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Influence of soiling on boar taint in boars

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ABSTRACT

It has been suggested that skatole, one of the main compounds responsible for boar taint, can be lowered by keeping pigs clean, as skatole can be absorbed through skin and/or lungs (Hansen, Larsen, Jensen, HansenMoller & Bartongade, 1994). With this experiment, we further investigated this hypothesis by comparing extremely clean with extremely dirty animals with regard to the occurrence of boar taint. One group of boars was washed daily and pens were mucked on and littered down daily (CLEAN), a second group of boars was rubbed with faeces daily (DIRTY) and a third group of boars was kept in control conditions (CONTROL). The treatment was performed during the last four weeks before slaughter.

According to the standardised consumer panel evaluations, boars subjected to extra soiling had a higher concentration of boar taint than boars that were kept extra clean. In contrast, expert panels judged general meat flavour to be inferior in CLEAN than CONTROL pigs. The home consumer panel, the hot iron method, and laboratory analyses, i.e., the presence of indole, skatole and androstenone in fat and serum, all showed no significant differences. So no clear indications towards skatole reduction by improving cleanliness of pigs were found.

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1. Introduction

In spite of heavy social pressure to ban surgical castration without anaesthesia, this procedure is still a commonly performed measure to prevent boar taint, an off-odour released by the heating of meat and fat of some boars. The main contributing compounds to this unpleasant odour are skatole (faecal-like odour) and androstenone (urine-like odour) (Claus, Weiler & Herzog, 1994; Rius & Garcia-Regueiro, 2001). While the production of entire boars would be more ethical, this would only be feasible with a low prevalence of boar taint and a rapid, inexpensive, and reliable detection technique appropriate for use at the slaughterhouse.

Hansen et al. (1994) suggested that skatole can be lowered by keeping pigs clean. They observed that pigs kept at high stocking density and in dirty pens had higher skatole concentrations than pigs that were kept at low stocking density and in clean pens. They hypothesised that skatole may be absorbed through the skin and/or the lungs and eventually accumulates in fat. Studies with radioactive skatole confirmed the absorption through the skin, with a higher absorption through the skin of the belly (40%) than through the skin of the back (6%). Absorption through the lungs in rabbits was found at

high temperatures. Effects were found in a summer as well as in a winter experiment, but skatole concentrations were clearly higher during summer (Hansen, 1998).

As literature about this topic is limited, we further investigated this hypothesis by comparing the occurrence of boar taint in extremely clean and dirty boars. To simulate the best- and worst-case scenario in our study, pigs were either washed daily or rubbed daily with their faeces.

2. Materials and methods

2.1. Animals and management

In each of three consecutive rounds with three weeks difference, twenty-one boars of three weeks old (Piétrain \times Rattlerow Seghers crossbred sow) were randomly allocated to three treatment groups. The effect of genetic background was minimised by allocating littermates to different treatment groups.

In the control group (CONTROL), pens were mucked out every weekday. In the dirty group (DIRTY), pigs were rubbed with their own manure once a day, and pens were only mucked out when necessary. In the clean group (CLEAN), pigs were washed daily, and pens were mucked out and littered down daily. These treatments were performed each morning. The boars were treated from week 22 to week 26 (slaughter). Before week 22, all boars were kept under the same standard, control conditions.





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All groups were kept in pens $(2.9 \text{ m} \times 2.6 \text{ m})$ with concrete floors and stocking rate ranging between 1.1 and 1.9 m²/pig (max. 7 pigs/pen). Up to the weight of 50 kg, ill or dead piglets were replaced (three boars were replaced because of lameness, one because of an infected penis). Ill or dead boars belonging to CONTROL were removed but not replaced, in order to not interfere with ongoing behavioural studies. Two-phase feed was given ad libitum: feed 1 from 20 to 50 kg and feed 2 from 50 kg to slaughter. The pigs had unlimited access to water at all times. Blood samples of the boars were collected to study the evolution of boar taint compounds during the experimental period. Samples were taken 5 times: at the start of the experiment (week 22), and then each week until one day before slaughter. Blood was sampled by venopuncture of the vena jugularis. Serum was separated by centrifugation at 3000 rpm and stored at -80 °C until analysis.

Pigs were fasted for 24 h before slaughtering. After 1 h of transport and about 3 h of lairage at the slaughterhouse, the pigs were slaughtered by exsanguination after electric stunning. Boars of the first, second and third round were slaughtered on 26/01/2008, 18/03/2008 and 8/04/2008, respectively.

