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Influence of chicory roots (*Cichorium intybus* L*)* on boar taint in entire male and female pigs

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Abstract

Boar taint is an off-flavour of pork caused primarily by a microbial breakdown product, skatole and a testicular steroid, androstenone. As skatole is produced in the large intestine from tryptophan, it is possible that some 'bioactive' ingredients could modify protein fermentation and, in the process, diminish boar taint. The aim of this study was to examine the effect of inulin-rich chicory roots (Cichorium intybus L.) on boar taint. In the first of three trials individually penned, entire males and females were given an organic concentrate in which 0·25 of the daily energy intake was replaced with crude chicory roots for 9 or 4 weeks prior to slaughter. In the second trial, entire male pigs were given diets that included, either crude chicory roots, dried chicory roots, or inulin (extracted from chicory roots) for 6 weeks pre-slaughter. In the third trial, intact male pigs were given the dried chicory diet for either 2 or 1 week before slaughter. In all trials the chicory diets were offered on a scale at 0·95 of the Danish recommendation for energy intake, and pig performance was compared with a control group given the organic concentrate at 0·95 of recommended energy intake plus silage ad libitum. In trial 1 an additional control group was offered the organic concentrate at a daily energy intake level of 1·0 of Danish recommendations. The pigs in trials 1, 2, and 3 were slaughtered at an average live weight of 118, 124, and 110 kg, respectively, in order to ensure that they had achieved sexual maturity. Overall, skatole concentrations in blood plasma and backfat at slaughter were reduced to almost zero levels by including crude or dried chicory or inulin in the diet. This occurred irrespective of sex and length of feeding period (1 to 9 weeks). In trial 3 a significant effect on blood plasma concentration was observed after 3 days of feeding a diet containing dried chicory. The only significant reduction in plasma androstenone levels was detected in pigs given the crude chicory for a 9 week duration in trial 1. The production and proportion of lean was generally not affected by the addition of either form of chicory to the diets in trials 1 and 2. Therefore, dried chicory may be the most suitable form for commercial use because it: had no initial adverse effects on food intake, consistently reduced skatole without reducing performance, was easy to handle throughout the entire year and is relatively inexpensive.

Keywords: *androstenone, boar taint, chicory, inulin, skatole.*

Introduction

Boar taint is an off-flavour that affects the acceptability of pig meat. It is caused by skatole, androstenone and possibly other less known compounds of fat (Weiler *et al*., 1995 and 2000; Bonneau *et al*., 2000; Matthews *et al*., 2000; Babol *et al*., 2002). Skatole (3-methylindole) is produced in the large intestine by microbial breakdown of tryptophan; the compound exhibits an intense faecal odour (Babol and Squires, 1995; Vold, 1970). The pheromone androstenone is a testicular steroid with a characteristic urinary odour (Patterson, 1968; Bonneau, 1982), enhanced by skatole concentrations above 0·15 μg/g (Godt *et al*., 1996). Many studies have focused on the acceptability of meat in relation to boar taint. Without castration, boar taint is a problem in 5 to 10% of intact male pigs, having a skatole concentration in backfat above 0·25 μg/g when slaughtered at 100 kg live

weight (Hansen-Møller and Kjeldsen, 1998; Udesen, 1998). However, even values above 0·15 μg/g in backfat enhance the off-odour in both female and castrated as well as intact male pigs (Gibis, 1994; Godt *et al*., 1996; Gibis *et al*., 1998).

For welfare reasons it is likely that castration of male piglets without analgesia will be prohibited in the near future in several European countries. Legislation prohibiting castration of pigs is already scheduled to take effect in 2009 in Norway (Landbruksdepartementet, 2002) and Switzerland. A similar ban is under consideration for the Danish organic pig production. This calls for alternative means and approaches to avoid or reduce the boar taint problem.

One such approach is the use of relevant 'bioactive' ingredients i.e. foods that exert biological effects not directly related to the nutritive value. Chicory (*Cichorium intybus* L.) is a bioactive crop that may positively affect eating quality in intact male and female pigs. The hypothesis of a direct effect of chicory on the taste of pork is based on the presence of fructo-oligosaccarides (inulin) (Claus *et al*., 1994; Jensen and Jensen, 1998) and to a lesser extent sesquiterpene lactones (bitter compounds) in the roots (Bais and Ravishankar, 2001). Inulin reduces glucose uptake and thus glycogen accumulation and functions as a soluble dietary fibre (prebiotic) in humans (Tomasik and Tomasik, 2003). It also functions as a prebiotic in pigs (Tungland, 1998) due to fermentation in caecum and colon, stimulating the growth of beneficial bacteria such as *Bifidobacteria* and *Lactobacillus*. This bacterial growth may positively influence the sensory (eating) quality of the meat by reducing, among other things, the numbers of bacteria producing the putrefactive products skatole, indole, and p-cresol in caecum and colon (Takahashi *et al*., 1996; Jensen and Hansen, 2006). However, a direct effect of sesquiterpene lactones is also likely (Rees and Harborne, 1985; Choi *et al*., 1998; de-Kraker *et al*., 1999 and 2001; Bais *et al*., 2000; Bais and Ravishankar, 2001).

