pig welfare, tail docking is also an issue of animal welfare since it causes acute trauma and pain. Several studies (Noonan et al., 1994; Prunier et al., 2005; Sutherland et al., 2008) have demonstrated that this procedure causes acute stress according to physiological and behavioral parameters.

Tail biting is an abnormal behavior since it rarely occurs in natural conditions; it is a form of aggression that appears in modern pig industry (Sutherland et al., 2008). It also seems to be a multifactorial problem: housing, feed management, stocking density, gender, genetics, tail length, lack of substrate, bad ventilation, temperature fluctuations and draughts all have an influence (Sutherland et al., 2008). Low health status can also provoke biting behavior (Moinard et al., 2003). According to Hunter et al. (2001) the most important influencing factor is whether or not tails are docked. Houpt (2005) indicated the lack of oral stimulation as the most important cause. In a natural environment, pigs spend about 7 hours per day on rooting. When held in other conditions, e.g. confined in pens, it is difficult to express this behavior in the same proportion. Boredom will make pigs nibble each other's tail. Regular nibbling will cause the tail to bleed and the blood will attract other pigs to bite the tail even more (Houpt, 2005). As a result, wounds are formed in a way that the tail and even the spine can be damaged. Deeper infections, abscesses, paralysis, pyemia and even death can occur. Other than the obvious animal welfare problem, there is also a financial loss for the farmer caused by a decreased weight gain and the rejection of carcasses in the slaughter house (Moinard et al., 2003). The severity of the biting problem depends on the intensity of the biting behavior, the number of pigs participating in the biting and the farmer's way of intervening with the problem. A bleeding tail often stimulates the interest and therefore also the further biting behavior of pigs (Taylor et al., 2010).

Environmental enrichment can be used to prevent tail biting. The enrichment objects should encourage a lasting interest, not just an initial interest that quickly declines. These objects should preferably be ingestible, destroyable and not rootable. Objects that were initially attractive because of their novelty but wherein pigs lost interest later on were not that helpful (Van de Weerd et al., 2005). The most applied method to prevent tail biting on intensive pig farms is still tail docking. This approach eliminates the target but not the cause so that the pigs might focus on another target like ears and flanks (Houpt, 2005).

#### Method

Tail docking occurs directly or some days after birth with a regular bistouri knife, scissors or a heated knife and mostly without anesthesia or analgesia. When a heated knife is used, tails are snipped off and the wound is sealed at the same time. The length of the removed tail varies from a little top to almost the complete tail where the tail remainder is reduced up to approximately 1.5 cm. Piglet's tails are usually cut shorter in case of biting problems (Chermat, 2006). According to Belgian law, amputation of the tail is only allowed if it is not applied routinely and if tail biting cannot be prevented by adapting farm management. When pigs are older than 7 days, the procedure can only be carried out by a veterinarian and anesthesia combined with long term analgesia should be used (Royal Decree of 17/12/2008 concerning the permitted operations on vertebrates).

#### Behavioral effects

It is known that tail docking is painful since the peripheral nerves in the tip of the tail are already fully developed in newborn piglets (Simonsen et al., 1991). Different behaviors can be observed after tail docking such as restlessness, stamping the paws, headshaking, vocalizations, tail wagging and jamming the tail between the legs. Most of these behaviors like tail wagging, jamming the tail between the legs and vocalizations are already shown in the first minute after tail docking (Sutherland et al., 2008). Tail wagging can be a consequence of irritation of the skin because the tail is damaged. This behavior therefore is exhibited both after tail docking and tail biting (Kiley-Worthington, 1976).

Noonan et al. (1996) observed suckling behavior right after tail docking in puppies. This may be explained by the fact that endorphins are released during suckling, which have an analgesic effect. This reason can also be applied on piglets. Another study (Sutherland et al., 2008) noticed that piglets scratched their hindquarters on the ground ('scooting') after tail docking to try and mitigate the pain.

#### Growth rate and mortality

Torrey et al. (2009) found no influence of tail docking on growth rate during the first 14 days after birth. In that study, piglets with the lowest and highest birth weights were removed from the study which may have influenced possible differences in growth rate.

# Teeth clipping/grinding

#### Reason

Piglets are born with 8 sharp teeth; 4 canines and 4 incisors (Brown et al., 1996). With these teeth, piglets can injure littermates and the mother sow (Barnett, 2007). From the first day after they are born, piglets establish a teat order. Competition is the highest the first hours

after birth and diminishes when the teat order becomes more stable. Piglets can wound each other during this competition for a good teat when they bite littermates sideways on the snout (Fraser, 1975). These bites can result in serious injuries with a risk of infection. Piglets can also hurt the udder of the sow with their sharp teeth (Brookes and Lean, 1993). To prevent these injuries to some extent, canines are often shortened by farmers (Brown et al., 1996). Prunier et al. (2005) investigated plasma metabolites and stress hormones and found that shortening teeth is less stressful if piglets are only 1 day old. Based on that study, teeth should be shortened as fast as possible after birth.

Shortening teeth of piglets cannot be carried out routinely in Belgium, and if so, only during the first week of life. Farmers can only shorten canines when there are injuries at the udder of the sow or at the ears or tails of other piglets, and when changes in management cannot remedy the problem. If piglets are older than 7 days, the procedure can only be performed under anesthesia and with long term analgesia and it should be done by a veterinarian (Royal Decree of 17/12/2008 concerning the permitted operations on vertebrates).

Brown et al. (1996) investigated the consequences of shortening piglets' canines in outdoor systems and observed no damage to sows' udders, both when piglets had intact and shortened teeth. Brookes and Lean (1993) and Hutter et al. (1994) did observe less damage to udders when piglets' teeth were shortened as opposed to piglets with intact teeth. The different results of these studies can be explained by the fact that sows in outdoor systems can escape more easily from their piglets.

#### Method

Most farmers shorten piglets' teeth by clipping the tips of the canines or grinding them (Barnett, 2007). Teeth can be shortened completely (to the gums) or partially (just the tips) (Weary and Fraser, 1999). Grinding teeth takes more time than clipping them (Hutter et al., 1994; Lewis et al., 2005). This means that piglets need to be handled longer, which can result in more stress (Lewis et al., 2005). On the other hand, clipping the teeth can cause injury to the gums or fragmentation of the teeth which leaves sharp bits of tooth behind. These sharp pieces can lead to wounds of the mouth and tongue that can get infected. Lewis et al. (2005) observed more injuries when piglets' teeth were clipped and Hay et al. (2004) found more negative effects when teeth were clipped like fragmentation, bleeding, infection of the teeth clipping is more painful than teeth grinding. Gallois et al. (2005) observed less biting behavior in pigs which teeth were clipped. The reason for this is that biting increases the

pressure on the teeth which worsens the pain. Gallois et al. (2005) therefore concluded that decreased biting behavior towards other piglets can be seen as an advantage of teeth clipping but damage to the tooth itself is an important disadvantage. According to Hay et al. (2004), shortening canines, whichever technique is used, leads to teeth lesions and intense pain.

# Behavioral effects

Right after teeth clipping, piglets chewed more frequently without having anything in their mouth compared to piglets with intact teeth (Noonan et al., 1994; Lewis et al., 2005). Noonan et al. (1994) reported that this pointless chewing was due to shortening of the teeth and the irritation, pain and discomfort it causes as a result of remaining pieces of tooth or blood in the mouth. This behavior only occurred right after the procedure of teeth shortening (Lewis et al., 2005).

Lewis et al. (2005) looked at the behavior of piglets with clipped teeth, ground teeth and intact teeth. They found that piglets with shortened teeth (clipped or ground) were more inactive (standing, sitting, lying down with eyes open) and less active (suckling, playing, walking) than piglets with intact teeth. Piglets with clipped teeth slept more than piglets with intact or ground teeth. According to Mullington et al. (2000), sleeping can be a consequence of the activation of the immune system as a result of infection. These authors stated that inflammatory cytokines are found to be sleep-promoting factors. Prolonged sleeping can be seen as an indicator of infection, and is used by animals as both a response and a combat to infection (Lewis et al., 2005). Prolonged sleeping in this case can thus be an indication of infection as a result of injuries caused by teeth clipping.

Immediately after the procedure, piglets with ground teeth visited the udder of the sow more frequently than piglets with clipped or intact teeth (Lewis et al., 2005). Piglets with ground teeth appeared to have lower skin temperature which made them seek the warmth of the sow. The lowered body temperature may be explained by the fact that grinding is possibly more stressful than clipping because of the noise and the warmth produced during grinding and the additional time needed for completing the procedure resulting in more heat loss (Lewis et al., 2005).

### Growth rate and mortality

Gallois et al. (2005) and Lewis et al. (2005) found no differences in growth rate between piglets with intact, clipped or ground teeth. However, Gallois et al. (2005) observed that piglets with deep wounds as a consequence of the teeth shortening procedure grew slower.

This conclusion that growth was mainly influenced by pain and injuries caused by the procedure was already stated by Robert et al. (1995) and Weary and Fraser (1999).

Another way growth can be influenced is by the possible loss of advantage during teat competition when piglets' teeth are shortened. Piglets with intact teeth can use their teeth to push away other piglets and conquer a good nipple (Robert et al., 1995). Weary and Fraser (1999) confirmed this, finding that piglets with intact teeth grew better than piglets with clipped teeth in the first week of lactation. They also found that piglets with partially clipped teeth grew better than piglets with completely clipped teeth. This is probably because partially clipped teeth still gave an advantage in teat competition compared to completely clipped teeth.

### Iron injection

#### Reason

When piglets are born, the blood level of iron is low. The concentration of iron diminishes strongly during the first 10-14 days of life. The milk of the mother sow only provides them with approximately 1 mg a day, while the daily need of a piglet is 7-8 mg (Loncke et al., 2008). When they do not receive enough iron, piglets will develop anemia. Symptoms of anemia are lethargy, increased sensitivity to infectious diseases and delayed growth (Kegley et al., 2002). Therefore it is important that suckling piglets receive enough iron from an external source to maintain their hemoglobin concentration (Brown et al., 1996).

#### Method

When piglets are kept in a natural environment, they can usually obtain enough iron from the soil (Loncke et al., 2008). Brown et al. (1996) confirmed that piglets can meet the need for iron when kept in an outdoor system, at least when the soil contains enough iron. On intensive pig farms, piglets are kept indoor so they cannot obtain iron from the soil and they have an increased risk on anemia. Therefore, piglets need to receive iron within the first 3 days of life to satisfy their need until they can eat feed which contains enough iron (Barnett, 2007). An injection with iron is the most common used method and can be given in the neck or in the ham (Robert et al., 1983). Injection of iron can also have negative side-effects. It costs time and money and when hygiene is not taken into account, abscesses can develop which is detrimental for health and growth of the piglets. When the injection is given in the muscles of the ham, spots on the ham can occur even after slaughter, which means an economical loss.

Too much iron can sometimes also cause death, especially in piglets with a shortage of vitamin E (Brown et al., 1996).

Iron can also be administered orally, shortly after birth (Schweigert et al., 2000). Oral administration is not invasive which makes it possibly less stressful for newborn piglets than an injection (Kegley et al., 2002). A disadvantage of oral administration is the fact that it is unsure whether piglets ingest enough iron (Marchant-Forde et al., 2009). Another aspect is the bioavailability of the iron in piglet's body. The bioavailability of iron ingested orally depends on what it is bound to and to the timing of delivery in relation to the closing of the gut to the transport of large molecules (Marchant-Forde et al., 2009). Newborn piglets can absorb large intact organic molecules from the gut by means of pinocytosis. This is only possible the first few hours after birth, until the closing of the gut. This suggests that iron absorption would be reduced if oral administration is postponed too long.

#### Growth rate and mortality

Iron administration is necessary in order to prevent the piglets from getting anemic. When the hemoglobin concentration did not drop below 6g/100ml, growth and health status were not affected (Smith, 1990). Anemic piglets were more prone to diseases, grew more slowly, were lethargic and had a higher mortality rate (Brown et al., 1996).

#### Vaccination

Next to iron injection, piglets also receive other injections like vaccination against *Mycoplasma hyopneumoniae* and an injection of antibiotics. These injections may also imply a stressful experience for the piglets. Although it is recognized that needle injections can be painful, hardly any investigation has been conducted (Marchant-Forde et al., 2009). Research in human medicine however has made it clear that needle injections can be painful (Goodenough et al., 1999; Uman et al., 2008).

#### Ear tagging (identification)

#### Reason

Every pig owner in Belgium, farmers as well as private persons, must register and identify his animals by means of an ear tag. In a central system called Sanitel, data of all pigs in the country are stored so that every pig can be traced back during its entire lifetime and after slaughtering. This system is designed to ensure food safety and traceability. The code on an ear tag consist of the country code (e.g. BE), a pig herd code and a 6 digit number unique for each pig. Each piglet should get an ear tag before weaning and when a pig is moved to a farmer other than its birth farm, it should receive a new, additional ear tag.

### Method

An ear tag consists of 2 parts; one part with a sharp pin and the other with a hole in the middle. The two parts are placed in a special device, they are placed at opposite sides of the ear and then the parts are clicked together through the ear so the ear tag is attached to the ear .

### Behavioral effects

Marchant-Forde et al. (2009) compared ear tagging with ear notching, a different method of identification. Based on vocal measurements they concluded that ear tagging is not pain free. After application of the tag, piglets shook their head and scratched the tagged ear with their paws (Faucitano and Schaefer, 2008).

### Growth rate and mortality

No studies have been conducted yet on the effect of ear tagging on growth rate and mortality of piglets.

## Ethical reflections on painful procedures

Animal welfare criteria were described by the Farm Animal Welfare Council (1979) in five freedoms:

- Free from thirst and hunger;
- Free from discomfort;
- Free from pain, injury and disease;
- Free to express normal behavior;
- Free from fear and distress.

Inflicting a painful procedure, like the ones described above, on an animal can therefore be seen as a violation of animal welfare. However, to place things in a more complete context, the principle of proportionality should also be considered. This means that unavoidable harm and distress caused to the animal should be in proportion to the benefit for the animal that is to be expected from the treatment (Cazaux, 2001). Taking this into account, not all pain and other forms of stress should be considered as a violation of welfare (Lips, 2004). Tail docking

is a procedure that is performed to prevent tail biting. Tail biting can lead to serious injuries and therefore a serious deterioration of pig welfare. It can be said that tail docking is in proportion to the benefit that the treatment gives the animal (no tail biting, no wounds or injuries); although it should also be considered that tail docking is a multifactorial problem and that other causes should be handled before deciding to dock tails. Tail docking is often an 'easy' way to try and solve a biting problem but it is not a solution. It treats the symptoms but does not eliminate the problem (Bracke et al., 2004; Moinard et al., 2003; Paul et al., 2007). In most cases the biting shifts from the tails to the ears or flanks when tails are docked. It is even more important when trying to solve a biting problem, to look at the management of the farm and the production system on its own. Climate factors (e.g. ventilation) often contribute, as well as boredom because of too little environmental enrichment, too little taste experience with the feed, high stocking densities... (Van de Perre, 2011). All these factors are as important, if not more important, to work on.

The same line of thought can be followed for teeth clipping or grinding. The aim of the procedure is to avoid wounds to piglets' heads and the sow's udder and can be seen as proportional to the benefits, especially the grinding. The proportionality of teeth clipping can be discussed because this technique causes wounds to the gums and the mouth, so it is questionable if the benefits outweigh the disadvantages of the technique.

In the case of injections, it is obvious that the pain of this minor intervention does outweigh the benefits obtained by the procedure. Anemia is prevented by iron injection, and diseases are prevented by vaccination.

With castration on the other hand, there is no discussion on the proportionality of the pain of the procedure, because the procedure brings no benefits to the animals. Castration is only carried out because it benefits humans. Even the argument that stress and injuries can occur because of fighting on a sexual mature age in grouped housing, is not justified because many animals are slaughtered before that age is reached (Lips, 2004).

### Avoiding pain via anesthetics: underlying mechanisms and alternatives

# Underlying mechanisms of pain

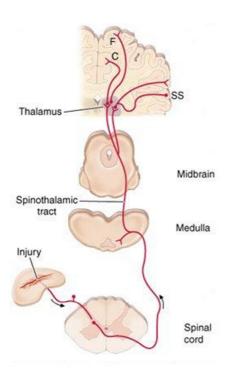
There are 3 main manifestations of pain. Pain is an essential physiological protective mechanism, acting as an alarm system that notifies the body of the presence of some dangerous, noxious stimulus in the environment. Also, once tissue damage has occurred, pain is a promotor of repair by creating a region of localized hypersensitivity (known as

inflammatory pain) surrounding the injured part. This hypersensitivity minimizes movement of, or contact with, the injury until healing has occurred. Finally, there is so-called neuropathic pain, which does not seem to have any beneficial function but results from disturbances in neural and non-neural cells that provoke maladaptive changes in the neurons of the sensory system. This can cause spontaneous, persistent pain and severe hypersensitivity (Basbaum and Woolf, 1999).

Nociceptors are free nerve endings of primary sensory nerves disseminated throughout the skin, muscles, joints and organ walls (Calvino and Grilo, 2006) which have high thresholds requiring noxious stimuli to activate them and which produce pain (Basbaum and Woolf, 1999). There are several classes of nerve fibers that transmit information about the type of painful stimulus being experienced. Thinly myelinated Aδ fibers, for example, respond to changes in temperature and to mechanical stimuli. The other major class of nociceptors are C-fibers or C mechano-heat receptors which express a sensation of burning. Because these fibers are unmyelinated, they are relatively slowly conducting information. Following a painful stimulus, if sufficient numbers of a particular type, or types, of nociceptor are activated, an afferent cascade will be created. The afferent volley travels along the peripheral nociceptor and enters the spinal cord via the dorsal horn. Within the dorsal horn, the terminal of the afferent nociceptor synapses with a dorsal horn neuron, and depending on the intensity of stimulation this may be sufficient to produce a postsynaptic output (Figure 1.2) (Brooks and Tracey, 2005).

An acute stimulus will trigger a series of events leading to excitatory pain signals reaching the brain via the spinal cord. When the stimulus is of short duration, so is the neuronal response. However, when a longer, more chronic stimulus exists, sensitization may occur at either the peripheral and/or the central level (Brooks and Tracey, 2005). Sensitization means that there can be a reduction in the threshold for activation, an increase in the response to a given stimulus, or the appearance of spontaneous activity (Besson, 1999). Hyperexcitability of peripheral nociceptors may occur after localized inflammation in the tissues, and this may cause exaggerated responses to normally painful stimuli. This is called primary hyperalgesia. Then again, sensitization may occur at the level of the dorsal horn neuron, so-called central sensitization. Amplification mechanisms, which are still not fully understood, then enable peripheral neurons which are not normally associated with pain to evoke painful sensations. Such centrally mediated sensitization may explain the event of secondary hyperalgesia,

whereby mechanical stimulation around the initial injury site (i.e. in normal skin) inflicts pain (Brooks and Tracey, 2005).



**Figure 1.2** Transmission system for nociceptive messages. Noxious stimuli activate the sensitive peripheral ending of the primary afferent nociceptor. The message is then transmitted over the peripheral nerve to the spinal cord, where it synapses with cells of origin of the major ascending pain pathway, the spinothalamic tract (Fauci et al., s.a.).

### Anesthetics

Anesthetic techniques can be divided in local anesthesia and general anesthesia (loss of conscience). Furthermore, anesthetics can be administered through injection, inhalation or by nose spray. The great advantage of anesthesia is that pain during painful procedures is avoided, resulting in improved animal welfare (EFSA, 2004). In the EU, the use of anesthetics is restricted to veterinarians. Also, veterinary medicines for animals destined for human consumption are subjected to the Maximum Residue Limits (MRL) guideline. A product can only be used if it is on the list of products for which an MRL is registered (List I) for a certain animal species (for pigs this is the case for azaperone and flunixine), or on the list of products that are not subjected to MRL's (List II) for a certain animal species (aspirin, ketamine, ketoprofen, xylazine and adrenalin for pigs) (European Medicines Agency, 2008).

We will further elaborate on anesthesia suited for piglet castration because these anesthetics may also be applicable to other painful procedures in piglets.

# Local anesthesia

### Injection

Usually a 0.5, 1.0 or 2% lidocaine concentration is used during castration. Concerning the MRL guideline, lidocaine is on list II but only for horses. Lidocaine, not registered for use in pigs, can be injected in the testicle, in the scrotum (subcutaneous administration at the site of injection) or in the spermatic cord although injection in the spermatic cord is technically more difficult. Moreover, lidocaine can immerse directly in the blood flow which can lead to inadequate anesthesia and even toxic symptoms in young piglets. After administering in the testicles, lidocaine can diffuse into the spermatic cords in about 10 minutes (Ranheim and Haga, 2006). A lidocaine injection in the testicles reduces pain related screaming on the moment of castration (White et al., 1995; Marx et al., 2003) and decreases ACTH and cortisol concentrations (Prunier et al., 2002). Zankl et al. (2007) on the other hand, found that piglets castrated under local anesthesia (with procainehydrochloride, procainhydrochloride + epinephrine or lidocainehydrochloride) had similar or even higher cortisol levels compared with piglets castrated without anesthesia. These authors also found no effect of local anesthesia during castration on the healing process of the wound. White et al. (1995) demonstrated that lidocaine reduced the number of high frequency screams and decreased heart rate during the tearing of the spermatic cord. In the study of White et al. (1995), there was also a decrease of blood pressure during and after castration when lidocaine was used. Ranheim and Haga (2006) discovered that castration with an injection of lidocaine in the testicles or in the spermatic cord is less painful than castration without local anesthesia. Their findings were based on heart rate measurements, mean arterial pressure and electroencephalography (EEG) and they found no difference in analgesia between an intratesticular injection and an injection in the spermatic cord.

Ranheim et al. (2005) used radioactive markers to demonstrate that lidocaine concentration (2,5  $\mu$ Ci/ml) in the testicles and the spermatic cord was the highest 3 minutes after intratesticular injection. Therefore they recommended an exposure time of 3 minutes. Bupivacaine, a long working local anesthetic, was tested as an alternative for lidocaine because of its longer effect. But the induction of bupivacaine is slower, it is also not registered

for use in pigs in the EU (EFSA, 2004) and it is more toxic for the heart than lidocaine (Udelsmann et al., 2008).

Procaine is a local anesthetic that was frequently used in the past, but is now replaced by other local anesthetics such as lidocaine that acts faster and longer. Lidocaine also causes less side-effects and diffuses more easily in the tissue (Ranheim and Haga, 2006).

When castration is performed under anesthesia, piglets have to be picked up twice (for injection and -after an exposure time- for castration) which causes extra stress for piglets and demands more labor. Moreover, the anesthetic injections in the testicles or in the spermatic cord are painful.

### General anesthesia

#### Injection

General anesthesia in pigs can be obtained by a combination of ketamine and azaperone. This combination has several disadvantages however. Animals still make aversive movements during castration, although these are reduced compared with castration without anesthesia. Poor healing of the wound and mortality occur more often than in control groups where no anesthesia is used; blood pressure drops and coordination decreases when anesthesia wears off. This can cause piglets to get beneath the sow and be crushed by the sow (McGlone and Hellman, 1988). Lahrmann (2006) only found hypothermia as a disadvantageous side-effect when using ketamine and azaperone for anesthesia. Azaperone (without ketamine) only causes sedation in pigs, no analgesia. It stimulates the respiratory system and decreases arterial blood pressure as a result of vasodilatation after intramuscular injection. This vasodilatation also results in loss of body warmth which can cause hypothermia. The working time of azaperone can be 2-3 hours (Axiak et al., 2007), which keeps the piglets from returning to their normal behavior quickly. Because of all the above reasons, the use of azaperone alone is not recommended for this purpose.

#### Inhalation

General anesthesia through inhalation is fast, causes a good muscle relaxation and loss of consciousness. The advantage of inhalation anesthesia compared to injection is that the animals can be castrated or treated immediately after induction. Induction time is short and piglets don't have to be picked up twice as is necessary with injection anesthesia.

In the past, several inhalation anesthetics for piglets have been tested, e.g. isoflurane, halothane, sevoflurane and carbon dioxide  $(CO_2)$ . The use of isoflurane, halothane and sevoflurane is not recommended when there is no system for gas removal. An extra problem is that isoflurane, halothane and sevoflurane may cause hyperthermia in some stresssusceptible pig breeds like the Piétrain whose influence is very much present in the Belgian pig population (Abbott Laboratories, 2001; EFSA, 2004). The halothane gene is associated with the possible development of the porcine stress syndrome (PSS). This syndrome is related to several symptoms like trembling, muscle contractions and an increased body temperature. PSS can be evoked by inhalation of halothane (Geers et al., 1992; Velarde et al., 2007). The economical and practical aspects of halothane use were investigated by Jaggin et al. (2001). Time needed for each piglet castrated was one minute longer with anesthesia  $(2,3 \pm 0,3 \text{ min})$ than without anesthesia  $(1,3 \pm 0,4 \text{ min})$ . Emissions of halothane in the environment were generally below 5 ppm. Kohler et al. (1998) compared an anesthetic technique with 80% CO<sub>2</sub> and 20% O<sub>2</sub> with halothane anesthesia. Their study demonstrated that recovery was smooth for both groups and that all piglets regained consciousness within 2 minutes. Meanwhile, halothane is no longer on the European market (EFSA, 2004).

Isoflurane is well tolerated by a lot of species like mammals and birds (Hodgson, 2007). In a study of Walker et al. (2004), a special anesthetic device was tested with a respiration bag and a mask with gas removal system to prevent gas from being released from the system and inhaled by the person performing the anesthesia. Both the administering of isoflurane and a combination of isoflurane and N<sub>2</sub>O was tested. Eyelid reflex disappeared after 36,5 seconds (average) and mean induction time was 123 seconds for the combination of isoflurane and N<sub>2</sub>O. The authors stated that the use of isoflurane or the combination of isoflurane and N<sub>2</sub>O was safe, quick and reliable if there was a system to prevent isoflurane from being released into the environment (EFSA, 2004).

