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Catherine Larzul, L. Fontanasi, E. Tholen, M. van Son

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s01

Genetic approaches for rearing entire males

C Larzul[1*], L Fontanesi[2], E Tholen[3], M van Son[4]
[1]GenPhyse, Université de Toulouse, INRA, ENVT, France, [2]University of Bologna, Department of Agricultural and Food Sciences, Italy, [3]Institute of Animal Sciences, Animal Genetics, Germany, [4]Norsvin, Norway
[*]catherine.larzul@inra.fr

Introduction

As far as entire male breeding is concerned, genetic studies have been mainly focused on boar taint either for the understanding of genetic determinism of boar taint or the implementation of selection to reduce boar taint risk without impairing progress on production or reproduction traits. In that respect, the genetic determinism of androstenone and skatole has been studied for a few decades now. Recently, more attention has been paid to other traits of interest mainly related to damaging behaviour and processing meat quality.
Nevertheless, application of selecting against boar taint has been limited in breeding schemes to obtain low boar taint lines. The aim of the review is to present a state of the art and some new opportunities regarding genetic studies that could contribute in the future to facilitate entire male breeding.

Genetic determinism of traits of interest for entire male breeding

Boar taint phenotypes have a high, moderately or low heritability values for androstenone, skatole and human nose score (HNS), respectively and they are moderately to highly correlated (Parois *et al.*, 2018). The identification of genes involved in either androstenone or skatole metabolism has been realised from analysing metabolic pathways in either testis or liver (Zadinova *et al.*, 2016). Several expression studies have been carried out, in both testis and liver, with microarrays technology, and recently, mRNA sequencing provides an interesting development for digging in whole genome expression studies (*e.g.* Drag *et al.*, 2017). Studies for detection of QTL have located several loci throughout the genome (Große-Brinkhaus *et al.*, 2015) but up until now, no causal mutation has been evidenced with an effect on boar taint risk. Results are expected in the near future from whole sequence data analyses and functional studies using gene editing.

Implementation in breeding schemes

The comparison between breeds shows that some breeds or lines present a lower risk of boar taint like the Pietrain breed. In general, the fatter the line, the higher the risk of boar taint (Parois *et al.*, 2018). Selection against boar taint by direct selection (Haberland *et al.*, 2014) or by HNS has proven to be efficient, at least in sire lines (Backus *et al.*, 2018). But all breeding objectives are to be balanced and an important topic is the relationships between boar taint risk and other traits of economic interest. The genetic correlations with production traits such as growth rate, feed efficiency or carcass quality are mostly favourable, and selection applied in sire lines should tend to decrease boar taint. In dam lines, the genetic relationships with reproductive traits still need to be further explored. Genetic markers are being used by exploiting favourable haplotypes. Genomic selection is, to our knowledge, being used to reduce boar taint in some countries, but there have been limited publications on this experience. Another potential source of progress is genome editing for variants decreasing levels of boar taint, but it still has to be evaluated and European legislation will slow down its expansion.

Conclusion

Recently, one of the most promising innovations for selecting against boar taint risk has been the implementation of human nose scoring at the slaughterhouse. To enlarge breeding against boar taint, relevant predictors are still needed that can directly be measured on live animals. In that respect, genomic information and a better understanding of genes involved in boar taint metabolism will provide useful tools for breeding entire males.

Acknowledgements

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s02

Breeding against boar taint: Recent research activities within German pig breeding organizations

C Große-Brinkhaus[1], I Brinke[1], K Roth[1], C Looft[3], J Trautmann Mörlein[2], D Mörlein[2], E Tholen[1*]
[1]Institute of Animal Science, Animal Genetics Group, University of Bonn, Germany; [2]Department of Animal Sciences, Meat Science Group, University of Göttingen, Germany; [3]Animal Breeding and Husbandry, Neubrandenburg University of Applied Sciences, Germany
[*]etho@itw.uni-bonn.de

Application

Performance testing schemes and selection tools against boar taint are currently being established within German pig breeding organizations.

Introduction

Breeding against boar taint is a sustainable alternative to painful surgical castration of male piglets without anesthesia. However, to date the proportion of entire male slaughters in Germany stagnates on a low level (~10%). Frequently mentioned reasons for this are: "incidence of tainted carcasses", "penile injuries", "changing payment systems for entire boars" and "processing difficulties of boar carcasses". Within finalized and on-going projects the effectiveness of breeding against boar taint has been evaluated within the sire breed Pietrain (Pi) and dam lines Landrace (LR)/Lage White (LW) of four German breeding organizations.

Material and Methods

In all projects androstenone (And) and skatole (Ska) were recorded according to the SIDA-HS-SPME-GC/MS protocol, and most purebred and Pi×F1 pigs were genotyped. For about 1 000 carcasses, boar taint perception was recorded by a 10 member trained sensory panel (SENS) on a scale from 0 to 5 (Trautmann *et al.*, 2014). In total 4 000 records for And/Ska and 1 000 records for SENS originating from slaughter pigs (Pi×F1) were available. In the on-going project GIFER ~4 000 entire LR/LW boars are tested for And/Ska. Additionally, the sexual hormone profile of 500 tested males and selected female full sibs were recorded at standardized live weights in order to quantify the genetic relationship between boar taint and fertility.

Results

11.6% of the PixF1 boars had a SENS Score >3 (Figure 1) which can be defined as a conservative threshold for perceivable boar taint deviations. A nonlinear function of And/Ska can be used to fit the risk thresholds for SENS with high accuracy (Mörlein *et al.* 2016). Heritabilities for And, Ska and SENS were 0.6, 0.5 and 0.4, respectively. Genetic correlations between SENS and And/Ska were high (0.65/0.94). Accuracy of genomic prediction was evaluated within all crossbred animals and showed reliabilities up to 0.5. Moreover, the incidence of tainted boar progenies (SENS >2) can be reduced below 1% if only AI-boars with best boar taint breeding values are used. Interpretable antagonism between fertility and boar taint as well as genetically linked QTL for both trait complexes could not be detected within the Pi population and is currently investigated within the dam line breeds.

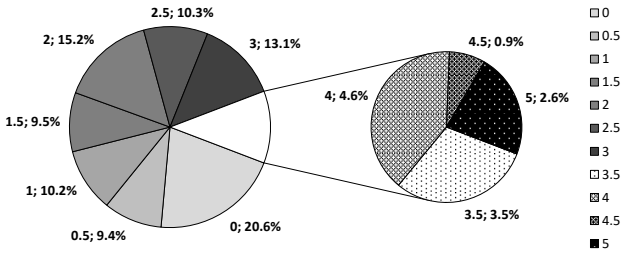


Figure 1 Distribution of human nose scores

Conclusion

Selection tools using And/Ska or verified SENS information are very effective to reduce incidence of boar taint. However, reflecting the on-going discussion in Germany, breeding organizations are still lacking clear signals to include boar taint into the breeding objectives of their nucleus populations.

Acknowledgments

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s03

Genome-wide DNA methylation analysis reveals candidate epigenetic biomarkers of boar taint in pigs

X Wang[1], M Drag[2], H N Kadarmideen[1*]
[1]*Department of Bio and Health Informatics, Technical University of Denmark, Kongens Lyngby, Denmark,* [2]*Department of Veterinary and Animal Sciences, University of Copenhagen, Frederiksberg C, Denmark*
[*]hajak@dtu.dk

Application

The identified candidate epigenetic biomarkers could be used in breeding program against boar taint to avoid surgical castration.

Introduction

The offensive flavour of boar taint (BT) is primarily caused by the accumulation of skatole and androstenone in the non-castrated male pigs (Drag *et al.* 2017). Surgical castration is an effective solution but results in reduced leaner carcass values and increased urinary nitrogen, in addition to animal welfare concerns (European Commission, 2010).

Material and methods

Nine samples of three different BT levels were analysed using Reduced representation bisulfite sequencing (RRBS) data. Methylation levels of cytosine were analysed by R package methylKit. Differentially methylated cytosine (DMC) was defined with regard to CpG islands, CpG island shores and the proximity to the nearest transcription start site (TSS) using R package genomation. Gene ontology (GO) enrichment and pathway terms were analysed by GenCLiP 2.0 software.

Results

The rate of uniquely mapped clean read pairs was 48.7%. The mean distribution of cytosine methylation rate in CpG, CHG and CHH sites were 49.0%, 0.9% and 0.7%, respectively. The distribution of DMC annotation within CpG islands, CpG island shores and other regions were 57.2%, 14.7% and 28.1%, as well as 5.30%, 1.22%, 3.79% and 89.7% in promoter, exon, intron and intergenic regions, respectively. Co-analysis of differentially expressed (DE) genes and significant DMCs found 32 significant co-identified genes. Joint analysis of GO terms and pathways revealed that DMAP1, EGFR and PEMT were very important in regulating gene expression underlying BT (Table 1).

Table 1 Gene annotation of DE genes associated with DMCs

Chromosome	Gene	Gene description	Structure	DMC
6	DMAP1	DNA methyltransferase 1 associated protein 1	Up 10K upstream	$^{a}2$
9	EGFR	Epidermal growth factor receptor	Gene body	$^{a2b1}3$
12	PEMT	Phosphatidylethanolamine N-methyltransferase	Gene body	$^{a2b2}9$

Note: $^{a/b}$Number of DMCs located within CpG islands/CpG island shores.

Conclusion

To our knowledge, we are the first to report epigenetic mechanisms and epigenetic markers using genome-wide DNA methylation profiles for BT in pigs. Results especially for PEMT, could be used in biotechnology and breeding industries.

Acknowledgements

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s04

Mining whole genome resequencing data to identify functional mutations in boar taint-candidate genes

G Schiavo[1], S Bovo[1], S Cheloni[1], A Ribani[1], C Geraci[1], M Gallo[2], G Etherington[3], F Di Palma[3], L Fontanesi[1*]
[1]University of Bologna, Department of Agricultural and Food Sciences, Italy [2]ANAS, Italy, [3]Earlham Institute, Norwich Research Park Innovation Centre, United Kingdom.
[*]luca.fontanesi@unibo.it

Application

Boar taint is an economically relevant meat defect that can be found in meat of sexually mature entire boars. Mining whole genome resequencing data can recover information on the variability of boar taint related genes. The information coming out from this study could highlight polymorphisms useful to understand the genetic architecture of boar taint.

Introduction

Boar taint is a urine-like flavour, strongly disliked by the consumers, caused by the concentration of androstenone and skatole in the fat tissues of mature non-castrated male pigs. To avoid this problem, male pigs are usually castrated at early age. Boar taint is, in part, genetically determined, opening opportunities to reduce this problem using selection programs (Robic *et al.*, 2008, 2016). Whole genome resequencing data from many individuals of different pig breeds have been recently produced using next generation sequencing and datasets are publicly available from DNA databases. In this work, we used these data together with novel whole genome re-sequencing data obtained from several commercial breeds, resulting in a final dataset of 132 individual pigs or DNA-pools. Data derived from several pig breeds and wild boars from different countries. We targeted 135 genes affecting production traits and involved in androstenone and skatole metabolic pathways with the aim to identify polymorphisms useful to describe the genetic variability of the selected traits.

Materials and methods

The final dataset included individuals of 28 different domestic breeds, 3 wild boar groups, and 8 pooled whole genome resequencing datasets generated from 35 individuals each (from Italian commercial and autochthonous breeds). A Q30 was chosen to filter reads. Regions with a depth coverage between 4x and 80x were kept. Reads were mapped on the *Sscrofa11.1* genome version. Variants were called with *samtools* v1.3 and analysed with Ensembl VEP tool (Maclaren *et al.*, 2016).

Results

At a population level, 102 957 single nucleotide polymorphisms (SNPs) were discovered on the targeted genes. About 14% of these SNPs (n = 14 440) were on 27 genes involved in the androstenone and skatole biochemical pathways. Only 53% of these variants were already present in dbSNP (Sherry *et al.*, 2001, build 151). Focusing on VEP results in genes involved in boar taint, 96.2% of these variants affects untranslated or intronic regions, 139 were in coding regions and 69 affected the protein structure. Seventeen of them were considered deleterious after SIFT analysis (Kumar *et al.*, 2009).

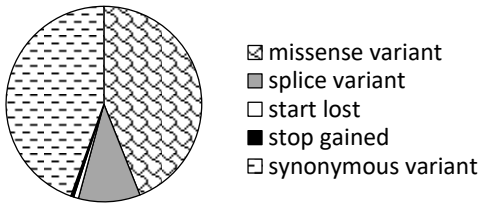


Figure 1 Consequences of the variants affecting coding regions in boar taint related genes considering the whole analysed dataset.

Conclusions

This study enriched the current dbSNP repository of 1263 new variants in the pig genome. In silico prediction indicated that variants in boar-taint related genes might affect their function and might be useful to explain the genetic variability of boar taint phenotype measurements in pigs. Further studies are needed to validate these results.

Acknowledgements

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s05

Characterisation of expression quantitative trait loci associated with androstenone and skatole by sequencing of RNA from porcine testis

M H Drag[1,5], L J A Kogelman[2], H Maribo[3], L Meinert[4], P D Thomsen[1], H N Kadarmideen[1,5*]
[1] *Department of Veterinary and Animal Sciences, Faculty of Health and Medical Sciences, University of Copenhagen, Grønnegårdsvej 7, 1870 Frederiksberg C, Denmark*
[2] *Danish Headache Center, Department of Neurology, Danish Headache Center, Rigshospitalet Glostrup, Faculty of Health and Medical Sciences, University of Copenhagen, Denmark*
[3] *SEGES, Danish Pig Research Center, Axeltorv 3 1609 København V, Denmark*
[4] *Danish Meat Research Institute (DMRI), Danish Technological Institute, Gregersensvej 9 2630 Taastrup, Denmark*
[5] *Department of Applied Mathematics and Computer Science, Technical University of Denmark, Richard Petersens Plads, Building 324, 2800 Kgs. Lyngby, Denmark*
[*]Haja N Kadarmideen, hajak@dtu.dk

Application

A powerful strategy for discovery of candidate genes and genetic regions associated with complex traits is based on the identification of expression quantitative trait loci (eQTLs; Westra and Franke, 2014). The approach has been used on important meat quality traits in pigs (González-Prendes *et al.*, 2017). Analysis of eQTLs may be highly relevant in the development of DNA test for boar taint.

Introduction

Surgical castration to avoid boar taint is an effective solution but is debated due to animal welfare concerns. Gene-based selection for low boar taint has been proposed as concentrations of the main compounds responsible for boar taint (androstenone and skatole [AS]) exhibit heritability (Strathe *et al.*, 2013). The aims of this study were to identify expression quantitative trait loci (eQTLs) associated with summed concentrations of AS.

Material and methods

Gene expression profiles were obtained by RNA-Seq from the testis of 32 Duroc × Landrace/Yorkshire (DLY) male pigs with extreme high and low sire estimated breeding values (EBVs) for androstenone. The pigs were genotyped using loin muscles and AS concentrations were measured by a rapid high-performance liquid chromatographic (HPLC) method on back fat (Hansen-Møller, 1994). The software Matrix eQTL (Shabalin, 2012) was used for *cis* eQTL identification. Significant (FDR < 0.05) eQTLs were analysed with a regression model to identify eQTLs associated to summed concentrations of AS.

Results and conclusions

A relatively high correlation was found between sire EBVs of androstenone and analytical concentrations in the DLY offspring in back fat ($r^2 = 0.48$, $n = 57$), which confirmed heritability of the trait. A total of 462 *cis* eQTLs were found to have significant (FDR < 0.05) association with AS concentrations and genes associated with the eQTLs included *CYP1A2, CYB5D1*, *MAP2K4* and *RDH16*. To our knowledge, this is the first report on eQTLs in testis associated with AS. The top eQTL was associated with *CYP1A2*, which was previously found to be involved in the metabolism of skatole (Rasmussen and Zamaratskaia, 2014). Hence, this study revealed a specific SNP associated with its expression, which might be a potential genetic marker.

Acknowledgements

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s06

Fine mapping of QTL regions for boar taint using whole genome re-sequencing data

M van Son[*1], R Agarwal[2], E Grindflek[1], H Grove[2], M P Kent[2], S Lien[2]
[1]Norsvin, Hamar, Norway, [2]Centre for Integrative Genetics (CIGENE), Department of Animal and Aquacultural Sciences, Faculty of Biosciences, Norwegian University of Life Sciences, Ås, Norway.
*maren.van.son@norsvin.no

Application

Fine mapping of QTL regions on SSC5 and SSC7 for androstenone and skatole using newly detected SNPs from whole genome re-sequencing data identified genetic markers explaining 2.6-5.4 % of the phenotypic variance for these traits. The markers can be used in selection for lower levels of boar taint.

Introduction

Boar taint is an unpleasant taste and/or smell from the meat of some boars and the problem is typically solved by routine castration of male piglets. Due to animal welfare concerns, alternative solutions to surgical castration are desirable, and genetic selection has been proposed as a promising alternative (Merks *et al.*, 2009). Unfavourable correlations to testosterone and oestrogens are challenging and have slowed down implementation. The aim of this study was to fine map QTL regions on SSC5 for androstenone and SSC7 for skatole to identify candidate genes and genetic markers suitable for selection.

Material and Methods

Re-sequencing data from 23 Norwegian Duroc and 24 Norwegian Landrace boars were used for SNP detection in QTL regions. For the association study, 911 Duroc and 1207 Landrace boars with androstenone and skatole measurements were used, and the boars were genotyped with newly detected SNPs. The pipeline for read mapping, SNP detection, genotyping and association mapping can be found in our published papers (van Son *et al.*, 2017a, 2017b).

Results

In the SSC5 QTL region in Duroc, 94 SNPs at 23.03-24.27 Mb were found significant and the most significant SNP explained 5.4 % of the phenotypic variance for androstenone. The SNPs were in high LD and the haplotype block contains at least four positional candidate genes (*HSD17B6, SDR9C7, RDH16* and *STAT6*). In the SSC7 QTL region, the most significant SNPs were found in Landrace at 78.4-79.5 Mb (*Sscrofa10.2*), explaining 5 % of the phenotypic variance for skatole. The SNPs are located in an intergenic region with extensive LD. In Duroc, the most significant SNP in the SSC7 QTL region was found at 80 Mb, explaining 2.6 % of the phenotypic variance for skatole. This SNP was located in the gene *GMZH-like*. None of the SNPs investigated were associated with levels of testosterone or estrogens, making them particularly interesting for selection purposes.

Conclusion

Fine mapping using whole genome sequence data narrowed down the QTL regions investigated and identified genetic markers and candidate genes. High LD made it difficult to identify the causative mutation on SSC5 whereas intergenic SNPs in Landrace on SSC7 could imply regulatory roles.

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s07

Effect of *CYP2E1* gene polymorphism and nutrition on skatole level in crossbred pig

K Zadinova*, R Stupka, J Citek, N Lebedova
Department of Animal Husbandary, Faculty of Agrobiology, Food and Natural Resources, Czech University of Life Sciences Prague, Prague,Czech Republic
*Zadinova@af.czu.cz

Application
The level of skatole can be affected by multiple factors (gene, nutrition, age). Therefore, it is appropriate to reduce it to combine several methods to remove boar taint in pig breeding.

Introduction
Skatole is formed by L-tryptophan degradation in colon. Earlier studies report that feeding diets supplemented with Jerusalem artichoke to entire male pigs can reduce skatole levels in adipose tissue. The initial step of skatole metabolism in the liver involves mainly the enzyme CYP2E1. The association of SPN (c.1423G>A) in *CYP2E1* with skatole level was described by Zadinová *et al*., 2017.

Material and Methods
A total of 25 boars (LWS x Pn) x (LWD x L) were assigned to two different dietary treatment groups with 0% (C) or 8,2 % (E) Jerusalem artichoke included in the diet from 14 days before slaughter. Pigs were genotyped for c.1423G>A according to Zadinová *et al*., 2017 and skatole level was measured according to Okrouhlá *et al*. (2016). Microbiological analysis of 0.5 g of faecal samples from the rectum was also performed. The data were analysed with ANOVA using the GLM procedure in SAS.

Results
Feeding Jerusalem artichoke significantly decreased skatole levels in adipose tissue (P<0.0057; Table 1). Feeding Jerusalem artichoke increased the levels of *Lactobacilli* (P<0.03) and decreased the levels *of E. coli* (P<0.02) in day 12 faeces. Differences in skatole levels were observed between the different genotypes of this SNP. Animals with genotype *AA* had the lowest level of skatole (0.027 µg/g) compared to *GG* (0.041 µg/g) and *AG* (0.038 µg/g). These differences were not statistically significant but this can result from the low number of animals in the experiment.

Table 1 Effect of the inclusion of 8.2% dried Jerusalem artichoke compared to a control diet

	Diets groups			
	C-control	E-experiment	SE	P-value
Skatole (µg/g backfat)	0.0459	0.0246	±0.01	0.0057
Lactobacilli (log CFU/g faeces)				
- Day 0	7.84	7.95	±0.7	0.70
- Day 5	7.52	7.70	±0.5	0.36
- Day 12	7.54	8.13	±0.7	0.03
Escherichia coli (log CFU/g faeces)				
- Day 0	6.75	7.12	±0.5	0.90
- Day 5	7.06	7.28	±0.5	0.29
- Day 12	7.23	6.50	±0.7	0.02

Conclusion
In conclusion, the results of this study show that feeding Jerusalem artichoke decreased the levels of skatole in adipose tissue of crossbred pigs. This could be due to increased competition from probiotic bacteria in the gastrointestinal tract. For SNP c.1423G>A, the genotype *AA* could be associated with reduced skatole level in adipose tissue, but further studies are needed to confirm this result.

Acknowledgments
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s08

Effect of divergent selection for cortisol level on boar taint

C Larzul[1*], E Terenina[1], Y Billon[2], L Gress[1], R Comte[3], A Prunier[3], P Mormede[1]
[1]GenPhySE, Université de Toulouse, INRA, ENVT, Castanet-Tolosan, [2]GenESI, Surgères, [3]PEGASE, INRA, Agrocampus Ouest, St-Gilles, France
[*]catherine.larzul@inra.fr

Application

Selection for a more active HPA axis might induce a higher boar taint risk in entire male pigs due to its incidence on steroidogenesis.

Introduction

Animal breeding is more and more challenged by antibiotics use, environment control with climate change, feed competition and animal welfare. To face these challenges, animal breeding and genetics can provide a sustainable option by improving animal robustness. It is known that the hypothalamic-pituitary-adrenocortical (HPA) axis plays a major role in metabolic regulations and adaptive responses and could provide a potential leverage for more robust animals (Mormede and Terenina, 2012). The HPA axis activity was estimated by plasma cortisol level measured one hour after injection of ACTH. The present study investigates the consequences of selecting for HPA axis activity on *post mortem* traits of interest for entire male breeding focusing on boar taint compounds.

Material and methods

The test was performed on piglets at 6 weeks of age. From a base population of Large White pigs, two divergent lines were selected for 3 generations, for high (HC) and low (LC) post ACTH cortisol levels, respectively. At the 3rd generation of selection, the divergence between the two lines was about 5 genetic standard deviations (Larzul *et al.*, 2018). At 158 days of age, thirty-two entire male pigs were slaughtered with or without mixing before slaughtering. A blood sample was collected at exsanguination for testosterone (ng/ml), estradiol (pg/ml) and cortisol analysis (ng/ml). Urine was collected at carcass evisceration directly from the bladder for adrenaline, noradrenaline, dopamine, deconjugated and total cortisol analysis (µg/g creatinine). A fat sample was taken at the neck level 24 hours post mortem for androstenone and skatole analysis (µg/g). Skin lesions were counted on the carcass. All measurements were log transformed for statistical analysis. Data were analysed with a model including line and pre-slaughter treatment and their interaction.

Results

The interaction between line and pre-slaughter treatment was not significant. Adrenaline, noradrenaline, and deconjugated cortisol were not affected by mixing nor line effect. Line effect was nearly significant on skatole and testosterone level (P=0.0625 and P=0.0735, respectively). The number of skin lesions was increased with mixing due to fighting. The cortisol was elevated in both plasma and urine for the HC line but only urinary cortisol was increased by mixing. Androstenone was higher in the HC line and in a less extent estradiol.

Table 1 Effect of line and pre-slaughter mixing, lsmeans and P-value for log transformed traits. Lsmeans with different letters are significantly different at the P<0.05 level.