The objective of the present study was to evaluate the effect of various finishing strategies using crude and dried chicory roots on the boar taint compounds skatole, indole, and androstenone in incact male and female pigs. To obtain information on related sensory properties such as flavour, odour, and texture, sensory profiling was carried out on selected meat samples and will be reported elsewhere (D.V. Byrne and L.L. Hansen, unpublished).

Material and methods

Experimental animals and design

All experimental animals of the three trials were Danish crossbred pigs of Duroc sire × zigzag crossbred dam of Danish Landrace × Large White (D × (L × Y) produced at Research Centre Foulum. In trial 1 both intact male (no. = 16) and female pigs (no. = 16) were used, but only intact male pigs were used in trial 2 (no. = 32) and 3 (no. = 16). In all trials pigs were allocated to treatments according to live weight and litter, and in trial 1 also according to sex.

In trial 1, the pigs were kept in litters and offered an organically certified concentrate according to scale (1·00 of daily energy intake according to Danish recommendations by Madsen *et al.* (1990)) and *ad libitum* clover grass silage according to organic farming practices for 4 weeks prior to initiation of the experiment (day 0), at a mean live weight of 55 kg. Organic pigs in Denmark must have access to some kind of roughage e.g. clover grass silage or chicory roots according to organic farming practices. At the beginning of the experimental period (day 0) the pigs were allocated to four treatments (ConCtrl, OrgCtrl, CC4 and CC9) and moved to individual pens. For details on treatments and food composition see Tables 1 and 2. In treatment OrgCtrl the *ad libitum* clover grass silage was not expected to provide the last 0·05 of daily energy intake because at very high concentrate intakes, clover grass silage intake is very limited (Danielsen *et al*., 2000). During the 1st week of allocation of crude chicory (CC9 and CC4) the pigs were adapted to the chopped roots by gradually increasing the ration to the planned 0·25 of the daily energy intake according to individual appetite. Male pigs of treatment CC9 were fed for 63 days (i.e. 9 weeks) with chicory, while female pigs were feed CC9 for 65 days before slaughter. Correspondingly, male pigs of treatment CC4 were fed the last 28 days (i.e. 4 weeks) with chicory and the female pigs 30 days before slaughter. At the end of the trials, pigs were consuming 3 kg crude chicory per day. The 0·95 energy level was chosen instead of 1·00 in the two chicory treatments because some pigs might not consume their full allowance of chopped crude chicory roots or concentrate because of the bulk and bitter taste of the roots.

In trial 2, the individually penned pigs were allocated to the four treatments (OrgCtrl, CC6, DC6, and I6) and for 4 weeks prior to initiation of the experiment (day 0), all pigs were offered organic concentrate according to scale plus *ad libitum* clover grass silage. From day 0 to day 42 (i.e. 6 weeks) or day 44 (slaughter days) pigs of the 4 treatments (OrgCtrl, CC6, DC6, and I6) were given their food according to Tables 1 and 2. Pigs were adapted to the crude chicory from day 0 to 7 (i.e. week 1) in treatment CC6 as in trial 1 whereas the dried chicory in treatment DC6 was eaten without an adaptation period.

Trial 3 was performed in parallel with trial 2 utilizing two of the same diets. Sixteen pigs were kept in individual pens from day −7 to day 0 and offered an organic concentrate and *ad libitum* clover grass silage. Eight pigs were then given the dried chicory diet for either 1 (day 7–14, treatment DC1) or 2 weeks (days 0 to 14, treatment DC2) before slaughter at day 15 (Tables 1 and 2).

Experimental foods

An inulin-rich variety of chicory (*Cichorium intybus* L. var. Orchies) was grown organically and used after 2 to 3 months (trial 1) or 2 to 4 months (trial 2 and 3) of storage. Crude chicory roots were chopped by a fast mincer (Wiencken, Copenhagen, Denmark) and added on top of the concentrate at feeding. Fructan level in the roots was determined as described by Larsson and Bengtsson (1983). In trial 2 and 3 some of the chopped roots were dried in a drying cupboard at a temperature just below 65°C for 48 h. Protein concentration in diets were collected at the time of slaughter in trial 1 and trial 2 (corresponding to the diet in

Table 1 *Experimental design: control and experimental diets used in trial 1 (9 weeks), trial 2 (6 weeks) and trial 3 (2 weeks) (according to Danish recommendations by Madsen et al.* (1990) *all energy values are given as % of total energy intake per day*)

	Treatment			Chicory roots		
	Code	Organic conc.	Clover grass silage	Crude (CC)	Dried (DC)	Inulin (I)[¶]
Trial 1						
Organic concentrate only	ConCtrl	100				
Org. concentrate + silage	OrgCtrl	95	*ad libitum*			
Org. conc. + crude chicory 4 weeks	CC4	95[†]/70[‡]	*ad libitum*[†]	25[‡]		
Org. conc. + crude chicory 9 weeks	CC9	70		25		
Trial 2						
Org. concentrate + silage	OrgCtrl	95	*ad libitum*			
Org. conc. + crude chicory 6 weeks	CC6	70	–	25		
Org. conc. + dried chicory 6 weeks	DC6	70	–		25	
Org. conc. + inulin 6 weeks[¶]	I6	70	–			14
Trial 3						
Org. conc. + dried chicory 1 week	DC1	95[§]/70[‖]	*ad libitum*[§]/-[‖]		[§]/25[‖]	
Org. conc. + dried chicory 2 weeks	DC2	70	–	–	25	–

[†] Control treatment (OrgCtrl) day 0 to 35 (i.e. 5 weeks).
[‡] Supplemented with chicory (CC4) day 35 to 63 (i.e. the last 4 weeks before slaughter).
[§] Control treatment (OrgCtrl) day 0 to 7 (i.e. 1 week).
[‖] Supplemted with chicory (CC1) day 7 to 14 (i.e. the last week before slaughter).
[¶] Raftiline®HP, Orafti Ltd, Belgium (produced from chicory roots).

