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Abstracts of Poster Presentations

Nutritional quality and the adequacy of donkey milk in the diet of allergic children

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Recent investigations have shown that donkey milk (DM) can be considered hypoallergenic in cases of cow milk protein allergies (CMA) and that it has a high acceptance rate by the children. Since DM could represent a natural alternative for allergic babies, the aim of this study was to evaluate the nutritional quality and adequacy of DM in the feeding of children with IgE-mediate cow's milk protein allergy and Food Protein Induced Enterocolitis Syndrome. DM was supplied by a donkey farm from central Italy conforming with EU regulation 853/2004. Eighteen bulk milk samples were taken monthly and analysed for gross and mineral composition, fatty acid profile, and vitamin D content. After an allergological work-up, nutritional plans including DM were prepared for 22 allergic children referring to the Allergy Unit of Meyer Children Hospital of Florence (Italy). Fat and vitamin D supplementations were supplied according to the age of the children. The nutritional state of the babies was evaluated considering weight and length /stature at the beginning and the end of the study. The gains were calculated in terms of Z-score ($Z=x-|X|/|$ s.d.). Mean and s.d. of milk chemical composition were calculated, and Z-score values were analysed by t-test. Similarly, to human milk, lactose was 7.05±0.150%, and proteins were 1.59±0.137%, with caseins representing about 50% of the total protein. Fat and ash were 0.31±0.053% and 0.37±0.022%, respectively. Like human milk, calcium and potassium were the main minerals and were 633.31±137.440 and 653.32±69.21 mg/L respectively, while zinc content was 3.16±1.500 mg/L. Unsaturated fatty acids were 48 g/100g of fat (Table 1). In addition, DM was richer in n3 linolenic acid (7.52±2.49 g/100g of fat) and vitamin D (1.97±0.454 µg/100ml), than both human and cow milk. The infants increased in weight and length/stature similarly to the reference population. In conclusion, DM was found to be nutritionally adequate using supplementations according to the age of the children. Further investigations in the field of farming techniques and genetic improvement of dairy donkeys are still ongoing.

The behaviour of dairy cattle on the day of calving

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The objective of this study was to use automated technology to describe the behaviour of cattle on the day of calving, for both normal calvings and assisted calvings. Data were collected from 32 multiparous and 12 primiparous Holstein dairy cattle (parity 0-7, mean 1.2 ± 0.2) to describe normal calving behaviour. Assisted calving was observed in 14 animals (parity 0-3, mean 1.1 ± 0.2). Animals with assisted calvings were matched to cows that had an unassisted calving based on parity, locomotion score, calf breed, calf sex, month and year of calving. An IceQube (IceRobotics Ltd., South Queensferry, United Kingdom) was fitted to cows 4 weeks prior to their expected calving date. Data for lying time, number of steps, motion index (total motion) and the total number of standing and lying bouts (postural transitions) were summarised into -2h periods prior to analyses. All analyses and data manipulations were carried out and investigated using R (version 3.4.4). Mixed effect models were used to determine behavioural changes in the -24h and -12h before calving. Piecewise regression with mixed-effects analyses were run for each behavioural variable to find breakpoints in the behavioural data. The behaviour of dairy cattle undergoes numerous changes on the day of calving. For cows that calved without assistance, there was an effect of -2h period on the number of postural transitions, total step count, and motion index ($P < 0.001$) and lying time duration ($P = 0.02$) in the -12h before calving. Interactions on the day of calving were found between -2h period and parity for the number of postural transitions, step count and motion index. In support of this finding, the piecewise regression analyses concluded that behavioural changes on the day of calving occurred at different time points for primiparous cows and multiparous cows. On the day of calving, assisted cows had 16.5% more postural transitions compared to non-assisted cows ($P < 0.001$). These findings indicate that parity should be considered when predicting the time of calving, and the number of postural transitions could be used as an indicator of animals that require assistance at calving.

