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THE EFFECT OF DIETARY FIBRE CONTENT ON SKATOLE AND INDOLE PRODUCTION IN FAECES OF IMMUNOCASTRATED MALE PIGS

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Original scientific paper

SUMMARY

The effect of dietary fibre content on intestinal production of indolic compounds was studied in immunocastrated pigs (IC). In addition, entire males (EM) and IC were compared on control diet (with low fibre content). For the study 32 crossbred pigs were assigned, within a litter to 4 treatment groups; 24 pigs were immunocastrated (at the age of 77 and 112 days) and 8 pigs were kept as entire males (EM). IC were split into three groups (IC_H, IC_M and IC_L) fed three diets differing in crude fibre (34, 60 and 80 g/kg dry matter, respectively) and net energy (NE) (10.0, 9.3, 8.5 MJ NE/kg/DM, respectively). EM were fed high NE i.e. low fibre diet. The experiment started when pigs were 84 days old and finished at the age of 172 days, when pigs were sent to slaughter. Skatole and indole concentrations were determined in the samples of intestinal content taken from caecum (CE), ascending (AC) and descending colon (DC). The concentration of indole was the highest in CE and proximal part of the colon, while skatole concentration increased in the distal parts of the large intestine. Concentrations of indolic compounds did not differ between EM and IC that were fed the same diet. Lowering dietary NE by inclusion of high fibre ingredients reduced the production of indole in the intestinal content of IC pigs, whereas the production of skatole was not affected.

Key-words: indole, skatole, dietary fibres, pig; entire males; immunocastrates

INTRODUCTION

Skatole (3-methyl-indole) is a product of bacterial degradation of tryptophan in the hind-gut of pigs, which is absorbed, and if not metabolised, deposited in the adipose tissue (Claus et al., 1994). Skatole and androstenone (testicular steroid) are held responsible for 'boar taint,'a sex-specific odour of meat from entire males (EM) (Lundström et al., 2009). It has been demonstrated that androstenone hindered skatole metabolism in the isolated pig hepatocytes (Doran et al., 2002) explaining higher skatole concentration in male pigs than in castrates. However, the levels of fat skatole can be also critically high in castrated males (Škrlep et al., 2012). Data in the literature shows that dietary factors are very important for the skatole production and that deposited skatole levels in the fat can be reduced by dietary means (Wesoly and Weiler, 2012), such as ingredients with high dietary fibres. However, the effect is not consistently shown (Aluwé et al., 2009) and it seems to depend on the level of inclusion and other dietary factors. Skatole level in adipose tissue is proportional to its production in the hind gut (Claus et al., 1993). The effect of sex on skatole production in the hind gut was not given much of attention, but higher levels of its concentration in fat of EM than castrates were mainly related to anabolic (Claus et al., 1994) or androstenone effect (Doran et al., 2002). In order to investigate the effect of increased dietary fibre content on intestinal production of indolic compounds, a feeding trial with different levels of NE in the diet achieved by different fibre content was conducted using IC pigs. Additionally, in regard to skatole production, IC pigs were also compared to EM.

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MATERIAL AND METHODS

Study is a part of bigger experiment performed in the frame of doctoral thesis of Nina Batorek Lukač made at INRA, Saint-Gilles, France in accordance with French laws on animal experimentation. Thirty-two crossbred pigs were used, 24 pigs were immunocastrated (IC) at the age of 77 and 112 days and 8 pigs were kept as EM. Pigs were assigned within litter to four treatment groups and individually housed during the experiment from 77 to 172 days of age. IC received three different diets (based on wheat, corn and barley), differing in net energy (NE) content (Table 1). Diet NE was reduced by inclusion of high fibre sources (wheat bran, soybean hulls, dried beet pulp) and by reduced quantities of added oil (Labussière et al., 2014). A high NE/low fibre diet (HNE) was given to EM (n=8) and IC from group IC_H (n=8), pigs in group IC_M (n=8) received medium NE/fibre diet (MNE) and pigs of group IC_L (n=8) received a low NE/high fibre diet (LNE, Table 1).