		Unmixed		Mixed			P-value	
Traits	N	HC line	LC line	HC line	LC line	RMSE	Mixing	Line
Plasma Cortisol	29	4.29^{a}	2.99^{b}	4.45^{a}	3.48^{b}	0.52	0.1076	<.0001
Androstenone	32	0.19^{a}	-0.54^{b}	0.19^{a}	-0.20^{ab}	0.64	0.451	0.021
Estradiol	29	3.84	3.29	3.86	3.5	0.58	0.588	0.049
Dopamine	29	2.87^{a}	2.04^{b}	2.81^{a}	2.49^{ab}	0.60	0.390	0.018
Skin lesions	32	3.63^{a}	3.53^{a}	4.46^{a}	4.58^{a}	0.58	<.0001	0.970
Urinary Cortisol	29	4.45^{a}	3.94^{b}	4.76^{a}	4.39^{ab}	0.43	0.025	0.011

Conclusion

This experiment confirmed the effect of mixing on cortisol levels and skin lesion scores. It also provided evidence that selection for cortisol level influences steroidogenesis in entire male pigs.

Acknowledgements

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s09

Genetic determinism of boar taint and relationship with meat traits

C Dugué*[1], A Prunier[2], M J Mercat[3], M Monziols[3], B Blanchet[4], C Larzul[1]
[1]*GenPhySE, université de Toulouse, INRA, ENVT, Castanet-Tolosan,* [2]*PEGASE, INRA, Agrocampus Ouest, St-Gilles,* [3]*IFIP, Le Rheu,* [4]*UEPR, INRA, Le Rheu, France.*
*claire.dugue@inra.fr

Application The genetic parameters estimated between two selection criteria against boar taint and meat traits give insight in improving meat quality in entire male pigs.

Introduction Entire male meat may have a major quality defect called boar taint, partly due to the presence of androstenone in fat. The purpose of this study is to evaluate the feasibility of a selection to directly decrease the androstenone level in back fat, or indirectly by a selection on the level of estradiol in plasma, a criterion highly correlated to androstenone level (Parois *et al.,* 2015) and to estimate the consequences on meat production and quality traits in purebred or crossbred pigs.

Material and Methods Pure Pietrain and Pietrain Large White crossbred pigs were measured for hormones levels (estradiol and testosterone) in plasma one week before slaughter. They were also measured for growth traits (average daily gain, feed conversion ratio, average daily feed intake), carcass composition (carcass yield, lean percentage) and quality traits (pH in *Ld* and ham, drip loss and intramuscular fat in loin and back fat androstenone level). The number of skin lesions was measured at three stages. Carcass additional measures were obtained by computerized tomography (CT) (loin eye area, loin eye density, femur density, ham muscle/bone length ratio). The number of measured animals varied from 712 purebred and 736 crossbred for growth traits to 553 purebred and 556 crossbred for CT measurements. Genetic parameters were estimated with the VCE6 software.

Results As expected heritabilities were of medium values for estradiol level (0.23 in purebred, 0.17 in crossbred) and high values for androstenone level (0.57 in purebred and 0.71 in crossbred). A selection to decrease back fat androstenone level in purebred would lead to a decrease in testosterone level (r_g=0.8), feed conversion ratio (r_g=0.47) and skin lesions number at fattening stage entrance (r_g=0.89), and to an increase in carcass yield (r_g=-0.5), lean percentage (r_g=-0.47), and in purebred pigs, increase in ham muscle/ bone length ratio (r_g=-0.51) and pH of ham (r_g=-0.4). This selection should also decrease femur density (0.27), average daily consumption (r_g=0.39), feed conversion ratio (r_g=0.51) and drip loss (r_g=0.4) and increase loin eye area (r_g=-0.5) in crossbred pigs. A selection to decrease estradiol in purebred would decrease back fat androstenone level in purebred (r_g=0.89) and in crossbred (r_g=0.8) and feed conversion ratio (r_g=0.55). In purebred, it would increase loin eye area (r_g=-0.46), ham muscle/ bone length ratio (r_g=-0.38) and ham pH (r_g=-0.47). The favourable effects on crossbred pigs would be a decrease of skin lesions number at fattening stage entrance (r_g=0.99) and increase of carcass yield (r_g=-0.46) and lean percentage (r_g=-0.44). It would lead to unfavourable effects on testosterone level in purebred (r_g=0.82) and in crossbred (r_g=0.57).

Table 1 Expected effects of a selection against androstenone or against estradiol

Selection against	Effects	Purebred	Crossbred
Androstenone h^2 = 0,57 in purebred h^2 = 0,71 in crossbred	Favourable effects on:	CT carcass measures Growth traits Technological quality	CT carcass measures Growth traits Technological quality Carcass composition Skin lesions
	Unfavourable effects on:	Testosterone level	CT bone measures
Estradiol h^2 = 0,23 in purebred h^2 = 0,17 in crossbred	Favourable effects on:	Androstenone level Growth traits Carcass composition Technological quality	Androstenone level Skin lesions Carcass composition
	Unfavourable effects on:	Testosterone level	Testosterone level

Conclusion Heritability and genetic correlations indicate that a selection to decrease estradiol level would have overall favourable effects on meat production and quality traits.

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Influence of terminal sire on fattening traits of gilts and castrated, immunocastrated and entire male pigs

G Kušec[1], I Djurkin Kušec[1*], M Škrlep[2], K Gvozdanović[1], V Margeta[1]
[1]*Faculty of Agrobiotechnical Sciences Osijek, University of Osijek, V.Preloga 1, 31000 Osijek, Croatia*,
[2]*Agricultural Institute of Slovenia, Hacquetova ulica 17, 1000 Ljubljana, Slovenia*
[*]idurkin@pfos.hr

Application When considering the introduction of immunocastrated and/or entire male pigs in the pork chains, the influence of breed on fattening traits should be taken into account.

Introduction At present time, it can be speculated that surgical castration is to be abandoned soon and that entire male pigs as well as immunocastrates will be introduced in the pig production in a larger scale than nowadays. The differences in fattening traits between genders have been well documented (Bonneau *et al.*, 1994; Batorek *et al.*, 2012; Puls *et al.*, 2013). The aim of the present study was to investigate the influence of terminal sire breed on some fattening traits of four different pig genders (gilts, barrows, immunocastrated pigs and entire males).

Material and methods The investigation was carried out on 240 fatteners reared on a commercial farm in Croatia. Sows were inseminated using the semen from three terminal sires: line A (Pietrain x Large White), line B (Pietrain NN genotype) and C (Pietrain * Duroc x Large White). Their offspring were assigned in groups according to the physiological status: surgically castrated males (SC, 60 animals), immuno-castrated males (IC, n=60), entire males (EM, n=60) and gilts (G, n=60). Immunocastration was performed using Improvac^{TM} vaccine at the age of 72 and 147 days. Live weight (LW) was measured at weaning (W, age = 25 days), at the time of first vaccine application (V1, age = 72 days), at the beginning of early fattening period (EF, age = 116 days), at the time of second vaccine application (V_2, age = 147 days), and before slaughter (FW, age = 168 days). Based on those measures, average daily gains (ADG) were calculated for different periods of fattening: from W to V1 (FP - feeder period), from V1 to EF (PP – porker period), from EF to V2 (EFP – early fattening period), from V2 to FW (LFP – late fattening period). At the age of 168 days, the pigs were slaughtered in a local abattoir.

Results Terminal sire line (S) significantly influenced LW of the fatteners at V_1 and EF (Table 1). Physiological status (T) had significant influence on the LW of fatteners at EF, V_2 and FW, while significant T*S interaction was observed for LW at EF, V2 and FW. Terminal sire line influenced ADG in the FP and EFP periods. Physiological status significantly affected ADG in following periods: PP, EFP, LFP and the total ADG. Significant interaction effect (T*S) was observed for FP and total ADG.

Table 1. Influence of terminal sire line, physiological status, and their interaction on fattening traits in the investigated growth periods

	S	T	T*S
LW - Live weight (kg)			
W – Weaning (25 days)	0,120	0,956	0,775
V_1 – time of the first vaccine (72 days)	<0,001	0,712	0,014
EF – start of early fattening period (116 days)	0,001	<0,001	0,047
V_2 – time of second vaccine (147 days)	0,997	<0,001	0,004
FW – before slaughter (168 days)	0,092	<0,001	0,041
ADG - Average daily gain (g)			
FP – Feeder period (25–72 days)	<0,001	0,672	0,007
PP – Porker period (72 -116 days)	0,131	<0,001	0,408
EFP – Early fattening period (116-147 days)	0,001	0,002	0,582
LFP – Late fattening period (147- 168 days)	0,168	<0,001	0,098
Total average daily gain (25-168 days)	0,161	<0,001	0,044

S: Terminal sire; T: Physiological status

Conclusion In the present study, significant effects of the sire line, physiological status and their interaction on the fattening traits were observed in most of the fattening periods. The highest LW and ADG were accomplished by immunocastrated pigs, descendants from sires of the line C, followed by the line B entire males and surgical castrates of the line A.

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Production of female pigs by embryo sexing using the porcine amelogenin gene

M Anastasiadou[*1], G Michailidis[2]
[1]*Research Institute of Animal Science, General Directorate of Agricultural Research, Hellenic Agricultural Organisation "Demeter", Paralimni Giannitsa, Greece*
[2]*Laboratory of Physiology of Reproduction of Farm Animals, Department of Animal Production, School of Agriculture, Aristotle University of Thessaloniki, Greece*
[*]marmogeo@gmail.com

Application

It is well known that surgical castration of boars without pain relief is considered unacceptable and the stakeholders of the pork industry committed to voluntarily end surgical castration of male pigs in Europe by January 1st, 2018. As the production of entire males or immunocastrates results in new challenges in order to reduce boar taint, one alternative would be the production of females. This could be achieved using reproductive biotechnologies, and in particular embryo transfer of female embryos using embryo sexing technology.

Introduction

Pre-determination of sex in livestock offspring is of critical importance for the efficient production of the world's food supply. Although several methods of sex identification exist, including cytogenetic analyses, H-Y antigen, measurement of X-linked enzymes and Y-chromosome-specific probes, polymerase chain reaction (PCR) amplification of sex specific DNA sequences is a relatively simple, rapid, inexpensive and highly accurate method. During the last years, molecular sexing was based on amplification of SRY gene. However, recent studies are focused on amplification of genes present in both males and females.

Material and Methods

In the present study, we designed a specific non degenerate primer pair for amplification of the porcine amelogenin (AMEL) gene. *AMEL* gene is present in both X (*AMELX*) and Y (*AMELY*) porcine chromosomes and based on the differences in the introns of the two alleles, we aligned the sequences of *AMELX* and *AMELY* genes and designed a non degenerate primer pair for PCR amplification in a single reaction. DNA was extracted from 50 male and 50 female pigs, as well as from 20 porcine embryos produced *in vitro*.

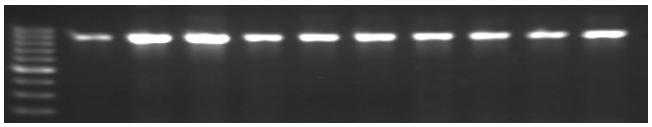


Figure 1 PCR amplification in porcine female DNA samples

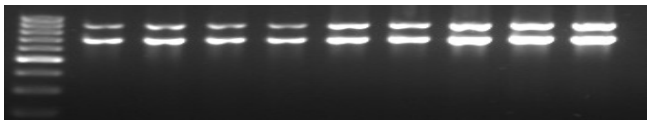


Figure 2 PCR amplification in porcine male DNA samples

Results

PCR performed in DNA extracted from 50 male and 50 female pigs revealed the 100% accuracy of this method, by successfully confirming the sex of each DNA sample tested (Figures 1 and 2). Furthermore, we successfully applied this PCR method for the sex determination of individual porcine embryos (Figure 3).

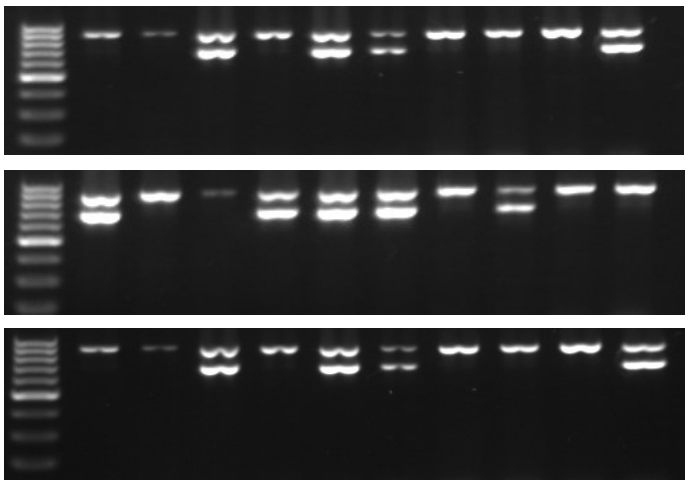


Figure 3 PCR amplification in porcine blastocysts DNA samples

Conclusion

This study shows that the present method is a simple, cheap and highly precise method for porcine sex identification, which can be applied in breeding programs to facilitate manipulation of the sex ratio of offspring.

The lysine requirement of growing entire male pigs (10-15 weeks of age): a dose-response study

S Millet[1*], M Aluwé[1], J De Sutter[2], W Lambert[3], B Ampe[1], S De Campeneere[1]
[1]Flanders research institute for agriculture, fisheries and food (ILVO), Belgium, [2]Orffa Belgium NV, Belgium, [3]Ajinomoto Eurolysine S.A.S., France
[*]sam.millet@ilvo.vlaanderen.be

Application

The current study shows that the standardised (SID) ileal digestible lysine: energy ratio for optimal performance of boars between 10 and 15 weeks of age is considerably higher than the current feeding practices for barrows and gilts in Belgium (around 9.5 g/kg). Data can be used by nutritionists to adjust dietary specifications of entire male pigs.

Introduction

With the production of entire male pigs, there is renewed interest in the determination of nutrient requirements of male pigs. Therefore, we designed a dose-response trial to determine the optimal dietary SID lysine level in the diet of growing entire male pigs.

Material and methods

In the present study, we evaluated the effect of increasing dietary SID lysine levels (6 levels: 7.5; 8.5; 9.5; 10.5; 11.5; 12.5 g/kg) on growth performance of entire male pigs (Piétrain boar x Ra-Se genetics hybrid sow) from 10 to 15 weeks of age. The CP level was fixed at 170 g/kg and all diets were formulated to meet or exceed the ideal amino acid (AA) profile for pigs (Tybirk *et al.*, 2016). Glutamic acid was included to keep diets isonitrogenous. Because of the high energy content assigned to glutamic acid, net energy level decreased slightly from the lowest to highest lysine level (from 9.9 to 9.6 MJ/kg). Amino acid analysis revealed slightly higher lysine levels than anticipated and requirements may therefore be slightly underestimated. The animal performance variables were subject to an ANOVA procedure, with bodyweight at start and feed as fixed factors and weaning series as random factor. Results were further explored with Tukey's post hoc test for all pairwise comparisons. Linear and quadratic models were fitted for the effect of SID lysine concentration on the response parameters to deduce a theoretical optimal SID lysine level. The pen was considered as the experimental unit and for all measurements, the pen average was used. Results were considered significant if $P < 0.05$.

Results

Both for daily gain and feed conversion ratio, animals on the diet with 10.5g SID LYS showed significantly better performance than the 3 groups with lower SID lysine levels. Daily gain increased from 674±51 g/day at the lowest level (7.5 g/kg) to 833±90 g/day at the highest level (10.5g/kg). Similarly, feed conversion ratio improved from 2.11±0.05 to 1.77±0.06 g/g with dietary SID LYS level between 7.5 g and 10.5 g/kg. For daily gain, it was not possible to fit a linear plateau model, but a quadratic plateau model gave an optimal SID lysine level at 12.95 g/kg. As this breakpoint is outside the measured range, it should be interpreted with caution. For feed conversion ratio, optimal SID lysine levels were 10.65 and 12.11 g/kg (1.09 or 1.25g SID LYS/MJ NE), for a linear and quadratic plateau model, respectively. The slope of the linear plateau model indicates that an improvement in FCR of 0.11 can be expected with each g increase of the dietary SID lysine level, between 7.5 and 10.6 g/kg.

	SID LYS level							
	7.5	**8.5**	**9.5**	**10.5**	**11.5**	**12.5**	SEM	P-value
Bodyweight at 10 weeks, kg	25.5	24.9	25.4	24.8	24.9	26.1	0.4	
Bodyweight at 15 weeks, kg	49.0^{a}	50.3^{ab}	51.4^{bc}	54.0^{d}	52.6^{cd}	55.3^{d}	0.7	0.002
Daily feed intake, kg	1.42	1.43	1.39	1.47	1.38	1.45	0.02	0.117
Daily gain, g	674^{a}	724^{ab}	744^{b}	833^{c}	790^{bc}	833^{c}	13	<0.001
Feed conversion ratio, g/g	2.11^{d}	1.97^{c}	1.87^{b}	1.77^{a}	1.75^{a}	1.74^{a}	0.02	<0.001

Conclusion

In the measured range of 7.5 to 12.5 g/kg SID lysine, increasing the lysine level clearly improved performance.
The dietary SID Lysine level for minimal FCR is 10.65 g/kg (1.09 g SID LYS/MJ NE) according to a linear plateau model and 12.11 g/kg (1.25g/MJ NE) according to a quadratic plateau model for growing entire male pigs from 10 to 15 weeks of age.

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Entire males fed high-protein and –energy diets

H Maribo
Danish Pig Research Centre, Copenhagen, Denmark, Feeding efficiency, SEGES
hma@seges.dk.

Application

The results indicate that a revision of the current Danish nutrient standards may be required according to increase productivity for entire males and reduce boar taint in the future. However, one trial is not enough to provide an unequivocal recommendation for feeding of entire males.

Introduction

Danish nutrient standards are based on production of castrates and female pigs and do not consider the high potential for lean meat gain of entire males. However, does entire males respond to the more energy and protein (Rasmussen, 2010) and whether this feeding strategy might lead to a lower degree of boar taint at slaughter.
Boar taint is mainly attributed to two substances: skatole, which is affected by nutrition whereas androstenone is affected by age/weight. When the growth rate of entire males is increased, they will be slaughtered younger, and this may lower androstenone levels i.e. the level of boar taint (Maribo and Jensen 2013, Maribo, 2017).

Material and Methods

The trial was conducted in one herd and comprised a total of 1 387 male pigs from 30 to 105 kg assigned to two groups: control diet and a diet plus 15% protein and 8% energy. Growth and feed uptake was recorded. Boar taint were determined in fat as skatole (ppm) (Hansen-Møller and Andersen 1994) and at line detection of Human nose.

Results

Daily gain increased 63 g for entire males which cut 4.6 days to slaughter by increasing energy and protein in the diet. Gross margin increased by 10%. Boar taint tended to be lower in the entire males in group 2 but skatole did not differ between groups.

Entire males	Control	Plus diet	Difference	P value
Number of pigs	684	703	-	-
Number of pens	46	47	-	-
Start weight, kg	29.0	29.2	+0.2	-
Carcass weight, kg	82.0	84.1	+2.1	<0.0001
Lean meat, %	60.7	60.6	-0.1	0.5248
Daily gain, g/day	1,084	1,147	62.5	<0.0001
Days 30-110 kg	73.8	69.2	-4.6	-
Feed uptake, FUgp/day	2.54	2.67	0.13	<0.0001
Feed utilisation, FUgp//kg	2.34	2.33	-0.01	0.1773

Conclusion

The results indicate that a revision of the current nutrient standards may require an optimization to production of entire males in the future. This to utilize growth capacity and reduce boar taint.

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Impact of sainfoin in entire male diets on nitrogen turnover and nutrient digestibility

E Seoni[1*], G Battacone[2], F Dohme-Meier[1], G Bee[1]
[1]*Agroscope, Posieux, Switzerland,* [2]*Dipartimento di Agraria, University of Sassari, Sassari, Italy*
[*]eleonora.seoni@agroscope.admin.ch

Application

The current findings suggest that condensed tannins from sainfoin reduces nitrogen absorption and digestibility resulting in greater faecal N-losses and concomitantly lower urinary N losses

Introduction

Various studies with ruminants (Scharenberg *et al.*, 2007; Grosse Brinkhaus *et al.*, 2016) have shown that offering legumes rich in condensed tannins (CT) have, among other things, a marked impact on the nitrogen (N) turnover since a shift of N excretion away from the urinary route to faeces occurs. From an environmental point of view, this shift is believed to be beneficial as ammonia losses from faeces are slower and less excessive than urine. These findings could also be of relevance for pig production as its environmental impact, especially the N utilization and ammonia emission, is of concern.

Material and methods

Forty-eight Swiss Large White entire male pigs (EM) originating from 12 litters and weighing on average 24.8 ± 5.1 kg were assigned within litter to 4 experimental diets: a control grower-finisher diet (T0) with no added tannins and 3 grower-finisher diets supplemented with 5 (T5), 10 (T10) and 15% (T15) of sainfoin, respectively. The grower (25-60 kg BW) and finisher (60-105 kg BW) diets were formulated according to the Swiss feeding recommendations for swine and were isocaloric and isonitrogenous. The experimental diets were offered *ad libitum* in a pelleted form. All pigs were reared in group pens, equipped with automatic feeders and individual pig recognition system (SchauerMaschinenfabrik GmbH. & Co KG, Prambachkirchen, Austria). The pigs were switched from the grower to the finisher diets when the BW of the individual pig reached > 60 kg the day of weighing. To assess the N-balance in the grower (50.0 ± 3.2 kg BW) and finisher (75.0 ± 4.7 kg BW) period, 2 metabolic trials, each lasting 7 d, were performed with all pigs where individual feed intake was monitored and total amount of urine and faeces were collected. In the diets, faeces and urine N-content was determined. Data were analysed with a one-way ANOVA using litter and experimental groups as fixed effects and mean differences tested using the Tukey post-hoc test.

Results

Despite similar N intake, urinary N excretion of T15 pigs was 32% lower ($P < 0.01$) compared to T0 and T5 pigs in the grower period and 32% lower ($P < 0.001$) compared to T0, T5 and T10 pigs in the finisher period. Concomitantly, faecal N excretion tended ($P < 0.06$) to be 56% lower and was 79% greater ($P < 0.001$) in T15 and T10 pigs compared to T0 pigs with intermediate values for T5 pigs in the grower and finisher period respectively. When expressed as percentage of total N intake, urinary N excretion tended ($P = 0.08$) to be lower by 27% and was lower ($P < 0.01$) by 31% in T15 compared to T0 pigs with intermediate values for T5 and T10 pigs in the grower and finisher period, respectively. Faecal excretion linearly increased ($P < 0.001$) by 56% from T0 pigs to T15 pigs in the grower period and was 64% greater ($P < 0.001$) in T15 pigs compared to T0 and T5 pigs in the finisher period. Body N retention was greater ($P < 0.05$) in T0 pigs compared to T5 pigs, with intermediate values for T10 and T15 pigs in the grower period whereas no treatment differences were observed in the finisher period. Moreover, urinary urea levels were numerically lower in T15 pigs compared to T0 pigs in both periods.

Conclusion

In conclusion, with increasing dietary CT inclusion a distinct shift in N excretion from urine to faeces was observed. This could be beneficial in reducing urinary ammonia emission from pig production.

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Testicular development, sex hormones and boar taint in pig lines divergent for residual feed intake

A Prunier[1*], Y Billon[2], S Ferchaud[2], R Comte[1], W Gros[1], J Ruesche[3], H Gilbert[3]
[1]*PEGASE, INRA, AGROCAMPUS OUEST, 35590 Saint-Gilles, France*
[2]GenESI*, INRA, 17700 Surgères, France*
[3]*GenPhySE, INRA-Université de Toulouse, INPT, ENVT, 31326 Castanet-Tolosan, France*
[*]armelle.prunier@inra.fr

Application

Pigs diverging by genetic selection on their residual feed intake differ highly in their testicular activity during the pubertal period pointing out the links between the regulation of the nutrient use and the reproductive axis.

Introduction

Improving feed efficiency and rearing entire male pigs are relevant strategies to reduce feed cost and environmental waste in the pig production. The major constraint for rearing entire male pigs being boar taint, an experiment was performed to determine the consequences of a divergent selection on residual feed intake (RFI) which is the difference between an animal's actual feed intake and its expected feed requirements for maintenance and growth (Gilbert *et al.*, 2017). Pigs from the HRFI line are low performers that "waste" nutrients whereas pigs from the LRFI line are good performers that "spare" nutrients.

Material and methods

Purebred French Large White male pigs from the 9th generation of selection were reared in two batches (n = 19 to 24 pigs/line/batch, n = 45 pigs from 18 litters and n = 43 pigs from 15 litters in the L and H lines respectively). Blood samples were drawn at 15 and 166 ± 1 days of age (mean ± SD) for testosterone (T) and oestradiol-7β (E2) analyses (immunoassays, ST AIA-Pack hsE2, ST AIA-Pack testosterone, Tosoh). Pigs were weighed at regular intervals. Pigs were slaughtered at 167 ± 1 days of age in July (Batch 1) and August (Batch 2) 2016. A back fat sample was collected on the carcass in the neck area for boar taint compound analysis by HPLC (Batorek *et al.*, 2012). The genital tract was removed for testis and epididymis weighing after dissection. Ratios between organ weight and live weight at slaughter were calculated for statistics. All data were analysed by ANOVA using R, including line, batch and their interaction as fixed effects and litter as a random effect.

