

Oral presentation

## Prevention of boar taint in pig production. Factors affecting the level of skatole

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### Introduction

The indolic compound skatole is produced by microbial degradation of the amino acid tryptophan and has been detected in the rumen of cows and the caecum and colon of pigs. Together with the testicular steroid androstenone skatole is considered to be the main contributor to boar taint.

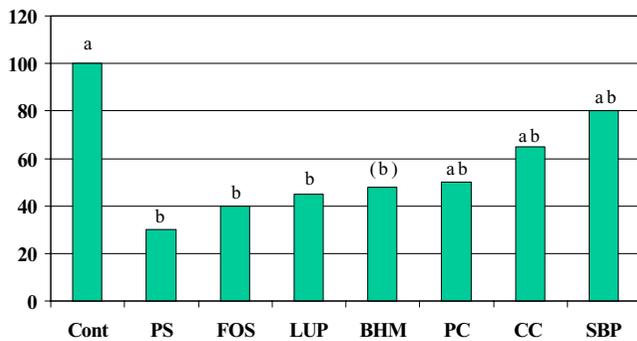
Several studies in our laboratory have shown that microbial degradation of tryptophan by pig gastrointestinal bacteria results mainly in the two volatile indolic compounds indole and skatole, skatole via a transient accumulation of indole acetic acid (IAA). Analogously with indole and skatole, indole propionic acid is also an end product of tryptophan metabolism, and has recently been shown to be produced in small amounts in the hind gut of pigs [1]. While the conversion of tryptophan to indole and IAA is performed by a variety of bacteria, only very few bacteria species are capable of further degradation of IAA to skatole [2]. The production of skatole increases through the colon and reaches a maximum in the distal part of the large intestine. From the intestinal tract, a portion of the produced skatole is absorbed to the blood and transported via the portal vein to the liver, where more than half of the amount absorbed is degraded by cytochrome P450. It has been demonstrated that the liver extraction is relatively constant within the individual animal, but varies between animals, and between sexes, showing a lower elimination rate of skatole in male pigs than female pigs [3]. Skatole avoiding degradation in the liver is deposited in peripheral tissues and due to the lipophilic characteristics of skatole the majority accumulates in the adipose tissue. To summarize, the amount of

skatole stored in adipose tissue depends on the concentration in the peripheral blood, which is influenced by the production in the large intestine and the degradation capacity of the liver. As the production of skatole is the incipient cause of tainted meat a limitation of the microbial fermentation of tryptophan in the large intestine would be an effective approach to solve the problem with boar odour caused by skatole.

### Factors affecting the level of skatole

#### Diet composition

One essential tool for manipulating the microbial activity and such the production of skatole in the gastro-intestinal tract is diet composition [4]. Several studies have provided evidence that the amount and type of protein and carbohydrate present in the feed has a substantial effect on nitrogen metabolism, thereby influencing the formation of skatole [5,6]. In accordance with this, it has been shown that diets containing a protein source with low pre-caecal digestibility stimulate skatole production [7,8] whereas diets with high content of fermentable carbohydrates that escape digestion in the small intestine have been shown to reduce the production of skatole, however, the results vary. Øverland et al., [9] and Van Oeckel et al., [10] found no effect of diets rich in sugar beet pulp, whereas Jensen et al. [8], Kjeldsen [11], Knarreborg et al. [1] and Whittington [12] observed significant reduction in skatole levels in pigs fed sugar beet pulp. Fructooligosaccharides has also been shown to reduce skatole production in both *in vitro* [13] and *in vivo* models [14,2]. Raw potato starch has also been shown to reduce the production of skatole [2,15,16]. In the study of Jensen and Jensen, [2] seven different fibre sources were investigated