All procedures were approved by the ethics committee.

Longissimus thoracis et lumborum samples (Kauffman, Smulders, Hartman, Habel & Bergstrom, 1990) with backfat layer (30 cm around the 13th rib) were taken at the slaughterhouse 24 h after slaughter. The samples were trimmed of visible fat and cut into slices of 2.5 cm, and backfat was cut into pieces. Each individual piece was vacuumpacked and stored at -20 °C until tests with consumer and expert panels were performed. Samples were thawed by keeping the samples at 4 °C overnight. For laboratory analyses of boar taint compounds, the fat samples were vacuum-packed and stored at -80 °C until analysis.

3. Measurements

3.1. Soiling

The effectiveness of the treatments was evaluated twice weekly by scoring cleanliness, before the animals were treated for that day. The pig surface was divided into 9 parts (head/neck, back, rear, ears (L, R), shoulder/front leg (L, R), and sides (L, R). All were scored on a scale from 0 (0% soilage) to 5 (80–100% soilage). Mean soilage score per pig was further used for statistical analysis. Ambient temperature was

recorded with a data logger (For Escort iLog temperature/humidity data loggers, Escort), from week 9 to week 26. The average ambient temperature was 15.3 ± 1.1 °C.

3.2. Boar taint detection

Boar taint was detected using the different methodologies described by Aluwé et al. (2009): a trained expert panel to evaluate the sensory quality of meat and fat samples, and laboratory analysis of the main boar taint compounds in fat samples (skatole, androstenone and indole), the hot iron method (a fast sensory assessment at the slaughterhouse consisting of heating neck fat with a hot iron), a standardised consumer panel to evaluate the sensory quality. The main characteristics of these different detections methods are given in Table 1. In addition to these methods, a home consumer panel was performed and the evolution of serum boar taint compounds during treatment was also investigated.

Cut-off values for the hot iron method, expert and consumer panels were taken at the corresponding value of a neutral or acceptable evaluation of the sample. Cut-off concentrations for indole, skatole and androstenone in fat were set at 0.10 (Moss, Hawe & Walker, 1993), 0.20 and 0.50 ppm (Babol & Squires, 1995), respectively. This allowed the calculation of the percentages of off-odour and off-taste animals.

3.2.1. Home panel

One hundred sixteen families were recruited from the staff of the ILVO institute and Ghent University to participate. Each family (the cook and a taster) was given three samples, randomly selected from each treatment group. The families were not informed that they were evaluating meat from boars. No instructions were given towards the preparation of the samples, but attention was given to the fact that each sample should be prepared separately and cooks were asked to describe the amount of butter/oil, salt, pepper, other spices, sauce used and other remarks. Samples were scored by the cook and a taster for taste, flavour, odour and tenderness on a scale from 1 (very good) to 7 (very bad). Additionally, the cook was asked to score the aroma during cooking on the same scale.

3.2.2. Laboratory analysis of serum

For extraction and clean-up, 2 mL of diethylether was added to a 1 mL serum sample, together with 200 ng of the internal standards

Table 1

Boar taint detection overview.

Method	Hot iron	Standardised consumer panel	Home consumer panel	Expert panel	Laboratory analyses
Sample	Fat	Meat	Meat	Fat Meat	Fat
Methodology	Heating neck fat with a hot iron (30 W)	Grill 1800 W, 3 min	No restrictions were given	Fat: Microwave Meat: Grill, 1800 W, 3 min	LC-MS (Verheyden et al., 2007).
Parameters	Odour	General Odour Flavour Tenderness	Odour Flavour	General Androstenone Skatole	SKA ^a AND IND
Scale/unit Cut-off	Neutral: 1 Bad: 4 >1.5	Good: 1 Bad: 5/6 >3	Good: 1 Bad: 7 >3	Neutral: 1 Bad: 7 ≥3	ppm SKA>0.20 ppm AND>0.50 ppm IND>0.10 ppm
Number of assessments	At least 1 out of same three androstenone-sensitive and trained persons	6 consumers/sample	6 cooks/sample 6 tasters/sample	6 experts/sample	1/sample
Where	Slaughterhouse	Cafetaria hospital	At home	ILVO	Lab. Of Chemical Analysis

^a SKA = skatole, AND = androstenone, and IND = indole.

2-methylindole and 5 α -androstan-3-one (10 ng/µL). Then the samples were rigorously vortexed during 1 min and centrifuged at 13,500×g for 20 min at 4 °C to obtain phase separation. Finally, the extract was dried under a gentle stream of nitrogen at 30 °C and redissolved in 300 µL ACN/H2O (30/70) prior to analysis.

