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Effect of lairage timing & duration on rumen physiology and muscle glycogen

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Executive summary

Dark cutting is a significant issue for the Australian feedlot industry. The depletion of muscle glycogen reserves due to reduced dietary glucose precursors and/or physiological stress are the primary factors associated with dark cutting. It has been commonly accepted that feedlot cattle consuming high energy diets receive adequate nutrition to ensure muscle glycogen concentration is maximised prior to dispatch from the feedlot. However, dark cutting remains a significant issue in feedlot cattle in Australia. The aim of the present study was to determine the optimal timing and duration of lairage to reduce the incidence of dark cutting in Australian feedlot cattle.

To determine the effects of timing and duration of lairage on rumen physiology, muscle glycogen, and carcass traits of feedlot cattle, a 2 x 2 factorial design was used to compare two dispatch times from the feedlot (afternoon prior to slaughter and morning of slaughter), and two slaughter times (morning slaughter and afternoon slaughter). British-type heifers (n=400) sourced to target the domestic market with an average entry weight of 379.8 kg were fed for 63 to 65 days, gained on average 2.17 kg/d, and dressed at 54.18%, yielding carcasses of 273.1 ± 25.0 kg with 7.1 ± 2.0 mm rib fat. To understand the effects of transportation and lairage on rumen physiology, a subset of cattle (n=120) received reticulorumen boluses to measure the pH and temperature of the rumen. Cattle from a single home pen were randomly allocated to four treatment pens and pens were randomly allocated to treatments. The four treatments included 1) AM ship, AM slaughter; 2) AM ship, PM slaughter; 3) PM ship, AM slaughter; and 4)PM ship, PM slaughter. Two replicates were completed. Full transportation and lairage audits were conducted for every pen of cattle. Hot carcass weight, dressing percent, eye muscle area, rib fat, P8 fat, hump height, chiller assessment pH, Aus-Meat meat colour, objective Hunter colorimeter scores, were recorded for all carcasses. Intramuscular glycogen, rumen pH, and rumen temperature were measured for a subset of the cattle (n=120).

For the complete data set of 400 heifers, cattle slaughtered in the morning realised a 2.4 kg advantage ($P < 0.01$) in hot carcass weight and a 0.5% advantage ($P < 0.01$) in dressing percent as compared to cattle slaughtered in the afternoon. Cattle slaughtered in the morning had less carcass shrink and thus produced heavier carcasses and higher dressing percentages. We hypothesize this is due to reduced tissue shrink and greater muscle glycogen, as intramuscular glycogen levels for bolused heifers were 0.15% higher ($P < 0.01$) for cattle slaughtered in the morning as compared to the afternoon.

For the complete data set of 400 heifers, chiller assessment pH of cattle slaughtered in the morning was 0.03 pH units lower ($P < 0.01$) than cattle slaughtered in the afternoon. Aus-Meat meat colour of cattle slaughtered in the morning was 0.23 colour scores lower on average than cattle slaughtered in the afternoon. Specifically, a shipping time by slaughter time interaction was detected ($P < 0.01$). Cattle shipped the morning of slaughter and slaughtered that afternoon displayed the highest ($P < 0.05$) chiller assessment pH and darkest ($P < 0.05$) meat colour as compared to all other treatments.

In regard to time off feed prior to slaughter, bolused heifers that were dispatched the morning of slaughter had 0.15% higher ($P < 0.01$) muscle glycogen levels as compared to cattle shipped the afternoon prior to slaughter. Interestingly, cattle that were shorter time off feed also had higher ($P < 0.01$) rumen temperature, due to metabolic heat production, and lower rumen pH, due to acid

production by ruminal microbes. These results suggest that cattle that are shorter off feed continue to produce glucose precursors required for glycogen synthesis in the hours leading up to slaughter. This coupled with a reduction in the mobilization of glycogen stores prior to slaughter results in higher muscle glycogen levels.

Results suggest that a reduction in the period of time off feed prior to slaughter, reduction in the duration of lairage, and an increase in the duration of chilling reduces the incidence of dark cutting, increases hot carcass weight, increases dressing percent, improves meat colour, and increases muscle glycogen levels. Through cross-sector discussions of these results, the red meat industry has the opportunity to drive change in the areas of transportation, lairage, and chilling to achieve optimal beef yield and quality.

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1 Background

Meat colour influences purchasing decisions and acceptance of fresh beef at the retail level (Hughes et al., 2014). Problems with meat colour, specifically dark cutting, are a major concern for the beef industry (Lu et al., 2018, Voisinet et al., 1997). Dark cutting beef is often discounted at the retail level (Janloo, 1999). Hence, there is a need to reduce the incidence of dark cutting beef carcasses (Hughes et al., 2014). To reduce the incidence of dark cutting, we must understand the factors that lead to this condition and scientifically test interventions that may reduce the incidence of dark cutting.

Reduced antemortem levels of muscle glycogen lead to reduced postmortem lactic acid accumulation and impede pH decline following death (Ashmore et al., 1973, Scanga et al., 1998). This results in beef with an elevated ultimate pH, increased light absorption, and increased water-binding ability that appears dark, firm, and dry (Tarrant and Sherington, 1980, Scanga et al., 1998).

It is commonly accepted that long-term physiological stressors prior to slaughter reduce and can deplete muscle glycogen levels resulting in dark cutting beef with elevated ultimate pH (Ashmore et al., 1973). Low levels of muscle glycogen are typically associated with stress from mustering, transportation, and slaughter which increase adrenaline levels, mobilizing and reducing levels of muscle glycogen (Chulayo et al., 2016, Hughes et al., 2014, Knee et al., 2007). However, dark cutting is an extremely complex multifactorial condition and the exact factors that lead to the development of this condition are not well understood (Scanga et al., 1998, Voisinet et al., 1997).

Reduced dietary glucose due to fasting during transportation and lairage can also lead to reduced levels of muscle glycogen (Tarrant and Sherington, 1980). Knee et al. (2007) found that feed quality and energy levels of the ration have the potential to increase muscle glycogen levels and reduce the incidence of dark cutting beef in pasture-fed cattle. It has been commonly accepted that feedlot cattle consuming high energy diets and gaining a minimum of 0.8 – 1.0 kg/d receive adequate nutrition to ensure muscle glycogen concentration is maximised prior to dispatch from the feedlot (Pethick et al., 2000). However, dark cutting remains a significant issue in feedlot cattle in Australia and thus further research is required to minimise the incidence of dark cutting. In feedlot cattle in Australia, there appears to be a seasonal elevation in the incidence of dark cutting during the summer heat stress period and early autumn when large variations in temperature occur in short periods of time leading up to slaughter.

Although muscle glycogen concentration is maximised prior to departure from the feedlot, the period of time from feedlot dispatch to slaughter may impact muscle glycogen concentration at slaughter. Cattle are not typically fed during transport and lairage and therefore it is expected that muscle glycogen concentration will decline throughout this period. As the period of time an animal remains off feed increases, the level of substrate available for microbial fermentation and subsequent glycogen synthesis decreases.

