

Editorial: When I was but a lad

Christopher H Knight

UK

Full-fat corn germ in diets for dairy cows as an alternative to ground corn

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Brazil and Sweden

Abstract

The experiments reported in this research paper address the effects of replacing ground corn (GC) with full-fat corn germ (FFCG) on nutrient intake and digestibility, nitrogen utilization efficiency, performance, and predicted methane production in dairy cows fed cactus cladodes and sugarcane. We hypothesized that the inclusion of FFCG in the diet would not alter the performance of lactating cows but would reduce the predicted methane production *in vivo*. Ten multiparous Holstein cows at 90 ± 10 d of lactation and yielding 24.2 ± 3.5 kg milk/d were assigned to dietary treatments consisting of different levels of replacement of GC by FFCG (0; 25%; 50%; 75% and 100% of diet dry matter) in a replicated 5 x 5 Latin square design with 21-d periods. Methane production was predicted using an automated gas *in vitro* production system. Except for ether extract intake, which increased, the intake of all nutrients decreased linearly with the replacement of GC by FFCG. The digestibility of dry matter, organic matter and neutral detergent fiber reduced, whereas the digestibility of ether extract increased linearly with FFCG. There were no changes in the digestibility of crude protein. The nitrogen intake and daily excretion in urine and feces decreased, while nitrogen use efficiency increased linearly. There was no significant effect of diets on nitrogen balance or microbial protein synthesis and efficiency. The yield of protein, lactose and total solids in milk showed a quadratic behavior. On the other hand, milk fat yield and energy-corrected milk yield decreased linearly with the replacement of GC by FFCG. No effect on pH or ammonia nitrogen was observed. The production of methane (CH_4 , g/kg DM) and total CH_4 (g/d), and CH_4 intensity decreased linearly with the replacement of GC by FFCG. In conclusion, FFCG has been shown to be an effective source of fat to reduce methane production in dairy cows, partially supporting our initial hypothesis. However, as it decreases milk fat production, it is not recommended to replace more than 50% of GC by FFCG for lactating cows fed cactus cladodes and sugarcane.

Effect of postruminal supply of linseed oil in dairy cows: 1. Production performance and fate of postruminally available α -linolenic acid

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Abstract

Triacylglycerols (TAG) are the primary sources of preformed fatty acids (FA) for lipid synthesis in the mammary gland. However, polyunsaturated FA escaping ruminal biohydrogenation are selectively incorporated into cholesterol esters (CE) and phospholipids (PL). The aim of the current experiment was to study the effects of abomasal infusion of increasing amount of linseed oil (L-oil) on plasma distribution of α -linolenic acid (α -LA) and its transfer efficiency into milk fat. Five rumen-fistulated Holstein cows were randomly distributed in a 5 \times 5 Latin square design. Abomasal infusion of L-oil (55.9% α -LA) was performed at the rate of 0, 75, 150, 300, and 600 mL/d. Concentrations of α -LA increased quadratically in TAG, PL, and CE; a less steep slope was observed with an inflexion at an infusion rate of 300 mL L-oil per day. The increase in plasma concentration of α -LA was of a lower magnitude in CE as compared with the other two fractions, resulting in a quadratic decrease in relative proportion of this FA circulating as CE. The transfer efficiency into milk fat increased from 0 to 150 mL L-oil infused, and a plateau was maintained thereafter with greater levels of infusion (quadratic response). This pattern resembles the quadratic response of the relative proportion of α -LA circulating as TAG, and the relative concentration of this FA in TAG. Increasing the postruminal supply of α -LA partly overcame the segregation mechanism of absorbed polyunsaturated FA in different plasma lipid classes. Proportionately more α -LA was then esterified as α -LA, at the expense of CE, increasing its efficiency of transfer into milk fat. This mechanism appears to be surpassed in its turn when L-oil infusion was increased over 150 mL/d. Nevertheless, the yield of α -LA in milk fat continued to increase, but at a slower rate at the highest levels of infusion.

