

# Impacts of Palm Kernel Expeller (PKE) supplementation on venison quality attributes

Sara Walden, Jamie Ward, Geoff Asher, Carolina Realini, Alastair Ross, Wendy Bain, John Mills, Noby Jacob, Yash Dixit, Sam Hitchman, Jack Somerton, Harold Henderson, Jim Morton, Sue Mason, Nicola Schreurs and Cameron Craigie

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#### Inquiries or requests to:

Cameron Craigie cameron.craigie@agresearch.co.nz Food & Bio-based Products Group, AgResearch Ltd Lincoln Research Centre, Private Bag 4749, Christchurch, New Zealand

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Dr Carolina Realini Acting Meat Quality Team Leader Meat Quality Team Food & Bio-based Products Group

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# 1. Executive Summary

The aim of this study was to investigate the impacts of Palm Kernel Expeller (PKE) as a supplementary feed in the New Zealand deer farming system on the meat quality attributes and the overall consumer acceptability of NZ farm-raised venison. For this study, two classes of red deer were used: rising two-year old hinds and rising one-year old stags. These animals were randomly placed into two treatment groups: pasture only and 50%PKE:50% pasture groups.

The animals were finished on these feeding treatments for 6 weeks and then processed according to standard industry practice at Duncan and Company's Otago Venison deer slaughter plant. The left loin was subsampled and used for the consumer sensory analysis using a panel of 102 untrained consumers at Lincoln University's Wine, Food and Molecular biosciences department and rapid evaporative ionisation spectrometry (REIMS) testing at AgResearch Lincoln. The right loin was subsampled for drip loss and pH, mechanical tenderness, Intramuscular fatty acid content, colour stability, myofibrillar fragmentation index (MFI) and sarcomere length and the confinement odour and microbial measurements (Lactic Acid Bacteria and Aerobic Plate Count).

Deer from the pasture-fed treatment had higher dressing out percentages than those fed PKE supplement (P<0.05). PKE Supplementation had little impact on carcass characteristics with no statistically significant effects on, carcass weight, GR, or leg weights or yields.

The consumer sensory analysis found that PKE supplementation had no statistically significant impact on venison sensory attributes overall liking, degree of tenderness and flavour. The consumers did rate that the juiciness of the venison from the pasture only treatment to be higher (P<0.1) than the venison from the PKE treatment. There were no significant differences among the different types of venison on the quality ratings assigned by respondents. Overall, venison resulted in high levels of consumer satisfaction with 43% of consumers rating samples as 'good everyday', 28% as 'better than everyday' and 14% as 'premium quality' regardless of the type of venison.

As expected, venison from the pasture-only group had higher PUFA, omega 3 content and a higher n3/n6 ratio (higher is better). PKE supplementation resulted in venison having higher intramuscular fat, saturated fatty acid and monounsaturated fatty acid contents compared to pasture-only finishing. PKE supplementation had no statistically significant impact on the levels of omega 6, L-omega 3, EPA+DHA components of venison intramuscular fat.

PKE supplementation impacted the composition of venison intramuscular fat, with some fatty acids increasing and others decreasing. The impacts of the differences in terms of nutritional quality may be explained by the higher intramuscular fat content in PKE-supplemented venison. There may be differences in fat melting point arising from the fat compositional differences, which could alter the mouthfeel of eating venison. The essential fatty acid alpha-linolenic acid (C18:3 n3) was statistically significantly higher in pasture-only venison.

PKE supplementation had no statistically significant effect on the colour (appearance) of venison in the vacuum pack after 8 weeks storage. During the 14-day retail display small differences in colour were detected by the colourimeter, However, these significant differences are unlikely to be detected by consumers.

PKE supplementation had no statistically significant impacts on pH<sub>24h</sub>, mechanical tenderness after 21d chilled aging, sarcomere length, confinement odour, persistent odour, pH, Aerobic Plate Count (APC) and Lactic Acid Bacteria (LAB) after 8 weeks chilled storage. However, venison from deer supplemented with PKE had significantly higher drip loss, drip loss percentage and MFI% than the pasture only group suggesting that the PKE supplementation may increase the rate of post mortem proteolysis (aging).

REIMS detected a total of about 5000 metabolite features. Of the metabolite features detected, over 1000 were significantly different between pasture fed and PKE fed deer. This result suggests that the REIMS device could be used for real-time verification of some product provenance factors.

#### Summary

Supplementary feeding of pasture-fed R1 stags and R2 hinds with PKE for 6 weeks pre-slaughter at 2kg/head per day had minimal impacts on growth, carcass or venison quality characteristics.

# 2. Background

Prime New Zealand farm-raised venison comes from animals <2 years of age. In New Zealand deer farming systems, the vast majority of venison is finished on grazed pasture, while many farm systems use crop supplementation in winter animals are most commonly slaughtered off pasture not crop. Palm Kernel Expeller (PKE) use has increased in deer farming systems in recent years although anecdotally most use appears to be with non-finishing stock classes (e.g. breeding hinds and velveting stags). Deer Industry New Zealand (DINZ), venison processors and other industry leaders were concerned about potential negative impacts of the use of PKE supplementation on the quality attributes and consumer acceptability of farm-raised venison.

PKE is a palm kernel by-product produced when oil from the fruit of the palm is extracted by screw presses, it is considered a medium quality energy feed that is high in dietary fibre and is a good source of protein (Lyu et al. 2018). Supplementation with PKE is very common in New Zealand Dairy farming systems. All PKE is imported in to New Zealand from tropical regions where Oil Palms are grown.

Currently the effects of PKE supplement on deer carcass characteristics and meat quality is not well characterised, but some research on other palm kernel (PK) by-products (e.g. Palm Kernel Cake (PKC) and Palm Kernel Meal (PKM)) and species (e.g. lamb, goat, cows, chicken and pigs) has been undertaken. In these studies animals were fed the PK as a concentrate diet and for 12-weeks or longer prior to slaughter or during production.

Wyngaard and Meeske (2017) found that milk yield and milk fat content from Jersey cows grazing kikuyu-ryegrass pasture did not change with the inclusion of different levels of PKE in the concentrates given. The body weight, body score, total volatile fatty acid and mean ruminal pH also did not differ between the treatments (0, 20% and 40% PKE). According to Freitas et al. (2017), the carcass characteristics of lamb did not differ between treatments where the inclusion of PKC increased in the concentrates (0%, 10%, 20% and 30% PKC). In both cases these animals were on the feeding treatments for 12 weeks before slaughter and were also grazing pasture (predominantly ryegrass). However, these reports did not evaluate the effects of PKE compared to pasture on the carcass and meat quality traits of the animals, instead these papers were focused on the effects of replacing parts of concentrates with PKC or PKE.

There are differences in venison quality attributes of stags and hinds. According to Kudrnáčová et al. (2018), stags have higher muscle and bone content and lower intramuscular fat (IMF) content than the hinds. Stags also have shorter sarcomere length than hinds which affects the tenderness of the meat (Purchas et al. 2010; Craigie 2012). Reports have found that there were colour differences between the genders and that venison from stags have a slightly lighter colour (higher L\* value) than hinds. According to Razmaitė et al. (2017), males have a higher amount of saturated fatty acids (SFA) and a lower amount of polyunsaturated fatty acids (PUFA) than hinds (18 months

to 36 months of age). One study found that because of these trait differences, the organoleptic attributes can also be different between stags and hinds. It was reported that venison from stags tends to be tougher than the venison from hinds (Purchas et al. 2010). None of the above studies utilised a consumer panel, which is the ultimate way to establish the impacts of different production and processing factors on venison eating quality.

The purpose of this study was to investigate the effect of Palm Kernel Expeller (PKE) supplementation vs pasture only as in a finishing regime, on the meat quality of venison from rising-yearling (R1) stags and rising-two-year-old (R2) hinds.

# 3. Methods

#### 3.1 Animals

The animals were born and raised on the AgResearch Invermay farm in Mosgiel, Otago, New Zealand. The study was approved by the Invermay animal ethics committee (AE14538). There were two classes of animal, rising-two-year old (R2) females that had failed to conceive following their first puberty, so were non-pregnant or 'dry' (n=20), and rising-one-year-old (R1) males (n=40). At slaughter it was noted that two of R1 males had been assigned the incorrect gender at weaning and were in fact females, so were excluded from the analysed data, they were both in the Pasture only feeding treatment. For this trial, 'dry' R2 females were selected on the basis that they tend to be a fatter stock class than R1 males, enabling a broader assessment of the impacts of PKE supplementation on venison quality.

All animals were red deer (*Cervus elaphus scotticus, hippelaphus or pannonensis*) or crosses of these subspecies. The R2 females were by 11 different sires, the R1 animals were sired by 4 different sires, which represented very diverse genetic lines. R2 females were randomly allocated ten per group to treatment groups and the R1 animals were randomly allocated twenty per group to treatment groups balanced by sire. Final groups were mixed sex with 10 R2 females, 18 R1 males and 2 R1 females (pasture group) and 10 R2 females, 20 R1 males in the PKE group. At the start of the transition week, then at the trial start and fortnightly after that animals were weighed to the nearest 0.5kg using weigh crates mounted on load cells recording electronic ID (EID) and live weight on a TSi 2 weigh scale and animal database (Gallagher Ltd Hamilton NZ). Body condition score (BCS) was also assessed at these time points using the 1-5 scale of Audigé et al. (1998) in 0.5 increments. Animal temperament (aggression, agitation when confined, ease of handling and exit speed) was scored at the mid weigh-point (day 29 of the trial) following Schutz et al. (2016).

At the start of the trial transitioning period on 20 August 2018 the mean live weights were 91.8kg (SD 13.1kg) for the Pasture group with the Pasture R2 female mean 104.9kg (SD 12.2kg) and R1 male mean 86.4kg (SD 6.7kg), whereas the PKE group was 92.2kg average (SD 12.7kg), R2 female mean 107.2kg (SD 5.8kg), and R1 male mean 84.7kg (SD 7.3kg).

All animals were grazed together for 12 days on the main Invermay hill farm, before being transported approximately 3km down to the flat farm. The flat area was used as it is fenced into small paddocks of approximately 0.25ha which is less than 10% of the size animals were accustomed to on the hill area of the farm. The two classes of animals were forced into social interactions, which they may have been able to avoid in larger paddocks, this adjustment to a new environment was likely reflected in a loss of live weight across both groups at the start of the 6-week treatment period. At the completion of the 6-week feeding treatment period all animals were yarded in the late morning, had the final live weight and BCS measured, velvet antler was removed by a veterinarian for any males that required it and they were transported approximately 300m to

Duncan & Company's Otago Venison Deer Slaughter Plant (DSP) in the late afternoon, where they were then held in lairage, fasted, for slaughter the following morning.

## 3.2 Grazing Treatments

The treatment groups were pasture only (Pasture) and 50% pasture: 50% PKE (PKE), based on total dietary metabolisable energy (ME (MJ/kgDM) offered. PKE was estimated to be at 11MJME/kgDM and diets were balanced assuming that the pasture offered was also at 11MJME/kgDM. The PKE group was offered a very high rate of PKE supplementation to try and allow expression of maximal differences between treatments. The feeding treatments occurred over a 6-week period, with one week of transitioning prior to this for the PKE group. Pasture quantity was measured using a rising plate meter (Farmworks Precision Farming Systems, Manawatu, NZ) and the quality was estimated using visual assessment of dead material, clover and herbs and green leaf, using the Q-Graze software package. Feed allocation was calculated using the Deer Feed application, and PKE was assumed to be 90% dry matter (DM) with an 85% utilisation rate. Pastures were ryegrass dominant and the initial dietary pasture quality was assessed at approximately 10 MJME/kgDM as they contained 30-40% dead material and only 0-2% white clover or herbs, average pasture mass was 2050kgDM/ha for the PKE group.

Moving into September pasture quality improved and dietary metabolisable energy exceeded 10.5 MJME/kgDM after the first week of September, with spring pasture growth rates exceeding 50kgDM/day. Pasture quality or quantity was not assessed after this point as quality was assumed to be approximately 11 MJME/kgDM and pasture masses were 2500kgDM/ha or greater. Pasture and PKE allocations were based on live weight at the start of the trial thereafter on each fortnightly weight measurement and were targeted so that high growth rates of 250g/day for R2 females, and 500g/day for R1 males were possible. This equated to offering of 3.6kgDM/head at 11MJME/kgDM at the start of the trial 50% of which equated to 2.0kg of PKE, PKE offering was increased relative to changing live weight with a final maximum offer of 2.5kg/head/day.

PKE was introduced to the PKE group incrementally over the course of the transition week in long skinny feeding troughs. The initial offering was 0.5 kg/head offered once a day, this was increased to 2.0kg offered half in the morning and half in the afternoon, this split feeding of the PKE offering continued throughout the trial. Not all PKE was consumed during the transitioning phase, nor was it during the first few days of the 6-week feeding, the feeders were identified as the problem and the PKE was fed in lines on the ground and was readily consumed after this point. Where the pastures in PKE groups paddocks exceeded 2 days allocation they were break fenced with electric fencing, in to smaller areas to ensure that 50% of the diet consumed was PKE.

#### 3.3 Processing

Animals were slaughtered at Duncan and Company's Otago Venison DSP on 11 October 2018 by captive bolt, and exsanguination and were electrically stimulated immediately following this. Each individual animal's identity (visual ear tag) and time of death was recorded at the point of slaughter . Samples were tracked through the slaughter floor, by slaughter sequence and before they entered the chillers were tagged with the DSP carcass ticket as well as two other carcass tickets with unique ID's to trace samples through boning. On the slaughter floor after all gutting, trimming and meat inspection hot carcass weight (HCW) was recorded, as was fat depth over the 12th rib (GR). Carcasses were then spray chilled following the DSP standard procedures.

On 12 October 2018 approximately 24 hours after slaughter the carcasses were boned out. The loins consisted of both *m. longissimus dorsi* and *multifidus* and were a short loin plus the last two ribs of a standard boneless loin, due to cutting specifications on the day. Short loins were from the posterior end of the animal from approximately the 6th lumbar vertebrae forward to the 12th thoracic vertebrae. Loins were boned off each side of the carcass animals left hand side first then the right. Each loin sample was accompanied by a uniquely identified carcass ticket. Both loins were weighed to the nearest 1g with the silverskin and fat cap-on (i.e. cap-on), then they were mechanically deskinned (i.e. cap-off) and those weights also recorded. Bone-in rear-leg pair weight was recorded after the shoulders and loins were boned off the carcass. The pelvis and legs were separated from the torso between *6th lumbar vertebrae* and *sacrum* and the pair of rear-legs with hocks removed but including the pelvis (aitch-bone) were weighed to the nearest 100g.

#### 3.4 Meat sample collection

At 24 hrs post mortem, carcasses were fabricated according to standard industry practice at Duncan Venison, Mosgiel, Otago, New Zealand. The left and right short loins from each carcass were recovered, the silver skin was removed before further processing into sub-samples.

The right short loin was transported chilled approximately 300m to the Invermay Meat Laboratory for the intramuscular fat (IMF), mechanical tenderness, colour stability and confinement odour testing. The mechanical tenderness and storage colour testing was carried out at the Invermay Meat Laboratory and then other sub-samples of right loins were transported to Grasslands Research Centre and the Hopkirk Research Institute, Palmerston North for IMF, colour stability, confinement odour and microbial testing. Another sub-sample of each right loin was frozen and placed in small bags and transported to Massey University meat laboratory in Palmerston North for the MFI and sarcomere length testing.