Sevoflurane is mostly used for mask induction in small and exotic animals (Holski and kretz, 1999; Lapin et al., 1999; Cravero and Rice, 2001; Hodgson, 2007). A comparative study on isoflurane and sevoflurane was performed by Hodgson (2007). Piglets (6-10 days old) were anesthetized with isoflurane or sevoflurane and then castrated. An inhaler suitable for liquid anesthetic injection was used. Induction of anesthesia was easily achieved with both sevoflurane and isoflurane. Piglets were placed in a box after castration, where they lay still for a short while and subsequently rolled to a sternal position. After that, they stood up again. They were able to move well afterwards, without too much balance problems and there were no surgical problems or mortality due to the treatments. Induction times were shorter (44,0 s  $\pm$ 

7,5 for isoflurane vs. 47,5 s  $\pm$  8,7 for sevoflurane) but recovery periods longer for isoflurane (140,6 s  $\pm$  51 vs. 122,5 s  $\pm$  43 for sevoflurane) (Hodgson, 2007). Another study of Hodgson (2006) on the use of isoflurane anesthesia determined a recovery period of 122 s  $\pm$  44.

Carbon dioxide  $(CO_2)$  is used for stunning in slaughter houses and as an anesthetic or euthanasia product in laboratories (Forslid, 1987; Ring et al., 1988; EU Council Directive, 1993; Martoft et al., 2001). For slaughtering pigs, only high concentrations (> 80% CO<sub>2</sub>) are used for anesthesia (Kohler et al., 1998). These high concentrations are recommended to keep the aversion period as short as possible (Nowak et al., 2007). Certain concentration dependent reactions, like restlessness and hyperventilation, were observed during induction with CO<sub>2</sub> (Kohler et al., 1998). These authors concluded that general anesthesia with CO<sub>2</sub> could be induced quickly and safely and that castration could be executed without any response, but that stress was not reduced. Svendsen (2006) on the other hand stated that the aversion before loss of consciousness is compensated by the fact that piglets were under complete anesthesia and analgesia during castration. Gerritzen et al. (2008) observed heavy breathing as the only typical behavior during induction with an anesthetic mixture of 70% CO<sub>2</sub> and 30% O<sub>2</sub>. In the same study it was concluded that anesthetizing piglets with a mixture of 70%  $CO_2$  and 30% O<sub>2</sub> leads to a period of unconsciousness and analgesia long enough to castrate the animals under anesthesia. An important advantage of the CO<sub>2</sub>-method is that there is no system needed for gas removal and that it can be easily implemented on a pig farm (EFSA, 2004). In a well-ventilated stable CO<sub>2</sub> concentrations maintained low when CO<sub>2</sub>-anesthesia was used for piglets.

#### Intranasal anesthesia

Axiak et al. (2007) compared intranasal versus intramuscular administration of a combination of ketamine, climazolam and azaperone. Their study demonstrated that piglets (4-7 days old) that were anesthetized intranasally had a shorter recovery period but that those piglets reacted stronger to castration than piglets that were anesthetized with an intramuscular injection.

### Underlying mechanisms of anesthetic techniques

Because it is not relevant to the present study to elaborate on all anesthetic techniques, only the techniques used in the study will be briefly discussed.

#### CO<sub>2</sub> anesthesia

Respiration is regulated by the medulla oblongata in the brain and is based on arterial pH. The concentration of  $CO_2$  and  $O_2$  in the blood influences pH. When  $CO_2$ -concentration rises, pH drops and lung ventilation will increase. The animal will breathe faster and deeper and, in case of  $CO_2$  anesthesia, inhale even more  $CO_2$ . Because of rapid diffusion of  $CO_2$  in all tissues, an acid pH change occurs throughout the organism; also in the cerebrospinal fluid. The narcosis begins when cerebrospinal fluid pH falls below 7.10 and reaches a maximum when the cerebrospinal fluid pH approaches 6.80. A cerebrospinal fluid pH of 6.80 appears to represent the critical level of cerebral pH associated with lack of responsiveness to painful stimuli (Eisele et al., 1967). Gerritzen et al. (2008) analyzed behavior, EEG and ECG recordings from piglets castrated under  $CO_2$ -anesthesia (70%). Based on the EEG results they found piglets were unconscious in the 70%  $CO_2$ -30%  $O_2$  mixture. Furthermore, none of the piglets used in the experiment displayed a behavioral, EEG or ECG response to castration; which implies insensibility.

In the Netherlands, CO<sub>2</sub>-anesthesia during castration has already been used for a few years. However, some doubts are circulating on the controllability and proper use of the technique. The comment is that there is no way of knowing whether a farmer actually uses the device to anesthetize his piglets before castration or not. Although correct, the same remark can be made for the other available alternatives. The use of analgesics has the same problem as the use of anesthetics: it is difficult to verify whether the products are actually used by the farmer. Also for immunocastration a validated watertight method is not yet available, since testis growth does not correlate for 100% with having vaccinated correctly nor with the risk for boar taint (Neirynck, personal communication).

Farmers nowadays have a choice between 4 alternatives: raising entire males, immunocastration, surgical castration with analgesia and surgical castration with anesthesia. The farmer will choose for the alternative he believes in the most and will therefore be more motivated to use the technique chosen. If a farmer would choose  $CO_2$ -anesthesia, the benefits of the technique (e.g. working in a more peaceful and quiet way, no extra costs for boar taintcontrol in the slaughterline) should convince him to effectively use the technique. It has no use to oblige people to use techniques when they are not convinced to use them.

# Anesthesia through Zoletil<sup>®</sup> and Xyl-M<sup>®</sup>

Zoletil<sup>®</sup> consists of tiletamine and zolazepam. Tiletamine is a dissociative anesthetic that resembles ketamine. Zolazepam is a benzodiazepine with a sedative, anxiolytic and

myorelaxing effect. Zolazepam increases the depression of the central nervous system (CNS) induced by tiletamine. It also enhances relaxation of the muscles and improves recovery after anesthesia. Zoletil<sup>®</sup> induces a state of unconsciousness that is called dissociative because the connections with the brain are selectively breached before causing an anesthetic sensorial blockage. Analgesia is the result of this interruption in conduction of sensorial information to the brain.

The active substance of Xyl-M<sup>®</sup> is xylazine, which is a non-narcotic analgesic. It causes sedation, analgesia and muscle relaxation, and has a depressing effect on the CNS (Veterinary e-Compendium, 2011). The combination of tiletamine and zolazepam with xylazine produces better muscle relaxation and longer duration of anesthesia than tiletamine and zolazepam alone (Lin et al., 1993).

### Analgesia

Several Non-Steroidal Anti-Inflammatory Drugs (NSAIDs) are permitted for the use in piglets. These are sodiumsalicylate (no withdrawal period but discouraged with piglets younger than 4 weeks), metamizol (withdrawal period of 3 days), flunixine (withdrawal period of 24 days), tolfenamic acid (withdrawal period of 3 days) and meloxicam (withdrawal period of 5 days) (Van Beirendonck et al., 2009). Various studies are available on the effects of pre-emptive analgesics. A study on piglets of 6-7 weeks of age showed a reduction of ACTH and cortisol before castration and the day after when piglets were injected with flunixine 15 minutes prior to castration (Prunier et al., 2002). An intravenous injection with butorphanol 30 minutes prior to castration of 8 week old piglets had no effect on the reduction of growth that was observed the day after castration (McGlone et al., 1993). Kluivers-Poodt et al. (2007) compared the behavior of piglets that received different treatments during 4 days after castration. Treatments were unanesthetized castration, castration under local anesthesia (lidocaine) combined with meloxicam, castration with meloxicam and a sham-castration group. Results of that study demonstrated that animals treated with meloxicam showed less pain-related behavior the first days after castration.

# Immunocastration

With immunocastration, male pigs are actively immunized against Gonadotrophic Releasing Hormone (GnRH). GnRH is a neuropeptide produced by the hypothalamus and stimulates the secretion of luteinizing hormone (LH) and follicle stimulating hormone (FSH) by the pituitary. LH and FSH control the production of testicular steroids like testosterone and androstenone. The immunologically active ingredient in the vaccine is a protein comprising a synthetic, incomplete analogue of natural GnRH conjugated to a larger immunogenic carrier protein. The antigen cannot itself bind to the GnRH receptor sites on the pituitary gland due to its size and configuration, and therefore has no direct hormonal activity (Clarke et al., 2008). Antibodies bind to the pig's GnRH, which prevents stimulation of the pituitary gland to produce LH and FSH, which in turn suppresses testicular function and reduces testosterone and androstenone levels. Skatole levels partially depend on the presence of testicular steroids and can therefore also be reduced by the vaccine (Dunshea et al., 2001). The level of boar taint of vaccinated pigs is similar to those of surgically castrated pigs (Giersing et al., 2006). The administration of the product must be repeated with at least 4 weeks between the two injections. The second injection must be given 4-5 weeks before slaughter (Zamaratskaia et al., 2008). The production of testosterone and boar taint only diminishes after the second injection. Pigs can be delivered to the slaughter house 4-8 weeks after the second injection; delivering too early or too late increases again the chance on a higher androstenone level.

Immunocastration has several advantages. Until the second vaccination, immunocastrated pigs grow at a similar rate as entire male pigs (Dunshea et al., 2001). Immunocastrates have higher average daily gain and better feed conversion ratio than barrows (Dunshea et al., 2001; Turkstra et al., 2002). They also have higher average daily feed intake than entire males (Zeng et al., 2002; Oliver et al., 2003; Pauly et al., 2009). Immunocastrated pigs have an overall higher feed efficiency and their carcasses are leaner compared to barrows (Pauly et al., 2009). A disadvantage of the method is that the vaccine is not species-specific. This means that it has the same effect on humans when vaccinated (de Roest et al., 2009). Accidental self-injection can thus cause (temporary) infertility in humans. The consumption of meat from vaccinated

De Roest et al. (2009) looked at the economic picture of immunocastration and mentioned the calculations of Raaflaub et al. (2008). Improvement in feed efficiency compared to barrows would generate a benefit of 4.44 per slaughter pig. Vaccinating the pigs twice was estimated as a cost of 3.65 per pig, a lower yield at slaughtering was estimated as 1.70 per pig and the extra cost of cutting off the testicles in the slaughter house was estimated at 0.30 per pig. However, the cost of boar taint detection was not taken into account. Nevertheless, based on this information, immunocastration would generate an extra cost of 1.21 per pig in comparison to the traditional surgical castration. An important remark is that possible benefits

animals however is harmless to humans and completely safe.

depend on the type of pigs that are used in different countries. When pigs are slaughtered at a high weight (like in Italy), feed conversion rate is high and the increase of feed efficiency as a result of immunocastration will cause a reduction of production costs that is higher than the extra costs of vaccination. When feed efficiency is already high (like in Denmark), a further improvement will not be sufficient to account for the increase in vaccination costs (de Roest et al., 2009).

# **Raising entire males**

Raising entire males excludes surgical castration. Male piglets are not castrated, with the risk of boar taint in the meat. Geneticists are conducting studies to breed animals with lower androstenone levels via genetic selection. The purpose is to identify animals with a lowered genetic capacity to accumulate androstenone in fat while maintaining the normal values of testicular steroids characteristic for entire males. The development of genetic markers to identify these pigs should lead to the selection of pigs with low androstenone levels but with a normal growth (Squires, 2006). Another solution would be to have reliable detection methods for boar taint, so tainted carcasses can be sorted out at the slaughter line.

Entire males consume less feed (1.88 vs. 2.23 kg/day) but are more feed efficient (2.43 vs. 2.69 kg feed/kg gain) than barrows (Pauly et al., 2009). Lower average daily feed intake was confirmed by several studies (Dunshea et al., 2001; EFSA, 2004). The advantages of boars compared to castrates were summarized by de Roest et al. (2009) as follows: boars have superior growth rate up to 13%, boars eat up to 9% less feed, their feed conversion is up to 14% more efficient, and they are generally leaner than castrates by up to 20%. Possible drawbacks are the incidence of boar taint, infrastructural costs for the separate housing of male and female pigs and the fact that entire males may show higher aggressive and sexual activities than castrates which can generate negative welfare issues or economic consequences for farmers if such behavior causes injuries or reduced efficiency (de Roest et al., 2009). The costs and benefits of raising entire males will depend on the percentage of males with boar taint at slaughtering (de Roest et al., 2009).

# Measuring pain: gold standard, how to detect pain in farmed animals?

Because of the lack of a gold standard for pain assessment in animals, a wide range of tools is currently used. According to Bath (1998), measurement of pain has been attempted in several ways, each with their advantages and disadvantages:

- Bio-chemical indicators such as cortisol, endorphins and lymphokines. The problem is that changes in concentrations may result from many causes and they do not measure the perception of pain as such. They do help to underpin the assessment in an objective way.
- Physiological. Blood pressure, heart rate and immunological measurements are similarly not necessarily or solely related to poor animal welfare, but may be used as research tools.
- Pathological. Measurements of lesions, disease incidence or organ changes suffer from the same problems as the previous 2 categories but may be useful in surveys and research.
- Production. The recording of meat, milk, fiber or reproductive performance is of value in that good production cannot occur in the face of very poor welfare, but sub-optimal welfare may not be reflected in lower production figures.
- Behavioral. Well-researched ethograms (normal behavioral repertoire) are essential for this to be meaningful and do give a more objective measurement, even though indirect, of the perception of the animal.
- Extrapolation. This clearly has many dangers, especially when extrapolation is made between very different families or genera. However, some degree of extrapolation is needed to improve our understanding of perceptions.
- Combination. It should be clear that a combination of these ways of assessment is superior to any on its own, and assessments should be combined whenever possible. Ultimately it must be conceded that welfare evaluation is not a tangible measurement but a value judgment.

None of the available tools has yet provided a sensitive, reliable and repeatable method of 'measuring' pain (Vinuela-Fernandez et al., 2007). In order to develop an assessment tool for a particular painful condition, the underlying pathology needs to be identified since different pain responses might be elicited depending on the type of lesion or the type of structures implicated (Vinuela-Fernandez et al., 2007). A description of some assessment tools is given below.

# **Behavior**

Despite many theoretical and practical limitations, observation and careful interpretation of behavior and physical signs remain essentially the only clinically useful means to assess animal responses to injury, according to Hansen (1997). Using behavior to assess pain has the

major advantage of being non-invasive, so any effects of assessment on the animal are limited (Rutherford, 2002). Many studies demonstrated that behavior is the most important indicator of pain (Bateson, 1991; Bath, 1998; Morton and Griffiths, 1985; Rutherford, 2002; Short, 1998). According to Short (1998), animals communicate their pain perception predominantly by behavioral patterns and physiologic changes in the sympathetic nervous and endocrine system. Quantification of behavioral patterns has been used to assess pain caused by procedures normally carried out without any anesthesia or analgesia in lambs, calves and piglets. These studies have identified pain related behaviors that are quantifiable by either measuring the frequency of an event behavior or the duration of a state behavior (Vinuela-Fernandez et al., 2007). Although pain related behavior should be looked at, it must also be noted that injury is routinely accompanied by other stressors that also affect (normal) behavior. Pain-induced alterations of behavior may include loss of normal behaviors, expression of new and uncharacteristic behavior, and expression of behavior designed to limit pain or adapt to loss of function (Hansen, 1997). In human medicine, pain detection is mostly based on (aberrant) behavioral observations. For those reasons, behavior is the most used parameter to assess pain in farm animals (Vinuela-Fernandez et al., 2007). Detailed knowledge of normal behavior and pain behavior of a specific species, combined with a careful judgement of the (possibly aberrant) behavior is an absolute must before the presence or absence of pain in the animal can be interpreted.

Martini et al. (2000) described symptoms of acute and chronic pain in animals:

Symptoms of acute pain:

- avoidance reflex
- vocalization elicited by touch or voluntary movement of the animal
- licking or self mutilation
- scratching
- rolling
- restlessness
- moving reluctantly
- modification of heart- and breathing frequency

Symptoms of chronic pain:

- licking of body parts
- moving reluctantly
- modification of behavior

- altering feed and water intake
- lameness

According to Hansen (1997) an objective methodology is to record rigidly defined behaviors in a present/absent format, because this approach allows the recognition of significant behavioral effects that would have otherwise gone unnoticed. These rigidly defined behaviors are described in an ethogram. An ethogram for piglets, that also contains pain related behavior, was described by Hay et al. (2003) (Table 1.1).

Because behavior is the most used parameter to assess pain in farm animals, as described above, the experiments in this thesis are based on behavioral observations. Observations were carried out by putting the ethogram of Hay et al. (2003) in a present/absent format, and in that way behavior was scored for every individual piglet. Hay et al. (2003) used both a scan sampling method and continuous observations. In the present thesis, 2 methods of behavior observations were also used. With the first method, behavior was observed continuously for a period of 10 minutes. Pens were observed at random within a sequence according to a scan sampling procedure. During each 10 minute lasting observation period, the frequency of each behavior was recorded every minute for each piglet in accordance with a continuous focal sampling procedure (Fraser, 1978). It was possible that a piglet could show a sequence of different behavioral events within a given single minute; however, the number of times each defined behavioral category was displayed was taken into account by calculating the frequency of that behavioral category, i.e. per minute for the total number of observed piglets. All observations were performed by a single observer standing in the central corridor of the pig house. The total observation time per pen, in combination with the number of pens was assumed to be sufficient to obtain an adequate understanding of the treatment effects studied (Hay et al., 2003; McGlone et al., 1993). After evaluation of the used method, it was adapted. Although the first method was not incorrect, there was room for improvement, which was done for following experiments. With the second method, behavior of all barrows in one pen was scored and then the observer moved to the next pen and scored behavior of those piglets; according to a scan sampling method. This was repeated until every pen was scored 30 times. The adapted method has the advantage of covering a wider time range for each pen.

### Vocalization

Animals can only express themselves through behavior and vocalization. Screaming is used to assess pain reaction in babies (Ranger et al., 2007). Weary et al. (1998) investigated the vocal

response to acute pain during unanesthetized piglet castration and showed that an increased number of high frequency screams is a reliable indicator for the pain caused by castration. Marx et al. (2003) distinguished 3 types of vocalizations: grunts, squeals and screams, which increased when a piglet was in pain. Besides the frequency, the intensity of vocalization can also be measured (Jourdan et al., 2001).

#### Zootechnical results (feed conversion, growth rate, sickness sensitivity)

According to Weary et al. (2006) there are commonly three approaches for pain evaluation in animals: (1) physiological response such as plasma cortisol concentrations, (2) behavioral observations and vocalizations, and (3) zootechnical parameters such as feed intake, growth rate, sickness prevalence (recording of lungs, heart and liver lesions during slaughter) and mortality. These zootechnical parameters are measured easiest, but do not reflect however what is going on with the animal at that exact moment. They rather give a summary of what has happened with the animal during the interval between observations (Weary et al., 2006). Furthermore, possible infections of wounds caused by the interventions can also be looked at.

### Physiological and biochemical parameters

Both pain and stress can induce an increase of adrenalin, corticosteroids, glucose, lactate and free fatty acids in plasma. The problem is that changes in concentration can be the result of many reasons, e.g. animal handling, and that not the perception of pain itself is measured (Dawkins, 2003; Weary et al., 2006). Another limitation is the occurrence of a "ceiling-effect" (Mellor et al., 2000; Molony and Kent, 1997).

The electrocardiogram (ECG) was used to assess the changes in frequency and other characteristics related to effectiveness of the heart function (Dubin, 1999). Kluivers-Poodt et al. (2007) used this technique during their study on  $CO_2$ -application during piglet castration in order to evaluate the status of anesthesia, analgesia, aversion and pain.

# **Animal integrity**

Integrity can be described as the wholeness (completeness) and intactness of the animal and its species-specific balance, as well as the capacity to sustain itself in an environment suitable to the species. It therefore implies that the animal is intact or whole (complete), which is an attribute of the animal itself, not just some value being placed on it (Bovenkerk et al., 2002). When placing the whole issue of management practices for piglets in this perspective, it can

be said that castration of piglets as well as tail docking and teeth resection threatens the integrity of the piglet because it affects the 'intactness' of the animal. Moreover, these practices are carried out not for the wellbeing of the animal itself but more for the convenience of humans. Animal integrity is considered by some as an important aspect of welfare (Bovenkerk et al., 2002), as an extension of the classical approach by FAWC, and it can therefore be interesting to consider in the whole discussion on painful procedures for piglets and animal welfare.

## Aim and outline of the thesis

In 2010 different actors of the European pig chain, European retailers and ngo's published a declaration which states that they want to stop unanesthetized piglet castration. As a first step, castration may only be carried out with local or general anesthesia or analgesia starting from 01/01/2012. A second step that is described in the declaration is to abolish surgical castration of piglets ultimately on 01/01/2018. Animal welfare is also gaining consumers' interest and concern. This was demonstrated by several media campaigns of animal welfare organizations over the last few years, which are getting more and more attention of the public. It is acknowledged nowadays that consumer attitudes towards food production methods are not only dependent on an analytical assessment of risk, benefit, economics and nutrition alone. Ethical and moral considerations may also have an influence on establishing the societal acceptability of a particular production process (Frewer et al., 2005). Because piglet castration is a sensitive issue, it is important to explore consumers' opinion on this topic. For that reason the opinion of Flemish consumers on unanesthetized piglet castration and three possible alternatives was investigated in the second chapter. This gives an idea of the public's need to improve animal welfare.

In the second chapter, results demonstrated that castration under anesthesia was the alternative that was most accepted by consumers. Therefore an alternative method, castration under  $CO_2$ -anesthesia was studied in the third chapter. The hypothesis was that piglets will experience less pain and discomfort during and after castration when anesthetized with  $CO_2$  (100%, 25s) prior to castration and thus improving their overall welfare. The analgesic capacities of  $CO_2$  during the procedure were also studied and compared to so-called ideal anesthesia with tiletamine and zolazepam combined with xylazine.

Castration is not the only event that may threaten piglets' welfare. Piglets are subjected to several painful management practices, especially during their first week of life. Tail docking, teeth clipping or grinding, ear tagging, injections and castration of male piglets are the most common procedures. In the fourth chapter, not only castration but painful management procedures in general are looked at. The hypothesis of this chapter was that reducing painful interventions during the first week of life results in better zootechnical performance of the piglets, reduced piglet mortality and that the overall welfare, indicated by behavioral criteria, would improve.

In the fifth chapter, the objectives of chapter 3 (anesthesia) and 4 (all painful management procedures) are combined. The hypothesis was that the bundling of painful management practices (carrying out all procedures on one moment in time), whether or not under  $CO_{2}$ -anesthesia influences positively the behavior and/or production results of piglets. The bundling of these painful events on a later age also has the advantage that the piglets only have to be picked up once (time saved for the farmer) and that the piglets are left alone (not handled) the first days of their life which also gives weaker piglets the chance to make a good start.

Chapter 2: Belgian consumers' opinion on pork consumption concerning alternatives for unanesthetized piglet castration

Chapter redrafted after: Van Beirendonck, S., Driessen, B. and Geers, R. (2012). Belgian consumers' opinion on pork consumption concerning alternatives for unanesthetized piglet castration. Journal of Agricultural and Environmental Ethics, DOI 10.1007/s10806-012-9378-y, in press.

### Abstract

Male piglets in Belgium are still castrated unanesthetized in the first week of life, but animal rights organizations, supermarkets, and some consumers no longer accept this method in terms of animal welfare, and are pushing the pig industry to apply available alternative methods. This major change in pig husbandry will increase production costs without a guarantee for return of investment by consumers. Therefore, it is important to know the opinion of consumers on this matter. A questionnaire was used to collect data from 1018 people through face to face interviewing in Flanders. A chi-square test and Fisher's exact test were used to analyze the data. Results show that in spite of several media campaigns of animal welfare organizations over the past few years, still half of the Flemish respondents were not aware of the problem of unanesthetized piglet castration. However, after being informed, the majority wanted unanesthetized castration to be banned. Although the concern about animal welfare implications was very high, the willingness to pay extra for alternatives was low.

## Introduction

Male pigs reared for meat production have been castrated for centuries to reduce aggressiveness and to improve handling. An additional advantage of castration, and today the most important reason, is the prevention of boar taint. Boar taint is an aversive smell or taste that can be released when heating meat of entire male pigs. Castration can be executed without anesthesia or analgesia within the first week of life (European Commission, 2001), and is detrimental to animal welfare (Van Beirendonck et al. 2011). The reason for exempting piglets during the first week of life from the requirement of anesthesia and analgesia may be associated with the belief that it was presumed for a long time that neonates experience less pain because of the immaturity of their neural development (Anand 1990; Fitzgerald 1994; Andrews and Fitzgerald 1994). However, research in human medicine has indicated that perception of pain in neonates and children is comparable to pain experienced by adults. According to the principle of analogy based on similarities between humans and higher animals, it is adopted that pain is analogous to humans and animals (Hendriksen and Boumans 2006). Previous studies (Taylor et al. 2001; Hay et al. 2003; Henke and Erhardt

2004) observed that castration causes pain to piglets, on the moment of castration but also days after the procedure.

Consumers, organizations, and individuals being concerned with animal welfare and production systems, and supermarkets are pushing the pig industry to search alternatives for unanesthetized piglet castration, which is no longer acceptable for animal welfare. Colruyt, one of the biggest supermarket chains in Belgium, refuses meat from castrated male pigs. They demand that their suppliers deliver only immunocastrated male pigs.

The quest for applicable alternatives is not only a matter of animal welfare, but also depends on the perception of the public. Consumer acceptance on food products resulting from new technologies depends on both processing considerations and quality characteristics associated with the product itself. The concern of the public on new technologies may have as much as an influence in terms of impact on consumer decisions as the quality considerations associated with the product itself (Frewer et al. 1997). It is acknowledged nowadays that consumer attitudes towards food production methods are not only dependent on an analytical assessment of risk, benefit, economics and nutrition alone. Ethical and moral considerations may also have an influence on establishing the societal acceptability of a particular production process (Frewer et al. 2005).

Many consumer studies that are carried out focus on reactions to pork flavor or odor from pigs treated according to different alternatives to unanesthetized surgical castration (Font I Furnols and Oliver 1999). Fewer studies have been done on consumer acceptance regarding the methodology of these alternatives (Vanhonacker et al. 2009). In Flanders, a study has been carried out (Vanhonacker et al. 2009) on consumer acceptance of immunocastration. However, to our knowledge, no study has been carried out yet comparing different alternatives. Therefore, it was the purpose of the present study to investigate consumer's opinion in Flanders, the Dutch speaking part of Belgium, with respect to three different alternatives: castration under anesthesia, immunocastration, and raising entire males.