Table 2 *Composition of control and experimental diets in trials 1, 2, and 3. Silage was offered to some control treatments (OrgCtrl) but it was rarely eaten and is therefore not included*

	Trial 1–treatments[†]		Trial 2 + 3–treatments[†]			
	ConCtrl and OrgCtrl	CC4 and CC9	OrgCtrl	CC6[‡]	DC6[‡], DC1[§] and DC2[§]	I6[‡]
Dry matter (g/kg)	890	890/250	880	880/250	900	900
Contents (g/kg wet matter)						
Crude chicory	0	564	0	563	0	0
Dried chicory	0	0	0	0	276	0
Inulin (Raftiline®HP)	0	0	0	0	0	163
Rape seed cake	145	63	145	63	105	121
Peas	240	103	239	103	173	201
Wheat	223	97	222	97	161	186
Barley	220	95	220	95	159	184
Oat	50	22	50	22	36	42
Soya bean	100	43	100	43	72	83
Vitamins and minerals	22	13	22	13	17	18
Marker (Cr_2O_3)	–		2	1	2	2

[†] Treatments include ConCtrl and OrgCtrl offered concentrate, CC offered concentrate and crude chicory roots, DC offered organic concentrate mixed with dried chicory roots and I offered organic concentrate mixed with inulin.
[‡] Trial 2 only.
[§] Trial 3 only.

trial 3) were determined according to the Kjeldahl method using a Kjeltec autosampler system 1035 (Foss Tecator, Höganäs, Sweden) whereas fat (hydrochloric acid-fat) was extracted with diethyl ether after acid hydrolysis (Stoldt, 1952). Ash was determined in accordance to the Association of Official Analytical Chemists (1990). Scandinavian Feed Units for pigs for net energy (FUp = feed units pigs; 1 FUp = 7·38 MJ) were calculated according to Boisen and Fernandez (1997). In trial 2 (and 3) sugars (glucose, fructose, sucrose and fructans) were determined by the method of Larsson and Bengtsson (1983). Starch and non-starch polysaccharides (NSP) were analysed as described by Bach Knudsen (1997). Klason lignin was measured according to Teander *et al.* (1994).

Animal sampling and analysis

Pigs were weighed on days 0, 63 and 65 (at slaughter) in trial 1 and day 0; days 42 and 44 (at slaughter) in trial 2, while they were weighed days 0 and 15 in trial 3.

Blood samples from the vena jugularis for plasma were collected in heparinized vacuum tubes on days 0 and 56 in trial 1; days 0 and 41 in trial 2 for analysis of androstenone and skatole. In trial 3, blood was collected and analysed for androstenone, skatole, and indole just before feeding on days 0, 3, 7, 10, and 14. Androstenone was measured by direct immunochemical analysis of the blood plasma samples (Tuomola *et al.*, 1997). Skatole in blood plasma was measured according to the high-performance liquid chromatography method described by Hansen-Møller (1998) and modified by omitting the column switching procedure and injecting the protein-precipitated plasma directly on a Hypersil 3 μm 3 × 60 mm column. The lower limit of quantification was 0·12 μg/l. Indole analyses were performed and expressed in relation to skatole content in plasma.

In trial 1, male pigs (16) were slaughtered by the end of week 9 (day 63) and the 16 female pigs 2 days later due abattoir limitations. On both slaughter days, 2 × eight pigs

were delivered at 08:00 and 10:30 h after a short transportation time (5 to 10 min) to the abattoir and kept in individual pens. Pigs were slaughtered alternately according to treatment and litter from the time of arrival with an avg. interval of 15 to 20 min. The same protocol was used for the pigs in trial 2 although 16 pigs from the four heaviest litters that were slaughtered the first day (day 42 and day 44). In trial 3 all 16 pigs were slaughtered alternately according to treatment and litter on day 15. In all trials backfat samples were collected 45 min post slaughter and skatole equivalents were measured by the automatic spectrophotometric method described by Mortensen and Sørensen (1984). All pigs in trials 1, 2, and 3 were slaughtered at an average live weight of 118, 124, and 110 kg, respectively, to ensure sexual maturity of intact male pigs had been reached, thereby securing potential high levels of boar taint (high skatole and androstenone concentrations) in meat and fat (Hansen *et al.*, 1997b; Babol *et al.*, 2004; Zamaratskaia *et al.*, 2004).

Statistical analysis

All statistical analyses were carried out using the program package from Statistical Analysis Systems Institute (2000).