The left loins remained food grade and were vacuum packed, then the carcass ticket was placed in a vacuum pack bag and packaged inside the second bag with the loin, these were then boxed as standard and stored in the chiller at Duncan & Company at approximately -1.5°C. After 8 weeks

the samples were transported in chiller transport to Lincoln University, Lincoln, Christchurch for the sensory analysis. Cooked and raw sub-samples from the left loins used in the sensory trial were frozen and transported to AgResearch Lincoln Research Centre for the RIEMS testing.

# 3.5 Consumer Sensory Analysis

The consumer sensory analysis was carried out at Lincoln University. The sensory evaluation study was approved by the Lincoln University Human Ethics committee (Application No: 2018-44). The consumers were required to answer a series of demographic and frequency of meat consumption questions before the tasting was carried out (Appendix 7.2). In preparing the loins, approximately 2 cm of the raw loins were cut off from the short end of the loins and then the four loins (Past-F, Past-M, PKE-F and PKE-M) were cooked in a convection oven to an internal temperature of 71°C. The cooked samples were then cut into small bite sizes  $(1.5 \times 1.5 \times 1.5 \times 1.5 cm^3)$  and these were then wrapped in individual coded aluminium and served to the consumers. Over 10 sessions, 104 consumers rated these four samples on the following sensory attributes using a 0-100 hedonic scale: overall liking, flavour, degree of juiciness and tenderness. The consumers were then asked to rate the quality of each meat sample of which they judged them to be, using the following four categories: 'unsatisfactory', 'good every day', 'better than every day' and 'premium' product.

# 3.6 Intramuscular fatty acid composition and content

The intramuscular fatty acid content and composition was measured according to the same procedure as lamb (Craigie et al. 2017). A direct trans-methylation method was employed for fatty acid analysis, which combined the extraction and esterification of lipids in a single step to generate fatty acid methyl esters (FAME). From each sample, 300 mg of freeze dried meat powder were placed into Kimax 15 mL tubes to which 4 mL of toluene and 300 µL of internal standard (C11 TAG in toluene) were added with a volumetric pipette under a fume hood. Then 4 mL of 5% methanolic sulphuric acid was added. After adding the solutions the tubes were tightly fitted with caps and mixed thoroughly then incubated at 70°C for 2 h, with mixing every 30 min during the incubation time. The tubes were left to equilibrate to room temperature for 25 min before 5 mL of saturated NaCl was added. The tubes were shaken vigorously and then centrifuged at 2300 rpm for 2 min into separate solvent layers.

The top FAME layer was transferred into 1.5 mL GLC auto sampler vials (Thermo Fisher). The GLC was a Shimadzu GC-2010 plus (Shimadzu Corporation, Kyoto, Japan) with a flame ionisation detector (FID). The column was a Restex RTX 2330 column of 105 m length, 0.25 mm i.d., and 0.20  $\mu$ m film thickness. The thermal programme had an initial temperature of 175°C, this was held for 17 min, the temperature was increased to 220°C at a rate of 6°C per minute and held for 10 min. The carrier gas was hydrogen with a linear velocity of 50 cm/s (3.05 mL/min). The injection volume

was 1  $\mu$ L, with a split ratio 80:1. The injector temperature was 260°C, the detector temperature was 300°C.

The peaks were identified and quantified by using internal standard (C11:0) and theoretical FID response factors. The equations for generating the response and conversion factors to quantify individual fatty acids from the FAME's were obtained from American Oil Chemists' Society Official Methods.

This method generates quantitative data for 34 fatty acids and their corresponding triglycerides (TAG). From the FA data, the following nine key FA attributes were calculated (note: some FA were not present in venison):

- 1) Omega-3: Sum of C18:3 n3, C20:5 n3, C22:5 n3 and C22:6 n3.
- 2) Omega-6: Sum of C18:2 n6, C20:3 n6 and C20:4 n6.
- 3) The n3/n6 ratio: Omega-3/Omega-6.
- Saturated fatty acids (SFA): Sum of C10:0, C12:0, C14:0, C15:0, C16:0, C17:0, C18:0, C20:0 and C24:0.
- 5) Monounsaturated fatty acids (MUFA): Sum of C14:1, C16:1, C17:1, C18:1 t9, C18:1 t11, C18:1 c9, C18:1 c11, C20:1, C22:1 and C24:1.
- Polyunsaturated fatty acids (PUFA): Sum of C18:2 n6, C18:3 n3, C20:3 n6, C20:4 n6, C20:5 n3, C22:5 n3, C22:6 n3, and 9 11 CLA.
- 7) Long-chain Omega-3 (L Omega-3): Sum of C20:5 n3, C22:5 and C22:6 n3.
- Eicosapentaenoic plus Docosahexaenoic acid (EPA+DHA): Sum of C20:5 n3 and C22:6 n3.
- 9) The atherogenic index (AI), which is a marker for cardiovascular disease, determined by the formula:

 $AI = 0.0378 \times C12 : 0 + 0.1512 \times C14 : 0 + 0.0378 \times C16 : 0 + 0.0018 \times C18 : 0 - 0.0178 \times MUFA - 0.0248 \times PUFA$ 

In addition to the FA attributes, the amount of intramuscular fat (IMF) was calculated by summing the TAG's and using the freeze drying conversion factor (see freeze drying) to convert from a freeze dried powder to the as supplied loin (wet weight).

#### 3.6.1 Freeze drying

The weight of frozen meat samples, in their trays complete with ID tag was electronically recorded using a calibrated 3 decimal place balance, connected to a laptop computer and barcode scanner. As samples were weighed, they were placed in ziplock bags. Prior to freeze drying, a straw was placed within the bag, making sure it reached to the bottom to allow full air circulation during freeze drying. Samples were placed on a large metal tray and inserted into a pilot scale Cuddon FD80 freeze dryer set to -30°C (Cuddon Freeze Dry, Blenheim, NZ). The meat was left at -30°C for 3 h

with a vacuum applied before stepping to -10°C for 4 h and 0°C for a final 72 h. On completion the sample tray was removed from the bag and weighed, with the tray weight also recorded. Freeze dried samples were then ground to a fine powder using a small domestic electric coffee grinder.

The moisture fraction was calculated using the following equation:

$$Moisture \ fraction = 1 - \frac{(weight of dried sample + tray (g) - weight of tray (g))}{(weight of wet sample + tray (g) - weight of tray (g))}$$

The conversion factor (weight of dried sample – weight of bag and tag) / (weight of wet sample – weight of bag and tag) was applied to the weight of the freeze dried meat to convert from concentration per gram of freeze dried meat to the concentration of the wet meat.

#### 3.7 Colour and Colour Stability

#### 3.7.1 Vacuum-packaged chilled storage

The vacuum-packaged chilled storage (VP colour) measurement was carried out at Invermay Agricultural Centre, Dunedin. The samples were vacuum packed, and chill aged at -1.5°C. Using a colourimeter (CM 700d Red Colour, CM700AE-16/006 Konica Minolta, Osaka) which was calibrated using a white calibrated cap with the CIE standard Illuminant D65 and at a 10° angle, the colour of venison was recorded every 14 days (Thursday 25<sup>th</sup> October, Thursday 8<sup>th</sup> November, Thursday 22<sup>nd</sup> November and Thursday 6<sup>th</sup> November). The colour measurement was carried out in triplicates (three different points on the meat) and the results were recorded as CIE Lab colour coordinates (Bixley et al. 2013). The L\* represents the lightness of the colour (black=0 and white= 100), a\* represents the redness of the colour (green (-) and red (+)) and b\* represents the yellowness of colour (blue (-) and yellow (+)). These triplicates were measured through the sealed vacuum pack.

#### 3.7.2 Retail display colour stability

The retail display colour stability (RD colour) was carried out at the Hopkirk Research Institute. RD colour was measured following the VP colour measurement and was measured over a period of 14 days. The samples were first trimmed to allow the fresh meat surface to be exposed to the flat surface in the new vacuum pack. These samples were then covered and stored in the chiller at 4°C. On days 1, 2, 5, 7 and 14, the colour of these packed samples was measured through the packaging using the same method used for the VP colour measurement (Bixley et al. 2013).

#### 3.8 Drip loss and pH

Drip loss was calculated for the samples reserved for tenderness testing during the 21 d storage period. The initial weight was recorded at 24 hours, and then the final weight was recorded once the sample was blotted dry with a paper towel immediately after aging. Drip loss is defined as the difference in weight after aging expressed as a percentage of the 24h weight.

The ultimate pH of venison loin samples was measured using a temperature compensated Hanna 99163 pH meter with a FC232D combined pH/temperature probe (Hanna Instruments, Rhode Island, USA). The pH meter was calibrated using a two-point calibration of pH buffers at pH 7.00 and pH 4.01 prior to use. The probe was inserted into each end of the loin sample reserved for tenderness testing at 24 h *post-mortem* and the average of these readings was taken as the final pH<sub>24</sub> reading.

#### 3.9 Tenderness

The meat tenderness and pH were measured at the Invermay Agricultural Centre. Venison tenderness was measured according to Cervena brand requirements after 21 days chill aging in a vacuum pack at 0°C using a MIRINZ tenderometer (Macfarlane & Marer 1966) (MIRINZ Inc., Hamilton, New Zealand). For each cooked venison loin sample (average weight = 258.1 g), ten 10 mm×10 mm cross sectional sub-samples were prepared and the shear force (kgF) was tested perpendicular to the muscle fibre direction. The average shear force value (kgF) of the 10 replicates is reported as the final reading of shear force for each meat sample received.

#### 3.10 Myofibrillar Fragmentation Index and Sarcomere length

Myofibrillar fragmentation index and sarcomere length were measured at the Massey University. Myofibrillar fragmentation index was measured by filtration through 231µm mesh following a standard homogenization (Purchas et al. 1997). Sarcomere length was measured by laser diffraction (helium-neon gas laser, 632.8 nm, Melles Griot, Carlsbad, CA, USA) (Coleman et al. 2016).

#### 3.11 Confinement Odour Analysis

The confinement odour analysis was carried out at the Hopkirk Research Institute. The samples were collected from the fridge and placed on a small table at room temperature allowing easy access from all sides. Three panellists were positioned around the table and quickly sniffed the samples immediately after they were opened. Confinement odour dissipates rapidly (Johnson 1991), therefore three panellists are the highest practicable number that can assess the sample in a short time. The samples were then kept at room temperature for 10 min after which the panellists sniffed them again. Each panellist described the type of aroma (notes) they noticed. The same

three people performed the analysis of all the samples. The presence of confinement odour was defined as the detection of odour in the first sniff but not in the second. If only one person detected the presence of confinement odour it is given a score 1, if two detected confinement odour it is scored 2 and if all three panellists detected the confinement odour it was scored 3. Confinement odour was only identified if all three panellists did not detect any odour in the second sniff. Samples exhibiting odours after 10 min were considered persistent odours. Those samples without odour in either evaluation were reported as "1" in accordance with the hedonic scale references from Gribble et al. (2014).

# 3.12 Microbial Analysis

The microbial analysis was carried out at the Hopkirk Research Institute. The surface of the meat pack and scissors used for opening the packs were disinfected by alcohol wipes prior to the bag being opened. Gloves were changed between samples. The surface of each sample was swabbed by the wet–dry swab method (AgResearch 2009). Briefly, the surface of the meat was delineated by a sterile 25 cm<sup>2</sup> circular cardboard template and a pair of sterile swabs moistened with peptone diluent (Fort Richard Laboratories, Auckland, New Zealand) were rubbed vertically, horizontally and diagonally across the entire exposed area. The process was repeated using a pair of unmoistened sterile swabs. After swabbing, both pairs of swabs were placed into a 15-mL sterile peptone diluent tube containing five–six glass beads which were then shaken for 2 min using a vortex mixer. Decimal dilutions of the swab suspensions were prepared with sterile 0.1% peptone diluent prior to enumeration of selected bacteria.

Aerobic bacteria counts (APC) were obtained by plating out a 0.1 mL aliquot of each dilution on plate count agar (Fort Richard Laboratories) prior to incubation at 25 °C for 72 h (AgResearch 2013). Lactic Acid Bacteria were enumerated by plating out a 0.1 mL aliquot of each dilution onto MRS agar prior to incubation at 25 °C for 72 h (AgResearch 2001). Plates with 30–300 colony forming units (cfu) were selected for enumeration. The results were transformed to logarithms and expressed as  $log_{10}$  cfu/cm<sup>2</sup> of meat.

# 3.13 Rapid Evaporative Ionisation Mass Spectrometrey (REIMS)

Rapid Evaporative Ionisation Mass Spectrometry (REIMS) was carried out at the AgResearch Lincoln Research Centre, Lincoln, using a commercial REIMS system (Waters, Wilmslow, UK) coupled to a quadrupole-time of flight (qTOF) mass spectrometer (Xevo G2-XS, Waters). Samples (40 raw and 40 cooked venison loin) were provided as 2 x 2 x 1 cm<sup>3</sup> medallions that were cut while semi-frozen using a diathermy knife (Erbe Medical UK Ltd, Leeds, UK) at a voltage of 15 V. Isopropanol was infused into the REIMS source at 100 uL/min using a syringe pump (Harvard). Mass spectra were collected at 0.5 Hz between 50-1500 m/z in TOF mode (i.e. no fragmentation). The Venturi pump of the REIMS was cleaned with ethanol every 30 burns. Each sample was burnt

3 times, and the average of the three burns used for further data analysis. Each sample was analysed in both positive and negative ionisation modes.

REIMS chronograms were processed into individual files using ProGenesis Bridge with the naturally occurring C18:1 fatty acid as a lockmass (m/z 281.2481 in negative ionisation mode) and then normalised to total peak intensity using ProGenesis QI (Waters). Tentative compound identification was based on accurate mass data matched against the Human Metabolome Database.

#### 3.13.1 REIMS Data Analysis

The relationship between REIMS fingerprints, feed and gender were explored using multivariate statistics. Initial data analysis used Principal Components Analysis (PCA), while Orthogonal Projection to Latent Structures-Discriminant Analysis (OPLS-DA) was used for targeted data analysis (i.e. focusing on specific comparisons such as feed). Multivariate data analysis was performed using Simca 15.0 (Umetrics AB, Umea, Sweden).

#### 3.14 Statistical Data Analysis

Carcass, meat quality and sensory traits were analysed by analysis of variance fitting Diet (PAST, PAST-PKE), Gender-Age (rising-two-year old females (F-R2) and rising-one-year old males (M-R1)) and their interaction using R (R-Core-Team 2018). Means and standard errors of difference between means were calculated using the R package predictmeans.

For brevity Gender-Age is referred to Gender or Sex.

# 4. Results

## 4.1 Animal Performance and handling

Throughout the 6 weeks of the trial, the live weight gain of PKE supplemented animals was higher than those on pasture without supplementary feeding (Table 1). Similarly, the live weight gain was higher for R1 stags than R2 Hinds. There was little impact of PKE supplementation on the body condition score. There was minimal impact of PKE supplementation on animal handling scores, although PKE-supplemented animals were slightly quieter in the weigh crate.