LC-MSⁿ analysis: the HPLC system consisted of a Finnigan Surveyor MS Pump Plus and a Finnigan Surveyor Autosampler Plus (Thermo Electron, San José, CA, USA). Chromatographic separation was achieved using reversed phase chromatography with gradient elution. Separation was performed using a Symmetry C18 column (5 µm, 150 mm×2.1 mm, Waters, Milford, MA, USA). The mobile phase consisted of a mixture of MeOH (A) and 1% acetic acid (B) and was pumped at a flow rate of 0.3 mL/min. A linear gradient was used starting with a mixture of 40% A. The MeOH percentage was increased from 40 to 100% in 7 min. Between each sample, the column was allowed to equilibrate at initial conditions (10 min). Analysis was carried out using a LTO linear ion trap mass analyser (Thermo Electron) equipped with an atmospheric pressure chemical ionisation (APCI) interface. Data acquisition was carried out by Xcalibur 2.0 software (Thermo Finnigan, Austin, TX, USA). Optimal ionisation source working parameters were: vaporiser temperature, 400 °C; sheath gas, 40 arbitrary units (a.u.); auxiliary gas, 5 a.u.; capillary temperature, 275 °C; capillary voltage, 2 V; and tube lens voltage, 25 V. Data acquisition was performed in full scan mode and in product ion scan mode, using as precursor ion the protonated molecular ions in accordance with Verheyden et al. (2007). When applied, the normalised collision energy was between 30 and 75%, the isolation width (IW, m/z) was 2.0, the activation time was 30 ms and the activation Q was between 0.25 and 0.50.

4. Statistical analysis

Boar taint detection variables were transformed by Box–Cox transformation to ensure a normal distribution (Neter, Kutner, Nachtsheim & Wasserman, 1996). Difference in boar taint according to the different detection methods was evaluated, based on the scores (per animal, the average scores for the expert panel and for the consumer panel) and concentrations of boar taint compounds in fat with ANOVA, with treatment, replicate and treatment × replicate as fixed factor, and the animal as the experimental unit. The effect of treatment on serum concentrations of boar taint compounds was evaluated in time by repeated measurements (Statistica 8.0, Statsoft, Tulsa, USA). Tukey's post hoc test was used to compare pair-wise differences between treatments. Pearson correlations between soilage scores and boar taint detection parameters and correlations in between the different detection parameters were also checked (significance level of 0.05).

Table 2

Average scores or levels \pm st. dev. for the different boar taint detection parameters and proportion of cases above cut-off value.