Trial 1. The GLM procedure was used to calculate the least-squares means and standard errors of the means for boar taint and production variables. The GLM models included the fixed effects of diet, sex, and animal replicate (litter) as well as interaction between diet and sex. The boar taint variables were skatole or androstenone in the blood 1 week before slaughter (both arithmetic and log-transformed data) or skatole in backfat at slaughter (both arithmetic and log-transformed data). Preliminary statistical analyses of logarithmically transformed data of skatole in blood and backfat and androstenone in blood to ensure normal distribution gave results similar to those of non-transformed data and therefore non-transformed data were used throughout. In addition, a GLM procedure was conducted with androstenone in the blood plasma just before initiation of the experiment (day 0) as a covariate to take out the covariance (correlation) from androstenone at day 0 in the androstenone level in the blood day 56. In the case of the production variables live weight, warm carcass weight, FUp per kg gain, and daily gain, a GLM procedure was conducted with initial live weight just before initiation of the experiment (day 0) as a covariate to take out the covariance. In the case of the variable lean meat proportion, a GLM procedure was conducted with warm carcass weight as a covariate to take out the co-variance. Performance results of the production variables FUp per kg gain, and daily gain, of all 4 treatments were calculated for the whole experimental period (63 days for entire male pigs and 65 days for female pigs).

Trial 2. The GLM models were applied as in trial 1, including the fixed effects of diet, replicate (litter) and slaughter day, as well as the interactions between diet and replicate and between diet and slaughter day. The boar taint variables were skatole, indole and androstenone (arithmetic and log-transformed data) in the blood plasma day 0 and day 41. Skatole in backfat at slaughter was analysed using both arithmetic and log-transformed data. In addition, the GLM procedure was used with androstenone in blood (arithmetic and log-transformed data) at day 0 as a covariate in the model with androstenone in blood at slaughter (day 41).

Trial 3. The GLM procedure was used to calculate the least-squares means and standard errors of the means for the boar taint variables in blood and backfat (arithmetic and log-transformed data). The variables (skatole, indole and androstenone) in blood plasma for each day (3, 7, 10, and 14) were analysed separately and for skatole in backfat day 15 using the GLM procedure. The models included the fixed effect of diet and animal replicate (litter) and the value of the corresponding variable just before initiation of the experiment (day 0) as a covariate to take out the co-variance (correlation) when appropriate.

Results

Trial 1

Root yield of chicory was 30 t/ha, and the inulin (fructan) concentration was 150 g/kg crude root. Roots contained 1·14 FUp (equal to 8·44 MJ net energy) and 53 g digestible protein per kg. After the week of adaptation in which the individual pigs were offered increasing amounts of the bitter crude chicory roots, all pigs consumed daily allocations of chicory roots and were consuming 3·0 kg roots per day before slaughter. Several pigs consumed the bitter chicory before the standard diet. All daily concentrate rations were also eaten but intake of silage of the pigs on treatment OrgCtrl was minimal. Each pig was given 3·43 (ConCtrl), 3·26 (OrgCtrl), 3·25 (CC4), and 3·25 (CC9) FUp per day at the time of slaughter.

All pigs appeared healthy and no differences between treatments were noted. Daily gain, warm carcass weight and live weight at slaughter of animals on the two chicory treatments CC4 and CC9 corresponded to those on the OrgCtrl treatment also offered 0·95 of energy level but were lower than for the ConCtrl treatment pigs given 1·0 of energy requirements (Table 3).

Food conversion ratio of the CC4 treatment was significantly lower ($P < 0{\cdot}05$) than controls (Table 3) but the proportion of lean meat in the chicory-fed pigs was not negatively affected, despite the fact that the diet had a lower content of crude protein compared with the two control treatments. Pigs were given 552 g (ConCtrl), 526 g (OrgCtrl), 425 g (CC4), and 425 g (CC9) digestible protein per day.

Irrespective of sex and experimental period, all chicory-fed pigs had skatole concentrations in backfat at slaughter (Figure 1a) and skatole concentrations in blood plasma after 21 and 56 days on trial; (Table 4), that were not significantly different from zero. Both chicory treatments were equally effective in reducing skatole in blood and backfat compared with the two control treatments ($P < 0{\cdot}001$). The two control treatments contained only one of 8 intact male pigs with a backfat skatole concentration at slaughter above the current Danish limit of 0·25 μg/g (0·30 μg/g), 2 intact male pigs had

Table 3 *Production results (least-square means (LSM) and standard error of LSM (s.e.)) for trial 1 (four entire male and 4 female pigs per treatment), and trial 2 (eight entire male pigs per treatment)*

	Trial 1						Trial 2					
	Treatment						Treatment					
	ConCtrl	OrgCtrl	CC4	CC9	s.e.	Significance of treatment	OrgCtrl	CC6	DC6	I6	s.e.	Significance of treatment
Live weight (kg)												
Day 0	55·0	55·5	55·5	55·5	2·6		82·9^{a}	84·3^{a}	84·6^{a}	83·6^{a}	4·0	
Day 42/day 44	–	–	–	–	–	–	126·8^{a}	122·8^{bc}	126·7^{a}	120·2^{c}	1·3	*
Day 63/day 65	123·8^{a}	117·2^{b}	116·4^{b}	115·1^{b}	1·3	***	–	–	–	–	–	
Warm carcass weight (kg)	94·7^{c}	89·4^{d}	88·0^{d}	86·4^{d}	1·2	***	95·3^{a}	92·6^{ab}	94·0^{ab}	91·2^{b}	1·1	**
Lean meat percent	59·0^{e}	59·9^{ef}	60·1^{f}	59·8 ^{ef}	0·34	*	59·3^{a}	59·5^{a}	59·6^{a}	59·5^{a}	0·24	
Daily gain (kg)	1·070 ^{g}	0·968 ^{h}	0·908 ^{h}	0·934 ^{h}	0·024	***	0·932^{a}	0·851^{ab}	0·894^{ab}	0·808^{b}	0·033	**
Fup per kg gain‡	3·05^{i}	3·18^{i}	3·53^{j}	3·22^{i}	0·08	*	2·89^{d}	3·24^{e}	2·98^{d}	3·05de	0·08	§