# 4.2 Carcass Characteristics

Palm Kernel Expeller supplementary feed did not have a significant effect on the carcass characteristics of the animals except for the dressing percentage. The dressing percentage of the deer from the pasture only group was significantly higher than the PKE group (P<0.05) (Table 1). The pasture only group had a dressing percentage of approximately 1% higher than the PKE group. The gender of the deer did have a significant effect on the carcass characteristics except for the leg/HCW percentage, the left and right cap off/on percentage and the cap off left/right percentage. For the characteristics with a significant sex effect (P<0.01 or P<0.05), values for the hinds were higher than for the stags (Table 1). The leg/HCW, left and right cap off/on and cap off left/right percentages were not significantly different between the treatment groups or the genders. However, there was a significant interaction between the group and sex for the right cap off/on percentage (P<0.05) (Table 1).

Animal/Carcass	Pas	ture	PK	E		P-valu	le
Characteristics							
	F	М	F	М	Group	Sex	Interaction
LW (kg)	105	86.4	107	84.7	0.685	0.000 <sup>1</sup>	0.368
LW 0 <sup>2</sup> (kg)	101	85.8	105	84.1	0.952	0.000	0.186
LW 1 <sup>3</sup> (kg)	104	91.6	107	89.5	0.778	0.000	0.300
LW 2⁴ (kg)	105	95.8	110	95.4	0.672	0.000	0.348
BCS W2 <sup>4</sup>	3.4	3.3	3.4	3.2	0.625	0.214	0.739
Aggression LW2 <sup>4</sup>	1.2	1.2	1.3	1.2	0.856	0.455	0.638
Agitation LW2 <sup>4</sup>	2.6	2.7	2.9	2.2	0.114	0.032	0.013
Handling Ease LW2 <sup>₄</sup>	2.8	3.2	3.2	2.4	0.332	0.634	0.183
Exit Speed LW2 <sup>4</sup>	2.8	2.2	1.8	2.2	0.207	0.747	0.160
In Crate LW2⁴	1.8	1.7	1.5	1.1	0.011	0.165	1.8
LW 3⁵ (kg)	105	98.1	111	99.5	0.244	0.001	0.336
BCS (LW3)	3.4	3.3	3.6	3.3	0.559	0.032	0.328
LWG <sup>6</sup> W0-W3 (g/day)	100	292	154	367	0.000	0.000	0.597
GR (mm)	6.0	3.94	5.8	3.85	0.539	0.000	0.863
HCW (kg)	60.4	55.1	62.6	55.2	0.617	0.000	0.498
Dressing (%)	57.5	56.2	56.2	55.6	0.014	0.013	0.390
LP (kg)	23.2	21.1	24.1	21.1	0.587	0.000	0.468
LP/HCW (%)	38.4	38.2	38.4	38.3	0.873	0.466	0.966
L SL Cap on (g)	1240	1100	1340	1100	0.512	0.000	0.259
R SL Cap on (g)	1220	1060	1290	1060	0.676	0.000	0.367
L SL Cap off (g)	810	707	824	719	0.664	0.000	0.960
R SL Cap off (g)	824	709	815	721	0.915	0.000	0.652
Total SL Cap on (g)	2460	2160	2630	2160	0.580	0.000	0.298
Total SL Cap off (g)	1630	1420	1640	1440	0.781	0.000	0.843
L cap off/on (%)	65.2	64.6	63.4	65.5	0.925	0.468	0.196
R Cap off/on (%)	67.7	67.0	64.9	68.1	0.814	0.130	0.017
Cap off L/R (%)	98.1	99.9	101.0	99.7	0.431	0.828	0.164
SL Cap on/ HWC (%)	4.08	3.91	4.18	3.91	0.746	0.002	0.411

Table 1: Carcass characteristics of venison from rising yearling male and rising-two-year old female deer fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets

Live weight (LW), fat depth over the 12th rib (GR), hot carcass weight (HCW), LP (leg pair weight), L (left side of animal), R (right side of animal), SL (short loin)

<sup>1</sup> (P-value 0.000 denotes  $\leq$  0.001)

<sup>2</sup> Live-weight at start of experiment (29/08/2018)

<sup>3</sup> Live-weight at day 13 of trial (11/09/2018)

<sup>4</sup> Live-weight at day 29 of trial (27/09/2018)
 <sup>5</sup> Live-weight at day 42 of trial (10/10/2018)

<sup>6</sup> Daily Live-weight gain over the 42 days of the trial

#### 4.3 Consumer Sensory Analysis

The consumers rated that there was no significant difference between the feed treatments for the degree of tenderness (P=0.71), liking of flavour (P=0.798) and the overall liking (P=0.502) of the venison samples. The sex of the animal had no significant impact on the sensory attributes (Table 2). The consumers rated that the degree of juiciness of the venison from the pasture only treatment to be higher than the venison from the PKE group (P<0.1). The juiciness for the venison from the pasture only group scored 6 points more than the PKE group on the hedonic scale (Table 2). The quality ratings between the treatment groups (P=0.542) and the genders (P=0.602) were not significantly different. The consumers overall rated that the quality of the venison to be 43% 'good everyday', 28% 'better than everyday' and 14% 'premium quality'' regardless of treatment group or sex (Fig. 1).

Table 2: Sensory attribute scores (0-100 hedonic scale, where 0 is dislike extremely, and 100 is like extremely) of venison from rising yearling male and rising-two-year old female deer fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets

Pasture		PKE		P-value		
Female	Male	Female	Male	Group	Sex	Interaction
69.0	71.7	70.4	68.0	0.714	0.974	0.405
62.6	65.6	58.3	58.1	0.092	0.683	0.635
59.7	57.7	58.4	57.5	0.798	0.619	0.844
60.9	60.0	58.4	57.9	0.502	0.834	0.956
2.6	2.6	2.3	2.2	0.542	0.602	0.970
	69.0 62.6 59.7 60.9	69.071.762.665.659.757.760.960.0	69.071.770.462.665.658.359.757.758.460.960.058.4	69.071.770.468.062.665.658.358.159.757.758.457.560.960.058.457.9	69.071.770.468.00.71462.665.658.358.10.09259.757.758.457.50.79860.960.058.457.90.502	69.071.770.468.00.7140.97462.665.658.358.10.0920.68359.757.758.457.50.7980.61960.960.058.457.90.5020.834

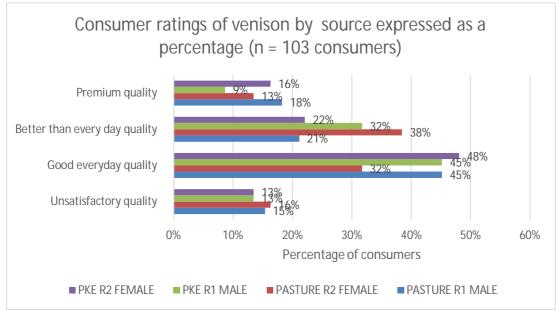


Figure 1: Percentage of overall quality level ratings by the consumers

#### 4.4 Intramuscular fatty acid content and composition

The intramuscular fatty acid percentage (IMF%) was significantly higher in the PKE treatment group and the R2 female deer (P<0.01). The differences in IMF% were quite small, the R2 female had a mean IMF% >0.34% than the R1 males and the PKE group had a mean IMF% >0.32% than the pasture only group. A significant interaction between the gender and feed treatment was identified for IMF % (Table 4).

The omega 3 fatty acids (n3) present in the venison from the pasture only group were significantly higher than the PKE group (P<0.01). The pasture only group had >0.5 mg of n3 than the PKE group. There was no significant sex effect on the total n3 present in the venison (P=0.917). The total omega 6 fatty acids (n6) in the venison from the R2 females were significantly higher than the R1 males (P<0.01). The R2 females had >0.6 than the R1 males. The treatment group had no significant effect on the amount of n6 fatty acids present in the venison. The n3/n6 ratio was significantly higher in the venison from the pasture only group and the R1 males (P<0.01). The difference between the treatment groups was 0.13mg and the difference between the sex of the animal was 0.09 mg. There were no significant differences between the treatment groups (P=0.309) and the sex of the animals (P=0.868) for long-chained omega 3 fatty acids present in the venison (Table 3).

The total saturated fatty acid (SFA) and monounsaturated fatty acid (MUFA) content present in the PKE group was significantly higher than the pasture only group (P<0.01) (Table 3). These fatty acids were significantly higher in the R2 females compared to the R1 males. The interaction between the sex and the group was significant for the total MUFA (P<0.05) and there was a slight interaction identified for the total SFA (P=0.07). The total poly-unsaturated fatty acids (PUFA)

present in the pasture group was significantly higher compared to the PKE group. The venison from the R2 females had a slightly higher total PUFA compared to the R1 males (P=0.07).

The following fatty acids and their corresponding triglycerides were significantly different between the treatment groups and genders: C12:0, C14:0, iso C17:0, antesio C17:0, C18:1 t11 and 9 11 CLA. Each of these fatty acids and their corresponding triglycerides were significantly higher in the venison from the PKE treatment group than the pasture only group (P<0.01 or P<0.05). Venison from R2 females had higher amounts of C14:0, iso C17:0, antesio C17:0, C18:1 t9 and 9 11 CLA than R1 males (P<0.01 or P<0.05). However, the meat from the R1 males did have a significantly higher amount of C12:0 compared to the R2 females (P<0.01).

The following fatty acids and their corresponding triglycerides had significant group and gender effects, and interaction: C14:1, C16:0, C16:1, C18:c11, C18:1 c9 and C22:1. Venison from the PKE group had significantly higher amounts of C14:1, C16:0, C16:1 and C18:1 c9 (P<0.01) than the pasture only group. The PKE group was also higher in C18:1 c11 and C22:1 than the pasture group (P<0.05). Venison from R2 females had higher amounts of C14:1, C16:0, C16:1, C16:0, C16:1, C18:1 c11 and C18:1 c9 (P<0.01). The C22:1 fatty acid was significantly higher in the R1 males (P<0.01). Significant treatment group and sex interactions were identified for these fatty acids and their corresponding triglycerides (p<0.05).

The amount of iso C15:0 and iso C16:0 found in the venison were significantly different between the genders of the deer. These fatty acids were significantly higher in the venison from the R2 females compared to the R1 males (P<0.01). There were significant sex effects on the following fatty acids and their corresponding triglycerides: antesio C15:0, C15:0, C17:0, C18:0 and C18:2 n6. The venison from the R2 females had significantly higher amounts of these fatty acids and their corresponding triglycerides than the R1 males (P<0.01).

The feeding treatments had significant effects on the C17:1 and C18:3 n3 fatty acids (P<0.01). C17:1 was significantly higher in the venison from the PKE group, while the C18:3 n3 was significantly lower in the PKE group. The C20:4 n6 fatty acids and their corresponding triglycerides were higher in the PKE group compared to the pasture group (P<0.1).

C18:1 t9, C22:2, C20:5 n3, C24:0, C24:1 and C22:6 n3 fatty acids and their corresponding triglycerides were not significantly different between the treatment groups or the genders (Tables 4 &5).

C22:5 fatty acid and its corresponding triglyceride was not significantly different between the treatment groups (p=0.215) or the genders (p=0.759), however a significant treatment and sex interaction was identified (p<0.1). Eicosapentaenoic plus Docosahexaenoic acid (EPA+DHA) present in the venison was not significantly different between the treatment groups (p=0.415) or the gender of the deer (p=0.941). The atherogenic index (AI) was significantly higher in the R2 females (p<0.05) and the PKE group (p<0.01) (Table 3).

Table 3: Intramuscular fatty acid content and composition of venison from R1 stags (Males) and R2 hinds (Females) fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets for six weeks prior to slaughter.

Composition (mg)	Pas	ture	PK	Έ		P-val	ue
	Female	Male	Female	Male	Group	Sex	Interaction
Sum of SFA	7.75	6.91	9.86	7.59	0.003	0.000 <sup>1</sup>	0.066
Sum of MUFA	4.09	3.68	6.42	4.76	0.000	0.000	0.013
Sum of PUFA	6.65	6.54	6.65	5.67	0.048	0.070	0.152
Omega 3	2.71	2.88	2.39	2.20	0.000	0.917	0.156
Omega 6	3.90	3.64	4.20	3.45	0.830	0.006	0.173
n3/ n6 ratio	0.690	0.793	0.572	0.640	0.000	0.000	0.195
L-omega 3	1.59	1.73	1.83	1.72	0.309	0.868	0.155
EPA + DHA	0.064	0.048	0.064	0.045	0.415	0.941	0.254
AI	-0.016	-0.038	0.049	0.02	0.000	0.026	0.751
SUM FA	20.0	18.6	25.0	19.8	0.005	0.000	0.037
Unreported FA	0.75	0.74	1.08	1.03	0.000	0.560	0.703
C10:0	0.000	0.000	0.000	0.000	-	-	-
C12:0	0.067	0.089	0.083	0.100	0.006	0.000	0.701
C14:0	0.459	0.391	0.829	0.675	0.000	0.020	0.362
iso C15:0	0.023	0.002	0.032	0.00	0.728	0.000	0.096
anteiso C15:0	0.054	0.041	0.066	0.046	0.105	0.000	0.397
C14:1	0.170	0.137	0.25	0.16	0.005	0.000	0.053
C15:0	0.086	0.076	0.094	0.076	0.581	0.004	0.346
iso C16:0	0.029	0.004	0.036	0.000	0.784	0.000	0.066
C16:0	3.81	3.21	5.11	3.53	0.004	0.000	0.028
iso C17:0	0.136	0.116	0.188	0.149	0.000	0.000	0.132
C16:1	0.747	0.669	1.21	0.795	0.000	0.000	0.012
anteiso C17:0	0.055	0.038	0.065	0.044	0.025	0.000	0.525
C17:0	0.076	0.070	0.089	0.697	0.287	0.002	0.118
C17:1	0.043	0.045	0.052	0.052	0.003	0.568	0.621
C18:0	3.25	3.06	3.65	3.12	0.209	0.012	0.248
C18:1 t9	0.000	0.005	0.039	0.009	0.345	0.293	0.982
C18:1 t11	0.242	0.163	0.320	0.22	0.001	0.000	0.583
C18:1 c9	2.18	1.92	3.65	2.76	0.000	0.000	0.031
C18:1 c11	0.668	0.685	0.890	0.71	0.017	0.037	0.014
C18:2 n6	3.04	2.79	3.20	2.55	0.419	0.002	0.162
C20:0	0.000	0.000	0.000	0.000	-	-	-
C18:3 n3	1.77	1.88	1.33	1.2	0.000	0.828	0.144
9 11 CLA	0.041	0.012	0.063	0.024	0.010	0.000	0.386
C20:4 n6	0.861	0.855	0.995	0.897	0.077	0.215	0.288
C22:1	0.037	0.0516	0.049	0.053	0.050	0.001	0.067
C22:2	0.010	0.0276	0.000	0.002	0.157	0.418	0.112
C20:5 n3	0.766	0.839	0.876	0.827	0.473	0.805	0.173
C24:0	0.005	0.000	0.007	0.027	0.467	0.121	0.914
C24:1	0.000	0.005	0.000	0.003	0.658	0.121	0.726
C22:5	0.655	0.722	0.766	0.721	0.038	0.759	0.083
C22:6 n3	0.168	0.165	0.186	0.176	0.213	0.675	0.824
	$\frac{0.100}{0.000}$		0.100	0.170	0.004	0.075	0.024