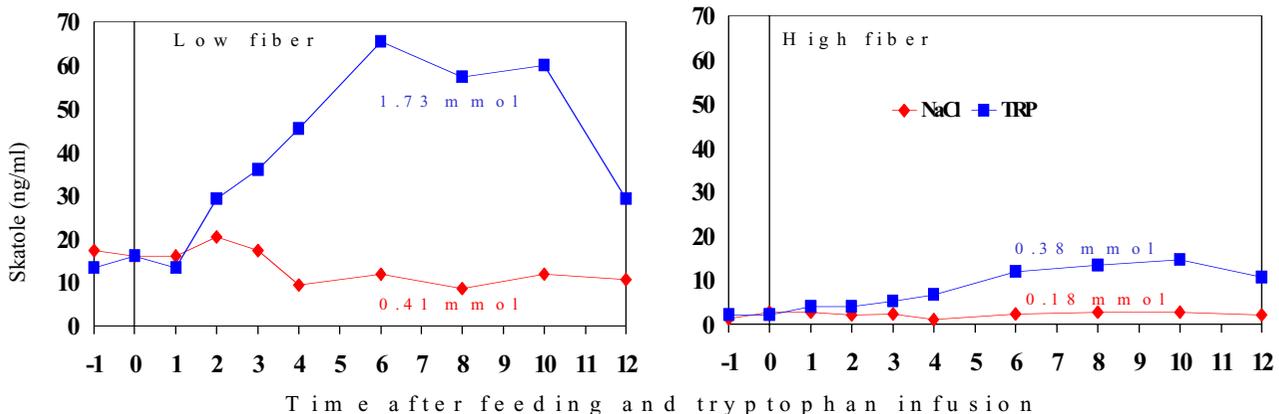
**Figure 1**  
Effect of various dietary fibre sources on the concentration of skatole in blood plasma. The diets were: a control diet based on barley and soya bean (Cont), seven diets with the same basal composition as the control, but with addition of 100 g kg<sup>-1</sup> of either raw potato starch (PS), fructooligosaccharides (FOS), lupins (LUP), barley hull meal (BHM), palm cake (PC), coconut cake (CC) or sugar beat pulp (SBP). All blood samples were taken 3 hours after the morning feeding. The control diet for each individual animal was given the value 100 and the other diets were related to this. The value for each fibre source represent three to four replicates with different animals.

for their effect to reduce skatole production. Raw potato starch, fructooligosaccharides and lupins were found to be most effective (Figure 1).

The exact mechanism of how fibre-rich diets affect skatole deposition in back fat is not known, but several contradic-

tory hypotheses have been presented. Firstly, in the presence of extra dietary fibre, more undigested protein will reach the large intestine with consequently more degradation of tryptophan to skatole. Secondly, more fermentable carbohydrates in the hindgut will increase the microbial activity in the gastrointestinal tract [17] resulting in more tryptophan incorporated as bacterial protein, further increased amounts of carbohydrates will decrease the activity of the proteolytic bacteria resulting in less tryptophan available for skatole production. Thirdly, extra dietary fibre results in more bulky material in the large intestine and an increased water binding capacity, leading to a dilution of skatole resulting in less contact of skatole with the intestinal wall and consequently, decreased skatole absorption [2]. Further, dietary fibre decreases the intestinal transit time and as such may decrease skatole absorption from the gut. Recently it has been hypotized that carbohydrates with high pre-caecal digestibility will increase cell debris formation in the small intestine, resulting in more tryptophan entering the large intestine and as such higher skatole formation [18]. On the other hand carbohydrates with low pre-caecal digestibility will decrease skatole production due to an increased butyrate production that inhibits apoptosis and as such less tryptophan available for skatole production.

That the production of skatole in the hindgut is dependent on the composition of the diet is illustrated by a series of experiments [19] where the production of skatole in the hindgut and the absorption of skatole to the portal blood was investigated after tryptophan was infused into the caecum of pigs fed either a low or a high fibre diet. Figure 2 shows the absorption pattern of skatole over a period



**Figure 2**  
Effect of dietary fibre on absorption patterns (Vp-Vj differences) of skatole to the portal blood following tryptophane (4,9 mmol) or saline infusion in the caecum. The area under curves (AUC) represent the total amount of skatole absorbed. Both diets were similar except that the high fibre diet was added 100 g sugar beet pulp per kg.