Results

Except for fat skatole (S), the interaction between line and batch, as well as the batch effect, were never significant ($P > 0.1$). Live weight at birth and age at slaughter were similar in both lines ($P > 0.1$) whereas average daily gain (ADG) during lactation was higher ($P < 0.01$), ADG during the post-weaning ($P < 0.001$) and the fattening periods ($P < 0.5$) were lower in the LRFI line, in agreement with previous data (Gilbert *et al.*, 2017). Consequently, live weight at slaughter was lower ($P < 0.02$) in LRFI (100 ± 2 kg, mean ± SEM) than in HRFI (106 ± 2 kg) pigs. Percentages of testes and of epididymes were higher in LRFI than in HRFI pigs ($P < 0.001$). Plasma concentrations of T and E2 at 15 days of age and plasma T at 166 days were similar in both lines ($P > 0.1$), whereas plasma E2 at 166 days and fat concentration of androstenone (A) were higher in LRFI than in HRFI pigs ($P < 0.01$). Fat concentration of S was about three times higher in LRFI pigs from Batch 1 than in the three other subgroups of pigs ($P < 0.05$). A summer heat wave probably explains this phenomenon in pigs predisposed to high S by high E2 and A in agreement with previous literature (Parois *et al.*, 2018).

Conclusion

Overall, these data indicate a higher testicular activity in LRFI than in HRFI pigs, in agreement with a smaller exploratory dataset from the 6th generation of the same selection experiment (Prunier *et al.*, 2016), and from domestic fowls (Morisson *et al.*, 1997). It reflects more likely an impairment of this activity in HRFI pigs than an improvement in LRFI pigs.

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Finishing heavy boars for lower taint, suitable welfare and optimal performance

L Martin[1*], A Frias[1], R P R Da Costa[1,2], M A P Conceição[1,2], R Cordeiro[3], A Ramos[1,2]
[1]Inst Politec Coimbra, Portugal, [2]CERNAS, Portugal, [3]Uzaldo Lda., Portugal
[*]luisam@esac.pt

Application This work can be useful to producers that consider finishing heavier boars without surgical or pharmacological castration.

Introduction Entire male pig production is considered economically and environmentally advantageous due to superior feed efficiency and growth rates when compared to barrows or immunocastrated males (Albrecht *et al.,* 2012). Finishing heavy boars is challenging not only because of aggressive behaviour, but also due to boar taint. Inulin may address this problem (Aluwé *et al.,* 2017), although dosage needs to be refined. This study evaluated three aspects. Firstly, the behaviour of boars subjected to two housing treatments and three diets, with 0%, 3% and 6% of inulin. Inulin is abundant in chicory roots but exists as well in other plants so it is our purpose to know exactly its content in the feeds used. Secondly, assess average daily weight gain and carcass yield to understand the implications of diet, housing and behaviour on the pig's performance. Thirdly, evaluate blood metabolites related to boar taint (skatole and indole) and cortisol.

Material and methods A 3x2 factorial design experiment took place with 60 cross boars (114 kg ± 9.5) subjected to 3 different diets and 2 housing conditions. Three pens allowed for normal animal density (1 m^2.pig^{-1}), whereas the other 3 were larger (1.9 m^2.pig^{-1}) and contained an additional drinker and two additional environmental enrichment items. Groups were fed, ad libitum, with isoproteic (15.5 %CP), isoenergetic (2.3 Mcal NE) diets containing of 0, 3 or 6 % inulin (from Fibrofos 60%) for seven weeks. Every raw component of the mix was analysed for inulin (AOAC 999.03). *In loco* individual skin lesions were assessed (Welfare Quality®) every two weeks. Intake was on average 2.7 $kg.day^{-1}$ per pig. To assess cortisol, indole and skatole, blood was collected at the end of the trial. The plasma obtained was maintained at - 80ºC. Cortisol concentration was measured using a competitive enzyme immunoassay kit (ENZO). Skatole and indole were analysed by HPLC-FL (Claus *et al.*, 1993). A RP-C18 column (5µm, 125mmx4.6mm) was used at 40°C, with an isocratic gradient, flow rate 1 $mL.min^{-1}$, the mobile phase was water:acetonitrile:2-propanol. Excitation and detection wavelengths were set at 220nm and 271nm respectively and the internal standard was 2-methylindole. Data were analysed with a STATISTICA software (version 8).

Results On average skin lesions were not serious and were of similar intensity for the two housing treatments. The diet as well did not had a large influence on skin lesions score but the group that was fed with 6% inulin had a lower score. Skin manure on the belly and face was higher where the animal density was higher. Diets with higher inulin tended to result in animals with higher levels of skin manure. More liquid excreta were observed in the groups with higher inulin intake and pen floors of these groups were persistently more wet and slippery throughout the trial. Our results for inulin showed that wheat (grain and bran) soya and rape meal had higher contents of inulin and corn grain had the lowest. Around 3% of inulin in diets came from the basic feeds and the rest from Fibrofos 60%. The pelleting process caused an inulin reduction of 1.4 to 1.8% in the feed mixes. The statistical analysis for pig performance did not show any significant differences between treatments for average daily weight gain or carcass yield. Our results showed a significant decrease in skatole and indole with the increase of inulin in the diet when compared with the control (0%); but there was no difference between 3 and 6%. The improved housing (low density) caused a decrease in plasma cortisol but an increase in skatole.

Conclusion Boar performance was not impaired by inulin or lower density pens. Intensity of skin lesions were low and did not differ with the treatments. Animal dirtiness was higher for higher stocking density and higher intake of inulin (coincided with more liquid faeces). Blood skatole and indole significantly decreased with inulin intake. The housing improvement (low density and enriched environment) caused an increase in skatole but the cortisol decreased. In parallel studies (unpublished) we found that pigs in improved housing tended to be more active and interact more with the environment but were less aggressive with each other. Next, we intend to assess welfare and boar taint when cheaper inulin feeds are used for shorter periods in pigs.

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Weight of sex glands as an on line tool to discriminate entire males from immunocastrates

N Batorek Lukač[1], G Fazarinc[2], M Prevolnik Povše[1], M Škrlep[1], M Čandek-Potokar[1*]
[1]*Agricultural Institute of Slovenia, Hacquetova ulica 17, SI-1000 Ljubljana, Slovenia*
[2]*University of Ljubljana, Veterinary Faculty, Gerbičeva 60, SI-1000 Ljubljana, Slovenia*
[*]meta.candek-potokar@kis.si

Application
On-line assessment of successful immunocastration based on the size of reproductive organs.

Introduction
Active immunisation against the hypothalamic GnRH i.e. immunocastration (IM) is an alternative to surgical castration of male pigs in prevention of boar taint. However, some pigs may escape the effective immunisation (so called non-responders). It is important to find a simple and reliable way to determine IM effectiveness at slaughter line, as chemical analysis of substances responsible for boar taint (androstenone and skatole) is laborious and expensive. A meta-analysis of published studies demonstrates a clear-cut impact of IM on the size of reproductive organs (Batorek *et al.*, 2012a) which implies the potential for on-line detection method of IM efficiency.

Material and Methods
Data on weights of testes, vesicular and bulbourethral gland of 55 entire males (EM) and 76 immunocastrates (IC) were obtained from our previously published experiments (Škrlep *et al.*, 2010; Škrlep *et al.*, 2012; Batorek *et al.*, 2012b; Kubale *et al.*, 2012). Pigs varied in body weight (103.6 ± 13.5 kg) and IM-to-slaughter lag (2 to 9 weeks). To distinguish IC and EM, a discriminant analysis (SPSS 21.0 for Windows) was made with cross-validation using leave-one-out method.

Results
The results (Table 1) indicate that the correctness of discrimination was high but not ideal, and it was better for IC than EM. The success rate of correct prediction of IC was practically 100%, because one misclassified IC was likely a non-responder.

Table 1: Results of discriminant analysis[a]

Criterion	Success rate, % (IC, EM)	Misclassified IC (n, %)	Misclassified EM (n, %)
Testes	91.6 (94.7, 87.3)	(4, 5.3 %)	(7, 12.7 %)
Bulbourethral gland	93.1 (96.1, 89.1)	(3, 3.9 %)	(6, 10.9 %)
Vesicular gland	90.8 (98.7, 80.0)	(1, 1.3 %)*	(11, 20.0 %)
All three organs	96.2 (98.7, 92.7)	(1, 1.3 %)*	(4, 7.3 %)

* based on androstenone level (0.34 μg/g) this pig was likely a non-responder, implying 100% correctness for IC.
[a]The results presented were published in Čandek-Potokar *et al.*, 2014.

Conclusion
Weight of reproductive organs may serve as a reliable tool to test the effectiveness of IM at slaughter, but the dissection is still time consuming (around 10 min per pig).

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Effect of the method of castration on growth performance and boar taint.

K Zadinova*, R Stupka, J Citek, N Lebedova
[1]*Faculty of Agrobiology, Food and Natural Resources, Czech University of Life Sciences Prague, Prague, Czech Republic*
*zadinova@af.czu.cz

Application The method of castration can affect growth performance and the quality of pork meat.

Introduction Fattening of the entire male pigs, when compared with other pig categories (barrows and gilts), appears to be more profitable, mainly due to better growth intensity, feed conversion and improved carcass leanness (Kim *et al.,* 2013). However the occurrence of so called "boar taint" in boars meat represents a significant problem. Boar taint can be eliminated by castration or immunocastration. This method prevents the occurrence of boar taint while preserving the positive effects of testicular steroids and anabolic hormones occurring in males (Zamaratskaia *et al.,* 2008).

Material and methods A total of 52 pigs of the D × (LW × L) crossbreed were assigned into 3 groups: boars (n = 18); immunocastrates (n = 16); barrows (n = 18). Barrows were surgically castrated on the 3rd day after birth. The immunocastrates were treated with Improvac® in two dates: pigs were 94 and 115 days old. All pigs were fed *ad libitum* by commercial diets. The pigs were slaughtered at 154 days of age. Androstenone and skatole level was measured according to Okrouhla *et al.* (2016). The following indicators were measured in weekly intervals (daily feed intake (kg), feed conversion ratio (kg), average daily gain (g), live weight (kg). Lean meat (%), backfat thickness (mm) and intramuscular fat (%) were measured after slaughter according to Walstra and Merkus (1995). The data were analysed with ANOVA using GLM procedure in SAS.

Results Overall growth performance did not differ between immunocastrates and boars. On the other hand, significant differences between barrows and the other groups were observed. The greatest back fat thickness, the lowest lean meat percentage and the greatest daily feed intake (P<0.05) were observed in barrows. The levels for androstenone were 2.381µg/g (boars), 0.533µg/g (immunocastrates), 0.185µg/g (barrows) and for skatole were 0.221µg/g (boars), 0.064µg/g (immunocastrates), 0.05µg/g (barrows).

Table 1 Growth performance parameters and meat quality traits of boars, immunocastrates and barrows.

Variable	Boars	Immunocastrates	Barrows	P
Body weight (kg)	106.2[a]	106.99[a]	105.9[a]	NS
ADG (g/day)	1169[a]	1181[a]	1193[a]	NS
Feed intake (kg/day)	2.62[a]	2.68[a]	2.83[b]	*
Feed conversion ratio	2.29[a]	2.23[a]	2.38[a]	NS
Carcass weight (kg)	79.2[a]	80.7[a]	81.8[a]	NS
Lean meat (%)	60.3[a]	59.5[a]	58.5[b]	*
Backfat thickness (mm)	11.5[a]	12.6[a]	17.1[b]	*
IMF (%)	2.18[a]	2.13[a]	2.31[a]	NS

* P<0.05

Conclusion In conclusion, boars and immunocastrates achieve better growth performance compared to barrows. Levels of androstenone and skatole were below the threshold (1µg/g and 0.25µg/g) for immunocastrates. Immunocastration reduces boar taint and positively influences the growth parameters of the pigs.

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Influence of housing conditions on antibody formation and testosterone after Improvac vaccinations

K Kress*[1], U Weiler[1], V Stefanski[1]
[1]*Behavioral Physiology of Livestock (460f), University of Hohenheim, Stuttgart, Germany.*
*kress.kevin@uni-hohenheim.de

Application

The use of immunocastration in male pigs can become a sustainable alternative to current castration practices, with high acceptance among consumers and producers on the European pig market.

Introduction

Immunocastration of male pigs is an effective alternative to prevent boar taint and to reduce aggressive behaviour among pigs. To achieve a high acceptance of immunocastration along the pork chain, the reliability of the method is crucial. The amount of "non-responders" is discussed controversially, and ranges between 0% and 30% have been reported. The reasons for the high variability are unknown. (Zamaratskaia and Rasmussen, 2015; Sødring *et al.*, 2018)

Material and Methods

The effect of housing conditions on the reliability of immunocastration was studied in immunocastrates (IM) and entire males (EM) under standard, enriched and stressful (mixing) conditions, with 6 IM and 6 EM in each group. Mixing events were performed 2 weeks around 1st vaccination and 3 weeks around 2nd vaccination. In total, 4 blood samples (B1-4) were collected per pig for the measurements of anti-GnRH-antibody level (binding of ^{125}I-GnRH) and testosterone concentrations (RIA). Further samples and observations (fat for androstenone and skatole determinations; testes weight; number of penile injuries) were collected at the slaughter line. The data was analysed with IBM SPSS (Version 24), with Spearman-Rho, U- and H-Test.

Results

Preliminary analysis given here represents 50% of the final number of IM (N=24) and EM (N=24) in this project. IM had plasma testosterone concentrations of 0.35±0.05 ng/ml at slaughter, which confirms successful immunocastration. In addition, all IM had androstenone levels less than 0.24 µg/g fat and skatole levels lower than 0.03 µg/g fat. GnRH-binding in B2 and B3 as well as B3 and B4 were positively correlated ($p<0.05$), while testes weight and GnRH-binding at slaughter were negatively correlated ($p<0.05$). In contrast to IM, EM had high plasma testosterone levels at slaughter (51.9±6.7 ng/ml; $p<0.001$). 71% of EM revealed androstenone values above 1.0 µg/g fat and 3 out of 24 EM had skatole levels above 0.15 µg/g fat. Compared to EM, IC had significantly lower testosterone levels at slaughter ($p<0.001$; n=24). Moreover, IM had not only significantly less penile injuries per animal than EM, but the number of IM with no injuries was also much higher.

Conclusion

The present data suggest that immunocastration is highly effective in reducing gonadal activity even under different housing conditions. Consequently, testosterone and androstenone concentrations were markedly suppressed in IM. Immunocastration also significantly reduces welfare problems associated with penile injuries compared to EM.

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Development of long-term, pre-finishing immunocastration protocols for male Iberian pigs. 1:Efficacy

F I Hernández-García*, M Izquierdo, M A Pérez, A I Del Rosario, N Garrido, A Montero
Center of Scientific & Technological Research of Extremadura (CICYTEX), Spain.
*francisco.hernandez@juntaex.es

Application Voluntary end of male pig castration is expected in the EU in the near future, but standard immunocastration (IC) protocols need to be adapted for Iberian (IB) pig males due to their long life cycle.

Introduction Male IB pigs are castrated to avoid boar taint, to improve fat accretion and to ease free-range management. In conventional pig production, a two-dose protocol of vaccine against GnRH is used to inhibit testosterone secretion and boar taint (Batorek *et al.*, 2012). However, this standard protocol is somewhat reversible and must be adapted for Iberian pigs due to their long life cycle, which usually ends at over 15 months (m) of age. In previous studies, we developed a long-term, 3-dose, pre-finishing IC protocol for male Iberian pigs whose efficacy seemed to be influenced by nutritional level. In addition, the last vaccination dose of the protocol coincided with the middle of *montanera* (*Mt*; acorn-feeding free-range period), thus complicating management. In extensive Iberian pig production systems, body weight (BW) and body condition score (BCS) are usually heterogeneous, due to the hierarchical competition and stress provoked by the required feed restriction during the long pre-finishing period. In fact, IB pigs cannot be fed *ad libitum* for a long time because of their obesity-prone genotype. The objectives of the present studies were the following: Study 1 aimed to improve the efficacy of the protocol by a short-term increase in feeding intake. Study 2 aimed to further adjust the protocol to suit the chronology of *Mt*.

Material and methods Study 1: Pigs (IC males; ICM; n=47) were fed concentrate in an extensive system and immunized against GnRH at 11, 12 and 14 m of age. Pigs were slaughtered at 16 m. Treated pigs (23 ICM) were submitted to a 15-day *ad libitum* (*alb*) feeding period starting at the 3rd vaccination, during which hopper-type feeders were set in large corrals to minimize competition and stress. The remaining ICM formed the Control group. Entire males (EM; n=5) were used as general controls. The *in vivo* data collection included monthly BW, testicular and body composition echography and blood sampling for anti-GnRH antibodies and testosterone. *Postmortem* data included carcass and meat quality assessment, androstenone and skatole levels in lumbar backfat and morphometry of the reproductive tract. Testis volume was measured by a water displacement technique.
Study 2: Our hypothesis was that improving BCS homogeneity before *Mt* would enhance and homogenize testicular atrophy. Control pigs (Ctrl; n=18 IB males) were immunized at 10.5, 12 and 13.5 m. Treated pigs (Trt; n=17 IB males) were immunized at 10.5, 11.5 and 13 m, with a 15-day *ad libitum* feeding period starting at the 3rd dose. Pigs started *Mt* at 13.5 m and were slaughtered at 16 m. To further determine the effect of nutrition level on the efficacy of IC, 15 IB x Duroc males (AdLib group) were fed *alb* with concentrate during the growth and finishing phases in a regular extensive system, immunized at 8, 9 and 10 m and slaughtered at 13 m. Data collection was similar to that of Study 1.

Results Study 1: Treated group reached 100% efficacy, as all animals had <150-g testes (threshold for blood testosterone presence in our earlier studies). In contrast, 4/24 Control ICM had >150-g testes. Testicular weight was significantly different among the 3 groups (EM: 381 ± 12 g; Control ICM: 103 ± 18 g; Treated ICM: 74 ± 6 g). Hormonal data are not yet available.
Study 2: The Adlib and Trt protocols showed a 100% efficacy (all testes <150g). The Ctrl group had greater testicular weight than the other 2 groups and also had 2 animals with ~150 g testes (medium sized; 160 and 146 g). Testicular and epididymal weights were significantly smaller for AdLib and Trt than for Ctrl pigs. Bulbourethral gland weight was significantly smaller for Trt than for Ctrl pigs. Echographycally estimated testis volume and actual volume were highly correlated (R^2=0.88). Blood testosterone was basal for all AdLib and Trt pigs and 16/18 Ctrl pigs, being positive for the 2 Ctrl pigs having medium-sized testis. Fat androstenone and skatole were basal for the basal-testosterone pigs (fat samples were not available from these 2 Control, medium-sized testis pigs). For Adlib and Trt groups, IC effect lasted for the 3-month post-immunization fattening period, which was 1 month longer than in our previous studies (Martinez-Macipe *et al.*, 2016), thus improving intramuscular fat deposition in Ctrl and Trt groups, which reached similar values to those usually reported for castrate Iberian pigs.

Conclusions
Nutritional level management can be used to improve IC efficacy, and treatment is compatible with *montanera* system.

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Sex neutralization of heavy pigs from Iberian Peninsula breeds: solutions and limitations

R Charneca[1*], F I Hernández-García[2], M Izquierdo[2]
[1]*Instituto de Ciências Agrárias e Ambientais Mediterrânicas, Universidade de Évora, Portugal,*
[2]*Center of Scientific & Technological Research of Extremadura (CICYTEX), Spain.*
[*]rmcc@uevora.pt

Introduction

Alentejano (AL) and Iberian (IB) pigs are two native breeds from Iberian Peninsula. Both breeds present: low prolificacy (Charneca *et al*., 2012; Fernandez *et al*., 2008) and low growth rate, but compensatory and relatively high growth rate (Freitas, 1999; Daza *et al*., 2005) during the fattening/finishing phase called "Montanheira" in Portugal and "Montanera" in Spain. Recent studies show that both breeds are genetically related (Muñoz *et al*., 2017). After weaning, pigs are mostly raised in free-range system and during the fattening period pigs have access to acorns and grass from the Mediterranean forest, usually from November to March (Charneca *et al*., 2017; Lopez-Bote, 1998). For the certification of some dry-cured products from these breeds, like the high-grade hams, the animals can only be slaughtered with an age of at least 14 months and a bodyweight ranging from 145-210 kg since the quality of the products is related to the age, weight and diet of animals (Daza *et al*., 2007; López-Bote, 1998). Because of that high slaughter age, sex neutralization is essential for the chain production success. Until now the gonadectomy of both males (avoid boar taint and aggressive and sexual behaviour) and females (avoid mating by wild boars) is a common practice, however, the foreseen voluntary end of surgical castration (SC) without pain relief in the EU requires the use of alternatives in these swine breeds management.

Options, main problems and risks

The options for sex neutralization are SC with pain relief and Immunocastration (IC). However, while SC with pain relief needs the development of a feasible and economically worthwhile procedure to be usable alternative, the two doses IC protocols already used in intensive swine genotypes and systems aren't effective in these heavy and aged pigs (Zamaraskaia and Rasmussen, 2015), so they have to be adjusted and tested in this case. Additionally, besides manipulation difficulties (extensive raising areas, poor facilities, low manpower), several risks must be considered when IC is proposed, namely: its effectiveness, the effects on performance, carcass, meat and dry-cured products characteristics (including sensorial) and finally, the acceptance by the industry and consumers.

State of the art of IC of females and males

A 3 doses pre-pubertal IC of Iberian gilts have long-term successful effects (ovarian quiescence). Protocol is relatively easy to apply at farm as it allows gender mixing before immunization and doesn't include vaccination during fattening period being effective until the usual slaughter age (Hernández-García *et al*., 2013). So far, no adverse effects of IC females have been detected both on performance or carcass traits (Gómez-Fernández *et al*., 2013, Martínez-Macipe *et al*., 2016). For males a 3 dose protocol is also needed but in this case the immunisation efficacy has been variable, although a 100% efficacy was recently reached with a protocol in which the 3rd dose was administrated before the acorn-feeding (Hernández-García *et al*., 2018) and when IC is concomitant with a specific nutritional plan (*ad libitum* feeding during 15 days since 3rd dose administration). In this last protocol besides desired effects on reproductive organs development and backfat androstenone and skatole levels, the supplemented pigs also presented greater foreleg, loin and prime-cuts yields than pigs with a standard 3 doses IC protocol. Immunocastrated males present higher growth rate than SC males from 40 to 105Kg of weight (Seiquer *et al*., 2017) and also present less redness and saturation and higher drip loss in their muscles at about the same weight (100kg; Nieto *et al*., 2017). At heavier weights (155 kg) IC males presented leaner carcasses, less intramuscular fat, higher shear force and rancidity than SC males (Martinez-Macipe *et al*., 2016). According to Martinez-Macipe *et al*., (2015) the effects of IC on male meat quality seem limited. Some studies were made on fresh products from IC males (e.g. García-Gudiño *et al*., 2017) but no information is available regarding cured products (including sensorial).

Conclusions

Because of their long production cycle, which is necessary to obtain the high-grade products, sexual neutralization is needed in Alentejano and Iberian pigs. An effective IC protocol seems more easy to achieve in females than males, and the absence of possible detrimental effects of IC of females are more "clear" than in males. No studies were found on the IC effects on dry-cured products quality and finally, no studies of IC are available for AL breed. Therefore, further studies for protocol optimization and impact of IC on final high grade products from AL and IB pigs are needed.

Acknowledgments

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References: full references list and full open-access cited papers can be sent upon request to the corresponding author.

Is it possible to reduce androstenone by dietary means?

G Zamaratskaia[*1], MK Rasmussen[2]
[1]*Department of Molecular Sciences, Swedish University of Agricultural Sciences, Uppsala, Sweden.*
[2]*Department of Food Science, Aarhus University, Tjele, Denmark.*
[*]galia.zamaratskaia@slu.se

Application

If positive research findings on dietary effect on androstenone continue to grow, there is reason for optimism over the possible elimination of boar taint by dietary means thus improving animal welfare and meat quality.