Assessing the use of thermal imaging in detection of pyrexia in pre-weaned calves

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An elevation in body temperature can be an indicator of acute illness, such as respiratory disease in pre-weaned calves. Core body temperature is commonly measured using a rectal thermometer. However, this method is invasive and involves handling of the calf. An alternative method using thermal imaging of the area in and around the medial canthus of the eye was investigated. Thermal images were taken daily from 125 Holstein calves (8-40 days of age) under on-farm conditions. Simult aneous measurements of rectal temperature (oC), ambient temperature (oC), relative humidity (%) and wind speed were also taken. Water vapour density (g/m3) was calculated retrospectively. A restricted maximum likelihood approach (REML) was used to predict rectal temperature using thermal image data corrected for environmental parameters. The correlation between rectal temperature and thermal image temperature was 0.28. The inclusion of ambient temperature (p 38.8oC, thermal imaging had 55% and 73% sensitivity and specificity respectively, and 6% sensitivity and 100% specificity at determining temperatures >39.5oC. Therefore more developmental work is required in the use of thermal imaging for the prediction of rectal temperature in calves.

Data driven disease detection – an example in dairy calves

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Early detection of disease in dairy animals is important, as it allows for early treatment which reduces the severity and duration of the disease for the affected individual, reduces disease spread to group-members and reduces costs and antibiotic use for the farm. Changes in behaviour can be used as indicators of early-stage disease. Remote sensing devices can be used to detect these changes, allowing behaviour to be monitored remotely and continuously over long periods. Studies have been carried out in adult cattle, but little has been done for calves. This study (funded by AHDB UK) aimed to determine whether changes in activity and feeding behaviour can be used as early-warning indicators of disease in dairy calves. One hundred male Holstein pre-weaned calves (age ~8-40 days) were used in the study. Each calf was fitted with an Axivity® monitor on the hind-leg to record activity. Feeding behaviour was recorded through automatic milk feeders. To detect presence of disease, each calf was assessed daily using a modified version of the 'Wisconsin Scoring System' which is the current best practice method for detecting ill health in calves and involves recording rectal temperature, coughing, nasal and ocular discharge. Activity and feeding variables were extracted for each calf. The peak day of the most extreme illness event (i.e. highest scoring day of disease event with the highest Wisconsin score) was identified for each calf. Data from ill and healthy calves were paired for analysis, with similar age and weight. Data for 22 events were analysed. The results showed that ill calves lay for longer, fed for a shorter time and had fewer feeder visits (with consumption) each day compared to healthy calves (means and sems for ill and healthy calves respectively: daily lying (h): 17.6±0.3; 16.7±0.2; $P<0.01$; feeding time (mins): 19.3±1.4, 22.8±1.5; $P<0.05$; visits: 2.1±0.2, 3.2±0.4; $P<0.05$). Changes in lying behaviour and feeder visits were also evident up to 2 days before the day of illness. The results show that activity and feeding behaviour can be used to detect disease in calves, and changes in behaviour can be used as early-warning indicators of disease.

Microarray-based transcriptome profiling of sheep fed with a high iodine-supplemented diet

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Iodine (I) is an essential trace element for both humans and animals. It has only one, but vit al, role in the body as a constituent of thyroid hormones, and its importance for livestock has been well-documented over the past years. The I content of farm animals' diet is important not only due its impact on the animals' homeostasis but also on humans who eventually consume the I-containing food (e.g., milk) obtained from those animals. Since little information have been reported about the effects of I-supplemented diet on transcriptomic profile in small ruminants, the aim of this study was to investigate the role of I supplementation (40 days) on gene expression regulation in sheep. Using a microarray we identified a total of 254 gene differentially expressed in the I group using a Log2FC > 2 and a p-value Our data highlight the influence of iodine supplementation on enhance effects of thyroid on growth performance and productive traits. Thus, our findings contribute to the growing body nutrigenomics research in ruminants.