Table 1. Chemical composition and nutritional value of the experimental diets

	LNE	MNE	HNE
Analysed chemical composition, g/kg of DM			
Ash	60	56	54
Crude protein (N × 6.25)	180	175	185
Starch	416	470	518
Ether extract	33	34	42
Crude fibre	80	60	34
GE, MJ/kg of DM	18.17	18.17	18.31
Nutritional value[1]			
DE, MJ/kg of feed	12.70	13.28	13.87
NE, MJ/kg of feed	9.29	9.83	10.83

LNE – low net energy/high fibre diet; MNE – medium net energy/medium fibre diet; HNE – high net energy/low fibre diet; GE – gross energy; DE – digestible energy; NE – net energy; DM – dry matter

Feed and water were provided *ad libitum*. The experiment started when pigs were 84 days old and finished at the age of 172 days, when pigs were sent to slaughter. Samples of intestinal content from caecum (CE), ascending (CA) and descending colon (DC) were taken at evisceration and immediately frozen in liquid nitrogen. Skatole and indole concentrations in the intestinal samples were determined by HPLC according to the method of Denhard et al. (1991). Analysis of variance with fixed effects of treatment group and intestinal section was performed (GLM procedure of SAS Inc., Cary, USA). If significant effect ($P<0.05$) was noted, least square means between treatment groups were compared using Tukey's test.

RESULTS AND DISCUSSION

In the present study, the highest indole production was observed in CE and proximal part of the colon (CA), while skatole concentration was higher in the distal parts of the large intestine (Figure 1) implying that the main sites of indole and skatole production differ within the intestines, and agrees with previously reported data (Claus et al., 1994; Knarreborg et al., 2002). It can be also observed that indole production decreased gradually with increasing inclusion of dietary fibre (i.e. decreasing dietary NE content), whereas skatole concentration of the intestinal content was not affected by higher dietary fibre content (Figure 2). Knarreborg et al. (2002) also studied the effect of low and high non-starch polysaccharides (NSP) diet (i.e. low and high fibre diet) and reported an effect on both, skatole and indole i.e. absorption from intestines into bloodstream was lower in high NSP diet. The differences in results between their and our study might be explained with ingredients used. Namely the main source of NSP in their diet was sugar beet pulp whereas in the present one it was wheat bran and soybean hulls (see discussion later on). Secondly, in their study EM were used while in the present one, IC pigs were used. However the latter does not seem to be a major factor, and it is important to highlight, that in the present study the concentrations of indole and skatole were comparable between EM and IC fed the same dietary regime (fed high NE, low fibre diet) implying that intestinal skatole production is similar in castrated and uncastrated pigs and that the differences between these two sex categories are mainly related to differences in skatole liver metabolism (Doran et al., 2002).

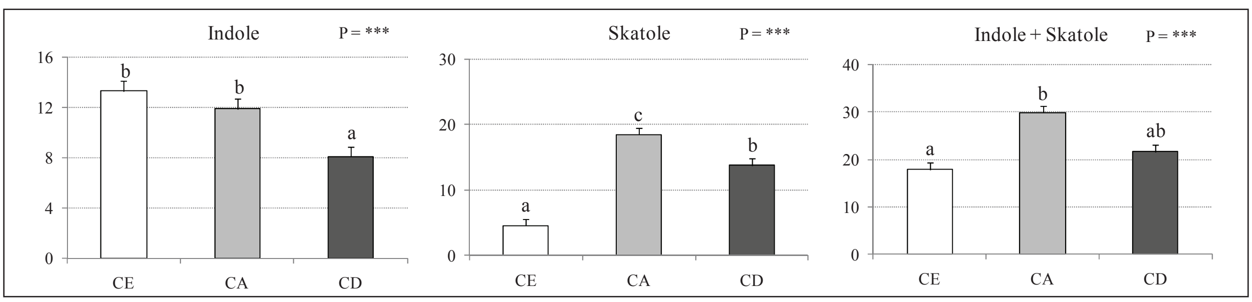

Figure 1. Intestinal concentrations of indole and skatole (μg/g, least squares means ± standard error) according to sampling locations (CE – caecum; CA – ascending colon; CE – descending colon)