### Materials and methods

### Research approach and sampling

Survey data were collected through face to face interviewing in Flanders during October and November 2009. A total number of 1018 people (aged 16-80 years) were interrogated on

social events, markets, in railway stations.... All data were collected by the same person. The 5 provinces of Flanders were investigated, and respondents were chosen in such a way as to obtain a sample of wide diversity of the population in terms of gender, age, family size, and living environment (Table 1). Before executing the questionnaire, the places, times ... for sampling were carefully chosen. The questionnaires were taken on different places in each province and both on weekdays and in weekends to obtain a sample of wide diversity, although probably not being completely statistically representative for the total Flemish population. Hence, findings mainly apply within the characteristics of the sample and generalization to the overall population needs critical reflection (Vanhonacker et al. 2009).

Characteristic	Percentage			
Gender				
Male	50.6			
Female	49.4			
Living environment				
Rural	44.8			
Urban	55.1			
Age				
16-25 years	38.5			
26-40 years	31.2			
41-54 years	21.5			
55+ years	8.7			
Profession				
Laborer	13.5			
Clerk	18.1			
Self-employed	9.1			
Student	35.7			
Unemployed	7.3			
Retired	4.7			
Civil servant	6.6			
Housewife/houseman	5.0			
Education level				
High school	34.38			
Higher education (university	33.79			
college)				
University	31.63			
Responsible for buying meat?				
Yes	39.19			
No	60.81			
Personal agricultural background				
None	93.4			
Beef cattle	2.9			
Pigs	0.5			
Fruit	1.5			

Table 2.1 Percentages of socio-demographic characteristics of the survey participants (n = 1018)

Dairy cattle	1.7
Cheese production	0.1
Familial agricultural background	0.1
None	55.2
Beef cattle	44.4
Pigs	0.2
Fruit production	0.1
Dairy cattle	0.1
Family size	
1	8.6
2	19.8
3	20.1
4	26.9
5+	24.5
Average consumption of pork per	
week	
Don't know	0.1
Never	0.6
Rare	19.6
1-2 times a week	61.5
3-4 times a week	17.9
>4 times a week	0.3

# Questionnaire

All aspects to be evaluated by respondents were explained first in an objective manner before questions were formulated, so that the risk for suggesting answers and yeah-saying was minimized. As an example of how things were objectively explained to respondents, the explanation of unanesthetized piglet castration was given as follows: "During unanesthetized piglet castration, the testicles are removed without anesthesia or pain relief. According to the law, the procedure must be carried out before the piglets reach the age of one week. This is a quick and cheap method, but also painful and stressful for the animals." Another example is the description of immunocastration: "The purpose of immunocastration is to make castration unnecessary by stemming the development of the testicles. The injected hormone like substances influence the hormonal system of the pig so that puberty is delayed. This means that the risk of boar taint is reduced strongly, but according to the available knowledge it cannot be guaranteed a 100% that boar taint cannot occur. Hence, a detection method has to be applied in the slaughter line."

Respondents were selected at random, since it was only possible to interview one person at the same time, followed by the next one being available without knowing in advance her/his qualifications, so that these qualifications could not have influenced the selection. In order to know whether or not an interviewed person's background might be associated with her/his content of answering, the sampled population was divided into classes for statistical analysis. After noting their socio-demographic characteristics, respondents were consecutively asked about the issue of unanesthetized castration, the need for alternatives, the acceptability of three proposed alternatives, their preference and their willingness to pay extra (Table 2) for these alternatives. To give respondents an idea of the concept of boar taint, which was unknown for the majority, they were given the chance to smell a skatole solution (0.2 ppm). The alternatives that were proposed to respondents were immunocastration, raising entire males, and castration under anesthesia and analgesia (it was not specified which type of anesthesia would be used). The total interview took about 10 minutes.

### Table 2.2 Questionnaire

# Awareness

Are you aware of male piglets being castrated?

Are you aware of the fact that this happens without anesthesia or analgesia?

Do you know the reason for piglet castration?

## Need for alternatives

Do you think alternatives to unanesthetized piglet castration are necessary?

Do you think unanesthetized piglet castration should be banned?

# Acceptability

Do you think castration with anesthesia and analgesia is an acceptable alternative?

Do you think raising entire males is an acceptable alternative?

Do you think immunocastration is an acceptable alternative?

# Preference

Which of the proposed alternatives do you prefer?

# Willingness to pay extra

Would you be willing to pay an extra price for pork from pigs castrated with anesthesia and analgesia?

Would you be willing to pay an extra price for pork from pigs that were not castrated (entire males)?

Would you be willing to pay an extra price for pork from pigs that were immunocastrated?

### Statistical analysis

All data were inserted in an Excel-file, and percentages were calculated through the proc freq procedure in SAS 9.2 (SAS Inst, Inc, Cary, NC, USA 2008). The influence of different sociodemographic data on the answers to the questions was tested through a chi-square test and where needed a Fisher's exact test. A significance level of 0.05 was used. A statistically significant result means that the variability within one class variable is associated with or explained by the variability within the other class variable. It was assumed that within each socio-demographic group or class all other characteristics were distributed at random, so that confounding effects did not create biases.

# Results

Only the demographic criteria that resulted in significant p-values (p<0.05) are shown in tables.

### Awareness

Of all the respondents, 45.9% stated they were aware of the issue of piglet castration . This was associated with the living environment (p = 0.0422), education level (p = 0.0448), and quantity of pork consumption per week (p = 0.0131) (table 3 and 4).

Of all respondents, 69.8% could not explain the reason for piglet castration. From the 30.2% who claimed to know the reason, 72.0% said it had something to do with odor and/or taste of the meat; 13.1% actually used the term "boar taint." The average consumption of pork was statistically associated with the answer respondents gave to the question concerning the reason for piglet castration (p = 0.0158) (table 3).

		Pork consumption per week			
	Never	Seldom	1-2 times	3-4 times	More
	( <b>n</b> = 6)	(n = 199)	(n = 626)	(n = 183)	(n = 3)
<i>Were you aware of the issue of</i>					
piglet castration before the					
questionnaire?'					
No	16.7	60.8	54.0	49.2	0.0
Yes	83.3	39.2	46.0	50.8	100.0
'Do you know the reason for					
piglet castration?'					
No	50.0	72.4	71.4	63.4	0.0
Yes	50.0	27.6	28.6	36.6	100.0
'Do you think alternatives are					
needed?'					
No	0.0	4.0	2.6	6.6	66.7
Yes	100.0	96.0	97.4	93.4	33.3
'Should unanesthetized castration					
be banned?'					
No	16.7	33.2	25.6	30.1	100.0
Yes	83.3	66.8	74.4	69.9	0.0
'Is anesthetized castration					
acceptable to you?'					
No	16.7	0.0	1.4	2.2	33.3
Yes	83.3	100.0	98.6	97.8	66.7

Table 2.3 Association of pork consumption per week with the answers to several questions (%)

# Need for alternatives

Concerning alternatives for unanesthetized piglet castration, 96.3% of the respondents felt that it was necessary to find an alternative, and 72.0% wanted unanesthetized castration to be banned. Opinion on the need for alternatives was statistically associated with the pork consumption per week (p = 0.0018), and the opinion on a ban for unanesthetized piglet castration was influenced by gender (stronger support for a ban among female respondents; p

= 0.0188), education level ( stronger support for a ban among higher educated respondents; p = 0.0445) and pork consumption per week ( stronger support for a ban among respondents that never eat pork; p = 0.0208) (Table 3 and 4).

'Were you awar	e of the issue of piglet castration?'	Yes	No	
Living environm	Living environment			
	Rural	49.3	50.7	
	Urban	43.0	57.0	
Education				
	High school	40.6	59.4	
	Higher education	49.4	50.6	
	University	47.8	52.2	
'Should unanes	thetized castration be banned?'	Yes	No	
Gender				
	Male	68.7	31.3	
	Female	75.4	24.6	
Education				
	High school	67.4	32.6	
	Higher education	75.3	24.7	
	University	73.6	26.4	

**Table 2.4** Association of living environment, gender and education level with the awareness of piglet castration and the opinion on a ban for unanesthetized castration (%)

# Acceptability of alternatives

Respondents were asked if they found the proposed alternatives acceptable, after being given a brief objective explanation on each of the alternatives. Figure 1 shows that castration under anesthesia had the most positive response, followed by immunocastration, while raising entire males was considered the least acceptable. The acceptance of immunocastration was associated with the gender of respondents (more men than women found this acceptable; p =0.0199) and responsibility for buying meat for the family (more respondents who are not responsible for buying meat found it acceptable; p = 0.0223) (Table 5). The pork consumption per week on the other hand, was associated (p = 0.0033) with the acceptability of castration under anesthesia (Table 3).

"Do you think	t immunocastration is an acceptable	Yes	No
alternative?"			
Gender			
	Male	52.4	47.6
	Female	45.1	54.9
Responsible	for		
buying meat?			
	Yes	44.4	55.6
	No	51.7	48.3
"Are you willin	g to pay an extra price for pork from	Yes	No
immunocastrate	d pigs?"		
Gender			
	Male	17.3	82.7
	Female	25.1	74.9
Family with an			
agricultural	Yes	17.6	82.4
background	No	24.0	76.0
Living environm	ent		
	Rural	24.3	75.7
	Urban	18.5	81.5
"Are you willin	g to pay an extra price for pork from	Yes	No
pigs castrated ur	nder anesthesia?"		
Gender			
	Male	37.1	62.9
	Female	43.7	56.3

**Table 2.5** Association of living environment, gender, responsibility for buying meat and agricultural background with the acceptability and willingness to pay an extra price for alternatives (%)

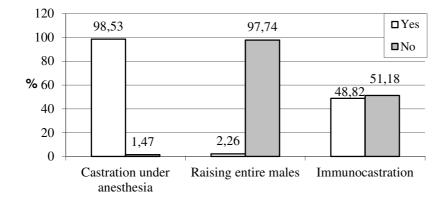
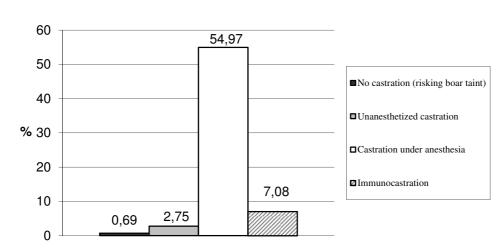


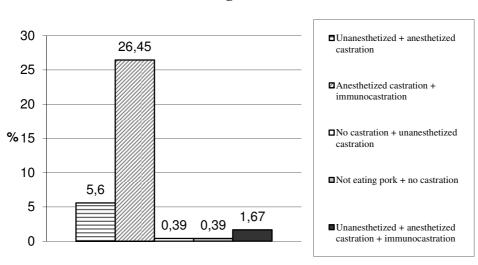
Figure 2.1 Judgment of respondents on the acceptability of proposed alternatives for unanesthetized castration

# Preference

When respondents preferred just one of the alternatives, castration under anesthesia had the most followers. When respondents equally preferred more than one alternative, the combination of castration under anesthesia and immunocastration was the most popular (Figure 2).



#### Figure a



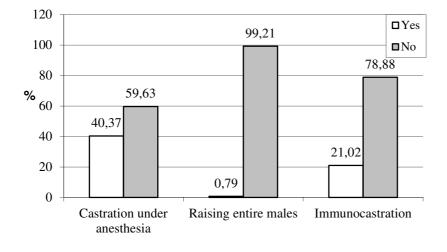
**Figure b** 

**Figure 2.2** Preference (%) on alternatives for unanesthetized piglet castration. a) represents the respondents who preferred just one of the alternatives; b) represents the respondents who equally preferred more than one alternative. Together, figure a and b represent all respondents (100%)

## Willingness to pay extra

Besides acceptability, the willingness to pay an extra price for meat of pigs that were treated with one of the alternatives was questioned. In general, respondents were not willing to pay extra. Willingness to do so was however greatest for pork from pigs castrated under anesthesia (Figure 3). Willingness to pay extra for immunocastration was influenced by (p = 0.0199), agricultural background of family (p = 0.0306) and living environment (p = 0.0242). Of the male respondents, 17.3% claimed to be willing to pay extra, compared to 25.1% of the female respondents. Of the respondents who had family with an agricultural background, 17.6% was willing to pay extra for immunocastration, as opposed to 24.0% for respondents without a familial agricultural background. Respondents living in a rural area were more willing to pay extra than respondents living in the city; 24.3% compared to 18.5%.

For castration under anesthesia, the willingness to pay extra was only associated with gender (p = 0.0306). The same as for immunocastration, female respondents were more willing to pay extra (43.7%) than male respondents (37.1%).



**Figure 2.3** Willingness to pay an extra price for pork when alternatives for unanesthetized piglet castration have been used

# Discussion

It is known that the sensitivity to animal welfare might be subjective and potential solutions vary between countries (Bonneau 1998; Dijksterhuis et al. 2000). Therefore, this survey was carried out in Flanders, the Dutch speaking part of Belgium, to obtain region specific information. Previous studies on single potential alternatives have been executed in Flanders, but to our knowledge none of them have investigated all three potential alternatives questioned in the present study. The three discussed alternatives were considered by Huber-Eicher and Spring (2008) as feasible on a short term. Immunocastration is a method that is already being used in several countries like Australia, New Zealand, Brazil, Russia, and Switzerland (Prunier et al. 2006, Vanhonacker et al. 2009). In Norway, Switzerland, and the Netherlands, surgical castration without anesthesia and/or analgesia is prohibited (Vanhonacker et al. 2009). In Ireland and the United Kingdom, where surgical castration is prohibited by law, entire males are raised and slaughtered at a lower weight. In Spain, Portugal, Greece, and Denmark a part of the male piglet population is also not being castrated, and they are also slaughtered at a lower final weight (EFSA 2004). In 2010 different actors of the European pig chain, European retailers and ngo's published a declaration that states that they want to stop unanesthetized piglet castration. As a first step, castration may only be carried out with long acting local or general anesthesia or analgesia according to established methods, starting from 01/01/2012. A second step is to abolish surgical castration of piglets ultimately on 01/01/2018 (European Declaration on alternatives to surgical castration of pigs, 2011).

It is important to know consumers' opinion, both as consumer and as citizen, because that information is used by lobbyist groups, either from welfare activists or farmers, to steer politicians into the direction they like to be followed. Eventually, the law must be fair, correct, and non-discriminating, for which scientific based analysis might contribute as an independent consultancy. Legislation is not necessary when stakeholders do agree voluntarily on a production system guaranteeing their different individual interests. A solution for which science also is able to provide independent and value-free information. In fact, methods are developed to measure pain in case of castration, debeaking, dehorning, injections, notching, and other interventions applied daily for managing husbandry systems. Hence, society is able to value and to weigh out that information against alternatives to agree on the solution combining the largest possible extent of all interests involved. The main objective of this paper was to find out how scientific knowledge is matched by consumers' opinion on castration of piglets, so that information campaigns might need to be improved to assure societal concern on pork producing methods.

#### Awareness

Striking was the low consumers' awareness regarding piglet castration, i.e., 45.9%, regarding the reason for it (only 30.2% knew why piglets are castrated) and regarding the fact that it takes place without anesthesia or analgesia (only 45.9% was aware of this). Vanhonacker et al. (2009) found that approximately 40% of respondents in Flanders were aware of piglet castration. A possible explanation for this is the fact that increasing urbanization and separation of food production from consumption has resulted in ignorance of modern production methods (Harper and Henson 2001). Although consumers' awareness in general is low, inhabitants of rural areas are slightly more aware than inhabitants of the city.

Only half (45.9%) of the respondents were aware of the lack of anesthesia or analgesia for piglet castration. Although this was influenced by living environment, education level and pork consumption per week, the awareness was in general very low. The fact that this is still the case in spite of several media campaigns, can be explained by the alienation of livestock production (Harper and Henson 2001; De Tavernier et al. 2005) or by "voluntary ignorance"

(Harper and Henson 2001). The latter means that people eat meat and ignore that it originates from animals especially raised for human consumption.

## Acceptability of alternatives and preference

Almost all of the respondents, 98.5%, agreed with the proposition that castration under anesthesia and analgesia is acceptable when pain during and after castration can be treated. Huber-Eicher and Spring (2008) also found the highest agreement for castration under anesthesia (82.4%) compared with immunocastration (52.8%) and raising intact males (41.3%). The authors claimed that this method is the most transparent to the consumer and that it reliably eliminates the risk of boar taint, which can explain the found result.

Few respondents thought of raising entire males as a good alternative. In an investigation of Swiss consumers opinion (Huber-Eicher and Spring 2008), the researchers found a relatively high disagreement for techniques of raising entire males ("no castration, sort out meat with boar taint and destroy it" and "no castration, sort out meat with boar taint and produce specific boar-products"). The results of the present study agree with this. A possible explanation for the low acceptability of entire males can be found in the smelling of the skatole solution when the concept of boar taint was explained at the beginning of the questionnaire. Most respondents found this smell unacceptable and this may have influenced their opinion on raising entire males because boar taint cannot totally be excluded with this alternative.

On immunocastration, opinions were not decisive. About half of the respondents found this unacceptable while the other half thought of this method as acceptable. These results agree with the results of Huber-Eicher and Spring (2008) who found the lowest disagreement for castration under anesthesia (17.6%) and the strongest disagreement for immunocastration (47.2%), "no castration, sort out meat with boar taint and destroy it" (42.4%) and "no castration, sort out meat with boar taint and produce specific boar-products" (40.2%). Vanhonacker et al. (2009) found that 60% of their respondents had a general appreciation for the concept of immunocastration. However, they only investigated immunocastration as an alternative, as opposed to unanesthetized surgical castration.

Font i Furnols et al. (2008) investigated the acceptance of meat from immunocastrated pigs based on odor and flavor, and found that Spanish consumers did not differentiate between cooked meat from immunocastrated pigs, surgically castrated pigs, and female pigs. Vanhonacker et al. (2009) also found that consumer acceptance of immunocastration does not

seem to be a problem for Flemish consumers, and that it was preferred over unanesthetized surgical castration. Results of the present study agree with this.

The acceptance of immunocastration was associated with gender in the sense that women accepted the method less than men. Huber-Eicher and Spring (2008) found similar results. In their study more women than men stated they would not buy meat from immunocastrated pigs and more women disagreed with immunocastration as an alternative method. The acceptance of immunocastration was also influenced by the fact if the respondent was responsible for buying meat in her/his family. Respondents that were not responsible accepted the method more. It could be possible that people who are responsible for buying meat are more concerned for the origin and the production methods of the meat.

## Willingness to pay extra

Although the willingness to pay extra for alternatives was influenced by some factors like gender and living environment, respondents were in general not very keen on paying an extra price, while 96.3% agreed that there should be an alternative and 72% wanted unanesthetized piglet castration to be banned. This behavior can be explained by the "double standard." In their role of citizens, people want farmers to pay more attention to animal welfare, but opposed to this, in their role of consumers, they are not willing to support the extra costs for it (De Tavernier et al. 2005). When discussing consumers' willingness to pay a price premium for animal welfare attributes, it must be kept in mind that the attitudes expressed by most people rarely inform their purchasing behavior. They can claim to be concerned about animal welfare without buying welfare-friendly products, and even when they do buy these products this may be for motives other than just concern about animal welfare (Verbeke 2009). This attitude-behavioral intention gap has also been described by Vermeir and Verbeke (2006). Furthermore, it should be noted that self-reported behavior may be subject to tendencies of socially desirable answering patterns or general beliefs (Verbeke and Viaene 1999).

# Conclusion

In spite of several media campaigns of animal welfare organizations over the past few years, still about half of the Flemish respondents were not aware of the problem of unanesthetized

piglet castration. However, after being informed, the majority wanted unanesthetized castration to be banned. Among the three questioned alternatives, acceptability was the highest for castration under anesthesia and analgesia and the lowest for raising entire males. Immunocastration was judged acceptable by about half of the respondents. The low acceptance of raising entire males may be due to the fact that respondents were possibly influenced by the smelling of the skatole solution which was presented to them to give them an idea of boar taint. This demonstrates that the way of presenting sensitive issues to consumers is very important. When reliable detection methods might become available, informing the public may alter the opinion on raising entire males. Although the concern about animal welfare implications was very high, the willingness to pay extra for alternatives was low, which might prevent the return of investment for the farmers since production costs will increase.

Chapter 3: Behavior of piglets after castration with and without CO<sub>2</sub>-anesthesia

#### Abstract

Surgical castration of male piglets without anesthesia is a routine management practice conducted on commercial pig farms. For animal welfare reasons, it would be beneficial to develop methods of practical pain relief. The objective of this study was to evaluate the effect of providing CO<sub>2</sub> anesthesia before castration on the behavior of piglets for up to 8 d after castration in comparison with piglets castrated without anesthesia. In 3 successive replicates, the behavior of 186 male piglets castrated with (n = 95) or without (n = 91) anesthesia was observed for up to 8 days after castration. All piglets in a given replicate were castrated on the same day, before 8 days of age. Behavioral observations were carried out in accordance with a continuous focal sampling procedure that began immediately after castration and continued for a period of 1 week. Barrows anesthetized with CO<sub>2</sub> before castration displayed more interactive behaviors during the overall observation period than the other barrows (P =0.0412), which may indicate better welfare. Assessing all observation periods separately, differences in activity at the udder, lying, walking, and interactive behaviors appeared to support the beneficial effect of providing CO<sub>2</sub> anesthesia before castration. However, these differences varied over time between treatment groups. The most important conclusion was that piglets castrated with or without CO<sub>2</sub> anesthesia displayed behaviors indicative of pain and discomfort for up to 6 days after castration. Therefore, additional analgesia may be necessary to eliminate the long-term pain caused by castration even in piglets anesthetized with CO<sub>2</sub> before castration.

## Introduction

In the previous chapter, results showed that consumers found castration under anesthesia the most acceptable alternative for unanesthetized piglet castration. Therefore, we conducted a study using carbon dioxide gas ( $CO_2$ ) to castrate piglets under anesthesia.

Carbon dioxide gas is currently being used in several countries to stun pigs prior to exsanguination during slaughter. Carbon dioxide gas is usually administered at concentrations above 80% to minimize the aversion period experienced by the animal and reduce the risk of inadequate anesthesia (Nowak et al. 2007). Kohler et al. (1998) concluded that  $CO_2$ -anesthesia with a concentration of 80% can be induced safely and rapidly in pigs and that castration can be performed without any reaction (struggling, vocalization, defensive

reactions), but stress induced by handling and manipulation before castration is not reduced. Administering  $CO_2$  can cause behavioral signs of aversion in pigs. Svendsen (2006), on the other hand, stated that aversion before losing consciousness is compensated by the fact that piglets experience complete anesthesia and analgesia during castration. Gerritzen et al. (2008) observed that heavy breathing was the only typical behavior that piglets exhibited when exposed to a mixture of 70%  $CO_2$  and 30%  $O_2$ . They also concluded that the period of unconsciousness and analgesia achieved by this gas mixture was long enough to castrate pigs without them experiencing pain.

Trembling behavior has been observed in piglets, lambs and dogs for several days after castration, which indicates that castration causes pain (Wemelsfelder and van Putten, 1985; Molony et al., 1997; Morton and Griffiths, 1985). Other pain-specific behaviors observed in piglets after castration include spasms, tail wagging, scratching and huddling up, as well as changes in lying and suckling behavior (McGlone et al. 1993; Taylor et al. 2001; Hay et al. 2003; Llamas Moya et al. 2008). However, the effect of  $CO_2$  -anesthesia prior to castration on behavioral aspects of pain experience displayed by piglets after castration has not yet been studied to our knowledge.

The main hypothesis of this study is that piglets will experience less pain and discomfort after castration when anesthetized with CO<sub>2</sub> prior to castration and thus improving their overall welfare. The conditional hypothesis is, that CO<sub>2</sub> administration induces complete anesthesia. Complete anesthesia means the loss of consciousness (mental block), elimination of pain (sensible block), muscle relaxation (locomotory block) and repressing reflexes (autonomous block). A preliminary experiment was conducted to test this conditional hypothesis. Therefore, the effect of CO<sub>2</sub>-anesthesia was compared to anesthesia with Zoletil<sup>®</sup> combined with Xyl-M<sup>®</sup> and to a control group (no anesthesia during castration). Intramuscular injection of tiletamine-zolazepam and xylazine is commonly used for veterinary surgical procedures and to produce short-term restraint (Lefkov and Müssig, 2007). According to Lefkov and Müssig (2007), this drug combination is popular because of its smooth induction and recovery profile. Henrikson et al. (1995) also stated that the combination of Zoletil<sup>®</sup> and Xyl-M<sup>®</sup> produces anesthesia and analgesia, and is characterized by reliable and rapid induction.

# **Materials and Methods**

## Animals and Housing

Hybrid pigs (Piétrain x Hypor), heterozygous for the halothane gene, were used. Piglets were raised in the same housing conditions at the Zootechnical Centre – K.U.Leuven R&D (Belgium, Europe). Management of this system was based on the "all in – all out" principle for each room. The farrowing pen had a lying area for the piglets and a sow lying/suckling area provided with rails for piglet protection. Piglets had ad libitum access to water and a commercial diet throughout the study. They were individually marked with an ear tag within the first 24 h after birth and weaned on the same day at approximately 28 d of age.

The farrowing house was temperature controlled using floor and air heating so that the piglets were kept within their thermal neutral zone. The housing environment was automatically controlled with a computerized heating and ventilation system, so that the required temperature was managed independently of the outside temperature. The selected temperature of the farrowing rooms was based on information advised in McGlone and Pond (2003) and was dependent on piglet body weight.

## Preliminary experiment

A total of 99 male piglets from 18 litters were observed in two successive replicates. The number of gilts and boars within a litter was more or less equal, but the total number was not standardized. All the barrows from a single litter received the same castration treatment. Other painful interventions, such as tail docking, ear tagging and iron injection, were standardized for each piglet, and applied without anesthesia or analgesia within the first three days of life, thus avoiding a confounding effect with the experimental treatment.

## Main experiment

A total of 186 male piglets from 38 litters were used in 3 successive replicates. The number of gilts and boars within a litter was more or less equal, but the total number was not standardized. Cross fostering was applied during the week of birth in order to balance litters for body weight and number of piglets. All barrows from a single litter received the same castration treatment. Other painful interventions, such as tail docking, ear tagging and iron injection were standardized for each piglet and applied without anesthesia or analgesia within the first 3 days of life, thus avoiding a confounding effect with the experimental treatment.

#### Experimental procedure

Piglets were handled in accordance with the Belgian law on the protection of animals and the experimental protocol was approved by the Ethical Committee of the K.U.Leuven on the use of experimental animals.

