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Effects of dietary components including garlic on concentrations of skatole and indole in subcutaneous fat of female pigs

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ABSTRACT

The results reported here showed that threshold concentrations of skatole and indole in rice-bran oil for Singaporean consumers were 0.028 μg/g and 0.051 μg/g, respectively, and that skatole and indole levels in subcutaneous fat of pigs can be affected by diet. In Experiment A, 31 female pigs were fed with diets based on plant products only (P) or plant plus animal by-products (AP), with added levels of garlic essential oil from zero to 2.15 g/kg feed. Concentrations of skatole and indole increased with increasing garlic concentration ($P<0.001$). In Experiment B, P and AP diets were fed to 47 female pigs with different dietary lipid sources (fish oil, tallow, and a mix of linseed oil and soya oil). Skatole and indole concentrations were higher in backfat of pigs fed with the AP diet ($P<0.05$), but were unaffected by the type of lipid.

1. Introduction

Meat is an important source of nutrients, but the main reason why people eat meat is because they like its aroma, flavour and texture (Farmer, 1994). A survey of pork consumption in Singapore found flavour to be an important aspect of acceptance (Leong, Purchas, Morel, & Wilkinson, 2008a). Diets fed to pigs can produce pork flavours that are considered normal and acceptable for some consumers, but intense and undesirable for other consumers. Boar taint is considered as a classical example of an undesirable flavour in pork due in part to the presence of skatole (Hansson, Lundström, Fjelkner-Modig, & Persson, 1980; Vold, 1970; Walstra & Maarse, 1970). In Singapore, consumers often associate the presence of mutton flavour as an undesirable pork attribute that is more common in pork from western countries like Australia, Canada and New Zealand, than in pork from countries such as Indonesia and China (Leong et al., 2008a; Leong, Purchas, Morel, & Wilkinson, 2008b).

Taints and foreign-flavours in meat can be the result of dietary components that result in the generation or transfer of undesirable compounds to the product. There have been a number of studies into the feasibility of changing the flavour profile of pork by feeding pigs with different plant and animal materials. These include the direct transfer of aromatic components rosemary, oregano, ginger, garlic or chicory that influence sensory quality (Hansen, Agerhem, Rosenvold, & Jensen, 2002; Janz, Morel, Wilkinson, & Purchas, 2007). A widely studied example is the transfer of aromatic components from fish that result in a fishy flavour in pork (Jaturasitha, Wudthithumkanaporn, Rurksasen, & Kreuzer, 2002; Kjos, Skrede, & Overland, 1999; Lauridsen et al., 1999). Several studies showed feeding rape seed meal or cake of poor quality to pigs and cows has resulted in inferior odour and flavour of the cooked meat (Andersen & Sørensen, 1985; Hertzman, Goransson, & Ruderus, 1988). There have also been reports of positive effects of certain feed components such as chicory roots (Jensen & Hansen, 2006) and potato starch (Claus, Losel, Lacorn, Mentschel, & Schenkel, 2003; Zamaratskaia, Babol, Andersson, Andersson, & Lundström, 2005) on the aroma and flavour of pork due to reduction of skatole. Other compounds from the husbandry and housing system also cause off-flavours in pork products (Maw, Fowler, Hamilton, & Petchey, 2001).

Dietary aromatic components from plants may also affect the performance of pigs, as illustrated by the preference of pigs for a garlic-containing diet shown by a higher total feed intake (Janz et al., 2007). Horton, Blethen, and Prasad (1991) noted that garlic is often added to pet foods to improve palatability. Garlic has also been added to the diets of race horses to increase intake and boost performance (Horton et al., 1991). Krusinski (2001) reported a feed preference amongst finisher pigs for diets containing garlic within a herb mixture.

The objectives of this paper were, first, to establish the threshold levels of skatole and indole for Singapore consumers, and, secondly, to investigate the effects of diets, including those containing garlic, animal products, and several different lipids on the concentrations of skatole and indole in the adipose tissue of pigs.

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2. Materials and methods

2.1. Threshold test for skatole and indole

Sensory thresholds were determined by using the forced-choice ascending concentration method of limits described by the American Society for Testing and Materials, designation E679-04 (ASTM, 2004). The test aims to determine a practical value close to the threshold, based on a minimum testing effort. It helps to determine a very approximate best estimate determination threshold for each panellist. The procedure involved presentation of three samples at a time with each set consisting of two blanks and one test sample. The odd sample, was at all times the test sample under consideration. All evaluations were conducted in a sensory panel room containing 8 separated booths at 21 ± 1 °C.

The panel consisted of three Chinese males and five Chinese females between 20 and 30 years of age initially selected using British Standards Institution (BSI) (British Standards Institution, 1993) methods. They received 2 weeks of training (5 sessions) on the discrimination and recognition of skatole and indole. All the panel members had at least 2 years in panel assessment of meat and meat products.

Samples for threshold tests were prepared by spiking a neutral lipid base of rice bran oil (Tong Seng Produce Pte Ltd, Singapore) with the required concentration of either indole or skatole (Sigma-Aldrich Co, Ltd, Poole, UK). Samples of 20 mL were prepared in 50 mL amber glass screw-top bottles that were heated to 60–65 °C and held for 20 min before evaluation by panellists to ensure that sufficient of the compounds volatilised within the headspace for sniffing. For sensory threshold tests, six ascending concentrations of skatole and indole in binary steps were used (0.0125, 0.025, 0.05, 0.10, 0.20, 0.40 μg/g). The concentrations were selected based on the range of 'First Approximation' thresholds obtained from the trained panel following earlier tests.

