A blinkered view of tunnel vision

Christopher H Knight
UK

Supplementation of hydrogenated fat-embedded calcium gluconate improves milk fat content and yield in multiparous Holstein dairy cattle

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Abstract

This research communication reports the responses to supplementing dairy cattle with a hydrogenated fat-embedded calcium gluconate feed additive. The role of hindgut health in ruminant performance and wellbeing is an area of growing interest. Various prebiotic compounds have been used to promote lower gut health in various non-ruminant species. Calcium gluconate, a prebiotic compound, has previously been observed to increase milk fat yield when fed to ruminants in a form capable of resisting fermentation in the rumen, though the mechanism(s) behind this response remain unclear. The objective of this study was to compare the responses of lactating cattle to two different supplementation levels of a hydrogenated fat-embedded calcium gluconate (HFCG) product to evaluate a potential linear dose response. Forty-six lactating Holstein dairy cattle were used in a 3 x 3 replicated Latin square design with 28 d periods to evaluate a previously used dose of HFCG (approximately 16 g/d) with both a negative control and a dose of 25 g/d. Supplementation of multiparous animals with 16 g/d HFCG significicantly ($P<0.05$) increased milk fat yield and content relative to the negative control, and subsequently improved gross feed efficiency ($P<0.05$); additionally, the presence of a potential non-linear dose response was observed for these parameters. Responses when supplemented with 25 g/d HFCG did not differ from the negative control. No production responses were observed in primiparous animals. The mode of action of HFCG, in addition to the potential differential response in primiparous animals remains unclear and warrants further investigation.

Using machine learning methods to predict dry matter intake from milk mid-infrared spectroscopy data on Swedish dairy cattle

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Abstract

In this research communication we compare three different approaches for developing dry matter intake (DMI) prediction models based on milk mid-infrared spectra (MIRS), using data

collected from a research herd over five years. In dairy production, knowledge of individual DMI could be important and useful, but DMI can be difficult and expensive to measure on most commercial farms as cows are commonly group-fed. Instead, this parameter is often estimated based on the age, body weight, stage of lactation and body condition score of the cow. Recently, milk MIRS have also been used as a tool to estimate DMI. There are different methods available to create prediction models from large datasets. The main data used were total DMI calculated as a 3-day average, coupled with milk MIRS data available fortnightly. Data on milk yield and lactation stage parameters were also available for each animal. We compared the performance of three prediction approaches: partial least square regression, support vector machine regression and random forest regression. The full milk MIRS alone gave low to moderate prediction accuracy (R^2=0.07-0.40), regardless of prediction modelling approach. Adding more variables to the model improved R^2 and decreased the prediction error. Overall, partial least square regression proved to be the best method for predicting DMI from milk MIRS data, while MIRS data together with milk yield and concentrate DMI at 3-30 days in milk provided good prediction accuracy (R^2= 0.52-0.65) regardless of the prediction tool used.

Correlations of whole blood heavy metals with serum immunologic and oxidative markers during the early dry period and transition period of dairy cattle

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Abstract
Immunologic and oxidative alterations have been reported around calving in dairy cattle. In addition, the levels of heavy metals rise in the blood around parturition, which might affect body systems. Therefore, in this Research Communication we evaluate the changes in whole blood lead (Pb), arsenic (As), and cadmium (Cd) around calving, in comparison with the beginning of the dry period, and assess the correlations of these elements with immunologic factors and oxidative markers. Samples were collected from 30 clinically healthy dairy cows in the early dry period (-6w), one week before expected calving (-1w), and one week postpartum (+1w). The highest concentrations of Pb, As, and Cd were observed at -1w and all the three elements decreased after parturition leading to significantly lower As and Cd, compared to -1w (P<0.05). The lowest levels of tumor necrosis factor-alpha, immunoglobulin G, interleukin 4, interleukin 10 and haptoglobin were found at -1w simultaneous with the highest measures of the heavy metals, with tumor necrosis factor-alpha being significantly lower at this time (P<0.05). At -6w, As concentration was significantly (P<0.05) correlated negatively (r=-0.366) and positively (r=0.417) with total antioxidant capacity and malondialdehyde, respectively. Furthermore, at -1w Pb and As had significant (P<0.05) negative correlations with interferon gamma (r=-0.502) and interleukin 4 (r=-0.483), respectively. After parturition, Pb was observed to be negatively correlated with total

antioxidant capacity (r=-0.538, P<0.05). The observed results revealed that the alterations in immunologic factors and antioxidant capacity around parturition were correlated with Pb and As levels.