$^{a, b, c, d, e, f,g,h,i,j}$ Within rows, least-square means not sharing a common superscript letter differ significantly ($P < 0·05$).
‡ FUp = Scandinavian Feed Units pigs; 1 FUp = 7·38 MJ.
§ Approaching significance ($P < 0·1$).

a concentration of 0·15 μg/g in the backfat, while the chicory fed intact male pigs varied from 0·01 to 0·03 μg/g. The combined overall least-square means for both sexes are given in blood plasma (Table 4). No interaction was found between sex and treatment for blood or backfat skatole levels. Plasma skatole concentrations were not significantly influenced by sex. Backfat skatole concentrations of intact males were higher ($P < 0·05$; Figure 1a). A decrease ($P < 0·05$) in the androstenone level was observed in CC9 compared with ConCtrl, when results were adjusted for androstenone levels at day 0 (covariate; Table 4).

Trial 2

The root yield for chicory grown for trials 2 and 3 was 40 t/ha, and the inulin content was 122 g/kg crude root. Chemical analyses of the diets showed a marked difference in the level of fructans (low molecular (LM) sugars such as inulin) between the control diet and the three experimental diets (Table 5). The overall concentration of fructan in the diets was such that the individual pigs were given a total of 36 g, 429 g, 446 g, and 428 g/day in OrgCtrl, CC6, DC6, and I6, respectively. As in trial 1, the pigs consumed bitter chopped crude roots without problems. Dried chicory roots mixed with the concentrate were given without a need for an adaptation period presumably because the dried chicory roots were less filling, (may be less bitter) and slightly sweet. Protein levels of the diets varied (Table 5); mean daily protein intake of each pig was 489 g (OrgCtrl), 398 g (CC6), 404 g (DC6), and 369 g (I6). Energy content of the diets was similar. Each pig was given 3·15 (OrgCtrl), 3·19 (CC6), 3·26 (DC6), and 2·80 (I6) FUp per day at the time of slaughter. Due to an error, I6 pigs received less energy per day (0·14 from inulin plus 0·70 from concentrate) compared with the other three treatments (0·95) and consequently the daily weight gain was significantly lower than for OrgCtrl (Table 3). The overall production results of the DC6 treatment were comparable with those of the OrgCtrl treatment and tended to be better than for treatments CC6 and I6.The lean content was almost identical for all treatments (Table 3) despite the differences in diet protein levels.

Irrespective of formulation, all chicory-fed entire male pigs (groups CC6 and DC6) showed a significant reduction in skatole concentrations in blood plasma compared with the control pigs (Table 4). Furthermore, skatole concentrations of the chicory-fed pigs were not significantly different from zero. Also the I6 treatment pigs had less ($P < 0·001$) skatole compared with the OrgCtrl treatment, however, this level was higher ($P > 0·05$) than zero. At the end of the 6-week trial period all chicory- and inulin-fed entire male pigs had less skatole concentrations in backfatthan those of

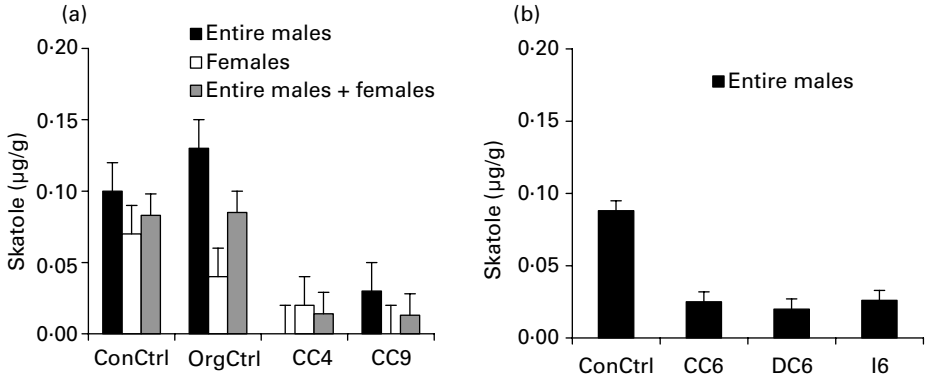

Figure 1 (a) Skatole in backfat (μg/g, least-square means ± standard error) in entire male (no. = 4), female pigs (no. = 4), both entire male and female pigs (no. = 8) at slaughter in trial 1 (significance of treatment: $P < 0·001$) and (b) entire male pigs in trial 2 (no. = 8) (significance of treatment: $P < 0·001$).