**Table 1: Influence of dietary fibre on microbial degradation of tryptophan (TRP) to indole, indole propionic acid (IPA) and skatole in the hind gut of male pigs.**

Fibre content	Indole*)		IPA*)		Skatole*)		Total indoles*)	
	Low	High	Low	High	Low	High	Low	High
Basic production in the hind gut, 12 h	960	290	10	30	480	230	1450	550
Total production following infusion of TRP, 12 h	4360	2000	370	800	1760	540	6490	3340
Production caused by infusion of TRP	3400	1710	360	770	1280	310	5040	2790
Infused TRP	4900	4900	4900	4900	4900	4900	4900	4900
Percent of infused TRP converted	69%	35%	7%	16%	26%	6%	103%	57%

\*) Values in  $\mu\text{mol}$

from 1 hour before to 12 hours after infusion of either tryptophan or saline.

Both the effect of available tryptophan and the effect of fibre on skatole are convincing. With the low fibre diet, the hindgut bacteria transform 26% of the infused tryptophan into skatole (Table 1) resulting in significant increase in skatole concentration in the portal blood (Figure 2).

Approximately 70% of the produced skatole was recovered in the portal blood. On the high fibre diet, however, only 6% of the infused tryptophan was converted to skatole in the gastrointestinal tract, resulting in a lower increase in portal blood skatole concentration. Again approximately 70% of the skatole produced in the hindgut was absorbed to the portal blood. Also the basal skatole production in the hindgut and absorption to the portal blood were affected by the fibre content in the diet. With the high fibre diet only 230  $\mu\text{mol}$  skatole was produced during the 12 hours, while more than twice as much (480  $\mu\text{mol}$ ) was produced on the low fibre diet. These results strongly show the usefulness of in vitro measurements of skatole production and the use of portal absorption to study the effect of diet on the production and absorption of microbial metabolites in the large intestine, and confirm the important effect of diet composition on the amount of skatole produced and absorbed from the gut. Further, the results point at the use of fibre rich diets as a relevant way to reduce boar taint due to skatole in practical pig production.

#### Feeding strategies

Two feeding strategies that have a marked effect on the gastrointestinal ecosystem are liquid feeding [20] and the structure of the feed (pellets vs. meal/fine vs. gorse) [21]. While use of liquid feed has been shown to reduce the level of skatole [22], it has never been investigated if the feed structure has any effect on skatole levels.

#### Feed intake (fasting)

Studies with different feeding levels during the growth period of the pigs showed that the general feed intake had no effect on the skatole level, whereas a 12 hours redraw of feed from the pigs prior to slaughter did reduce the skatole level [11]. This is in agreement with that reduced amount of digesta entering the large intestine results in a lower fermentation of non-digested protein and the very rapid degradation of skatole in the liver. However, Anderson et al. [22], were unable to confirm that a 12 hours redraw of feed reduced skatole.

#### Environmental factors

Skatole levels depends on environmental conditions [23], and it has been shown that pigs raised in a clean environment have a lower skatole level than those raised in dirty environments [24].

#### Puberty (age)

Androstenone and other testicular steroids might be involved in the skatole level by regulation of skatole metabolism in the liver [16]. Androstenone is a pheromonal steroid produced in the testes of mature male pigs together with other steroids and its levels are primarily affected by puberty stage. Using in vitro experiments Doran et al. [25] have shown that androstenone suppress the induction of enzymes involved in skatole metabolism. Zamaratskaia et al. [16] found a positive correlation between the testicular hormones testosterone and oestrone sulphate and skatole level suggesting that these compounds have an influence on skatole pattern, and the authors suggest that slaughter of entire male pigs at a weight below 100 kg can be used to avoid boar taint.

#### Genetics

There are strong indications of genetic influences on skatole levels in pigs. These include differences in skatole levels between breeds [26], significant heritability estimates for levels of skatole in fat [27] as well as indications of the presence of a major gene affecting boar taint due to skatole [28].

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