1 (P-value 0.000 denotes  $\le$  0.001)

Table 4: Intramuscular triglyceride content and composition of venison from R1 stags (Males) and R2 hinds (Females) fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets for six weeks prior to slaughter.

composition (mg)	Past	ture	PK	E		P-val	ue
	Female	Male	Female	Male	Group	Sex	Interaction
IMF%	2.1	1.95	2.62	2.08	0.005	0.000 <sup>1</sup>	0.037
SUM TAG	21.0	19.5	26.2	20.8	0.005	0.000	0.037
Unreported TAG	0.785	0.775	1.13	1.08	0.000	0.560	0.703
C10:0	0.000	0.000	0.000	0.000	-	-	-
C12:0	0.072	0.095	0.089	0.107	0.006	0.000	0.701
C14:0	0.485	0.413	0.876	0.713	0.000	0.020	0.362
iso C15:0	0.024	0.002	0.034	0.000	0.728	0.000	0.096
anteiso C15:0	0.056	0.043	0.069	0.048	0.105	0.000	0.397
C14:1	0.179	0.145	0.265	0.169	0.005	0.000	0.053
C15:0	0.090	0.080	0.099	0.080	0.581	0.004	0.346
iso C16:0	0.030	0.004	0.038	0.00	0.784	0.000	0.066
C16:0	4.00	3.38	5.37	3.71	0.004	0.000	0.028
iso C17:0	0.142	0.012	0.197	0.156	0.000	0.000	0.132
C16:1	0.785	0.703	1.27	0.835	0.000	0.000	0.012
anteiso C17:0	0.058	0.040	0.068	0.046	0.025	0.000	0.525
C17:0	0.080	0.073	0.093	0.073	0.287	0.002	0.118
C17:1	0.045	0.048	0.054	0.055	0.003	0.568	0.621
C18:0	3.40	3.20	3.81	3.26	0.209	0.012	0.248
C18:1 t9	0.000	0.005	0.004	0.009	0.345	0.293	0.982
C18:1 t11	0.254	0.170	0.335	0.230	0.001	0.000	0.583
С18:1 с9	2.28	2.01	3.82	2.88	0.000	0.000	0.031
C18:1 c11	0.698	0.717	0.931	0.742	0.017	0.037	0.014
C18:2 n6	3.18	2.92	3.35	2.67	0.419	0.002	0.162
C20:0	0.000	0.000	0.000	0.000	-	-	-
C18:3 n3	1.85	1.95	1.39	1.25	0.000	0.828	0.144
9 11 CLA	0.043	0.013	0.066	0.025	0.010	0.000	0.386
C20:4 n6	0.901	0.894	1.04	0.939	0.077	0.215	0.288
C22:1	0.039	0.054	0.051	0.055	0.050	0.001	0.067
C22:2	0.010	0.003	0.000	0.002	0.157	0.418	0.112
C20:5 n3	0.799	0.875	0.913	0.863	0.473	0.805	0.173
C24:0	0.005	0.012	0.007	0.015	0.467	0.121	0.914
C24:1	0.000	0.005	0.000	0.003	0.658	0.110	0.726
C22:5	0.683	0.752	0.798	0.751	0.215	0.759	0.083
C22:6	0.174	0.171	0.193	0.183	0.384	0.675	0.824

1 (P-value 0.000 denotes  $\leq 0.001$ )

#### 4.5 Colour and Colour Stability

#### 4.5.1 Vacuum-packaged chilled storage colour

L\* colour coordinates measured on  $25^{\text{th}}$  October (2-weeks post-slaughter- Colour day 14) were significantly higher in the venison from the PKE group than the pasture group (P<0.01) (Table 5). The a\* (redness) colour coordinates were higher in the venison from the pasture only group than the PKE group (P<0.10). The b\* (yellowness) colour coordinates were not significantly different between treatment groups (P=0.127). L\* (lightness) (P<0.01), a\* (P<0.05) and b\* (P<0.01) coordinates were significantly influenced by the gender of the deer. The meat from the R1 males had a higher L\* and b\* and a lower a\* coordinate than the R2 females (Table 5).

L\*, a\* and b\* coordinates measured on 8<sup>th</sup> November (colour day 28) were significantly different between the genders of the animals (P<0.01) (Table 5). R1 males had higher L\* and b\* coordinates than R2 females, while R2 females had higher a\* values than R1 males. The a\* coordinate was higher in venison from the PKE group than the pasture only group (P<0.1). The feeding treatment had no significant effect on the L\* (P=0.133) and b\* (P=0.935) coordinates (Table 5).

L\* and b\* coordinates measured on  $22^{nd}$  November (colour day 42) were significantly higher in R1 males than R2 females (P<0.01) (Table 5). The venison from the PKE group had a significantly higher L\* coordinate than the pasture group (P<0.01). The feeding treatments had no significant effect on the a\* (P=0.109) and b\* (P=0.902) coordinates. There were no significant differences in a\* values between the genders (P=0.173).

L\*, a\* and b\* colour coordinates measured on 6<sup>th</sup> December (colour day 56) were significantly different between the sex of the animals (P<0.01 for L\* and a\*, P<0.05 for b\*). The R1 males had higher L\* and b\* coordinates than R2 females, while the R2 females had higher a\* values than R1 males. The L\* (P=0.246), a\* (P=0.267) and b\* (P=0.928) were not significantly different between the treatment groups (Table 5).

VP Colour	Past	ure	PK	E		P-val	lue
Measurements							
	Female	Male	Female	Male	Group	Sex	Interaction
Day 14							
(25/10/2018)							
L*	31.6	33.7	32.2	34.5	0.003	0.000 <sup>1</sup>	0.420
a*	10.8	10.6	10.5	10.3	0.055	0.013	0.667
b*	6.18	6.50	6.26	6.72	0.127	0.003	0.528
Day 28							
(8/11/2018)							
L*	30.4	32.9	30.2	33.6	0.133	0.000	0.110
a*	10.6	10.3	11.0	10.5	0.070	0.002	0.180
b*	6.10	6.60	5.90	6.70	0.935	0.000	0.206
Day 42							
(22/11/2018)							
L*	30.3	32.6	30.7	33.4	0.001	0.000	0.330
a*	10.4	10.2	10.3	10.0	0.109	0.173	0.673
b*	6.10	6.60	5.90	6.60	0.902	0.000	0.250
Day 56							
(6/12/2018)							
L*	30.5	32.7	30.3	33.5	0.246	0.000	0.265
a*	10.9	10.5	10.9	10.2	0.267	0.002	0.420
b*	6.12	6.59	6.22	6.51	0.928	0.021	0.597

Table 5: Vacuum packaged chilled storage (VP) colour of venison *m. longissimus lumborum* from R1 stags (Males) and R2 hinds (Females) fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets for six weeks prior to slaughter.

<sup>1</sup> (P-value 0.000 denotes  $\leq$  0.001)

Between days 14 and 28 the colour of the venison became significantly darker (- $\Delta$ L\*) from the R2 females of both treatment groups (P<0.05). The redness (- $\Delta$ a\*) of the venison from the PKE group increased significantly more than the pasture group (P<0.01). In the pasture only group the yellowness (+ $\Delta$ b\*) increased significantly more in the venison from the R1 male (P<0.01). The total colour difference was higher in the R2 females than the R1 males for both treatments (P<0.1) (Table 6).

By day 42, there were no significant differences between the treatment groups and sex of animal for  $\Delta L^*$ ,  $\Delta a^*$  and  $\Delta E$  (Table 6). The yellowness increased significantly in venison from the pasture group (P<0.05) and the R1 males (P<0.05). At the end of the 10-week storage period there were no significant differences in colour differences for the L<sup>\*</sup>, a<sup>\*</sup>, b<sup>\*</sup> coordinates or the total colour difference (E) in the venison from the treatment groups or the sex of the deer (Table 6).

Change in VP colour	Past	ure	PKE			P-value		
	Female	Male	Female	Male	Group	Sex	Interaction	
Days 14-28								
$\Delta L^*$	-1.19	-0.76	-2.02	-0.98	0.167	0.029	0.378	
$\Delta$ a*	-0.25	-0.14	0.52	0.17	0.004	0.503	0.160	
$\Delta \mathbf{b^*}$	-0.059	0.082	-0.380	-0.025	0.141	0.059	0.399	
$\Delta \mathbf{E}$	1.44	1.22	2.17	1.31	0.286	0.085	0.284	
Days 14-42								
$\Delta L^*$	-1.28	-1.11	-1.30	-1.12	0.977	0.504	0.981	
∆ <b>a*</b>	-0.42	-0.29	-0.40	-0.27	0.837	0.312	0.984	
$\Delta \mathbf{b^*}$	-0.090	0.089	-0.390	-0.071	0.034	0.016	0.475	
$\Delta \mathbf{E}$	1.46	1.40	1.57	1.54	0.504	0.799	0.947	
Days 14-56								
$\Delta L^*$	-1.09	-0.99	-1.85	-1.27	0.363	0.516	0.636	
$\Delta$ a*	0.096	0.006	0.400	-0.093	0.838	0.164	0.319	
$\Delta \mathbf{b^*}$	-0.060	0.091	-0.046	-0.235	0.211	0.950	0.325	
$\Delta \mathbf{E}$	2.07	2.01	2.01	1.99	0.905	0.910	0.957	

Table 6: Vacuum packaged chilled storage (VP) colour change of venison over 10-week period from R1 stags (Males) and R2 hinds (Females) fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets for six weeks prior to slaughter.

#### 4.5.2 Retail display colour stability

The feeding treatments had no significant effect on the colour coordinates: L\* (P=0.975), a\* (P=0.794) and b\* (P=0.987) on day 1. The R1 males had higher L\* and b\* coordinates than R2 females (P<0.01). The sex of the animal had no significant effect on the a\* coordinate (P=0.105) (Table 7).

On day 3, the feed treatments had no significant effect on the colour coordinates: L\* (P=0.286), a\* (P=0.569) and b\* (P=0.396). The sex of the deer had no significant effect on the a\* coordinate (P=0.469). The venison from the R1 males had a significantly higher L\* (P<0.01) and b\* (P<0.05) than R2 females (Table 7).

The feeding treatments had no significant effects on the L\* (P=0.222) and b\* (P=0.339) on day 5. The a\* coordinates were higher in the venison from the PKE group than the pasture group (P<0.1). The sex of the animals had no significant effect on the a\* coordinate (P=0.446). The venison from R1 males had significantly higher L\* and b\* coordinates than the R2 females (P<0.01) (Table 7).

The L\* coordinate of venison from the pasture only group was higher than the PKE group on day 7 (P<0.1) (Table 7). The a\* coordinate was significantly higher in the PKE group than the pasture only group (P<0.01). L\* and b\* coordinates were significantly higher in R1 males than R1 females (P<0.01). The a\* coordinates were not significantly different between the genders (P=0.292). The treatment group had no significant effect on the b\* coordinate (P=0.666).

The feeding treatments had no significant effect on the L\* (P=0.566) or the b\* (P=0.347) on day 14. The a\* coordinate was significantly higher in the venison on the PKE treatment than the pasture only treatment (P<0.05). The sex of the deer had no significant effect on the a\* coordinate (P=0.370). Venison from the R1 males had significantly higher in L\* and b\* coordinates than the R2 females (p<0.01) (Table 7).

Colour Measurements	Past	ure	PK	E		P-val	ue
	Female	Male	Female	Male	Group	Sex	Interaction
Day 1							
L*	35.2	37.2	35.1	37.2	0.975	0.000 <sup>1</sup>	0.920
a*	10.1	10.4	9.9	10.4	0.794	0.105	0.680
b*	5.81	6.44	5.68	6.50	0.987	0.000	0.541
Day 3							
L*	35.2	37.4	35.1	36.4	0.286	0.004	0.389
a*	10.6	10.5	10.2	10.6	0.569	0.469	0.303
b*	5.90	6.37	5.63	6.26	0.396	0.023	0.717
Day 5							
L*	34.4	36.9	34.2	36.2	0.222	0.000	0.532
a*	10.7	10.7	10.8	11.2	0.084	0.446	0.456
b*	5.63	6.64	5.53	6.40	0.339	0.000	0.724
Day 7							
L*	35.0	37.1	34.3	36.4	0.094	0.000	0.970
a*	10.5	10.7	11.1	11.3	0.006	0.292	0.889
b*	5.72	6.63	5.80	6.70	0.666	0.000	0.974
Day 14							
L*	35.2	37.2	35.1	36.9	0.566	0.000	0.769
a*	10.9	10.9	11.1	11.7	0.019	0.370	0.215
b*	5.98	6.68	6.18	6.84	0.347	0.001	0.920

Table 7: Retail display colour stability (RD) colour of venison from R1 stags (Males) and R2 hinds (Females) fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets for six weeks prior to slaughter.

<sup>1</sup> (P-value 0.000 denotes  $\leq 0.001$ )

There were significant differences in changes in venison colour between day 1 and day 2 (Table 8). The venison from the R2 females became significantly darker ( $-\Delta L^*$ ) than the R1 males (P<0.01) and there was a significant interaction between sex and feeding treatment for  $\Delta L^*$  (P<0.05). The redness of the venison from the PKE group increased slightly more than the pasture group (P<0.1), while it increased significantly more in the venison from the R1 males than the R2 females (P<0.05). Overall the total colour difference ( $\Delta E$ ) between day 1 and 2 was significantly higher in venison from the PKE group and the R1 males (P<0.01).

Between days 1 and 7, there were no significant colour changes for  $\Delta L^*$ ,  $\Delta b^*$  and  $\Delta E$  between the treatment group or sex of the deer (Table 8). However the redness of the venison from the PKE group increased significantly more than the pasture group (P<0.05).

By day 14 there were significant changes in venison colour between the treatment groups and genders (Table 8). The venison from the R1 males on the PKE treatment was darker than the males and females from the pasture group (P<0.1). The redness of the venison increased significantly more in the PKE group the pasture group (P<0.01). The total colour difference was significantly higher in the PKE group (P<0.01) and the males from the PKE group (P<0.05). There was a significant interaction between the groups and sex of animals (P<0.05) for  $\Delta E$ .

Change in colour	Past	ure	PK	E		P-val	ue
	Female	Male	Female	Male	Group	Sex	Interaction
Days 1-2							
$\Delta L^*$	0.167	-0.270	0.447	-2.28	0.120	0.008	0.045
∆ <b>a*</b>	0.190	0.556	0.478	1.33	0.080	0.048	0.403
$\Delta b^*$	0.116	0.033	0.287	-0.212	0.882	0.252	0.407
$\Delta \mathbf{E}$	0.985	1.58	1.54	2.73	0.010	0.008	0.325
Days 1-5							
$\Delta L^*$	-0.800	-0.331	-0.897	-1.06	0.225	0.700	0.467
∆ <b>a*</b>	0.543	0.260	0.883	0.727	0.127	0.435	0.823
$\Delta b^*$	-0.185	0.202	-0.150	-0.107	0.301	0.238	0.360
$\Delta \mathbf{E}$	1.43	1.37	1.65	2.06	0.154	0.672	0.535
Days 1-7							
$\Delta L^*$	-0.146	-0.110	-0.788	-0.866	0.114	0.968	0.904
$\Delta$ a*	0.403	0.309	1.14	0.909	0.019	0.570	0.810
$\Delta b^*$	-0.095	0.193	0.125	0.196	0.651	0.329	0.566
$\Delta \mathbf{E}$	1.64	1.72	1.86	2.14	0.252	0.582	0.749
Days 1-14							
$\Delta L^*$	-0.020	-0.008	-0.030	-0.366	0.068	0.104	0.198
∆ <b>a*</b>	0.796	0.428	1.16	1.22	0.000 <sup>1</sup>	0.174	0.099
$\Delta b^*$	0.163	0.243	0.502	0.338	0.148	0.695	0.512
$\Delta \mathbf{E}$	1.58	1.57	1.73	2.32	0.003	0.035	0.044

Table 8: Retail display colour stability (RD) change of venison from R1 stags (Males) and R2 hinds (Females) fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets for six weeks prior to slaughter (from day 1 of new trimming to day 14).