Introduction

It is generally believed that in the regulation of androstenone, dietary factors are of minor importance. However, androstenone metabolism can be modified by dietary ingredients through up-regulation of androstenone-metabolising enzymes. From practical perspective, it is of great interest to find dietary ingredient(s) with androstenone-reducing effect as an alternative to invasive methods such as surgical castration.

Material and Methods

In the present study we attempted to summarize existing data on the effects of nutritional factors on androstenone levels. The main focus was on research performed in Scandinavia over the period from 2007 to 2017.

Results

Interestingly, a number of studies suggested the effect of dietary components on androstenone. For example, Chen *et al.* (2007) demonstrated that plasma free androstenone levels significantly decreased in the pigs fed raw potato starch for 2 weeks. Fat androstenone levels were also slightly lower in these pigs, although this decrease did not reach statistical significance. It was suggested that most androstenone in plasma is present as sulphoconjugates, which are formed in the liver and testes (Sinclair and Squires, 2005). It is likely that androstenone sulphation also occurs in other tissues, e.g. the intestine. Intestinal sulphotransferases can be either up-regulated by resistant starch or products of its fermentation and speed up intestinal androstenone sulphation making androstenone less available for accumulation in fat. Enzymes involved in hepatic androstenone metabolism can also be regulated by diet. Feeding male pigs with 10% dried chicory root for 16 days before slaughter reduced fat androstenone levels by up-regulation of major androstenone-metabolising enzyme 3β- hydroxysteroid dehydrogenase (Rasmussen *et al*., 2012; Figure 1). This study, however, was performed on limited number of animals and further research is needed to elucidate effect of chicory root and its active component on androstenone metabolism.

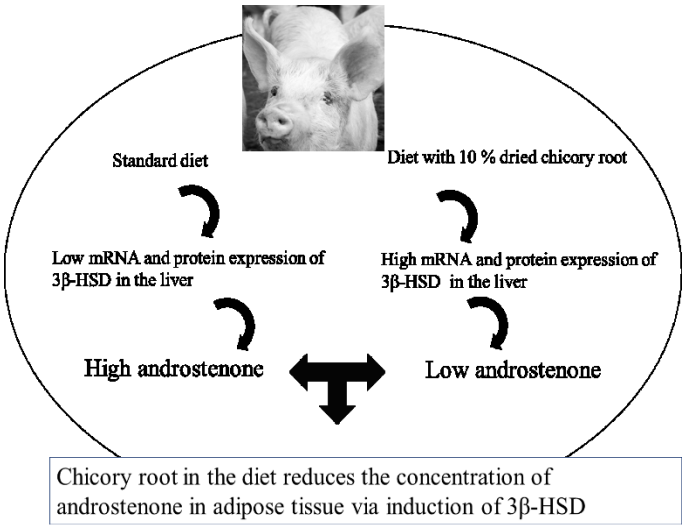


Figure 1 The effect of chicory root on fat androstenone

Conclusion

There is a major interest in understanding physiological processes involved androstenone metabolism to find a way to reduce androstenone through the dietary means.

Acknowledgments

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Health and welfare issues regarding surgical castration of male piglets and its alternatives

E von Borell[1*], A Prunier[2], U Weiler[3]
[1]*Martin-Luther-University Halle-Wittenberg, Halle, Germany,* [2]*INRA, Saint-Gilles, France,* [3]*University of Hohenheim, Stuttgart, Germany*
[*]eberhard.vonborell@landw.uni-halle.de

Application
Status quo on health and welfare issues regarding surgical castration and entire male production**.**

Introduction
Surgical castration without anaesthesia is increasingly recognised as a painful and welfare relevant procedure, although the systematic use of anaesthesia with or without additional analgesia for pain relief during surgical castration is currently only practiced in some countries where this was mandated or part of their national quality assurance scheme. Representatives of the European pig sector endorsed the European Declaration (2010) on alternatives to surgical castration of pigs, aiming to stop surgical castration of male pigs by 2018 (Boars, 2018). However, it is estimated that currently up to 70% of the male pigs within the EU are still surgically castrated without anaesthesia.

Current knowledge on effectiveness of pain intervention
From scientific studies, it is evident that the effectiveness of pain intervention during and after surgical castration is only given when anaesthesia is combined with pre-emptive analgesia, although it has to be considered that additional stress and associated health and welfare risks may be imposed by the intervention itself. Previous surveys among stakeholders evidenced that the acceptance and likelihood of anaesthesia implementation will depend on authorization of farmers to do the pain interventions after special training (CASTRUM, 2017). However, such protocols are difficult to apply taking into account legal and economic constraints.

Entire males and immunocastration
Immunocastration is an already licensed and practiced alternative performed by farmers. Although this intervention was proven to be effective with less impact on health and welfare of pigs, this method has not achieved a broad application due to the resistance of some chain actors that fear low acceptance of this method by consumers. The widely favoured option of fattening entire males poses a risk for injuries due to unwanted sexually motivated behaviours such as mounting and penis biting under current housing and management conditions, unless pigs are slaughtered before puberty in order to reduce critical behaviours and to limit boar taint.

Conclusion
Raising entire young males and immunocastration seem to be the preferred short term and widely usable options, whereas fattening of matured heavy entire males as a sustainable and effective long-term goal would necessitate the implementation of new genetic schemes as well as new housing and management systems and a significant effort in research.

Acknowledgements
The review was prepared within the framework of the Cost Action CA 15215 IPEMA.

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Video analysis of entire male pigs' aggressive behaviour: Effects of inulin and housing conditions

M Fonseca[1], M A P Conceição[1], M A Ramos[1], L Martin[1], A Frias[1], R Cordeiro[3], N Lavado[2], R P R da Costa[1*]
1Polytechnic of Coimbra, Coimbra Agriculture School (ESAC), Bencanta, 3045-601 Coimbra, Portugal
2Polytechnic of Coimbra, Coimbra Institute of Engineering (ISEC), Portugal
3Uzaldo, ltd, R Balastreira 8, 3090-649, Coimbra, Portugal
[*]ross@esac.pt

Application

To reduce aggressive behaviour of entire male pigs through a diet of 0%, 3% and 6% of inulin (chicory compound) and placed under two housing conditions: standard and improved.

Introduction

Surgical castration, a painful procedure when done without anaesthesia, is a practice in many countries and represents a welfare issue (Marchant-Forde *et al.*, 2009). Castration with anaesthesia in piglets can be a difficult task for large pig industries. Rearing of uncastrated male pigs and for a longer period to gain maximum growth rate could be a possible solution. This, however, presents difficulties in good welfare practice because male pigs express their dominance and mating urge through aggressive behaviours that can cause serious lesions and a state of permanent stress in submissive individuals. Consequently, meat quality could be affected, therefore urging for studies to seek ways to reduce this effect.

Material and Methods

This study was conducted at a Portuguese commercial farm for 49 days using 60 crossbreed pigs (Pietrain X F1 [Large White x Landrace]) aged 5.5 months and weighing 114 ± 9.5 kg. The 60 pigs were distributed in 6 pens (A, B, C, D, E and F), in groups of 10 individuals and given 3 diet treatments and two housing conditions both with slatted (stripped area) and concrete floors (white area) (Fig. 1). Pens A, B and C with diet of 0%, 3% and 6% inulin respectively and standard housing (1m^2/pig; one drinker; one feeder; one environmental enrichment item) and pens D, E and F with diet of 0%, 3% and 6% inulin respectively and improved housing (1.9 m^2/pig; two drinkers; one feeder; three environmental enrichment items). Six CCTV cameras recorded for 24 hours but only the peak activity diurnal hours (7:30 h/day/video), specific behaviours (Table 1) and five days (3 days in November; 2 days in December 2017) were assessed. Video behavioural analysis of selected behaviours was performed, using BORIS program. SPSS software was used for statistical analyses.

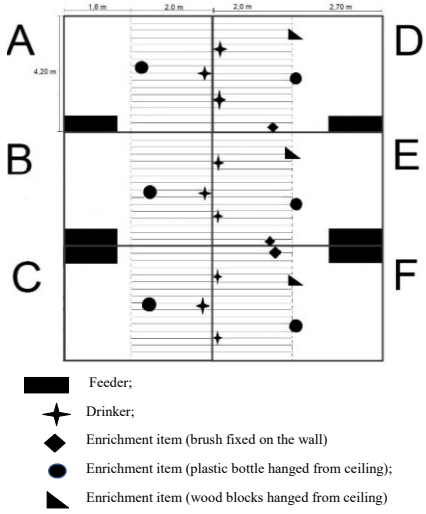


Figure 1 Experimental design of the 6 pens

Table 1 Behavioural ethogram

Behavioural categories	Behaviour
Exploratory	Interaction with Objects Explore Environment
Agonistic – Medium Aggression	Lever Head/Body Knock Parallel/Inverse Press Chase Mount Attempt
Agonistic – High Aggression	Mount Sexual Action Bite

Results

Diet had an effect in reducing aggression ($F_{2,24}$=5.44, p=0.011)) where 3% of inulin was more efficient than 0% (p=0.002). Six percent was less effective than 3% but not significantly (p=0.057). Although a higher number of aggressions was found in standard housing compared to improved housing, the difference was non-significant (U = 89.00, p = 0.330), taking into consideration the number of days analysed. Pen A had the highest aggressive behaviour occurrences (309.20 ± 68.78) compared with E (175.43 ± 66.25) ($\chi^2(5)_{KW}$ =11.46, p=0.043).

Conclusion

The 3% of inulin diet was the most efficient in reducing the number of aggressive behaviours. Further studies are needed on environmental enrichment and housing to find the best solution to reduce aggressive behaviour among boars bred after 5 months of age.

Acknowledgments

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Influence of raising entire boars and gilts during fattening in one unit on pubertal development, growth performance and welfare problems

J Bütfering, V Stefanski, U Weiler*
[1]*Institute of Animal Science, University of Hohenheim, Stuttgart, Germany.*
*weiler@uni-hohenheim.de

Introduction

Pork production with entire male pigs requires the definition of best practice conditions to make use of the higher anabolic potential of entire males and to minimize boar taint and welfare problems as well. The effect of a mature male on female puberty is well known but the effect of the presence of the other sex during the fattening period is less clear. Thus, in a field study, the effect of raising pigs either in single sex units (entire males or gilts only) or in a unit with alternating pens of gilts and entire males was evaluated.

Material and Methods

Entire males and gilts (JSR-hybrid x Pietrain) were raised from 31.5 kg to slaughter at 119-123 kg in three separate units (8 pens/unit; 33 animals/pen). Two units were single sex units (U-M: 8 pens with entire males; U-F: 8 pens with gilts), in the third one (U-MF) pens with entire males and with gilts were alternating (4 pens each). Two groups of "focus animals", slaughtered at an age of either 189 or 196 days from all units, were further sampled at the abattoir to analyse testosterone in blood plasma, androstenone, skatole and indole concentrations in adipose tissue in boars according to Wesoly *et al.*, 2015. Penile injuries were determined as described earlier (Weiler *et al.*, 2016). In females only uteri and the ovaries were collected at the slaughter line to evaluate pubertal development. Data were analysed with SPSS after log transformation (androstenone, skatole). The effect of unit and age were analysed using a univariate analysis of variance.

Results

Daily gain in entire males from U-M was significantly lower than in those from U-MF [742 ± 84 g (n=247) vs 820 ± 74 g (n=122); $p<0.001$]. These differences in growth performance coincided with a higher testicular activity in the focus animals (Table 1), resulting in significant differences in testosterone in blood and androstenone but not on skatole in fat. Penile injuries tended to be more severe in U-MF (50% vs 41.3% with 4 and more injuries/entire males, ns). In contrast to males, pubertal development in females was not affected.

Table 1 Endocrine and boar taint parameter at slaughter in entire males focus animals (LS-means, (n)) raised with entire males (U-M) or entire males/gilts (U-MF)

Parameter	U-M		U-MF		RMSE	P-value
Testosterone ng/ml (n)	11.9	(53)	17.1	(45)	1.873	0.01
Androstenone µg/g fat (n)	0.77	(100)	1.57	(79)	0.163	0.001
Skatole ng/g fat (n)	68.6	(100)	81.1	(79)	12.4	ns

Conclusion Raising boars together with gilts hastened the pubertal development in entire males as characterized by higher testosterone at slaughter. This coincides with a better growth performance but increases the risk for higher boar taint levels and welfare problems.

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Level of boar taint in young fattening boars as related to health and rearing conditions

M Škrlep[*1], N Batorek Lukač[1], U Tomažin[1], M Prevolnik Povše[1,2], D Škorjanc[2], M Čandek-Potokar[1,2]
[1]*Agricultural Institute of Slovenia, Ljubljana, Slovenia,*
[2]*University of Maribor, Faculty of Agriculture and Life Sciences, Hoče, Slovenia*
[*]martin.skrlep@kis.si

Application
Inadequate rearing conditions can increase the risk of boar taint.

Introduction
Whereas accumulation of androstenone is largely under genetic control (Robic *et al.*, 2008), the level of skatole is mainly influenced by steroid hormones (e.g. androstenone) inhibiting its metabolism and by nutrition (Zamaratskaia and Squires, 2009). Rearing conditions may also be important and were analysed in the present study.

Material and Methods
Entire male pigs (EM; n=102) of the same crossbreed and nucleus herd, fed similar corn-based diet were used. One group of EM (n=29) was housed in individual pens (2.7 m^2) and slaughtered at the age of 193 days and 122 kg live weight (LW). The second group of EM was reared in group pens at initial stocking density of 0.7 m^2/pig. At 169 days, slow growing and injured pigs (n=22, LW=74.5kg) were slaughtered, while the remaining were reared for further 35 days at 1.0 m^2/pig and slaughtered at 127 kg LW. Levels of androstenone and skatole were determined in back fat by HPLC and analysis of variance was performed to assess the difference due to housing (individual vs. group) and rearing circumstances (inferior vs. improved).

Results
Comparison of individual and group housing (Table 1) showed 35% higher skatole levels in group housed EM while having similar androstenone level and slaughter weight. Although slaughtered earlier and at lower body weight, the injured and slow growing EM reared at higher stocking density, had 43% higher back fat skatole than their older counterparts reared at lower stocking density. Level of androstenone in the former group (younger, lighter pigs) was, however, 68% lower, indicating their sexual immaturity.

Table 1 Effect of rearing conditions on boar taint compounds (μg/g liquid fat)

	Housing			Rearing circumstances		
Trait	Group	Individual	P-value	Inferior*	Improved**	P-value
Androstenone	0.58±0.07	0.53±0.10	0.650	0.19±0.10	0.59±0.07	0.020
Skatole	0.17±0.01	0.11±0.02	0.016	0.30±0.03	0.17±0.02	<0.001
Live weight, kg	126.7±1.5	122.4±2.0	0.096	74.5±2.3	126.7±1.5	<0.001

*0.7 m^2/pig, slaughtered earlier due to low performance and injuries; ** 1.0 m^2/pig.

Conclusion
Inferior living conditions can cause a significant rise in fat skatole level. Even if EM are slaughtered at younger age, high welfare measures should be considered if boar taint occurrence is to be completely avoided.

Acknowledgments
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Pork production with immunocastration: welfare and environment

V Stefanski[*1], E Labussière[2], S Millet[3], U Weiler[1]
[1]*Behavioral Physiology of Livestock, University of Hohenheim, Stuttgart, Germany.*
[2]*PEGASE, Inra, Agrocampus Ouest 35590, Saint-Gilles, France*
[3]*Flanders research institute for agriculture, fisheries and food (ILVO), Melle, Belgium*
[*]volker.stefanski@uni-hohenheim.de

Application
Optimized pork production with immunocastration as an environmental, economic and socially sustainable alternative to systems with surgically castrated (SC) and entire males (EM).

State of the art and research gaps
Societal pressure to end surgical castration of piglets without anesthesia and pain relief is increasing. Two alternatives to surgical castration currently exist: entire males or immunocastrated males (IC), both with advantages and disadvantages. Considering animal integrity, EM appeared to be the preferred choice. However, welfare problems especially with regard to aggression (i.e. penile injuries) exist (Weiler *et al.*, 2016). Research has shown that immunocastration may solve some of the welfare issues associated with EM production, because IC display less aggressive and sexual behavior than EM (Rydmer *et al.*, 2010). Compared to surgically castrated males, EM and IC grow more efficiently and have a better feed conversion ratio. Despite increased nutrient requirements for the animals, both probably imply lower nutrient excretion, a lower carbon footprint per kg pork and higher nitrogen efficiency (Millet *et al.*, 2018). Because IC can be considered as EM until the second vaccination, after which their feed intake increases drastically while protein deposition remains stable, optimized feeding of IC must be adjusted to minimize excessive amino acid and nitrogen supply. Thus, immunocastration has the potential to be a serious alternative with advantages on animal welfare, ecology and economy. More research is nevertheless required on aspects of animal welfare, especially on the effect of the stability of immunocastration on behavior under varying and/or stressful housing conditions as well as on the feed of IC after the second vaccination to further enhance the economic and environmental sustainability.
The ERA-Net project "SuSI" (*Sustainability in pork production with immunocastration*) launched in 2017 aims to critically evaluate and optimize pork production with IC as an environmental, economic and socially sustainable alternative to systems with SC or EM. The goals are addressed by a multidisciplinary consortium of eight partners with wide expertise covering the overall pork supply chain (https://susi.uni-hohenheim.de). SuSI will thus substantially contribute to close research gaps on aspects of animal behavior, welfare, nutrition and environment.

Conclusion
Precondition for a success of immunocastration in the European market is, among others, an optimized production process and the confirmation of reliability of immunocastration under varying conditions.

Acknowledgements
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Performance and welfare assessment in entire males compared to anti GnRH vaccinated boars

E von Borell[1*], M Weber[2], S Ebschke[1]
[1]*Institute of Agricultural and Nutritional Sciences, Martin-Luther-University Halle-Wittenberg, Theodor-Lieser-Str. 11, 06129 Halle, Germany;* [2] *State Centre for Animal Management and Technology Sachsen-Anhalt, Lindenstr. 18, 39606 Iden, Germany*
[*]eberhard.vonborell@landw.uni-halle.de

Introduction

Immunization against GnRH is practiced as one of the alternatives to surgical castration and raising entire males (EM). The aim of the present study was to assess animal welfare and performance associated with this alternative as well as feeding options to affect organoleptic perception and consumer acceptance positively.

Material and Methods

A total of 185 male pigs were either fattened as EM with a standard protein/energy adapted diet for boars SBD (n=46) or were vaccinated against GnRH (Improvac®) and fed with SBD (n=45), an energy reduced boar diet ERD (n=47) and a diet suited for barrows BD (n=47). They were kept in groups of 12 in four replicates with identical randomly allocated pens. DFI was recorded individually. Animal based indicators like skin alterations, performance and carcass traits were evaluated on farm and abattoir. Furthermore, carcass quality, sensory testing at consumer level and animal behaviour (resting, locomotion, feeding, agonistic interaction) were estimated.

Results

DWG did not differ between groups until the 2nd vaccination but subsequently increased for anti GnRH vac. boars ($p<0.05$; Table 1). FCR was lower for both EM and vac. M fed the SBD in relation to ERD and BD ($p<0.05$).

Lean meat percentage did not differ between groups but dressing percentage was at least 1.3 % higher when fed with SBD (Table 2). IMF was lowest in EM (1.15 %) and differed from all other vaccinated groups ($p<0.05$).

Juiciness was unaffected by treatment but tenderness was slightly reduced in EM vs. vac. fed with SBD ($p=0.046$; Table 3). Flavour was consistently rated higher in all vac. groups ($p<0.05$) and did not differ within these groups. The overall consumer assessment of meat from EM was negatively perceived compared to vac. boars ($p<0.05$).

Table 1 Overall fattening performance

	Ent. Males SBD		Improvac® SBD		Improvac® ERD		Improvac® BD	
	Average	s	Average	s	Average	s	Average	s
Weight 2nd Vac., kg	91.0	10.4	91.9	9.5	92.7	10.6	90.6	9.0
Live Weight, kg	125.9^{a}	8.4	130.0^{b}	8.0	128.7^{ab}	7.3	129.0^{ab}	6.9
Overall DWG, g	874^{a}	96	941^{b}	90	927^{b}	89	920^{b}	89
Overall FCR, kg/kg	2.51^{a}	0.21	2.53^{a}	0.21	2.63^{b}	0.22	2.65^{b}	0.23

Table 2 Carcass traits

	Ent. Males SBD		Improvac® SBD		Improvac® ERD		Improvac® BD	
	Average	s	Average	s	Average	s	Average	s
Slaughter weight, kg	97.5	6.5	98.9	5.7	97.1	5.7	98.2	5.2
Carcass yield, %	77.4^{a}	1.0	76.2^{b}	1.7	75.5^{b}	1.8	76.2^{b}	1.5
Lean meat, %	61.4^{a}	2.1	58.3^{b}	2.3	58.7^{b}	2.1	57.0^{c}	2.2
Drip loss, %	2.4	1.2	2.7	1.4	2.3	1.3	2.3	1.1
Intramuscular fat, %	1.15^{a}	0.51	1.28^{ab}	0.60	1.41^{bc}	0.77	1.46^{c}	0.69

Table 3 Sensory evaluation by consumers rating from 1 (worst) to 6 (best)

	Ent. Males SBD	Improvac® SBD	Improvac® ERD	Improvac® BD
N	46	45	47	47
Juiciness	3.72 ± 0.95	4.00 ± 1.03	4.03 ± 0.96	3.74 ± 0.83
Tenderness	3.64^{a} ± 0.85	4.02^{b} ± 0.92	4.04^{a} ± 0.96	3.81^{ab} ± 0.83
Flavour	3.40^{a} ± 0.65	3.70^{b} ± 0.66	3.83^{b} ± 0.59	3.68^{b} ± 0.64
Overall	3.37^{a} ± 0.65	3.71^{b} ± 0.71	3.85^{b} ± 0.63	3.64^{b} ± 0.68

Increased resting ($p<0.01$), less agonistic interaction ($p<0.001$) and riding events ($p<0.001$) were only observed in groups fed the BD. Skin damage alterations, especially on head and flank, were slightly higher in EM during the whole observation period.

Conclusion

Fattening of EM may create animal welfare and consumer acceptance problems. The advantage of improved body and carcass performance in EM can be best utilised when providing them with an adapted boar diet and vaccination against boar taint.

Penile injuries in immunocastrated and entire male pigs of one fattening farm

S Reiter[1], S Zöls[1], M Ritzmann[1], V Stefanski[2], U Weiler[2*]
[1]*Clinic for Swine, LMU Munich,* [2]*Institute of Animal Science, Hohenheim University, Stuttgart, Germany*
[*]weiler@uni-hohenheim.de

Application

To avoid welfare problems in pork production with entire males (entire males) the incidence of penile injuries has to be reduced. Immunocastrated boars (immunocastrates) were systematically compared in frequency and severity of penile injuries to entire males.

Introduction

The use of entire males for pork production in Germany is still limited. In addition to problems with boar taint, behavioural problems due to aggressive behaviour of entire males to establish social rank order (Rydhmer *et al.*, 2013) and sexual behaviour have to be solved. Recently the relevance of penile injuries for welfare problems in entire males have been reported (Isernhagen, 2015; Weiler *et al.*, 2016). Thus, immunocastration was evaluated for the consequences on frequency and severity of penile injuries.

Material and Methods

A total of 407 boars were raised from about 30 kg (11 weeks of age) to 120-125 kg in a commercial fattening farm, which had been selected for the study due to a known experience in rearing entire males. The animals were assigned to the two treatment groups entire males (n = 215) and immunocastrates (n = 192) in the fattening unit. The immunocastrates were vaccinated twice [1st and 6th week of fattening (fwk), age: 12 and 17 weeks)] with Improvac® (2 mL s.c., Zoetis Deutschland GmbH, Berlin, Germany) and entire males remained unvaccinated. Animals were delivered in 4 batches to the slaughterhouse: 12 fwk (batch1), 13 fwk (batch2), 15 fwk (batch3) and 16 fwk (batch4), respectively. At the slaughter line, the genital tract (penis, covered by the preputial sheet) was excised during evisceration, and further evaluated as described by Weiler *et al.* (2016).

Results

Incidence and size of penile injuries (wounds, scars, hematomas) were evaluated at slaughter. 75.8% of entire males and 48.4% of immunocastrates exhibited injuries at the pars libra of the penis. Scars were observed in 71.2% of entire males and 44.8% of immunocastrates. Scars/animal were significantly influenced by treatment (immunocastrates vs. entire males), batch and treatment x batch and increased with age in entire males (batch1: 2.61 ± 3.05, batch4: 3.59 ± 3.47), but not in immunocastrates (batch1: 2.00 ± 3.02, batch4: 1.22 ± 1.91). Fresh wounds were obvious in 17.2% of entire males and 8.3% of immunocastrates. Number of wounds per animal was only influenced significantly by treatment and was lower in immunocastrates than in entire males.