Table 4 *Skatole levels in blood (four entire male plus four female pigs per treatment) and androstenone (four entire male pigs/treatment) levels in trial 1 and skatole, androstenone, and indole levels in eight entire male pigs in trial 2 (least-square means (LSM) and standard error of LSM (s.e.))*

	Trial 1						Trial 2					
	Treatment						Treatment					
	ConCtrl	OrgCtrl	CC4	CC9	s.e.	Significance of treatment	OrgCtrl	CC6	DC6	I6	s.e.	Significance of treatment
Skatole (μg/l)												
Day 0							4·6[a]	3·2[a]	2·8[a]	3·8[a]	1·3	
Day 42							3·49[a]	0·32[b]	0·11[b]	0·68[b]	0·3	***
Day 8	1·82[a]	2·13[a]	0·13[b]	0·08[b]	0·36	***						
Androstenone (μg/l)												
Day 0	3·8	2·9	3·9	4·4	1·2		18·2[a]	23·0[a]	18·2[a]	13·5[a]	4·0	
Day 42							27·0[a]	27·1[a]	23·2[a]	27·0[a]	3·5	
Day 56	17·7[c]	13·5[cd]	13·7[cd]	9·1[d]	1·6	§						
Indole (μg/l)												
Day 42							1·4[a]	1·8[a]	1·2[a]	1·1[a]	0·3	

[a, b, c, d] Within trial and within rows, LSM not sharing a common superscript letter differ significantly ($P < 0·05$).
§ Approaching significance ($P < 0·1$).

Table 5 *Chemical analysis (g/kg dry matter) of the diets used in trial 2 (the pigs of the control group (OrgCtrl) rarely ate the offered silage, which is therefore not included in the analysis)*

	Treatment			
	OrgCtrl	CC6	DC6	I6
Protein	195	162	155	164
Fat	67	53	49	53
Ash	56	52	56	49
Low molecular sugars (total)	42	210	233	197
Glucose	1	1	2	1
Fructose	<1	3	16	<1
Sucrose	28	67	60	24
Fructan (inulin)	13	139	155	172
Starch	377	284	260	316
Dietary fibres (total)	203	183	189	184
Klason lignin	48	44	33	36
Non-starch polysaccharides (total)	155	139	156	148
Cellulose	43	40	44	42
Non-cellulosic polysaccharides[†]	112 (31)	99 (29)	112 (51)	106 (35)
Feed Units for pigs (FUp)[‡] per kg dry matter	1·14	1·15	1·14	1·13
FUp per pig per day	3·15	3·19	3·26	2·80

[†] Values in brackets denote the fraction of non-cellulosic polysaccharides that was insoluble.
[‡] 1·0 FUp = 7·38 MJ.

corresponding control-fed pigs ($P < 0·001$; Figure 1b); although in trial 2, eight intact males of the control treatment (OrgCtrl) had low skatole levels in backfat at slaughter that ranged from 0·03 to 0·13 μg/g (avg. 0·088 μg/g), which is well-below the off odour limit but corresponds to normal skatole levels (0·90 to 0·95) of the Danish entire male pig population. Plasma indole and androstenone concentrations of chicory- and inulin-fed pigs were not affected by treatment (Table 4). Plasma androstenone concentrations increased ($P < 0·01$) from day 0 (84 kg live weight) until day 41 (124 kg live weight).

Trial 3

The health status and final body weights of the pigs at slaughter were identical (90·2 and 110·2 kg) for both treatments, and the pigs consumed full rations from day 0. After only 3 days of being fed the dried chicory diet, plasma skatole concentrations of DC2 animals were lower ($P < 0·001$ than DC1 pigs offered the control organic concentrate diet (Figure 2a). Moreover, plasma skatole concentrations were reduced to nearly undetectable levels within 1 week and and remained low during the 2nd week of the experiment. A similar reduction in blood skatole concentrations were observed in DC1 animals during the second week of the experiment being fed the dried chicory diet as DC1 animals after 3 days chicory feeding (day 10) only showed slightly higher blood skatole concentration ($P < 0·05$) than DC2 animals. By the end of the experiment, plasma skatole concentrations in the blood of the two treatments were similar and barely detectable. The skatole concentration in backfat at slaughter was also low and similar in both treatments (DC1: LS mean 0·026 (s.e. 0·001) μg/g and DC2: LS mean 0·020 (s.e. 0·001); $P > 0·05$). Although plasma indole levels were low at the beginning of the experiment, further reductions ($P < 0·01$) were evident within a few days after dietary addition of chicory (Figure 2b). Plasma androstenone levels were not, however, affected by feeding chicory for 1 or 2 weeks (Figure 2c).

