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Effect of branched-chain fatty acids, 3-methylindole and 4-methylphenol on consumer sensory scores of grilled lamb meat

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ABSTRACT

Tenderness, flavour, overall liking and odour are important components of sheepmeat eating quality. Consumer assessment of these attributes has been made for carcasses from the Information Nucleus Flock (INF) of the Co-operative Research Centre for Sheep Industry Innovation. The concentrations of three branched chain fatty acids, 4-methyloctanoic (MOA), 4-ethyloctanoic (EOA) and 4-methylnonanoic acids (compounds related to 'mutton flavour' in cooked sheepmeat) and 3-methylindole and 4-methylphenol (compounds related to 'pastoral flavour') were determined for 178 fat samples taken from INF carcasses. Statistical modelling revealed that both MOA and EOA impacted on the 'Like Smell' consumer sensory score of the cooked meat product ($P < 0.05$), with increasing concentration causing lower consumer acceptance of the product. None of the compounds though had an effect on the liking of flavour. Obviously, reducing the effect of MOA and EOA on the odour of grilled lamb will improve consumer acceptance of the cooked product but other factors affecting the eating quality also need to be considered.

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1. Introduction

Tenderness, sheep meat flavour, overall liking and cooking odour are regarded as important components of the eating quality of sheep meat (Pleasant, Thompson, & Pethick, 2005; Pethick, Hopkins, D'Souza, Thompson, & Walker, 2005b). For odour, two aromas have often been associated with cooked sheepmeat. The first aroma, generally labelled 'mutton' flavour, is usually associated with an animal's age while the second, generally described as 'pastoral' flavour, is associated with an animal's diet (Young & Braggins, 1998).

Mutton flavour, regarded as the characteristic flavour associated with the cooked meat of older sheep, becomes more pronounced as the meat is being cooked and has been cited as one of the historical reasons that sheepmeat consumption has been low in some markets (Young & Braggins, 1998). Branched chain fatty acids (BCFAs; 4-methyloctanoic (MOA), 4-ethyloctanoic (EOA) and 4-methylnonanoic acids) are the chemical compounds that are accepted as the main contributors for this flavour and research continues to explore the role of these compounds and their contribution to 'mutton' odour (Young, Berdagué, Viallon, Rousset-Akrim, & Theriez, 1997).

'Pastoral' flavour becomes evident as a result of cooking the meat of pasture fed ruminants (Young et al., 1997). In Australia, the feed for the domestic flock is pasture-based with grain feeding used in summer and autumn, depending on the availability of pasture from irrigation and the length of the dry period (Rowe, 1986; Wales, Doyle, & Pearce, 1990; McFarland, Curmow, Hyder, Ashton, & Roberts, 2006). Untrained taste panels of Australian consumers are not able to distinguish between meat product obtained from lambs finished on pasture and grain-based diets (Pethick et al., 2005a). This verifies that although pasture is the main feed material for sheep in Australia, Australian consumers are habituated to the presence of pastoral flavour in locally produced sheepmeat. 3-Methylindole, also involved with 'boar' taint in pigs, and to a lesser extent 4-methylphenol (*p*-cresol) are the main compounds implicated as contributors to 'pastoral' flavour (Young, Lane, Priolo, & Fraser, 2003).

The Co-operative Research Centre for Sheep Industry Innovation (Sheep CRC) has been conducting research aimed at understanding the links between a range of selected phenotypes and animal genetics. This work included evaluating cooked meat products, using consumer sensory panels according to Meat Standards Australia (MSA) protocols (Thompson et al., 2005a). As far as we are aware, no study has been performed which examines whether there is a relationship between the compounds responsible for 'pastoral' and 'mutton' flavours in sheepmeat (in either lamb or older animals) and consumer sensory

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attributes. The aim of this study was therefore to identify the effect of BCFAs (4-methyloctanoic (MOA), 4-ethyloctanoic (EOA) and 4-methylnonanoic (MNA) acids), 3-methylindole and 4-methylphenol measured in sheep fat on consumer sensory attributes of grilled lamb meat.

2. Materials and methods

2.1. Fat samples

The samples used were taken from lamb carcasses from the Information Nucleus Flock (INF) of the Cooperative Research Centre for Sheep Industry Innovation (Sheep CRC, Armidale, New South Wales, NSW) and the design of the INF has been presented elsewhere (Fogarty, Banks, van der Werf, Ball, & Gibson, 2007). The age of the lambs ranged from 215 to 362 d. The results presented in this paper are based on 178 samples taken from a subset of 760 animals of the 2009/2010 cohort lamb progeny, selected from the Katanning (Western Australia, WA) and Kirby (NSW) research flocks for sensory testing. A summary of the nutritional history of these animals is shown in Table 1 (Ponnampalam, Butler, Jacob, Pethick, Ball, Hocking Edwards, Geesink, & Hopkins, this issue). The lambs were slaughtered at two separate abattoirs (Tamworth, NSW and Katanning, WA) on four separate dates at Tamworth and three dates for Katanning.