	CONTROL		CLEAN		DIRTY		p-value
	Score	%У	Score	%	Score	%	
n	18		21		20		
Slaughterhouse							
Hot iron (%>1.5) ^x	1.3 ± 0.5	17	1.5 ± 0.7	29	1.3 ± 0.6	15	0.555
Experts							
Fat odour							
General (%≥3)	1.8 ± 0.6	0	1.8 ± 0.5	6	1.9 ± 0.5	5	0.848
Androstenone ($\% \ge 3$)	1.5 ± 0.4	0	1.5 ± 0.4	0	1.6 ± 0.4	0	0.580
Skatole ($\% \ge 3$)	1.3 ± 0.4	0	1.4 ± 0.2	0	1.4 ± 0.4	0	0.864
Meat odour							
General (%≥3)	1.4 ± 0.2	0	1.7 ± 0.9	10	1.5 ± 0.3	0	0.191
Androstenone ($\% \ge 3$)	1.2 ± 0.2	0	1.4 ± 0.5	5	$1.3 \pm .3$	0	0.182
Skatole ($\% \ge 3$)	1.1 ± 0.1	0	1.3 ± 0.4	0	$1.2 \pm .2$	0	0.407
Meat flavour							
General (%≥3)	1.3 ± 0.4^{b}	0	1.8 ± 0.8^{a}	10	$1.6\pm0.4^{\mathrm{ab}}$	5	0.027
Androstenone ($\% \ge 3$)	1.2 ± 0.2	0	1.3 ± 0.4	0	1.3 ± 0.3	0	0.134
Skatole ($\% \ge 3$)	1.1 ± 0.3	0	1.4 ± 0.5	0	1.2 ± 0.3	0	0.094
Consumers							
Standardised panel							
General (%>3)	$2.8\pm0.4^{\mathrm{ab}}$	28	$2.6\pm0.4^{ m b}$	19	2.9 ± 0.4^a	30	0.031
Odour (%>3)	2.8 ± 0.3	28	2.8 ± 0.4	24	2.9 ± 0.5	35	0.420
Flavour (%>3)	2.8 ± 0.4^{ab}	33	$2.6\pm0.5^{\rm b}$	14	3.0 ± 0.4^{a}	30	0.017
Home panel							
Cook: cooking odour	3.3 ± 0.4	0	3.4 ± 0.4	0	3.4 ± 0.4	5	0.828
Cook: odour	3.3 ± 0.4	0	3.2 ± 0.3	0	3.2 ± 0.4	0	0.754
Cook: flavour	3.1 ± 0.6	6	3.0 ± 0.5	0	3.0 ± 0.5	0	0.906
Taster: odour	3.2 ± 0.4	0	3.3 ± 0.3	0	3.4 ± 0.4	0	0.561
Taster: flavour	3.0 ± 0.4	0	3.1 ± 0.5	0	3.1 ± 0.5	0	0.995
Lab analyses							
Fat							
Indole, ppm (>0.10)	0.03 ± 0.02	0	0.09 ± 0.19	15	0.04 ± 0.05	5	0.911
Skatole, ppm (>0.20)	0.05 ± 0.06	7	0.05 ± 0.07	5	0.11 ± 0.19	15	0.220
Androstenone, ppm (>0.50/>1.00)	0.53 ± 0.31	40/13	1.46 ± 2.93	45/30	0.56 ± 0.71	33/11	0.437
Serum							
Indole, ng/mL	2.1 ± 1.6		2.2 ± 3.5		1.7 ± 2.3		0.617
Skatole, ng/mL	5.2 ± 10.0		1.9 ± 3.6		8.7 ± 22.6		0.253
Androstenone, ng/mL	1.9 ± 1.6		3.9 ± 4.4		1.8 ± 2.6		0.676

^{ab} Values with a same superscript in the same row are not significantly different.

^x Cut-off value between brackets.

^y Proportion of animals above cut-off value(s).

5. Results

Treatments were found to be effective in reducing or inducing soiling (p<0.001), with an average score of 1.0 ± 0.1 , 0.7 ± 0.1 and 2.0 ± 0.2 for CONTROL, CLEAN and DIRTY pigs, respectively. Scores might be considered rather low for the dirty group, but it should be remembered that soiling was scored about 24 h after soiling. By that time, a fair amount of dirt had already dried and fallen off.

Treatment did not significantly affect indole, skatole and androstenone concentrations in serum (Table 2). A time effect was found only for serum indole, but no further differentiation at certain time points could be made by Tukey's post hoc test. This indicated no evolution in boar taint compounds due to treatment. Similarly, treatment did not affect the concentration of indole, skatole and androstenone analysed in fat. The hot iron method, the assessment by the home panel and the odour of fat and meat as scored by the expert panel also revealed no significant differences between treatments.

However, the standardised consumer panel did give significantly better scores for general taste and flavour for CLEAN compared to DIRTY pigs. This was also reflected in the only significant correlation that was found between the soiling score and the detection parameters, namely the correlation between soiling score and flavour as scored by consumers (r = 0.30, p = 0.021). Intriguingly, the expert panel scored general meat flavour from CONTROL to be significantly superior than that from CLEAN pigs.

Significant correlations between skatole and androstenone versus the other detection parameters are given in Table 3. Significant correlation with skatole is limited to the hot iron method and the expert evaluation of fat samples (general, androstenone). For androstenone, more significant correlations were found, namely with the hot iron method, the standardised consumer score for odour, and several expert evaluation parameters for fat and meat. With the exception of the expert evaluation of fat odour, all detection parameters were more correlated with androstenone than with skatole. No significant correlations between laboratory analysis of skatole/androstenone and the home consumer panel were found. Indole level in fat was only correlated with the serum indole level (r=0.35, p=0.010). For skatole and androstenone, correlation between the level in fat and the level in serum was 0.49 and 0.56, respectively.

6. Discussion

With the exception of the standardised consumer panel evaluations, none of the other boar taint detection methods – not even those specifically designed to detect skatole concentrations – indicated an elevated concentration of boar taint in boars subjected to extra soiling than in boars that were kept extra clean. Nor did serum analysis of

Table 3
Pearson correlation coefficients between different detection methods.