Effect of postruminal supply of linseed oil in dairy cows: 2. Milk fatty acid profile and oxidative stability

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Abstract

Our objective was to study the effect of increasing postruminal supply of linseed oil (L-oil), as a source of *cis*-9, *cis*-12, *cis*-15 18:3, on milk fatty acid profile and to assess the resulting impact on the development of volatile degradation products during the storage of homogenized milk. Five Holstein dairy cows fitted with a rumen cannula were randomly distributed in a 5 \times 5 Latin square design. Abomasal infusion of L-oil was performed at the rate of 0, 75, 150, 300, and 600 mL/d during periods of 14 d. The concentration of *cis*-9, *cis*-

12, *cis*-15 18:3 in milk fat increased linearly with L-oil dose. Concentrations of primary (conjugated diene and triene hydroperoxides) and secondary oxidation products (1-octen-3-one, propanal, hexanal, *trans*-2+*cis*-3-hexenals, *cis*-4-heptenal, *trans*-2, *cis*-6-nonadienal *trans*-2, *trans*-4-nonadienal) increased during 11 d of storage at 4°C of homogenized milk under fluorescent light. The magnitude of the increase (difference between final and initial measurements) was linearly greater for all nine lipid oxidation products evaluated in response to increasing level of infusion. Results of the current experiment have shown that milk enriched in *cis*-9, *cis*-12, *cis*-15 18:3 via post-ruminal supply of L-oil is highly prone to oxidative degradation. This low oxidative stability, exposed under controlled experimental conditions, would represent a major obstacle to those who aim to market milk enriched in polyunsaturated fatty acids.

Effect of crude glycerin supplementation via drinking water on feed intake, water intake and productive performance of dairy ewes

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Brazil

Abstract

This study was performed to determine the effects of crude glycerin (CG) supplementation in drinking water on DM and nutrient intake, milk production, milk composition, and serum glucose. Twenty multiparous Lacaune × East Friesian ewes were randomly distributed into four dietary treatments throughout the lactation cycle. Treatments consisted of doses of CG supplementation via drinking water as follows: (1) no CG supplementation, (2) 15.0 g CG/kg DM, (3) 30.0 g CG/kg DM, and (4) 45.0 g CG/kg DM. DM and nutrient intake were reduced linearly with CG supplementation. CG linearly reduced water intake when expressed as kg.day⁻¹. However, no effect of CG was observed when it was expressed as a percentage of body weight or metabolic body weight. The water to DM intake ratio was increased linearly with CG supplementation. No effect of CG doses on serum glucose was observed. The production of standardized milk decreased linearly with the experimental doses of CG. Protein, fat, and lactose yield were linearly reduced with the experimental doses of CG. Milk urea concentration was quadratically increased with CG doses. Feed conversion was quadratically increased by treatments during the pre-weaning period ($P < 0.05$), in which the worst values were observed when the ewes were supplemented with 15 and 30 g CG/kg DM. The N-efficiency was linearly increased with CG supplementation in drinking water. Our results suggest that dairy sheep can be supplemented with CG up to 15 g/kg DM in drinking

water. Greater doses are not beneficial for feed intake, milk production, and the yield of milk components.

Predicting live weight using body volume formula in lactating water buffalo

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Mexico, South Africa and Brazil

Abstract

Live weight (LW) is an important piece of information within production systems, as it is related to several other economic characteristics. However, in the main buffalo-producing regions in the world, it is not common to periodically weigh the animals. We develop and evaluate linear, quadratic, and allometric mathematical models to predict LW using the body volume (BV) formula in lactating water buffalo (*Bubalus bubalis*) reared in southeastern Mexico. The LW (391.5 ± 138.9 kg) and BV (333.62 ± 58.51 dm³) were measured in 165 lactating Murrah buffalo aged between 3-10 years. The goodness-of-fit of the models was evaluated using the Akaike Information Criterion (AIC), Bayesian Information Criterion (BIC), coefficient of determination (R²), mean squared error (MSE) and root MSE (RMSE). In addition, the developed models were evaluated through cross-validation (k-folds). The ability of the fitted models to predict the observed values was evaluated based on the RMSEP, R², and mean absolute error (MAE). LW and BV were significantly positively and strongly correlated ($r=0.81$; $P<0.001$). The quadratic model had the lowest values of MSE (2788.12) and RMSE (52.80). On the other hand, the allometric model showed the lowest values of BIC (1319.24) and AIC (1313.07). The Quadratic and allometric models had lower values of MSE and MAE. We recommend the quadratic and allometric models to predict the LW of lactating Murrah buffalo using BV as a predictor.