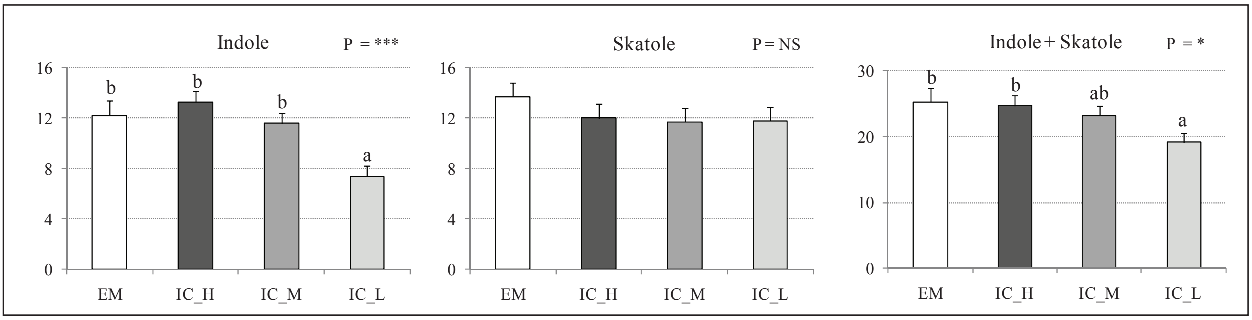

Figure 2. Overall intestinal concentrations (including caecum, ascending colon, descending colon) of indole and skatole (μg/g, least squares means ± standard error) according to treatment groups (EM – entire males, fed high energy/low fibre diet; IC_H – immunocastrates (IC) fed high energy/low fibre diet; IC_M – IC fed medium energy/medium fibre diet; IC_L – IC fed low energy/high fibre diet)

The amount of skatole stored in adipose tissue depends on the rate of its production, intestinal transit time, intestinal absorption and hepatic metabolism rate (Zamaratskaia and Squires, 2008). Generally, the production of skatole and its resorption into peripheral bloodstream is highly correlated ($r=0.79$, $P>0.001$; Claus et al., 1993) whereas skatole deposition depends strongly upon blood plasma concentrations. Therefore reducing skatole production in the intestine can be considered as an effective way for reducing skatole deposition in the adipose tissue. Skatole production can be manipulated by alterations in feed composition e.g. fermentable carbohydrates, like inulin and raw potato starch being effective in reducing skatole levels in gut and fat tissues (Kjos et al., 2010; Vhile et al., 2011). Concerning dietary fibres, as reviewed by Jensen (2006), the exact mechanism of how fibre-rich diets affect skatole deposition in back fat is not clear. The level of skatole production depends on the sufficient availability of carbohydrates (for fermentation) to supply gut bacteria with energy, reducing the skatole production (Jensen et al., 1995). Additionally, dietary fibres can reduce skatole absorption by increasing faecal wet weight and decreasing intestinal transit time (Zamaratskaia and Squires, 2009). Studies interested in the effect of NSP on skatole deposition (Knarreborg et al., 2002; Hansen et al., 2008: Aluwé et al., 2009) show variable response and imply that dietary influences should be considered from a broader perspective of diet composition/ingredients. It is possible that pH of intestinal content (which we didn't measure) varied according to the diet because dietary carbohydrates alter faecal pH and composition (Canh et al., 1998). Jensen et al. (1995) showed the importance of intestinal content pH for production of indolic compounds; with high pH favouring the production of indole and low pH favouring the production of skatole. The amount and source of fermentable carbohydrates in pig diets influence volatile fatty acid concentration and pH of faeces (Canh et al., 1998) which are key factors influencing pathways of microbial production of indolic compound (Jensen et al., 1995). In regard to indole (and sum of indole and skatole) production it is worth noting that LNE (high fibre) diet significantly reduced its production denoting a positive effect in respect of boar taint control (Figure 2). Although indole levels in fat tissue are generally low and seem unproblematic *per se*, its contribution to boar taint is important due to synergy with other compounds (Annor-Frempong et al., 1997). In general, the reduction of indole concentration in faeces in pigs fed LNE/high fibre diet can be considered as positive.

CONCLUSION

Production of indolic compounds varies along the intestinal tract; the concentration of indole was the highest in caecum and proximal part of the colon, while skatole concentration increased in the distal parts of the large intestines. Production of indolic compounds was comparable in entire and immunocastrated male pigs fed the same diet (high NE). Lowering dietary NE by inclusion of high fibre ingredients reduced the production of indole in the intestinal content of immunocastrated pigs, whereas the production of skatole was not affected. Nevertheless, in view of boar taint control, it can be considered that high fibre diet has a positive effect.

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