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## Factors affecting feeding and activity behaviours in healthy pre-weaned artificially reared calves

Riley, BR; Duthie, C-A; Corbishley, Alex; Bowen, J M; Mason, CS; Bell, DJ; Haskell, MJ

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an individual's behaviour varies around its mean). Here we use methods from the behavioural ecology literature to quantify between and within individual variation in calf feeding behaviour.

*Materials and methods:* A computerised milk feeder recorded the feeding behaviour of 64 Holstein-Friesian heifer calves, housed in small groups, for 35 days at the Centre for Dairy Science and Innovation, University Of Nottingham, UK. 57 000 visits to the feeder were summarised into different feeding behaviours, calculated per calf per day. Calves were health scored twice weekly and clinically sick calves (n = 14) excluded from the analysis. We calculated repeatability (the proportion of total variation explained by between individual differences) by use of multi-level models and variance partitioning. We calculated the coefficient of variation in predictability (the degree to which individuals differ in their residual variation around their respective means) by use of double-hierarchical generalized linear models. Finally, we use a multivariate double-hierarchical model to test if calves that have more meals had higher feeding rates. All analysis was carried out in R (version 4.1.0).

*Results:* Repeatability estimates indicate that individual calves displayed substantial between individual differences in their average behaviour for feeding rate, meal frequency and feeding time (Table 1). Minimal between individual differences was observed for meal size. The coefficient of variation in predictability shows that calves with predictable and unpredictable feeding rates were present in our study, but for other behaviours calves were more homogenous in the degree to which their behaviour varied around their respective means. Outputs from the multivariate model indicate that behavioural types for feeding rate and meal frequency were positively correlated (R = 0.35 (CI: 0.06 – 0.60)) indicating that calves with higher feeding rates had more meals.

Conclusion: Implementing a behavioural ecology approach is a useful way to quantify differences in feeding behaviour.

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### 94. Factors affecting feeding and activity behaviours in healthy pre-weaned artificially reared calves

B. Riley<sup>a,b,\*</sup>, C.A. Duthie<sup>a</sup>, A. Corbishley<sup>b</sup>, J.M. Bowen<sup>a</sup>, C. Mason<sup>a</sup>, D.J. Bell<sup>a</sup>, M. Haskell<sup>a</sup>

<sup>a</sup> SRUC, Edinburgh, UK

<sup>b</sup> University of Edinburgh, Edinburgh, UK

\* Corresponding author. B. Riley.

**Application:** An understanding of factors affecting healthy calf behaviour will guide the development of behaviour-based tools for disease detection. **Introduction**: Behaviour is increasingly being researched as a tool for early disease detection in artificially reared calves. However, it is important to understand how other factors such as sex, and breed may affect healthy calf behaviour. This is to improve the sensitivity and specificity of behaviour-based tools, as alert accuracy is a major concern for animal keepers when discussing technology-based disease detection.

*Materials and methods:* One hundred and fourteen dairy bred calves from two herds (herd A = 43 calves, herd B = 71 calves) entered group pens of 12-14 calves at approximately seven days of age and left the study when weaning began at 40 days of age. Calves were bedded on straw with *ad-libitum* access to racks of straw and concentrate and a single automatic milk feeder with an automated weighing platform (*Biocontrol*). Seven litres of acidified milk replacer (mixed at a rate of 150g/L) were available daily. Activity behaviours were recorded using a leg-mounted 3-axis accelerometer (*Iceqube*). Health was scored daily using the Wisconsin calf health score method. To ensure only days where calves were healthy were included, Data was removed for days when medication was given, health scores were intermediate or high, and for 3 days either side. Calves with at least 10 consecutive healthy days were included and the middle 10 days of the healthy period were taken to achieve a balanced dataset. For parameters with a normal distribution a generalised linear mixed model was constructed using REML in R with animal number nested within group as random effects. Fixed effects tested were live weight, age, sex, herd, season of birth, age of inclusion into the group, dam parity, birth weight and sire breed type (beef or dairy). Model selection was carried out using the Akaike information criterion.

