Final report

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Prepared by:	**Robyn Warner and Peter Watkins**
	CSIRO Division of Food & Nutritional Sciences
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Meat & Livestock Australia Limited
Locked Bag 991
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Effects of Feeding Regimes on Lamb Meat Flavour – Sample Collection and Processing

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Abstract
Samples of loin and topside were obtained from 65 carcasses, derived from lambs grazing on feeding regimes of 100% plantain, lucerne and ryegrass. The samples are presently in storage for MSA consumer panel and trained sensory analysis as well as chemical analysis of branched chain fatty acids (BCFAs, 4-methyloctanoic, 4-ethyloctanoic and 4-methylnonanoic acids), aldehydes and ketones, and the compounds responsible for pastoral flavour in sheep meat (*p*-cresol and 3-methylindole).

Project objectives
To collect samples of loin and topside from 108 lambs, and arrange sampling, cut-up and storage of muscles for future analysis.
The samples collected will be for;

- MSA consumer panel analysis,
- trained sensory analysis and
- chemical analysis of BCFAs (4-methyloctanoic, 4-ethyloctanoic and 4-methylnonanoic acids), volatile components (aldehydes and ketones) and pastoral flavours (*p*ara-cresol and 3-methylindole).

Success in achieving milestone
The samples were successfully collected for the MSA consumer panel and trained sensory analysis, and chemical analysis of BCFAs, *p*-cresol and 3-methylindole. There was some problems with number of animals, misallocation of animals for slaughter and an insufficient number of samples from each carcass. It is believed that this will not impact detrimentally on the outcomes of the study.

Details
The experimental design involved seven treatments which were applied across 28 paddocks, with four replicate paddocks for each grazing treatment and 9-12 animals allocated to each replicate paddock. We had originally proposed to sample 108 animals for the monoculture grazing treatments (100% lucerne, 100% plantain, 100% ryegrass). The 9-12 animals per paddock included ewes and wethers. Ralph Behrendt (DPI-Vic) needed the ewes for another independent experiment. This meant that only 69 wethers were left for sampling of the monoculture treatments across the four replicate paddocks. The day prior to slaughter, the animals were removed from the paddocks and grouped into two mobs; one containing the 69 animals from the monoculture treatments, *viz.* 100% plantain, lucerne and ryegrass and the other containing the remaining animals from other grazing treatments. All of the animals were transported from DPIV-Hamilton and delivered to CRF-Colac on the afternoon of Tuesday, June 7 2011.

The animals were slaughtered on the CRF premises and chilled on Wednesday with boning of the carcasses performed on Thursday. Collection of the samples was performed by DPI-Vic. staff, contracted by CSIRO to

perform this task. A CSIRO technician also came down from Brisbane, to assist with the boning, as DPI-Vic. needed additional staff. Furthermore, since the meat was destined for human consumption, CSIRO procedures require that the cold chain is maintained for shipment of the meat product , thusrequiring refrigerated transport The CSIRO technician was able to fulfil this duty. Striploins and topsides were obtained from both sides of each of the 69 carcasses. On the day of slaughter, fat samples, from over the rump site, were also collected from each carcass, for measurement of branched chain fatty acid (BCFA) concentrations (compounds responsible for "mutton" flavour) and *p*-cresol and 3-methylindole (compounds responsible for "pastoral" flavour).

After discussion with the sensory scientists in CFNS, North Ryde, the number of samples required for training of, and analysis by, the trained sensory panel was determined. It was also decided that the sample and cooking protocols used for the MSA consumer panel would be used for the meat presented to the trained sensory panel. Thus Alan Gee was asked to prepare all samples for the trained sensory panel and the MSA consumer panel. Ultimately, samples from 36 carcasses would be presented to the trained sensory panel, for which 20 samples weighing >35g would be required from each carcass. For the training, a similar number of samples would be required from each carcass, because samples displaying the typical range in flavour to that ultimately consumed are required for training. Thus it was calculated that we would require all of the striploins from both sides of the 68 carcasses.

Subsequently, Eric Ponnampalam (from DPIV) informed us that he required some samples from the striploin, in order to conduct retail colour shelf-life studies as well as measurements of IMF, Vitamin E and LCFA's. There was also discussion between MLA and Matt McDonagh, at DPIV. Matt McDonagh indicated that he wanted to measure the eye muscle area and fat depth at the $12^{th}/13^{th}$ rib and, at a later stage, also indicated he wanted to take samples for histochemistry and fibre typing. This meant that not all striploin was available from each carcass for sensory sampling, and that less sample was available for consumer and trained sensory analysis than desirable. Furthermore, when the ear tag identification numbers had been checked, two days after slaughter, it was found that all animals from one replicate paddock of the 100% lucerne treatment were missing. This had been replaced by a 50%ryegrass:50% lucerne treatment. Evidently, an error had occurred when the sheep were collected from the paddock.