Rational design of food products as promoters of metabolic health

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Dietary components have effects on human metabolism which go beyond their immediate energy content. The composition of macronutrients has decisive impacts on how nutrients are metabolised. E.g., a very low carbohydrate, high fat diet leads to weight loss, whereas a carbohydrate rich and fat rich diet leads to weight gain.
Micronutrients and other dietary components (like pre- and probiotics) can also have significant effects on the regulation of whole body metabolism. This is mediated by the interaction of the gut microbiome with intestinal signalling circuits, like the bile acid system. A better understanding of the interactions between diet, microbiome and intestinal signalling will support the rational design of food products, including dairy products, with a beneficial metabolic health profile.

Integration of multiple sensor data streams to assist in the detection of mastitis

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Incidence of mastitis within the dairy industry is estimated to cost the UK dairy industry around £200Million GBP annually in terms of lost production. This paper examines the combination of two independent sensor technologies for early detection of mastitis. Behavioural monitoring collars that measure feeding and ruminating times were combined with milk component analysis and quarter conductivity from an au tomated milking robot. Combining information from both sensors reduces the prevalence of false positives and increases the specificity of health alerts. Accurately detecting illnesses can enable early treatment which can significantly benefit both farm profitability and animal welfare, increasing short- and long-term milk production, reducing milk refusals, and reducing use of medication in treatment of animals. 200 Holstein-Freisian cows of varying parity and stage of lactation were milked over 4 Fullwood Merlin2 robots providing per-quarter conductivity and yield measurements per milking, and bulk fat, protein, and lactose milk content. Farm operatives were challenged to monitor and identify animals with mastitis. Table 1 shows a summary of the trial findings. The collar alerts (both rumination and feeding behavior) showed strongest predictive ability, followed by milk conductivity and time between milkings. In 74% of the cases the feeding/rumination signatures alert ed at least one day before the condition was noted by the farmer. More than 90% of cases were detected at least as early as the farm observations. Given that this was a trial where the farm operatives were specifically tasked with maintaining a close watch on the herd for welfare related indicators, this performance is impressive. Conductivity changes were typically observed after either rumination or eating alerts from the collar. Conductivity alerted in 25% of cases at least 1 day in advance of the farmer, was equal to or in advance of the farmer in 50% of cases. Changes in the fat to protein ratio were not observed to be a strong indicator for mastitis; only 13% of cases had a drop in fat:protein ratio in advance of the farmer.

Challenges and potential of dairy processing sludge recycle for agronomic use

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Dairy processing sludge (DPS) is the by-product of the wastewater treatment process at milk processing facilities (Figure 1) concerning the production of a range of products like butter, cheese, milk powders, cream, and whey powders. Increased worldwide demand for dairy products creates an added challenge of DPS management, which has associated environmental regulation. Sustainable recycling of DPS could maximise industrial symbiosis for operational performance and resource efficiency. In Europe, dairy industry accounts for processing of about 160 million tonnes of milk, which is the largest in the world. After the European milk quotas abolition in 2015, milk production across the EU countries increased significantly. For example, the Irish dairy industry has experienced a rise of up to 46% in 2017 compared to the average milk production of the reference years 2007 to 2009. This has posed the challenge of tackling more dairy wastewater treatment and subsequent sludge generation. The present study was aimed to estimate the amount of DPS and to develop a seasonal database of nutrient and metal content of the dominant DPS type across Ireland. Heretofore, this has not been investigated and there is lack of knowledge pertaining to the nutrient and metal content of this waste stream. This is unfortunate as many of these wastes are being land spread without knowledge of their fertiliser equivalent value. Herein, a database of nutrients and trace metals composition of this waste stream was developed consisting of 63 seasonal sludge samples from 9 major dairy processing plants in Ireland. The r esults indicated that DPS samples are heterogeneous in respect to the wastewater treatment process, and therefore the concentrations of N, P and K varied significantly across sludge type. Heavy metal concentrations were significantly lower in DPS than those regulated by the EU for controlling metals accumulation in agricultural land due to sludge recycling. Overall, the outcomes demonstrate that dairy industry derived sludge is an important secondary resource for nutrient recovery to benefit agri-based recycling and circular economy. The characterization profile from this study would serve as a reference database for future investigation on the valorisation process of DPS.