Salivary crystallization pattern: a possible unconventional tool for timing of insemination and early pregnancy diagnosis in zebu cows

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Abstract

The present study assessed if salivary crystallization pattern (ferning pattern formed as a result of the higher levels of salt content in the dried sample) could be used for estrus detection and for diagnosis of pregnancy/non-pregnancy in dairy cows. Saliva and blood samples were collected from non-pregnant cycling cows (Sahiwal breed; n=20) on alternate days from the day of estrus till next estrus. Then, all the cows were inseminated and saliva and blood sampling were continued further for a period of 22 days post-insemination. Pregnancy diagnosis was carried out on day 45 post-insemination and eight cows were found to be pregnant. The salivary crystallization pattern and estradiol:progesterone ratio during estrous cycle and during pregnancy were compared among these cows. Six types of salivary crystallization patterns were discerned; distinct patterns such as branch-like, fern-like, fir-like and combinations of these. Fern-like pattern was observed in all the cows on the day of estrus (first measurement day) and furthermore, all of the cows that subsequently became pregnant had fern-like salivary crystallization pattern at the time of insemination. Saliva of all the pregnant cows showed branch-fir type of crystallization pattern on day 16 post-breeding while only 50% of non-pregnant cows showed this pattern on day 16 of estrous cycle. The appearance of fern-like pattern was positively and significantly related to estradiol:progesterone ratio (r=0.86; p>0.001). The findings were validated on a separate group of cycling cows (n=32). We can conclude that salivary crystallization pattern might serve as a non-invasive and cost effective and easy to use cow-side tool for estrus detection and early pregnancy/non-pregnancy diagnosis in cows upon validation on a larger sample size.

Dairy animals and breast cancer: reflections on a long-term study from the 1970s that was never done

Malcolm Peaker
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Abstract

In this short commentary I recall a long-term experiment that was sketched out to determine if the low incidence of mammary cancer in dairy animals reflects a low incidence in these species generally or is the result of a protective effect of early pregnancy and long lactations. Although that experiment was never done, I discuss these questions in the light of developing knowledge on the incidence of cancer in ruminants generally and in the mammary gland in particular.

Mammary lipid secretion: a reassessment

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Abstract

Mammary lipid secretion is generally held to be unique and remarkably uniform between the many different orders of mammals. It produces a unit membrane bounded milk fat globule (MFG). The unit membrane is separated from the lipoprotein boundary of what was the cytoplasmic lipid droplet (CLD) boundary by a uniform layer of cytoplasmic proteins. In 3- 8% of the MFG in all species examined this cytoplasmic layer widens to include cytoplasmic organelles which are referred to as "crescents". This defines the MFG secretion as apocrine indicating a closely regulated process which minimises the loss of mammary epithelial cell (MEC) cytoplasm. The apocrine nature of the secretion might be expected since the evolution of the mammary gland is considered to be from an apocrine secreting skin gland. This short Research Reflection Review is designed to investigate the exact cytoplasmic interactions which allow such efficient lipid secretion. There are two main scenarios: one which assumes that the observed close association between CLD and GV results in the CLD being released as a consequence of sequential exocytosis of the content of the associated GV. The second assumes that the CLD and the MEC apical plasmalemma interact in some way which causes the CLD to rise out of the cytoplasm enveloped in the plasmalemma. Here I present the evidence for the two possibilities. The first scenario is favoured, but the second cannot be ruled out.

Antimicrobial susceptibility and biofilm forming ability of staphylococci from subclinical buffalo mastitis

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Turkey

Abstract

The starting objective of this research communication was to determine the prevalence of subclinical mastitis in buffalo in Turkey. We also seeked to isolate and identify staphylococci, determine their antimicrobial susceptibilities and biofilm-forming abilities as well as

investigating the presence of biofilm-related genes and microbial surface components recognizing adhesive matrix molecules. A total of 107 (66.9%) staphylococci (28 *S. aureus* and 79 coagulse-negative staphylococci, CoNS) were isolated from 160 mastitic milk samples collected from 200 lactating water buffalos. The staphylococci were especially resistant to beta-lactams except for cefoxitin but were less resistant to the other antimicrobials that were tested. Based on the Congo red agar method, 92.9% of the *S. aureus* and 70.9% of the CoNS isolates were positive for biofilm-forming ability, while all *S. aureus* and 97.5% of CoNS isolates were positive by a microtitre plate analysis. The presence of *ica*A and *ica*D genes was not always correlated with biofilm synthesis, and even in the absence of these genes, the isolates were able to synthesize biofilm.