Piglets within a given replicate were castrated on the same day but before 8 days of age (European Directive 2001/93/EG). The age of piglets at castration ranged from 2 to 7 days because of different dates of birth. Piglets were weighed on the day of castration. Between weighing and castration, male piglets were vaccinated for *Mycoplasma hyopneumoniae* (Stellamune Mycoplasma, Intervet) and treated with 0.5 ml amoxicilline (Duphamox, Fort Dodge). Surgical castration was performed by the company veterinarian following the common castration technique. The piglet was held in the hand of the technician (in a head down position), a single transverse incision was made with a scalpel and then the testicles were removed by cutting the spermatic cords. Finally, a disinfectant (Cyclospray, Eurovet) was sprayed onto the wound. After castration the piglets were returned to their pen.

#### Preliminary experiment

In the Netherlands (Animal Sciences Group, WUR) experiments were conducted with a gas mixture of 70% CO<sub>2</sub> and 30% O<sub>2</sub>. The research group started with low concentrations and then gradually increased the concentration of CO<sub>2</sub> until they reached a sufficient anesthesia level. They monitored anesthesia by EEG, ECG and pH, pCO<sub>2</sub> ABE, glucose and lactate in the blood. Based on their results they concluded that piglets were unconscious and insensible during the procedure (Gerritzen et al., 2008). In the present experiment a concentration of 100% CO<sub>2</sub> was used. Higher concentrations give shorter aversion periods during induction and are also used to stun pigs in the slaughter house. Nowak et al. (2007) recommended to keep the aversion period as short as possible, and therefore use high concentrations. Although 100% CO<sub>2</sub> was administered, measurements in the gas mask during a preliminary experiment showed that the concentration of CO<sub>2</sub> in the mask never exceeded 90% (not published).

Piglets were anesthetized with 100%  $CO_2$  for a period of 25 seconds before castration (group I), anesthetized with a combination of Zoletil<sup>®</sup> and Xyl-M<sup>®</sup> injection before castration (group II), or castrated without anesthesia (group III). The  $CO_2$  gas was administered via a mouth mask at a concentration of 100% in order to reduce the duration of aversion. Gerritzen et al. (2008) applied 70%  $CO_2$  during at least 90 s. For the Zoletil and Xyl-M group, a waiting time

of 8 minutes was respected after injecting the piglets with the anesthetics, to make sure piglets were anesthetized properly. In both replicates 9 pens were available with 3 pens for each treatment group.

### Main experiment

Piglets were either anesthetized with 100%  $CO_2$  for a period of 25 seconds before castration (experimental group I) or castrated without anesthesia (experimental group II). Carbon dioxide gas was administered via a mouth mask at a concentration of 100% in order to reduce the duration of aversion. Table 2.1 shows the experimental design, consisting of 2 treatment groups. In the first, second and third replicates, 8, 18 and 12 pens were used respectively, with 4, 10 and 6 pens respectively assigned to the  $CO_2$ -anesthesia treatment. The  $CO_2$  group consisted of 95 piglets from 20 litters; the unanesthetized group contained 91 piglets from 18 litters. It was not the purpose of the present study to examine whether or not piglets show pain-related behavior after castration because it has been extensively described in literature that piglets experience pain after castration. The goal was to study whether or not  $CO_2$ -anesthesia had an influence on that pain experience, therefore only castrated treatment groups with and without  $CO_2$  were included in the experiment.

After castration, barrows were marked visually with a colored sign on their back and returned to their pen. Individual piglet behavior was scored according to the ethogram validated by Hay et al. (2003) (Table 2.2).

Experimental group I	Experimental group II
Anesthetized with CO <sub>2</sub> , then castrated	Castrated without anesthesia
n = 95 piglets (20 pens)	n = 91 piglets (18 pens)

 Table 3.1 Experimental design main experiment.

## **Behavioral Observations**

#### Preliminary experiment

The experimental procedure took place from the morning till noon. In the afternoon the behavior of each barrow was observed within each pen according to a scan sampling procedure. The behavior of all barrows in one pen was scored, and then the observer moved to the next pen and scored behavior of those piglets. This was repeated until every pen was scored 30 times. This procedure has the advantage of covering a wide time range for each

pen. The following days, the procedure was repeated in the morning as well as in the afternoon (i.e. 60 observations per piglet per day).

All behavioral categories were mutually exclusive and are described in Table 1.1. All observations were performed by a single observer standing in the central corridor of the pig house. It was expected that no effects on behavior would remain after six days, as was reported by Hay et al. (2003). The observations started on Wednesday and ended on Tuesday (there were no observations on Saturday and Sunday). The total observation time per pen, in combination with the number of pens (i.e. 18) was assumed to be sufficient to obtain an adequate understanding of the treatment effects studied (Hay et al., 2003; McGlone et al., 1993).

#### Main experiment

Immediately after castration, behavior of each barrow within each pen was observed continuously for a period of 10 min. Pens were observed at random within a sequence according to a scan sampling procedure. The duration of each observation period (i.e. 10 min, multiplied by the number of pens) was considered adequate to achieve the objective of the present study. The main objective of the study was to observe piglet behavior for up to 8 days after castration in relation to anesthesia treatment, since pain-related behaviors were observed in piglets after castration for up to 5 days by Hay et al. (2003). The fact that castration causes pain shortly after the procedure is well-known, since most studies made observations within 24 h after castration (McGlone and Hellman, 1988; Taylor et al., 2001; Llamas Moya et al., 2008). The observation frequency in these studies was high (e.g. every 3 min), but on a limited number of piglets (n = 80). Thirty-eight pens were included in the present study, which represented a total of 186 male piglets. Each pen was observed for a period of 10 min, 2 times a day, giving a total of 13 h of continuous behavioral observations a day for the 38 pens. Hence, in order to make it feasible to combine observations that spanned over 8 days and 186 animals, a sample frequency of 10 min per pen was chosen. It was assumed that pen sampling matched time sampling, so that the observed behaviors per pen were a representative sample of the evolution of pain experienced by the animals over time from castration to day 8. Moreover, the applied statistical method made it possible to consider the individual piglet as the experimental unit, so that about 5 male piglets per time frame of 10 min were available during observation of a pen.

During each 10 min observation period, the frequency of each behavior was recorded every minute for each piglet in accordance with a continuous focal sampling procedure (Fraser,

1978). It was possible that a piglet could show a sequence of different behavioral events within a given single minute; however, the number of times each defined behavioral category was displayed was taken into account by calculating the frequency of that behavioral categories were mutually exclusive and are described in Table 1.1. All observations, i.e. two 10-min sessions per pen per day (am and pm) for 6 days, were performed by a single observer standing in the central corridor of the pig house. It was expected that no behavioral effects due to castration treatment would continue after day 6, as reported by Hay et al. (2003). The observations started on Wednesday and ended the next Wednesday (there were no behavioral observations on Saturday and Sunday, so there were 3 days of observation followed by a 2-day break and then another 3 days of observation). The total observation time per pen, in combination with the number of pens (i.e., 38) was assumed to be sufficient to obtain an adequate understanding of the treatment effects studied (Hay et al., 2003; McGlone et al., 1993).

### Statistical Analysis

All data were analyzed using the SAS 9.2 (SAS Inst, Inc, Cary, NC, USA) software. A significance level of 0.05 was used. Behaviors were grouped for analysis in order to have a sufficient number of observations within each grouped category, so that the empirical estimation could be carried out on a sufficient number of observations. A condition of the applied statistical model is that the performance of the empirical estimation passes the convergence test (Anonymous, 2006). Hence, lateral lying, ventral lying and sleeping were grouped under "lying"; teat seeking, suckling and udder massage were grouped under "udder activity"; huddled up, trembling, spasms, scratching and tail wagging were grouped under "pain-related behaviors"; nosing, chewing, licking, playing and aggression were grouped under "interaction behaviors"; walking and running were grouped under "walking"; and sitting, standing and kneeling were grouped under "postures". The complete sequence of suckling behavior was estimated to last about 2 min, consisting of 5 typical different phases, with the behavior preliminary to suckling being very variable (Fraser, 1980). This information was considered to be a biologically sound argument for grouping these behaviors into a single category representing nursing behavior, which is reported to be sensitive to castration (McGlone and Hellman, 1988). Moreover, grouping of a sufficient number of physiologically related behavioral events into a single category increased the probability of matching the power requirements of the applied statistical model. Huddling and trembling behaviors were not regarded as thermoregulatory behaviors, since environmental temperature in the farrowing/nursing room was engineered according to the piglet's thermal neutral zone. This was confirmed by the displayed temperature and by observation of piglet lying behavior, i.e. that of the female piglets being neutral for castration (Geers et al., 1986).

Data were not normally distributed and were dichotomized using the median as cut-off value. The binary data were analyzed using the logistic mixed model, with fixed effects being treatment, observation period, piglet weight and age at castration, with the piglet as random effect. Random effects accounted for the variability between the piglets within and between litters. The applied procedure made it possible to allocate a random effect to a variable (Anonymous, 2006), so that piglets could be regarded as the experimental units. There were 2 factors considered in defining this random effect: variability between litters and variability between individual piglets. When taking into account the lowest level in the model, i.e. individual piglets, the variability between piglets partially represented the variability between litters. Furthermore, Fraser (1978) analyzed behavior of group housed piglets with the individual piglet being regarded as the experimental unit. After analysis of the data for the overall period of observations, data were also analyzed in the same way for each morning and afternoon observation period. Values of the calculated frequency are presented as means  $\pm$  SEM for each behavioral category. The unit of values mentioned in the text is mean frequency per minute.

### Results

### **Preliminary experiment**

Piglets of group I (CO<sub>2</sub>) recovered from anesthesia within a few minutes. Piglets of group II (Zoletil<sup>®</sup> and Xyl-M<sup>®</sup>) recovered within a period of 2 hours.

#### Behavior, overall period of observation

When looking at all observation periods together, there were no significant differences in behavior between treatment groups.

#### Behavior, per observation period

In the afternoon of observation day 1, piglets of the group III walked less than piglets of the group I (p = 0.0126) and piglets of group II (p = 0.0101).

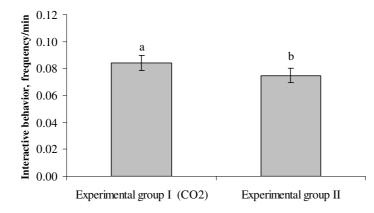
In the afternoon of observation day 4, piglets of group II were lying down less than piglets of group I (p = 0,0051) and piglets of group III (p = 0,0081).

#### Main experiment

In order to fully understand the effect of  $CO_2$ -anesthesia on the behavior of piglets after castrations, results were reported for all data over time and for each observation period separately. Attention was focused on the complementary behavioral events that are considered to be indicative of pain and discomfort, and also on the so-called positive behaviors (i.e. interactive behavior and social cohesion) that are important for animal welfare (Fraser and Broom, 1990; Blackshaw et al., 1997).

#### Observation periods taken together

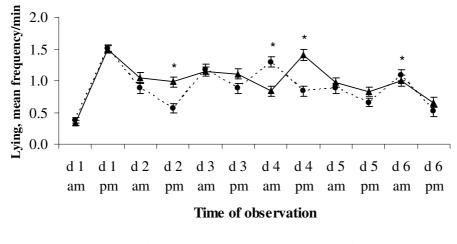
Assessment of all observation periods taken together found that the only behavioral category that showed a significant difference was interactive behaviors (nosing, chewing, licking, playing and aggression) (Figure 3.1). Barrows in the anesthetized group displayed 13% more interactive behaviors than barrows from the unanesthetized group (0.0842  $\pm$  0.00548 vs. 0.0748  $\pm$  0.00523; *P* = 0.0412).



**Figure 3.1** Occurrence of interactive behaviors (mean frequency per minute) over all observation periods. Significant differences (P < 0.05) are indicated by different letters. Means and SEM are from non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

#### Observation periods separately

*Lying.* During the afternoon observation periods on day 2 (P = 0.0004) and 4 (P < 0.0001), barrows in the CO<sub>2</sub>-group (respectively 0.980 ± 0.0763 and 1.409 ± 0.0836) laid down about 71% more than unanesthetized piglets (respectively 0.567 ± 0.0708 and 0.834 ± 0.0836). During the morning observation periods on day 4 (P = 0.0008) and 6 (P = 0.0245), barrows in the unanesthetized group (respectively 1.295 ± 0.0801 and 1.089 ± 0.0843) laid down 55% and 9% more, respectively, than barrows in the anesthetized group (respectively 0.837 ± 0.0830 and 0.996 ± 0.0839) (Figure 3.2).

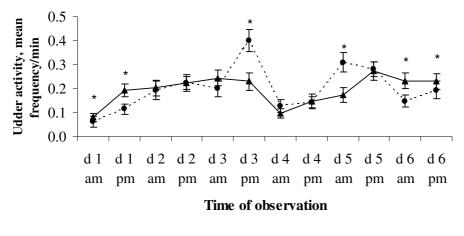


Experimental group I (CO2) ··· Experimental group II

Figure 3.2 Occurrence of lying (mean frequency per minute) performed by piglets castrated with or without  $CO_2$ -anesthesia. Means with an asterisk differ (P < 0.05) from means without an asterisk within the same observation time. Means and SEM are from the non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

*Udder activity.* Barrows in the CO<sub>2</sub>-group were 26% more active at the udder than piglets in the unanesthetized group during the morning observation periods on day 1 (0.0789  $\pm$  0.0169 vs. 0.0627  $\pm$  0.0233; P = 0.0259), 69% more during the afternoon observation periods on day 1 (0.193  $\pm$  0.0276 vs. 0.114  $\pm$  0.0201; P = 0.0372), 56% more during the morning observation periods on day 6 (0.231  $\pm$  0.0326 vs. 0.148  $\pm$  0.0242; P = 0.0245) and 19% more during the afternoon observation periods on day 6 (0.239  $\pm$  0.0330 vs. 0.193  $\pm$  0.0359; P = 0.0149). However, during the afternoon observation periods on day 3 (P = 0.0424) and the morning observation periods on day 5 (P = 0.0167), barrows in the

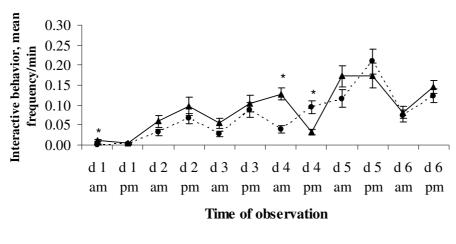
unanesthetized group (respectively  $0.399 \pm 0.0467$  and  $0.309 \pm 0.0396$ ) performed more udder activity than barrows in the CO<sub>2</sub>-group (respectively  $0.229 \pm 0.0367$  and  $0.172 \pm 0.0311$ ), 74 and 80%, respectively (Figure 3.3).



Experimental group I (CO2) •••• Experimental group II

Figure 3.3 Occurrence of activity at the udder (mean frequency per minute) performed by piglets castrated with or without CO<sub>2</sub>-anesthesia. Means with an asterisk differ (P < 0.05) from means without an asterisk within the same observation time. Means and SEM are from the non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

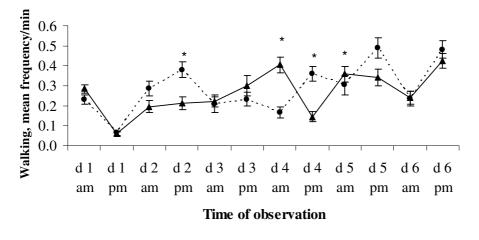
Interactive behavior. During the morning observation periods on day 1 (P = 0.0188) and 4 (P = 0.0008) barrows in the CO<sub>2</sub>-group (respectively 0.0116 ± 0.00330 and 0.128 ± 0.0156) performed more interactive behaviors than barrows in the unanesthetized group (respectively 0.00110 ± 0.00110 and 0.0385 ± 0.00921), 955 and 228%, respectively. During the afternoon observation periods on day 4, on the other hand, barrows in the unanesthetized group (0.0945 ± 0.0152) performed 199% more interactive behaviors than piglets in the anesthetized group (0.0316 ± 0.00706) (P = 0.0045) (Figure 3.4).



Experimental group I (CO2) ···• Experimental group II

**Figure 3.4** Occurrence of interactive behavior (mean frequency per minute) performed by piglets castrated with or without  $CO_2$ -anesthesia. Means with an asterisk differ (P < 0.05) from means without an asterisk within the same observation time. Means and SEM are from the non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

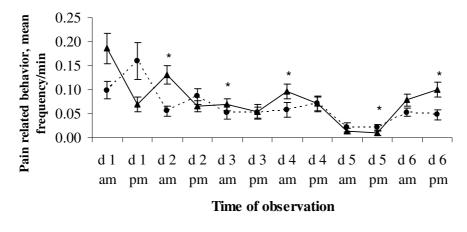
*Walking.* Barrows in the unanesthetized group walked around more during the afternoon observation periods on day 2 (0.381 ± 0.0396 vs. 0.213 ± 0.0311; P = 0.0039) and day 4 (0.360 ± 0.0382 vs. 0.145 ± 0.0253; P < 0.0001), 79 and 148%, respectively. Piglets in the anesthetized group walked around more during the morning observation periods on day 4 (0.406 ± 0.0390 vs. 0.166 ± 0.0296; P < 0.0001) and day 5 (0.359 ± 0.0402 vs. 0.303 ± 0.0476; P = 0.0011), 145 and 19%, respectively (Figure 3.5).



Experimental group I (CO2) ···• Experimental group II

Figure 3.5 Occurrence of walking (mean frequency per minute) performed by piglets castrated with or without  $CO_2$ -anesthesia. Means with an asterisk differ (P < 0.05) from means without an asterisk within the same observation time. Means and SEM are from the non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

**Pain-related behavior.** During the morning observation periods on day 2 (0.131  $\pm$  0.0192 vs. 0.0549  $\pm$  0.0104; P = 0.0376), day 3 (0.0684  $\pm$  0.0130 vs. 0.0527  $\pm$  0.0150; P = 0.0191), day 4 (0.0958  $\pm$  0.0148 vs. 0.0582  $\pm$  0.0156; P = 0.0477) and day 6 (0.100  $\pm$  0.0154 vs. 0.0473  $\pm$  0.0105; P = 0.0028), barrows of the CO<sub>2</sub>-group displayed more pain-related behaviors (138%, 30%, 65% and 113%, respectively) compared with piglets in the unanesthetized group. During the afternoon observation periods on day 5, piglets in the unanesthetized group displayed 75% more pain-related behaviors (0.0220  $\pm$  0.00973 vs. 0.0126  $\pm$  0.00584; P = 0.0109) (Figure 3.6). Figure 3.6 shows a larger difference between the mean values of pain-related behavior during day 1, compared with the significant differences. However, these differences between the mean values on day 1 were not statistically significant. The explanation is that variability with respect to showing pain-related behavior among barrows was large.

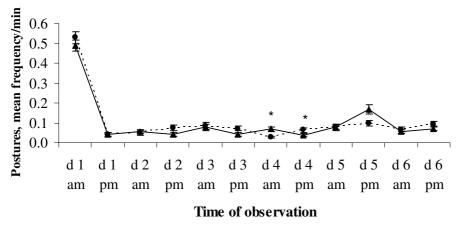


Experimental group I (CO2) ···· Experimental group II

Figure 3.6 Occurrence of pain-related behavior (mean frequency per minute) performed by piglets castrated with or without CO<sub>2</sub>-anesthesia. Means with an asterisk differ (P < 0.05) from means without an asterisk within the same observation time. Means and SEM are from the non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

**Postures.** During the morning observation periods on day 4, barrows in the anesthetized group displayed 139% more postures than those in the unanesthetized group  $(0.0684 \pm 0.0107 \text{ vs. } 0.0286 \pm 0.00591; P = 0.0079)$ . During the afternoon observation periods on day 4; however, barrows in the unanesthetized group performed 64% more postures  $(0.0637 \pm 0.0104 \text{ vs. } 0.0389 \pm 0.00767; P = 0.0359)$  (Figure 3.7). Figure 3.7 shows a

larger difference between the mean values of postures for afternoon observations on day 5, compared with the significant differences. However, this difference between mean values on the afternoon of day 5 was not statistically significant. The explanation is that individual variability with respect to showing sitting, standing and kneeling among barrows was large.



Experimental group I (CO2) ··· Experimental group II

Figure 3.7 Occurrence of postures (mean frequency per minute) performed by piglets castrated with or without  $CO_2$ -anesthesia. Means with an asterisk differ (P < 0.05) from means without an asterisk within the same observation time. Means and SEM are from the non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

*Social cohesion.* Whether or not a piglet isolates itself from other piglets is considered a measure of social cohesion (Table 1.1). There were no differences in social cohesion between the 2 treatment groups.

# Discussion

#### **Preliminary experiment**

#### Behavior, overall period of observation

There were no differences in behavior between group I and II when considering the overall period of observation. This may be an indication that  $CO_2$  matches the anesthetic properties of the combination of Zoletil<sup>®</sup> and Xyl-M<sup>®</sup>. The fact that there is no difference between anesthetized piglets (group I and II) and piglets of group III, confirms the findings of

Gerritzen et al. (2008) that anesthesia relieves the pain on the moment of the procedure but not the pain afterwards.

#### Behavior, per observation period

In the afternoon of observation day 1 piglets of group III walked around the least. Llamas Moya et al. (2008) suggested that castrated piglets may avoid certain behavioral activities, such as walking, in order to minimize pain. Walking can thus be interpreted as increased restlessness when piglets are in pain, which implicates that piglets that were anesthetized during castration (both group I and II) are in more pain than piglets of group III in the first hours after the procedure. A possible explanation for this is that they are anesthetized during the procedure, but when anesthesia wears off the pain kicks in more strongly. This should be interpreted as a delayed pain experience rather than an additional pain experience.

In the afternoon of observation day 4, piglets of group II were lying down (resting) the least. It could be that they were more agitated, but it concerns observation day 4 (i.e. 5 days after castration) so it is difficult to say whether this is still an effect of the anesthesia treatment.

### Main experiment

#### Effects of Treatment on Behavior: Overall Period of Observation

Anesthetized barrows interacted more with one another than barrows in the unanesthetized group. Interactive behaviors in this study included frequency of nosing, chewing, licking, aggression and playing. Lower levels of interactive behaviors such as play displayed by castrated piglets may indicate poor welfare (Llamas Moya et al., 2008). Blackshaw et al. (1997) specifically described playing behavior as an indication of positive animal welfare. Conversely, Hay et al. (2003) suggested that reduced oral exploration, such as licking and chewing, may be associated with the experience of pain. These conclusions suggested by other researchers agree with our findings that the lower level of interactive behaviors (less playing, licking and chewing) performed by unanesthetized barrows as compared with anesthetized barrows could be an indication of a different state of welfare.

#### Effects of Treatment on Behavior: per Observation Period

The frequency of lying behavior was greater for barrows in the anesthetized group than for the unanesthetized group during the afternoon observation periods on day 2 and 4, though the opposite was true during the morning observation periods on day 4 and 6, i.e. day 6 and 8 after castration, respectively. In the present study, a beneficial effect of anesthesia during castration could be advocated, according to Torrey et al. (2009), because of a greater frequency of lying behavior the first 6 days after castration. A lower frequency was observed from day 6 on, on the other hand, which matched the point of view of McGlone and Hellman (1988) and Hay et al. (2003). Both McGlone and Hellman (1988) and Hay et al. (2003) observed a greater frequency of lying behavior within the first hours after castration, mutually excluding suckling activity and considered this an indication of poor welfare.

Udder activity differed between the 2 treatment groups over the observation periods. During the morning and afternoon observation periods on day 1 and 6, barrows in the CO<sub>2</sub>-group displayed more activity at the udder than unanesthetized barrows. During the afternoon observation periods on day 3 and morning observation periods on day 5; however, barrows in the unanesthetized group were more active at the udder. Noonan et al. (1996) suggested that increased udder activity may be related to the experience of pain as endorphins (i.e. endogenous opioids) may be released during suckling, which may have an analgesic effect. This would indicate that at the day of castration (day 1), anesthetized piglets possibly experienced more pain. This should be interpreted as a delayed pain experience rather than an additional pain experience. This finding also confirms the results of Gerritzen et al. (2008) that CO<sub>2</sub> induces analgesia only for a short period of time (i.e. the time during anesthesia). During the morning observation periods on day 1 and 4, barrows in the CO<sub>2</sub>-group did show more interactive behaviors. During the afternoon observation periods on day 4, however, barrows in the unanesthetized group displayed more interactive behaviors. The explanation previously given concerning the beneficial aspect (overall period of observation), can also be applied here (Torrey et al., 2009).

Llamas Moya et al. (2008) suggested that castrated piglets may avoid certain behavioral activities, such as walking, in order to minimize pain. During the morning observation periods on day 4 and 5, barrows in the CO<sub>2</sub>-group walked around more than barrows in the unanesthetized group. During the afternoon observation periods on day 2 and 4, on the other hand, the opposite was found. These alternating observations of walking behavior could suggest that both treatment groups in fact experienced pain as result of castration and therefore adjusted their walking patterns, though at different times.

The postures performed by piglets differed significantly between the 2 treatment groups, but only during the observation periods on day 4. During the morning observation periods on day 4, barrows in the CO<sub>2</sub>-group sat, kneeled and stood more, while during the afternoon of that same day barrows in the unanesthetized group displayed these behaviors more frequently. Taylor et al. (2001) observed that the frequency of sitting and standing postures increased after castration in piglets. Other studies (McGlone & Hellman 1988; McGlone et al. 1993; Kielly et al. 1999) however, found that piglets spent less time standing after castration. Concerning postures, the literature appears to be contradictory, which suggests that changes in posture are not a fully reliable indicator of pain in response to castration in piglets (Hay et al. 2003).

In the present study, barrows in the CO<sub>2</sub>-group displayed more pain-related behaviors during the morning observation periods on day 2, 3 and 4 and afternoon observation periods on day 6. Only during the afternoon observation periods on day 5 did barrows in the unanesthetized group show more pain-related behaviors. These results contradict the hypothesis of the present study that piglets anesthetized with CO<sub>2</sub> during castration would suffer less pain after castration; however, these results confirm the finding that CO<sub>2</sub> induces analgesia but only for a short period of time (Gerritzen et al., 2008). In other words, piglets under CO<sub>2</sub>-anesthesia did not feel pain during castration, but it appears as though the anesthetic effects of the CO<sub>2</sub> wore off quickly, so additional analgesia may be necessary to avoid the long term (postoperative) pain experienced by piglets in response to castration. Another point of view is that the parameters used to assess pain-related behaviors used in this study did not discriminate between actual pain and discomfort, which originated from different sources. In other words, the specificity and sensitivity of the parameters used in this study may have been too low for detection of actual pain. Nevertheless, these behaviors can be considered to be part of a pig's normal ethogram, in which the expressions of both discomfort and pain are equally accepted as indications of reduced animal welfare.