Inline estimation of multiple heat damage tracers in milk by front-face fluorescence

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In order to evaluate the thermal damage in milk, a series of chemical indicators such as lactulose, HMF, furosine, etc., have been proposed. But the methods of analysis available must be performed in a specialized laboratory, being laborious, expensive and/or slow. Techniques for the determination of 5-hydroxymethylfurfural (HMF), sulfhydryl / thiol (-SH) groups, whey proteins (native, denatured bound to casein and denatured in the form of soluble aggregates), vitamins A, B2, and C, furosine, lactulose, and casein micelle particle size were implemented. Likewise, front-face fluorescence determination instrumentation and methods were implemented. Prediction models for a large number of thermal damage reference indicators were developed. Through the information extracted from the fluorescence analysis, a number of prediction models were successfully obtained for most traditional reference markers of thermal damage using a few fluorescence-based predictors. The most relevant models obtained were: 1) [-SH]: R2 = 0.92, SEP = 0.01 µmol/L-cysteine L; 2) [HMF]: R2 = 0.94, SEP = 6.54 µmol/L; 3) [lactulose]: R2 = 0.91, SEP = 1.8 10-7 mol/L; 4) [furosine]: R2 = 0.92, SEP = 9.15 mg/kg protein; 5) [native whey proteins]: R2 =0.97, SEP 35.3 µg/mL; 6) [denatured whey protein aggregates]: R2 = 0.70, R2 = 134 µg/mL; 7) [bound whey protein]: R2 = 0.98, SEP = 38.7 µg/mL; 8) casein micelle size, R2=0.99, SEP 2.77 nm; 9) [ascorbic acid]: R2 = 0.95, SEP = 0.72 ppm; 10) [riboflavin]: R2 = 0.94, SEP = 0.01 ppm; 11) [retinol]: R2 = 0.87, SEP = 2.47·10-2 mg/L. Results obtained encouraged usage of front-face fluorescence as a rapid method for thermal damage assessment in milk.

Mastitis research at Moredun Research Institute

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Pressure to reduce the use of antimicrobials in agriculture continues to grow as a result of the threat to human health from antimicrobial resistance. The development of alternatives to antimicrobials in mastitis control is therefore seen as an industry and social priority. Despite years of research, few commercial mastitis vaccines are available. However, the development of alternative measures such as vaccines has been hampered by the poor knowledge of pathogenesis of mastitis and the immune response in the mammary gland. At Moredun Research Institute over the past years, we developed models of cattle and sheep mastitis with the aim of understanding the factors involved in the pathogenesis and the immune response of mastitis. Specifically, we developed a cattle and sheep Streptococcus uberis model and sheep Mannheimia haemolytica and Staphylococcus aureus models. These in vivo models allowed us to characterize the immune response to the infection and the re-infection by analysing cytokines and specific immune cells phenotypes present in the mammary gland. In particular, we found that resolution of intramammary infection might be related with a strong cellular Th17 type response in the mammary gland thus a mastitis vaccine that targets this type of response is desirable. Beside in vivo models, we use in vitro models to investigate specific aspects host pathogen interactions such as the ability of mastitis pathogens to adhere and invade the mammary epithelial cells and their ability to evade killing the bactericidal activity of host immune cells such as macrophages and neutrophils.

