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# Fatty acid composition of subcutaneous adipose tissue from entire male pigs with extremely divergent levels of boar taint compounds — An exploratory study

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#### ABSTRACT

This exploratory study investigated the variability of fatty acid composition in entire male pigs with extremely divergent levels of boar taint compounds. Fatty acids were quantified in back fat samples from 20 selected carcasses of Pietrain\*F1 sired boars (average carcass weight 84 kg) with extremely low (LL) or extremely high (HH) levels of androstenone, skatole, and indole. Concentrations of polyunsaturated fatty acids (PUFA) were significantly (p < 0.05) increased in LL boars (23.4%) compared to HH boars (19.7%). This was mainly due to increased levels of linoleic acid (C18:2 n – 6) and  $\alpha$ -linolenic acid (C18:3 n – 3). Correspondingly, unsaturated fatty acids (SFA) were significantly lower (p < 0.05) in LL boars (35.2%) compared to HH boars (37.7%). The findings are discussed with respect to potential effects on flavor formation in boar fat and meat. Further research is needed to study the gender specificity and the interplay of the synthesis and the metabolism of steroids, lipids, and the clearance of skatole in pigs.

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#### 1. Introduction

Back fat (subcutaneous adipose tissue) in pigs is mainly composed of water, collagen and lipids (which consist mainly of triacylglycerols). The concentrations of fatty acids determine the firmness of back fat and its technological as well as its nutritional quality (Wood, Enser, Whittington, & Moncrieff, 1989). Surgical castration of male piglets is routinely applied in most countries to prevent off-odors and because it reduces sex-specific behavior as the animals mature. The EU has imposed a voluntary ban on this practice. This is the background for the present study of the consequences for meat and fat quality of boars. Numerous studies have indicated that the accumulation of androstenone, skatole, and indole are the main cause for decreased consumer acceptance of boar meat as reviewed by Lundström, Matthews, and Haugen (2009) and Xue and Dial (1997). Previous studies have also identified various other volatiles such as aldehydes and short chain fatty acids as potential causes for off-odors in boar fat samples that contain very low levels of androstenone, skatole and indole (Rius, Hortos, & Garcia-Regueiro, 2005). Recently, the skatole metabolite 2-aminoacetophenone was suggested as another candidate for contributing to off-flavors in boar meat (Fischer et al., 2014).

It has also been suggested that the back fat composition itself may affect the release (volatilization) of androstenone and skatole and, subsequently, their impact on olfactory perception (Rius et al., 2005). Besides such effects of matrix composition on flavor release (Chevance & Farmer, 1999), fatty acids themselves directly contribute to flavor formation in pork, e.g. during oxidative processes, see for example Larick, Turner, Schoenherr, Coffey, and Pilkington (1992). Aldehydes resulting from fatty acid oxidation such as hexanal were shown to have very low detection thresholds (Abraham, Sánchez-Moreno, Cometto-Muñiz, & Cain, 2012) and are described as off-flavor in foods (Brunton, Cronin, Monahan, & Durcan, 2000). In general, the susceptibility of fats to oxidative deterioration increases with the degree of unsaturation, i.e., their oxidative stability is impaired with increasing levels of MUFA and, especially, PUFA (Shahidi & Zhong, 2010).

Pig genetics and diet have been shown to affect the degree unsaturation in fat tissues; and carcass fatness was shown to be inversely related to unsaturation (Wood et al., 2008). With respect to gender, sex specific differences in fat composition have been reported before (Barton-Gade, 1987; Wood et al., 1989). The carcass weight in those studies was, however, much lower than today. A recent meta-analysis using studies from 1990 until 2010 confirmed previous findings that boars have a higher average PUFA content than castrates while no difference between entire males and female pigs was found (Pauly,







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Luginbühl, Ampuero, & Bee, 2012). At higher weights and with higher physical maturity, however, gender differences are likely to be stronger.

Until the work of Wood et al. (1989) it had not been possible to say whether the differences in fatty acid composition were explained by differences in fatness between the sexes. The authors concluded that "the difference in sex hormone metabolism between males and females may be responsible for the effects on composition. This causes the skin to be thicker in entire males, testosterone possibly having a primary effect on the synthesis of collagen in skin and back fat". The biosynthesis of androstenone in the testes is closely linked to the synthesis of anabolic testicular hormones such as testosterone (Claus, Weiler, & Herzog, 1994).

To the best of our knowledge, no study has reported relationships between key boar taint compounds and fatty acid composition. This exploratory study thus was aimed specifically at (i) investigating the variability of fatty acid composition of entire male pigs and (ii) comparing entire males with extremely divergent levels of boar taint compounds.

#### 2. Animals, material and methods

No animal care approval was required from the University of Göttingen for these experiments because only samples of subcutaneous adipose tissue from carcasses were used.