## Conclusion

The preliminary experiment showed no differences in behavior between group I and II when considering the overall period of observation. This may be an indication that  $CO_2$  matches the anesthetic properties of the combination of Zoletil<sup>®</sup> and Xyl-M<sup>®</sup>. The main objective of the present study was to evaluate the effects of  $CO_2$ -anesthesia during castration on the behavior

of piglets after castration. The observed differences in behaviors were not conclusive for any of the behavioral categories studied. However, the fact that barrows castrated under  $CO_{2}$ anesthesia displayed more interactive behaviors during the overall observation period than unanesthetized barrows may be an indication of better welfare. The fact that the observed behavioral differences with respect to lying, and interactive behaviors continued for up to 6 or 7 days after castration support this conclusion. However, all barrows that were castrated under anesthesia also displayed behaviors indicative of pain and discomfort. Therefore, piglets may need to be provided with additional analgesia to eliminate the pain caused by castration even if piglets are anesthetized with  $CO_2$  prior to castration. By way of conclusion, for post operative effects, male piglets appeared to benefit to a certain extent from the use of  $CO_2$ anesthesia for castration though this conclusion still needs to be confirmed regarding the use of  $CO_2$ -anesthesia in the performance of other painful interventions whether or not in combination with castration.

Castration is not the only event that may threaten piglets' welfare. Piglets are subjected to several painful management practices, especially during their first week of life. In order to improve overall welfare, not only castration has to be dealt with, but painful management procedures in general. In the following chapter it was therefore investigated if reducing painful interventions during the first week of life results in better zootechnical performance of the piglets, reduced piglet mortality and if the overall welfare, indicated by behavioral criteria, is improved.

Chapter 4: Improving survival, growth rate and animal welfare in piglets by avoiding teeth shortening and tail docking?

Chapter redrafted after: S. Van Beirendonck, B. Driessen, G. Verbeke, L. Permentier, V. Van de Perre, R. Geers (2011). Improving survival, growth rate and animal welfare in piglets by avoiding teeth shortening and tail docking? Journal of Veterinary Behavior: Clinical Applications and Research, 7:2, 88-93.

## Abstract

Piglets are subjected to several painful procedures during their first week of life, including ear tagging, teeth clipping or grinding (although routinely prohibited in Europe), tail docking (although routinely prohibited in Europe), needle injections (vaccination and iron injection), and castration for male piglets. All these management practices may cause pain and stress to the newborn piglets. The hypothesis of this experiment was that reducing painful interventions during the first week of life would result in better zootechnical performance of the piglets, reduced piglet mortality, and that the overall welfare would be improved. To investigate this, the 4 lightest piglets of the experimental group (EE) were not subjected to tail docking and teeth clipping or grinding. The 4 lightest piglets of the control group (CL) and the other piglets of the experimental group (EC) and the control group (CC) received treatments as the common practice. There were differences in behavior, but there were no differences regarding weight at weaning between CL and EE. However, mortality rate was higher in the lightest piglets, that is, CL (34.1%) and EE (23.0%), whereas mortality rate in the other piglets was much lower (9.0% for CC and 9.3% for EC).

## Introduction

In the first week of life, a number of painful management practices are carried out on piglets (Vinuela-Fernandez et al., 2007): they get a numbered ear tag, teeth are shortened (although routinely prohibited in Europe), tails are docked (although routinely prohibited in Europe), they receive needle injections (vaccination and iron injection) and male piglets are castrated. Teeth shortening is not routinely allowed in Europe but is still frequently applied to minimise wounds and lesions of the skin in other piglets and the sow's udder. Two techniques are used: clipping and grinding. The first days after teeth shortening, the intervention seems to have some advantages (fewer skin lesions on both the piglets and the sows' udder) (Fraser 1975, Brookes and Lean 1993). On the other hand, more recent work indicates growth advantages for piglets of which the canines were not shortened (Weary and Fraser 1999, Delbor et al., 2000, Bataille et al., 2002). Several authors (Gallois et al., 2002; Noonan et al., 1994) stated that shortening canines, whichever technique is used, leads to tooth lesions and pain.

Tail docking is also not routinely allowed. Tails are docked directly or some days after birth (within the first 7 days), frequently without anesthesia or analgesia. They are snipped off with a heated knife so the wound is sealed at the same time. Sometimes an ordinary cutter or bistouri knife is used, and antiseptic products might be applied on the wound. Tail docking is considered to be painful (Meunier-Salaun et al., 2002; Noonan et al., 1994; Simonsen et al., 1991) since the peripheral nerves in the tip of the tail are already fully developed in newborn piglets (Simonsen et al., 1991).

The hypothesis of this experiment was that reducing painful interventions during the first week of life results in better zootechnical performance of the piglets, reduced piglet mortality and that the overall welfare, indicated by behavioral criteria, is improved. The present study tries to bring new information into the debate on farm animal welfare by focusing on the lightest piglets, which is a novel aspect compared to other reports.

## **Materials and Methods**

#### Animals and Housing

A total of 295 piglets from 22 litters were observed from birth to weaning. Berndtson (1991) described a method for selecting or assessing the number of replicates for animal experiments. Based on his method and considering coefficient of variance (CV) and the difference to be detected, the sample size in the present experiment is sufficient to assess statistically significant differences with p < 0.05 and a power of 80 %. The number of female and male piglets within a litter was more or less equal, but the total number was not standardized. Cross fostering was carried out in litters up to 7 days old in order to balance weight and number of piglets per litter. Six out of 22 sows were primiparous, but they were spread over treatment groups. Housing conditions were the same as described in chapter 3.

## Experimental procedure

Piglets were handled according to the Belgian law on the protection of animals, and the experimental protocol was agreed by the Ethical Committee on the use of experimental animals.

Litters were randomly divided into two main groups, a control group and an experimental group, and these main groups were each divided into two subgroups (Table 3.1). A few hours after birth, all piglets were weighed individually to identify the lightest piglets within each litter. The number of gilts and boars within a litter was more or less equal, but the total number was not standardized.

Experime	Experimental group Control grou		l group
EE (n = 44)	EC (n = 105)	CL (n = 44)	CC (n = 102)
4 lightest piglets of	Other piglets in same	4 lightest piglets of	Other piglets in same
each pen	pen as EE	each pen	pen as CL
Tails and teeth left	All procedures (tail	All procedures carried	All procedures carried
intact; ear tagging,	docking, teeth	out as normal	out as normal
vaccination, iron and	grinding, ear tagging,		
antibiotic injection	vaccination, iron and		
and castration carried	antibiotic injection		
out as normal	and castration) carried		
	out as normal		

 Table 4.1 Experimental design.

Piglets were weighed, had their teeth shortened and tails docked, and received a numbered ear tag and iron injection within the first day of life. Within the first week of life, they were vaccinated for *Mycoplasma hyopneumoniae* (Stellamune Mycoplasma, Pfizer Animal Health), and they received an injection of antibiotics (Duphamox LA, Fort Dodge). These interventions were performed by the technicians while supervised by the company veterinarian.

However, all piglet castrations were performed by the company veterinarian following the common castration technique, i.e. with the piglet held in the hand of the technician (head down position). One transverse incision was made with a scalpel and the testicles were removed by cutting the spermatic cord. Finally, a disinfectant (Cyclospray, Eurovet) was sprayed on to the wound and the piglets were returned to their pen.

If cross fostering was necessary after the experimental procedure, this happened only between litters of the same treatment group. Body weight was measured every week until weaning.

### **Behavioral Observations**

Behavioral observations were carried out the day of birth and the day after, and subsequently every week until weaning. The technique used to observe behavior was the same as described in the main experiment of chapter 3. The behaviors were described in table 1.1 (p. 9).

#### Statistical Analysis

All data were analyzed using SAS 9.2 (SAS Inst, Inc, Cary, NC, USA, 2008) software. A significance level of 0.05 was used. Categories of behavior were grouped for analysis in order to have a sufficient number of observations within each grouped category, so that the empirical estimation could be carried out on a sufficient number of observations. A condition of the applied statistical model is that the performance of the empirical estimation passes the convergence test (Anonymous, 2006). Hence, lateral lying, ventral lying and sleeping were grouped under 'lying'; suckling, udder massage and looking for teat were grouped under 'udder activity'; huddled up, trembling, spasms, scratching and tail wagging were grouped under 'behavior indicative of discomfort'; nosing, chewing, licking, playing and aggression were grouped under 'interaction behavior'; walking and running were grouped under 'walking'; and sitting, standing and kneeling were grouped under 'postures'.

Behavioral data were not normally distributed; therefore data were dichotomized using the median as cut-off value. The binary data were analyzed using a logistic mixed model, with fixed effects for treatment and piglet weight, and with piglet as a random effect. The random effect accounts for the variability between the piglets within and between litters. The applied procedure (GLIMMIX) makes it possible to allocate a random effect to a variable (Anonymous, 2006), so that the piglets can be regarded as the experimental units. There are two factors to be considered in defining this random effect: the variability between litters and the variability between individual piglets. When taking into account the lowest level in the model, i.e. the individual piglets, the variability between piglets partially represents the variability between litters. Values are presented as means  $\pm$  SEM.

Data concerning body weight were analyzed using the generalized linear mixed model (MIXED) procedure (SAS, Inst, Inc, Cary, NC, USA, 2008). Mortality rates were analyzed using a logistic mixed model with fixed effects for treatment and parity of the sow, and with sow as a random effect.

# Results

#### Behavior from birth to weaning (4 weeks)

Piglets of the control group (CC) walked around more than the control piglets of the experimental group (EC) (p < 0.0001); the four lightest piglets of the control group (CL) walked around more than the lightest piglets of the experimental group (EE) (p = 0.0040) and the control piglets of the experimental group (EC) (p < 0.0001) (Table 3.2).

**Table 4.2** Walking (mean frequency per minute, minimum and maximum) observed during the entire nursery period. Significant differences at  $P \le 0.05$  are indicated by letters in the mean column. Treatment groups without a common superscript differ. Means, minima and maxima are from the non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

Treatment	Mean	Minimum	Maximum
CC	0.330 <sup>ab</sup>	0	1.4
CL	0.329 <sup>b</sup>	0	1.2
EE	0.274 <sup>ac</sup>	0	1.3
EC	0.264 <sup>c</sup>	0	1.2

CC and CL showed more activity at the udder than EE (p < 0.0001 for CC and p = 0.0021 for CL) and EC (p < 0.0001 for CC and p = 0.0009 for CL) (Table 3.3).

**Table 4.3** Udder activity (mean frequency per minute, minimum and maximum) observed during the entire nursery period. Significant differences at  $P \le 0.05$  are indicated by letters in the mean column. Treatment groups without a common superscript differ. Means, minima and maxima are from the non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

Treatment	Mean	Minimum	Maximum
CC	0.291 <sup>a</sup>	0	1
CL	$0.282^{a}$	0	0.9
EE	0.200 <sup>b</sup>	0	1
EC	$0.200^{b}$	0	1

## Growth rate

In the first week of life, CL grew the least (p = 0.0142 for CC; 0.0243 for EE and 0.0168 for EC). In the second week however, CC grew more than EC (p = 0.0112). In the third week, the

four lightest piglets of the experimental group (EE) (p = 0.0197) and of the control group (CL) (p = 0.0392) gained less weight than EC. In the last week before weaning, piglets of group EE put on less weight than piglets of the control group, both CC (p = 0.0105) and CL (p = 0.0139). Looking at the overall period from birth till weaning, the four lightest piglets of the experimental group (EE) gained less weight than CC (p = 0.0005) and EC (p = 0.0064) (Table 3.4).

	Corrected Means (g)	Lower	Upper
Birth-week 1			
CC	$0.286^{a}$	0.238	0.333
CL	$0.252^{b}$	0.203	0.300
EE	$0.288^{a}$	0.236	0.339
EC	$0.285^{a}$	0.236	0.334
Week 1-week 2			
CC	0.392 <sup>a</sup>	0.334	0.451
CL	0.376 <sup>ab</sup>	0.317	0.435
EE	0.369 <sup>ab</sup>	0.306	0.432
EC	0.364 <sup>b</sup>	0.303	0.424
Week 2-week 3			
CC	0.389 <sup>ab</sup>	0.318	0.459
CL	$0.368^{a}$	0.296	0.440
EE	0.365 <sup>a</sup>	0.288	0.441
EC	0.412 <sup>b</sup>	0.339	0.484
Veek 3-weaning			
CC	$0.187^{a}$	0.122	0.251
CL	0.201 <sup>a</sup>	0.129	0.274
EE	0.132 <sup>b</sup>	0.0618	0.201
EC	$0.164^{ab}$	0.0989	0.229
Birth-weaning			
CC	$0.350^{a}$	0.312	0.387
CL	$0.340^{ab}$	0.300	0.379
EE	0.315 <sup>b</sup>	0.276	0.355
EC	$0.342^{a}$	0.304	0.380

**Table 4.4** Daily weight gain (g) during nursery period. Means and confidence intervals are shown. Within a subcolumn, means without a common letter differ significantly at  $P \le 0.05$ .

### Comparison of weight at weaning

The four lightest piglets of the experimental group (EE) had a lower weaning weight than CC (p = 0.0001) and EC (p = 0.0062) (Table 3.5). There was no significant difference between the four lightest piglets of the experimental group and the four lightest piglets of the control group.

**Table 4.5** Weight at weaning (kg) for the different treatment groups. Data are presented as lsmeans  $\pm$  standard error. Within a row, means without a common letter differ significantly at P  $\leq$  0.05.

CC	CL	EE	EC
$8.65 \pm 0.135$	$8.21 \pm 0.265$	$7.60 \pm 0.230$	$8.30 \pm 0.127$
а	ab	b	a

### Mortality rate

Considering the four different treatment groups together, 14.9 % of all piglets died before weaning. In the group of the four lightest piglets of the control group, 34.1 % died before weaning. In the group of the four lightest piglets of the experimental group (tails and teeth left intact), 23 % died before weaning (Table 3.6). The most piglets died in group CL (p = 0.0004 versus CC and p = 0.0004 versus EC) and group EE (p = 0.0184 versus CC and p = 0.0098 versus EC).

**Table 4.6** Mortality rate and moment of death for the different treatment groups. Values without a common letterdiffer significantly at  $P \le 0.05$ .

Treatment	Mortality rate	Time of death
CC	9.0% <sup>a</sup>	88.9% in week 1
	n = 9	11.1% in week 2
CL	34.1% <sup>b</sup>	93.3% in week 1
	n = 15	6.7% in week 2
EC	9.3% <sup>a</sup>	80.0% in week 1
	n = 9	20.0% in week 2
EE	23.0% <sup>b</sup>	80.0% in week 1
	n = 11	20.0% in week 2
All piglets	14.9%	86.4% in week 1
	n = 44	13.6% in week 2

## Discussion

### Behavior from birth to weaning (4 weeks)

CC and CL showed more activity at the udder than EE and EC. Although Hay et al. (2003) found that piglets are less active at the udder in the first 2.5 hours after castration (when they are in pain), Noonan et al. (1996) found that endorphins are released during suckling, hence creating an analgesic effect. Therefore, Noonan et al. (1996) related painful procedures to increased udder activity. Transposing that finding to the results of the present study, it is possible that the four lightest piglets of the control group experienced more discomfort than the four lightest piglets of the experimental group. This might be due to the extra procedures they were subjected to -tail docking and teeth clipping- because this was the only difference between both treatments.

Comparing the two groups of lightest piglets, the control group (CL) walked around more than the experimental group (EE). Llamas Moya et al. (2008) stated that piglets in pain (e.g. after castration) avoid walking in order to minimize pain. Applying this on the present results, it would imply that the lightest piglets of the experimental group (EE) experience more pain than the lightest piglets of the control group (CL). A possible explanation could be that more weaker piglets have survived in the EE group, which are more sensitive to pain. This finding disputes the behavioral indications of udder activity, but can be explained by the fact that the difference in udder activity is mostly based on a difference seen in the first week of observations. Walking behavior differed between treatment groups more in the last week of observations. This would imply that in the beginning, after the painful procedures, EE benefit from the fact that their teeth and tails were left intact while a few weeks later this benefit has faded out and EE are possibly more sensitive to pain because of higher survival of weaker piglets. The fact that CC were more active at the udder than EC could possibly indicate that CC were in more pain than EC. A possible explanation for this may be that EC were slightly heavier (on average 1.60 kg) than CC (on average 1.45 kg) and therefore might have experienced relatively less inconvenience from the treatments.

### Growth rate

Until the last week before weaning, there was no difference in mean daily weight gain between the four lightest piglets of the control group (CL) and those of the experimental group (EE). In the fourth week, CL had a higher daily weight gain than EE. The explanation for this sudden change at the end of the nursery period could be that more piglets had died in the CL group in comparison with the EE group (34.1% versus 23.0%). In the CL group more of the lighter piglets had died, leaving behind the 'stronger', better growing piglets, while in the EE group more lighter piglets survived, lowering the mean daily weight gain of the group. Overall, growth rate dropped during the fourth week, which might be explained by a decreased milk production of the sows (King et al., 1997; Whittemore and Kyriazakis, 2005). It is known that smaller pigs more often lose the fight for the highest milk producing teat, so that they are more prone to a decreased growth rate in case of overall reduced milk production.

### Comparison of weight at weaning

There is no significant difference between the four lightest piglets of the experimental group and the four lightest piglets of the control group. Nevertheless, it must be considered that a larger number of piglets was weaned in the EE group, which is a beneficial effect.

#### Survival rate

Survival is reduced in low birth weight piglets (Baxter et al., 2008). However, Knol et al. (2002) stated that genetic correlation between piglet survival until weaning and birth weight is low, indicating that selection on birth weight as an indirect way to improve pre-weaning survival is doubtful. Tuchscherer et al. (2000) on the other hand, found that piglets that survived the nursing period were significantly heavier at birth than piglets that died before weaning. In the present study, this result was confirmed considering that more piglets died in the groups with the lightest piglets than in the other groups (34.1% of CL and 23.0% of EE vs. 9.0% of CC and 9.3% of EC). According to Tuchscherer et al. (2000), Herpin et al. (2002) and Baxter et al. (2008), the majority of pre-weaning deaths occurs within the first 72 h post-partum. The results found in the present study agree with these authors, in that most of the deaths occurred within the first week of life. The rest of piglet deaths occurred during the second week; in the last two weeks of the nursing period no piglets died. The total percentage

of piglets that died before weaning was 14.9, which is in agreement with Loncke et al. (2008) who stated that piglet mortality up to 15% is considered as acceptable in Belgium. These figures were obtained from a recent study in Belgium (Loncke et al., 2008) and are representative for Belgium. The last decennia, the number of piglets per sow per year has increased drastically because of a thorough selection on fertility and productivity. With the selection for larger litters, pre-weaning mortality has increased because the number of smaller piglets has increased too. On many pig farms, mortality during lactation exceeds 15% (Borges et al., 2005; Cutler et al., 2006). Although the aim is to decrease neonatal mortality significantly, the reality is that there is still a lot of work in this area for the pig sector.

## Conclusion

The lightest piglets seemed to show less pain related behavior when their teeth and tail were left intact. Moreover, mortality rate tended to be lower when compared with the lightest piglets of the control group. Further research specified on neonatal mortality would however be useful. There was no advantage in growth rate for the lightest piglets of the experimental group, which might be related to the tendency for higher survival of (lighter) piglets in this group, downgrading the mean value of growth rate.

The study also made it clear that behavioral observations are not the perfect way to study pain. But as mentioned in the general introduction, the perfect way to study pain in animals is yet to be found. Possibly the parameters used to assess pain-related behaviors in this study did not discriminate between actual pain and discomfort, which might originate from different sources. The specificity and sensitivity of the parameters used in this study may therefore have been too low for detection of actual pain. The differences between control groups demonstrate this, although it can also be seen as both groups showing signs of discomfort, only at different moments in time.

Reducing painful procedures was studied in the present chapter while in the previous chapter anesthesia during castration was investigated. It would be interesting to combine these 2 objectives and to perform all painful procedures on one moment in time, possibly under anesthesia. The detailed experiment is described in the next chapter.

Chapter 5: Painful standard management practices with piglets: does bundling and/or anesthesia improve animal welfare?

Chapter redrafted after: S. Van Beirendonck, B. Driessen, R. Geers (2011). Painful standard management practices with piglets: does bundling and/or anesthesia improve animal welfare? Journal of Anesthesia and Clinical Research 2:157, doi:10.4172/2155-6148.1000157.

### Abstract

Newborn piglets are subjected to several painful management practices during their first week of life. The objective of this study was to investigate whether the bundling of these painful management practices as such and in combination with anesthesia influenced the behavior and/or production results of piglets positively compared to a random application. There were 515 piglets included in this study, which consisted of two experiments. In the first experiment, management practices were carried out spread over the first week ("separate" group, n = 168) or bundled at one week of age ("together1", n = 144). In the second experiment management practices were all bundled at one week of age without anesthesia ("together2" group, n = 97), or bundled at one week of age while the piglets were anesthetized with 100% CO<sub>2</sub> ("anesthesia" group, n = 106). Behavior of the piglets in both experiments was observed from the day of birth until weaning. Behavioral categories were lying down, udder activity, walking, social cohesion, interactive behavior, pain related behavior and postures (sitting, standing and kneeling). Results demonstrate that piglets seem to cope better with pain if painful interventions are not combined. Moreover, the applied CO<sub>2</sub> anesthesia has facilitated the pain experience after treatment, since lying, interactive and walking behavior indicated more discomfort for the anesthetized piglets. Anesthetized piglets had only an advantage when considering nursing behavior.

## Introduction

The objective of this study was to investigate in a first experiment whether the bundling of painful management practices influences the behavior and/or production results of piglets. The bundling of these painful events on a later age also has the advantage that the piglets only have to be picked up once (time saving for the farmer) and that the piglets are left alone the first days of their life which also gives weaker piglets the chance to make a good start. In a second experiment, the objective was to examine whether the bundling of painful management practices with anesthesia influences the behavior and/or production results of piglets positively compared to a random application without anesthesia. The hypothesis that bundling of the procedures might improve welfare was based on the phenomenon of 'pain memory'. Studies on rats (Anand et al., 1998) and humans (Taddio et al., 1997; Peters et al.,

2005) have indicated that individuals who were repeatedly exposed to neonatal pain develop a lowered pain threshold. Wounding and tissue damage at a young age can cause changes in the central nervous system that persist after the wound is healed, and influence the behavioral response on painful procedures months later in the sense that they display a much stronger pain response (Taddio et al., 1997).

The absence of pain experience during the procedures for piglets of the anesthetized group is undoubtedly advantageous to piglet welfare, but the objective of the study was to look at the effects afterwards. The present study attempts to contribute to the debate on farm animal welfare and the discussion on painful management practices.

## Material and methods

### Animals and Housing

A total of 515 hybrid piglets (Piétrain x Hypor), being heterozygous for the halothane gene, out of 41 litters were used. Housing conditions were the same as described in chapter 3.

### Experimental design

Piglets were handled according to the Belgian law on the protection of animals, and the experimental protocol was agreed by the Ethical Committee on the use of experimental animals. Two experiments were conducted in 2 different trials.

### Experiment 1.

In the first experiment, the following 2 treatment groups were compared:

- Management practices carried out as normal, spread in the first week of life (SEPARATE, n = 168 out of 12 litters):
  - On day 1: iron injection, ear tagging, tooth resection, tail docking
  - On day 7: Vaccination for Mycoplasma hyopneumoniae (Stellamune Mycoplasma, Intervet), 0.5 ml Amoxycilline (Duphamox, Fort Dodge) injection, castration (male piglets, about half of the litter)
- Management practices all carried out together on day 7 (TOGETHER 1, n = 144 out of 12 litters).

All management practices were executed before the piglets reached the age of 1 week (EU Directive 91/630/EEG). Piglets were weighed regularly: on the day of birth, on the day of castration, at about 20 kg, 45 kg, 75 kg and before transport to the slaughterhouse ( $\pm$  110 kg). *Experiment 2.* 

In a subsequent experiment, bundling of management practices was examined further. Two treatment groups were compared:

- Management practices all carried out together on day 7, (TOGETHER 2, n = 97 out of 9 litters).
- Management practices all carried out together on day 7, piglets were anesthetized with 100% CO<sub>2</sub> before the procedures (ANESTHETIZED, n = 106 out of 8 litters).

Anesthesia with  $CO_2$  has generated different opinions. The noxious effect was too large according to Danneman et al. (1997), while Kohler et al. (1998) concluded that  $CO_2$  - anesthesia can be induced safely and rapidly. A concentration of 70%  $CO_2$  was considered admissible to avoid pain during castration by Gerritzen et al. (2008).  $CO_2$  is already being used to stun animals before slaughter. Only high concentrations above 80% are used for this to keep the aversion period as short as possible, i.e. about 70 sec, and reducing the risk of inadequate anesthesia (Nowak et al., 2007). Therefore, in this experiment 100%  $CO_2$  was used during 25s as being found to be the optimal duration in a preliminary experiment. Anesthesia was administered through a mouth mask.

All management practices were executed before the piglets reached the age of 1 week (EU Directive 91/630/EEG). Piglets were weighed regularly: on the day of birth, on the day of castration, at about 20 kg, 45 kg, 75 kg and before transport to the slaughterhouse ( $\pm$  110 kg).

### **Behavioral observations**

Behavioral observations were carried out in the same way for the 2 experiments, as described in the preliminary experiment of chapter 3 and in table 1.1 (p. 9). Piglets were observed the first time at the day of birth and behavior was followed up regularly until weaning (every Monday, Wednesday and Friday).

### Statistical Analysis

All data were analyzed with SAS 9.2 (SAS Inst, Inc, Cary, NC, USA 2008) software. A significance level of 0.05 was used. Categories of behavior were grouped for analysis in order

to have a sufficient number of observations within each grouped category, so that the empirical estimation could be carried out on a sufficient number of observations. A condition of the applied statistical model is that the performance of the empirical estimation passes the convergence test (Anonymous, 2006), being dependent on the number of observations within each category. Hence, lateral lying, ventral lying and sleeping were grouped under 'lying'; teat seeking, suckling and udder massage were grouped under 'udder activity'; huddled up, trembling, spasms, scratching and tail wagging were grouped under 'pain related behavior'; nosing, chewing, licking, playing and aggression were grouped under 'interactive behavior'; walking and running were grouped under 'walking'; and sitting, standing and kneeling were grouped under 'postures'.