Conclusion

It is concluded that immunocastration reduces welfare problems due to penile injuries as it reduces the frequency and severity of penile injuries in immunocastrates compared to entire males of the same age and weight.

Acknowledgments

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Influences of different vaccination protocols of immunocastration on endocrine parameters, growth performance, carcass quality, boar taint as well as on penile injuries and behavioural changes

S Reiter*[1], U Weiler[2], P Lindner[3], A Bauer[4], V Stefanski[2], M Ritzmann[1], S Zöls*[1]
[1]*Clinic f. Swine, LMU, Munich,;* [2]*Institute of Animal Sciences, University of Hohenheim, ,* [3]*Bavarian State Research Center for Agriculture Schwarzenau, Schwarzach am Main, Germany;* [4]*Department of Safety and Quality in Meat, Max Rubner-Institute, Kulmbach, Germany,*
*s.zoels@lmu.de

Application
Different vaccination schemes of a GnRH vaccine were evaluated for their effect on growth performance, carcass quality boar taint levels and welfare problems.

Introduction
Immunization of boars against GnRH (Improvac®, Zoetis Deutschland GmbH) is an effective alternative to surgical castration to avoid boar taint (Dunshea *et al.*, 2001) In this study different schemes of GnRH-vaccination were compared to surgical castration and entire males for the effects on growth, carcass quality and endocrine parameter, boar taint and behavior and compared to castrates and entire males.

Material and Methods 128 male piglets (PI x DE/DL) were assigned to five treatments (n=24/group): surgical castrates, entire males or three groups of Improvac-vaccinated boars (immunocastrates). Immunocastrates received Improvac vaccinations twice (2ml s.c.) either at an age of 8 and 12 weeks (immunocastrates-8/12), 12 and 16 weeks (immunocastrates-12/16) or 12 and 18 weeks (immunocastrates-12/18), respectively. Animals were slaughtered at 24 or 26 weeks (batch 1 and 2). Behavioral patterns (mounting attempts, mounting, fighting, biting) were evaluated by direct observations at V2 (second vaccination) and two weeks later. Feces were collected from focus animals every 2nd week (12th to 22nd week) and blood samples were taken during exsanguination at slaughter to analyze testosterone concentrations (Wesoly *et al.*, 2015) and GnRH binding with ^{125}I-GnRH. At slaughter, back fat samples were collected to determine androstenone and skatole concentrations (Fischer et al., 2011) as well as fatty acid composition. Additionally, penile injuries were recorded.

Results
V2 GnRH-vaccination led to a decrease in the number of mounting attempts and mountings in all immunocastrate groups two weeks later. Analysis by GLM repeated measures revealed no significant effect of time ($df = 1.34$, $F = 1.85$, ns) but a significant effect of time x group ($df = 6.98$, $F = 3.32$, $p<0.01$) on fecal testosterone concentrations. In all immunocastrate groups a significant decrease of fecal testosterone to levels observed in surgical castrates was measured in consequence to V2. In immunocastrates serum levels of testosterone were correlated to GnRH binding at slaughter ($r=-0.68$; $p<0.001$; $n=72$) and regression analysis revealed a significant effect of GnRH binding on testosterone concentrations ($B=-0.196$, $SE(B)=0.026$, $p<0.001$). Carcass weight was between 74.5 and 110.6 kg and similar in all groups. Feed per gain varied significantly between groups ($p<0.001$), with lowest values for entire males and the highest for immunocastrates-8/12. Group and batch had a significant effect on fatty acid composition (group: $p<0.001$, batch: $p <0.05$), except PUFA (group: $p<0.001$, batch: $p=0.76$). Entire males had significantly higher androstenone and skatole concentrations than all immunocastrate groups ($p<0.001$). The lowest levels of androstenone and skatole were measured in immunocastrates-12/16 and immunocastrates-12/18. In immunocastrates the percentage of animals with penile injuries was lower than in entire males (91.7%) and ranged between 16.7% (immunocastrates-8/12) and 41.7% (immunocastrates-12/18). Severe injuries in entire males were 12.5% and below 4.2% in all other groups.

Conclusion
GnRH vaccination prevents boar taint reliably, if applied according to manufacturer recommendation. The immunocastrates-8/12 vaccination protocol cannot be recommended with regard to boar taint. Vaccination reduced significantly welfare problems due to penile injuries. Immunocastrates (except immunocastrates-8/12) had an improved feed conversion ratio, leaner carcasses and lower PUFAs than surgical castrates. Thus, immunocastration offers a reliable and animal friendly alternative to surgical castration.

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Meat quality issues in entire male and immunocastrated pigs

M Škrlep[*1], M Čandek-Potokar[1,2]
[1]*Agricultural Institute of Slovenia, Slovenia,* [2]*University of Maribor, Faculty of Agriculture and Life Sciences, Slovenia*
[*]martin.skrlep@kis.si

Application

By replacing surgical castration with alternatives such as entire males or immunocastrates, the pork industry will have to deal with altered meat quality.

Introduction

Alternatives to surgical castrates (SC) like rearing of entire males (EM) and immunocastrates (IC) is becoming a reality imposing the European pig sector to confront many challenges. Besides solving issues related to management, economics, nutrition and welfare, there is also a problem of meat technological quality, potentially affecting processing aptitude and consumer acceptability (Lundström *et al.*, 2009; de Roest *et al.*, 2009).

Results and discussion

Due to the presence or sudden loss of the androgen potential, EM and IC represent metabolically very different status from SC. Differences in behaviour, feed intake and composition of tissue deposition during growth reflect in body composition and apparently also meat quality (Batorek *et al.*, 2012; Pauly *et al.*, 2012). In the case of EM, on top of the presence of unpleasant boar taint, EM were found to have less (and more unsaturated) intramuscular fat, even at the same carcass fatness as SC. Published meta-analyses (Batorek *et al.*, 2012; Pauly *et al.*, 2012; Trefan *et al.*, 2013) and several recent studies confirmed tougher meat of EM. Several reports have also shown inferior water holding capacity (WHC) of EM or IC than SC. Differences between these sex categories in meat colour and pH vary significantly between the studies and are inconclusive. Still, indications exist for EM to exhibit darker meat colour. The background of the reported differences is also not fully understood. Lower amount of intramuscular fat or higher collagen content in EM could directly affect meat tenderness. At the same time, higher collagen synthesis rate in EM could also mean more immature and thus soluble collagen. Indirectly, higher fat unsaturation and oxidative metabolic profile of EM can be related to higher protein oxidation, leading to the loss of protein solubility, denaturation, and myofibrillar aggregation resulting in lower WHC and tougher meat. On the other hand, indications for higher muscle proteolytic potential of EM than SC also exist (i.e. shown for proteolysis index in dry-cured ham and fresh muscle proteome). Other possible factors include the differences in carcass fatness and muscularity (affecting carcass cooling rate and thus muscle early *post-mortem* metabolism and myofibrillar shrinkage) along with pre-slaughter activity and susceptibility to stress (governing the nature of *post-mortem* muscle-to-meat conversion and muscle glycogen reserves).
Regarding meat quality of IC, they are mostly similar to SC but superior to EM in terms of higher IMF levels, better meat tenderness, although some indications for lower WHC and lighter meat colour exist. Their resemblance to either EM or SC depends on the length of the interval between effective immunisation and slaughter. As intensive metabolic changes occur during this period (increased growth and fat deposition) this could potentially induce changes of proteolytic and lipolytic properties of IC tissues. Yet, similarly to EM, the differences are not fully confirmed and aetiology of the processes poorly understood which demands further research, either in fresh or processed meat.

Conclusions

Although numerous studies address the characterization of meat quality of the alternatives to surgical castration, there is a lack of understanding of the aetiology of the differences. The research findings are often inconsistent, which may be assigned to numerous interfering factors, bringing the need for a new meta-analytical study.

Acknowledgements

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Quality control in entire male pig production with particular emphasis on boar taint detection

M Font-i-Furnols*[1], M Čandek-Potokar[2], N Panella-Riera[1], J E Haugen[3], I. Bahelka[4]
[1] IRTA, Finca Camps i Armet, 17121 Monells, Girona, Spain, [2] Agricultural Institute of Slovenia, Hacquetova ulica 17, 1000 Ljubljana, Slovenia, [3] Nofima AS, Osloveien 1, 1430 Ås, Norway,[4] Animal Production Research Centre, Hlohovska 2, 94992 Nitra, Slovak Republic
*maria.font@irta.cat

Application

Meat quality evaluation, including boar taint, in combination with carcass traits would upgrade classification system and optimization of meat processing.

Introduction

Today, pig grading at slaughter line only considers lean content of carcasses. Only some Swiss abattoirs perform a quality assessment of fat tissue that affects the price of the carcass (Bee and Kragten, 2014). Due to welfare concerns, there has been an increase of entire and immuno-castrated males in meat chain as alternative to surgical castration. To avoid tainted carcasses entering to the market, there is a need to consider the on/at line rapid detection of boar taint which would allow sorting them for specific processing and guaranteeing tainted-free meat. This is a challenge since there is a lack of rapid detection methods, and also of cut-off levels to guarantee consumer acceptance are not defined.

Material and methods

A multidisciplinary network has been established within IPEMA that will help to identify requirements for meat quality grading and on-line boar taint detection and to describe the technologies available for this purpose.

Results

On-line boar taint evaluation of skatole with a colorimetric method was developed in Denmark to sort out carcasses with high levels of skatole and in few EU countries, some abattoirs use the human nose to classify male carcasses according its boar taint level. Several works aimed to evaluate rapid methods for boar taint detection like human nose quality control (Trautmann *et al.*, 2014), as well as some technologies were tested for boar taint evaluation like sensors (Hart *et al.*, 2016), Raman spectroscopy (Sørensen *et al.*, 2015; Liu *et al.*, 2016) and mass spectroscopy (Borggaard *et al.*, 2017) providing promising results.

Conclusion

Advances in pig carcass grading at slaughter line are needed that would go further than lean meat content by including traits important for processing, eating quality, boar taint detection. Preferably they should be fast, non-destructive (or non-invasive), automatic or semi-automatic, on-line or at-line, accurate and precise, cost-effective and correlated with consumers' perception of quality.

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Fully automated and rapid at-line method for measuring boar taint related compounds in back fat

R I D Birkler*, C Borggaard, S Støier, B L W Lund
Danish Technological Institute, Danish Meat Research Institute, Gregersensvej 9, 2630 Taastrup, Denmark
*rub@teknologisk.dk

Application

An analytical method for quantification of the two major boar taint compounds skatole and androstenone has been developed, validated and prepared for full automation. The method and procedures have been designed with full automation in mind enabling 2880 samples to be analysed during an 8-hour workday.

Introduction

Producing entire male pigs entails a risk of boar taint. Therefore, a possible stop for surgical castration of pigs in the EU requires action towards sorting malodorous male pigs at the slaughter line. Existing analytical methods are either time consuming, demand manual labour or do not quantify both skatole and androstenone. They are currently not fit for implementation at a slaughterhouse due to high turnover time or total cost of analysis. To meet the requirements of a rapid at-line, analytical method for the simultaneous quantification of skatole and androstenone in back fat while keeping price to a minimum, the presented method was developed, validated and prepared for automation.

Material and methods

Sample preparation: To a 0.35 g back fat sample, 2.5 mL of slightly alkaline brine (0.08 M NaOH) and 2.5 mL acetonitrile with internal standards (skatole-d3 and androstanone) were added prior to homogenization. Post homogenization, samples were centrifuged to obtain phase separation. The upper organic phase was subsequently deposited to a LazWell™ plate. After evaporation of the organic solvent, the residue was analysed by laser diode thermal desorption tandem mass spectrometer (LDTD-MS/MS). Instrumentation: Sciex 6500 QTRAP MS-MS mass spectrometer (AB Sciex, MA, U.S.A) with a LDTD model 960S with built in atmospheric pressure chemical ionization (APCI) (Phytronix, Québec, Canada). The LDTD-MS/MS monitors two transitions for each compound. Compounds are quantified against a 6-point external calibration curve prepared by spiking castrate back fat samples at appropriate levels.

Results

The method is specific towards the quantified compounds in back fat samples. Trueness was examined by comparing quantitative results from in-house HPLC-UV or external GC-MS analyses. The method's limit of detection (LOD) and limit of quantification (LOQ) were determined by 10 repeated measurements on a spiked blank castrate back fat sample. The standard deviation (SD) was used to determine LOD (3 x SD) and LOQ (10 x SD). LOQ for skatole and androstenone are below the current sorting limit of skatole (0.25 µg/g). Results are shown in Table 1.

Table 1 Limit of detection (LOD) and limit of quantification (LOQ) for skatole and androstenone

	LOD	LOQ
Skatole	0.02 µg/g	0.05 µg/g
Androstenone	0.05 µg/g	0.1 µg/g

The precision of the analytical method was performed on whole back fat samples as well as homogenized back fat samples to minimize contribution from sample inhomogeneity. Double determinations were analysed on five different days. Results from homogenized back fat samples are presented in Table 2. For both compounds, the RSD% were below 6%. Based on the laboratory results and optimizations, a detailed plan for full automation was designed.

Table 2 Precision results (RSD%) for skatole and androstenone at two concentration levels(1)

	Skatole			Androstenone		
	Concentration	S_R (%)	S_r (%)	Concentration	S_R (%)	S_r (%)
Sample 1	0.21 µg/g	5.5	3.1	3.9 µg/g	3.1	2.6
Sample 2	0.90 µg/g	5.3	5.3	1.6 µg/g	2.7	2.7

(1) It should be noted that the evaluated samples were homogenized back fat samples. Homogenized samples were used in order to minimize influence from the natural sample inhomogeneity. S_R is reproducibility (RSD%) and S_r is repeatability (RSD%).

Conclusion

The developed analytical method simultaneously quantifies the two major boar taint compounds skatole and androstenone in back fat. The limits of quantification are below current or expected future sorting levels. The precision of the analytical method is below 6% for both compounds. In addition, the method meets the criteria for subsequent automation at the slaughter line. Once implemented, the method's capacity is 2880 samples during an 8-hour workday at a price of 1 €/sample. Time of analysis by LDTD-MS/MS is 10 seconds with a total time of 40 minutes from sampling at the slaughter line until the analytical results can be found in the abattoir database.

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Quality and sensory evaluation of meat from entire males after adding hydrolysable tannins to diet

I Bahelka[1*], O Bučko[2], E Hanusová[1]
[1]*National Agricultural and Food Centre – Research Institute for Animal Production, Lužianky, Slovakia,*
[2]*Slovak Agricultural University, Nitra, Slovakia*
[*]bahelka@vuzv.sk

Application

Supplementation of diet of entire male pigs with hydrolysable tannins had no negative effect on sensory properties of pork evaluated by consumers. On the contrary, this addition reduced the skatole concentration in adipose tissue of boars. Thus, tannin supplementation can be recommended for boar taint reduction at future feeding of entire males after foreseeable ban on surgical castration of piglets.

Introduction

Surgical castration of piglets minimizes an unpleasant off-flavour (boar taint) which otherwise can develop in meat when boars reach puberty and is generally rejected by the consumers. According to current trends (European declaration on alternatives to surgical castration of pigs), this practice has been considered as animal welfare problem and is likely to be abandoned. Therefore, fattening of entire males may become a more widespread practice in the EU countries. However, to avoid consumer dissatisfaction, sustainable methods for minimizing boar taint levels in the meat need to be developed. One of the options is the use of feed additives to the diet of uncastrated boars. The tannin compounds are widely distributed in many species of plants, where they play a role in protection from predation, possibly also as pesticides, and might help in regulating plant growth. A recent study by Čandek-Potokar *et al.* (2015) demonstrated that supplementation with 1 and 2 % extract of hydrolysable tannins induced activity of cytochrome CYP450, likewise potential positive effects on reduction of skatole concentration – one of two main compounds of boar taint. However, further studies are needed to validate first results.

Material and methods

Thirty pigs – progeny of Landrace sows and Yorkshire x Pietrain boars – from 6 litters were assigned to 3 groups (each of 10) with different level of tannins supplementation. They were housed by pairs in the pen and fed commercial diet (12.8 MJ ME, 17.2 % crude proteins). Trial started from 30 kg of live weight and finished at 122 ± 4 kg. Males diet was supplemented with 0, 1 or 2 % (T0, T1 and T2) of chestnut wood extract (Farmatan, Slovenia). A day after slaughter, pork quality parameters were measured and samples of *longissimus dorsi* muscle (100 g) for sensory evaluation as well as back fat for androstenone and skatole analyses were taken. Sensory properties of pork were evaluated 4 days after slaughter in consumer test. Statistical analysis was carried out using GLM procedure of SAS (SAS Inc. Cary, USA) with fixed effect of treatment group (tannin addition).

Results

Significant differences between T0 and T1, resp. T2 were found in electrical conductivity measured 24 hours in *musculus semimembranosus* (3.71 vs. 2.27, 2.55 μS). Other pork quality traits were not influenced by tannin supplementation. Effect of this addition on odour, flavour, juiciness and tenderness of pork was not significant comparing control and experimental groups. However, tannins significantly reduced skatole concentration in fat tissue at 2 % addition compared to control (0.011 vs. 0.34 μg/g, $P<0.05$). Effect of tannins on androstenone deposited in fat was not significant (0.14, 0.12 and 0.09 μg/g in T0, T1, T2).

Conclusion

There were no significant differences in sensory evaluation of pork from entire males after supplementation of boar diet with hydrolysable tannins. However, tannins might exert potential for boar taint prevention via reduced deposition of skatole in subcutaneous fat in entire male pigs.

Acknowledgments

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The quality of meat from Romanian pigs, surgical castrated *versus* entire male

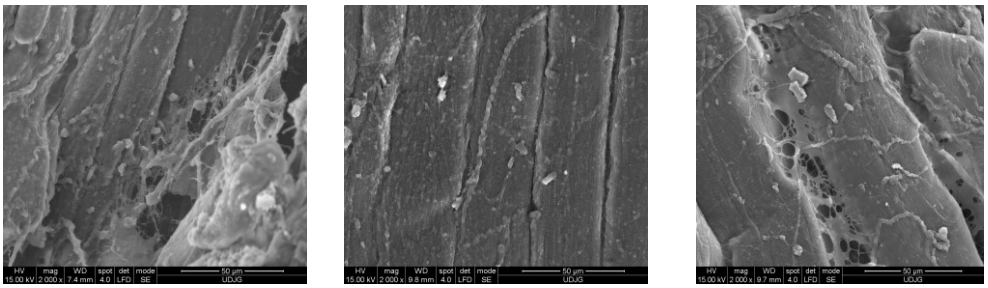
G Iordăchescu*[1], M Stoica[2], R M Dinică[3], A Cantaragiu[2], I Grecu[1], A O Mihalache[1]
[1]*SAIABA, Dunărea de Jos University,Faculty of Food Science and Engineering, Galați, Romania;* [2]*Department of General Sciences, Dunărea de Jos Univesity, Cross-border Faculty of Humanities, Economics and Engineering, Galați, Romania;* [3]*Chemistry, Physics and Environment, Dunărea de Jos University, Faculty of Sciences and Environment, Galați, Romania.*
*gabriela.iordachescu@ugal.ro

Application
The aim of the study is to evaluate the pork structure and the pork composition of indigenous Mangalita breed pigs.

Introduction
Romanian pig breeds have a number of characteristics related to the meat/fat ratio, age, the nutritional composition of the diet, the way of growth, characteristics that influence the quality of meat and meat products. The European Commission will end the piglet surgical castration from 2018 (Borrisser-Pairo *et al*., 2016) and one of the alternatives is meat consumption from entire males. This can influence the acceptance in terms of texture (Sorapukdee *et al*., 2013), odour, colour, and quality of products.

Material and methods
Fifteen indigenous Mangalita pigs (5 surgically castrated males, 5 entire males and 5 females) around 60 kg live weight were selected for slaughtering. Two types of tissues were compared: one sample of the muscle *longissimus dorsi* and another one from the neck muscle were collected at five hours after slaughter. The measurements were performed in triplicates using some methods like AOAC (chemical composition), L* a* b*parameters (colour), Scanning Electron Microscopy (structure), TLC chromatographic and FTIR spectrometric method (skatole and androstenone). ANOVA was used for statistical analyses.

Results
Generally, the castration showed no significant influence on chemical composition and on colour L*, a*, b* parameters. The contents in intramuscular fat were higher for castrated (according to literature data), providing better nutritional quality, while no significant influence on the meat structure was noticed (Figures 1a, 1b and 1c).



a *b* *c*

Figure 1 Scanning Electron Microscopy images showing the fibers of muscle neck from (a) castrated male, (b) entire male, (c) female

From the Figure 1a it can be noticed that the muscle cells are surrounded by endomysium connective tissue which keeps the muscle cells (fibres) firmly attached to one another. The myofibrils are strongly attached to the sarcolemma. Figure 1b shows a fine, uniform structure. The myofibrils are strongly attached to the sarcolemma. Figure 1c reveals the muscle cells that are surrounded by endomysium connective tissue.

Conclusion
The image analyses on pork coming from castrated male, entire male and female pigs show no significant influence on the meat structure. The physico-chemical composition of the castrated pig samples, entire male samples, and female samples did not present significant differences. Moreover, the quality of products obtained from entire male meat is similar to the quality of products obtained from castrated pigs. Therefore, the consumption of meat products obtained from entire male is a good alternative to consumption of meat products obtained from castrated pigs.

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Impact of immunological castration on the carcass quality of final hybrids in commercial Ukrainian pig farms

M Povod[1], O Kravchenko[2], A Getya[3*], T Kodak[2]
[1]Department of feed technology and animal feeding, Sumy National Agrarian University, Sumy, Ukraine, [2]Department of technology of processing of animal products, Poltava State Agrarian Academy, Poltava, Ukraine, [3]Department of genetics, breeding and animal biotechnology, National University of Life and Environmental Sciences of Ukraine, Kyiv, Ukraine.
[*]getya@ukr.net

Introduction

Nowadays civil society-pays attention not only to increasing of the productivity of pigs and improving of the quality of livestock products, but also to creation of comfortable conditions for animals, trying to develop new technological elements like immunological castration (Weiler *et al.,* 2016). Ukrainian producers also interested in such approach.

Material and Methods

Two groups of hybrid pigs were formed. Animals in one group were surgically castrated at the age of 4 days while in the other group they were twice vaccinated with Improvac vaccine in accordance to the instructions for use. All animals were slaughtered at a live weight of 100 kg. The thickness of the back fat was measured at the level of 6-7 thoracic vertebra (TI) and at sacrum (TII). Dressing percentage was calculated as the proportion of carcass weight without head and anterior legs to the animal live weight.

Results

After comparison of the carcasses of immunological and surgical castrated males it was established that the carcasses of immunologically castrated males had lower back fat thickness at the level of 6-7 thoracic vertebrae by 4.64 mm ($P<0.001$) and at sacrum by 2.73 mm ($P<0.05$) (Table 1), which can be explained by lower fat content and coincided with the results of other studies (Povod *et al.*, 2017).

Table 1 Carcass quality of experimental pigs (n=60)

Groups	LMP, % (Fat-o-Meat'er S71)	Backfat thickness, mm		Dressing percentage, %	Loss after cooling, %
		T I	T II		
Surgical castration	52.63 ± 0.83	23.57 ± 0.79	15.03 ± 0.86	71.43	1.36 ± 0.07
Immunological castration	54.84 ± 0.90	18.93 ± 0.71	12.30 ± 1.05	73.42	1.46 ± 0.19
RMSE	1.81	4.37	2.01	-	0.50
P-value	-	<0.001	<0.05	-	-

Conclusion

Introduction of immunological castration promotes the decreasing of intensity of deposition of subcutaneous fat as well as increasing of lean meat percentage in pig's carcasses.

Acknowledgments

The authors express their gratitude to general director of group of companies "Hlobyno" O.V.Kuzmynskyy for the assistance in organizing of research.

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Slovenian results within JRC-IRMM organised boar taint reference method ring test

Š Velikonja Bolta*, H Baša-Česnik
Central Laboratories, Kmetijski inštitut Slovenije, Ljubljana, Slovenia
*spela.bolta@kis.si

Application
Boar taint compounds determination in fat.

Introduction
JRC-IRMM in Geel organised inter-laboratory comparison study to test the method for the determination of boar taint compounds skatole, androstenone and indole in fat samples in 2014. Fifteen laboratories from 9 countries participated. In order to perform the proposed HPLC-APCI-MS/MS method, some adaptations had to be made to the equipment of the laboratory.