Oestrus Effects on Cow Behaviour and Milk Yield

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Detecting oestrus is crucial to maintaining a high reproductive performance in dairy cows. This study aimed to determine the effect of oestrus on cow behaviour and milk production. Data originated from the research farm of Scotland's Rural College (Crichton Royal Farm, Dumfries, UK), October 2013 to June 2018. The cows were loose-housed in four pens of 24 cubicles with rubber mattresses, top-dressed with sawdust/oat husks, and grooved concrete alleys with automatic scrapers. Cows were milked thrice daily in a milking parlour. Cows had *ad libitum* access to water from automatic water troughs (Hokofarm Group, Marknesse, The Netherlands) and to TMR from automatic feed stations (RIC, Hokofarm Group, Marknesse, The Netherlands). On a hind leg, cows wore a 3-dimensional activity sensor (IceQube, IceRobotics, South Queensferry, UK), measuring cow behaviour and triggering a heat alert during oestrus. Only confirmed oestrus events, defined as a heat alert with insemination leading to conception, and occurring between 5-300 days in milk (DIM) were analysed. There were 1461 confirmed oestrus events from 511 cows. Lying Time (LT, h/d), Motion Index (MI, an expression of overall activity, acceleration/d), Eating Time (ET, h/d), Drinking Time (DT, min/d) and Milk Yield (MY, l/d) were calculated. The day of confirmed oestrus was defined as day zero. Daily LT, MI, ET, DT as well as MY from days -5 to 5 were analysed in a linear mixed model with parity (1, 2, 3+), DIM, genetic line (High or Control MY line), housing (housed, grazing) as fixed effects, and cow as random effect. As expected, MI increased by 77% on days -1 and 0 ($P<0.001$ for both days). Conversely, LT decreased by 17% on days -1 and 0 ($P<0.001$ for both days). Affected for longer, DT decreased by 16% ($P<0.01$) and ET by 15% ($P<0.001$) on days -1, 0 and 1 during oestrus. MY decreased by 3% on days -1, 0 and 1, but only significantly compared to days -5, -2 and 5 ($P<0.05$). The extent of these effects, especially on cow time-budget, is important to consider and account for when detecting disease patterns. We thank InnovateUK (IUK #133601) for support.

The assessment of farm level heat stress risk in a Scottish dairy herd.

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An increased focus has been placed on heat stress resulting in production losses, increased disease incidence and decreased animal welfare (Polsky et al., 2017). Temperature humidity index (THI) combines relative humidity and environmental temperature to estimate the risk of heat stress. Maximum THI thresholds at which animals demonstrate heat stress vary dependent on geographical location and climate type: THI62 (NRC,1971), THI68 (Collier et al., 2009) and THI72 (Armstrong 1994). Currently, little data relate to the risk of heat stress to dairy cattle within the UK. The aim of this study was to identify if THI thresholds were surpassed at farm level in a Scottish dairy herd. A 500 cow Holstein herd in central Scotland was recruited. Cattle are housed indoors all year round, milked three times daily and fed a total mixed ration. Average yield is 9000L. HOBOTM data loggers were installed at a height of 2.5m, away from direct sunlight and drafts in four locations: high-yielding group, postpartum group, "close-up" dry cow, and collecting yard. Temperature and relative humidity data were recorded every 30 minutes between July 2017 and March 2018. All data were stored in Excel™ for descriptive analysis. The THI was calculated via $(1.8 \times T + 32) - [(0.55 - 0.0055 \times RH) \times (1.8 \times T - 26)$. The maximum and minimum THI across all four locations was THI74 in July and THI35 in February. The THI thresholds were exceeded on 84 days for THI62; 38, THI68; 6, THI72 between July and September. The percentage of total time the THI exceeded threshold on days with potential heat stress varied up to 84%, THI62; 40%, THI68; 17%, THI72. Additionally, the THI recorded in the collecting yard increased by up to 15% whilst compared to other management groups during milking periods.These data suggest a risk of heat stress on this Scottish farm during the summer months and an increased risk of heat stress during milking times. These data are part of a larger ongoing study comparing THI thresholds beyond which production output and disease incidence start to deteriorate in Scottish dairy cows.