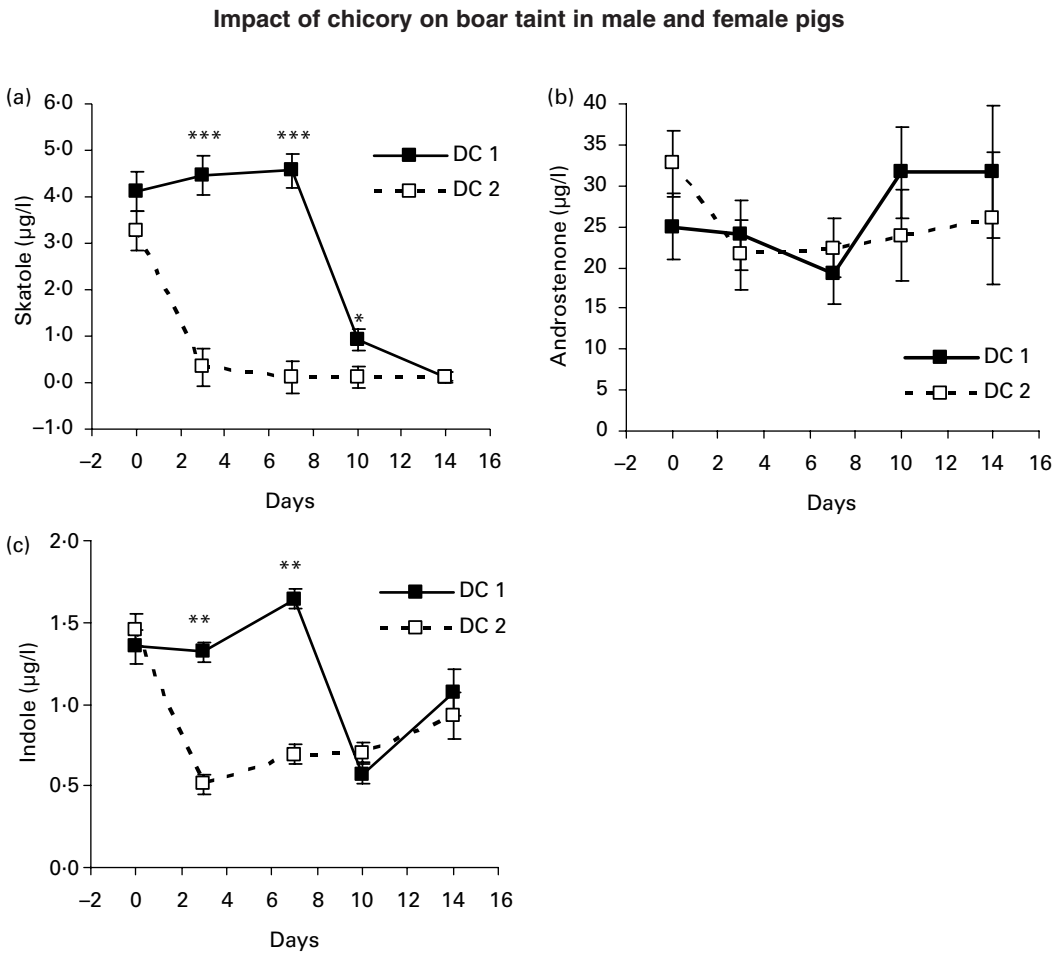

Figure 2 (a) Skatole, (b) androstenone and (c) indole in blood (µg/l, least-square means ± standard errors) from entire male pigs (no. = 8) given concentrate with dried chicory from days 7 to 14 (DC1) and from day 0 to 14 (DC2) (trial 3). Significant differences between treatments on given days are marked.

Discussion

Results of this study clearly demonstrate that feeding crude or dried chicory roots with a high content of inulin, or purified inulin strongly reduces the boar taint-associated compound skatole. Similar findings have been reported by Claus *et al.* (1994) and Jensen and Jensen (1998) for chemically purified inulin and fructo-oligosaccharides. Earlier Danish experiments have indicated skatole is the most important boar taint compound, as the majority of the human population is highly sensitive to this compound. Thus, it is important to reduce the skatole concentration in backfat below the current Danish limit of 0·25 µg/g to a proposed upper limit of 0·15 µg/g to avoid boar taint in more than 99% of all Danish entire male pigs slaughtered at 100 kg live weight (Godt *et al.*, 1996). In the remaining 1% of the Danish intact DLY crossbred male pig population with extremely high androstenone and with skatole concentrations approaching 0·25 µg/g or more, skatole enhances the off-odour and off-flavour of an odorous compound like androstenone so that the meat may be unacceptable to the consumers This conclusion may be an overestimation of the level of consumer acceptance of intact males when compared with other studies (Weiler *et al.*, 1995 and 2000; Bonneau *et al.*, 2000; Matthews *et al.*, 2000; Babol *et al.*, 2002) especially where androstenone sensitive consumers are concerned. Regardless, even this may involve only 1% of the pigs, this is a significant number of animals in any production system. One solution could be that androstenone-sensitive consumers might get the choice of buying branded meat from female pigs, but this will obviously not solve the skatole-based off-odour and off-flavour problem that also arises in pork from some female pigs (Bonneau *et al.*, 2000) and castrated male pigs (Gibis, 1994; Gibis *et al.*, 1998).

A strong correlation between skatole in blood plasma and backfat has been shown in several experiments ($r = 0{\cdot}90$ to 0·98, $P < 0{\cdot}001$) (Tuomola *et al.*, 1996; Hansen *et al.*, 1997a; Agergaard and Laue, 1998). In the case of androstenone, variable results have been obtained for the correlation of levels in blood and fat samples when the blood samples have been extracted with organic solvents before the analysis. The results have ranged from a lack of correlation (Malmfors and Andresen, 1975; Lundström *et al.*, 1978) to a high degree of association (Andresen, 1976; Groth and Claus, 1977; Booth *et al.*, 1986; Babol *et al.* 1999; Zamaratskaia *et al.*, 2004). On the other hand, the results from direct immunochemical analysis of non-extracted blood samples, such as those used in the current study, correlate well with the respective levels in backfat ($r = 0{\cdot}78$ to 0·89; $P < 0{\cdot}001$) (Tuomola *et al.*, 1997 and 2002). Due to these strong correlations, blood analysis has replaced repeated backfat analysis during the experimental period of the three trials.