Investigation of conceptus stimulated gene expression in buffalo peripheral blood mononuclear cells as potential diagnostic markers of early pregnancy

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Abstract

Exploration of novel strategies for early pregnancy diagnosis is pivotal in enhancing the reproductive potential and monetary gains from dairy herds. In buffalo, the trophoblast cells of the elongating conceptus secrete interferon-tau, that stimulates the transcription of various genes in peripheral blood mononuclear cells (PBMC) during the peri-implantation period. We explored the differential expression of classical (*ISG15*) and novel (*LGALS3BP* and

CD9) early pregnancy markers in PBMC of buffaloes during various stages of pregnancy. Natural heat was detected in buffaloes by assessing the vaginal fluid, and artificial insemination (AI) was done. Whole blood was collected from the jugular vein in EDTA-containing vacutainers for PBMC isolation before AI (0-day) and 20, 25 and 40 days post-AI. On day 40, transrectal ultrasonography examination was performed to confirm pregnancy. The inseminated non-pregnant animals served as control. Total RNA was extracted using the TRIzol method. The temporal abundance of *ISG15*, *LGALS3BP* and *CD9* genes in PBMC was compared between pregnant and non-pregnant groups (n=9 per group) using real time-qPCR. Results showed transcripts of *ISG15* and *LGALS3BP* were more abundant at 20 day in the pregnant group compared to the 0 day and 20 day values of the non-pregnant group. However, due to variability in expression, threshold (Ct) cycle of RT-qPCR alone could not distinguish pregnant and non-pregnant animals. In conclusion, *ISG15* and *LGALS3BP* transcripts abundance in PBMCs are potential candidate biomarkers for early prediction of buffalo pregnancy 20-days post-AI, but further work is required to allow the development of a reliable new methodology.

Flagella are an important virulence factor in the subclinical persistence of *Escherichia coli* in bovine mammary gland

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Brazil

Abstract

We compared the virulence profile and REP-PCR genotypes of *Escherichia coli* strains isolated from subclinical and clinical mastitis cases and dairy farm environments in Minas Gerais State, Brazil, to determine virulence factors and genotypes potentially associated with subclinical persistence in the udder. The virulence profile was obtained by the search for three virulence genes: *lpfA* (long polar fimbriae), *fliC* (flagella), and *escN* (type III secretion system). Subclinical isolates exhibited mainly the *fliC* gene (33.33%) and *fliC* + *escN* genes (30.30%). Clinical isolates exhibited mainly *fliC* + *escN* genes (50%) and environmental isolates the *lpfA* + *escN* genes (58.04%). Strains isolated from subclinical mastitis showed 6.75 times more positivity to *fliC* than environmental isolates. Thirty-four genotypes were observed in the REP-PCR analysis, and clinical mastitis isolates indicated more genetic proximity to dairy farm environment isolates than subclinical mastitis isolates. In conclusion, the results suggested that flagella may be an important virulence factor for mammary persistent *E. coli* infection in cattle, however, none of the *E. coli* REP-PCR genotypes were associated with subclinical infection.

Virulence factors, antimicrobial resistance and phylogeny of bovine mastitis-associated *Streptococcus dysgalactiae*.

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Brazil, USA and China

Abstract

We carried out a thorough genetic evaluation of *Streptococcus dysgalactiae* isolated from clinical bovine mastitis cases, as well as performing a phylogenetic analysis to represent the evolutionary relationship between *S. dysgalactiae* sequences. A total of 35 *S. dysgalactiae* strains were isolated from cases of clinical mastitis identified at a large commercial dairy farm located near Ithaca, New York. Whole-genome sequencing identified twenty-six antibiotic resistance genes, four of which were acquired genes, in addition to fifty virulence genes. Multi-locus sequence typing detected three new sequence types (STs). We conclude that a high proportion of this microorganism carries multiple virulence determinants and resistance genes, and that this indicates its potential to cause mastitis. Eight different STs were identified, of which ST453 (n=17) was the most prevalent and ST714, ST715, ST716 were novel STs.