An increased incidence of dark-cutting beef has been associated with extended duration of transportation and lairage (Janloo, 1999). Transportation, handling, and lairage of cattle prior to slaughter can be stressful and contribute to the depletion of muscle glycogen (Giannetto et al., 2011, Romero et al., 2013). The environmental conditions of lairage may also impact the incidence of dark cutting. For example, in feedlots, the incidence of heifers that produce dark cutting beef is lower in heifers housed in shaded pens as compared to unshaded pens (Mitlöhner et al., 2002). Cattle

subjected to an extended duration of lairage such as cattle that are held in lairage over the weekend for 36-84 h exhibited a higher incidence of dark cutting as compared to a typical duration of lairage of 12 h (Kreikemeier et al., 1998). Under Canadian commercial conditions, the incidence of dark cutting increased as the distance between the farm and slaughter plant increased (Jones and Tong, 1989). The effects of duration of transportation feed withdrawal (1 h vs 11 h) on carcass quality in Holstein Friesian calves were studied and found that an extended duration of transportation resulted in higher muscle pH 4h following slaughter (Fernandez et al., 1996).

However, Ferguson et al. (2007) found no difference in carcass traits (carcass weight, muscle glycogen concentration, pH 3 h post-slaughter, ultimate pH, and Minolta lightness objective meat colour) when comparing lairage times of 3 h vs 16 h for steers transported 150 km to slaughter. Fasting times were 14 h and 27 h, respectively. This study was conducted in April and May and meat quality (pH, temperature, muscle glycogen, ultimate pH, Minolta colour) was only evaluated on 15 animals per treatment (Ferguson et al., 2007). Thus, the climatic conditions were not typically associated with dark cutting and the sample size was small for a condition that is quite rare.

A primary objective of this work is to determine how the period of time off feed impacts ruminal pH which is associated with the level of substrate available for fermentation. During fermentation, ruminal microbes produce volatile fatty acids and other acids which reduce the pH of the rumen. When substrate for ruminal fermentation is reduced, speed of fermentation is reduced and subsequently less acid is produced. Therefore, it is expected that ruminal pH would be higher when substrate for ruminal fermentation is reduced, such as extended periods of time witout access to high energy feed. We hypothesized that cattle exposed to longer periods of transport and lairage will have higher ruminal pH, higher ultimate pH, and darker meat colour.

Physiological stressors also mobilise muscle glycogen and can have negative imacts on meat colour and ultimate pH. Transportation and lairage are stressful events for cattle due to changes in environmental conditions such as temperature, access to shade, animal handling, and comingling with other cattle. Lairage can be particularly stressful during summer months where heat stress is a major concern, particularly in environments without shade. This project aimed to assess the impact of lairage conditions and transportation on ruminal temperature as a measurement of core body temperature. We hypothesize that transportation and lairage will increase ruminal temperature in comparison to cattle remaining in the home pen. We hypothesized that cattle that experience extended durations of transportation and lairage will display elevated ruminal temperature as compared to cattle exposed to short durations of transport and lairage. Elevated ruminal temperature may be associated with darker meat colour, reduced muscle glycogen concentration, and higher ultimate pH.

2 Project objectives

This project aimed to determine the effect of time of dispatch (AM vs PM ship) from the feedlot, specifically morning or evening transportation, on rumen physiology, muscle glycogen levels at slaughter, and carcass characteristics.

Further, this project was designed to determine the effect of duration and timing of lairage (AM vs PM slaughter) at the abattoir on rumen physiology, muscle glycogen levels at slaughter, and carcass characteristics.

3 Methodology

3.1 Animal welfare

Animals were housed and used in accordance with the animal welfare principles established and maintained by Bovine Dynamics Pty Ltd.

3.2 Experimental design

Two replicates of a 2 x 2 factorial design evaluated the effects of time of dispatch (morning vs. evening transportation from feedlot to abattoir) and time of slaughter (morning vs. afternoon slaughter) on rumen physiology, muscle glycogen levels at slaughter, and carcass characteristics.

Treatment	Feedlot dispatch	Slaughter	Number of cattle
PMAM	6:00 p.m. Day Prior to Slaughter	9:00 a.m.	n_{total}=99, n_{Rep1}=47, n_{Rep2}=52
PMPM	6:00 p.m. Day Prior to Slaughter	3:00 p.m.	n_{total}=100, n_{Rep1}=49, n_{Rep2}=51
AMAM	7:00 a.m. Day of Slaughter	9:00 a.m.	n_{total}=101, n_{Rep1}=50, n_{Rep2}=51
AMPM	7:00 a.m. Day of Slaughter	3:00 p.m.	n_{total}=100, n_{Rep1}=49, n_{Rep2}=51

3.3 General

The experiment was conducted at a commercial feedlot located in Northern New South Wales, Australia. Replicate 1 was conducted from the 15th to the 22nd of February 2018. Replicate 2 was conducted from the 15th to the 22nd of March 2018.

At the feedlot, cloth shaded experimental pens with compacted gravel in-situ floors housed cattle at a stocking density of approximately 16.7 m^2 per animal. Four pens with an area of 836 m^2 were used. The concrete feed bunks were 22 metres in length, allowing for approximately 44 cm of bunk space per animal. Water troughs, with float activated water supplies, were 3 metres in length and shared between two pens. Fences were metal pipe with steel cabling.

Replicate 1 included 195 Angus heifers. Replicate 2 included 205 British type heifers. At the time of induction to the feedlot, heifers were implanted with 20 mg oestradiol benzoate and 200 mg testosterone propionate, vaccinated against Infectious Bovine Rhinotracheitis and *Mannheimia haemolytica*, and dewormed with an injectable abamectin. Heifers were fed a high energy steam-flaked wheat and barley finisher ration that exceeded NRC 2016 requirements for beef cattle (National Academies of Sciences and Medicine, 2016).

Seven days prior to slaughter, cattle (n_{total}=400, n_{Rep1}195, n_{Rep2}=205) were randomly drafted from their feedlot home pen into four experimental pens of equal dimensions as described above.

Importantly, cattle were not mixed with new cattle at this time. A random number generator was used to assign experimental pens to treatments.

At the time of drafting, 60 animals per replicate (n=15 per pen) received reticulorumen boluses that measure pH, temperature, and activity levels every 10 minutes. For replicate 1, 60 Angus heifers ranging in weight from 483-540 kg received boluses. For replicate 2, 60 British heifers (9 Angus and 6 Murray Grey heifers per pen) ranging in weight from 490-579 kg received boluses.

Cattle were fed ad libitum with feed deliveries at 10:30 a.m. and 1:00 p.m.