On the boning day, Matt McDonagh made a cut into one side of the carcass, in order to conduct his EMA and fat depth measurements and histochemistry samples. This left two sections that CSIRO could use on this side of the carcass. These were meant to be 30 and 10 cm, yet were in fact <26 cm and <10 cm in length. From the other side of the carcass, we had hoped to get 40 cm of striploin, but each of these were < 40 cm in length. The '40 cm' and '30 cm' sections of striploin were despatched to Alan Gee, at Cosign in Coffs

Harbour, where the samples were prepared from 36 carcasses (12 per treatment x 3 treatments) for MSA consumer panel. The remainder were prepared for the training of, and testing by, the trained sensory panel at CFNS, North Ryde. Table 1 shows the samples which have been collected and their intended purpose, i.e. for chemical measurement, or use with the trained sensory or MSA consumer panel. The remaining 10 cm of striploin is being held in reserve at CSIRO-Werribee (at-20 °C), if needed, for the training of the trained sensory panel. Otherwise it will be used for chemistry analysis, along with the topside samples. Table 2 shows the number of samples that Alan Gee and his team were able to obtain from each carcass, for consumer and trained sensory panels.

Impact of the reduction in carcass numbers from 108 to 69 - For the trained sensory panel, a labour intensive exercise, it was intended that only samples from 36 carcasses would be used. However, for the MSA consumer panel, a reduction in the number of samples could impact on the significance of the final statistical analysis.. For the chemical analysis, a sample size of 69 carcasses will be sufficient for statistical significance . For the purposes of statistical analysis, the experimental unit is the paddock, as opposed to the individual animal that is in the paddock. Thus the impact of less animals per paddock than originally proposed, on the statistical significance of the outcomes, will be less than in the case if the carcass/animal were the experimental unit. This is demonstrated in Table 3, using livewight gain as an example, where the effect of changing from 7 animals per plot (paddock) to 4 animals per plot is shown. For 4 animals per plot, there is an 80% chance of finding a difference of 22.5% or more in two treatment means. If the number of animals per plot is changed to 7, there is an 80% chance of finding a difference of 20.0% or more in two treatment means, viz. not a big difference.

Table 1: Samples in storage for chemical and sensory testing

Samples	Assay/Measurement	Storage and location
69 fat samples	- branch chain fatty acids - pastoral flavour	-20 °C at CFNS Werribee
5 steaks from 36 carcasses	MSA consumer panel	-20 °C, stored at Cosign
5-15 steaks from 68 carcasses	Training and conduct of trained sensory panel	-20 °C, stored at Cosign
<10 cm striploin	Chemistry	-20 °C at CFNS Werribee
Topside samples	Chemistry	-20 °C at CFNS Werribee

Table 2: List of samples obtained by Alan Gee for consumer (Cons Obt) and trained sensory (Sen obt) panel.

hook #	PLOT	TREAT	REP	CONS PANEL	Cons Obt	Sensory panel	Sens obt	Arrived
1	6	2	1	Yes	5	Yes	9	y
2	20	3	3	Yes	5	Yes	8	y
3	13	1	2	Yes	5	Yes	6	Y
4	22	1	4			Yes	12	y
5	22	1	4	Yes	5	Yes	7	y
6	24	2	4	Yes	5	Yes	9	y
7	9	2	2	Yes	5	Yes	7	y
8	26	3	4	Yes	5	Yes	7	y
9	20	3	3	Yes	5	Yes	7	y
10	11	3	2			Yes	13	y
11	22	1	4			Yes	13	Y
12	13	1	2		5	Yes	6	Y
13	11	3	2	Yes	5	Yes	6	y
14	18	2	3	Yes	5	Yes	8	y
15	11	3	2	Yes	5	Yes	10	y
16	2	3	1	Yes	5	Yes	7	y
17	18	2	3			Yes	13	y
18	2	3	1			Yes	13	y
19	26	3	4			Yes	13	y
20	17	4	3			Yes	12	y
21	17	**4**	3			Yes	16	y
22	22	1	4			Yes	12	Y
23	26	3	4			Yes	12	y
24	18	2	3	Yes	5	Yes	6	y
25	22	1	4	Yes	5	Yes	8	y
26	24	2	4	Yes	5	Yes	8	y
27	26	3	4	Yes	5	Yes	8	y
28	2	3	1	Yes	5	Yes	8	y
29	24	2	4			Yes	14	y
30	6	2	1	Yes	5	Yes	7	y
31	26	3	4	Yes	5	Yes	6	y
32	11	3	2			Yes	12	y
33	13	1	2	Yes	5	Yes	7	Y
34	13	1	2	Yes	5	Yes	8	Y
35	24	2	4			Yes	14	y
36	9	2	2	Yes	5	Yes	6	y
37	22	1	4	Yes	5	Yes	6	y
38	17	4	3			Yes	14	y
39	4	1	1	Yes	5	Yes	8	Y