Behavioral data were not normally distributed and were dichotomized using the median as cut-off value. The binary data were analyzed using the logistic mixed model, with the fixed effects being treatment and observation period (time related variations are therefore taken into account), and the random effect being piglet. Random effects accounted for the variability between the piglets within and between litters, hence also for a litter effect. The applied procedure made it possible to allocate a random effect to a variable (Anonymous, 2006), so that piglets could be regarded as the experimental units. There were 2 factors considered in defining this random effect: variability between litters and variability between individual piglets. When taking into account the lowest level in the model, i.e. individual piglets, the variability between piglets partially represented the variability between litters and the possible influence of sow's behavior on piglets' behavior. Sows' behavior was divided into three classes, the lowest class representing resting behavior and the highest class representing more restless behavior; and was added in the model as a covariate. Values are presented as means  $\pm$  SEM.

Data concerning body weight were analyzed using a linear mixed model (SAS, Inst, Inc, Cary, NC, USA 2008), with the litter effect taken into account through the random effect, and were covariated for starting weight.

Data analysis was the same for experiment 1 and 2. However, both data sets were not merged for a global analysis, because experimental conditions were not exactly the same. However, it can be assumed that all potential influential factors, but out of an intended treatment effect, were at random distributed in the same way between treatments within each experiment. Hence, a difference between treatments can be compared between experiments, especially in reference to the common treatment.

## Results

For both experiments, results are reported for all data taken together over time, and separately for every week before weaning, in order to infer the effect of the bundling of the painful management procedures, whether or not in combination with anesthesia, on the behavioral categories studied. Attention will be focused on the complementarities of the behavioral events that are considered to be indicative of pain and discomfort, and also on the so-called positive behavior (interactive behavior, social cohesion) that is important for animal welfare (Fraser and Brown, 1990; Blackshaw et al., 1997).

### **Experiment** 1

### Behavior, all observation periods together

Piglets of the "together1" group were lying down more but showed less activity at the udder, less interactive behavior, less walking and less postures than piglets of the "separate" group (Table 4.2).

**Table 5.2** Behavioral data of all observation periods of experiment 1 taken together, presented as means  $\pm$  SEM <sup>a,b</sup>Within a row and variable, means without a common superscript differ (*P* < 0.05). Only significant *P*-values are shown. Means and SEM are from the non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

	SEPARATE	TOGETHER1	<i>P</i> -value
Lying down	$0.592 \pm 0.00515^{a}$	$0.641 \pm 0.00526^{b}$	< 0.0001
Isolated behavior	$0.00584 \pm 0.000673$	$0.00696 \pm 0.000819$	
Postures	$0.0374 \pm 0.00177^{a}$	$0.0301 \pm 0.00169^{b}$	0.0259
Walking	$0.113 \pm 0.00298^{a}$	$0.0979 \pm 0.00315^{b}$	0.0009
Interactive behavior	$0.0419 \pm 0.00182^{a}$	$0.0289 \pm 0.00169^{b}$	< 0.0001
Pain related behavior	$0.00734 \pm 0.000946$	$0.00876 \pm 0.00115$	
Activity at the udder	$0.202 \pm 0.00406^{a}$	$0.189 \pm 0.00453^{b}$	0.0068

### Behavioral observations, per week

In week 3 and 4, piglets of the "separate" group were more active than piglets of the "together1" group by displaying more udder activity, interactive behavior, walking and postures, and less lying down and isolated behavior. More interactive behavior was also shown by the "separate" group in week 1. Piglets of the "together1" were more active at the udder in week 2 (Table 4.3). No differences were found for pain related behavior.

**Table 5.3** Behavioral data per week in experiment 1, presented as means  $\pm$  SEM. Only behavioral categories which differ significantly (P < 0.05) are shown. Means and SEM are from the non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

Week	Behavior	SEPARATE	TOGETHER1	<i>P</i> -value
1	Interactive	$0.0120 \pm 0.00244$	$0.00413 \pm 0.00178$	0.0230
2	Udder activity	$0.172 \pm 0.0065$	$0.210 \pm 0.00818$	0.0020
3	Lying down	$0.528 \pm 0.0110$	$0.653 \pm 0.0107$	< 0.0001
	Udder activity	$0.262 \pm 0.00914$	$0.166 \pm 0.00792$	< 0.0001
	Interactive	$0.0414 \pm 0.00392$	$0.0249 \pm 0.00330$	0.0052
4	Lying down	$0.457 \pm 0.00824$	$0.588 \pm 0.00914$	< 0.0001
	Udder activity	$0.200 \pm 0.00644$	$0.166 \pm 0.00678$	0.0005
	Interactive	$0.0861 \pm 0.00438$	$0.0568 \pm 0.00413$	< 0.0001
	Walking	$0.179 \pm 0.00620$	$0.125 \pm 0.00612$	< 0.0001
	Isolated	$0.00505 \pm 0.00114$	$0.00936 \pm 0.00167$	0.0427
	Postures	$0.0735 \pm 0.00427$	$0.0477 \pm 0.00380$	0.0004

### Body weight, from week 1 until slaughtering

Mean body weights at castration were 2.37 kg for the "separate" group and 2.16 kg for the "together1" group. There were no statistically significant differences on body weight between treatment groups from day of castration until slaughtering.

## **Experiment** 2

## Behavior, all observation periods together

The "together2" group showed more activity at the udder, but less sitting, standing and kneeling compared to the "anesthetized" group (Table 4.4).

**Table 5.4** Behavioral data of all observation periods of experiment 2 taken together, presented as means  $\pm$  SEM. <sup>a,b</sup>Within a row and variable, means without a common superscript differ (P < 0.05). Only significant *P*-values are shown. Means and SEM are from the non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

	<b>TOGETHER2</b>	ANESTHETIZED	<i>P</i> -value
Lying down	$0.544 \pm 0.00711$	$0.539 \pm 0.00752$	
Isolated behavior	$0.00521 \pm 0.00122$	$0.00823 \pm 0.00158$	
Postures	$0.0598 \pm 0.00307^{a}$	$0.0659 \pm 0.00332^{\rm b}$	0.0013
Walking	$0.141 \pm 0.00452$	$0.152 \pm 0.00518$	
Interactive behavior	$0.0250 \pm 0.00184$	$0.0294 \pm 0.00223$	
Pain related behavior	$0.00285 \pm 0.000652$	$0.00518 \pm 0.00120$	
Activity at the udder	$0.216 \pm 0.00531^{a}$	$0.200 \pm 0.00567^{b}$	0.0296

### Behavioral observations, per week

The piglets of the "together 2" group were more active in week 2 and 3, by showing more udder activity, walking and interactive behavior than the "anesthetized" group (Table 5). The latter group was walking most during week 1, lying most during week 2, sitting, standing and kneeling most in week 3, and showed most udder activity in week 4 (Table 4.5). No differences were found in relation to pain related behavior and isolated behavior.

Week	Behavior	TOGETHER2	ANESTHETIZED	<i>P</i> -value
1	Udder activity	$0.253 \pm 0.0140$	$0.158 \pm 0.0111$	0.0002
	Walking	$0.0798 \pm 0.00841$	$0.131 \pm 0.00924$	0.0464
2	Lying down	$0.623 \pm 0.0109$	$0.543 \pm 0.0133$	0.0168
	Udder activity	$0.183 \pm 0.00813$	$0.241 \pm 0.0105$	0.0003
	Walking	$0.102 \pm 0.00649$	$0.102 \pm 0.00787$	< 0.0001
3	Lying down	$0.452 \pm 0.0130$	$0.518 \pm 0.0136$	< 0.0001
	Udder activity	$0.232 \pm 0.0107$	$0.204 \pm 0.0122$	0.0022
	Interactive	$0.0469 \pm 0.00491$	$0.0376 \pm 0.00507$	0.0064
	Postures	$0.0827 \pm 0.00733$	$0.0828 \pm 0.00740$	0.0471
4	Udder activity	$0.214 \pm 0.0102$	$0.177 \pm 0.0107$	0.0155

**Table 5.5** Behavioral data per week in experiment 2, presented as means  $\pm$  SEM. Only behavioral categories which differ significantly (P < 0.05) are shown. Means and SEM are from the non-transformed data. Significant differences were obtained after analysis of the dichotomized data.

#### Body weight, from week 1 until slaughtering

There were no significant differences on body weight for the two treatment groups, from the day of the treatment until slaughtering.

### Discussion

### **Experiment** 1

*Lying.* Lying down of the piglets was considered by Molony and Kent (1997) as a way to protect themselves from their littermates when they are in pain, by shielding their painful body parts away from the other piglets. Following this reasoning, the differences in lying behavior in the present study turned out in the benefit of the "separate" group. This group was lying down the least during the complete observation period and in week 3 and 4.

*Udder activity.* Concerning activity at the udder, piglets of the "separate" group were more active than piglets of the "together1" group for the overall period of observation. This was also the case in week 3 and 4, while in week 2 piglets of the "together1" group were more active at the udder than piglets of the "separate" group. Noonan et al. (1996) correlated

increased udder activity with pain because of the release of endorphins during suckling, which have an analgesic effect. This was confirmed by Llamas Moya et al. (2008) and Taylor et al. (2001). A lactating domestic sow nurses every 45-70 minutes. The nursing lasts about 4-10 minutes but the actual milk intake occurs only during the brief milk ejection that starts 1-4 minutes after the beginning of the nursing and lasts only 15-20 seconds (Fraser, 1980). Llamas Moya et al. (2008) observed an increase in udder massaging behavior of 4% for castrated piglets in the hours after castration compared to non-castrated handled piglets. Following this line of thought, this would suggest that in week 3, 4 and the overall observation period, the "separate" group may have experienced more pain. Anand et al. (1998) found that repeated exposure to neonatal pain can lead to a lowered pain threshold, which is an explanation that can be applied to the "separate" group.

*Interactive behavior.* Piglets of the "separate" group displayed more interactive behavior than those of the "together1" group for the overall observation period and for the first and third week. In the fourth week however, piglets of the "together1" group showed more interactions. Interactive behavior in this study included nosing, chewing, licking, aggression and playing. Llamas Moya et al. (2008) found that castrated piglets were being less playful in comparison with sham-castrated piglets, which might indicate poor welfare. Blackshaw et al. (1997) specifically label playing behavior as a positive indication of animal welfare. Hay et al. (2003) suggested that reduced oral exploration, such as licking and chewing, may be associated with pain.

In the present study, this would suggest that the "separate" group might have a better state of welfare for the overall observation period and in the first and third week, but in week 4 the "together1" group might have a better state of welfare.

*Walking.* Piglets of the "separate" group tended to walk more than the piglets of the "together1" group in the overall period of observation and in week 4. Llamas Moya et al. (2008) already stated that piglets after castration, in pain, may avoid certain activities such as walking and postures like dog-sitting, to minimize pain. The reduced running around of piglets of the "together1" group, having to cope with a lot of painful procedures at the same time, may possibly be interpreted in this way.

*Isolated behaviour.* Concerning social cohesion, there was only a difference in the last week before weaning. Piglets of the "together1" group isolated themselves more from their

littermates than piglets of the "separate" group. Other studies (Hay et al., 2003; Llamas Moya et al., 2008) observed that piglets after castration, when they were in pain, tended to be more isolated than their pen mates. The most plausible explanation for this is that they tried to avoid contacts with pen-mates that might create more pain. In the present study there was only a difference between the treatment groups in week 4, where this could suggest a better welfare for the "separate" group. However, since all painful procedures took place in week 1, it is not certain that this behavioral difference is due to the painful procedures performed some weeks earlier.

*Postures.* Looking at postures, piglets of the "separate" group sat, stood and kneeled more than the other piglets in the overall observation period and in week 4. Taylor et al. (2001) observed increased sitting and standing after castration, which could be a result of the painful experience. Other studies (McGlone and Hellman, 1988; McGlone et al., 1993; Kielly et al., 1999) however, showed reduced standing after castration. Concerning postures, studies do not agree. Hay et al. (2003) therefore suggested that changes in posture are not fully reliable to assess pain in piglets.

*Pain related behaviour.* There are no differences observed between treatment groups, for the overall observation period and every week separately. It would be expected that piglets of the "separate" group, because of the prolonged (repeated) pain experience, would be more subjected to pain. However, apart from udder activity the other results (interactive behavior, walking), do not point in that direction.

*Body weight.* There were no differences in body weight between the two treatment groups, so that the treatment applied in the farrowing room had no effect on body growth until slaughter.

### **Experiment 2**

*Lying.* In week 2 and 3, the "anesthetized" group lay down more than the "together2" group. Applying the conclusions of Molony and Kent (1997) again, who considered lying down as a strategy of piglets to shield their painful body parts from other piglets; this could be interpreted as a better state of welfare for the "together2" group. In experiment 1 lying behavior was more beneficial for the "separate" group, which might indicate that in

experiment 2 the anesthesia procedure might have increased the impact of the treatment compared to the "together2" group. Hence, the statement of Svendsen (2006), that aversion before losing consciousness is compensated by the fact that piglets experience complete anesthesia and analgesia during castration, might not be completely true when considering the period after treatment.

*Udder activity.* For the overall observation period as well as in week 1, 2 and 3, piglets of the "together2" group displayed more udder activity. In discussing experiment 1, it is already mentioned that piglets in pain increased in udder activity because of the release of endorphins during suckling (Noonan et al., 1996). Extrapolating this to the results of experiment 2, there is an indication that piglets of the "together2" group perceived more pain than piglets of the "anesthetized" group.

*Interactive behavior.* Piglets of the "together2" group performed more interactive behaviors than piglets of the other group in week 3. Several studies (Blackshaw et al., 1997; Hay et al., 2003; Llamas Moya et al., 2008) linked interactive behaviors with an improved welfare. Results of the present study therefore speculate a better state of welfare for piglets of the "together2" group in week 3, based on interactive behavior. In experiment 1, interactive behavior was more beneficial for the "separate" group, which might indicate that the anesthesia procedure might have increased the impact of the treatment compared to the "together2" group.

*Walking.* Walking behavior shifted after 1 week. In the first week, piglets of the "anesthetized" group walked around more than piglets of the "together2" group, while in week 2 it was the contrary. Because treatments were only carried out at the end of the first week, the difference in week 2, right after the treatment, can be considered the most important. Llamas Moya et al. (2008) interpreted avoiding walking in piglets as a way to minimize pain after a painful procedure like castration. For the present study, that would imply that piglets of the "anesthetized" group experience more pain in week 2, after the treatments are carried out. This emphasizes again the additional impact of anesthesia on the pain experience afterwards.

*Postures.* For the overall observation period as well as week 3, piglets of the "anesthetized" group sat, stood and kneeled more than piglets of the "together2" group. As

already mentioned for experiment 1, a number of studies (McGlone and Hellman, 1988; McGlone et al., 1993; Kielly et al., 1999; Taylor et al., 2001; Hay et al., 2003) disagree on sitting and standing in piglets, which makes these behavioral parameters not fully reliable for assessing pain in piglets (Hay et al., 2003).

*Body weight.* There were no differences in body weight between the two treatment groups, so that the treatment applied in the farrowing room had no effect on body growth until slaughter.

## Conclusion

The painful effect of castration is demonstrated again, but the interaction with other painful interventions is new information. Piglets seem to cope better with pain if painful interventions are not combined. Moreover, the applied CO<sub>2</sub> anesthesia has facilitated the pain experience after treatment, since lying, interactive and walking behavior indicated more discomfort for the anesthetized piglets. Anesthetized piglets had only an advantage when considering nursing behavior. Although the beneficial effect of anesthesia during painful procedures is not really confirmed by the results, these results should be interpreted as a 'delayed' pain experience for anesthetized piglets rather than an additional pain experience. This experiment demonstrated that CO<sub>2</sub>-anesthesia is not a perfect technique. The main purpose, providing anesthesia during painful management practices, is realised but disadvantages are excitation during the induction phase and post-operative pain when anesthesia wears off. The question if anesthesia during a painful procedure outweighs the disadvantages of the technique is difficult to answer. Every source of pain or discomfort, including post-operative pain, is a threat to animal welfare. When it comes to gradation of pain in comparing operative pain to post-operative pain, it can only be said that pain is a subjective emotion. It is an individual decision which pain is experienced as 'worse'.

Chapter 6: General discussion, conclusions and perspectives for further research

Piglets on commercial pig farms are subjected to several painful management practices, especially in their first week of life. These practices are ear tagging, iron injection, teeth clipping (although routinely prohibited), tail docking (although routinely prohibited), vaccination, and castration of the male piglets. It has been studied and demonstrated in the past that all these procedures cause pain and/or reduce piglet welfare (Noonan et al., 1994; Goodenough et al., 1999; Taylor et al., 2001; Hay et al., 2003; EFSA, 2004; Hay et al., 2004; Henke and Erhardt, 2004; Prunier et al., 2005; Sutherland et al., 2008; Uman et al., 2008; Marchant-Forde et al., 2009).

Improving animal welfare is advantageous not only for the piglets, but also for the farmers. Animals in good condition indeed give better production results than animals in poor welfare conditions. Creating better piglet welfare also helps to improve the image of the pig sector towards the public opinion. Public concern on animal welfare has grown in the last few years as a result of multiple media campaigns of animal rights organizations.

All these aspects, improving piglet welfare, improving production results and the opinion of the public, were studied in the present thesis. In the next paragraphs, the main results are placed in a broader context.

Welfare of farm animals is not only important for the animals and the farmers (better animal welfare implies better zootechnical results), it is also a topic that is gaining consumer's interest and concern. It is acknowledged nowadays that consumer attitudes towards food production methods are not only dependent on an analytical assessment of risk, benefit, economics and nutrition alone. Ethical and moral considerations may also have an influence on establishing the societal acceptability of a particular production process (Frewer et al., 2005). Consumers have an indirect influence on the income of farmers since they are the ones that have to buy farmers' products. Piglet castration is a sensitive issue that has drawn the attention of the public as a result of several campaigns of animal right organizations. It is also known that the sensitivity to animal welfare might be subjective and potential solutions vary between countries (Bonneau, 1998; Dijksterhuis et al., 2000). Therefore the opinion of Flemish consumers on unanesthetized piglet castration and three possible alternatives was examined in the first chapter to form an idea of the Flemish public's concern to improve animal welfare. The three alternatives that were discussed were immunocastration, castration under anesthesia and raising entire males. All three methods were considered by Huber-Eicher and Spring (2008) as feasible on a short term. Results of the questionnaire demonstrated low consumers' awareness on piglet castration, the reason for it and the fact that it is carried out

without anesthesia or analgesia. This result confirms previous studies (Vanhonacker et al., 2009) and can be explained by the fact that increasing urbanization and separation of food production from consumption has resulted in ignorance of modern production methods and alienation of livestock production (Harper and Henson, 2001). Another possible explanation can be found in the so-called 'voluntary ignorance' (Harper and Henson, 2001) which means that people eat meat and ignore that it comes from animals especially raised for human consumption. When looking at the acceptability and preference of the proposed alternatives, castration under anesthesia was considered the most acceptable, followed by immunocastration, while raising entire males was found the least acceptable. The fact that castration under anesthesia gives people the idea pork will still be completely 'boar taintproof' while raising entire males does not, can explain that these alternatives are respectively the most and the least popular. Immunocastration on the other hand had both equal proponents and opponents. These results agree with the Swiss consumers questioned by Huber-Eicher and Spring (2008). When asked about the need for alternatives and the willingness to pay extra for it, respondents reacted divergent. The majority of respondents claimed that alternatives for unanesthetized piglet castration are necessary, and even wanted a ban on unanesthetized piglet castration. On the other hand, the willingness to pay an extra price for this was in general very low. This behavioral duality can be explained by the 'double standard': in their role of citizens people want farmers to pay more attention to animal welfare, but in their role of consumers they are not willing to support the extra costs for it (De Tavernier et al., 2005). When discussing consumers' willingness to pay a price premium for animal welfare attributes, it must be kept in mind that the attitudes expressed by most people rarely inform their purchasing behavior. They can claim to be concerned about animal welfare without buying welfare-friendly products, and even when they do buy these products this may be for other motives than just concern about animal welfare (Verbeke, 2009). This attitudebehavioral intention gap was already described by Vermeir and Verbeke (2006). Furthermore, it should be noted that self-reported behavior may be subject to tendencies of socially desirable answering patterns or general beliefs (Verbeke and Viaene, 1999).

Because castration under anesthesia was the most accepted alternative for Flemish consumers, as reported in the second chapter, the third chapter involved castration under  $CO_2$ -anesthesia. In the preliminary experiment of the third chapter,  $CO_2$ -anesthesia was compared to anesthesia with zolazepam, tiletamine and xylazine. No differences in behavior were found for the overall period of observation, which may be an indication that  $CO_2$  matches the

anesthetic properties of the combination of Zoletil<sup>®</sup> (tiletamine and zolazepam) and Xyl-M<sup>®</sup> (xylazine). An intramuscular injection of tiletamine-zolazepam and xylazine is commonly used as a preanesthetic for veterinary surgical procedures and to produce short-term restraint (Lefkov and Müssig, 2007). According to Lefkov and Müssig (2007), this combination of drugs is widely used because of its smooth induction and recovery profile. Henrikson et al. (1995) also stated that the combination of Zoletil<sup>®</sup> and Xyl-M<sup>®</sup> produces a complete anesthesia characterized by reliable and rapid induction. Hence, knowing that CO<sub>2</sub> induces the expected anesthetic properties, the main experiment of comparing castration under CO<sub>2</sub>anesthesia with unanesthetized castration could be carried out. The results of this main experiment demonstrated that, when looking at the overall period of observation, a difference in interactive behavior indicates a better welfare for anesthetized barrows (Blackshaw et al., 1997; Hay et al., 2003; Llamas Moya et al., 2008). When looking at observation periods separately, the differences in interactive behavior point in the same direction, in favor of anesthetized barrows. Differences in walking and udder activity shifted between treatment groups, which could mean that at one point one group showed behavioral states indicative of better welfare, while at other points it was the other treatment group. This is confirmed by the observation of pain-related behavior in both treatment groups and supports the statement of Gerritzen et al. (2008) that CO<sub>2</sub> induces analgesia only for a short period of time. CO<sub>2</sub> provides anesthesia and analgesia on the moment of castration, but when anesthesia wears off, so does the state of analgesia.

It can be questioned if behavioral observations are a reliable parameter to differentiate between pain and other emotional states of the piglet. The measurement and evaluation of pain is difficult and ultimately subjective (Bath, 1998). The most objective manner to score pain is therefore to observe behavior and compare it to the normal ethogram. Behavior is the parameter most often used to assess animal pain (Rutherford, 2002). According to Weary et al. (2006), the gold standard for validating response measures comes from studies that examine responses with and without a condition causing pain, and with and without analgesics known to be effective at treating this pain. It is therefore very important to be aware of what is normal behavior, which can be implemented by always including a control group in a study. Like this, an experimental group can always be compared to a 'normal behavior' control group. In our experiment, the unanesthetized barrows were the control group. In a more ideal situation, a sham-castration group should also be included in the study and act as a negative control group. Next to behavior, there are a lot of other parameters that can be used to assess pain but as discussed in the general introduction, there is no ideal pain

assessment parameter. Because measuring physiological parameters induces stress on its own, which biases the results, these were not included in the present study.

Castration is not the only painful procedure piglets are submitted to in their first week of life. Therefore it was studied in the fourth chapter if reducing painful events (in this particular case tail docking and teeth clipping) would improve growth rate, survival and animal welfare. In the experimental group, teeth and tails of the four lightest piglets of each litter (EE) were left intact. The rest of the litter (EC) was treated as usually, all procedures carried out. In the control group the four lightest piglets of each litter (CL) were submitted to all procedures, including tail docking and teeth clipping, and this was also the case for their littermates (CC). The behavioral results concerning udder activity indicated a better welfare for EE (Noonan et al., 1996). Walking behavior on the other hand implied that in the beginning, after the painful procedures, EE benefit from the fact that their teeth and tails were left intact while a few weeks later this benefit has faded out. Weaning weights were the same for both EE and CL, and growth rate only differed in the last week before weaning. In that last week, the four lightest piglets of the control group had a higher daily weight gain than the four lightest piglets of the experimental group. This difference can be explained by the higher survival rate in the experimental group. More piglets had died in the control group, leaving behind the 'stronger', better growing piglets while in the experimental group more lighter piglets survived, lowering the mean daily weight gain of the group.

In this experiment, the consequences of pain in the neonatal period on production results and behavior later in life were studied. It can be questioned though, whether or not it can be justified to apply pain inducing interventions in order to prevent possible problems at a later age. Consequences of neonatal pain on later life are extensively described in literature on human medicine, and are deemed important (Abdulkader et al., 2008). But it can also be considered that every single moment of pain has a negative impact on the welfare of the individual. Animal welfare was described in "Five Freedoms" by the Farm Animal Welfare Council (1979):

- Freedom from thirst and hunger;
- Freedom from discomfort;
- Freedom from pain, injury and disease;
- Freedom to express normal behavior;
- Freedom from fear and distress.

Every procedure that threatens one of these freedoms, like neonatal pain threatens the third freedom, can be regarded as a violation of animal welfare. Every painful procedure therefore has an impact on the welfare of piglets.