*Results:* Thirty-one calves were included in the analysis. Live weight, age, sex, herd, season of birth and age of inclusion into the group had significant effects on activity or milk feeding behaviours. Dam parity, birth weight and sire breed type had no significant effects on the behaviours studied. The results of the final models are presented in Table 1.

Table 1

Behaviour	Fixed effect	Level	Effect size	Confidence interval	P-value
Lying bouts /day	Season	Autumn	Reference	Reference	Reference
		Summer	6.8	2.6-10.8	0.01866
		Winter	-0.4	-3.4-2.7	0.78657
	Herd	В	Reference	Reference	Reference
		Α	4.0	1.1-6.5	0.00816
	Sex	Female	Reference	Reference	Reference
		Male	-3.2	-6.4-0.1	0.07107
Motion index/day	Age		-61.5	-96.2 - 25.8	0.000883
	Sex	Female	Reference	Reference	Reference
		Male	-653.98	-1360.5 - 3.3	0.065207
Motion index units/ standing bout	Age		-3.1	-4.9 - 1.0	0.00143
	Age at inclusion into group pen		-11.2	-18.88 - 2.2	0.00652
Visits to milk/day	Weight		0.08	0.02-0.14	0.00727
Volume milk/ visit	Weight		-5.4	-11.1 - 0.3	0.0454

*Conclusion:* When building behaviour-based disease detection tools, models should account for other calf factors including weight, age, and sex to improve accuracy. While there has been much research into changes in calf behaviour with social housing and greater milk allowances but there is very little literature on other factors that affect behaviour in pre-weaned calves.

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## 95. Examination of the bronchial lymph node and cranial lung lobe messenger RNA and micro RNA transcriptome response, following an experimental challenge to dairy calves with bovine herpes virus 1

S. O'Donoghue<sup>a,\*</sup>, D. Johnston<sup>a</sup>, B. Earley<sup>a</sup>, M.S. McCabe<sup>a</sup>, K. Lemon<sup>b</sup>, C. Duffy<sup>b</sup>, M. McMenamy<sup>b</sup>, S.L. Cosby<sup>b</sup>, J. Kim<sup>c</sup>, J.F. Taylor<sup>c</sup>, D.W. Morris<sup>d</sup>, S.M Waters<sup>a</sup>

<sup>a</sup> Animal and Bioscience Research Department, Animal & Grassland Research and Innovation Centre, Teagasc, Grange, Meath, Ireland

<sup>b</sup> Veterinary Science Division, Agri-food and Biosciences Institute, Stormont, Belfast, Northern Ireland, UK

<sup>c</sup> Division of Animal Sciences, University of Missouri, Columbia, Missouri, USA

<sup>d</sup> Discipline of Biochemistry, National University of Ireland, Galway, Ireland

\* Corresponding author. S. O'Donoghue.

*Application:* Differentially expressed microRNA's (miRNA's) and their target genes, may identify potential therapeutic targets for Bovine Respiratory Disease (BRD) infection.