Biofilm expression and antimicrobial resistance patterns of *Streptococcus uberis* isolated from milk samples of dairy cows in South Africa

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South Africa

Abstract

The research described in this Research Communication addresses the hypothesis that intramammary infections with *Streptococcus uberis* (*S. uberis*) are associated with biofilm formation, which limits antibiotic efficacy. This retrospective study investigated biofilm expression and antimicrobial resistance (AMR) patterns of 172 *S. uberis* infections. Isolates were recovered from milk samples of subclinical, clinical, and intramammary infection cases on 30 commercial dairy herds. We determined the presence and intensity of biofilm expression of *S. uberis* isolates *in vitro* in three somatic cell count categories to recognise their AMR patterns. An automated minimum inhibitory concentration system with a commercially available panel of 23 antimicrobial agents evaluated AMR, while biofilm determination was conducted using a microplate method. The study established that all the *S. uberis* isolates assessed expressed biofilm with the following varying degrees of intensities: 30 (17.8%) strong, 59 (34.9%) medium and 80 (47.3%) weak biofilms. The newly registered UBAC mastitis vaccine containing biofilm adhesion components may, therefore, be a viable option for proactive mastitis management under field conditions. No differences were identified between biofilm intensity and the three somatic cell count groups. Most *S. uberis* isolates indicated a high-level sensitivity to the antimicrobial agents tested. Resistances were present in 8.7%, 8.1% and 7.0% cases to rifampin, minocycline and tetracycline, respectively. Multidrug resistance was observed in 6.4%, emphasising AMR to antibiotics used in human medicine only. The low overall resistance suggests that farmers adhere to the prudent use of antimicrobials in the dairy industry.

Reducing milking frequency from 3 to 2 times daily in early lactation: effects on milk production, health and body condition.

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Abstract

The objective of this experiment was to investigate the effect of reducing early lactation milking frequency on milk yield and persistency through lactation and early lactation fat mobilization, measured by body condition score (BCS) and BHB in milk. We hypothesized that milking cows twice per day in early lactation before milking them 3 times per day for the remaining lactation would cause less fat mobilization in early lactation, a lower peak milk yield but improved persistency throughout lactation compared with milking cows 3 times per day for the entire lactation. The experiment took place on 2 commercial dairy farms in Denmark. All cows calving in a period of nine months (n=239) in their current first and later parities were randomly allocated at dry-off to 1 of 3 treatments based on expected calving date. The treatments were 1) cows milked 2 times per day for 1 week after calving, 2) cows milked 2 times per day for 4 weeks after calving and 3) cows milked 2 times per day for 7 weeks after calving. All cows were then milked 3 times per day for the remaining lactation. Milk yield peaked 3.3 and 3.6 days later and milk yield persistency improved with 18 and 19 g per day when cows were milked 2 times per day for 4 and 7 weeks, respectively, compared with milking 2 times per day for 1 week after calving. We found a significant highest milk BHB in treatment 2, but the underlying effect of milking cows 2 times per day for 4 weeks compared with 1 or 7 weeks was unclear. In conclusion, we did not confirm our hypothesis that milking cows 2 times per day compared to 3 times in early lactation would reduce fat mobilization and reduce peak yield. We did, however, find an improved milk yield persistency, which partially offset a numerical reduction in peak yield, and hence there was no significant effect of reducing early lactation milking frequency on total lactation (305 DIM) milk yield.

Probing of alpha, beta, and kappa-caseins polymorphic variants in Gangatiri cow milk with the use of polyacrylamide gel electrophoresis and HRAMS

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India

Abstract
This research communication aimed to probe the genetic polymorphisms of alpha, beta, and kappa caseins in Gangatiri cows (an indigenous Indian cattle). Detection of variants has received considerable research interest in the dairy industry and animal breeding in recent years as a source of good quality protein, but also of bioactive peptides that may be linked to health implications. The polymorphic nature of casein fractions and their association with milk production traits, composition, and quality also attracted several efforts in evaluating the allelic distribution of different casein locus as a potential dairy trait marker. Molecular techniques of polyacrylamide gel electrophoresis and high-resolution accurate mass-spectroscopy have been applied to this probe. Sequence analysis of different casein types in the cows showed the presence of four specific variants.