Each pen of cattle was transported from the feedlot to a large commercial abattoir in a separate "contractor approved" B-Double combination truck with air bags with 75 head capacity. Loading, dispatch, transport, unloading, and penning of cattle were audited. The distance of transportation was 75 km and the duration of transportation was approximately 90 minutes.

At the abattoir, unshaded lairage pens with dirt floors housed cattle at a stocking density of approximately 3.4 m^2 per animal. Cattle were not provided feed or hay during transportation and lairage. Water troughs, with float activated water supplies, were 1.8 metres in length and shared between two pens. Cattle received a gentle belly wash of non-potable water from spray nozzles on the floor of concrete-floored abattoir holding pens for 30 minutes approximately 2 hours prior to slaughter. Fifteen minutes prior to slaughter, cattle received an additional potable water wash delivered from overhead in a holding yard for approximately 30 seconds.

Cattle were stunned with a non-penetrative stunner and processed according to industry standards. Immediately following exsanguination via severance of the carotid artery and jugular vein, all cattle were electrically stimulated to facilitate further exacerbate exsanguination. Electronic radio frequency identification, time of stun, visual identification, and body number were recorded. Reticulorumen boluses were collected during evisceration. A rigidity probe was inserted into the loin to provide electrical stimulation to facilitate hide removal.

3.4 Rumen Physiology Measurements

Internal, wireless reticulorumen boluses which monitor pH, temperature, and activity levels with a measurement interval of 10 minutes (smaXtec Animal Care GmbH, Graz, Austria) were activated, tested with pH 4 and pH 7 buffer solutions, and then inserted into the animal within 4 hours of activation. Boluses were positioned into the applicator with the sensor tip at the head. The animal's neck and head were extended and straightened and then the bolus was administered. The boluses lodge in the reticulum. Two base stations were positioned within the experimental pens and fitted with solar panels and a repeater to allow real-time monitoring of rumen physiology via the smaXtec messenger 4.0 software application (smaXtec Animal Care GmbH, Graz, Austria). Following collection of boluses from the viscera, boluses were cleaned and re-tested in pH 4 and pH 7 buffer solutions to assess levels of pH drift throughout the study. Boluses were stored in pH 7 buffered solution between replicates (3 weeks).

3.5 Weather Measurements

Weather data including ambient temperature, relative humidity, solar radiation, and wind speed were recorded every 10 minutes and used to describe climatic conditions during transportation and lairage.

3.6 Performance Measurements

Live body weight measurements at induction and drafting were obtained using a single animal scale (Silencer Squeeze Chute, Moly Manufacturing Inc., Lorraine, Kansas, USA) set on four load cells that were professionally calibrated. Cattle were individually weighed at induction and drafting. Upon feedlot exit, cattle were weighed on a group scale. Exit weight was calculated by dividing the weight of the group scale by the number of head on the scale. Weight gain was calculated by subtracting the induction weight from the exit weight. Average daily gain was calculated by dividing the weight gain by the number of days on feed.

3.7 Muscle Glycogen Measurements

End glycogen content measures were taken on the carcasses of all cattle that received a reticulorumen bolus (n=120). Samples were taken at the time of chiller entry, approximately 60 minutes post-slaughter, from the dorsal region of the *Longissimus thoracis*, adjacent to the twelfth rib. Samples were frozen in liquid nitrogen directly after sampling and during transport to the laboratory. Samples were frozen at -20°C until laboratory analysis. Laboratory analyses for muscle glycogen levels were conducted according to methods described by Coombes et al., 2014.

3.8 Carcass Measurements

Hot standard carcass weight was recorded after evisceration and trimming. After chilling for approximately 15.8 h (range 10.5 to 20.3 h), carcass assessment was conducted by trained plant graders. Chiller assessment was conducted by qualified plant graders. Sex, body number, dentition, left side bruise, right side bruise, left hot standard carcass weight, right hot standard carcass weight, total hot standard carcass weight, eye muscle area at the *M. longissimus dorsi* quartering site, pH at chiller assessment at the *M. longissimus dorsi* quartering site, subcutaneous rib fat cold at the *M. longissimus dorsi* (Aus-meat standard site), Aus-meat meat colour, Hunter L, Hunter a, Hunter b, P8 fat, hump height cold, and ossification cold were recorded. PH meters were calibrated prior to each grading session and every two hours within a session by qualified plant graders. Temperature probes were calibrated. Objective meat colour was measured using a Hunter colorimeter at the time of grading by the principal investigator. Dressing percentage was calculated as the hot carcass weight divided by the draft weight weight times 100, as exit weights were measured on a group (not individual) basis.

3.9 Statistical Analyses

The experimental unit for this pilot experiment was defined as the individual animal. The experiment was initially analysed with all cattle and carcasses (n=400) as a 2 x 2 factorial design using the GLM procedure of SAS (SAS Institute Inc., Cary, North Carolina USA). Ship time, slaughter time, replicate,

the ship time x slaughter time interaction, and the ship time x slaughter time x replicate interaction were included in the model as fixed effects. Draft weight was used as a covariate in the model as it was determined to be statistically significant ($P < 0.05$) in initial models. Statistical significance of interactions and main effects were defined at $P < 0.05$ and a trend at $P < 0.10$ levels. If a significant shipping by slaughter time interaction was detected ($P < 0.05$), P – values for differences between simple effect means were determined using the PDIFF procedure (SAS).

The MEANS procedure was completed for induction weight, draft weight, exit weight, days on feed, weight gain, average daily gain, hot standard carcass weight, dressing percent, Hunter L, Hunter a, Hunter b, Aus-meat meat colour, chiller assessment pH, P8 fat, eye muscle area, hump height, ossification, rib fat, fat colour and duration of time from stunning to grading to calculate the mean, standard deviation, minimum and maximum of all variables.

The FREQ procedure was completed to calculate the percent of carcasses with a chiller assessment pH of greater than 5.70 and/or meat colour outside of the AUS-meat 1B-3 range per treatment. The FREQ procedure was used to determine the frequency of carcasses with each AUS-meat meat colour score per treatment. Transportation time and slaughter time were tested separately using the FREQ procedure described above. The Chi-squared test was used to test for differences in frequency data.

The CORR procedure tested for correlations between hot carcass weight, meat colour, chiller assessment pH, Hunter L, Hunter a, and Hunter b.

Further analyses were conducted on a subset of cattle (n=120). These cattle were randomised to treatment by weight at drafting which occurred 7 days prior transportation to the abattoir. These cattle (n=120) received reticulorumen boluses that recorded pH, temperature and activity levels every 10 minutes. A subset of these measurements including pH and temperature at the time of slaughter, 1 h before slaughter, 4 h before slaughter, 8 h before slaughter, 12 h before slaughter, 24 h before slaughter, and during transportation were selected for subsequent analyses. Intramuscular glycogen, Hunter L, Hunter a, Hunter b, meat colour, chiller assessment pH, hump height, P8 and rib fat, eye muscle area, dressing percentage, induction weight and hot carcass weight were measured for further analyses using the GLM procedure of SAS (SAS Institute Inc., Cary, North Carolina USA). Ship time, slaughter time, replicate, the ship time x slaughter time interaction, and the ship time x slaughter time x replicate interaction were included in the model as fixed effects. Draft weight was not significant in this sub-population, and therefore was not included as a covariate. Significant differences between simple effect means were determined using the PDIFF procedure (SAS) when a significant ship x slaughter time interaction was detected ($P < 0.05$).