The prevalence of coagulase-negative staphylococcus associated with bovine mastitis in China and its antimicrobial resistant rate: a meta-analysis

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China

Abstract

To contribute to the treatment decision and optimize coagulase-negative staphylococcus (CNS) control programs, we conducted a meta-analysis to investigate the epidemiology and antimicrobial resistance rates of coagulase-negative staphylococcus associated with bovine mastitis in China. Three databases (PubMed, Google scholar and China National Knowledge Infrastructure database) were utilized to obtain relevant publications. A total of 18 publications were included in our research, and 3 of them included antimicrobial resistant (AMR) test. The pooled prevalence of coagulase-negative staphylococcus was 17.28%. Subgroup analysis revealed that the prevalence was higher in South China than in North China, was higher in 2011–2020 than in 2000–2010 and was higher in clinical bovine mastitis cases than in subclinical cases. The pooled AMR were most resistant to β -lactams, followed by tetracyclines, quinolones, nitrofurans, lincosamides, sulfonamides, amphenicol and aminoglycosides. The pooled AMR rate of coagulase-negative staphylococcus was lower in 2011–2020 than in 2000–2010. Although the prevalence of CNS showed an increasing trend over 20 years, the AMR rate showed a decreasing trend, and the clinical type of mastitis was

the most frequent and the prevalence was highest in South China. Finally, CNS was most resistant to β -lactams amongst the eight groups of antimicrobial agents.

Seasonal, physiological and bacteriological risk factors for subclinical mastitis in dairy cows maintained under different farming conditions

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India

Abstract

Subclinical mastitis (SCM) is a major health problem of dairy animals in India and across the globe. An identification of potential risk factors of SCM can help for efficient udder health management in dairy animals. In this study, apparently healthy cows (HF crossbred: n=45; Deoni: n=43) were screened for SCM during different seasons through milk somatic cell count (SCC: reference test using 200×10^3 cells/ml as cut off value), California mastitis test (CMT) and differential electrical conductivity (DEC) test at an organized research farm. SCM positive milk samples (n=34) were inoculated in selective media for *Coliform sp*, *Streptococcus sp.* and *Staphylococcus sp.* and DNA was isolated (n=10) for species confirmation by 16s rRNA method. Both bivariate and multivariate models were used for risk assessment. We found the cumulative prevalence of 31% and 65% SCM in Deoni and crossbred cows, respectively. Screening of 328 crossbred cows under field conditions revealed point prevalence of 55% SCM. Multivariate analysis revealed stage of lactation (SOL), milk yield in previous lactation and test day milk yield in Deoni cows, as well as parity and mastitis treatment history in current lactation in HF crossbred cows as risk factors. SOL was a significant factor under field conditions. Receiver operated characteristic curve analysis revealed better accuracy of CMT than DEC. We found more mixed infections due to *Staphylococcus sp.* and *Streptococcus sp.* in culture, while 16s rRNA based molecular method revealed lesser-known pathogens associated with SCM. It is concluded that SCM prevalence rate is higher in crossbred than indigenous cows and these breeds have different risk factors for SCM. HF crossbred cows had similar SCM prevalence rate under different farming conditions, where CMT can be used for SCM diagnosis with excellent accuracy. The 16s rRNA method is useful for specific identification of lesser known and emerging mastitis pathogens.