In the most common management systems, painful procedures are spread over the piglets' first week of life. This means they are repeatedly confronted with pain. Because research in human neonates (Taddio et al., 1997, Peters et al., 2005) and rats (Anand et al., 1998) has demonstrated that repeated neonatal pain can lead to lowered pain thresholds and thus reduced welfare, it was studied in the fifth chapter if bundling of painful procedures (whether or not under anesthesia) would improve piglet welfare. Two experiments were carried out; in the first experiment bundling the procedures ('together1' group) was compared to separate implementation of the procedures ('separate' group), as common practice. Behavioral results of the first experiment showed positive results for the 'separate' group compared to the 'together1' group for lying, interactive, walking and isolated behavior. Only for activity at the udder the 'together1' group seemed to have an advantage. Results of the first experiment appear to demonstrate that piglets do not benefit from receiving all painful treatments at one moment in time when these procedures are carried out without anesthesia. Therefore in the second experiment bundling of painful management practices without anesthesia ('together2' group) was compared to bundling the procedures after anesthetizing the piglets with CO<sub>2</sub> (100%, 25s) ('anesthetized' group). In this experiment, some behavioral results implicated a welfare benefit for the 'anesthetized' group (activity at the udder) while other results demonstrated an advantage for the 'together2' group (lying, interactive behavior and walking). It would be expected that behavioral results would have pointed out an overall welfare benefit for the 'anesthetized' group. This is not entirely the case because CO<sub>2</sub>anesthesia provides anesthesia and analgesia only for a short period of time (Gerritzen et al., 2008); during the procedure but not for long afterwards. Although the beneficial effect of anesthesia during painful procedures is not really confirmed, these results should be interpreted as a 'postponed' pain experience for anesthetized piglets rather than an additional pain experience. The post-operative pain may be present in all treatment groups, but the absent pain experience during the procedures for piglets of the anesthetized group is still advantageous to piglet welfare. Next to behavior, there were no differences in weight gain or body weight between treatment groups so that the treatment applied in the farrowing room had no effect on body growth until slaughter.

Out of these results and the second experiment, it is clear that CO<sub>2</sub> has certain advantages for piglet welfare but that it is not an ideal solution because the pain after the procedures is still present. Concerning unanesthetized piglet castration and possible alternatives, this is still the main point of discussion; for the moment there is no ideal solution. All available alternatives have advantages and disadvantages. Local anesthesia has the disadvantage that it has to be injected in the testicle, which is also painful (Von Waldmann et al., 1994; Haga and Ranheim, 2005; Kluivers-Poodt et al., 2007). Administering an analgesic (e.g. meloxicam) partly relieves the pain, but does not completely eliminate it (Kluivers-Poodt et al., 2007). Immunocastration excludes surgical castration, which is beneficial to welfare, but the vaccine is not specific for pigs so humans can also be affected when they accidentally inject themselves. Nevertheless, the technique is presented to the public as a vaccination in order to induce association with disease prevention, i.e. a beneficial effect. However, it is in fact an intervention in the animal's hormonal system to change its functioning, i.e. against public opinion on hormonal applications. Hence, some countries are not willing to import pork from immunocastrated pigs, which is an important problem for countries where most of the production has to be exported. On top of that it is a serious investment (cost of the vaccine, more labor, adjusting the stable to raise boars and gilts separately) for pig farmers that are already in financial distress. Also, more aggressive behavior can possibly be seen before the second injection (until then, they are still entire males), which can lead to lesions and stress. Raising entire males also eliminates castration, and is most likely the ultimate solution, but for the moment the risk on boar taint can not be excluded yet and methods to sort out tainted carcasses at the slaughter line are not finalized yet. Like with immunocastration, aggressive behavior can also increase. For both these alternatives, immunocastration and raising entire males, farmers will also have to invest in separate housing for gilts and boars. However, the solution in the future will probably be to raise entire males, combined with a reliable and efficient method to sort out tainted carcasses. A detection method should be very short (seconds to minutes) to obtain a fast result and it should have a sufficiently high sample throughput (analysis capacity of several hundreds of carcasses per hour) to work in practice. Ideally, it should also have a performance that allows 100% correct classification of both acceptable and not-acceptable samples with regard to boar taint. Especially the number of false negatives should be zero (Lundstrom et al., 2009). Over the years several technologies have been investigated like chemical sensor array (electronic noses), mass-spectrometry fingerprinting, ultra-fast gas chromatography, gas-phase spectrometry and biosensors (Wäckers et al., 2011). On the moment, studies are also carried out (e.g. in Germany) to sort out tainted carcasses at the slaughter line by using a soldering iron. This soldering iron is applied to the exposed back fat of the carcass. The adipose tissue is heated, causing a volatilization of androstenone and skatole which can be detected by a trained operator at the slaughter line (Lundstrom et al., 2009). Of course, needing an extra operator at the slaughter line implies extra costs. Wäckers et al. (2011) recently published a study on the ability of wasps to perceive and learn the 3 boar taint compounds indole, skatole and androstenone. For skatole and indole the wasps scored good results, for androstenone on the other hand they showed a weak response. The authors concluded it was a promising method if enhanced training could improve the response to androstenone.

Although different research topics were investigated in the present study, there still remain a lot of opportunities for further investigation. It would be interesting to combine  $CO_2$ -anesthesia with an analgesic to avoid the pain afterwards, to see if that would eliminate most of the pain. If that would be the case, all painful management procedures could be carried out on one moment in time (timesaving for the farmer) without pain experience for the piglets. Also, it could be used as a temporary alternative for unanesthetized castration until the 'ideal solution' comes along or surgical castration will be forbidden. Moreover, the pig industry has engaged itself to stop unanesthetized castration from 01/01/2012 on; surgical castration as such will be forbidden from 01/01/2018.

Further research on reducing painful practices would also be useful to confirm the finding that tail docking and teeth clipping does not necessarily improve zootechnical results or animal welfare. If it could be confirmed that these procedures are redundant, it would mean an improvement of animal welfare for the piglets and a gain of time for the farmer.

By means of conclusion of the thesis, it can be said that reducing painful procedures (tail docking and teeth clipping) improves animal welfare and survival rate to a certain extent. Adjusting the painful procedures by using  $CO_2$ -anesthesia and/or bundling all procedures on one moment in time did not give clear results. Bundling of the procedures without anesthesia did not seem to benefit the piglets. Anesthesia with  $CO_2$  provides anesthesia and analgesia on the moment of the procedure, which improves animal welfare, but it wears off quickly resulting in postoperative pain. When questioning consumers' opinion on piglet castration, the most known procedure for the public, it became clear that general awareness on the subject is still low. On the other hand, when respondents were informed, they felt the need for

alternatives was high. If people will be willing to pay an extra price to improve piglet welfare is questionable and will be a lesson learned in time.

An important aspect that should be considered in the whole study of painful management practices on farm animals is animal integrity. As already explained in the introduction, integrity can be described as the wholeness (completeness) and intactness of the animal and its species-specific balance, as well as the capacity to sustain itself in an environment suitable to the species. All the procedures discussed in this thesis interfere with the integrity of pigs. Next to production results which are important for the farmers, animal welfare and integrity should also be kept in mind when interpreting results.

Appendix

# Enquête biggencastratie

#### Algemeen

- 1) Geslacht:
  - Man
  - □ Vrouw

#### 2) Woonplaats:

- □ Antwerpen
- Limburg
- Oost-Vlaanderen
- □ Vlaams-Brabant
- West-Vlaanderen

#### 3) Woont u:

- Op het platteland
- In de stad

### 4) Leeftijd: .....

#### 5) Wat is het hoogste opleidingsniveau dat u behaalde:

- Secundair onderwijs
- □ Hoger onderwijs (hoge school)
- □ Hoger onderwijs (universiteit)

#### 6) Beroep:

- □ Arbeider
- □ Bediende
- Zelfstandige
- □ Student
- □ Werkzoekende
- Pensioen
- Andere, nl: .....

#### 7) a) Heeft u een agrarische achtergrond?

- I Ja, welke?.....
- □ Nee

#### b) Heeft u iemand in uw familie en/of vriendenkring een agrarische achtergrond?

- 🗆 Ja
- □ Nee

8) Uit hoeveel leden bestaat uw gezin?.....

9) Hoeveel kinderen jonger dan 18 jaar telt uw gezin:.....

### Specifiek

10) Hoeveel maal per week consumeert u varkensvlees (gemiddeld op jaarbasis):

- D Nooit
- □ Zelden (minder dan 1 maal per week)
- □ 1 tot 2 maal per week
- □ 3 tot 4 maal per week

Meer

11) Bent u degene in het gezin die instaat voor de aankoop van voeding?

- Ja
- Nee

12) Kent u de achterliggende reden van biggencastratie?

□ Ja, nl: .....

□ Nee

Biggen worden gecastreerd omdat het vlees van 7-10 % van ongecastreerde, mannelijke slachtvarkens berengeur kan vertonen. Deze geur komt vrij bij het braden van het vlees, bij koude bereidingen is er dus geen probleem. Ook is niet iedereen even gevoelig aan berengeur. (De respondenten krijgen nu de kans om te ruiken aan een skatoloplossing)

13) Ervoer u reeds berengeur? (zoja, waar en wanneer?).....

14) Bent u bereid om verhit (gebakken, gekookt) vlees met een berengeur te eten zodat castratie onnodig wordt?

- П Ja
- Nee

15) Bent u bereid om koude producten waarin vlees met berengeur verwerkt zit (salami, gerookte worst) te eten die geen berengeur bevatten, tenzij ze opgewarmd worden?

- Ja
- Nee

16) Vind u de skatolgeur (concentratie = drempelwaarde voor berengeur) aanvaardbaar?

- Ja
- Nee

#### **Onverdoofde castratie**

Bij onverdoofde castratie worden de teelballen zonder verdoving verwijderd. Wettelijk mag dit enkel overdoofd gebeuren bij biggen die maximaal één week oud zijn. Dit is een snelle en goedkope procedure maar ook pijnlijk/ stresserend voor de dieren.

17) Bent u op de hoogte van het feit dat biggen onverdoofd worden gecastreerd?

- 🗆 Ja
- Nee
- 18) Vindt u dat er een alternatief moet komen voor onverdoofde castratie?
  - Ja
  - Nee
- 19) Vindt u dat onverdoofde castratie verboden moet worden?
  - П Ja
  - П Nee

#### Verdoofde castratie

Verdoving kan plaatselijk (injectie in de teelballen of het scrotum) of algemeen (injectie of inhalatie) gebeuren. Hierdoor gebeurt de castratie zelf pijnloos.

20) Vind u verdoofde castratie een acceptabel alternatief voor onverdoofde castratie?

🗆 Ja

□ Nee

21) Castratie onder verdoving betekent een hogere kostprijs (kosten veearts, verdovingsmiddelen). Bent u bereid meer te betalen voor varkensvlees waarvan de biggen werden gecastreerd onder verdoving?

- 🗆 Ja
- □ Nee

#### Intacte beren

Intacte beren zijn mannelijke varkens die niet gecastreerd worden. Daardoor is er een kans op berengeur. Methoden om karkassen met berengeur in de slachtlijn te detecteren worden onderzocht maar staan momenteel nog niet op punt.

22) Vindt u het kweken van intacte beren een acceptabel alternatief voor onverdoofde biggencastratie?

- 🗆 Ja
- □ Nee

23) Bent u bereid om meer te betalen voor varkensvlees als dieren onder deze vorm worden opgekweekt?

- 🗆 Ja
- □ Nee

#### Immunocastratie

Het doel van immunocastratie (vaccinatie tegen berengeur) is castratie onnodig maken doordat de ontwikkeling van de teelballen wordt afgeremd. De vaccinatie werkt in op het hormonaal systeem van het varken, waardoor de puberteit uitgesteld wordt, zonder evenwel 100% te garanderen dat de berengeur zich ontwikkelt, althans volgens de reeds beschikbare kennis.

24) Vindt u het toedienen van een vaccin dat tijdelijk de teelballen, de mannelijke hormonen en de berengeur reduceert een goed alternatief voor castratie bij biggen?

- 🗆 Ja
- □ Nee

25) Bent u bereid om meer te betalen voor vlees afkomstig van deze dieren?

- 🗆 Ja
- □ Nee

#### Algemeen

26) Na het invullen van deze enquête, wat verkiest u (meerdere antwoorden mogelijk)?

- Geen castratie waardoor het vlees het risico loopt op het bezitten van een berengeur
- Onverdoofde castratie
- □ Verdoofde castratie met pijnnazorg
- □ Immunocastratie
- □ Ik eet geen varkensvlees meer

35) Was u, voor het invullen van deze enquête, op de hoogte van de problematiek rond biggencastratie?

- 🗆 Ja
- □ Nee

**Cited References** 

Abbott Laboratories (2001). Sevorane.

http://www.safety.vanderbilt.edu/pdf/hcs\_mds/sevoflurane.pdf

- Abdulkader, H. M., Freer, Y., Garry, E. M., Fleetwood-Walker, S. M. and McIntosh, N. (2008). Prematurity and neonatal noxious events exert lasting effects on infant pain behaviour. Early Human Development 74, 351-355.
- Anand, K. J. S. (1990). Neonatal responses to anesthesia and surgery. Clinical Perinatology 17, 207-214.
- Anand, K. J. S. and Craig, K. D. (1996). New perspectives on the definition of pain. Pain 67, 3-6.
- Anand, K. J. S., Coskun, V., Thrivikraman, K. V., Nemeroff, C.B. and Plotsky, P. M. (1998). Long-term behavioral effects of repetitive pain in neonatal rat pups. Physiology & Behavior 66-4, 627-637.
- Andrews, K. and Fitzgerald, M. (1994). The cutaneous withdrawal reflex in human neonates: Sensitization, receptive fields and the effects of contralateral stimulation. Pain 51, 95-101.
- Animal Sciences Group (2007). Verdoofd castreren in de varkenshouderij. Rapport 73, Wageningen UR.
- Anonymous (2006). The Glimmix Procedure. SAS Institute Inc., Cary, NC, 256p.
- Axiak, S. M., Jaggin, N., Wenger, S., Doherr, M. G. and Schatzmann, U. (2007). Anaesthesia for castration of piglets: comparison between intranasal and intramuscular application of ketamine, climazolam and azaperone. Schweizer Archiv fur Tierheilkunde 149-9, 395-402.
- Barnett, S. W. (2007). Manual of animal technology. Iowa: Blackwell Publishing.
- Basbaum, A. I. and Woolf, C. J. (1999). Pain. Current Biology 9-12, 429-430.
- Bataille, G., Rugraff, Y., Meunier-Salaün, M. C., Brégeon, A. and Prunier, A. (2002).
  Conséquences comportementales, zootechniques et physiologiques de l'épointage des dents chez le porcelet. Journées de la Recherche Porcine en France 34, 203-209.
- Bateson, P. (1991). Assessment of pain in animals. Animal Behaviour 42, 827-839.
- Bath, G. F. (1998). Management of pain in production animals. Applied Animal Behaviour Science 59, 147-156.
- Baxter, E. M., Jarvis, S., D'Eath, R. B., Ross, D. W., Robson, S. K., Farish, M., Nevison, I.M., Lawrence, A. B. and Edwards, S. A. (2008). Investigating the behavioural and physiological indicators of neonatal survival in pigs. Theriogenology 69, 773-783.
- Berndtson, W. E. (1991). A simple, rapid and reliable method for selecting or assessing the number of replicates for animal experiments. Journal of Animal Science 69, 67-76.

Besson, J. M. (1999). The neurobiology of pain. The Lancet 353, 1610-1615.

- Blackshaw, J. K., Swain, A. J., Blackshaw, A. W., Thomas, F. J. M. and Gillies, K. J. (1997). The development of playful behavior in piglets from birth to weaning in three farrowing environments. Applied Animal Behaviour Science 55, 37-49.
- Bonneau, M. (1982). Compounds responsible for boar taint, with special emphasis on androstenone: a review. Livestock Production Science 9, 687-707.
- Bonneau, M. (1998). Use of entire males for pig meat in the European Union. Meat Science 49-1, 257–272.
- Bovenkerk, B., Brom, F. W. A. and Van Den Bergh, B. J. (2002). Brave new birds: the use of 'animal integrity' in animal ethics. Hastings Center Report 32-1, 16-22.
- Bracke, M. B. M., Hulsegge, B., Keeling, L. and Blokhuis, H. J. (2004). Decision support system with semantic model to assess the risk of tail biting in pigs:1. Modelling. Applied Animal Behaviour Science 98, 165-182.
- Breuer, K., Sutcliffe, M. E. M., Mercer, J. T., Rance, K. A., O'Connell, N. E., Sneddon, I. A. and Edwards, S. A. (2005). Heritability of clinical tail-biting and its relation to performance traits. Livestock Production Science 93, 87-94.
- Brookes, J. B. and Lean, I. J. (1993). Teeth clipping in piglets. Animal Production 56, 437.
- Brooks, J. and Tracey, I. (2005). From nociception to pain perception: imaging the spinal and supraspinal pathways. Journal of Anatomy 207, 19-33.
- Brown, J. M. E., Edwards, S. A., Smith, W. J., Thompson, E. and Duncan, J. (1996). Welfare and production implications of teeth clipping and iron injection of piglets in outdoor systems in Scotland. Preventive Veterinary Medicine 27, 95-105.
- Calvino, B. and Grilo, R. M. (2006). Central pain control. Joint Bone Spine 73, 10-16.
- Cazaux, G. (2001). Mensen en andere dieren Hun onderlinge relaties meervoudig bekeken. Leuven-Apeldoorn: Garant-Uitgevers.
- Cazaux, G., Van Gijseghem, D. and Carels, K. (2008). Dierenwelzijn in de Vlaamse landbouw en visserij op de onderzoeksagenda. Brussel: Departement Landbouw en Visserijafdeling, Monitoring en Studie. pp. 1-79.
- Chermat, A. (2006). Le cannibalism chez le porc charcutier: approaches zootechnique, physiologique et comportementale. Thèse de Docteur Vétérinaire, Ecole Nationale Vétérinaire de Nantes, France.
- Clarke, I., Walker, J., Hennessy, D., Kreeger, J., Nappier, J. and Crane, J. (2008). Inherent

food safety of a synthetic gonadotropin releasing factor (GnRF) vaccine for the control of boar taint in entire male pigs. International Journal of Applied Research in Veterinary Medicine 6:1, 7-14.

- Cravero, J. P. and Rice, L. J. (2001). Pediatric anesthesia. In: Clinical Anesthesia. 4<sup>th</sup> edition, Philadelphia: Lippincot-Williams and Wilkins.
- Danneman, P. J., Stein, S. and Walshaw, S. O. (1997). Humane and practical implications of using carbon dioxide mixed with oxygen for anesthesia or euthanasia of rats. Laboratory Animal Science 47 (4), 376-385.
- Dawkins, M.S. (2003). Behaviour as a tool in the assessment of animal welfare. Zoology 106, 383-387.
- de Roest, K., Montanari, C., Fowler, T. and Baltussen, W. (2009). Resource efficiency and economic implications of alternatives to surgical castration without anaesthesia. Animal 3:11, 1522-1531.
- De Tavernier, J., Lips, D. and Aerts, S. (2005). Dier en Welzijn. Tielt, Belgium: Terra-Lannoo nv.
- Delbor, C., Beaudeau, F. and Berger, F. (2000). Intérêt de la section des dents et de l'injection de fer pour des porcelets nés en plein air. Journées de la Recherche Porcine en France 32, 129-134.
- Dijksterhuis, G. B., Engel, B., Walstra, P., Furnols, M. F. I., Agerhem, H., Fischer, K., Oliver, M. A., Claudi-Magnussen, C., Siret, F., Beague, M. P., Homer, D. B. and Bonneau, M. (2000). An international study on the importance of androstenone and skatole for boar taint: II. Sensory evaluation by trained panels in seven European countries. Meat Science 54-3, 261–269.
- Dubin, D. (1999). Snelle interpretative van ECG's. Maarsen, Elsevier/De Tijdstroom.
- Dunshea, F. R., Colantoni, C., Howard, K., McCauly, I., Jackson, P., Long, K. A., Lopaticki, S., Nugent, E. A., Simons, J. A., Walker, J. and Hennessy, D. P. (2001). Vaccination of boars with a GnRH vaccine (Improvac) eliminates boar taint and increases growth performance. Journal of Animal Science 79, 2524-2535.
- Eisele, J. H., Eger, E. I. and Muallem, M. (1967). Narcotic proprties of carbon dioxide in the dog. Anesthesiology 28-5, 856-865.
- European Food Safety Authority (2004). Welfare aspects of the castration of piglets. The EFSA Journal 91, 1-100.
- EUFIC: The European Food Information Council (2010). What causes boar taint? http://www.eufic.org/page/en/page/FAQ/faqid/What-causes-boar-taint/

- European Medicines Agency (2008). Status of MRL procedures, MRL assessments in the context of Council Regulation (EEC) No 2377/90. EMEA/CVMP/765/99-Rev.22. <u>http://www.emea.europa.eu/pdfs/vet/mrls/076599en.pdf</u>
- Farm Animal Welfare Council (1979). Farm Animal Welfare Council, press statement. http://www.fawc.org.uk/pdf/fivefreedoms1979.pdf
- Fauci, A. S., Kasper, D. L., Braunwald, E., Hauser, S. L., Longo, D. L., Jameson, J. L. and Loscalzo, J. (s.a.). Harrison's Principles of Internal Medicine, 17<sup>th</sup> edition: http://www.accessmedicine.com
- Faucitano, L. and Schaefer, A. L. (2008). Welfare of pigs: from birth to slaughter.Wageningen, Versailles: Wageningen Academic Publishers.
- Faulkner, P. M. and Weary, D. M. (2000). Reducing pain after dehorning in dairy calves. Journal of Dairy Science 83, 2037-2041.
- Fitzgerald, M. (1994). Neurobiology of foetal and neonatal pain. In: Textbook of pain. 3<sup>rd</sup> Edition, London, Churchill Livingstone, 153-163.
- Font i Furnols, M., Gispert, M., Guerrero, L., Velarde, A., Tibau, J., Soler, J. and Hortós, M. (2008). Consumers' sensory acceptability of pork from immunocastrated male pigs. Meat Science 80, 1013–1018.
- Font i Furnols, M. and Oliver, M. A. (1999). Production and consumption of pork meat with different levels of boar taint. Food Science and Technology International 5, 367-375.
- Forslid, A. (1987). Transient neocortical, hippocampal and amygdaloid EEG silence induced by one minute inhalation of high CO<sub>2</sub> concentration in swine. Acta Physiologica Scandinavica 130, 1-10.
- Fraser, D. (1975). The 'teat order' of suckling pigs: II. Fighting during suckling and the effect of clipping the eye teeth. The Journal of Agricultural Science 84, 393-399.
- Fraser, D. (1978). Observations on the behavioural development of suckling and early weaned piglets during the first six weeks after birth. Animal Behaviour 26, 22-30.
- Fraser, D. (1980). A review of the behavioural mechanism of milk ejection of the domestic pig. Applied Animal Ethology 6, 247-255.
- Fraser, A. F., Broom, D. M. (1990). Farm animal behavior and welfare. Baillière Tindall, London, 437p.
- Frewer, L. J., Howard, C., Hedderley, D. and Shepherd, R. (1997). Consumer attitudes towards different food-processing technologies used in cheese production-the influence of consumer benefit. Food Quality and Preference 8-4, 271-280.
- Frewer, L. J., Kole, A., Van De Kroon, S. M. A. and De Lauwere, C. (2005). Consumer

attitudes towards the development of animal-friendly husbandry systems. Journal of Agricultural and Environmental Ethics 18, 345-367.

- Gallois, M., Le Cozler, Y. and Prunier, A. (2005). Influence of tooth resection in piglets on welfare and performance. Preventive Veterinary Medicine 69, 13-23.
- Geers, R., Goedseels, V., Parduyns, G. and Vercruysse, G. (1986). The group postural behaviour of growing pigs in relation to air velocity, air and floor temperature. Applied Animal Behavioural Science 16, 353-362.
- Geers, R., Decanniere, C., Villé, H., Van Hecke, P., Goedseels, V., Bosschaerts, L., Deley, J., Janssens, S. and Nierynck, W. (1992). Identification of halothane gene carriers by use of in vivo <sup>31</sup>P nuclear magnetic resonance spectroscopy in pigs. American Journal of Veterinary Research 53-9, 1711-1714.
- Gentle, M. J. (2001). Attentional shifts after pain perception in the chicken. Animal Welfare 10, 187-194.
- Gerritzen, M. A., Kluivers-Poodt, M., Reimert, H. G. M., Hindle, V. and Lambooij, E. (2008). Castration of piglets under CO<sub>2</sub>-gas anaesthesia. Animal 2 (11), 1666-1673.
- Giersing, M., Ladewig, J. and Forkman, B. (2006). Animal welfare aspects of preventing boar taint. Acta Veterinaria Scandinavica 48, 7-9.
- Goodenough, B., Thomas, W., Champion, G. D., Perrott, D., Taplin, J. E., von Bayer, C. L. and Ziegler, J. B. (1999). Unravelling age effects and sex differences in needle pain: ratings of sensory intensity and unpleasantness of venipuncture pain by children and their parents. Pain 80, 179-190.
- Haga, H. A. and Ranheim, B. (2005). Castration of piglets: the analgesic effects of intratesticular and intrafunicular lidocaine injection. Veterinary Anaesthesia and Analgesia 32, 1-9.
- Hansen, B. (1997). Through a glass darkly: using behavior to assess pain. Seminars in Veterinary Medicine and Surgery (Small Animal) 12:2, 61-74.
- Harper, G. and Henson, S. (2001). Consumer Concerns About Animal Welfare and the Impact on Food Choice. EU FAIR CT98-3678 Final Report 38 pp. http://europa.eu.int/comm/food/animal/welfare/eu\_fair\_project\_en.pdf.
- Hay, M., Vulin, A., Genin, S., Sales, P. and Prunier, A. (2003). Assessment of pain induced by castration in piglets: behavioural and physiological responses over the subsequent 5 days. Applied Animal Behaviour Science 82, 201-218.
- Hay, M., Rue, J., Sansac, C., Brunel, G. and Prunier, A. (2004). Long-term detrimental effects

of tooth clipping or grinding in piglets: a histological approach. Animal Welfare 13 (1), 27-32.