<sup>1</sup> (P-value 0.000 denotes  $\leq 0.001$ )

#### 4.5.3 Vacuum pack and bloom colour scores

The vacuum pack colour score was significantly higher in the venison from the R2 females than R1 males (P<0.05). The feeding treatment had no significant effect on the vacuum pack colour score (P=0.571). The bloom colour score of the venison was not significantly different between the treatment groups (P=0.636) or the genders (P=0.504) (Table 9).

Table 9: Vacuum pack and bloom colour scores for venison from R1 stags (Males) and R2 hinds
(Females) fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets for six weeks
prior to slaughter.

Colour scores	Pasture		PK	E	P-value			
	Female	Male	Female Male		Group	Sex	Interaction	
VP colour	1.3	1.1	1	1.1	0.571	0.025	0.100	
Bloom	1.1	1.3	1.2	1.1	0.636	0.504	0.068	

Vacuum pack colour (VP)

#### 4.6 Drip Loss and pH

The pH 24h post-mortem of the venison was not significantly different between the treatment groups (P=0.861) or the gender of the deer (P=0.421) (Table 10). The 24-hr drip loss (P<0.05) and drip loss percentage (P<0.05) were statistically significant between the treatment groups, as the venison from the PKE group had a >0.5% drip loss than the pasture only group. There were no significant differences in gender for either the drip loss (P=0.487) or the drip loss percentage (P=0.972) (Table 10).

Table 10: Drip loss and pH of venison from R1 stags (Males) and R2 hinds (Females) fed either
pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets for six weeks prior to slaughter.

Meat Quality Traits	Pasture		PK	E	P-value			
	Female Male		Female	Male	Group	Sex	Interaction	
рН	5.26	5.26	5.24	5.28	0.861	0.421	0.355	
Drip loss (g)	6.20	5.72	7.60	7.25	0.011	0.487	0.914	
Drip loss (%)	2.24	2.30	2.92	2.88	0.009	0.972	0.831	

#### 4.7 Tenderness

The venison from the R2 females was significantly (P<0.05) more tender than that of the R1 stags. Shear forces required to 'bite' the venison from the stags were almost 1kg higher. However, at  $\leq$ 4kg shear force all venison was very tender. There was no significant difference in shear force between the treatment groups (P=0.751) (Table 11).

pasiule-only (Fas	sule) and 307	o pasiure.	<u> J0 /0 FNL (F</u>		I SIX WEEP	s phot t	o slaughter.
	Pastur	e	PKE	P-value			
	Female	Male	Female	Male	Group	Sex	Interaction
Tenderness (kgF)	3.25	4.21	3.23	4.01	0.751	0.026	0.816

Table 11: Shear forces of venison from R1 stags (Males) and R2 hinds (Females) fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets for six weeks prior to slaughter.

#### 4.8 Myofibrillar Fragmentation Index and Sarcomere Length

The treatment group (P=0.739) and sex of the deer (P=0.164) did not have a significant effect on the sarcomere length of the venison (Table 12). The MFI percentage was significantly higher in venison from the pasture only group (P<0.01). The sex of the animal did not have a significant effect on the MFI percentage (P=0.578).

Table 12: Myofibrillar fragmentation index (MFI) and sarcomere length of venison from R1 stags (Males) and R2 hinds (Females) fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets for six weeks prior to slaughter.

Meat Quality Traits	Pasture		PKE		P-value		
	Female	Male	Female	Male	Group	Sex	Interaction
MFI (%)	97.1	97.5	94.3	95.0	0.004	0.578	0.868
Sarcomere Length (µm)	1.65	1.64	1.68	1.61	0.739	0.164	0.247

#### 4.9 Confinement Odour Analysis

The feeding treatment (P=0.809) and the sex of the animal (P=0.506) had no significant effect on the confinement odour score from the venison (Table 13). The pH was also not affected by the sex (P=0.250) or the treatment group (P=0.217). The venison from the R2 females had a significantly higher persistent odour score than the R1 males (P<0.01). The feeding treatment had no significant effect on the persistent odour (P=0.411).

Table 13: Confinement odour analysis of venison from R1 stags (Males) and R2 hinds (Females) fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets for six weeks prior to slaughter.

Confinement Odour score	Pasture		PK	E	P-value		
	Female	Male	Female	Male	Group	Sex	Interaction
Confinement Odour	1.2	1.2	1.1	1.2	0.809	0.506	0.851
Persistent Odour	1.6	1.4	1.1	1.1	0.411	0.001	0.389
рН	5.6	5.6	5.6	5.6	0.217	0.250	0.709

#### 4.10 Microbial Analysis

The number of aerobic bacteria (APC) found on the venison from the R1 males were significantly higher than the R2 females (P<0.01). The lactic acid bacteria count (LAB) was also significantly higher in the venison from the R1 males (P<0.05). The treatment groups did not have a significant effect on the APC (P=0.175) and LAB counts (P=0.065) (Table 14).

Table 14: Microbial analysis of venison from R1 stags (Males) and R2 hinds (Females) fed either pasture-only (Pasture) and 50% pasture: 50% PKE (PKE) diets for six weeks prior to slaughter.

Microbial Tests (log <sub>10</sub> cfu/cm <sup>2</sup> )	Past	ure	PK	E		P-value		
	Female	Male	Female	Male	Group	Sex	Interaction	
Aerobic bacteria count (APC)	1.32	1.64	2.73	4.76	0.175	0.008	0.391	
Lactic Acid bacteria count (LAB)	2.35	1.93	2.8	4.61	0.065	0.023	0.120	

# 4.11 Rapid Evaporative Ionisation Mass Spectrometry

REIMS detected 5469 ions in positive mode and 3079 ions in negative mode. Note, one ion does not equal an individual compound, and one compound can produce several different ions. In both modes using Principal Components Analysis (PCA) the difference between the feeding treatments and sexes were distinguished based on the metabolites measured by REIMS in venison (Fig 2). The variance between the samples explained by feed was about 6.5%. The metabolite differences were also distinguished between the cooked and uncooked venison.

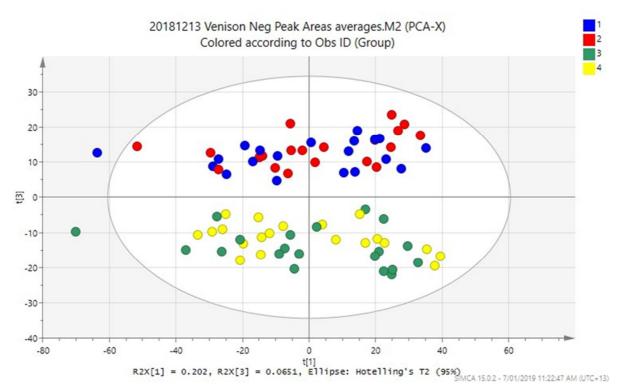


Figure 2: Principal Components Analysis plot of negative ionisation REIMS response coloured according to meat group. Groups 1 and 2 (blue and red dots) were fed pasture, while groups 3 and 4 (green and yellow dots) were fed PKE.

# 5. Discussion

## 5.1 Animal Growth and Handling

Live weight gain increased with PKE-supplementation for both R1 stags and R2 Hinds, while the body condition scores remained consistent. Although diets were designed to be equivalent on a metabolizable energy basis, PKE being a concentrate feed with a higher nutrient density, may have allowed greater energy intake at the same level of gut-fill. This trial was not designed to compare live weight gain between pasture and PKE supplementation. Such a trial would have a much greater number of assimilated groups of deer over a longer period of feeding (e.g. three months). With PKE supplement being fed twice per day, the level of human contact was much higher than the pasture-fed group. It was expected that this increased contact would result in quieter animals with lower handling scores. Whilst there was some evidence for this, both groups were very easy to handle. It is interesting to note that the increased growth rates in the PKE-supplemented group did not result in heavier carcasses.

#### 5.2 Carcass Characteristics

The carcass characteristics were significantly higher for R2 females than the R1 males which shows that the sex and age of the deer influenced the carcass characteristics in this trial. However, this should not concern the processors and industry leaders as the age of the deer at slaughter were <2 years old and at this age the venison produced is generally consider as premium quality in the market. For this trial, the age of this class of females was chosen for the basis of this hypothesis that the effects of PKE feeding on meat quality is more notable in R2 females than R1 males as this class had a better chance of laying down fat than the R1 males.

The dressing percentage of deer from the pasture only group was approximately 1% higher than the PKE group and this is influenced by live weight and HCW of the animals. The live weight can be influenced by a variety of factors including the time of feeding, dry matter of feed and the rate of passage of feed. The final live weight of the animals varied slightly, and it is possible the gut fill weight of the deer from the PKE group was higher than the pasture only group. This is possible as PKE is like most concentrates in which it is denser than the pasture and therefore causes more weight of feed in the gut and the live weight to be greater. Once the weight of the gut is removed from the animal, the carcass weight becomes less, making the dressing percentage greater. Deer fed PKE supplemented diet are more likely to have a higher gut fill weight and therefore it is recommended that farmers farm deer to have slightly heavier live-weights than those of which are on pasture only treatments to ensure that the targeted carcass weight requirements are meet.

The leg/HCW, left and right cap off/on and cap off left/right percentages were not significantly affected by the gender or treatment group of the deer. This was not unexpected as the deer produce relatively uniform carcass shapes when fed and grown well. From this 6-week trial, some

differences of interest were found and should be investigated further. In this trial the pasture was supplemented with a high rate of PKE (50%) and this level of supplementation is unlikely to be used by NZ deer farmers unless during extreme climatic events (e.g. droughts). Currently in NZ dairy cattle farmers are recommended to feed no more than 6kg/hd/day of PKE which is about a rate of <30% PKE. Therefore it is recommended that additional research into the effects of using lower rates (<30%) of PKE on deer are investigated.

#### 5.3 Consumer Sensory Analysis

The consumer sensory analysis found there were no significant differences between the feed treatments or the sex of the deer for the tenderness, flavour and overall liking of the venison samples. These findings were consistent with An et al. (2017) who reported that consumers found that addition of palm kernel meal (PKM) had no significant effect on the flavour, tenderness or the overall acceptability of pork (NOTE: the pigs were fed the PKM supplementary diet from weaning until slaughter (12-week period)). Filho et al. (2016) also reported that the flavour, tenderness, global acceptance and preference of bull meat did not change significantly with the addition of PKC.

The tenderness scores were consistent with the shear force measurements as the consumers scored as high as 71.1 for tenderness which indicates that the venison was very tender, and the shear force results were >4 kgF which is very tender for venison. The tenderometer found a significant difference between the sex of the animals, however with a difference of 1.0 kgF consumers were unable to detect this difference. REIMS found that there were differences in the meat compositions between the two treatments, which may have an impact on the eating quality of venison. However, the consumer overall liking and quality rankings were not significantly different between the treatment groups which indicate that the difference in meat compositions were not detectable by consumers.

The consumers did rate the juiciness of the venison from the pasture only group to be higher (P<0.1) than the PKE treatment group. This may possibly be a concern for processor and industry leaders as this may affect the consumer acceptability and the overall commercial value of the farmed venison. However, it is important to note that the quality ratings were not significantly different between the treatment groups or the genders, which indicate that level of satisfaction of the venison were similar between the groups and genders. It was possible that the juiciness of the venison was affected by the degree of cooking and resting time provided during preparation of the loins for the consumer taste trial. The loins were cooked to 71°C which is considered as well done for degree of doneness and this may have affected the consumers perception of juiciness as venison is generally recommended to be prepared rare or medium rare. The consumers for this trial were untrained and they had stated their consumption of venison was low, which may have affected their judgement on degree of juiciness of the venison samples.

There were no significant differences among the different types of venison on the quality ratings assigned by respondents. Most consumers (around 85%) indicated that venison samples were at least a 'good everyday quality' showing high level of satisfaction. However, few consumers (around 14%) were unsatisfied with the eating quality of the samples. This response could be partly associated to the low frequency of venison consumption of consumers being unfamiliar with this type of meat. Pasture supplementation with PKE or animal gender didn't seem to influence the eating quality of venison from the production conditions evaluated in this study. Overall, venison resulted in high levels of consumer satisfaction, with 43% of consumers rating samples as 'good everyday', 28% as 'better than everyday' and 14% as 'premium quality' regardless of the type of venison.

## 5.4 Intramuscular fatty acid content and composition

Venison from deer supplemented with PKE had a significantly higher IMF% than the pasture only group. This increased in IMF% was inconsistent with Bureš et al. (2015) findings that the IMF content was significantly higher in deer fed concentrate diets compared to grass-fed deer. However, the difference in IMF content between the two treatments was only 0.34% which is relatively small and is unlikely to have an impact on the consumer acceptability. As expected, the venison from the R2 female deer was significantly higher in IMF% than the R1 males which indicates the gender and/or age of the animal has an impact of the IMF content in venison. Comparing IMF% from genders at the same age or maturity is required to understand whether age or gender is the main driver for differences in IMF.

Venison from deer supplemented with PKE had significantly lower amounts of omega 3 fatty acids (n3) than the pasture only group. The omega 3 difference was 0.5 mg which is relatively significant from a health perspective as n3 are healthy for people and consumers are encouraged to have higher levels of n3 in their diets. This finding was not consistent with the findings of Ribeiro et al. (2018) which reported that the total n3 and n6 found in goat meat did not change significantly with the addition of palm kernel cake (PKC). Additional research will need to be completed to find out how the addition of PKE may have caused this fatty acid change and how the difference may change the sensory attributes of venison. PKE supplementation had no significant effect on the omega 6 fatty acids present in the venison, however the n3/n6 ratio was significantly higher in venison from the pasture only group. This result was not consistent with the findings of Ribeiro et al. (2018) which found that the n3/n6 ratio in goats did not change significantly with the addition of PKC, however due to the higher levels of n3 present in the venison from the pasture only group, the n3/n6 was expected in be higher in the pasture only group.

There were other significant differences in fatty acids that were observed between the treatment groups which were interesting discoveries and therefore could be investigated further.

## 5.5 Colour and Colour stability

## 5.5.1 Vacuum-packaged chilled storage colour

Two-weeks post-slaughter the venison from deer supplemented with PKE was statistically significantly lighter in colour than the pasture only group, however the actual difference was 0.7 which visually would not be noticeable by consumers. This is the same for the redness being lower in the venison from the PKE treatment because the actual difference was only 0.3. The venison was significantly lighter and yellower in the R1 males, while the redness was greater in the venison from the R2 females. However, the differences were small, and consumers would not visually be able to detect the colour differences.