Pathogens in milk of goats and their relationship with somatic cell count

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Slovak Republic, Czech Republic and France

Abstract

We evaluated the presence of bacterial pathogens in the milk of goats and their relationship with somatic cell count (SCC) and milk composition. The study was performed on a dairy farm in northern Slovakia. Half udder milk samples were collected from goats in June and July. The samples were divided on the basis of SCC into 4 bands (SCC1 lowest to SCC4 highest). Bacterial pathogens were only detected in 13% of samples. SCC3 and SCC4 had 15 and 25% positive samples respectively compared with SCC1 (2%) and SCC2 (14%). Coagulase negative staphylococci (CNS) were the most common isolates (73%), of which *Staphylococcus caprae* was the most frequently isolated (65%). In samples with $\geq 1000 \times 10^3$ cells.mL⁻¹ (SCC3, SCC4) there was higher somatic cell score (SCS) in the presence of a pathogen (7.48 ± 0.11) than without a pathogen (7.16 ± 0.05 , $P < 0.01$). Statistically significant but weak negative correlations were observed between SCS and lactose, dry matter and non-fat dry matter. In conclusion, a higher percentage of bacteriologically positive milk samples was observed in both SCC3 and SC4 groups but this does not explain the aetiology of high SCC in the milk of goats that are apparently free of bacteria. As a diagnostic tool, SCC is probably less useful in goats than in cows.

Detection of lytic phage infecting flavour-producing strain of *Lacticaseibacillus paracasei* in the dairy effluents of Kerala

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India

Abstract

The performance of the starter culture is a critical factor that decides the quality of fermented milk. Dahi is a fermented milk product popular in India made using a mixed starter culture of lactic acid bacteria comprising acid and flavour producers. The prevalence of bacteriophages in the dairy environment can critically affect the activity of these starter cultures resulting in starter failure. As there is little information available on the occurrence of bacteriophages in the dairy environment of Kerala, this research communication examines the presence of lytic

bacteriophages acting against three potential flavour-producing strains of *Lacticaseibacillus paracasei* (*Lc. paracasei*). Dairy effluent samples were screened for the presence of phages against the strains of *Lc. paracasei* by the multiple host enrichment method. Plates showing clearance zone in spot assay were confirmed for the presence of phages by double layer agar assay. The plaques obtained in the double-layer agar assay were purified for further identification by next-generation sequencing. A bacteriophage infecting one of the three strains of *Lc.paracasei* was detected by the plaque assay and the blast annotation of the bacteriophage sequence found 86.05% similarity of the phage to *Siphoviridae* family. The study endorses the need for monitoring phages in the dairy environment to control phage-related starter failure in the state of Kerala.

Sonication as a pre-processing step to reduce acid whey generation during Greek yoghurt manufacture

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Australia and Sri Lanka

Abstract

We investigated sonication as a pre-processing step to reduce acid whey generation during Greek yoghurt manufacture. The generation of a large amount of acid whey during the production of Greek yoghurt is an ongoing problem in the dairy industry and many studies are currently focusing on reducing it. We focused on the use of ultrasonication as a novel approach to minimize the casein fraction in the acid whey stream and simultaneously improve the gel properties. Ultrasound applied before the fermentation modified the structural properties and bonding behaviours of milk proteins, and enhanced the retention of casein in the yoghurt gel after the fermentation and straining steps. Therefore, the use of low frequency ultrasonication as a pre-processing step may have the potential to provide significant economic benefits to the Greek yoghurt manufacturing process. Moreover, it improved the nutritional and physicochemical properties compared to regular Greek yoghurts.

Effects of calcium chloride substitution on the physicochemical properties of Minas Frescal cheese

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Brazil

Abstract

The aim in this research paper was to investigate the effect of using calcium monophosphate (MCP) and MCP mixed with commercial phosphates salts, in total or partial replacement of calcium chloride (CaCl_2) in the manufacture of Minas Frescal cheese. Initially, model cheeses were made to perform the rheological analysis during the coagulation process. Of these, the five best treatments were chosen to carry out the production of Minas Frescal cheese, used only CaCl_2 and MCP, and partial replacements of MCP + polyphosphate, MCP + potassium monophosphate (MKP) and MCP. The cheeses showed no significant difference in physicochemical composition, yield and syneresis, however, the cheese with partial replacement of CaCl_2 by MCP + polyphosphate and MCP + MKP showed the highest hardness values, like the control. This demonstrates that it is possible to replace calcium chloride without significant changes in the physicochemical characteristics and yield of Minas Frescal cheese, and it is still possible to modulate the hardness of the cheese produced according to the type of calcium/phosphate source used. This allows the industry to replace the source of calcium in the manufacture of Minas Frescal cheese according to the desired hardness.