Material and Methods
All standard materials and SEC (size exclusion chromatography) column were sent by the organiser. Solvents used (methanol, toluene, nonane and 1-octanol) were HPLC gradient grade or higher (Sigma-Aldrich). The method provided by the organiser of inter laboratory comparison JRC-IRMM was used. The fat was liquefied, dissolved in methanol containing internal standards and SEC eluent and cleaned on SEC column. After addition of keepers, the extract was vacuum dried under nitrogen and dissolved in appropriate solvent for GC/MS or LC-MS/MS determination. GC separation was achieved on HP 5 MS, 30 m * 0.25 mm, 0.25 µm film chromatographic column with temperature gradient and LC separation on Luna 150 x 4,6 mm, 5 µm 100 A with gradient of 0.1% formic acid and 0.1% formic methanol.

Results
Each laboratory received 5 samples, 3 samples of spiked lard and 2 samples of pork fat tissue, already milled and frozen.

Table 1 Results of inter-laboratory comparison

Sample	Indole [µg/kg]			Skatole [µg/kg]			Androstenone [µg/kg]		
	R	G	L	R	G	L	R	G	L
T1	103	110	108	272	271	252	1946	1732	***2802***
T2	121	111	127	221	203	190	701	695	***998***
L1	105	87	66	104	112	99	4113	3300	4281
L2	386	343	461	1150	1140	1275	1159	981	1211
L3	1046	756	1311	362	387	396	335	316	386

R- reference value; G- GC-MS; L LC-MS/MS; ***bold italic*** results with $|z|>2$

Conclusion
The results obtained with GC-MS were better than those obtained with LC-MS/MS, but considering the circumstances, the results were good.
The proposed reference method is very time consuming.

Acknowledgments
This work was financially supported by the Slovenian Research Agency (grant P4-0133). The authors thank Mateja Fortuna for technical assistance.

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Parallel analysis of boar taint compounds in meat and in back fat

S Ampuero Kragten*, H Chavarria-Drecourt, G Bee
Agroscope, CH-1725 Posieux Switzerland
*silvia.ampuero@agroscope.admin.ch

Application

Most of the studies dealing with boar taint, whether from a breeding, nutrition or management point of view, but especially sensory and consumer tests including meat products are based on the quantification of boar taint compounds in adipose tissue instead of meat. The present results strongly suggest that the analysis of meat or meat products provide a more complete information and ultimately may help to overcome some of the discrepancy between sensorial and boar taint analysis.

Introduction

The lipophilic characteristic of androstenone (A) and, to a slightly lesser extent, skatole (S) and indole (I) facilitates the analysis of these compounds in adipose tissue or in the liquid fat and pure fat melted from it. Indeed, with an intramuscular fat level (IMF) ranging from 2 to 15 %, the expected concentrations of A, S and I in meat are well below the detection limits of most common analytical techniques. However, boar taint compounds are generally determined in adipose tissue whereas sensorial analysis with panelists or consumers are primarily performed in meat. Therefore, it seems legitimate to ask the question of how well correlated the concentration of boar taint compounds in adipose tissue and in meat are.

Material and methods

The present data are based on samples from 22 boars (Swiss Large White) reared and slaughtered at Agroscope at 170±5 days of age with an average body weight of 109±13 kg. The analysis was performed both, in adipose tissue (AT) from the neck region (stored under vacuum at -20°C) and in muscle (*Longissimus thoracis*, 10thrib, stored freeze-dried in powder form) from the same carcasses. Androstenone, S and I were quantified with HPLC, using the same extraction solution of methanol containing androstanone and 2-methyl indole as corresponding internal standards (IS). The analysis of AT was performed in duplicate in liquid fat after heating the AT in a microwave and subsequent extraction of boar taint compounds with methanol-IS (Pauly *et al.*, 2008; Ampuero Kragten *et al.*, 2011). For the analysis of muscle samples (triplicate), the extraction of liquid fat with petrol ether was performed first, followed by a similar procedure as the analysis of liquid fat from AT. The recovery rate of fat from muscle samples was 96%, and the overall recovery rate for A was 94% at 1.5 ppm. The contents in meat were calculated from the contents in muscle-liquid fat using the corresponding IMF value.

Results

Surprisingly, very low coefficients of determination (≤ 0.02) were found between A, S and I contents in meat, or IMF (liquid fat from meat), and liquid fat from AT. By eliminating the 3-4 most diverging points, the determination coefficients could improve to 0.6 to 0.7 for S but only to 0.3 for A. Furthermore, a determination coefficient of 0.18 was found between the A content in meat and the IMF. Although in the case of S the R^2 showed a higher value, 0.24, this was heavily dependent on two extreme points (Figure 1).

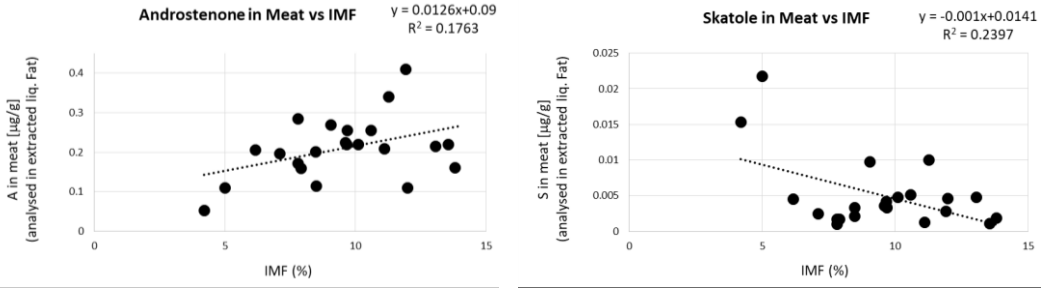


Figure 1 Androstenone and Skatole content in meat (analysed in extracted liquid fat) versus IMF.

Conclusion

The analysis of boar taint compounds is commonly performed in AT, which is the most practical way from an analytical point of view. However, the information thus obtained is probably not complete. Although the number of samples presented here is not sufficient for the determination of robust correlation coefficients, these results nevertheless show the need to carry out the analysis directly in meat or meat products, which are the main food commodities actually consumed.

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Effect of sampling location on concentration of boar taint compounds in fat tissue

N. Batorek Lukač*[1], Š. Velikonja Bolta[1], M. Škrlep[1], U. Tomažin[1], M. Čandek-Potokar[1,2]
[1]*Agricultural Institute of Slovenia, Ljubljana, Slovenia,* [2]*University of Maribor, Faculty of Agriculture and Life Sciences, Hoče, Slovenia*
*nina.batorek@kis.si

Application
Present study aims to explore whether sampling location on the carcass affects the concentration of skatole and androstenone in fat tissue.

Introduction
Boar taint, an offensive odor of meat from some, mostly entire, male pigs is caused by androstenone and/or skatole. Due to their lipophilic nature, both substances are accumulated in fat, however, little information is available whether the deposition is even among fat depots and consequently whether sampling location might have an effect on the concentration of boar taint compounds in fat tissue.

Material and Methods
Samples of leaf fat (LF), fat tissue from the belly (B) and backfat tissue at the position of withers (W), last rib (LR) and above *Gluteus medius* muscle (S) were collected from 4 entire males, 2 females and 6 castrated pigs on the left half-carcasses 45 min *post mortem* and analysed according to the method described in Batorek *et al.* (2012). Results were analyzed using the paired t-test procedure of SAS (PROC TTEST, SAS Inst., Cary NC, USA).

Results
Results are presented in Figure 1. No differences between sampling location were observed for androstenone concentrations (P>0.05), whereas concentrations of skatole determined in LF differed from other sampling locations (P<0.05).

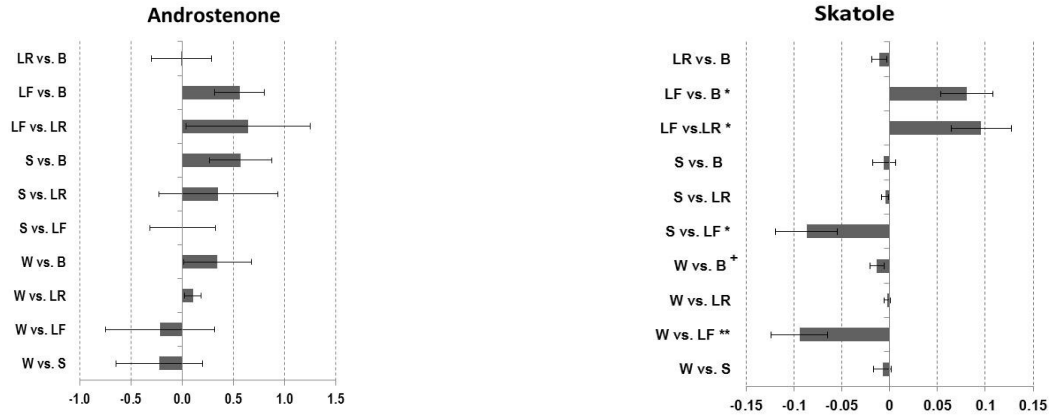


Figure 1 Difference in androstenone (n=4) and skatole (n=12) concentrations (µg/g liquid fat) between sampling locations (LR=back fat at the position of the last rib, B=belly fat, LF=leaf fat, S=backfat above Gluteus medius muscle, W=backfat at the position of withers,**P<0.01, * P<0.05, + P<0.10).

Conclusion
Sampling location may affect the concentration of skatole determined in fat tissue, whereas no effect seems to be present on androstenone concentration.

Acknowledgments
The authors acknowledge the financial support of the Slovenian Agency of Research (grant P4-0133).

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The effect of sex on performance and carcass quality in commercial pig farms

A Van den Broeke*, F Leen, M Aluwé, J Van Meensel, S Millet
ILVO (Institute for Agricultural, Fisheries and Food Research), Scheldeweg 68, 9090 Melle, Belgium.
*alice.vandenbroeke@ilvo.vlaanderen.be

Application

The choice between raising immunocastrates or entire males, as an alternative to barrows, has implications in terms of growth performance and carcass quality.

Introduction

Studies in research facilities show that immunocastration results in an increase in feed intake, accompanied with an increase in daily gain, causing alterations in carcass- and meat quality (Batorek *et al.*, 2012; Van den Broeke *et al.*, 2016). Performance and carcass quality depends on genetics, feeding and management strategy. Studies conducted in experimental conditions do not always reflect reality on pig farms. In this study, we wanted to investigate the differences between barrows, entire male pigs and immunocastrates under Belgian farm conditions: fed with commercial feed, housed in larger groups and at varying slaughter weights.

Material and Methods

In this study three farms were monitored with a total of 786 pigs (barrows, entire males, immunocastrates), divided over 126 pens. All pigs were a cross between a Belgian Piétrain and a hybrid sow. They were given regular commercial ad libitum multi-phase feed and free access to water. The aim was to vaccinate the immunocastrates for a second time 4 weeks before slaughter. The slaughter weight varied between 99 and 138 kg. Daily feed intake, daily gain and feed conversion ratio were recorded between 24 kg and slaughter.

Results

Feed intake was highest for barrows, intermediate for immunocastrates and lowest for entire males ($P < 0.001$). The daily gain of entire males was significantly lower compared to barrows and immunocastrates, which did not differ ($P < 0.001$). Feed conversion ratio or dressing percentage did not differ between immunocastrates and entire males, but both were significantly lower than barrows ($P < 0.001$). Meat percentage was highest and ham and back fat thickness lowest in entire males compared to barrows. Immunocastrates achieved an intermediate result between entire males and barrows ($P < 0.001$). This resulted in the highest overall carcass quality and therefore the best payout per kg of carcass for entire males, intermediary for immunocastrates and the lowest for barrows (Figure 1).

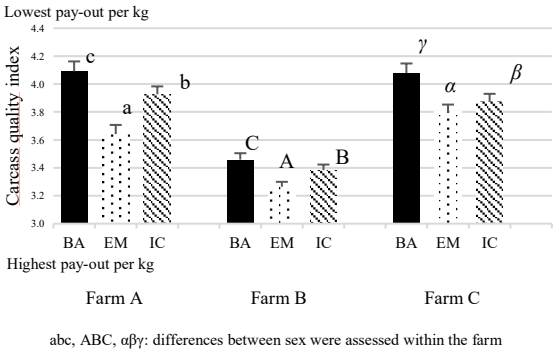


Figure 1 Carcass quality index

Conclusion

The effect of sex on performances and carcass quality in commercial pig farms are in line with results obtained in experimental settings. Although farm and slaughter weight has a significant effect on performances and carcass quality, in all cases, barrows had the worst feed conversion ratio and worst payout compared to the other sexes.

Acknowledgments

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Quality of pig carcasses of surgical- and immuno-castrated males slaughtered at different live weights in commercial Ukrainian pig farms

M. Povod[1], O. Kravchenko[2], A. Getya[3*], O. Kodak[2]
[1]*Sumy State National University, Ukraine,* [2]*Poltava State Agrarian Academy, Ukraine,* [3]*National University of Life and Environmental Sciences of Ukraine, Ukraine*
[*]getya@ukr.net

Application The results of this research can be used by the pork-producing farmers, which would like to increase the weight of pigs before slaughter.

Introduction There is a trend to slaughter the pigs at heavier live weights, because of better economic effectiveness of fattening (Piao *et al.*, 2004). Traditionally, Ukrainian pork producers slaughter the animals at live weights of 100-110 kg but they would like to increase the weight before slaughter, working with hybrid pigs. Increasing weight before slaughter might decrease carcass quality, especially in surgically castrated and entire males. In that regard, application of immunological castration can be seen as one of possible technological solution (Weiler *et al.*, 2016).

Material and methods The trial was organized at commercial farm conditions on hybrid animals (Yorkshire*Langrass*Maxgro). Two groups of 30 pigs were formed (surgical and immunological castrated males). Surgical castration (SC) was done at the age of 4 days. Immunological castration (IC) was performed by application of Improvac vaccine in accordance to the instructions for use. In each group, 10 animals were slaughtered after reaching live weight of 100, 110 and 120 kg, respectively. Carcass evaluation based on dressing percentage (DP) calculation was made using Fat-o-Meater S71 (Frontmatec, Denmark). The thickness of the backfat (BT 6/7) was measured at the level of 6-7 thoracic vertebra and at sacrum. Carcass grading was done using EUROP and Ukrainian scale. Sensory evaluation of carcasses after slaughter was performed.

Results Carcasses of immunocastrated males had lower backfat thickness measured at 6-7 thoracic vertebra: slaughtered at 100 kg – by 2.8 mm; slaughtered at 120 kg – by 3.3 mm, was coincided with the results of our previous study (Povod *et al.*, 2017a). Because of large variation of data within each group, none of the differences was significant. Dressing percentage of carcasses of immunocastrated males was slightly higher, but not significant, in all weights categories comparing to surgical castrated. Similar results were obtained in our former experiments (Povod *et al.*, 2017b). After slaughter at 120 kg weight 70% of carcasses of immunocastrated males were graded E + U, while in the group of surgically castrated males such carcasses were only 50%. No critical level of boar odour was detected at any slaughter weight.

Table 1 Results of slaughter traits of pigs at different live weights castrated with different methods.

	Traits (at different weight at slaughter)								
	BT 6/7 (mm)			DP (%)			E+U (%)		
	100 kg	110 kg	120 kg	100 kg	110 kg	120 kg	100 kg	110 kg	120 kg
SC	24.4±1.0	25.2±1.1	28.0±2.7	51.6±1.6	48.0±1.7	49.6±1.5	70	40	50
IC	21.6±1.6	25.2±2.2	24.7±1.2	52.0±1.1	49.4±1.8	51.0±1.0	80	60	70
RMSE	1.48	–	1.12	0.21	0.56	0.78			
P-value	–	–	–	–	–	–			

BT: back fat thickness; DP: dressing percentage, E+U: Carcasses classified as E and U lean meat content categories

Conclusion Increasing the live weight before slaughter until 120 kg did not affect negative the carcass quality of surgical and immunological castrated males. Carcasses of immunocastrated males had slightly higher dressing percentage and lower backfat thickness.

Acknowledgements The authors express their gratitude to general director of group of companies "Hlobyno" O.V. Kuzmynskyy for the assistance in organizing of research and IPEMA research team for professional advices delivered during the conducting of experiment.

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Retrospective by sex of the new EU lean meat content of pig carcasses

G Daumas
IFIP-Institut du Porc, BP 35104, 35601 Le Rheu Cedex, France
gerard.daumas@ifip.asso.fr

Application

The provided inputs on carcass composition should allow a better assessment of the technical and economical balance between entire and castrated males. Moreover, when the carcass grading methods will be updated, it seems worthwhile to consider separate slopes for entire males, females and castrated males.

Introduction

A new EU regulation on carcass classification applies from July 2018 (European Commission, 2017). This regulation changes the definition of the reference lean meat percentage (LMP) which becomes the LMP in the carcass (from total dissection). Nevertheless, each Member State can choose when he will update its grading methods. Sex is registered online in France during pig classification conversely to most of European countries. The production of entire males started in France in 2013 (13% of the males), increased quickly and is now stable (27% of the males in 2017), around 2.5 million a year. More than 95% of entire males are classified with the grading method CSB Image-Meater® (IM) approved by the EU in 2013 (European Commission, 2013). The aim of this work is to simulate the LMP change over the last five years in France for each sex.

Material and methods

A sample of 180 pigs was uniformly stratified on sex: 60 entire males, 60 females and 60 castrated males. Carcasses were measured online by the IM and the two fat depths (G3 and G4) and two muscle depths (M3 and M4) were collected. After overnight chilling, the left sides were cut and all the cuts were scanned by computed tomography (CT). The new LMP was calculated according to the unbiased pan-European CT procedure developed by Daumas and Monziols (2018). A prediction equation was calculated by a general linear model including the four IM depths as well as the interactions with sex. Model was selected with the stepwise procedure by using BIC. This equation was then applied on the annual classification averages from 2013 to 2017 (Uniporc Ouest, 2014-2018).

Results

The selected model included one fat depth (G3) and two muscle depths (M3 and M4), G3 and M3 coefficients depending on sex. The RMSE was 2.15. Removal of M4 only decreased the RMSE of 0.01. The fat coefficient of entire males was more than twice that of females, while that of castrates was in the middle. Differences on M3 coefficient had lower impact on LMP. Table 1 gathers the LMP by sex over the years 2013 to 2017. In 4 years, LMP increased, but differently according to sex: +0.11 for entire males, +0.25 for females and +0.35 for castrated males. In the same period the difference between entire and castrated males decreased from 4.02 to 3.78, while the difference between entire males and females decreased from 1.37 to 1.23; the difference between females and castrated males reduced from 2.65 to 2.55.

Table 1 Changes in LMP by sex over time

	Year				
Sexual type	2013	2014	2015	2016	2017
Entire males	62.13	62.28	62.25	62.19	62.24
Females	60.76	60.83	60.89	60.99	61.01
Castrated males	58.11	58.15	58.22	58.39	58.46

Conclusion

The prediction equation of lean meat content in pig carcass from split line measurements is sex-dependent. The slopes of both fat and muscle depth are affected. In four years, the differences of lean meat content between sexes have slightly been reduced.

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Sensory analysis of ham from entire male pigs raised with different feeding and housing conditions

R Pinto[1*], N Reis[1] C Barbosa[1,2], R Pinheiro[1,3], M Vaz Velho[1]
[1]*Escola Superior de Tecnologia e Gestão - Instituto Politécnico de Viana do Castelo, Portugal,* [2]*LAQV-REQUIMTE, Faculdade de Farmácia da Universidade do Porto, Portugal.,* [3]*CEB, Universidade do Minho, Portugal.*
[*]pintoricardo@ipvc.pt

Application
This study aims at evaluating sensory quality of cooked ham produced with entire male pigs raised under specific conditions in order to reduce or eliminate the boar taint.

Introduction
Rearing entire male pigs is one of the alternatives to surgical castration however there is the risk of boar taint occurrence (Wauters *et al.*, 2017). Boar taint is a sensory off-odour/off-flavour found in meat from entire male pigs due to two main compounds: androstenone and skatole (Font-i-Furnols, 2012). Literature indicates reduced incidence of boar taint when boars are fed with inulin (Aluwe *et al.*, 2013). Housing conditions and genetic selection can also have a favourable effect on boar taint reduction (Backus *et al.*, 2016). The production of pork from entire male pigs in enriched housing conditions seems also to be a promising alternative to castration, as improved husbandry aims at high standards of animal welfare and elimination of mutilations (Holinger *et al.*, 2015).

Material and methods
Meat from entire male pigs of a crossbreed (progeny of Large White x Landrace gilts sired by Pietrain boars), raised under six different conditions (normal housing versus improved housing, and fed 2 months prior to slaughter with different levels of added inulin: 0%, 3% and 6%) was used to produce six batches of cooked ham, processed under the same conditions. Samples were coded as N0%, N3%, N6%, C0%, C3% and C6%, where "N" means normal housing, "C" improved housing and 0, 3 and 6 correspond to percentage of added inulin in animal feed. A quantitative descriptive analysis was performed by 11 trained panellists, assessing odour and flavour of skatole and androstenone, texture and appearance, on a 1 to 10 scale. Perception of defects on overall appreciation and appearance was evaluated on a 1 to 5 scale, where scores below 3 meant there was a perceived problem. A total of eight coded samples, with the six conditions and two replicates to evaluate repeatability. ANOVA with a *post hoc* Fisher's LSD test was used to investigate significance of observed differences and Canonical Variates Analysis (CVA) with judges as replicates, was carried out to discriminate groups of samples.

Results
Results show significant differences ($p<0.05$) in the evaluation of attributes such as flavour and odour of androstenone and skatole, being N0%, N3% and N6% the samples with highest intensity scores meaning they had stronger boar taint odour and flavour. CVA separates N0% from the other for almost every condition. Ham from animals with improved housing conditions had better appearance scores than normal housing. No differences were found in texture and overall appreciation.

Conclusion
It can be concluded that the effect of inulin addition in pigs feed was not clear, but it was found that its addition could have positive effects when conjugated with better housing conditions. As expected, the presence of boar taint was perceived in meat samples where no addition of inulin and no improved conditions were applied.

Acknowledgements
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Quality of dry-cured hams from entire males in relation to boar taint level[1]

T Kaltnekar[1], M Škrlep[2], N Batorek Lukač[2], U Tomažin[2], M Prevolnik-Povše[2,3], E Labussière[4], L Demšar[5], M Čandek-Potokar[*2,3]
[1]Pršutarna Lokev na Krasu, Lokev, Slovenia, [2]KIS-Agricultural Institute of Slovenia, Ljubljana, Slovenia, [3] University of Maribor, Faculty of agriculture and life sciences, Pivola, Slovenia, [4]INRA UMR 1348 Pegase, Saint-Gilles and Agrocampus, Rennes, France, [5]University of Ljubljana, Biotechnical faculty, Dpt. Food science and technology, Ljubljana, Slovenia
[*]meta.candek-potokar@kis.si
The results presented in this abstract were published in Kaltnekar *et al.*, 2016.

Application
Boar taint could be more pronounced in dry-cured hams with lower salt content.

Introduction
In future, the rearing of entire males may take a significant share in European pig production. Information about the aptitude of entire males for dry-curing and final product sensory quality is lacking.

Material and Methods
Green hams (n=16; 12.8 ± 1.4 kg) from 8 crossbred entire males were processed for 16 months according to the rules of consortium Kraški pršut. Two salting regimes were applied; LS–shortened salting to the right hams or HS–standard salting regime to the left hams. Boar taint substances androstenone and skatole were quantified in back fat of dry-cured hams by HPLC (see Batorek *et al.*, 2012). Chemical, rheological, sensory traits were assessed as described in Kaltnekar *et al.* (2016). The median of androstenone concentration (0.78 µg/g liquid fat) was used to classify the hams into low (LBT) and high boar taint (HBT).

Results
Compared to LBT, HBT hams were more proteolysed, had softer texture, higher bitterness, pastiness and more pronounced off-flavours (Kaltnekar *et al.,* 2016). The effect was seen in HS and LS hams (no interaction). Exception was the off-flavours, which were perceived more intensely in low salt hams (Table 1).

Table 1 Effect of salting and boar taint level on perceived off-flavours (Kaltnekar *et al.*, 2016)

	HS		LS		P-value			
Off-flavours	LBT	HBT	LBT	HBT	Salting	Boar taint	S × BT	RMSE
Biceps femoris	0.8[a]	1.0[a]	1.3[a]	2.5[b]	0.000	0.002	0.014	0.3
Semimembranosus	0.6[a]	0.8[a]	1.2[a]	2.1[b]	0.001	0.006	0.071	0.3
Fat	0.9	1.1	1.0	1.6	0.254	0.081	0.330	0.4

LS- shortened salting; HS- standard salting; LBT-low boar taint; HBT-high boar taint

Conclusion
Our results indicate that off flavours are less perceived in saltier hams; either salt covers off-flavours or there are additive effects of boar taint substances and proteolysis products when salt content is low.