hook #	PLOT	TREAT	REP	CONS PANEL	Cons Obt	Sensory panel	Sens obt	Arrived
40	26	3	4			Yes	11	Y
41	20	3	3	Yes	5	Yes	7	y
42	22	1	4			Yes	12	y
43	24	2	4			Yes	12	y
44	26	3	4		5	Yes	8	y
45	22	1	4	Yes	5	Yes	7	y
46	6	2	1	Yes	5	Yes	6	y
47	24	2	4			Yes	13	y
48	6	2	1			Yes	13	y
49	13	1	2	Yes	5	Yes	7	Y
50	20	3	3			Yes	13	y
51	20	3	3	Yes	5	Yes	7	y
52	4	1	1	Yes	5	Yes	8	Y
53	18	2	3			Yes	15	y
54	13	1	2			Yes	12	Y
55	24	2	4			Yes	12	y
56	22	1	4			Yes	12	y
57	4	1	1	Yes	5	Yes	7	Y
58	9	2	2			Yes	13	y
59	2	3	1			Yes	11	y
60	17	4	3			Yes	12	y
61	18	2	3	Yes	5	Yes	6	y
62	9	2	2			Yes	12	y
63	4	1	1	Yes	0	Yes	0	Missing
64	11	3	2			Yes	15	y
65	9	2	2	Yes	5	Yes	7	y
66	6	2	1			Yes	12	y
67	24	2	4	Yes	5	Yes	6	y
68	17	4	3			Yes	15	y
69	Not in List						11	y
170	2	3	1	Yes	0	Yes	0	Missing
170	2	3	1		0	Yes	0	Missing

Table 3: Using live weight gain as an example, the statistical effect of changing from 7 animals per plot to 4 animals per plot

Example 1: 4 reps, 4 animals per plot		**Example 1: 4 reps, 5 animals per plot**	
Grand mean	263	Grand mean	263
Variance components		Variance components	
Rep	256	Rep	256
Plot	392	Plot	392
animals within plots	1555	animals within plots	1555
number of reps	4	number of reps	4
number of animals per plot	4	number of animals per plot	5
comparing treatment main affects means		comparing treatment main affects means	
the between plot mean square	3123.0	the between plot mean square	3515.0
s.e.d. of comparing treaments		s.e.d. of comparing treaments	
variance of plot mean (s^2)	780.8	variance of plot mean (s^2)	703.0
50% power d^2	1561.5	50% power d^2	1406.0
80% power d^2	3513.4	80% power d^2	3163.5
50% power d	39.52	50% power d	37.50
80% power d	59.27	80% power d	56.24
% difference of grand mean 50% power	15.0	% difference of grand mean 50% power	14.3
% difference of grand mean 80% power	22.5	% difference of grand mean 80% power	21.4
Example 3: 4 reps, 6 animals per plot		**Example 4: 4 reps, 7 animals per plot**	
Grand mean	263	Grand mean	263
Variance components		Variance components	
Rep	256	Rep	256
Plot	392	Plot	392
animals within plots	1555	animals within plots	1555
number of reps	4	number of reps	4
number of animals per plot	6	number of animals per plot	7
comparing treatment main affects means		comparing treatment main affects means	
the between plot mean square	3907.0	the between plot mean square	4299.0
s.e.d. of comparing treaments		s.e.d. of comparing treaments	
variance of plot mean (s^2)	651.2	variance of plot mean (s^2)	614.1
50% power d^2	1302.3	50% power d^2	1228.3
80% power d^2	2930.3	80% power d^2	2763.6
50% power d	36.09	50% power d	35.05
80% power d	54.13	80% power d	52.57
% difference of grand mean 50% power	13.7	% difference of grand mean 50% power	13.3
% difference of grand mean 80% power	20.6	% difference of grand mean 80% power	20.0