4 Results

Simple descriptive statistics of the research population are presented in Table 1. The research cattle were fed between 63 and 65 days on feed (average 64.3), had an average daily gain of 2.17 kg/head/day, provided dressed carcasses with an average hot standard carcass weight of 273.1 ± 25.0 Kg, and 7.1 ± 2.0 mm rib fat. Mean timing of stunning to grading was 15.77 ± 3.18 hours, but ranged from 10.48 to 20.25 hours. Longissimus muscle glycogen of the 120 head tested averaged 1.18%, but ranged from 0.34 to 1.86%. Meat colour at grading averaged 2.23 ± 0.78 (range 1.33 to

6.00). The preharvest cattle daily gain noted in this study well exceeds the minimum of 1.0 Kg/day as the minimum daily gain for feedlot cattle to have adequate intramuscular glycogen to prevent dark cutting beef (Pethick et al., 2000).

The ambient temperature and relative humidity at the feedlot in the week prior to slaughter are displayed in Table 2. The ambient temperature, relative humidity, and duration of lairage are displayed in Table 3.

For the complete data set of 400 heifers, cattle slaughtered in the afternoon had significantly reduced (0.5%) dressing percentages and yielded carcasses that were 2.4 kg lighter ($P < 0.01$) than cattle slaughtered in the morning (Table 4). For shipping treatments, arrival at the abattoir the day before slaughter numerically decreased hot carcass weight ($P = 0.12$) and resulted in a trend for decreased dressing percentage ($P < 0.10$) by 1.4 kg and 0.3%, respectively.

A significant ($P < 0.01$) shipping time by slaughter time interaction was detected for chiller assessment pH and Aus-meat colour score with carcasses from heifers shipped the morning of slaughter and slaughtered that afternoon having higher pH and darker colour ($P < 0.05$) compared to other treatments. Cattle both shipped and slaughtered in the morning produced carcasses superior for pH and meat colour compared to all other treatments. After careful interpretation of the interaction and pooling of treatment means to analyse main effects, morning slaughtered heifers had lower pH and lighter coloured carcasses ($P < 0.01$).

There was no significant effect of timing of shipping or slaughter on carcass Hunter L scores. A significant ($P < 0.01$) shipping time by slaughter time interaction was however detected for Hunter a (redness) and b (yellowness), with heifers shipped the afternoon before slaughter and slaughtered the following morning having the greatest values. Similar to chiller assessment pH and meat colour, upon comparison of main effects, morning slaughtered carcasses had greater ($P < 0.01$) Hunter a and b values.

On average, chilling times were 17.7 h for AM ship and AM slaughter, 12.9 h for AM ship and PM slaughter, 19.2 h for PM ship and AM slaughter, and 13.3 h for PM ship and PM slaughter.

Statistical analyses (Table 5) of the cattle (n = 120) subset that received reticulorumen boluses at drafting noted no influence of shipping time or slaughter time on hot carcass weight, possibly reflecting limited statistical power to detect differences in hot carcass weight. However, the main effects of time of shipping and timing of slaughter significantly reduced – by a similar quantum – intramuscular glycogen concentration, however only time of slaughter negatively influenced ($P = .06$) chiller assessment pH. Differences between morning and afternoon slaughter were small (5.53 versus 5.56, respectively. In this data subset, shipping time similarly did not influence any of the Hunter colour values, however slaughter time significantly reduced Hunter a and Hunter b values.

Late afternoon (PM) shipping time did not impact rumen pH during transport or 24 h before stunning. However, PM shipping did significantly increase rumen bolus pH values 12 h, 8 h, 4 h, and 1 h prior to slaughter versus cattle shipped in the morning of slaughter. Effects of slaughter time on rumen pH at stunning were less clear due to the presence of a significant shipping time by slaughter time interaction ($P = 0.02$). Rumen pH was lowest for the AM ship/ AM slaughter treatment, which was significantly different from both PM shipping treatments.

Rumen temperature at time of stunning was greater ($P < 0.01$) for cattle shipped or slaughtered in the morning versus afternoon (0.47°C and 0.27°C increase in rumen temperature for morning shipping and morning slaughter, respectively).

Similar to the complete dataset, Hunter a (redness) and b (yellowness) values were significantly greatest for cattle PM Ship and AM Slaughter > AM Ship and AM Slaughter > AM Ship and PM Slaughter > PM Ship and PM Slaughter.

For the complete data set (n = 400), cattle shipped and slaughtered in the AM produced no carcasses (P = 0.02) with either chiller assessment pH > 5.70 nor carcasses with an Aus-Meat meat colour greater than 3.0 (Table 6). Incidence of carcasses with both chiller assessment pH > 5.70 or meat colour > 3.0 was 10% for AM ship/PM slaughter and 7% for PM ship/AM slaughter and PM ship/PM slaughter. The distribution of carcasses by Ausmeat meat colour assignment is presented in Table 7.

Table 1. Descriptive statistics of study heifers (n=400).

Variable	Mean	Stdev	Minimum	Maximum
Entry weight, kg	379.8	35.6	275.0	488.0
Draft weight, kg	504.0	42.5	378.0	631.0
Exit weight, kg*	512.3	8.3	499.0	524.0
Days on feed, d	64.3	0.6	63.0	65.0
Average daily gain, kg†	2.17	0.38	1.09	3.67
Hot carcass weight, kg	273.1	25.0	193.5	358.0
Dressing Percent, %‡	54.18	1.85	46.05	59.57
Eye muscle area, cm^2	67.6	9.9	35.0	98.0
Rib fat, mm	7.1	2.0	3.0	16.0
P8 fat, mm	12.3	4.1	5.0	30.0
Hump height, mm	48.7	7.0	30.0	75.0
Ossification	157.6	19.8	120.0	230.0
Time from stun to grade, h	15.77	3.18	10.48	20.25
Chiller assessment pH	5.55	0.10	5.37	5.92
Muscle glycogen, %§	1.18	0.31	0.34	1.86
Meat colour‖	2.23	0.78	1.33	6.00
Hunter Lightness, L	33.14	3.39	24.01	64.91
Hunter redness, a	20.37	2.88	4.83	26.89
Hunter yellowness, b	16.27	3.04	4.16	23.21

*Exit weights were not measured individually.

†Average daily gain was reported as ((draft weight-entry weight)/days on feed at drafting).