Study of potential environmental factors predisposing ewes to subclinical mastitis in Greece

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Objectives were to investigate association of prevalence of subclinical mastitis with environmental (climatic and topographic) factors and to identify factors potentially predisposing ewes to the disease. Milk samples were collected from 2,198 sheep in 111 farms, in all 13 administrative regions of Greece, for bacteriological and cytological examination. Data on farm location were collected in the field using hand-held Global Positioning System Garmin units. The geo-references were resolved to specific farm level. Prevalence of subclinical mastitis was 0.260. Main aetiological agents were staphylococci (Staphylococcus aureus and coagulase-negative species), which accounted for 0.699 of all isolates recovered. In a multivariable mixed-effects analysis, the two environmental variables found to be associated with increased prevalence of subclinical mastitis were the minimum temperature of coldest month (coefficient: -0.084±0.033, P = 0.014) and the mean temperature for 30 days prior to sampling date (coefficient: 0.031±0.014, P = 0.029). The combination of the two factors points out that extreme temperatures adversely affect occurrence of subclinical mastitis. Although environmental factors are outside the control of farm managers, nevertheless the results can be of value in health management of flocks. During periods of adverse climatological conditions, when risk for mastitis would be increased, preventive measures for the disease should be applied meticulously. Moreover, in farms located in areas where extreme temperatures would occur frequently, long-term preventive measures should be taken.

Age reflects on the occurrence of bovine periodontal lesions

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Bovine periodontitis is a progressive infectious process that causes cumulative alterations readily seen in slaughtered animals and herds raised extensively. The periodontal lesions can be characterised by the formation of periodontal pockets, gingival recession, clinical attachment loss and premature loss of teeth. As a polymicrobial infection, its aetiology and pathogenesis are associated with the presence of a biofilm, periodontal pathogens and immune-inflammatory mechanisms. In periodontal disease, epidemiological studies are important to describe the health status of populations, elucidate the aetiology of the disease, identify risk factors, prevent its occurrence and design control measures. No published study has investigated the risk factors associated with periodontitis in cattle or other ruminants. The present study evaluated some possible risk factors associated with the occurrence of periodontal lesions in cattle slaughtered in the West of Scotland. From 250 cattle examined in an abattoir in the West of Scotland, 35 dental arches with periodontal lesions and 40 considered periodontally healthy were selected. Logistic regression analysis was used to evaluate the association between the independent variables, gender, age and breed with periodontitis. The average age of animals with periodontitis was 7.4 years (range 1.5 to 16.5 years) and for periodontally healthy animals was 2.9 years (range 1.4 to 10.6 years). For statistical analysis, the 75 animals were grouped into two categories, dairy (n=20) or beef (n=55) cattle. Age of animals was significantly associated with the presence of periodontal lesions. For every year of age, cattle were 1.53 times likely to have periodontitis ($p<0.001$). Gender was not significantly associated with periodontitis. Beef cattle were 0.36 times as likely to have periodontitis compared to dairy cattle ($p=0.054$).It is likely that bovine periodontitis has a significant impact on the welfare of affected animals, as it can be a painful, chronic condition leading to difficult feeding and consequent loss of body condition, increased susceptibility to disease and reduced productivity. We hypothesise that increasing age may not represent a risk factor outright for the development of bovine periodontitis but may simply reflect the cumulative exposure over time to environmental risk factors.

Table 1: Distribution of breed and sex of 75 cattle with periodontal lesions (n=35) and periodontally healthy (n=40) selected from 250 slaughtered animals in Scotland

Breed	Periodontitis (n=35)		Healthy (n=40)		Total
	F	M	F	M	
Limousin	6	2	17	1	26
Holstein Friesian	7	0	4	0	11
Aberdeen Angus	5	0	4	2	11
British Friesian	6	0	2	0	8
Simmental	3	0	3	0	6
Shorthorn	1	1	1	0	3
Belgian Blue	2	0	0	0	2
British Blue	1	0	1	0	2
Charolais	0	0	1	1	2
Belted Galloway	0	1	0	0	1
Blonde D´Aquitaine	0	0	1	0	1
Highland	0	0	0	2	1
Luing	0	0	1	0	1