Acknowledgements
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Effect of slaughter age of entire males on cooked ham quality

N Reis, R Pinto* ,C Barbosa, M Vaz Velho
Escola Superior de Tecnologia e Gestão do Instituto Politécnico de Viana do Castelo, Portugal.
*rpinto@ipvc.pt

Application

The use of entire males older than 8 months in the production of cooked ham.

Introduction

Boar taint is a sensory off-odour/off-flavour found in meat from entire male pigs due to two main compounds: androstenone and skatole (Font-i-Furnols, 2012). The incidence of boar taint is of concern when considering the use of entire males for pork production (Babol *et al.*, 2002). Entire male pigs usually attain puberty at 6–7 months old (Andersson *et al.*, 1999). The measurement of the overall consumer acceptability of entire male pork should be conducted with unselected consumers (Bonneau and Chevillon, 2012).

Material and Methods

Two batches of cooked ham from pigs slaughtered at 5/6 months (H5/6) and 8/9 months (H8/9) respectively, were produced following the same conditions and formulations. Texture Profile Analysis, colour test (CIE Lab system), pH and moisture content were determined. A consumer test (38 tasters) was performed to evaluate characteristics such as texture, aroma, taste, appearance and global appreciation, and preferred sample.

Results

Results showed that H8/9 had significantly higher values ($p < 0.05$) in the hardness and chewiness parameters, and no differences in elasticity and cohesiveness were found (Figure 1). There were also significant differences in colour parameters, H8/9 samples being darker, reddish and less yellow (Figure 2). There were no significant differences in pH and moisture. H5/6 were the preferred samples (for 67% of consumers), however with respect to overall appreciation and texture no significant differences were detected by tasters. The consumers preferred the appearance of H8/9 ($p < 0.05$), but the aroma and flavour of H5/6 achieved better scores.

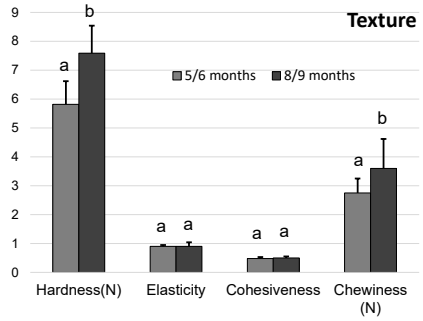


Figure 1 Texture profile analysis

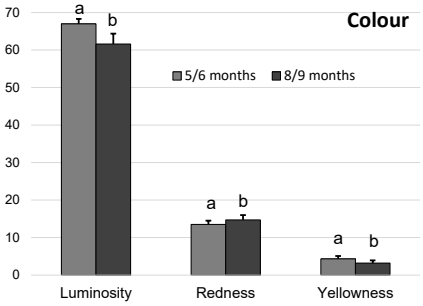


Figure 2 Colour analysis

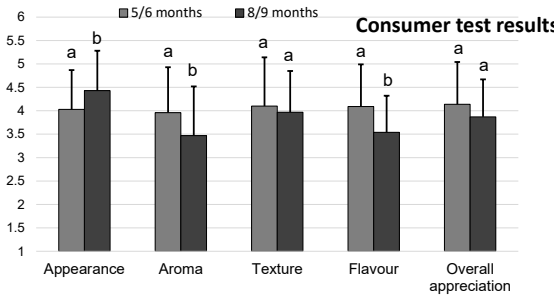


Figure 3 Consumer test results

Conclusion

Although most consumers preferred ham produced with pork slaughtered at 5/6 months, no differences in overall appreciation were found. Furthermore, the better scores in terms of appearance were achieved by H8/9. The results obtained lead to the conclusion that ham from pigs slaughtered aged 8/9 months is acceptable. Studies for improving flavour parameter of H8/9 hams are under consideration.

Acknowledgments

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Fermentation is not a strategy to mask boar taint

M D. Aaslyng*, S Støier, A Granly Koch
Danish Meat Research Institute, Gregersensvej 9, DK-2630 Taastrup,
[1]mdag@teknologisk.dk

Background

In a future scenario with slaughtering of entire males and sorting of the carcasses according to boar taint, an urgent need will arise to utilize the tainted meat in products in which the taint is masked. Fermentation has often been mentioned as a possibility (Stolzenbach *et al.*, 2009; Corral *et al.*, 2016]. During fermentation, aroma compounds are developed, specific for the actual starter culture, and different starter cultures could therefore have different masking potential. The aim of this study was to compare the masking effect of different starter cultures on boar taint, combining different bacteria, yeast and fungi.

Methods

Fermented sausages were produced using slightly tainted meat and fat of entire male pigs with an average skatole content in the neck fat of 0.28 µg/g (the meat) and 0.17 µg/g (the fat) and androstenone in the neck fat of 2.5 µg/g (the meat) and 2.2 µg/g (the fat). Four different starter cultures were tested: Texel XT-100, Bactoferm SM-194, Texel SP-362 and Texel SP Elite. The Texel SP Elite was produced with and without fungi (Texel PNT). Furthermore, one sausage of entire males and one sausage of castrates were produced simply using Glucono delta-lactone (GDL) to obtain a fast reduction in pH. Altogether, this resulted in seven different types of fermented sausages dried 20% and 30% resulting in 14 different samples. The sausages were profiled by a sensory panel, the content of skatole and androstenone was analysed before and after fermentation, and the microbial composition was analysed using traditional microbiological methods combined with 16s sequencing.

Results

No reduction was seen in the content of skatole or androstenone during fermentation. Contradictory, the concentration of the compounds was increased during drying. Independently of starter culture, boar taint was present in all the fermented sausages. No difference in flavour was seen between 20% and 30% drying loss even though the concentration of skatole and androstenone was increased. The 30% GDL sausage of entire males was the only sausage less intense in boar taint and with a more intense salami odour and flavour. The 16s sequencing did show that the GDL fermented sausages especially had *Pediococcus pentosaceaus* and *Lactobacillus sakei* and so did the sausage with Bactoferm SM-194 as starter culture without any masking effect on boar taint. It is therefore questionable if the masking occurring in the GDL sausage with 30% drying loss is due to the microbial composition.

Table 1 Skatole and androstenone contents and sensory assessment

	Drying loss	Skatole (µg/g)	Androstenone (µg/g)	Salami	Boar taint	Urine	Manure	Pungent	Sweat	Sour
Castrate-GDL				7.4	1.9	1.4	1.5	1.4	1.4	3.5
Entire male–GDL	20%	0.06±0.003	0.7±0.02	7.1	3.5	2.4	2.6	2.5	2.6	3.4
	30%	0.08±0.006	0.9±0.02							
Texel XT-100	20%	0.06±0.003	0.7±0.02	5.8	4.8	3.4	3.8	3.7	3.7	4.9
	30%	0.07±0.006	0.8±0.02							
Bactoferm SM-194	20%	0.06±0.003	0.7±0.02	5.8	4.7	3.5	3.7	3.7	3.3	5.2
	30%	0.07±0.006	0.9±0.02							
Texel SP-362	20%	0.06±0.003	0.7±0.02	5.8	4.5	3.7	3.8	3.9	3.8	5.0
	30%	0.07±0.006	0.8±0.02							
Texel SP Elite	20%	0.06±0.003	0.8±0.02	5.3	5.1	3.8	4.1	4.0	4.0	5.8
	30%	0.07±0.006	0.9±0.02							
Texel SP Elite + Texel PNT	20%	0.06±0.003	0.8±0.02	3.6	4.9	3.6	4.3	4.2	4.1	2.8
	30%	0.05±0.006	0.8±0.02							

Conclusion

Fermentation as a strategy to utilise tainted boar meat cannot be recommended on the background of the present study.

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Cooking meat for a long time at low temperatures does not decrease boar taint

M D Aaslyng*, S Støier, A Granly Koch
Danish Meat Research Institute, Gregersensvej 9, DK-2630 Taastrup,
*mdag@teknologisk.dk

Background
Slaughtering of entire males and sorting of the carcasses according to boar taint, will arise a need to utilize the tainted meat in products in which the taint is masked, or the content of skatole and androstenone is reduced. As both compounds are volatile, cooking for a long time has been suggested as a strategy to reduce the content of skatole and androstenone (Engesser *et al.*, 2017; Borrisser-Pairo *et al.*, 2017). It is therefore of interest to investigate if the concentration of skatole and androstenone is reduced during cooking for a long time at a low temperature, and the boar taint thereby becomes diminished.

Method
Pork from entire male pigs having in average 0.32 µg/g skatole and 1.34 µg/g androstenone in the neck fat was minced and mixed thoroughly. Pork patties were formed and either vacuum packed and cooked in a sous vide water bath at 58°C, 65°C or 75°C or cooked in an oven at 58°C or 75°C either vacuum packed or without packing. All patties were cooked for 6 hours. Skatole and androstenone were analysed in the raw meat and after cooking. As a control, meat from female pigs was cooked in the same way. All samples were analysed by a trained sensory panel estimating the intensity of manure, urine, pungent, sweat and boar taint relative to the female pork patty cooked similarly.

Results
The skatole concentration in the raw meat was 0.05 µg/g while it was between 0.05 and 0.07 µg/g in the cooked meat (Table 1). The concentration of androstenone in the raw meat was 0.15 µg/g while the content in the cooked meat was between 0.20 and 0.25 µg/g (Table 2). The more or less constant concentration of skatole and increased concentration of androstenone indicates that skatole, being most water soluble, has been 'washed out' in the cooking loss, while androstenone being mainly fat soluble, has stayed in the meat. A significant boar taint was present in all samples of meat from entire male pigs. A small variation existed between samples as the intensity was lowest in sous vide cooked samples at 75°C and the odour highest in sous vide cooked samples at 65°C while the flavour was highest in the vacuum-packed oven cooked samples at 75°C.

Table 1 Skatole contents in cooked meat

		Skatole Cooking temperature		
Cooking	Packaging	58 °C	65 °C	75 °C
Oven	None	0.06		0.07
Oven	Vacuum	0.06		0.06
Sous vide	Vacuum	0.05	0.06	0.05

Table 2 Androstenone contents in cooked meat

		Androstenone Cooking temperature		
Cooking	Packaging	58 °C	65 °C	75 °C
Oven	None	0.21		0.25
Oven	Vacuum	0.20		0.21
Sous vide	Vacuum	0.21	0.23	0.21

Discussion and conclusion
Even though it has been pointed out in the literature that sous vide cooking at 75°C could be a tool to diminish boar taint (Borrisser-Pairo *et al.*, 2017), we could not demonstrate any effect in this study, whatever the cooking temperature (58°C, 65°C and 75°C) and the cooking method (oven/sous vide water bath and with or without vacuum package).

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Effect of essential oil addition on masking boar taint in fresh pork sausage

B Šojić[*1], V Tomović[1], P Ikonić[2], B Pavlić[1], N Džinić[1], Z Zeković[1], T Tasić[2], N Batorek Lukač[3], I Tomašević[4], M Ivić[1], S Škaljac[1], M Jokanović[1]
[1]*Faculty of Technology Novi Sad, Novi Sad, Serbia,* [2]*Institute of Food Technology, Novi Sad, Serbia,* [3]*Agricultural Institute of Slovenia, Ljubljana, Slovenia,* [4]*Faculty of Agriculture, Belgrade, Serbia*
[*]bsojic@gmail.com

Application
Satureja montana essential oil was added in order to mask boar taint in fresh pork sausages.

Introduction
Boar taint is an off-flavour in the meat of entire male pigs, mainly related to presence of androstenone and skatole (Bañón *et al.*, 2003). Approximately 99% of consumers are sensitive to skatole (associated with naphthalene and faecal odour) (Martinez *et al.*, 2016). In order to reduce boar taint in meat and meat products, different spices and plant extracts were used (Martinez *et al.*, 2016). The aim of this study was to evaluate the effect of *Satureja montana* essential oil addition on masking boar taint in fresh pork sausages produced with two levels of skatole (0.2 ppm and 0.4 ppm).

Material and Methods
Fresh pork sausages (model system) were produced using meat and fat from castrates. Skatole (SK) and *Satureja montana* essential oil (EO) were added. The consumers (students and staff members of the Faculty of Technology Novi Sad) evaluated the samples on nine point hedonic scale from dislike very much (1) to like very much (9).

Results The results presented in Table 1 show that the control sample had significantly ($P<0.05$) higher overall liking score compared to all samples containing skatole. These results indicated that consumers can detect skatole odour and flavour at concentrations under 0.2 ppm. Relatively high overall score for sample SK2EO showed that addition of *Satureja montana* essential oil could have a positive effect on sensory acceptability of meat products with high skatole concentrations (0.4 ppm).

Table 1 Summary statistics from the consumer test data (attribute: overall liking*)

Sample	n	Mean	Median	StDev	StErr	Min	Max
Control	70	7.11[a]	7.0	1.66	0.20	2.0	9.0
SK1	70	5.86[b]	6.0	2.06	0.25	2.0	9.0
SK2	70	4.63[c]	5.0	2.63	0.31	1.0	9.0
SK1EO	70	5.43[b]	5.0	2.73	0.33	1.0	9.0
SK2EO	70	5.20[bc]	6.0	2.61	0.31	1.0	9.0

[abc] $P<0.05$. SK1-0.2 ppm skatole; SK2-0.4 ppm skatole; SK1EO-SK1+0.150 µL/g EO; SK2EO-SK2+0.150 µL/g EO

Conclusion
The data from this study showed that addition of essential oils and plant extracts in fresh pork sausages may be an alternative solution for commercializing of entire pigs.

Acknowledgments
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Raising entire males: A framework for sensory quality control

D Mörlein
University of Göttingen, Department of Animal Sciences, Germany
daniel.moerlein@uni-goettingen.de

Without doubt, there is a risk associated with the production of entire male pigs. Retailers usually rule out meat with deviant smell to prevent consumer complaints and often legislation requires sorting out of pigs with pronounced sexual odour. However, it is often questioned whether and/or how a reliable sensory evaluation can be achieved. Therefore, a framework for selection, training and monitoring of sensory assessors is presented; key aspects are highlighted.

Standardized procedures for selection of sensory assessors including reference materials are suggested given the variability of human olfactory acuity (Mörlein et al., 2013) which in turn affects the perception of boar taint (Meier-Dinkel et al., 2012; Trautmann et al., 2014). Both the capability to correctly discriminate androstenone and skatole at relevant concentration levels and the ability to correctly identify these substances need to be assured.

Once assessors are sufficiently trained, their performance should be quantified using statistical parameters such as sensitivity and specificity (Meier-Dinkel et al., 2015), area under the receiver operator characteristics (ROC) curve, and intra class correlations as measures for the assessors' agreement and repeatability. This approach has been demonstrated for objective comparison of different heating methods (Trautmann et al., 2016). A statistical simulation showed the importance of large enough sample sizes for such performance evaluations (Mörlein et al., 2015). As the performance of a single assessor usually is inferior to a group of assessors, preventive measures should be taken for slaughterhouse quality control given the risk of consumer complaints.

The quantitative relationship of key boar taint substances with the olfactory perception of deviant smell was modelled based using 1000+ boar carcasses (Mörlein et al., 2016). A nonlinearity of perception was shown as was the relatively higher importance of skatole. A curved approach is proposed instead of the so-called "safe box" when comparing sensory with chemical boar taint evaluation as it better reflects the sensory perception. The quantitative model can be used as reference / calibration for sorting schemes based on technical measurement of androstenone and or skatole.

A strictly consumer-driven approach is recommended. That is, both the assessor performance criteria and the sorting limits should be aligned with consumer acceptance data. Moreover, a recent study established the framework to assess how many consumers would be at risk to experience dislike at a given sorting threshold for androstenone and skatole (Christensen et al., 2019). By combining a model describing consumer acceptance of boar meat based on androstenone and skatole levels with a bivariate distribution model of androstenone and skatole levels in a representative set of boars, the expected risk of consumer dislike is predicted. Hence, the consequences of a given sorting scenario can be evaluated.

In conclusion, selection and training of sensory assessors should be handled with care. It is essential to objectively characterize the olfactory acuity of sensory assessors. In addition, methods for sample preparation, scales of evaluation and references should be carefully evaluated. It is furthermore critical to train the sensory memory of the assessors by presenting multiple samples that cover the whole universe of boar taint; depending on their androstenone and skatole levels, boar taint can vary substantially in terms of odour quality and quantity. A statistical framework should be applied to evaluate the assessors' performance; when doing so, confidence intervals for those parameters must not be neglected. Finally, it is of importance to calibrate any quality assurance system with consumer data. Further research is needed with regard to the long-term performance of slaughterhouse panels as boredom and adaptation may be distractors.

Acknowledgements This presentation was prepared within the framework of the Cost Action CA 15215 IPEMA.

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Challenges for pork industry: Profiling multi-dimensional consumers

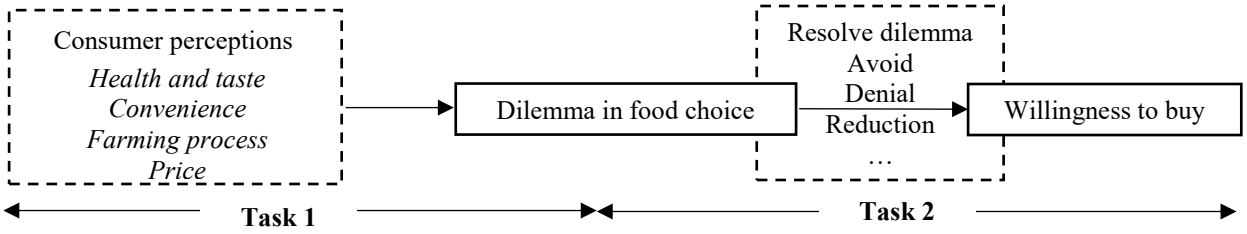
L Lin[1*], A Fischer[1], M Aluwé[2]
[1]*Social-Sciences, Marketing and Consumer Behaviour, Wageningen University, Wageningen, the Netherlands;*
[2]*Institute for Agricultural, Food and Fisheries Research, Melle, Belgium*
[*]li.lin@wur.nl

Abstract
In the pork industry, it is common to castrate male piglets in order to prevent boar taint. Consumers' concern for pig (animal) welfare emerges as the castration is predominantly done without analgesia and/or anesthesia. Today, there are three alternatives; all of which has pros and cons: castration with pain relief, production of non-castrated or entire male pigs (EM) which has the risk of boar taint, and immunocastration (IC) which involves a vaccine against GnRH to prevent boar taint. Studies on these methods focus on economic (production) and social (animal welfare) aspects. However, the success of the method finally depends on consumer acceptance. For example, the possible boar taint resulting from EM may lower pork quality, while the idea of a vaccine (IC) may be rejected by consumers.

Before applying EM or IC methods, stakeholders are eager to estimate customer acceptance/market acceptance. Questions such as, how do meat consumers experience animal welfare concerns, perceive new way of animal farming, evaluate product quality and price and make purchasing decisions, remain to be answered. This project will develop a multi-dimensional consumer profile and look for (and seek to explain) differences of profiles between social groups, and thus help to get a grip on the view of European market and consumers in accepting EM and IC practices.

Key concept: moral dilemma in food choice
Literature shows that consumers' perception of product quality involves four dimensions: taste, health, convenience, and process characteristics including natural production, animal welfare, GMO, etc. Together with product price, opposing evaluations of these dimensions can create a dilemma in food choice. For example, people who strongly embrace animal-friendly values are confronted with the higher price of the product. Those who preferred relatively traditional farms are worried about the treatment of animals in modern production. Such dilemma does not imply an outright rejection of meat consumption, but rather an unpredictable choice. Some consumers would give up the "health and taste" to exchange it with "convenience". Some are willing to pay more for a better farming process in considering better animal welfare and natural production.

Our research is particularly interested in consumers' moral dilemma in meat choice. Consumers for example show moral concern about the living conditions of animals and meanwhile prefer a lower price for the animal-friendly product. Our first task is to investigate the choice dilemma residing in meat consumption, with emphasis on the interaction between *perceptions of farming process characteristics* (animal-friendly vs. animal-unfriendly; naturalness vs. unnaturalness) and other perceptual factors, including *price, health and taste, and convenience*. To promote EM and IC methods, it is important to study how consumers resolve this dilemma. Literature shows resolutions of moral dilemmas related to animal welfare are *avoided, dissonance resolved (e.g. denial of the animal mind), or lead to behaviour changes (e.g. reduction in meat consumption)*. Our second task is thus to examine how meat consumers deal with moral dilemmas.



Citizen attitudes and consumer acceptability towards meat from boars and immunocastrates in Europe

M Aluwé[1*], L Tudoreanu[2], L Lin[3], A Fischer[3], M Font i Furnols[4]
[1]Flanders research institute for agriculture, fisheries and food (ILVO), Belgium, [2]USAMV of Bucharest, Faculty of Veterinary Medicine, Romania, [3]Marketing and Consumer Behaviour, Wageningen University, Netherlands, [4] IRTA-Food Industries, Finca Camps i Armet, Spain
[*] Marijke.Aluwe@ilvo.vlaanderen.be

Introduction

Alternatives for surgical castration are production of entire male pigs (EM) and immunocastrates (IM), or application of pain relief and/or anaesthesia (SCA). Market acceptance of alternatives is hampered in many countries for various reasons. Alternatives for surgical castration have different pros and cons for consumers. Heid and Hamm (2013) indicated that for SCA, it is mainly animal welfare aspect that plays a role in consumer willingness to pay; for IM it is food safety; and for EM, it is mainly taste and in a lesser extent also animal welfare.

Current knowledge and considerations

In general, consumer acceptance of SCA seems to be highest, followed by EM and IM and is least for surgical castration without pain relief (SC) if presented as option in the study, although differences between studies can be observed (Figure 1). Overall, the number of studies that evaluated consumer acceptance of IM is still low. Differences in consumer acceptances and preferences of alternatives can be assigned to consumer characteristics, but also to the research methodology used and the provided background information. A better understanding of which alternative is acceptable to whom will provide a better understanding of consumer attitudes. Another important issue is the lack of knowledge and familiarity of consumers with pig production and alternatives for surgical castration (Vanhonacker *et al.*, 2011). Moreover, consumers' unfamiliarity with boar taint can make it difficult for them to have a good idea about the acceptability of boar production.

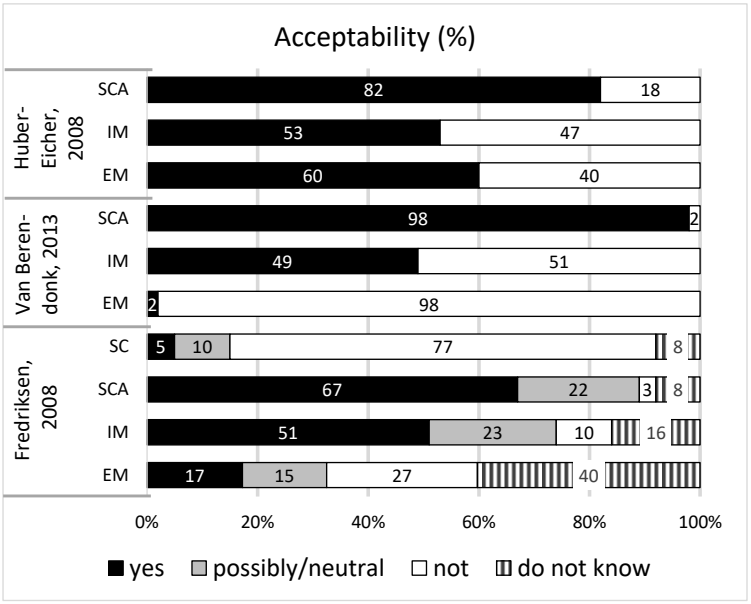


Figure 1 Acceptability of surgical castration without pain relief (SC), surgical castration with anaesthesia (SCA), immunocastration (IM) and production of boars (EM).

Conclusion

Communication and a harmonization towards production systems with the alternatives are crucial, but this needs to be tailored to the local and individual context as citizen attitudes and consumer acceptability may differ between and within countries. Combining sensory studies and attitude surveys providing audiovisual information and taking into account consumer segmentation seems to be an interesting approach for further research.

Acknowledgements

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Consumer expectations towards meat from castrated and immunocastrated pigs: a segmentation approach

A Claret[1], L Guerrero[1], M Font-i-Furnols[*1], A Dalmau[2]
[1]*IRTA-Food Industries, Finca Camps i Armet, 17121 Monells, Spain,*
[2]*IRTA-Animal welfare, Veïnat de Sies, 17121 Monells, Spain*
[*]maria.font@irta.cat

Application
Knowledge of type of consumers according to their acceptability and expectations towards immunocastrated meat to prepare optimal marketing strategies.