‡Dressing percent was calculated as ((Hot carcass weight/Draft weight) x 100%).

§Muscle glycogen was measured on a subset of cattle (n=120).

‖Meat colour was scored as 1A=1.00, 1B=1.33, 1C=1.67, 2=2.00, 3=3.00, 4=4.00, 5=5.00, 6=6.00.

Table 2. Ambient temperature and relative humidity at feedlot.

Variable		Mean	Stdev	Minimum	Maximum
Replicate 1*					
	Ambient temperature, °C	25.7	6.2	14.5	37.5
	Relative humidity, %	46.9	22.0	5.4	85.8
Replicate 2†					
	Ambient temperature, °C	27.3	5.1	19.0	38.1
	Relative humidity, %	45.5	15.4	19.4	85.8

*Measurements for replicate 1 were recorded from 5:00 p.m. on 15 February 2018 to 5:00 p.m. on 21 February 2018.

†Measurements for replicate 2 were recorded from 5:00 p.m. on 15 March 2018 to 5:00 p.m. on 21 March 2018. .

Table 3. Ambient temperature, relative humidity, and duration of lairage.

Variable			Mean	Stdev	Minimum	Maximum
Replicate 1						
	Lairage duration, h					
		AM ship, AM slaughter	1.6	0.3	1.2	2.0
		AM ship, PM slaughter	7.2	0.1	7.0	7.4
		PM ship, AM slaughter	13.0	0.2	12.8	13.3
		PM ship, PM slaughter	19.6	0.1	19.4	19.8
	Ambient temperature, °C					
		AM ship, AM slaughter	20.0	1.0	18.7	21.2
		AM ship, PM slaughter	25.6	4.0	18.7	31.3
		PM ship, AM slaughter	19.7	3.4	15.0	27.4
		PM ship, PM slaughter	21.5	4.4	15.0	30.1
	Relative humidity, %					
		AM ship, AM slaughter	59.0	3.2	55.0	63.0
		AM ship, PM slaughter	41.6	12.1	25.0	63.0
		PM ship, AM slaughter	60.9	10.9	36.0	77.0
		PM ship, PM slaughter	54.9	14.0	28.0	77.0
Replicate 2						
	Lairage duration, h					
		AM ship, AM slaughter	1.8	0.1	1.6	2.1
		AM ship, PM slaughter	7.2	0.2	6.9	7.6
		PM ship, AM slaughter	13.5	0.1	13.2	13.7
		PM ship, PM slaughter	19.3	0.1	19.1	19.5
	Ambient temperature, °C					
		AM ship, AM slaughter	22.0	1.0	20.6	23.7
		AM ship, PM slaughter	22.8	1.0	20.6	24.2
		PM ship, AM slaughter	20.7	1.1	19.3	23.1
		PM ship, PM slaughter	21.4	1.5	19.3	24.2
	Relative humidity, %					
		AM ship, AM slaughter	65.8	4.3	58.0	70.0
		AM ship, PM slaughter	61.4	4.9	54.0	70.0
		PM ship, AM slaughter	66.1	6.7	55.0	74.0
		PM ship, PM slaughter	64.6	6.5	54.0	74.0

Table 4. Effects of time of ship and time of slaughter on meat quality of finishing beef heifers (n=400).

	Ship			Slaughter			Ship*Slaughter					
							AM Ship		PM Ship			
Item	AM	PM	*P-value*	AM	PM	*P-value*	AM Slaughter	PM Slaughter	AM Slaughter	PM Slaughter	*P-value*	SE
Draft weight, kg[*]	498.2	510.1	<0.01	506.1	502.3	0.36	497.6	498.9	514.6	505.6	0.22	41.700
Hot carcass weight, kg	273.8	272.4	0.12	274.3	271.9	<0.01	275.8	271.9	272.8	272.0	0.09	9.155
Dressing percent, %[†]	54.33	54.03	0.10	54.43	53.94	<0.01	54.73	53.94	54.12	53.94	0.10	1.822
Eye muscle area, cm^2	67.7	67.6	0.90	67.6	67.7	0.88	67.7	67.7	67.5	67.7	0.86	8.366
Rib fat, mm	7.0	7.2	0.33	7.0	7.1	0.82	6.6^{a}	7.3^{bc}	7.5^{c}	6.8^{ab}	<0.01	1.769
P8 fat, mm	12.1	12.5	0.30	12.8	11.9	<0.01	12.8	11.5	12.8	12.2	0.32	3.563
Hump height, mm	48.6	48.7	0.90	48.5	48.9	0.59	48.0	49.3	49.0	48.4	0.16	6.596
Chiller assessment pH	5.55	5.55	0.86	5.54	5.57	<0.01	5.52^{a}	5.59^{c}	5.55^{ab}	5.55^{b}	<0.01	0.100
Meat colour[‡]	2.24	2.23	0.98	2.12	2.35	<0.01	2.01^{a}	2.46^{c}	2.23^{b}	2.23^{b}	<0.01	0.756
Hunter lightness, L	33.15	33.12	0.94	33.39	32.88	0.13	33.70	32.60	33.08	33.16	0.08	3.349
Hunter redness, a	20.41	20.33	0.76	21.43	19.31	<0.01	20.85^{b}	19.97^{c}	22.02^{a}	18.64^{d}	<0.01	2.562
Hunter yellowness, b	16.14	16.39	0.35	17.55	14.97	<0.01	16.76^{b}	15.52^{c}	18.34^{a}	14.43^{d}	<0.01	2.622

[*]Draft weight was included in the model as a covariate.

[†]Dressing percent was calculated as ((Hot carcass weight/Draft weight) x 100%).

[‡]Meat colour was scored according to AusMeat colour standards and converted to numerical scores. 1A=1.00, 1B=1.33, 1C=1.67, 2=2.00, 3=3.00, 4=4.00, 5=5.00, 6=6.00.

[a,b,c,d]Means with diffferent superscripts differ ($P < 0.05$).

Table 5. Effects of time of ship and time of slaughter on meat quality and rumen physiology of bolused finishing beef heifers.