Replacing 0·25 of the recommended daily energy intake with crude or dried chicory had a pronounced effect on the skatole concentrations in blood and backfat after only 1 week, and the effect clearly persisted for a 9-week feeding period, if results of all three trials are considered together. In the second trial, 0·14 inulin (corresponding to the amount of inulin (fructan) in 0·25 of chicory) reduced the skatole concentrations in blood and backfat to the same extent as 0·25 of chicory, which suggests the amount of inulin was most likely the most important factor responsible for the reductions in skatole concentration in blood and backfat. However, chicory may also have some additional beneficial effects on eating quality compared with inulin, perhaps due to the presence of bioactive bitter compounds in chicory roots.

Several studies have shown that rather large deviations in the amount of protein as well as the amount of the amino acid tryptophan in the diet did not change the skatole concentration in backfat (Pedersen *et al.*, 1986; Mortensen *et al.*, 1989; Bernal-Barragan, 1992). Therefore the chicory- and inulin-fed pigs were not supplied with further protein in the concentrate diet as the amount of protein needed for growth and lean meat in the chicory and inulin treatments in all three trials was fulfilled (0·15 to 0·16) according to current Danish recommendations for nutrients to slaughter pigs (Danish Bacon and Meat Council, 2002). This was also supported by similar levels of weight gain and lean content in the chicory treatments in trials 1 and 2 compared with the control treatments (OrgCtrl). In our opinion, the slight improvement in performance in trial 1 and 2 (mostly not significant) of the control treatment (OrgCtrl) given 0·95 energy level of concentrate plus *ad libitum* silage compared with the crude chicory-fed pigs on 0·95 energy level had little to do with the intake of silage but was more a function of crude chicory dietary adaptation, as this week was included in the performance results to show realistic practical performance results.

Plasma indole concentration was reduced within a few days after switching to chicory feeding in trial 3, while no reduction in indole was detected after 6 weeks of chicory and inulin feeding in trial 2. This difference of chicory/inulin feeding on the indole level in blood between trial 2 and 3 could originate from the difference in number of days the pigs were given the chicory/inulin. Microbial degradation of tryptophan to indole can be performed by many bacterial species; it is possible that the microbial ecosystem in the gastro-intestinal tract (GI), therefore requires an adaptation to the new diet to produce indole. This is in contrast to the skatole production, where a limited number of bacterial species are able of producing this substance; therefore, additional time would be required to detect differences in this substance. Indole results from this experiment demonstrate that chicory roots do not appear to function in the same way in the GI as zinc bacitracin and fermented liquid food, which all are associated with an increased production of indole in GI coinciding with a reduced skatole production (Agergaard *et al.*, 1998; Jensen *et al.*, 1998; Hansen *et al.*, 2000).

The androstenone concentration in the blood did not seem to be affected by feeding chicory or inulin and there is no clear explanation for the decrease in androstenone level in the four entire male pigs given crude chicory for 9 weeks before slaughter compared with one of the control treatments in trial 1. However, it is known that dietary fibre is able to reduce the amount of cholesterol in serum (Marlett *et al.*, 1994) by interrupting the entero-hepatic circulation of the steroid core of cholesterol. As androstenone and cholesterol follows the same biosynthetic pathway a prolonged feeding period with a dietary fiber as inulin could affect the androstenone content in the same way as cholesterol. If you consider no effect on androstenone in blood and backfat - by short time feeding - the best effect of chicory roots against boar taint perhaps may be obtained when the entire male pigs have not yet reached full sexual maturity in the interval 85 to 100 kg live weight (Claus *et al.*, 1994; Godt *et al.*, 1996). It is likely that with the very low levels of skatole obtained after chicory feeding, malodour from androstenone will not be enhanced and therefore will not result in serious boar taint problems in most cases, however, depending on the concentration of androstenone.

It was possible to produce *Cichorium intybus* L of the inulin-rich variety Orchies on clay type sandy soils under Danish organic production conditions. Unlike the traditional sugar-beet crop, chicory is somewhat insect-resistant possibly due to the inherent bitterness compounds found in the leaves and root (Rees and Harborne, 1985). Crop yields of 30 t and 40 t/ha on clay-type sandy soils under Danish organic production conditions were acceptable but further studies are needed.

Overall, it can be concluded from the present study feeding of 0·25 of crude and dried chicory significantly reduces skatole in blood and backfat (to almost zero levels) irrespective of sex and feeding period (1 to 9 weeks). Even as little as a 3-day feeding period of dried chicory resulted in a significant decrease in blood plasma skatole levels in trial 3.

From a holistic standpoint, it could be postulated that feeding dried chicory for as little as 1 week may ultimately provide a practically applicable way to reduce the sensory off-flavour problem boar taint, described as an unacceptable piggy/manure/urine-like odour and flavour character, in female and castrated males, as well as more importantly in pork from entire male pigs (D.V. Byrne and L.L. Hansen, unpublished). Further research may be expected to show which amounts and periods of dried chicory feeding that are needed to produce pork from entire male pigs on a commercial scale to ensure a low level of boar taint that is acceptable to the consumer.

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