Panellists were individually seated in well-ventilated booths and were each presented with one blind-coded, 3-AFC set of samples that consisted of one level of the test sample and two blank samples. The order of presentation of the samples within a set of three samples was randomised. In addition to determining whether there was an odd sample in the set, when an odd sample was chosen correctly, panellists were also required to give a rating on the level of difference of the odd sample from the other two based on a scale from 1 to 10 with 1 = "least different" and 10 = "most different." On each assessment day, six levels of indole or skatole were presented to the panellists in ascending order in six panel sessions separated 20 min apart. The assessments were replicated five times over a period of 5 weeks and were all carried out under red lights.

The Best Estimate Detection Threshold (BEDT) value for a panellist within a replicate was calculated as the geometric mean of the lowest concentration that was detected as being different and the next lowest concentration. The group BEDT detection values were calculated as the average geometrical means of the BEDT of the 5 replicates for each panellist and then by combining these across the 8 panellists. Reliability of BEDTs was estimated as inter-replicate correlations between individual means over the five replicates in each case.

2.2. Pig feeding experiments

The experiments were conducted in accordance with the "Massey University Code of Ethical Conduct for the Use of Live Animals for Research, Testing and Teaching" (Massey, 2008).

2.2.1. Experimental design for Experiment A

The objective of this experiment was to investigate the effects of different levels of dietary garlic essential oil (GEO) on skatole and indole concentrations in pork subcutaneous fat. GEO (product number: 31-05) was obtained from Kalsec Corporation, USA. Because garlic is one of the most popular flavoured plant materials used with pork in Singapore (Leong et al., 2008b), it was chosen to be tested for its ability to suppress undesirable mutton-like flavours in pork for the Singapore market.

The diets, animals, and experimental design were described by Leong, Morel, Purchas, and Wilkinson, 2010. In short, GEO was added directly to the diets of the 31 female pigs (Duroc × (Large White × Landrace)) grown on diets containing either animal-plus-plant products (AP diet) or plant products only (P diet) with four levels of GEO: 0, 0.55, 1.44 and 1.84 g/kg feed for the AP diet, and 0, 0.55, 1.44 and 2.15 g/kg feed for the P diet.

2.2.2. Experimental design for Experiment B

In Experiment B, the effects of dietary fish oil, tallow and plant oils with either animal plus plant components in the diet or plant components only were investigated.

Forty-seven female pigs (PIC hybrids, with a mean starting live weight 18.90 kg ± 1.75 (mean ± SD)) from a single commercial operation in the North Island of New Zealand, were assigned to one of six dietary treatment groups (as shown in Table 1 with PFS having two sub groups as explained below). They grew at a mean rate of 851 ± 56 g/day over a period of 84 days to produce an average carcass weight of 72.1 ± 4.7 kg. Growth rates and carcass weights did not differ significantly between treatments ($p > 0.05$). The pigs were kept in pens of six, but were fed individually twice daily. Water was available at all times. Individual feed intakes were measured daily and live weights recorded weekly.

In this experiment, the effects of (1) lipid type (soy bean oil, linseed oil, tallow and fish oil), (2) the period the fish oil was provided and (3) a dietary supplement (Sanovite™) containing conjugated linoleic acid (CLA), selenium, vitamin E and vitamin C on pig performance and pork quality were studied.

This experiment follows on from that reported by Janz et al., (2007) and Morel et al. (2008) in which the influence of diets supplemented with Sanovite™, with or without animal protein, on the growth performance, meat quality, and pork fatty acid profile from female pigs was studied.

The diet base was either a combination of animal and plant feedstuffs (AT and PTS), plant feedstuffs only (PO, POS) or plant feedstuffs combined with fish oil (PFS). The diets also differed depending on the presence or absence of the nutritional supplement Sanovite™ and Vitamin C. Diet POS, PTS and PFS contained Sanovite™ and Vitamin C.

The composition of the grower and finisher diets are given in Table 1.

The diets AT, PO, POS and PTS were fed over the whole experiment (84 days). The PFS was fed either between days 1 and 35 of the experiment (experimental group PFSe) or between days 36 and 56 of the experiment (experimental group PFSl). The total amount of fish oil fed per pig for groups PFSe and PFSl was the same at 2.31 kg. These two groups received the POS diet when diet PFS was not fed, which included the 28 days prior to slaughter.

2.3. Skatole and indole analysis in fat samples

Reference compounds indole, skatole, and the internal standard 2-methyindole were obtained from Sigma Aldrich, Singapore. They were of analytical reagent grade. Methanol and acetonitrile were HPLC grade (Sigma Aldrich, Singapore).

The samples were prepared as described by Tuomola, Vahva, and Kallio (1996) with minor modifications. Liquid nitrogen was used to freeze a fat sample (2.4–2.5 g) which was then crushed to powder using a pestle in a mortar (Biase, Franco, Goulart, & Antunes, 2002; Deveaud, Beauvoit, Reynaud, & Bonnet, 2007; Fraser, personal communication, 2008). The crushed sample was placed in a centrifuge