	Ship			Slaughter			Ship*Slaughter					
							AM Ship		PM Ship			
Item	AM	PM	*P-value*	AM	PM	*P-value*	AM Slaughter	PM Slaughter	AM Slaughter	PM Slaughter	*P-value*	SE
Entry weight, kg	392.50	394.88	0.55	397.62	389.77	0.05	395.07	389.93	400.17	389.60	0.49	21.645
Draft weight, kg	520.18	524.50	0.25	523.37	521.32	0.59	520.33	520.03	526.40	522.60	0.64	20.573
Hot carcass weight, kg	281.69	284.84	0.24	284.90	281.63	0.22	282.48	280.90	287.32	282.37	0.53	14.559
Dressing percent, %[*]	54.16	54.31	0.67	54.44	54.02	0.24	54.29	54.03	54.59	54.02	0.65	1.903
Eye muscle area, cm^2	71.32	71.92	0.69	70.93	72.30	0.36	70.70	71.93	71.17	72.67	0.93	8.119
Rib fat, mm	7.33	7.52	0.59	7.33	7.52	0.59	6.70^{a}	7.97^{b}	7.97^{b}	7.07^{ab}	<0.01	1.879
P8 fat, mm	12.35	12.85	0.38	13.08	12.12	0.09	12.77	11.93	13.40	12.30	0.82	3.134
Hump height, mm	50.00	50.50	0.71	50.17	50.33	0.90	49.17	50.83	51.17	49.83	0.27	7.352
Chiller assessment pH	5.54	5.55	0.77	5.53	5.56	0.06	5.51	5.57	5.54	5.55	0.13	0.090
Muscle glycogen, %	1.25	1.10	<0.01	1.25	1.10	<0.01	1.30	1.21	1.20	1.00	0.31	0.292
Meat colour[†]	2.14	2.21	0.64	2.12	2.23	0.42	2.02	2.27	2.22	2.19	0.29	0.719
Hunter lightness, L[‡]	33.59	33.98	0.60	33.88	33.69	0.80	34.54^{a}	32.64^{b}	33.23^{ab}	34.74^{a}	0.03	4.118
Hunter redness, a	20.79	20.29	0.33	21.64	19.44	<0.01	21.15^{ab}	20.43^{b}	22.12^{a}	18.45^{c}	<0.01	2.854
Hunter yellowness, b	16.56	16.42	0.78	17.88	15.11	<0.01	17.25^{ab}	15.87^{b}	18.50^{a}	14.34^{c}	<0.01	2.792
Rumen pH at stun	6.93	7.21	<0.01	7.06	7.08	0.80	6.85^{a}	7.02^{ab}	7.28^{c}	7.15^{bc}	0.02	0.361
Rumen pH 1h before stun	6.93	7.22	<0.01	7.08	7.07	0.91	6.88	6.98	7.29	7.16	0.08	0.357
Rumen pH 4h before stun	6.73	7.19	<0.01	6.94	6.98	0.68	6.66	6.81	7.23	7.15	0.16	0.413
Rumen pH 8h before stun	6.51	7.09	<0.01	6.72	6.87	0.05	6.39	6.63	7.05	7.12	0.28	0.405
Rumen pH 12h before stun	6.31	6.92	<0.01	6.50	6.73	<0.01	6.17	6.45	6.83	7.00	0.42	0.360
Rumen pH 24h before stun	6.55	6.53	0.84	6.67	6.42	<0.01	6.65	6.46	6.69	6.38	0.45	0.453
Rumen pH on truck	6.74	6.71	0.73	6.80	6.65	0.11	6.82	6.67	6.78	6.64	0.96	0.486
Temperature at stun	39.83	39.36	<0.01	39.74	39.45	<0.01	39.99	39.68	39.50	39.22	0.81	0.333
Temperature 1h before stun	39.81	39.17	<0.01	39.77	39.22	<0.01	40.17^{c}	39.46^{b}	39.37^{b}	39.98^{a}	0.02	0.368
Temperature 4h before stun	39.40	38.88	<0.01	39.36	38.93	<0.01	39.60	39.21	39.12	38.64	0.61	0.479
Temperature 8h before stun	39.87	39.04	<0.01	39.43	39.49	0.41	39.70^{c}	40.05^{d}	39.16^{b}	38.92^{a}	<0.01	0.359
Temperature 12h before stun	39.98	39.37	<0.01	39.84	39.50	<0.01	40.02^{c}	39.93^{c}	39.67^{b}	39.06^{a}	<0.01	0.496
Temperature 24h before stun	39.76	39.68	0.39	39.69	39.76	0.42	39.62^{a}	39.90^{b}	39.76^{ab}	39.61^{a}	0.01	0.460
Temperature on truck	39.95	40.39	<0.01	40.18	40.16	0.72	40.00	39.89	40.36	40.42	0.18	0.345

[*]Dressing percent was calculated as ((Hot carcass weight/Draft weight) x 100%).

[†]Meat colour was scored according to AusMeat colour standards and converted to numerical scores. 1A=1.00, 1B=1.33, 1C=1.67, 2=2.00, 3=3.00, 4=4.00, 5=5.00, 6=6.00.

[‡]For Hunter lightness, L, a level of significance of p<0.10 was used to test the ship by slaughter time interaction.

[a,b,c,d]Means with diffferent superscripts differ ($P < 0.05$).

Table 6. Frequency statistics for chiller assessment pH and Aus-Meat meat colour of study heifers (n=400).

Variable	Treatment				*P*- value
	AM Ship		PM Ship		
	AM Slaughter	PM Slaughter	AM Slaughter	PM Slaughter	
Chiller assessment pH, n					0.0203
> 5.70	0	10	6	7	
≤ 5.70	101	90	93	93	
Chiller assessment pH, %					0.0203
> 5.70	0.00	10.00	6.06	7.00	
≤ 5.70	100.00	90.00	93.94	93.00	
Aus-Meat meat colour*, n					0.0203
Darker than 3	0	10	6	7	
3 or lighter	101	90	93	93	
Aus-Meat meat colour*, %					0.0203
Darker than 3	0.00	10.00	6.06	7.00	
3 or lighter	100.00	90.00	93.94	93.00	

*Meat colour scores were assigned as 1A=1.00, 1B=1.33, 1C=1.67, 2= 2.00, 3=3.00.

Table 7. Frequency statistics for Aus-Meat meat colour of study heifers (n=400).

	Treatment				*P*- value
Aus-Meat Meat Colour*	AM Ship		PM Ship		
	AM Slaughter	PM Slaughter	AM Slaughter	PM Slaughter	0.0128
1B					
n	0	0	2	2	
%	0.00	0.00	2.02	2.00	
1C					
n	16	7	16	18	
%	15.84	7.00	16.16	18.00	
2					
n	78	62	66	62	
%	77.23	62.00	66.67	62.00	
3					
n	7	21	9	11	
%	6.93	21.00	9.09	11.00	
4	0	4	1	2	
n	0.00	4.00	1.01	2.00	
%					
5					
n	0	3	4	4	
%	0.00	3.00	4.04	4.00	
6					
n	0	3	1	1	
%	0.00	3.00	1.01	1.00	

*Meat colour scores were 1A=1.00, 1B=1.33, 1C=1.67, 2= 2.00, 3=3.00, 4=4.00, 5=5.00, 6=6.00.