Introduction
Iberian pigs are raised indoor and outdoor until high body weight and all are castrated to prevent pregnancies and to avoid boar taint (Martínez-Mancipe *et al.*, 2016). An alternative to castration is immunocastration. The aim of this work is to identify segments of consumers according to their expectations and acceptability of Iberian pork from 5 sex types.

Material and methods
Loins (n=83) from entire females (EF), castrated females (CF), immunocastrated females (IF), castrated males (CM) and immunocastrated males (IM), from Iberian pigs slaughtered at 155.7 ± 8.4 kg were used. A consumer study was carried out in Barcelona (n=151) and Madrid (n=101). Consumers evaluated one sample of each sex type at three information conditions (1) liking in blind condition, (2) expectations knowing sex type and (3) liking in informed condition. Cluster analysis and mixed model with repeated measures were used for data analysis.

Results
Results allowed defining 3 clusters according to their expectations (Table 1). In blind conditions there were no differences in liking between sexual types in cluster 3 while in cluster 1 CM was preferred and in cluster 2 CM was also preferred but not significantly different from CF. In cluster 1 all the sexual types presented negative disconfirmation. In clusters 2 and 3 negative or positive disconfirmation was detected. In informed conditions assimilation effect have occurred in some sexual type's scores of clusters 1 and 3.

Table 1 Expected liking scores of loins from different sex types (CF: castrated females, EF: entire females; IF: immunocastrated females; CM: castrated males; IM: immunocastrated males) by consumers[1]

	Sexual type[2]					
Cluster	CF	EF	IF	CM	IM	RMSE
1: No preferences (n=79)	5.9	5.3	5.7	6.1	5.8	1.95
2: Against castration and immunocastration (n=79)	3.3^{d}	6.7^{a}	4.2^{bc}	3.5^{cd}	4.4^{b}	1.96
3: Against immunocastration (n= 94)	6.1^{b}	7.1^{a}	2.9^{c}	6.0^{b}	2.8^{c}	1.84

[1]Linear scale from 0 (extremely dislike) to 10 (extremely like).
[2]a,b,c Different letters mean significant ($P<0.05$) differences between sex type within cluster.

Conclusion
Segments of consumers with different expectations towards meat from castrated and immunocastrated pigs were identified. They presented negative or positive disconfirmation expectations in most of the meat types and an assimilation effect in some of them.

Acknowledgements
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Consumer's opinion on animal welfare and pig castration in Croatia

I Djurkin Kušec[1*], G Kušec[1], L Guerrero[2], M Font-i-Furnols[2], I Tomašević[3]
[1]*Faculty of Agrobiotechnical Sciences Osijek, University of Osijek, Croatia,* [2]*Institute of Agrifood Research and Technology, Spain*, [3]*Faculty of Agriculture, University of Belgrade, Serbia*
[*]idurkin@pfos.hr

Application Immunocastration is a feasible alternative to surgical castration of pigs in Croatia and abandon of surgical castration should be supported.

Introduction Surgical castration without anaesthesia is still a routine practice in Croatia. However, due to the growing concern on animal welfare this practice is being seriously questioned in most European countries. Subsequently, a declaration of intention to abandon it has been signed by many different stakeholders of the pork chain (farmers' representatives, meat industry, retailers, scientists, veterinarians and animal welfare NGOs) (EU Declaration, 2010). A number of alternatives to surgical castration have been proposed (von Borell *et al.*, 2009), immunocastration and entire male production being the feasible ones at present (Cimerman, 2016). However, besides its advantage on animal welfare, the consumer's opinion should also be considered.
The aim of the present study was to understand Croatian consumer beliefs and attitudes towards pig castration and animal welfare, as well as to identify their willingness to pay more for meat of animals that were not surgically castrated and that were treated according to animal welfare legislation.

Material and methods A total of 301 consumers participated in the survey through self-administered questionnaires during 2017 in Croatia. The responders were selected by gender and age to follow the National distribution and no previous information was given to them about production practices and meat quality. Analysis of variance (ANOVA), principal component analysis (PCA) and linear regression analysis were computed using XL STAT (2017). Behavioural intention was measured using Cronbach's Alpha Coefficient (Cronbach, 1951). Analysis of variance included gender, place of origin, age, education and economic situation as fixed effects.

Results Using the answers from the questionnaire, Croatian consumers were divided into three clusters regarding their attitudes and believes towards pig castration. The consumers from the first cluster (N=88) believe that surgical castration is unnecessary procedure that produces pain to the animal. They believe that castrated pigs produce leaner meat, but the quality of their meat is no better than the meat from uncastrated pigs. They accept pig castration using vaccines. The consumers from the second cluster (N=87) believe that castration is necessary and easily performed, and that immunocastration improves pork quality. The consumers from the third cluster (N=126) believe that castration is necessary, easily performed and natural. They prefer to eat meat from castrated animals, however they do not believe that immunocastration improves meat quality. Using the same methodological approach, we identified three clusters of consumers according to their beliefs and attitudes towards animal welfare. The consumers from the first cluster (N=101) believe that animal welfare is very important and are concerned with the transport of animals and slaughter systems. The consumers in the second cluster (N=88) agree that animal welfare is very important and that animals for human consumption should be treated with dignity. The consumers from the third cluster (N=112) believe that animals raised according to animal welfare legislation produce meat of higher quality. Unlike consumers in other two clusters, these consumers do not believe that meat animals should grow in freedom.
In sociodemographic data, significant differences ($P < 0.05$) between clusters were found in the frequency of pork consumption and the level of finished studies. No significant differences between clusters in other investigated sociodemographic data (professional activity, economic situation, the classification of places of residence and place of origin) were observed.

Conclusion Croatian consumers are concerned about animal welfare and strongly believe that animals raised for human consumption should be treated with dignity. Generally, Croatian consumers prefer the meat from castrated pigs; however, they accept immunocastration as an alternative to surgical castration.

Acknowledgements
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Consumer's attitudes towards surgical castration of pigs in three Western Balkan countries

I Tomašević[1*], S Novakovič[1], I Djekič[1], D Nakov[2], L Guerrero[3], M Font-I-Furnols[3]
[1]*Faculty of Agriculture, University of Belgrade, Serbia,* [2]*Faculty of Agricultural Sciences and Food*, *Republic of Macedonia,* [3]*Institute for Food and Agricultural Research and Technology (IRTA), Spain*
[*]tbigor@agrif.bg.ac.rs

Application
This study investigates the acceptance of surgical castration of boars among consumers from three Western Balkan countries and if there is a willingness to find its modifications.

Introduction
Surgical castration of male piglets without anaesthesia is performed routinely by farmers or veterinarians in Western Balkan countries to eliminate the risk of boar taint in pork meat (Tomašević *et al., 2017*). The aim of the study was to investigate consumers' attitudes towards surgical castration of piglets in Serbia, Bosnia and Herzegovina (B&H) and Macedonia.

Material and methods
A representative consumer survey was carried out in these three countries in 2017. Over twelve hundred (1,287) questionnaires were answered by pork eaters. Likert scale data were considered as ordinal values and non-parametric statistical tests have been used since data were not normally distributed. Mann-Whitney U test has been performed to compare the statements between genders and age and Kruskal-Wallis H test between countries (p=0.05).

Results
Western Balkan consumers agree (5.2 ± 1.2) that surgical castration produces pain to the animal. They are significantly more likely to agree in Macedonia that meat from castrated pigs is of better quality (4.8±1.5) than in Serbia (4.6±1.3) and B&H (4.5±1.1). Macedonians agree the least (3.9±1.3) followed by more ambiguous consumers from Serbia (4.4 ± 1.1) and B&H (4.5±1.0) that meat from castrated pigs is more expensive. Macedonians are also significantly more likely to disagree (3.6 ± 1.6) that castration is not necessary, compared to Serbian (4.7±1.1) and B&H (4.9±0.9) consumers. On average, Western Balkan consumers, slightly disagree that meat from castrated pigs is leaner (3.7±1.1) and they neither agree nor disagree that they prefer to eat meat from castrated pigs (4.0±1.3) or that pig castration with vaccines improves pork quality (4.0±1.5). For consumers from Switzerland the term "immunocastration" lead to a negative connotation and was a bit scary to them (Huber-Eicher and Spring, 2008). On the other hand, about 70% of the total sample of consumers from Belgium, France, Germany and the Netherlands expressed a preference for the vaccine method (Vanhonacker and Verbeke, 2011).

Table 1 Attitudes among consumers from three Western Balkan countries about surgical castration of boars

	Disagree very strongly (1); Agree (4); Agree very strongly (7)		
Attitudes	Serbia	B&H	Macedonia
Meat from castrated pigs is of better quality	4.6 ± 1.3	4.5 ± 1.1	4.8 ± 1.5
Meat from castrated pigs is more expensive	4.4 ± 1.1	4.5 ± 1.0	3.9 ± 1.3
Castration is not necessary	4.7 ± 1.1	4.9 ± 0.9	3.6 ± 1.6
Surgical castration produces pain to the animal	5.2 ± 1.2	5.2 ± 1.2	5.2 ± 1.2
Pig castration with vaccines improves pork quality	4.0 ± 1.5	4.0 ± 1.5	4.0 ± 1.5

B&H: Bosnia and Herzegovina

Conclusion
There are differences in consumer's attitudes towards surgical castration by country of origin, but further work is needed to find segments of consumers according to their attitudes.

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Economical assessment of the alternatives for surgical castration: a comparison of the gross margin per pig

F Leen*, A Van den Broeke, M Aluwé, J Van Meensel, S Millet
ILVO (Institute for Agricultural, Fisheries and Food Research), Scheldeweg 68, 9090 Melle, Belgium.
*frederik.leen@ilvo.vlaanderen.be

Application

The effect of sex on technical performance provokes differences in gross margin, resulting in profit or loss for the farmer, depending on his castration decision.

Introduction

In recent years, the pig industry has been facing very difficult economic conditions. Contemporary intensive pig production is characterized by small margins between costs and revenues. As a result, small adjustments or anomalies in management, even with slight changes in revenues and costs, can significantly affect the economic performance of the pig farm. Among many aspects of managing the fattening farm or the farrow-to-finish farm, the decision to raise entire males (EM), immunocastrates (IC) or barrows (BA), can make an important difference in the technical and economic performance of the farm.

Material and Methods

In three trials, the effect of sex on the gross margin per pig was determined. Growth and feed intake curves were estimated using the Bridges model for growth (Bridges *et al.*, 1986) and the Giesen model for daily feed intake (Giesen *et al.*, 1988). With these curves, the evolutions in revenues and costs per pig were simulated. The gross margin per pig was calculated as the difference between the revenues on the one hand and the costs for feed and piglets, manure disposal, immunocastration and other variable costs.

Results

In all three trials, EM and IC were the better economic alternative compared to BA, mostly due to the lower feed conversion ratio and the improved conformation compared to BA (Figure 1). The feed conversion ratio of IC approached those of EM. In addition, in some cases IC received a higher price per pig due to a better conformation compared to EM. However, the extra costs for immunological castration per pig resulted in lower gross margins per pig for IC compared to EM. Still, these results must be considered in a context where an agreement has been concluded between the farmer and the slaughterhouse about delivering EM or IC. International market acceptance of EM and IC currently remains an obstacle for the entire pig chain to accept IC and EM as a solid alternative for BA.

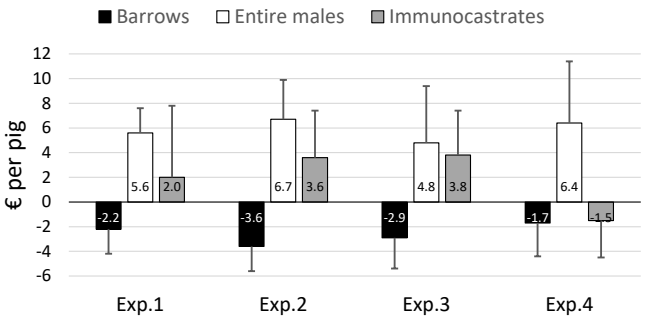


Figure 1 Gross margin per sex

Conclusion

The better technical performances of EM and IC translate into higher gross margins per pig compared to BA. Although these results sound promising, a full international market acceptance of these alternatives is needed to eliminate the risk of declining revenues.

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Economic effects of alternatives to piglet castration without anaesthesia on competitiveness in Germany

M Verhaagh
Thünen Institute of Farm Economics, Brunswick, Germany
mandes.verhaagh@thuenen.de

Application

The purpose of this study is to analyse the economic effects of alternatives to piglet castration without anaesthesia and to provide policy and practice advice.

Introduction

The ban of executing piglet castration without anaesthesia is an adaptation of pig production to an accept form of livestock farming in Europe. In addition to animal welfare aspects, an economic analysis is necessary in order to ensure a successful implementation on the farms and for understanding effects on competitiveness between countries.

Material and Methods

The farms represent typical regional farms and are surveyed using the standard operating procedure developed by agri benchmark to define typical farms. All scenarios for the alternatives of piglet castration without anaesthesia are quantified and based on scientific literature. The following processes are considered: Finishing of boars, vaccination against boar taint (here called immunocastration; with two and three vaccinations) as well as surgical castration with the use of Isoflurane and injection anaesthesia. Changes in the structure of billing systems of entire males in Germany and new knowledge about the immunocastration are making a renewal of economic analysis necessary. (Norda *et al*., 2017, Verhaagh and Deblitz, 2017).

Results

Composition and meaning of the enterprise names: DE_350_0 Sow farm with 350 breeding sows and DE_0_6000 Fattening farm with 6000 fattening pigs sold in Germany. Table 1 shows changes in profitability for investigated scenarios on typical farms in reference to the individual farm levels in 2016.

Table 1 Changes in profitability through alternative processes on typical farms

	Sow enterprises (€ kg/ LW)		Finishing enterprises (€ kg/ CW)			
Scenarios	DE_350_0	DE_800_0	DE_0_3600	DE_0_5000	DE_0_6000	DE_0_6300
Boars	0,42	0,15	2,58	1,85	2,62	0,26
Immuno (2)	0,42	0,15	0,78	0,47	1,29	-1,12
Immuno (3)	0,42	0,15	-0,20	-0,52	0,32	-2,11
Isoflurane	-12,21	-10,71	-	-	-	-
Injection	-10,86	-10,77	-	-	-	-

Conclusion

Boar finishing and the vaccination against boar taint are profitable strategies, despite the adjustments to the billing system (about three cents less per kg carcass weight). Whereby for these alternatives of production a problem can arise in the selling for the farmer. The cash cost for the use of anaesthesia rising the total costs of piglet production in Germany. While Denmark and the Netherlands have already made it possible for farmers to use the anaesthetic, German piglet producers are losing their competitiveness.

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The attitude of pig farmers from Vojvodina (Northern Serbia) regarding the alternative strategies for surgical castration of piglets

P Ikonić*[1], T Peulić[1], B Šojić[2], I Tomašević[3]
[1]*Institute of Food Technology, Novi Sad, Serbia,* [2]*Faculty of Technology Novi Sad, Novi Sad, Serbia,* [3]*Faculty of Agriculture, Belgrade, Serbia*
*predrag.ikonic@fins.uns.ac.rs

Application
The findings of this preliminary study allow having an overview on the opinion and willingness of Serbian pig farmers to use alternatives for surgical castration.

Introduction
In order to prevent boar taint, unpleasant off-flavor induced by accumulation of androstenone and skatole in meat, male piglets are routinely surgically castrated without pain relief in most of the EU countries and Serbia, as well. This practice is unacceptable from the standpoint of animal welfare. Thus, main stakeholders of the pork chain in EU voluntarily decided to stop surgical castration of piglets and implement more human alternatives (Tuyttens *et al.,* 2012; Candek-Potokar *et al.* 2015). The objective of this study was to evaluate the Serbian farmers' attitude towards immunocastration (IC) and production of entire males (EM) as alternatives for surgical castration of male piglets without anaesthesia (SC).

Material and Methods
Survey was conducted among the group of 21 pig producers having on average 390 sows and 4 400 fattening pigs per year. All the farms were located in Province of Vojvodina (Northern Serbia). The questionnaire created within EU PIGCAS project (Project No. 043969) was used with small modifications to investigate the opinion of farmers.

Results
About 80% of the farmers considered themselves to be uninformed or poorly informed about IC, while nearly 60% of them are well acquainted with the production of EM (data not shown). Producers' general preference was recorded as SC>IC>EM. Pronounced difference in the general ranking of practices was mostly associated with aspects of animal welfare and elimination of boar taint ($\chi^2 = 16.67$ and $\chi^2 = 20.67$, respectively) (Table 1). SC was ranked the most suitable practice in terms of profitability and consumer acceptance, while EM was perceived as the most appropriate alternative regarding the labor conditions and performance of the animals.

Table 1 Farmers' ranking of alternative strategies regarding different aspects of pig production from most appropriate (1) to least appropriate (3) (n=12)

	General	Labour	Animal welfare	Effectiveness against boar taint	Performance	Profitability	Consumer acceptance
SC	1.33	2.17	2.83	1.17	2.17	1.50	1.42
IC	2.17	2.17	2.00	1.83	2.25	2.17	2.17
EM	2.50	1.67	1.17	3.00	1.58	2.33	2.42
χ^2	8.67	2.00	16.67	20.67	3.17	4.67	6.50
p	<0.05	0.37	<0.05	<0.00	0.21	0.10	<0.05

Conclusion
The farmers' attitude toward traditionally performed practice SN is very positive. Introduction of alternative strategies will be rather difficult without legal requirements and strong financial support.

Acknowledgments
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Pork production with entire males and immunocastrates in Australia

D N D'Souza[1*], R J E Hewitt[1], R J van Barneveld[1]
[1]*SunPork Group, Murarrie, Queensland, Australia*
[*]darryl.dsouza@sunporkfarms.com.au

Introduction

Australian pork producers ceased castration in the late 1970's due to very 'light' slaughter live weights (~85kg), and the growth performance inefficiencies associated with surgically castrated animals. Almost all of the pork produced was consumed domestically. Since then, in the mid-1990s, Australia started to export chilled carcases to the Singaporean market. As the requirement in Singapore was only for female pigs or surgical castrates, this placed a relative increase in the number of males destined for the Australian fresh pork market. This brought with it a greater risk of Australian consumers being exposed to boar tainted pork, particularly since we have increased slaughter weights to ~105kg HSCW.

Detail

Meeting the consumer's need for lean and consistently high pork quality is a challenge for many pork supply chains and it is imperative to the future success of pork industries in an ever changing, highly competitive food market. Some key points in relation to entire male pig production systems and the issues that need to be addressed are:

- Entire male pigs have higher animal welfare issues as a consequence of socio-sexual behaviours and these also have a negative impact on carcase quality;
- Incidence of boar taint in entire male pigs is >30%, and reflects levels observed in entire male pig populations globally;
- There is a poor correlation between boar taint risk (based on consumer threshold values for androstenone and skatole) and carcase weight;
- Entire male pork has lower quality consistency, higher consumer fail rates and lower consumer re-purchase intention scores.

Immunocastration is a key technology that enables Australian pork producers to continue to grow entire male pigs in the most efficient manner, eliminates boar taint and negative behaviours associated with entire male pigs, and improves quality and consistency of pork products. Immunocastration allows pork producers flexibility in achieving a high-quality pork product for consumers for domestic and export markets, whilst also allowing supply chains to tailor the end carcase quality to reflect specific market requirements e.g. back fat thickness.

Australian retailers did not seem particularly keen to firstly acknowledge, and then eliminate, boar taint. However, much has changed in the last 10 years, with all retailers allowing the use of the boar taint vaccine (to eliminate boar taint and improve welfare). Adopting an eating quality focus, immunocastration has allowed individual supply chains to develop their eating quality assurance pathways to differentiate their brands in the market place.

The percentage of Australian pork exported is still relatively small by global standards. Nevertheless, Australian pork still commands a premium in markets like Singapore and Hong Kong. For the most part pork and pork products from immunocastrates exported to high-end markets like Hong Kong, Singapore and Japan are now commonplace and accepted. There are however exceptions, such as the chilled pork carcase market in Singapore that still stipulates a requirement for female pig carcases. Frozen pork or chilled pork cuts exported to Singapore can be from immunocastrates. It appears that the reasoning for this relates to the visibility of the testicle cut lines on immunocastrate carcases sold in the wet markets, and consumer perception being unable to differentiate entire male pigs from immunocastrates.

RSPCA Australia, a community-based charity that works to prevent cruelty to animals by actively promoting their care and protection, permits immunocastration as part of its Approved Farming Scheme Standards. Incidentally, physical castration is not permitted by the RSPCA. In 2011, Australian Pork Limited, the pig producer owned organisation supporting and promoting the Australian pork industry, established its position on immunocastration that supported the use of Improvac® in Australia.

Conclusions

Improvac® is a key technology that enables pork producers to continue to grow entire male pigs in the most efficient manner, eliminates boar taint and negative behaviours associate with entire male pigs, and improves quality and consistency of pork products. Market acceptance of immunocastration in domestic and export markets has been slow, but this has turned around in the last 3-5 years with increased retailer focus on animal welfare and eating quality. Importantly, this increased acceptance and use of immunocastration has not led to any consumer backlash.

Whole chain approach for moving from surgical castrate to entire male production

G B C. Backus
Connecting Agri and Food, Oostwijk 5, 5400 AM Uden, the Netherlands
g.backus@connectingagriandfood.nl

Application

The results of the study indicate that different segments of the pork supply chain provide directions for solutions to ending castration. Our findings also show that the cost-effectiveness of preventive measures to reduce boar taint prevalence varies greatly. These solutions affect the market acceptance of meat from entire male pigs.

Introduction

About 100 million male pigs are annually castrated in Europe. The interference in the animal's integrity is considered not welfare friendly, and is associated with the risk of infection. For the pig farmer it is an annoying and labour-intensive task. Castrating piglets has a negative influence on the utilization of feed, and producing boars saves costs and agricultural land. A supply chain approach for moving from surgical castration to entire male production requires commitment from retailers, being the gatekeeper to the consumer. The issue of ending castration of piglets is further complicated by the uneven distribution of benefits and risks across chain segments. This study presents a unified analysis of measures to reduce boar taint prevalence and detection methods for boar taint.

Material and methods

Effectiveness of measures to reduce boar taint was determined based on literature review and using data collected in observational and experimental studies. Benefits and costs associated with raising boars are calculated for Dutch conditions. These benefits and costs are not equally distributed across chain segments.

Results

Boars have a better feed conversion ratio and carcass quality, compared to barrows, generating a calculated net return of €7.71 per pig. At slaughterhouse level, plants are confronted with costs of testing for tainted carcasses and a price reduction for identified tainted carcasses. Changes in breeding programs become attractive only when they assure considerable decreases in tainted carcasses, otherwise the advantage due to reduction of tainted carcasses is outweighed by the decline in genetic progress in other important traits. High heritabilities are reported for Androstenone (0.54) and for Skatole (0.41). Changes in feeding and housing conditions may reduce Skatole content. Among effective feed measures reported are adding inulin (14 days before slaughter), beet pulp (14 days before slaughter) and grain mixtures (4 days before slaughter), with reductions in Skatole content ranging from 29% to 80%.
Lower farm level boar taint prevalence is associated with smaller group sizes (less than 30 pigs per pen), cleaner pens and pigs, wider gaps of the slats and a good quality of the floor.
At slaughter plant level, companies are confronted with costs of testing for tainted carcasses and price reductions for tainted carcasses. In the Netherlands, the cost of boar taint tests based on human nose scoring (HNS) is €1-2 per carcass. Valuable parts of tainted carcasses are not sold in fresh meat markets. Tainted carcasses are used for processed cold meat products. Given that an average price difference between meat used for sale in the fresh meat market and for production of processed meat products is €0.28 per kilogram, the return on tainted carcasses is ca. €25 lower than on not tainted ones. Implementing boar taint detection systems requires weighing false positives against false negatives. Falsely accepted carcasses hamper market acceptance, falsely rejected carcass reduce the value of marketed products. Kendall's W between consumer perception of odour and HNS was equal to 0.63. The human nose scoring system predicts boar taint quite accurately, to a level at which major pork supply chains accept it as a sufficient quality control measure.

Conclusion

Combinations of preventive measures to reduce boar taint that include breeding and appropriate feeding and housing conditions result in cost-effective solutions. Breeding requires a focus on the long-term perspective, while use of feed measures is an available short-term solution, however still costly. The uneven distribution of benefits limits the use of cost-effective measures. In a non-integrated chain, preventive and costly measures that reduce boar taint will only be implemented when incentives are imposed to induce farmers to prevent developing boar taint. The optimal combination of preventive measures and investments in boar taint testing can vary greatly with the boar taint threshold level.