After 8-weeks of vacuum-packed chilled storage, the PKE supplementation had a statistically significant effect on colour stability of venison. By day 54, the total colour difference was approximately  $\Delta E$ =2.0 and for each treatment the venison colour had increased in darkness (- $\Delta L^*$ ), redness (+ $\Delta a^*$ ) and decreased in yellowness (- $\Delta b^*$ ). These colour differences were expected as venison is a red meat and over time becomes purple or brown more rapidly than other meats. In this trial the colour difference over 8-weeks was quite small and this difference would not have any visual impact on the consumer acceptability of NZ farmed venison.

## 5.5.2 Retail display colour stability

The colour of the venison was not significantly different between the treatment groups once the loins were trimmed and the surface was exposed. There was a significant sex effect on the L\* and b\* coordinates for the venison. The colour of the venison from the R1 males was significantly lighter and yellower than the R2 females which corroborates the findings of Craigie (2012), however the actual colour difference would not be noticeable by consumers.

After 14-days in retail display conditions, the PKE supplementation had significant impact on the colour stability of the venison. The total colour difference ( $\Delta E$ ) and the increase in redness was significantly higher in the PKE group. However, the difference in  $\Delta E$  and  $\Delta a^*$  were quite small and are unlikely to be detected by the consumers.

## 5.5.3 Vacuum pack and bloom colour scores

The vacuum pack colour was significantly higher for venison from R2 females compared to R1 males. This is not likely to have impact on consumer acceptability as consumers will not see the venison in the vacuum-packed conditions and therefore is not a concern for processors and industry leaders. The bloom colour of the venison was not significantly different between the treatment groups or genders. Discolouration is likely to occur more rapidly in red meat than other meats and these could be caused by several factors such as the browning effect, pre-slaughter

stress syndrome, lipid oxidation, chemical and autolytic enzymatic reactions, bacterial spoilage or the breakdown of fat, protein and carbohydrates in the meat (Schuster et al. 2018).

## 5.6 Drip loss and pH

The pH measured 24 h post-mortem was not significantly different between the pasture only and PKE groups or the sex of the animals. The mean 24 hr pH from the R1 on the pasture only and PKE diets were 5.26 and 5.28, respectively, these pH values were very similar to the mean pH measured from R1 in the Deer Progeny Tests (DPT) where the mean 24 hr pH was 5.46. This indicates that the rate of pH decline was not influenced by the diet of which the animal is fed. This result agreed with Ribeiro et al. (2018) which reported that the addition of PKC did not change the ultimate pH of the goat meat. According to Geay et al. (2001), the decline in pH rate is greatly affected by the glycogen stores present in the animal at slaughter, cooling rate of meat and other pre- and post-slaughter procedures and therefore the pH of the meat at 24 h post mortem was the same between the treatment groups and genders.

The 24-hr drip loss and drip loss percentage were significantly higher in the venison from the PKE group. However, the actual drip loss differences in this trial were only 0.5% which is quite small and the mean drip losses for this trial were lower than what was found from the R1 deer in the DPT (3.4%). This difference in drip loss for venison could be a concern for the processors and industry leaders as the drip loss affects the final meat weight and the sensory eating qualities which in the end could contribute to commercial value decreasing. Therefore, additional research into the causes of this meat quality attribute may be required to determine if this increase in drip loss is a result of diet-related causes.

# 5.7 Tenderness

According to Kim et al. (2017), the shear force of cooked meat is related to the connective tissues present in the meat and these can contribute to the meat flavour and other sensory attributes. The shear force of the venison was not significantly different between the treatment groups and is comparable to the findings reported by Filho et al. (2015) and Santos et al. (2017). These studies reported that the shear force of meat from the Nellore bulls and lamb did not change significantly with the addition of PKC. According to Filho et al. (2015) a possible reason for there being no changes in shear force with the addition of a palm oil by-product supplement would be because the animals were the same age and size regardless of treatment group. This could also be one of the main reasons why the R1 males had significantly higher shear forces than R2 females as the animals were different ages.

The R1 males had a mean shear force of approximately 1KgF greater than the R2 females which is congruent with the findings of Craigie (2012), however these shear force values were still low, and the venison was still very tender. In comparison with the DPT the mean shear force for this trial was approximately 2kgF lower than DPT R1 mean shear force (6.58 kgF). It is important to note that if the shear force for cooked venison is <8kgF then it is considered tender for consumer consumption (Bickerstaffe et al. 2001). These shear forces should not be a concern for consumers and industry leaders because most consumers cannot detect differences <2 kgF. In the case of the DPT the consumer panel were able to detect 1.5 kgF differences. The cause of this slight difference could possibly be due to there being more connective tissue present in the R1 males or that the connective tissues present are significantly tougher than the connective tissues found in the R2 females (Purchas et al. 2010). However, further work will be required to confirm if the connective tissues were significantly different between the genders of the animals.

# 5.8 Myofibrillar fragmentation Index and Sarcomere length

The myofibrillar fragmentation index (MFI) was significantly higher in the venison from the pasture only group and there was no significant difference between the sex of the deer. MFI is the measure of myofibrillar proteins present in the meat which have not disintegrated, and this is associated with ageing process of meat. The venison from the PKE group had a significantly lower MFI than the pasture group, which indicates that the ageing process of the PKE venison had not progressed as much as the pasture-fed venison.

According to Olson et al. (1976), the MFI has a strong positive correlation with the tenderness of the meat and MFI approximately represents 50% of the variation in shear force values and sensory tenderness scores. This means as the MFI increases, the tenderness will increase too. The MFI was lower in the PKE group, however from the consumer sensory analysis in this study, the consumers rated that there was no significant difference in the degree of the tenderness of the venison between the treatment groups. This result suggests that lower MFI value did not influence the consumer acceptability of venison in terms of tenderness.

According to Purchas et al. (2010), the sarcomere length of stags (18-20 months) were significantly shorter than the hinds but these results were not consistent with the findings found in this study. There was no significant difference in sarcomere lengths between the treatment groups or the sex of deer. This finding was good and could explain why the tenderness of the treatment groups and gender were scored the same by the consumers in the sensory analysis. This is because meat tenderness is also associated with sarcomere lengths and according to Hutchison et al. (2014), meat is typically more tender when the sarcomeres are stretched, and contraction is prevented.

# 5.9 Confinement odour analysis

The confinement odour scores between the treatment groups and genders were not significantly different, which indicate that differences in intramuscular fat (IMF) content had no significant impact

on the odour produced during storage by the venison. With a mean score of approximately 1 been given to the venison samples this indicates that confinement odour was not detected, and that meat spoilage had not occurred in the vacuum pack storage. Overall the feeding treatments did not have an impact on persistent odour, however this attribute was scored significantly higher for the venison from the R2 females. However, the difference between the genders were quite small (0.1) and both classes of venison were scored on average less than 2 which indicate that no persistent odour was detected, and that little spoilage-related odour had occurred during storage. From the results of the confinement odour analysis, it indicates that PKE had little impact on the confinement odour produced from the venison during vacuum pack storage and would not have an impact on the consumer acceptability of NZ farmed venison.

## 5.10 Microbial Analysis

This study found that the addition of PKE as a supplement in the deer diet had no significant effect on the number of aerobic bacteria (APC) or lactic acid bacteria (LAB) present on the venison following 10-weeks of storage. However, the APC and LAB counts were significantly higher in the venison from the stags. This could be an area of concern for venison processors as the growth of LAB is one of the main spoilage factors that can cause off-flavours, off-odour and discolouration of the meat during storage (Schuster et al. 2018) and therefore reduce the shelf-life of the product. The growth of LAB has been known to cause a sour off flavour and odour which is from lactic acid produced by the bacteria.

The difference in microbial levels between the R1 males and R2 females could be possibly a result of the maturity or behavioural patterns of the animals. The main source of bacterial contamination would most likely be from the skinning of the animal. R1 males may have had a significantly higher level of bacteria because they may have been dirtier or were in a different coat phase (shedding) than the R2 females. This gives more opportunity for bacterial contamination to occur for R1 males during the slaughter and processing stages.

# 5.11 Rapid Evaporative Ionisation Mass Spectrometry

Both positive and negative modes were used in this measurement to broaden the range of compounds detected in the venison. This is because ionisation is a prerequisite for detection by mass spectrometry. Therefore, some compounds ionise better with a positive charge while others ionise better with a negative charge.

Based on the metabolites measured by REIMS in the venison, differences between the pasture only and the PKE group were distinguishable and these results suggests that what the deer have been fed is clearly affecting their meat composition. As the difference was much clearer for negative ionisation mode than positive ionisation mode (Fig 2), this strongly indicates that the differences

are likely to be strongest for lipophilic compounds. Further work will be done to see if these differences in meat composition are related to consumer preference scores.

# 6. Recommendations

- A high level of dietary supplementation with PKE of R1 male and R2 female deer on a
  pasture diet did not change any consumer perceptions of quality over that of pasture fed
  venison except for juiciness, which was perceived to be lower (P<0.1) for PKE. There were
  significant differences detected in IMF and omega 3 where these differences were positive
  in favour of pasture, the New Zealand deer industry may wish to investigate alternative feed
  sources that enhance these quality characteristics.</li>
- If consumers are exposed to farmed New Zealand venison from R1 males or R2 females that have been supplemented with PKE it is unlikely they will detect an unfavourable difference and other factors will likely have a much greater influence on consumer appeal.
- The drip loss of venison was significantly higher from the PKE group and additional work will be required to determine if this was caused by diet. This may have implications for venison processors, but the absolute differences were low.
- The shear forces of cooked venison were significantly higher in the stag group, the actual differences were small, and it is not clear whether this was due to the amount of connective tissues present in the venison or if the connective tissues present were tougher.
- Pasture supplementation with PKE or animal gender did not influence the sensory attribute of venison from the production conditions evaluated in this study.
- REIMS detected that the feeding treatment of deer affects the composition of the venison. The technology shows promise as a tool for real-time sample characterisation or testing. Future research could investigate if the differences in metabolites are related to the results from the consumer sensory analysis.

# 7. Acknowledgements

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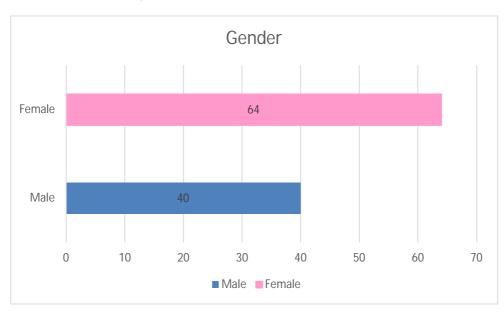
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# 9. Appendices

# 9.1 Consumer Sensory Analysis Demographic Questionnaire Results

Results from the Demographic Questionnaire which the consumers completed before the consumer venison tasting.