5 Discussion

The study was comprised of British-type heifers sourced to target the domestic market with an average entry weight of 379.8 kg that were fed for 63 to 65 days with an average daily gain of 2.17 kg. Cattle dressed at 54.18% on average, yielding carcasses of 273.1 ± 25.0 kg with 7.1 ± 2.0 mm rib fat. These descriptive statistics (Table 1) are representative of the type of cattle which are fed for the Australian domestic market. It is important that the results of this study are interpreted in the context of cattle type as further research is required to determine if the results of this study are consistent for cattle which represent different market types such as pasture-fed cattle, long-fed cattle, Wagyu cattle, Bos indicus cattle, cattle with very large carcass weights or elevated levels of subcutaneous fat. The inherent differences in fat levels, intramuscular glycogen content, and carcass surface area to weight ratio between cattle types may influence the chilling profile of carcasses, pH decline, meat colour changes post-mortem, and meat quality. However, the sample of cattle tested in this study are consistent with those fed for the domestic market in Australia and thus the results of this study are directly applicable to that population.

While it has been clearly demonstrated and accepted across the feedlot industry that cattle destined for slaughter should be on a high plane of nutrition to maximise muscle glycogen levels prior to feedlot exit (Pethick et al., 2000), the incidence of dark cutting beef remains a significant issue for the feedlot industry in Australia. For example, the heifers in the present study were all fed a high energy steam-flaked wheat and barley finisher ration that exceeded National Research Council 2016 requirements for beef cattle (National Academies of Sciences and Medicine, 2016). However, 5.8% (23/400) of the heifers displayed chiller assessment pH greater than 5.70 and Aus-Meat meat colour darker than 3.0. Previous work has suggested that cattle gaining a minimum of 1.0 kg/d is required to assure a level of muscle glycogen that is adequate to reduce the incidence of dark cutting (Pethick et al., 2000), however, dark cutting remains an issue for cattle gaining more than 1.0 kg/d such as the cattle in this study that gained on average 2.17 kg/day, with a range from 1.09 to 3.67 kg/d. Importantly, the 23 carcasses in the present study with Aus-Meat meat colour greater than 3.0 and chiller assessment pH greater than 5.70 gained 2.18 kg/d on average.

The average amount of time from stun to grade in the present study was 15.77 ± 3.18 hours and ranged from 10.48 to 20.25 hours. The duration of time from stun to grade was not included in the statistical model as a covariate as it is confounded by treatment and thus would not be appropriate. A limitation of the present study is that the duration of time from stun to grade was only controlled to a 10-hour window, ranging between 10.5 to 20.5 hours. It is suggested that future work in this area further control for the duration of time from stun to grade. However, this was beyond the context of the present study and presents logistical timing challenges for the individuals grading the carcasses.

The effect of time of slaughter on hot carcass weight and dressing percent are profound in the present study. Although the aim of the present study was to identify and test an intervention for dark cutting, the yield results present noteworthy economic returns and are thus further discussed. Specifically, cattle slaughtered in the morning produced carcasses 2.4 kg heavier ($P < 0.01$) than

cattle slaughtered in the afternoon regardless of time of ship. For dressing percent, cattle slaughtered in the morning dressed 0.5% higher (P <0.01) than cattle slaughtered in the afternoon (Table 4). Heifers that were shipped and slaughtered in the morning yielded carcasses that were numerically 3.9 kg heavier than cattle AM shipped and PM slaughtered, 3.0 kg heavier than cattle PM shipped and AM slaughtered, and 3.8 kg heavier than cattle PM shipped and PM slaughtered. Regardless of shipping time, cattle slaughtered in the afternoon had similar carcass weights. Cattle that are slaughtered in the morning had less carcass shrink and thus produced heavier carcasses and higher dressing percentages. Although not significant, perhaps due to a smaller sample size (n=120), similar numerical results were found in the subset of cattle where muscle glycogen levels were measured (Table 5). In this sample of bolused cattle, cattle slaughtered in the morning had numerically higher (P = 0.22) hot carcass weights (284.90 kg for AM slaughter and 281.63 for PM slaughter) and numerically higher (P = 0.24) dressing percentages (54.44% for AM slaughter and 54.02% for PM slaughter) than cattle slaughtered in the afternoon. We hypothesize this is due to reduced tissue shrink and lower mobilisation of muscle glycogen in cattle slaughtered in the morning as compared to cattle slaughtered in the afternoon. Intramuscular glycogen levels were 0.15% higher (P < 0.01) for cattle slaughtered in the morning as compared to the afternoon which reflects greater glycogen stores in cattle slaughtered in the morning as compared to those slaughtered in the afternoon.

Cattle that were shipped the day before slaughter and slaughtered the following afternoon had the longest duration of lairage, approximately 19 hours (Table 3), and had the lowest (P < 0.01) levels of muscle glycogen which was on average 1.00%. These findings demonstrate that the greater the amount of time an individual spent in lairage, the lower the level of intramuscular glycogen at slaughter. These findings are consistent with those of Janloo et al. (1999) and Kreikemeier et al. (1998) who found a increase in the incidence of dark cutting with an extended duration of lairage. This suggests that a greater amount of glycogen is metabolised as the duration of lairage increases. This is further confounded by the amount of time without feed increases as the duration of lairage increases and thus there is a reduction in the glucose precursors required to synthesize glycogen.

Reduced antemortem levels of muscle glycogen lead to reduced postmortem lactic acid accumulation and impede pH decline following slaughter (Ashmore et al., 1973, Scanga et al., 1998). This results in beef with an elevated chiller assessment pH, increased light absorption, and increased water-binding ability that appears dark, firm, and dry (Tarrant and Sherington, 1980, Scanga et al., 1998). Although we expected the cattle with the lowest muscle glycogen levels, those cattle PM shipped and PM slaughtered, would have the highest chiller assessment pH and highest meat colour scores, this was not found in the present study. This suggests that muscle glycogen levels may not fully explain the differences in meat colour and chiller assessment pH identified in the present study. We hypothesize that cattle slaughtered in the afternoon may not achieve complete post-mortem changes in meat colour and pH due to insufficient hours of chilling. Cattle slaughtered in the afternoon had higher chiller assessment pH, darker meat colour, and lower Hunter redness and yellowness scores compared to cattle slaughtered in the morning. Cattle slaughtered in the afternoon were chilled for 5.32 (P < 0.01) hours less than cattle slaughtered in the morning. On average, chilling times were 17.7 h for AM ship and AM slaughter, 12.9 h for AM ship and PM slaughter, 19.2 h for PM ship and AM slaughter, and 13.3 h for PM ship and PM slaughter. Although these findings suggest carcasses should be chilled for a minimum for 18 hours to allow for complete

conversion of muscle to meat and associated changes in pH and meat colour, ensuring adequate chilling time will not solve the issue of dark cutting as 6.06% of carcasses from the PM ship AM slaughter treatment exhibited Aus-Meat meat colour greater than 3 and chiller assessment pH greater than 5.70 as compared to 0 carcasses from the AM ship AM slaughter treatment. Cattle transported to the abattoir on the morning of slaughter and slaughtered that same morning displayed the lowest ($P < 0.06$) chiller assessment pH, most ideal Aus-Meat meat colour ($P < 0.01$), and lowest ($P = 0.02$) frequency of carcasses with chiller assessment pH > 5.70 and Aus-Meat meat colour darker than 3 (Table 6).