#### 2.1. Samples

Back fat samples for this study were taken from boar carcasses from a project which had been constructed to evaluate boar taint levels and performance data in approximately 1000 Pietrain sired crossbred males in Germany. The paternal line was purebred Pietrain, the maternal line was German Large White × German Landrace. Pigs were raised in a performance testing station (Haus Düsse) using standardized feeding (16.0% crude protein, 1.0% lysine; 13.4 MJ/kg) according to the requirements for pig performance testing in Germany (ZDS, 2007), and were slaughtered in a commercial abattoir. After chilling at slaughter, samples of subcutaneous fat were taken from the neck, individually packed in polyethylene bags under vacuum, and stored at -20 °C until analysis. Twenty samples for fatty acid analysis were selected from the samples in storage, based on their levels of androstenone and skatole. The animals studied here originated from seven sires and 14 dams. Ten samples had very low (LL) and ten had very high (HH) levels of androstenone, skatole and indole. Of the pigs selected for the present study, the average hot carcass weight yielded about 85.6 kg (range: 77.0 to 95.5 kg); average age at slaughter was 180 days (range: 153 to 212 days). Average back fat thickness was 18 mm at 13/14th rib (range: 12 to 28 mm). Average lean meat yield was 61% (range: 53.8 to 64.7%) as estimated using the 'Bonner Formel' according to the procedures described in ZDS (2007). Care was taken to have a balanced variation of back fat thickness within groups.

#### 2.2. Laboratory analyses

Androstenone was quantified using gas chromatography mass spectrometry (GC–MS), skatole and indole via liquid chromatography with fluorescence detection (HPLC) as described previously (Mörlein, Grave, Sharifi, Bücking, & Wicke, 2012). Results are given in ng per g melted back fat. Fatty acid analysis was performed in duplicate using gas chromatography with flame ionization detection as described previously (Koch et al., 2011). Individual fatty acids are given as percent of total detected fatty acids, and summarized as saturated fatty acids (SFA), monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA). All laboratory analyses were completed within six months after sampling.

#### 2.3. Data analysis

Analysis of variance (ANOVA) was used to study differences between selected groups using SAS software (The SAS Institute Inc.; Cary, NC, USA). The experimental unit was the individual pig, and the fixed effect included in the model was boar taint compound level group (LL, HH). One-way ANOVA was performed using the GLM procedure in SAS 9.3; normality was checked using the UNIVARIATE procedure. Differences between LS-means were tested for statistical significance using the PDIFF option. Multivariate analyses were performed using The Unscrambler 10.3 (CAMO Software AS; Oslo, Norway). Principal component analysis (PCA) was performed to elucidate correlations between fatty acid composition, carcass data and boar taint compound levels. For PCA, data were standardized (1/standard deviation) and mean centered (mean = 0); full cross validation was performed.

#### 3. Results

#### 3.1. Carcass characteristics and boar taint compounds

Carcass characteristics, intramuscular fat, and boar taint compound levels of the selected animals are shown in Table 1. Hot carcass weight, age at slaughter, and back fat thickness were not significantly different (p > 0.05) between LL and HH boars. Lean meat yield was significantly higher (p < 0.05) in LL (62.9%) compared to HH boars (59.2%). Due to the selection, androstenone, skatole and indole were significantly (p < 0.01) higher in HH than in LL boars.

#### 3.2. Fatty acid composition

Results of fatty acid analysis with respect to boar taint levels are shown in Table 2. Saturated fatty acids were significantly (p < 0.05) increased in entire males with high levels of androstenone and skatole (= HH boars). This was mainly due to significantly higher levels of myristic acid (C14:0), palmitic acid (C16:0), and arachidic acid (C20:0) in HH animals. The level of monounsaturated fatty acids (MUFA), especially oleic acid (18:1 n – 9 cis), did not (p > 0.05) differ between LL and HH boars. Polyunsaturated fatty acids (PUFA) were significantly (p < 0.05) increased in boars with low levels of androstenone and skatole (= LL boars). This was mainly due to increased levels of linoleic acid (C18:2 n – 6) and  $\alpha$ -linolenic acid (C18:3 n – 3).

## 3.3. Relationship of fatty acid composition, carcass characteristics and boar taint compounds (PCA)

Fig. 1 shows the results of the principal component analysis using the individual fatty acid levels as input variables; carcass data, summed fatty acids (SFA, MUFA, PUFA) and boar taint compounds were used with very low weights ('passified') such as they were not contributing to the model but used for graphing only. As to be seen from the score plot that shows the distribution of samples in the space of the input

#### Table 1

Carcass characteristics (LS-means) of entire males selected for very low levels of androstenone and skatole (LL, n = 10) compared to very high levels (HH, n = 10).

	LL	HH	s.e.	F-value	p-Value
Hot carcass weight, kg	86.6	84.7	2.00	0.47	0.4998
Age, d	177.7	182.2	5.41	0.35	0.5641
Back fat thickness, mm	17.7	18.5	0.13	0.19	0.6688
Lean meat yield <sup>a</sup> , %	62.9	59.2	0.62	17.99	0.0005
Intramuscular fat, %	1.04	1.19	0.13	0.69	0.4164
Androstenone, ng/g <sup>b</sup>	97.7	2983.7	305.53	44.61	<.0001
Skatole, ng/g <sup>b</sup>	37.5	464.2	38.41	61.71	<.0001
Indole, ng/g <sup>b</sup>	33.9	331.9	68.67	9.41	0.0066

<sup>a</sup> Estimated (according to ZDS, 2007).

<sup>b</sup> Given in ng/g melted fat.

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