At 24 h post-mortem the *longissimus thoracis et lumborum* (LTL) and *semimembranosus* (SM) muscles were excised from the carcass, and were vacuum packed and stored at 2 °C to age for 5 days. Subcutaneous fat and silver skin were removed, and 5 steaks from each muscle of 15 mm thick were cut and frozen at –20 °C for subsequent sensory testing and chemical analysis. The LTL and SM were assessed by MSA consumer panels, as described by Pannier, Pethick, Geesink, Ball, Jacob, and Gardner (this issue). Briefly, the steaks were cooked by grilling on a Silex S-165 clam shell grill unit (Silex Grills Australia Pty Ltd, Marrickville, NSW, Australia) set at 220–230 °C. The cooking was controlled by a timer to produce a constant medium degree of doneness (internal temperature of about 65°) and then rested for 2 min prior to tasting (Thompson et al., 2005b). The MSA testing panels consisted of untrained consumers who were familiar with sheepmeat and consumed a meal of cooked meat at least once per fortnight. Details on recruitment of the consumers are given elsewhere (Thompson et al., 2005b). The untrained consumers were used to assess the steaks for tenderness, juiciness, liking of the flavour ('Like Flavour'), liking of the smell ('Like Smell') and overall liking ('Overall Like') based on a 1 to 100 score. Consumers also graded the samples into the following categories; unsatisfactory, good every day (3 star), better than every day (4 star), or premium (5 star). Every muscle was tasted 10 times by 6 different consumers, and the individual consumer scores with the mean score of the 10 consumer scores per sample were recorded. There was a total of 43 sampling sessions, with 60 consumers per session, which assessed the grilled meat. The mean of the 'Like Flavour' and 'Like Smell' consumer scores for each sample was used for the subsequent statistical analysis. The associated subcutaneous fat samples (20 g), taken from over the *gluteus medius* muscle site at 1 h post-slaughter, were stored at –20 °C. The samples were transported respectively from NSW and WA to Werribee (Victoria) at –20 °C for chemical analysis. The samples were kept at this temperature until required for

analysis. The cohort of 178 fat samples were selected to be representative of the range of the mean consumer flavour scores of LTL, according to sire type (Terminal ($n = 122$), Maternal ($n = 31$) or Merino ($n = 25$)) and production site (Kirby ($n = 89$) and Katanning ($n = 89$)). The mean hot carcass weight was 24.7 ± 0.3 (standard error) kg while the mean GR fat depth was 16.5 ± 0.5 mm.

2.2. Chemicals

4-Methyloctanoic (MOA), 4-methylnonanoic (MNA), 4-ethyloctanoic (EOA) and undecanoic acids as well as 4-methylphenol (MP) and 3-methylindole (MI) were purchased from Sigma-Aldrich (Castle Hill, NSW, Australia) and used without purification. Divinylbenzene/Carboxen®/polydimethylsiloxane (DVB/Car/PDMS) solid phase micro-extraction (SPME) fibres were obtained from Sigma-Aldrich. The SPME fibres were pre-conditioned at 280 °C for 90 min. Solvents used were of pesticide grade quality. Nitrogen and helium were ultra-high purity grade (Coregas, Altona, Vic., Australia). All other reagents were of analytical reagent grade.

2.3. Measurement of branched chain fatty acids

The fat samples were wholly melted by heating 4×5 g portions between 3 and 5 min (sufficient to melt but not cook the fat) in a domestic microwave oven, ensuring homogeneity of the sample. A sample of the liquid fat (1 g) was injected into a Unitrex sweep co-distillation unit (SGE, Ringwood, Vic.) and heated at 200 °C for 1 h under a flow (200 mL min^{-1}) of nitrogen. Each batch of ten samples included one spiked recovery fat sample containing the internal standard, undecanoic acid (C_{11} FA, $1.00 \mu\text{g mL}^{-1}$). The released compounds were purged through the Unitrex unit and collected onto a trap. The trap, consisting of Tenax®, a glass wool plug and sodium sulphate, was eluted with 5 mL diethyl ether:hexane (20:80). The organic phase was concentrated to 1 mL and, after the addition of the internal standard ($1.00 \mu\text{g mL}^{-1}$), the sample was treated with (*N,O*)-bisilyltrifluoroacetamide at 60 °C for 30 min and the free fatty acids (including BCFAs) were derivatised as the trimethylsilyl (TMS) esters.

The fatty acid-TMS esters were separated by injection (1 μL) onto a DB5-MS fused silica capillary column (J&W, $30 \text{ m} \times 0.25 \text{ mm i.d.} \times 2.5 \mu\text{m}$ film thickness) in a Varian 3400 gas chromatograph (GC) and detected by a Saturn 2000 ion trap mass spectrometer (MS) operating in full scan mode. The septumless programmable injector (SPI) was programmed starting at 45 °C and increased to 325 °C at a rate of $180 \text{ }^\circ\text{C min}^{-1}$. The GC oven was held at 75 °C for 2 min then increased to 300 °C at a rate of $10 \text{ }^\circ\text{C min}^{-1}$ and held at this temperature for 8 min. Helium was used as the carrier gas at a constant pressure of 105 kPa. The MS transfer line was held at 280 °C. Mass spectra were acquired using an ion source temperature of 220 °C and an electron multiplier voltage of 2400 V. The MS was calibrated using FC43 (Varian, Inc., Springvale, Vic.).

Quantitation of the BCFAs was performed using the Varian Saturn Workstation 2000 software. For calibration, the standards were in the range of 0.02 to $1.00 \mu\text{g mL}^{-1}$ (or mg kg^{-1} effective concentration in sheep fat) and the standard solutions were similarly derivatised using (*N,O*)-bisilyltrifluoroacetamide at 60 °C for 30 min. The following ions were used for quantitation; MOA-TMS ester, $m/z = 215.0$, EOA-TMS

Table 1
Summary of nutritional history of 2009/2010 lamb progeny used in this study^a.

Site	Early post weaning		Late post weaning	
Katanning	Pasture	Concentrate	Pasture	Concentrate
	Green annual grass and subclover	60:40 Lupins and oats supplementary fed in lick feeders	Dried senesced pasture, annual grass and subclover	60:40 Lupins and oats
Kirby	Improved pasture	Lupin	Grazing oats	Supplementary fed in lick feeders Prime lamb finisher

^a Ponnampalam et al., this issue.

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