Results of the Demographic Questionnaire

Figure 3: Number of male and female consumers

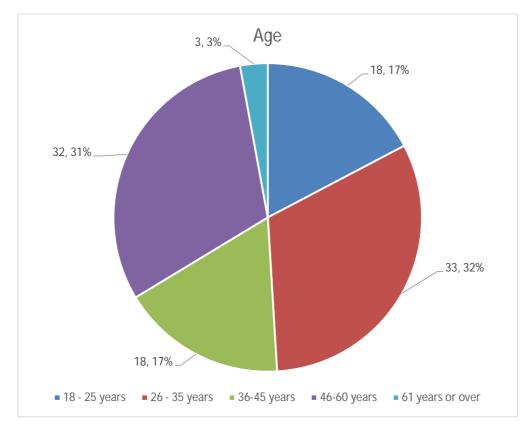


Figure 4: Percentage of different age groups of consumers

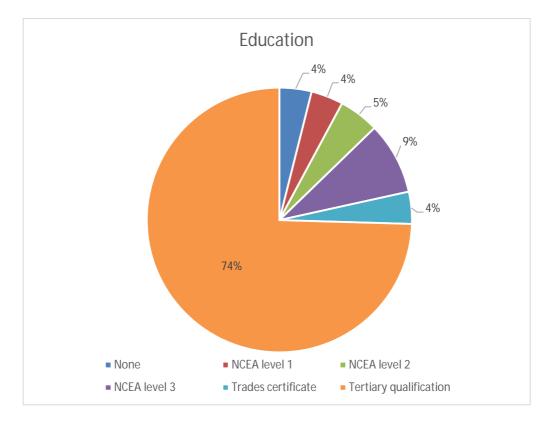


Figure 5: Percentage of Educational levels of the consumers

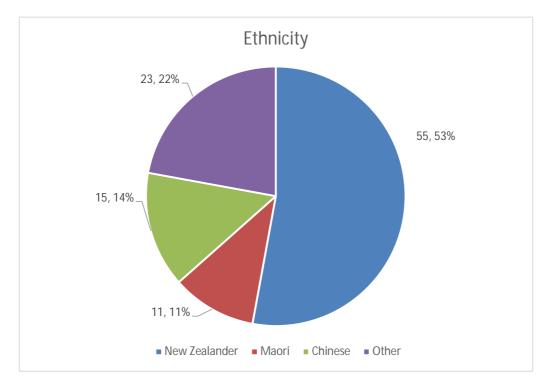


Figure 6: Percentage of different Ethnic groups in the consumers

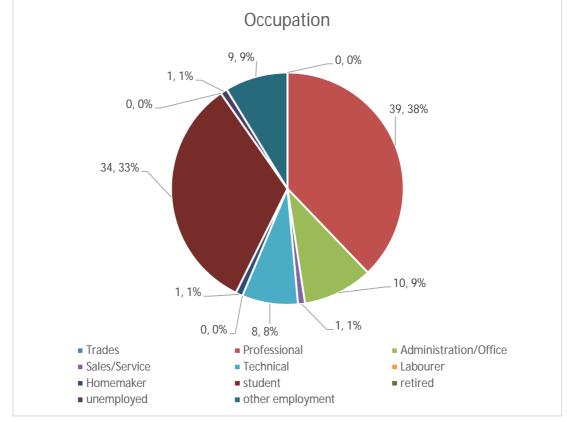


Figure 7: Percentage of consumers in different occupations

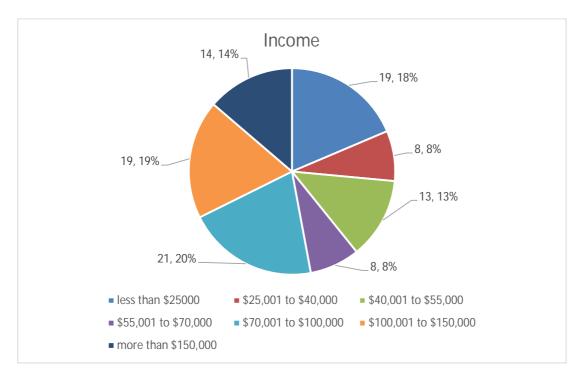


Figure 8: Percentage of consumers in different income levels

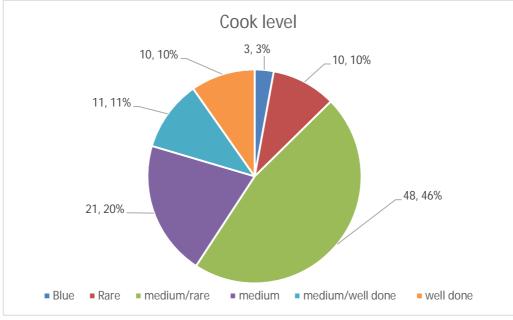


Figure 9: Percentage of desired level of doneness of meat among the consumers

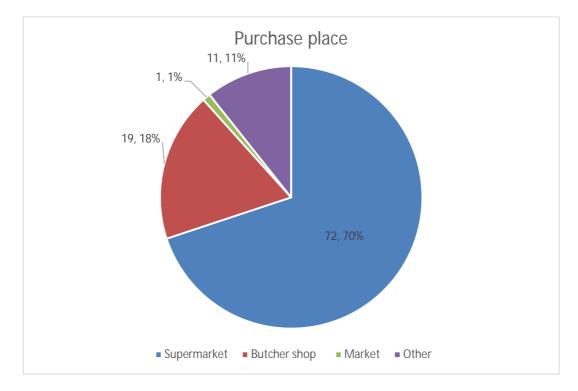


Figure 10: Percentage of preferred purchaser place for meat among the consumers

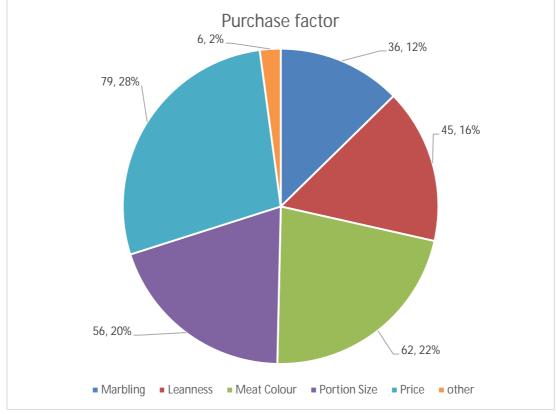


Figure 11: Percentage of different quality factors for the purchase of red meat

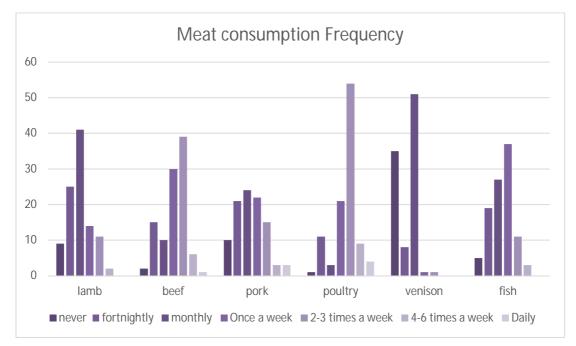
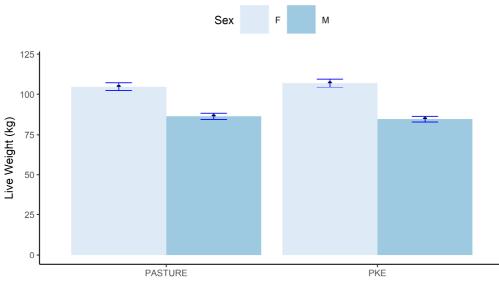


Figure 12: Percentage of consumption frequency of different types of meat by consumers

## 9.2 Graphs of findings

## 9.2.1 Carcass Characteristics



Predicted means for "Group:Sex" with Stder Bars

Group

Figure 13: Plot of the pre-trial live-weight (20/08/2018) of the deer from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

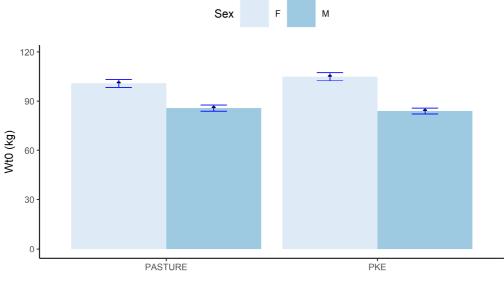
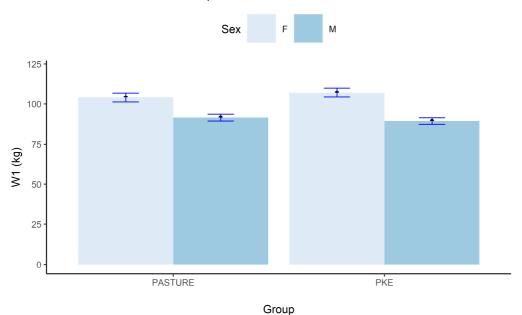




Figure 14: Plot of live-weights (W0) at the start of the six-week feeding trial (29/08/2018) of the deer from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 15: Plot of live-weights (W1) at, day 13 of trial (11/09/2018) of the deer from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

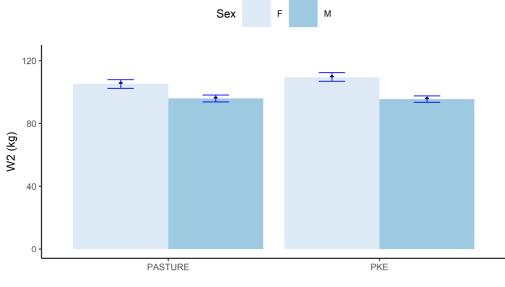
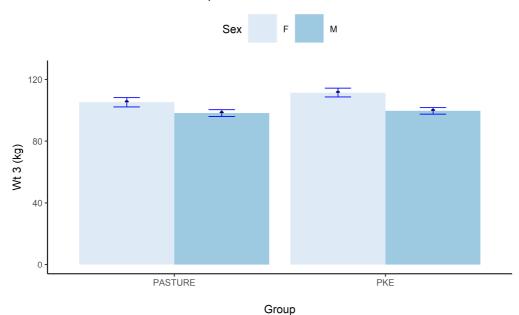




Figure 16: Plot of live-weights (W2) at, day 29 of trial (27/09/2018) of the deer from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 17: Plot of live-weights (W3) at, day 42 of trial (10/10/2018) of the deer from the two different treatment groups (pasture only and 50%PKE:50%pasture)

# Predicted means for "Group:Sex" with Stder Bars

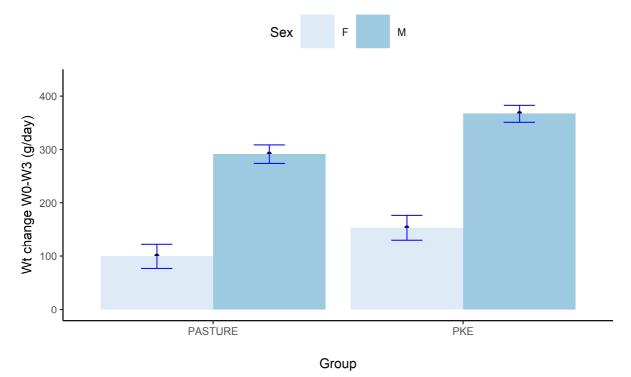
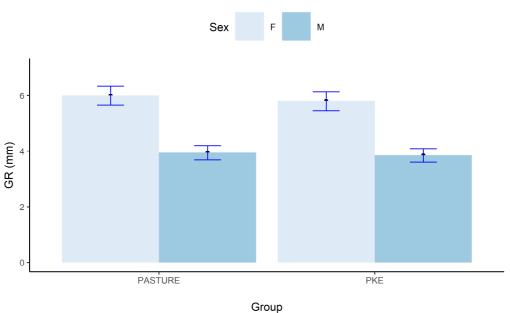


Figure 18: Plot of daily live-weight gain from 29/08/2018 to 10/10/2018 (42 days) of the deer from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 19: Plot of fat depth over the 12th rib (GR) of the deer from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

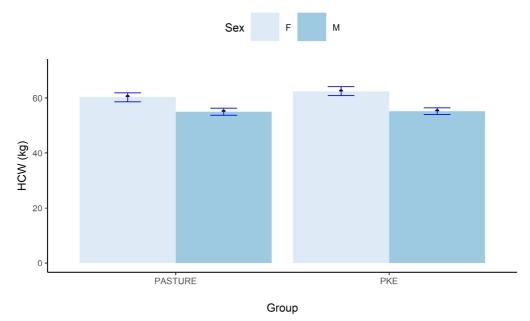
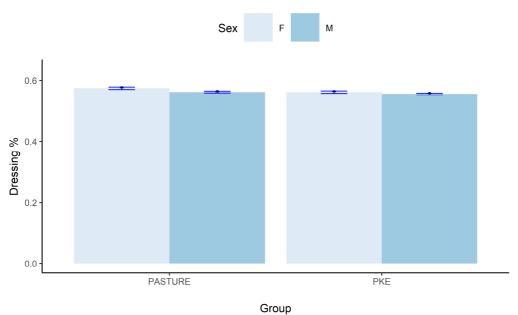


Figure 20: Plot of hot carcass weight of the deer from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 21: Plot of dressing percentage of the deer from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

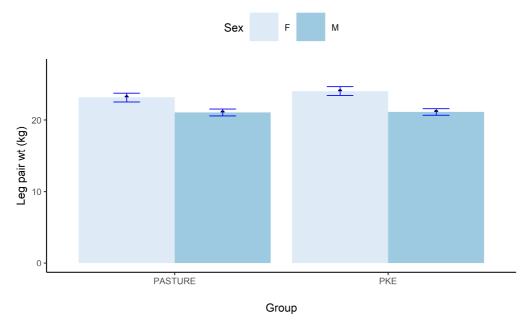
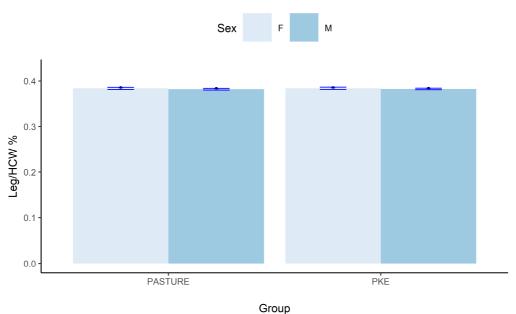
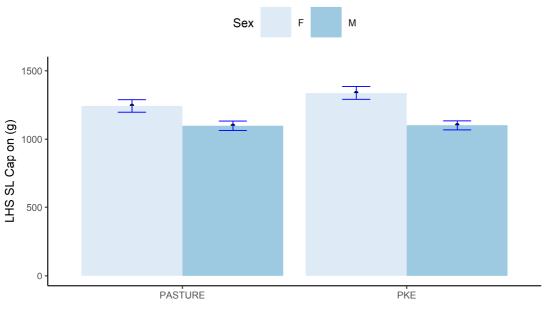


Figure 22: Plot of leg pair weight of the deer from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

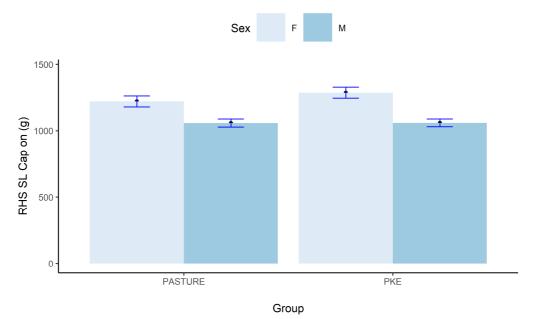
Figure 23: Plot of leg pair weight/HCW percentage of the deer from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Group

Figure 24: Plot of LHS SL cap on from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 25: Plot of RHS SL cap on from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

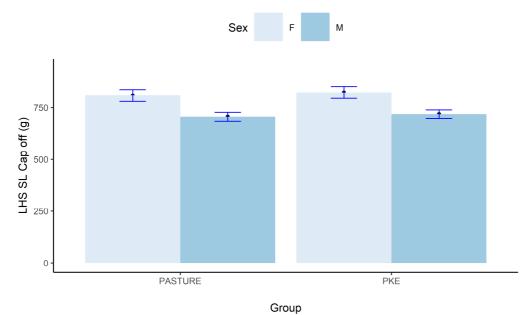
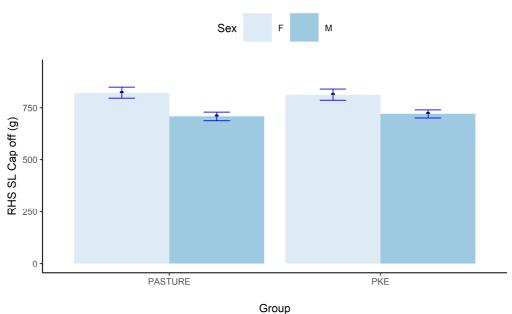


Figure 26: Plot of LHS SL cap off from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 27: Plot of RHS SL cap off from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

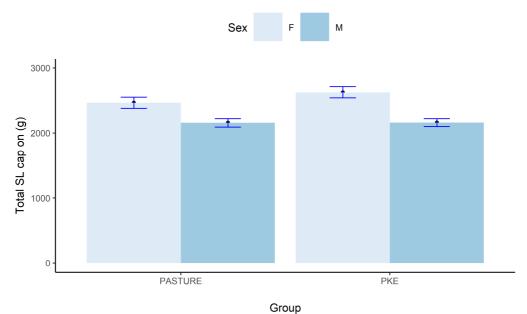
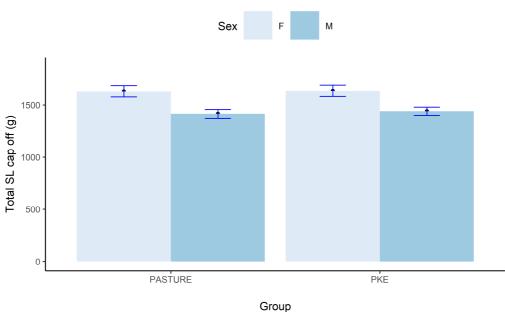


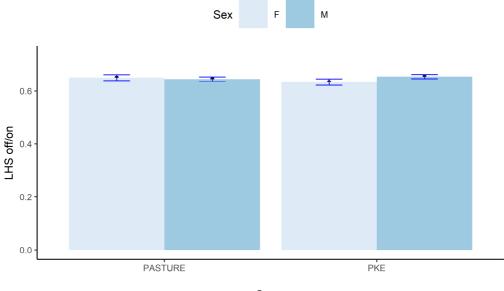
Figure 28: Plot of total SL cap on from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

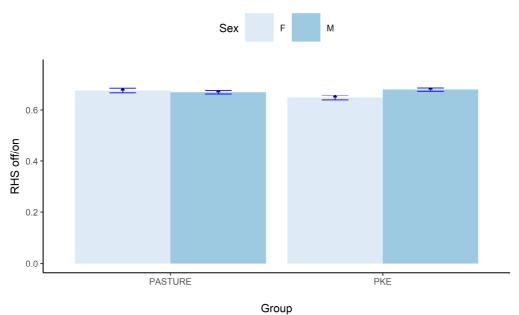
Figure 29: Plot of total SL cap off from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars



Group

Figure 30: Plot of LHS SL cap off/ cap on percentage from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 31: Plot of RHS SL cap off/ cap on percentage from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