Effects of time and temperature of storage on chemical and nutritional characteristics of raw milk for Provolone Valpadana PDO cheesemaking: a multivariate approach

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Italy

Abstract

We evaluated the possibility of increasing the storage temperature of raw milk for Provolone Valpadana cheesemaking, to identify the most suitable conditions of time and temperature for a pre-maturation process. We used Principal Component Analysis (PCA) to analyse the overall effects of different storage conditions on chemical, nutritional and technological characteristics of the raw milk. Four different thermal storage cycles, two at fixed temperature/time (6 and 12 °C for 60 hours) and two with two-phase thermal cycle (10 and 12 °C for 15 hours, followed by refrigeration at 4 °C for 45 hours) were studied. Although a moderate heterogeneity among raw milks from the 11 producers of Provolone Valpadana cheese was observed, PCA revealed the critical aspects of the extreme storage conditions (60

hours of refrigeration). Some samples resulted in anomalous behaviors, probably related to unexpected fermentation phenomena occurring with increasing storage temperature. The acidification and the increase in the contents of lactic acid, soluble calcium, and degree of retinol isomerization observed in the anomalous samples can compromise the technological functionality of milk. Conversely, the storage with a two-phase thermal cycle did not lead to variations in any measured characteristic, suggesting that mild refrigeration conditions (10 or 12 °C for 15 hours followed by 4 °C for 45 hours) could be a good compromise in favoring milk pre-maturation without altering its quality characteristics.

The effect of different production methods on quality parameters of Hatay Künefe cheese

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Abstract

We investigated four different production methods of Künefe cheese, emphasizing their industrial importance. The four methods used fresh Künefe cheese (FKC), salted Künefe cheese (SKC), Boru type Künefe cheese (BKC) and culture-added processed Künefe cheese (CPKC), all used in the production of Künefe, a syrupy dessert unique to the Middle East. FKC was produced as a result of curd formation from raw milk with rennet and then the fermentation of curd. SKC was manufactured using the salting method in addition to FKC production. BKC was obtained using the dry cooking process with emulsifying salts applied to cheese curd. CPKC was produced by applying heat treatment to raw milk and using starter culture before adding rennet that was different to Boru-type Künefe cheese. The composition, color, meltability, texture and sensory properties of the Künefe cheeses were examined. As a result of statistical analysis, it was determined that the effect of different production methods on the composition, meltability, texture (except springiness and cohesiveness) and sensory properties was significant in all cheeses ($p < 0.05$). CKPC was the most appropriate cheese in various properties.

Eggnog: process optimization and characterization of a dairy based beverage

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India

Abstract

Eggnog, a dairy based beverage, comprises both milk and egg proteins. We aimed at optimizing the eggnog formulation using Box-Behnken design of response surface methodology. The combined effects of milk (60-75), cream (25-35) and eggnog base (6-8, all three as g 100 mL⁻¹) were investigated on heat coagulation time, viscosity and thermal gelation temperature. ANOVA indicated that experimental data were well explained by a quadratic model with high check values ($R^2 > 0.94$) and non-significant lack of fit tests. Based on the responses, an optimized formulation of eggnog with 60.0 milk, 25.0 cream and 6.50 eggnog base (as g 100 mL⁻¹), could be considered best for manufacturing eggnog with desired attributes. This optimized formulation was characterized for physico-chemical, microbial and sensory attributes and the results indicated significantly higher fat and protein content than control formulation, but lesser lactose and total sugar content. Significantly higher viscosity, heat stability and lower thermal gelation temperature were also observed for the optimized formulation. Coliform, yeast and mold, *E. coli* and *Salmonella* counts were not detected in any sample but a significantly lower total plate count was observed for the optimized formulation.

Book review: Advances in sustainable dairy cattle nutrition

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UK