- Hendriksen, C. F. M. and Boumans, I. J. M. M. (2006). Een rapport over de ontwikkelingen in en rondom het proefdiergebruik en alternatieven voor dierproeven. De proefdierbarometer. http://www.vet.uu.nl/nca/userfiles/other/Proefdierbarometer.pdf
- Henke J. and Ehrhardt W. (2004). Anästhesie und Analgesie beim Klein- und Heimtier sowie bei Vögeln, Reptilien, Amphibien und Fischen. Schattauer Verlag, 369-405.
- Henrikson, H., JensenWaern, M. and Nyman, G. (1995). Anaesthetics for general anaesthesia in growing pigs. Acta Veterinaria Scandinavica 36 (4), 401-411.
- Herpin, P., Damon, M. and Le Dividich, J. (2002). Development of thermoregulation and neonatal survival in pigs. Livestock Production Science 78, 25-45.
- Hodgson, D. (2006). An inhaler device using liquid injection of isoflurane for short term anesthesia in piglets. Veterinary Anesthesia an Analgesia 33, 207-213.
- Hodgson, D. (2007). Comparison of isoflurane and sevoflurane for short-term anesthesia in piglets. Veterinary Anesthesia and Analgesia 34, 117-124.
- Holyoake, P. K., Broek, D. J. and Callinan, A. P. L. (2004). The effects of reducing the length of canine teeth in sucking pigs by clipping or grinding. Australian Veterinary Journal 82:9, 574-576.
- Holzki, J. and Kretz, F. J. (1999). Changing aspects of sevoflurane in paediatric anaesthesia. Pediatric Anaesthesia 9, 283-286.
- Houpt, K. A. (2005). Domestic animal behaviour. Iowa: Blackwell Publishing.
- Huber-Eicher B. and Spring P. (2008). Attitudes of Swiss consumers towards meat from entire or immunocastrated boars: A representative survey. Research in Veterinary Science 85, 625–627.
- Hunter, E. J., Jones, T. A., Guise, H. J., Penny, R. H. C. and Hoste, S. (2001). The relationship between tail biting in pigs, docking procedure and other management practices. The Veterinary Journal 161, 72-79.
- Hutter, S., Heinritzi, K., Reich, E. and Ehret, W. (1994). The effect of different methods of tooth resection in piglets. Revue de Medicine Veterinaire 145 (3), 205-213.
- IASP (1979). The need of a taxonomy. Pain 6, 247-252.
- Improvac (2009). Improvac: zichtbaar meer winst. Louvain-La-Neuve, N.Y.: Pfizer Animal Health.
- Jaggin, N., Kohler, I., Blum, J. and Schatzmann, U. (2001). Castration of newborn piglets under inhalation anesthesia with halothane. Der Praktische Tierarzt 82, 1054-1061.

- Jourdan, D., Ardid, D. and Eschalier, A. (2001). Automated behavioural analysis in animal pain studies. Pharmacological Research 43-2, 103-110.
- Kegley, E. B., Spears, J. W., Flowers, W L. and Schoenherr, W. D. (2002). Iron methionine as a source of iron for the neonatal pig. Nutrition Research 22, 1209-1217.
- Kielly, J., Dewey, C. E. and Cochran, M. (1999). Castration at 3 days of age temporarily slows growth of pigs. Journal of Swine Health Production 7-4, 151-153.
- Kiley-Worthington, M. (1976). The tail movements of ungulates, canids and felids with particular reference to their causation and function as display. Behaviour 56, 69-115.
- King, R. H., Mullan, B. P., Dunshea, F.R. and Dove, H. (1997). The influence of piglet body weight on milk production of sows. Livestock Production Science 47, 169-174.
- Kluivers-Poodt, M., Hopster, H. and Spoolder, H. A. M. (2007). Castration under anaesthesia and/or analgesia in commercial pig production. Rapport 73, Animal Sciences Group Wageningen UR, Lelystad.
- Knol, E. F., Leenhouwers, J. I. and Van Der Lende, T. (2002). Genetic aspects of piglet survival. Livestock Production Science 78, 47-55.
- Kohler, I., Moens, Y., Busato, A., Blum, J. and Schatzmann U (1998). Inhalation anaesthesia for the castration of piglets: CO<sub>2</sub> compared to halothane. Journal of Veterinary Medicine 45, 625-633.
- Lahrmann, K. H., Kmiec, M. and Stecher, R. (2006). Piglet castration with ketamine/azaperone-anesthesia: Concurring with animal welfare, practical, but economic? Der Praktische Tierarzt 87-10, 802-809.
- Lapin, S. L., Auden, S. M. and Goldsmith, L. J. (1999). Effects of sevoflurane anaesthesia on recovery in children: a comparison with halothane. Pediatric Anaesthesia 9, 299-304.
- Lefkov, S. H. and Müssig, D. (2007). Tiletamine-zolazepam and xylazine is a potent cardiodepressive combination: a case report. Journal of the American Association for Laboratory Animal Science 46 (6), 63-64.
- Leidig, M. S., Hertrampf, B., Failing, K., Schumann, A. and Reiner, G. (2009). Pain and discomfort in male piglets during surgical castration with and without local anaesthesia as determined by vocalisation and defence behaviour. Applied Animal Behaviour Science 116, 174-178.
- Lewis, E., Boyle, L. A., Lynch, P. B., Brophy, P. and O'Doherty, J. V. (2005). The effect of two teeth resection procedures on the welfare of piglets in farrowing crates. Part I. Applied Animal Behaviour Science 90, 233-249.
- Lin, H. C., Tyler, J. W., Wallace, S. S., Thurmon, J. C. and Wolfe, D. F. (1993). Telazol and

xylazine anesthesia in sheep. Cornell Veterinarian 83-2, 117-124.

- Lips, D. (2004). Op zoek naar een meer diervriendelijke veehouderij in de 21ste eeuw. Proefschrift voorgedragen tot het behalen van de graad van Doctor in de Toegepaste Biologische Wetenschappen, K.U.Leuven.
- Llamas Moya, S., Boyle, L. A., Lynch, P. B. and Arkins, S. (2008). Effect of surgical castration on the behavioural and acute phase responses of 5-day old piglets. Applied Animal Behaviour Science 111, 133-145.
- Loncke, R., Dewulf, J., Vanderhaeghe, C., De Kruif, A. and Maes, D. (2008). Non-infectious causes of piglet mortality before weaning. Part 1: Factors related to the piglet. Vlaams Diergeneeskundig Tijdschrift 78, 20-27.
- Lundström, K., Malmfors, B., Fjelkner-Modig, S. and Szatek, A. (1986). Consumer testing of boar meat in Sweden. Swedish Journal of Agricultural Research 13, 39-46.
- Lundstrom, K., Malmfors, B., Stern, S., Rhydmer, L., Eliasson-Selling, L., Mortensen, A. B. and Mortensen, H. P. (1994). Skatole levels in pigs selected for high lean tissue growth rate on different dietary protein levels. Livestock Production Science 38-2, 125-132.
- Lundstrom, K., and Zamaratskaia, G. (2006). Moving towards taint-free pork: Alternatives to surgical castration. Acta Veterinaria Scandinavica 48-1, 1-4.
- Lundstrom, K., Matthews, K. R. and Haugen, J.-E. (2009). Pig meat quality from entire males. Animal 3:11, 1497-1507.
- Maes, D., Mateusen, B., Van Soom, A., Verdonck, M. and de Kruif, A. (2002). Castreren van biggen en problemen met berengeur. Vlaams Diergeneeskundig Tijdschrift 71, 125-132.
- Magalhaes-Sant'Ana, M., Sandoe, P. and Olsson, I. A. S. (2009). Painful dilemmas: the ethics of animal-based pain research. Animal Welfare 18, 49-63.
- Marchant-Forde, J. N., Lay, D. C. Jr., McMunn, K. A., Cheng, H.W., Pajor, E. A. and Marchant-Forde, R. M. (2009). Postnatal piglet husbandry practices and well-being: The effects of alternative techniques delivered separately.Journal of Animal Science 87, 1479-1492.
- Martini, L., Lorenzini, R. N., Cinotti, S., Fini, M., Giavaresi, G. and Giardino, R. (2000). Evaluation of pain and stress levels of animals used in experimental research. Journal of Surgical Research 88, 114-119.
- Martoft, L., Lomholt, L., Kolthoff, C., Rodriguez, B. E., Jensen, E. W., Jorgensen, P. F., Pedersen, H. D. and Forslid, A. (2001). Effects of CO<sub>2</sub> anaesthesia on central nervous system activity in swine. Laboratory animals 36, 115-126.
- Marx, G., Horn, T., Thielebein, J., Knubel, B. and von Borell, E. (2003). Analysis of pain

related vocalization in young pigs. Journal of Sound and Vibration 266, 687-698.

- McGlone, J. J. and Hellman, J. M. (1988). Local and general anesthetic effects on behaviour and performance of 2- and 7-week-old castrated and uncastrated piglets. Journal of Animal Science 66, 3049-3058.
- McGlone, J. J., Nicholson, R. I., Hellman, J. M. and Herzog, D. N. (1993). The development of pain in young pigs associated with castration and attempts to prevent castration-induced behavioral changes. Journal of Animal Science 71, 1441-1446.
- McGlone, J. J. and Pond, W. (2003). Pig production Biological principles and applications. Thomson Delmar Learning, Clifton Park, 395p.
- McMeekan, C., Stafford, K. J., Mellor, D. J., Bruce, R. A., Ward, R. N. and Gregory, N. (1999). Effects of a local anaesthetic and a non-steroidal anti-inflammatory analgesic on the behavioural responses of calves to dehorning. New Zealand Veterinary Journal 47-3, 92-96.
- Mellor, D. J., Cook, C. J. and Stafford, K. J. (2000). Quantifying some responses to pain as a stressor. In: Moberg, G. P., Mench, J. A. (Eds.), The Biology of Animal Stress. CAB International, Amsterdam. pp. 173-198.
- Mellor, D. J. and Stafford, K. J. (2004). Animal welfare implications of neonatal mortality and morbidity in farm animals. The Veterinary Journal 168, 118-133.
- Melzack, R. and Wall, P. D. (1983). The Challenge of Pain. Basic Books, New York.
- Moinard, C., Mendl, M., Nicol, C. J. and Green, L. E. (2003). A case control study of on-farm risk factors for tail biting in pigs. Applied Animal Behaviour Science 81, 333-355.
- Molony, V. and Kent, J. E. (1997). Assessment of acute pain in farm animals using behavioral and physiological measurements. Journal of Animal Science 75, 266-272.
- Molony, V., Kent, J. E., Hosie, B. D. and Graham, M. J. (1997). Reduction in pain suffered by lambs at castration. Veterinary Journal 153, 205-213.
- Morton, D. B. and Griffiths, P. H. M. (1985). Guidelines on the reception of pain, distress and discomfort in experimental animals and an hypothesis for assessment. Veterinary Record 116, 431-436.
- Mullington, J., Korth, C., Hermann, D. M., Orth, A., Galanos, C., Holsboer, F. and Pollmächer, T. Dose-dependent effects of endotoxin on human sleep. American Journal of Physiology – Regulatory, Integrative and Comparative Physiology 278, 947-955.
- Noonan, G. J., Rand, J. S., Priest, J., Ainscow, J. and Blackshaw, J. K. (1994). Behavioural observations of piglets undergoing tail docking, teeth clipping and ear notching. Applied Animal Behaviour Science 39, 3-4.

- Noonan, G. J., Rand, J. S., Blackshaw, J. K. and Priest, J. (1996). Behavioural observations of puppies undergoing tail docking. Applied Animal Behaviour Science 49, 335-342.
- Nowak, B., Mueffling, T. V. and Hartung, J. (2007). Effect of different carbon dioxide concentrations and exposure times in stunning of slaughter pigs: Impact on animal welfare and meat quality. Meat Science 75, 290-298.
- Oliver, W. T., McCauly, I., Harrell, R. J., Suster, D., Kerton, D. J. and Dunshea, F. R. (2003). A gonadotrophin-releasing factor vaccine (Improvac) and porcine somatotropin have synergistic and additive effects on growth performance in group-housed boars and gilts. Journal of Animal Science 81, 1959-1966.
- O'Reilly, K. M., Harris, M. J., Mendl M., Held, S., Moinard, C., Statham, P., Marchant-Forde, J. And Green, L. E. (2006). Factors associated with preweaning mortality on commercial pig farms in England and Wales. Veterinary Record 159 (7), 193-196.
- Paul, E. S., Moinard, C., Green, L. E. and Mendl, M. (2007). Farmers' attitude to methods for controlling tail biting in pigs. Veterinary Record 160, 803-805.
- Pauly, C., Spring, P., O'Doherty, J. V., Ampuero Kragten, S. and Bee, G. (2009). Growth performance, carcass characteristics and meat quality of group-penned surgically castrated, immunocastrated (Improvac®) and entire male pigs and individually penned entire male pigs. Animal 3:7, 1057-1066.
- Peters, J. W. B., Schouw, R., Anand, K. J. S., van Dijk, M., Duivenvoorden, H. J. and Tibboel, D. (2005). Does neonatal surgery lead to increased pain sensitivity in later childhood? Pain 114, 444-454.
- Prunier, A., Mounier, A. M., Bregeon, A. and Hay, M. (2001). Influence of tail docking, tooth resection and castration on plasma cortisol, ACTH, glucose and lactate in piglets. Proceedings of the 4<sup>th</sup> International Conference on Farm Animal Endocrinology, Parme, Italy, 7-10 October 2001.
- Prunier, A., Hay, M. and Servière, J. (2002). Evaluation et prevention de la douleur induite par les interventions de convenance chez le porcelet. Journées de la Recherche Porcine en France 34, 257-268.
- Prunier, A., Mounier, A. M. and Hay, M. (2005). Effects of castration, tooth resection, or tail docking on plasma metabolites and stress hormones in young pigs. Journal of Animal Science 83, 216-222.
- Prunier, A., Bonneau, M., von Borell, E. H., Cinotti, S., Gunn, M., Fredriksen, B., Giersing,

M., Morton, D. B., Tuyttens, F. A. M. and Velarde, A. (2006). A review of the welfare consequences of surgical castration in piglets and the evaluation of non-surgical methods. Animal Welfare 15, 277-289.

- Raaflaub, M., Genoni, M. and Kampf, D. (2008). Economical impacts of alternatives to the castration of piglets without pain relief. Schweizerische Hochschule fur Landwirtschaft (SHL), Zollikofen, Switzerland.
- Ranger, M., Johnston, C. C. and Anand, K. J. S. (2007). Current controversies regarding pain assessment in neonates. Seminars in Perinatology 31, 283-288.
- Ranheim, B., Haga, H. A. and Ingebrigtsen, K. (2005). Distribution of radioactive lidocaine injected into the testes in piglets. Journal of Veterinary Pharmacology and Therapeutics 28-5, 481-483.
- Ranheim, B. and Haga, H. A. (2006). Local anesthesia for pigs subject to castration. Acta Veterinaria Scandinavica 48 (Suppl I), S13.
- Ring, C., Erhardt, W., Kraft, H., Schmid, A., Weinmann, H. M., Berner, H. and Unshelm, J. (1988). CO<sub>2</sub> anaesthesia of slaughter pigs. Fleischwirtschaft 68, 1304-1307.
- Robert, E. and Taylor, I. (1983). Scientific farm animal production. New York: Macmillan Publishing Company.
- Robert, S., Thompson, B. K. and Fraser, D. (1995). Effect d'une coupe selective des dents à la naissance sur la survie et la croissance des porcelets de faible poids. Journées de la Recherche Porcine en France 27, 1-4.
- Rutherford, K. M.D. (2002). Assessing pain in animals. Animal Welfare11, 31-53.
- Salak-Johnson, J. L. and McGlone, J. J. (2007). Making sense of apparently conflicting data: stress and immunity in swine and cattle. Journal of Animal Science 85, E81-E88.
- Schweigert, F. J., Gürtler, H., Baumane, A., Wahren, M. and Leo, M. (2000). Effect of iron supplementation on plasma levels of vitamins A, E and C in piglets. Livestock Production Science 63, 297-302.
- Short, S. E. (1998). Fundamentals of pain perception in animals. Applied Animal Behaviour Science 59, 125-133.
- Simonsen, H. B., Klinken, L. and Bindseil, E. (1991). Histopathology of intact and docked pigtails. British Veterinary Journal 147-5, 407-412.
- Smith, W. J. (1990). A study of the iron requirements of the intensively reared baby piglet. DPM thesis, Royal College of Veterinary Surgeons.
- Squires, J. (2006). Possibilities for selection against boar taint. Acta Veterinaria Scandinavica 48, 19-21.

- Sutherland, M. A., Bryer, P. J., Krebs, N. and McGlone, J. J. (2008). Tail docking in pigs: acute physiological and behavioural responses. Animal 2-2, 292-297.
- Svendsen, O. (2006). Castration of piglets under CO<sub>2</sub> anaesthesia. Volume 1 of Proc. 19<sup>th</sup> IPVS Congr., Copenhagen, Denmark.
- Svendsen, O. (2006). Castration of piglets under carbon dioxide (CO<sub>2</sub>) anaesthesia. Journal of Veterinary Pharmacology and Therapeutics 29 (1), 54-55.
- Taddio, A., Katz, J., Ilersich, A. L. and Koren, G. (1997). Effect of neonatal circumcision on pain response during subsequent routine vaccination. The Lancet 349, 599-603.
- Taylor, A. A., Weary, D. M., Lessard, M. and Braithwaite, L.A. (2001). Behavioral responses of piglets to castration: the effect of pig age. Applied Animal Behaviour Science 73, 35-45.
- Taylor, N. R., Main, D. C. J., Mendl, M., Edwards, S. A. (2010). Tail-biting: a new perspective. The Veterinary Journal 186, 137-147.
- Torrey, S., Devillers, N., Lessard, M., Farmer, C. and Widowski, T. (2009). Effect of age on the behavioural and physiological responses of piglets to tail docking and ear notching. Journal of Animal Science 87 (5), 1778-1786.
- Tuchscherer, M., Puppe, B., Tuchscherer, A. and Tiemann, U. (2000). Early identification of neonates at risk: traits of newborn piglets with respect to survival. Theriogenology 54, 371-388.
- Turkstra, J. A., Zeng, X. Y., van Diepen, J. T. M., Jongbloed, A. W., Oonk, H. B., van de Wiel, D. F. M. and Meloen, R. H. (2002). Performance of male pigs immunized against GnRH is related to the time of onset of biological response. Journal of Animal Science 80, 2953-2959.
- Udelsmann, A., Lorena, S. E. R. D. S., Girioli, S. U., Silva, W. A., De Moraes, A. C. and Andreollo, N. A. (2008). Hemodynamic effects of local anesthetics intoxication. Experimental study in swine with levobupivacaine and bupivacaine. Acta Cirurgica Brasileira 23-1, 55-64.
- Uman, L. S., Chambers, C. T., McGrath, P. J. and Kisely, S. (2008). A systematic review of randomized controlled trials examining psychological interventions for needle-related procedural pain and distress in children and adolescents: an abbreviated Cochrane review. Journal of Pediatric Psychology 33-8, 842-854.
- Van Beirendonck, S., Driessen, B. and Geers, R. (2009). Biggencastratie onder verdoving. Vlaams Diergeneeskundig Tijdschrift 78, 239-248.
- Van Beirendonck, S., Driessen, B., Verbeke, G. and Geers, R. (2011). Behavior of piglets

with or without carbon dioxide anesthesia. Journal of Animal Science 89:10, 3310-3317.

- Van de Perre, V. (2011). Evaluation of pigs' welfare and meat quality in relation to housing, transport and slaughterhouse procedures. Proefschrift voorgedragen tot het behalen van de graad van Doctor in de Bio-ingenieurswetenschappen.
- Van de Weerd, H. A., Docking, C. M., Day, J. E. L. and Edwards, S. A. (2005) The development of harmful social behaviour in pigs with intact tails and different enrichment backgrounds in two housing systems. Animal Science 80-3, 289-298.
- Vanhonacker, F., Verbeke, W. and Tuyttens, F. (2009). Belgian consumers' attitude towards surgical castration and immunocastration of piglets. Animal Welfare 18, 371-380.
- Velarde, A., Cruz, J., Gispert, M., Carrion, D., Ruiz de la Torre, J. L., Diestre, A. and Manteca, X. (2007). Aversion to carbon dioxide stunning in pigs: effect of carbon dioxide concentration and halothane genotype. Animal Welfare 16, 513-522.
- Verbeke, W. and Viaene, J. (1999). Beliefs, attitude and behaviour towards fresh meat consumption in Belgium: empirical evidence from a consumer survey. Food Quality and Preference 10, 437-445.
- Verbeke, W. (2009). Stakeholder, citizen and consumer interests in farm animal welfare. Animal Welfare 18, 325-333.
- Vermeir, I. and Verbeke, W. (2006).Sustainable food consumption: exploring the consumer "attitude-behavioral intention" gap. Journal of Agricultural and Environmental Ethics 19, 169-194.
- Vinuela-Fernandez, I., Jones, E., Welsh, E.M. and Fleetwood-Walker, S. M. (2007). Pain mechanisms and their implication for the management of pain in farm and companion animals. The Veterinary Journal 174, 227-239.
- Von Waldmann, K.-H., Otto, K. and Bollwahn, W. (1994). Ferkelkastration Schmerzempfindung und Schmerzausschaltung. Deutsche Tierarztliche Wochenschrifte 101, 105-109.
- Wäckers, F., Olson, D., Rains, G., Lundby, F. and Haugen, J.-E. (2011). Boar taint detection using parasitoid biosensors. Journal of Food Science 76-1, s41-s47.
- Walker, B., Jaggin, N., Doherr, M. and Schatzmann, U. (2004). Inhalation anaesthesia for castration of newborn piglets: experiences with isoflurane and isoflurane/N<sub>2</sub>O. Journal of Veterinary Medicine A 51, 150-154.
- Weary, D. M., Braithwaite, L. A. and Fraser, D. (1998). Vocal response to pain in piglets. Applied Animal Behaviour Science 56, 161-172.
- Weary, D. M. and Fraser, D. (1999). Partial tooth-clipping of suckling pigs: effects on

neonatal competition and facial injuries. Applied Animal Behaviour Science 65, 21-27.

- Weary, D. M., Niel, L., Flower, F. C. and Fraser, D. (2006). Identifying and preventing pain in animals. Applied Animal Behaviour Science 100, 64-76.
- Wemelsfelder, F. and van Putten, G. (1985). Behavior as a possible indicator for pain in piglets. Instituut voor Veeteeltkundig Onderzoek "Schoonoord", Zeist, The Netherlands.
- White, R. G., DeShazer, J. A., Tressler, C. J., Borcher, G. M., Davey, S., Waninge, A., Parkhurst, A. M., Milanuk, M. J. and Clemens, E. T. (1995). Vocalization and physiological response of pigs during castration with or without a local anesthetic. Journal of Animal Science 73, 381-386.
- Whittemore, C. T. and Kyriazakis, I. (2005). In: Whittemore's Science and Practice of Pig Production. 3rd edn. Blackwell Publishing, pp 86, 1
- Zamaratskaia, G., Rydhmer, L., Chen, G., Madej, A., Andersson, H. K. and Lundstrom, K. (2005). Boar taint is related to endocrine and anatomical changes at puberty but not to aggressive behavior in entire male pigs. Reproduction in Domestic Animals 40, 500-506.
- Zamaratskaia, G., Andersson, H. K., Chen, G., Andersson, K., Madej, A. and Lundstrom, K. (2008). Effects of a gonadotrophin-releasing hormone vaccine (Improvac<sup>TM</sup>) on steroid hormones, boar taint compounds and performance in entire male pigs. Reproduction in Domestic Animals 43, 351-359.
- Zankl, A., Ritzmann, M., Zöls, S. and Heinritzi, K. (2007). Analysis of efficacy of local anaesthetics administered prior to castration of male suckling piglets. Deutsche Tierärztliche Wochenschrift 144-11, 418-422.
- Zeng, X. Y., Turkstra, J. A., Jongbloed, A. W., van Diepen, J. Th. M., Meloen, R. H., Oonk,
  H. B., Guo, D. Z. and van de Wiel, D. F. M. (2002). Performance and hormone levels of immunocastrated, surgically castrated and intact male pigs fed ad libitum high- and low-energy diets. Livestock Production Science 77, 1-11.

List of publications

## Articles in internationally reviewed scientific journals

Van Beirendonck, S., Driessen, B., Verbeke, G. and Geers, R. (2011). Behavior of piglets after castration with or without carbon dioxide anesthesia. *Journal of Animal Science 89:10*, 3310-3317.

Van Beirendonck, S., Driessen, B. and Geers, R. (2011). Painful standard management practices with piglets: does bundling and/or anesthesia improve animal welfare. *Journal of Anesthesia and Clinical Research 2:157*, doi:10.4172/2155-6148.1000157.

Van Beirendonck, S., Driessen, B., Verbeke, G., Permentier, L., Van de Perre, V. and Geers, R. (2011). Improving survival, growth rate, and animal welfare in piglets by avoiding teeth shortening and tail docking. *Journal of Veterinary Behavior: Clinical Applications and Research* 7:2, 88-93.

Van Beirendonck, S., Driessen, B. and Geers, R. (2012). Belgian consumers' opinion on pork consumption concerning alternatives for unanesthetized piglet castration. *Journal of Agricultural and Environmental Ethics*, DOI 10.1007/s10806-012-9378-y, *in press*.

## Meeting abstracts, presented at international conferences, published in proceedings or journals

S. Van Beirendonck, B. Driessen, R. Geers (2009). Piglet castration under CO<sub>2</sub>-anaesthesia: the effect of CO<sub>2</sub>-gas on blood parameters. *Abstract book of the 15<sup>th</sup> PhD symposium on applied biological sciences*, Leuven (Belgium), 6 November 2009 (p. 30).

S. Van Beirendonck, B. Driessen, R. Geers (2010). The effect of meloxicam administration on the behaviour of piglets after castration. *Book of abstracts of the 61<sup>st</sup> Annual Meeting of the European Association for Animal Production, Heraklion (Greece), 23-27 August 2010* (p. 141). Wageningen, the Netherlands: Wageningen Academic Publisher.

S. Van Beirendonck, B. Driessen, R. Geers (2010). Consumers' opinion on alternatives for unanaesthetized piglet castration. *Book of abstracts of the 61<sup>st</sup> Annual Meeting of the European Association for Animal Production 2010, Heraklion (Greece), 23-27 August 2010* (p. 134). Wageningen, the Netherlands: Wageningen Academic Publisher.

## National publications (reviewed and non-reviewed)

S. Van Beirendonck, B. Driessen, R. Geers (2009). Biggencastratie onder verdoving. *Vlaams Diergeneeskundig Tijdschrift* 78, 239-248.

S. Van Beirendonck, V. Vandeperre, R. Geers, B. Driessen, J. Van Thielen (2009). Kan het couperen van staartjes bijtgedrag vermijden? *Landbouw&Techniek* 22, 14-15.

Driessen, B., Van Beirendonck, S., Geers, R. (2009). La castration des porcelets sous anesthésie au CO<sub>2</sub>: la meilleure alternative ? *L'essentiel du porc* 5, 11-12.

J. Van Thielen, S. Van Beirendonck, B. Driessen, R. Geers (2010). Wil de consument meer betalen voor alternatief? *Landbouw&Techniek* 7, 16-17.