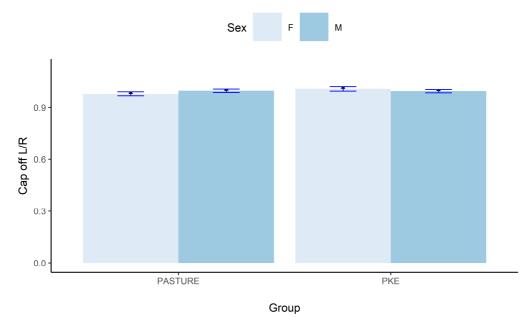
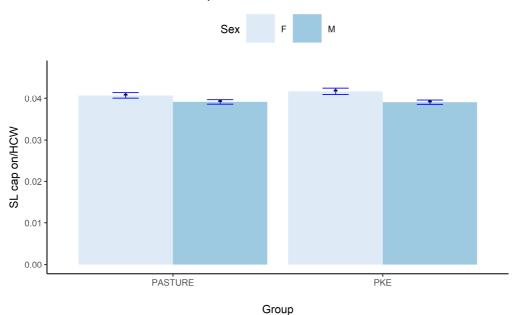


Figure 32: Plot of cap off LHS/RHS percentage from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 33: Plot of SL cap on/HCW percentage from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

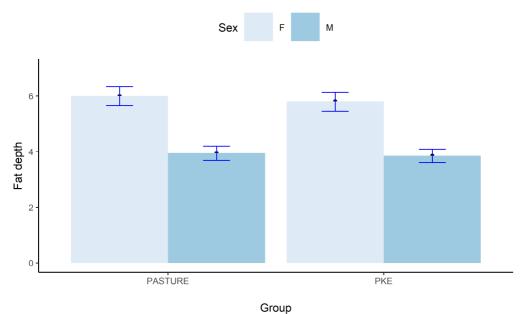
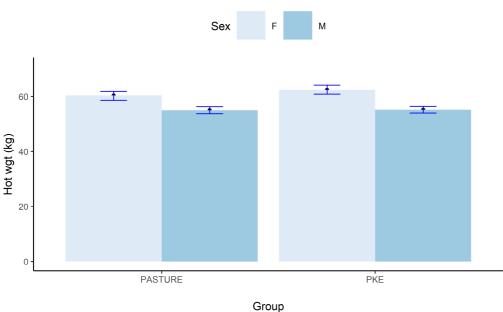


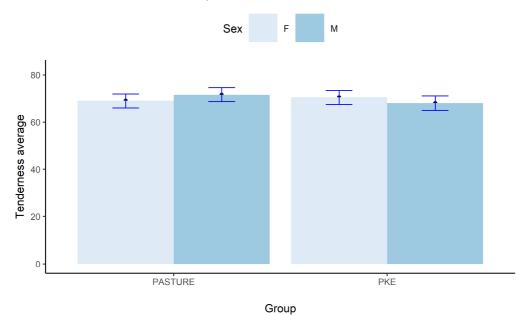
Figure 34: Plot of fat depth from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

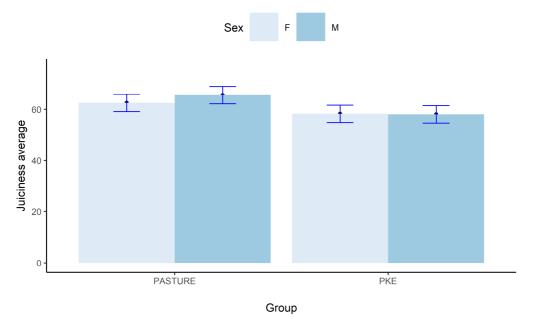
Figure 35: Plot of hot carcass weight from the two different treatment groups (pasture only and 50%PKE:50%pasture)

### 9.2.2 Consumer sensory analysis



Predicted means for "Group:Sex" with Stder Bars

Figure 36: Plot of the tenderness scores for the venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 37: Plot of the juiciness scores for the venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

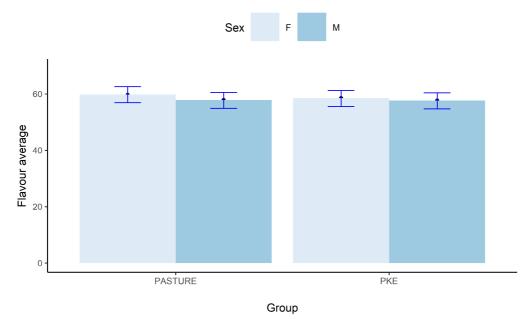
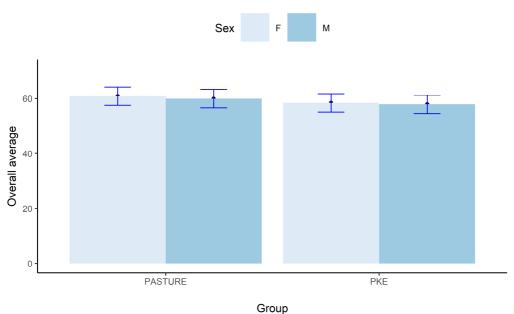


Figure 38: Plot of the flavour scores for the venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 39: Plot of the overall liking scores for the venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)

## 9.2.3 Venison Intramuscular fatty acid content and composition

#### 9.2.3.1 Fatty acid composition

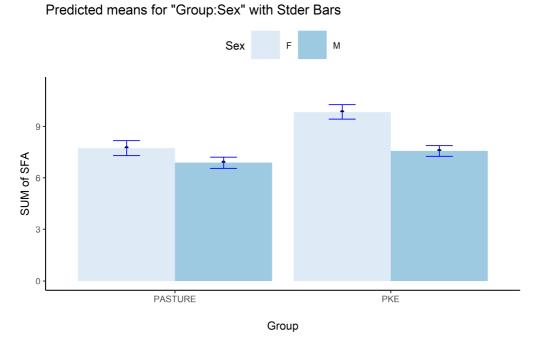
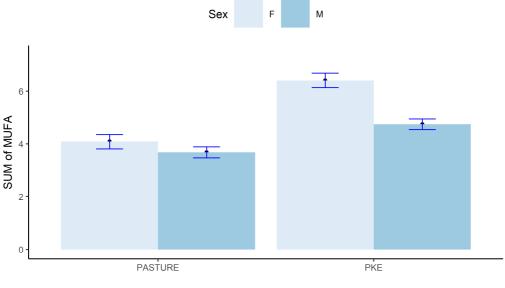


Figure 40: Plot of the total saturated fatty acids (SFA) presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)



#### Predicted means for "Group:Sex" with Stder Bars

Group

Figure 41: Plot of the total mono-unsaturated fatty acids (MUFA) presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

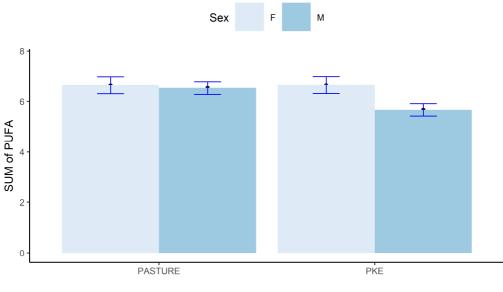
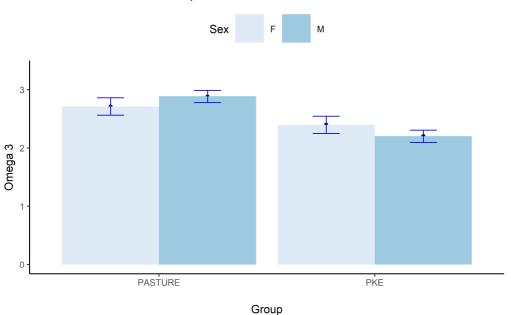




Figure 42: Plot of the total poly-unsaturated fatty acids (PUFA) presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 43: Plot of the omega 3 fatty acids presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

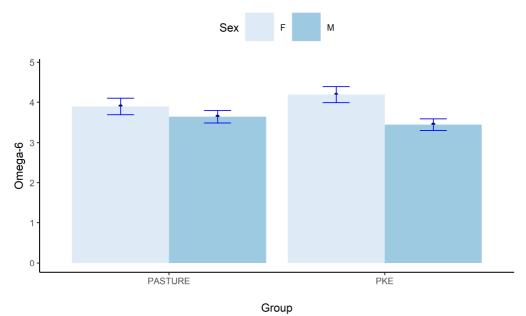
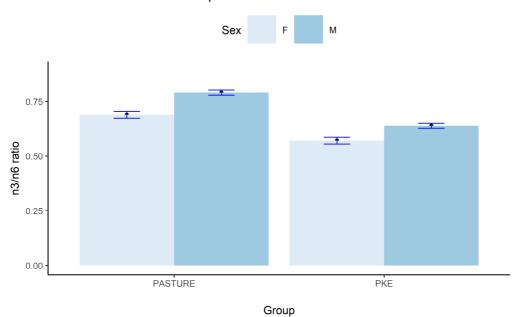


Figure 44: Plot of the omega 6 fatty acids presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 45: Plot of the omega 3/omega 6 ratio presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

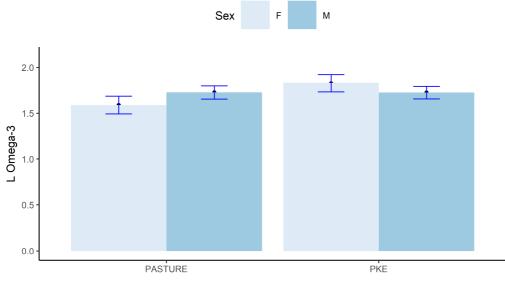
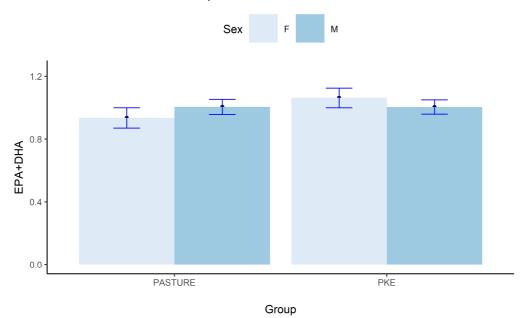




Figure 46: Plot of the long-chained omega 3 fatty acids presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 47: Plot of the EPA + DHA fatty acids presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)

### Predicted means for "Group:Sex" with Stder Bars

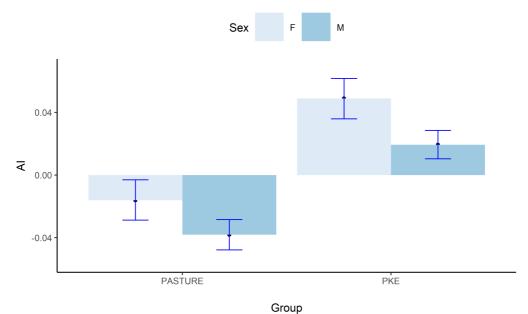
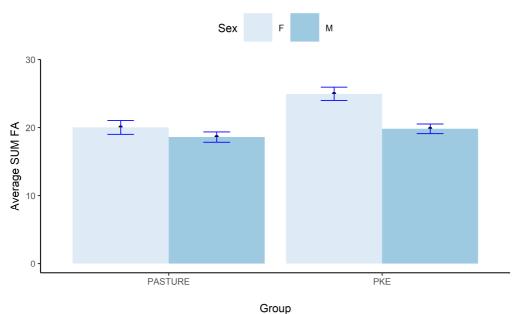


Figure 48: Plot of the AI of the venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 49: Plot of the average total fatty acids presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars

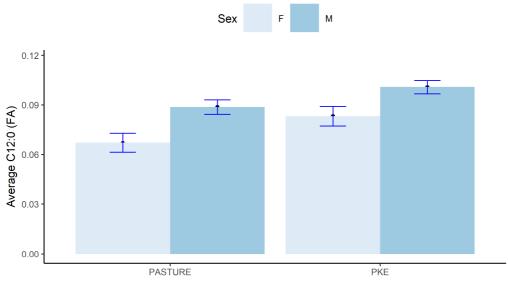
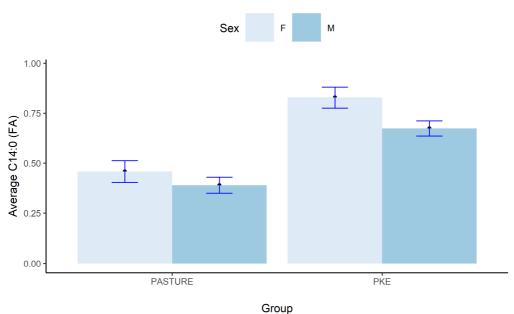




Figure 50: Plot of the average C12:0 fatty acids presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 51: Plot of the average C14:0 fatty acids presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)

### Predicted means for "Group:Sex" with Stder Bars

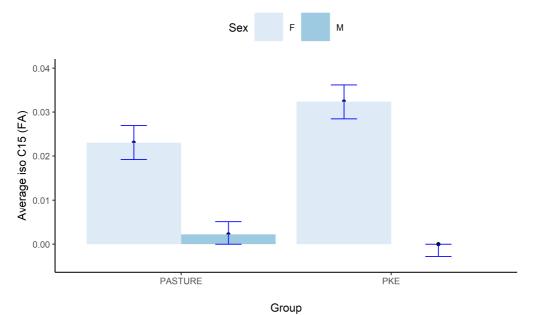
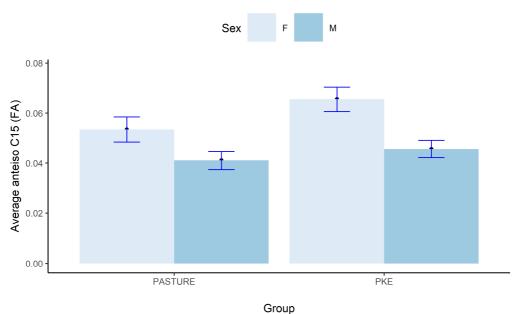


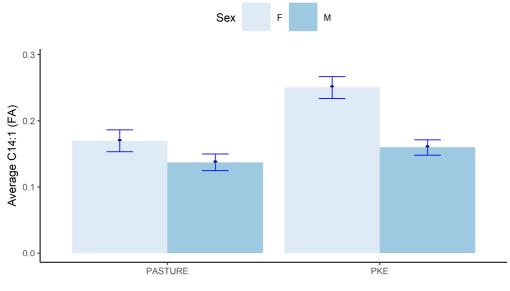
Figure 52: Plot of the average iso C15:0 fatty acids presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

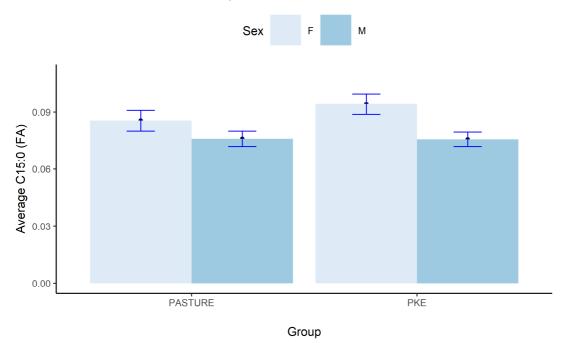
Figure 53: Plot of the average anteiso C15:0 fatty acids presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)

Predicted means for "Group:Sex" with Stder Bars



Group

Figure 54: Plot of the average C14:1 fatty acids presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)



Predicted means for "Group:Sex" with Stder Bars

Figure 55: Plot of the average C15:0 fatty acids presented in venison from the two different treatment groups (pasture only and 50%PKE:50%pasture)