Introduction: BRD is a primary cause of morbidity and mortality in cattle of all ages, both in Ireland and internationally. Bovine Herpes Virus 1 (BoHV-1), a double-stranded DNA virus of the alphaherpesvirus subfamily, is a primary BRD causative pathogen. There are limited data on the host transcriptome response (lung tissue) in dairy calves and on the microRNA (miRNA) transcriptome following BoHV-1 infection. Therefore, the study objective was to elucidate the changes in gene expression, and the miRNA transcriptome, following an experimental challenge with BoHV-1 in dairy calves. *Materials and methods:* Holstein-Friesian calves (149.2  $\pm$  23.82 days) were either challenged with BoHV-1 inoculum (10<sup>7</sup>/mL x 8.5mL per animal) (n = 12) or mock challenged with sterile phosphate buffered saline (PBS) (n = 6). Calves were euthanised on day 6 post-challenge. Lungs were scored for the presence of lesions and bronchial lymph node and cranial lung lobe tissues were collected. Tissue aliquots were flash frozen at -80°C for RNA-Seq and small RNA-Seq. Total RNA (including small RNA's) was extracted from tissue aliquots using the Qiagen RNeasy Plus Universal Mini Kit. RNA sequencing was performed using an Illumina Novaseq 6000 (150 bp paired-end) and small RNA-Seq samples were sequenced on an Illumina NextSeq 500 (75 bp single-end). RNA-Seq analysis, from quality assessment to differential expression, was undertaken and sequenced reads were aligned to the ARS-UCD1.2 bovine reference genome. Small RNA-Seq analysis was performed and data were aligned to the ARS-UCD1.2 reference genome. Messenger RNA (mRNA) targets of differentially expressed (DE) miRNA's were determined using Targetscan and context ++ scores were calculated using Targetscan and ViennaRNA. Target genes with a weighted context + + percentile rank of 99 were used for subsequent analysis. Pathway and functional enrichment analysis was performed on DE genes and DE miRNA's (P < 0.05, FDR < 0.05) using the ClusterProfiler package in R. Results: Multidimensional scaling (MDS) revealed a clear separation between the BoHV-1 challenge and control groups based on bronchial lymph node gene expression changes. A total of 337 genes were DE in the bronchial lymph node (P < 0.05, FDR < 0.1, fold-change > 2) between the two treatments. Additionally, there were 334 DE genes in the healthy cranial lobe tissue and 67 DE genes in the lesioned cranial lobe tissue. A total of 62, 55 and 14 miRNA's were DE (P < 0.05, FDR < 0.1, fold-change > 1.5) in the bronchial lymph node, healthy cranial lobe and lesioned cranial lobe tissues, respectively. The KEGG pathways enriched (P < 0.05, FDR < 0.05) across all tissues were 'Influenza A' and 'Herpes Simplex Infection'.

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Conclusion: The miRNA's involved in the bronchial lymph node and cranial lung lobe transcriptome response to BoHV-1 and their target genes, could

## 96. Intake, growth and carcass traits of steers offered grass silage supplemented with barley- and maize-based rations containing flaked peas, flaked beans, maize dried distillers grains or maize gluten feed

R. Kennedy<sup>a,b,\*</sup>, A.P. Moloney<sup>a</sup>, E.G. O'Riordan<sup>a</sup>, A.K. Kelly<sup>b</sup>, M. McGee<sup>a</sup>

potentially contain variants which influence susceptibility to BoHV-1.

<sup>a</sup> Teagasc, Grange, Meath, Ireland

<sup>b</sup> University College Dublin, Belfield, Dublin, Ireland

\* Corresponding author. R. Kennedy.

*Application:* Indigenous cereals and legume-protein sources can replace imported energy/protein feed ingredients as supplements to grass silage for beef cattle.

*Introduction:* In Northern Europe there is growing interest in exploiting indigenous cereals (barley) and legumes rather than imported cereals (maize) and by-products (e.g. maize-based) as animal feedstuffs. There is little published information comparing the relative feeding value of rolled barley and maize meal, and beans, peas, maize dried distillers grains (MDD) and corn gluten feed (CGF) as protein (+energy) sources in concentrate rations offered as a supplement to grass silage. The objective of this study, therefore, was to determine intake, growth and carcass traits of beef cattle offered grass silage supplemented with rolled barley or maize meal-based rations containing legumes or maize by-products.

*Materials and methods:* Late-maturing suckler-bred steers (n = 80; 575 kg, s.d. 21.3; 18-months, s.d. 1.0) were blocked by sire breed and weight, and within block were randomly assigned to one of eight supplement treatments, formulated to be isonitrogenous (135g crude protein (CP)/ kg dry matter (DM)): 1.Rolled barley (622g/ kg fresh weight basis) plus flaked peas (300 g/ kg), 2.Rolled barley (722g/ kg) plus flaked beans (200g/ kg), 3.Rolled