	Skatole		Androstenone	
	r	Р	r	р
Hot iron	0.29	0.033	0.43	0.001
Consumers: Odour	0.15	0.265	0.27	0.047
Experts: Fat odour general	0.41	0.002	0.31	0.026
Experts: Fat odour androstenone	0.43	0.001	0.23	0.096
Experts: Meat odour general	0.08	0.539	0.65	< 0.001
Experts: Meat odour androstenone	0.05	0.714	0.62	< 0.001
Experts: Meat odour skatole	-0.12	0.387	0.51	< 0.001
Experts: Meat flavour general	0.14	0.296	0.53	< 0.001
Experts: Meat flavour androstenone	0.38	0.004	0.43	0.002
Experts: Meat flavour skatole	-0.11	0.438	0.52	< 0.001
Serum: Skatole (week 26)	0.49	< 0.001	0.16	0.332
Serum: Androstenone (week 26)	0.16	0.283	0.56	< 0.001

boar taint compounds, which reveals evolution during treatment for each individual boar, show any difference between treatments. Expert panels even judged general meat flavour to be inferior in CLEAN as compared with CONTROL pigs. These findings, therefore, provide only little support to the statement by Hansen et al. (1994) that skatole concentration can be lowered by keeping pigs clean.

The lack of significant differences between the groups could be due to the low average skatole concentrations, in line with similarly low concentrations in our previous study with these hybrids (Aluwé et al., 2009). In an international study, performed by Walstra et al. (1999), average skatole concentrations differed according to the origin of the boars and ranged between 0.10 and 0.17 ppm. Literature also describes lower skatole concentrations in winter, the timing of our experiment, compared to those in summer (Hansen et al., 1994; Walstra et al., 1999). Gibis (1994) found the highest skatole concentrations in the months of March, June and July. In our study, a numerical increase - albeit not significant - in fat skatole concentrations for DIRTY compared to CLEAN and CONTROL pigs was found, as well as an increase in the proportion of pigs with a skatole level above the cut-off of 0.20 ppm. Hansen et al. (1994) found a comparable influence for indole as for skatole. Our study showed no significant effect for indole nor for skatole. The low average skatole concentration can hinder further significant reduction. On the other hand, if skatole levels are already low, the need to further reduce the level of skatole is also low, as the prevalence of odour/flavour problems due to skatole is yet limited and more focus is needed on androstenone.

The verdicts of the expert panel and the standardised consumer panel contradict one another. It is possible that the experts scored meat from CLEAN pigs worse than CONTROL pigs because of the numerically higher androstenone concentrations in the CLEAN group. This was also reflected in the results from the hot iron method, i.e., a higher percentage of pigs with boar taint in the CLEAN compared to the DIRTY and CONTROL groups. Both methods are performed by androstenone sensitive persons. With the exception of the expert evaluation of fat odour, all detection parameters were more correlated with androstenone than with skatole. Bonneau et al. (1992) also found a closer relationship between the expert scoring of fat/cooking odour and fat androstenone level than for skatole level. As only the standardised consumer panel scored the DIRTY pigs worse than CLEAN, other effects on flavour, apart from boar taint, due to the different hygiene treatments may cause this difference. However, studies that have compared the influence of different management systems, like free range pigs or organic pigs with intensive fattening systems did not indicate differences in meat taste or off-odour (Van der Wal et al., 1993) or flavour and aroma (Jjönsall et al., 2002) respectively. Compared with a consumer panel, the experts and the hot iron method might overestimate the boar taint problem due to the experts' training and selection towards androstenone sensitivity. For the standardised consumer panel, androstenone may be less important than skatole, especially because cooking odour is not taken into account in our panel and androstenone sensitivity was not tested. In the present study and in our previous studies with standardised consumer panels, no significant difference in odour between treatments was found, although difference in flavour was found. Consumers also reacted negatively towards the unseasoned grilled meat presented in the standardised consumer panel. This may explain why the meat received generally lower scores than in home panels. The results of the home panels may be a better representation of the actual consumer acceptance of boar meat, but it may be harder to find differences between treatments. While the standardised consumer panels indicated a small improvement of boar taint by cleaning the pigs, the home panels minimised the occurrence of boar taint in the meat samples received.

In general, the findings from the consumer panels seem too weak to confirm the statement that cleanliness affects boar taint in pigs.

7. Conclusion

Only the standardised consumer panel evaluations indicated an elevated level of boar taint in boars subjected to extra soiling compared to the boars that were kept extremely clean. This was not confirmed by laboratory analyses or expert scores. We found no clear indication of skatole reduction by improving the cleanliness of pigs.

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