To further understand the physiology contributing to these differences in meat quality and yield, measurements of reticulorumen pH and temperature were analysed for a subset (n=120) of the cattle (Table 5). Cattle that were transported to the abattoir the evening prior to the day of slaughter (PM ship) had higher ($P < 0.01$) rumen pH 1h before stun, 4h before stun, 8h before stun, and 12h before stun than cattle transported to the abattoir on the morning of slaughter (AM ship). This is likely associated with the period of time cattle were withheld from feed prior to stunning. Cattle shipped the evening before slaughter (PM ship) experienced a longer period of time off feed prior to slaughter as compared to cattle in the AM ship treatment. As the period of time off feed increases, substrate for ruminal fermentation is reduced, rate of fermentation is reduced and subsequently less acid is produced by ruminal microbes resulting in higher pH of the rumen. As expected, within treatments and for all treatments, as the period of time off feed increased, there was a steady increase in rumen pH. These results suggest that cattle that are transported to the abattoir the evening before slaughter which experience extended durations of time off feed have reduced dietary glucose available for ruminal fermentation as compared to cattle transported on the morning of slaughter and thus have higher ruminal pH.

Rumen temperature at time of stunning was greater ($P < 0.01$) for cattle shipped or slaughtered in the morning versus afternoon (0.47 and 0.27 °C increase in rumen temperature for morning shipping and slaughter, respectively. These differences are likely a result of increased heat of fermentation in the rumens of cattle shipped or slaughtered in the morning as compared to cattle that have been withheld from feed for a longer period of time. In summary, cattle that are shorter time off feed (e.g. AM ship/AM slaughter), are hotter due to higher metabolic heat production, have lower rumen pH in the hours directly prior to slaughter due to continued acid production in the rumen, and have higher intramuscular glycogen as there is a continuous level of substrate available for glycogen synthesis.

The project fulfilled all objectives. Specifically, this work determined that shipping cattle on the morning of slaughter increased muscle glycogen levels, reduced the pH and increased the temperature of the rumen in the hours leading up to slaughter and at the time of slaughter, tended to increase dressing percent and hot carcass weight. This work clearly demonstrated a 0.49% advantage in dressing percent and 2.4 kg advantage in hot carcass weight for cattle slaughtered in the morning versus afternoon. Cattle slaughtered in the morning also exhibited lower chiller assessment pH and more ideal subjective and objective meat colour as compared to cattle slaughtered in the afternoon. Cattle slaughtered in the morning displayed a 0.15% advantage in muscle glycogen as compared to cattle slaughtered in the afternoon.

Interestingly, cattle transported to the abattoir on the morning of slaughter and slaughtered that same morning displayed the numerically highest muscle glycogen level, numerically highest hot carcass weight, numerically highest dressing percent, lowest chiller assessment pH, lowest Aus-Meat meat colour score, and lowest frequency of cattle with pH > 5.70. These results suggest that the incidence of dark cutting may be reduced by reducing the period of time off feed prior to slaughter to less than 4 hours, reducing the duration of lairage to less than 3 hours, and increasing the duration of chilling time to approximately 18 hours.

While this study was only a pilot of 400 head, it raises significant questions in regards to the influence of duration of lairage, time of slaughter, and duration of chilling which require further research. Although future research in these areas will require increases in sample size, likely to large pen trials, this work must be completed with impeccable attention to detail including full transportation audits to confirm times of feedlot dispatch and abattoir arrival. Additionally, strict surveillance of research cattle is required at the abattoir to confirm location and conditions of lairage as well as time of slaughter and washing conditions. In the present study, rumen boluses were inserted one week prior to slaughter when cattle were drafted to exit pens. Importantly, no cattle were comingled at drafting. However, it would be ideal to minimise handling of cattle in the two weeks directly proceeding slaughter to reduce stress levels. If rumen boluses were inserted at induction and cattle were slaughtered by home pen, not draft pen, this may provide the opportunity to remove working cattle within 2 weeks prior to slaughter.

6 Conclusions/recommendations

The present study provides multiple areas for practical application in the red meat industry. The incidence of dark cutters was lowest in cattle that were transported the morning of slaughter and slaughtered in the morning as compared to cattle that were transported the evening before slaughter or transported on the morning of slaughter and slaughtered that afternoon. Thus, a shorter duration of lairage and slaughtering cattle in the morning was shown to reduce the incidence of dark cutters. It appears that cattle slaughtered in the morning are less likely to cut dark. However, this may be influenced by the duration of chilling time prior to grading since cattle slaughtered in the morning had longer chiller duration than cattle slaughtered in the afternoon. These results suggest that the incidence of dark cutting may be reduced by minimising the period of time off feed prior to slaughter to less than 4 hours, reducing the duration of lairage to less than 3 hours, and increasing the duration of chilling time to approximately 18 hours. Therefore, we recommend future discussions between the feedlot and processing sectors be held to determine how the period of time off feed prior to slaughter and lairage times can be shortened and chiller times can be maintained to a minimum of 18 hours.

For the red meat industry, to achieve full value from the findings of this study, the results must be clearly communicated to industry.

Presently beef producers are not made aware of the time that their cattle arrive at an abattoir, the period of time their cattle remain in lairage, the conditions of lairage, the time that cattle are slaughtered, or the time of grading. The present work demonstrated that these conditions have a

significant impact on hot carcass weight, dressing percent, and chiller assessment pH which influence the price that producers receive for their cattle. To drive improvement in these areas, it would be beneficial to provide feedback to producers including the time cattle arrive to the abattoir, time of slaughter, and time of grading. To drive change in these areas, producers, transporters, processors, abattoir veterinarians, and qualified graders will need to work together and develop a plan to achieve optimal transportation and lairage conditions to achieve optimal carcass yield and quality. Logistically, it will be challenging to reduce the period of time off feed and the period of time in lairage, and increase the chilling period, however, the results of this study clearly demonstrate it is possible and the benefits are significant.

7 Key messages

Slaughtering cattle in the morning reduced the frequency of dark cutting and increased hot carcass weight as compared to slaughtering cattle in the afternoon.

In the present study, the ideal conditions to reduce the incidence of dark cutting in British-type heifers fed a high energy steam-flaked wheat and barley finisher ration for approximately 60 days with an average hot carcass weight of 225 – 325 kg were found to be:

1) Period of time off feed prior to slaughter should be a maximum of 4 hours.
2) Lairage duration should be a maximum of 3 hours.
3) Chilling duration should be a minimum of 18 hours

We urge the red meat industry to not simply put these recommendations in the 'too hard basket' and to organise cross-sector discussions on how to drive change in the areas of transportation, lairage, and chilling to achieve optimal beef yield and quality.

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