Protein modifications due to homogenisation and heat treatment of cow milk

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New Zealand

Abstract

This research paper aimed to locate protein modifications caused by treatment of milk and determine if the modification locations were consistent. The majority of milk for consumption is homogenised using pressure and heat, and this causes changes in the location of proteins in the milk as well as protein modifications. To investigate these proteomic changes, raw milk was pasteurised (72 °C, 15 s), then, to separate the treatment for homogenisation, heated at these different pressures and temperatures: 45 °C without no pressure applied, 45 °C with 35 Mpa, 80 °C without pressure applied and 80 °C, with 35 MPa. Proteomic analysis was done after separating the milk into three fractions: whey, casein and cream. Protein modifications in each fraction were examined and we found Maillard products as well as oxidation to be of interest. The proteins were also further identified and characterised to compare protein modification sites and differences in proteins present in the cream resulting from homogenisation and/or pasteurisation. This experiment showed that both heat and pressure during homogenisation can cause increases in protein modifications as a result of oxidation or the Maillard reaction. Total cysteine oxidation and total proline oxidation differed between treatments although this was only significantly different for cysteine. It was observed that protein modifications occurred in the same location in the protein sequence rather than in random locations which we highlighted by examining α-S1-casein, lactadherin and β-lactoglobulin.

Pasteurized milk quality in Brazil: a cross-sectional study over five years

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Abstract

This research communication delineates the quality of pasteurized cow milk sold in Brazil from 2015 to 2020. A cross-sectional study was performed gathering 1,749 samples, which were evaluated for microbiological and physicochemical parameters, including *Salmonella* spp., total and thermotolerant coliforms, freezing point, alkaline phosphatase and lactoperoxidase. The proportion of compliant and non-compliant samples was compared through the years and jurisdiction of the inspection services. Interactions between the design and response variables were assessed by log-linear analysis. Overall, a considerable non-conformity rate (12%) was found for at least one microbiological or physicochemical

parameter. Post-pasteurization contamination by coliforms was the major challenge for dairy industries. Notably, the non-compliance rate for freezing point increased during the SARS-CoV-2 pandemic. In addition, the ability to comply was linked to the type of inspection service. Thus, it is suggested that the SARS-CoV-2 pandemic is affecting the dairy industries in Brazil, and we strengthen the need for more studies monitoring the quality of milk over the years, which could assist industries and regulatory agencies to ensure the compliance of pasteurized milk.

Fibrillar aggregates in powdered milk

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Poland

Abstract

This research paper addresses the hypothesis that powdered milk may contain amyloid fibrils. Amyloids are fibrillar aggregates of proteins. Up to this time, research on the presence of amyloids in food products are scarce. To check the hypothesis we performed thioflavin T fluorescence assay, X-ray powder diffraction, atomic force microscopy and fluorescence microscopy imaging. Our preliminary results show that commercially available milks contain fibrils that have features characteristic to amyloids. The obtained results can be interpreted in two opposite ways. The presence of amyloids could be considered as a hazard due to the fact that food products may induce amyloid related diseases. On the other hand, the presence of amyloids in traditionally consumed foodstuffs could serve as proof that fibrils of food proteins do not pose a threat for consumers.

Effect of mucin on β-lactoglobulin and lactose interaction

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Abstract

Functions of mucin, as the major macromolecular component in saliva or gastric fluids, are drawing increasing attention in the context of understanding the oral processing or digestion of dairy foods at the molecular level. This study was designed to investigate the interactions between β-lactoglobulin (BLG)-lactose, mucin-lactose and BLG-lactose-mucin at the molecular level under different temperature and pH conditions using fluorescence spectroscopy in combination with scanning electron microscopes (SEM). It is the first study of its kind. There was no lactose-dependent quenching on BLG fluorophore in the range of 0-10 mM lactose concentration. On the contrary, there was a continuous increase in the

fluorescence intensity of the BLG protein when the lactose concentration increased, especially at 25°C. BLG-lactose complex became thermally unstable at 37°C and 45°C. Moreover, BLG exhibited a pH dependent conformational change and had higher fluorescence intensity at pH 3 than pH 6.8. The fluorescence result was in correspondence with SEM images where we observed lactose crystals gathering around and on the BLG molecule, but lactose molecules could not be seen in the presence of mucin. It was anticipated that mucin molecules interacted with BLG-lactose complex via electrostatic attraction and formed an extra protective layer around the BLG molecules to avoid solvent exposure.

Evaluating the antagonistic effect of *Lactobacillus acidophilus* against Shigatoxigenic and non-toxigenic *Escherichia coli* strains in bioyogurt

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Abstract

This study was conducted to determine the effect of acidophilus yoghurt (yoghurt fortified with *Lactobacillus acidophilus)* in comparison to traditional plain yoghurt (*St. thermophilus* and *L. bulgaricus* starter cultures) on the survival of three pathogenic *Escherichia coli* strains; Shigatoxigenic O157 (STx O157), non-toxigenic O157 (Non-STx O157) and shigatoxigenic non-O157 (STx O145). After six days of refrigerated storage of laboratory-manufactured yoghurt inoculated with the three strains of *E. coli* separately, all were eliminated in acidophilus yoghurt, while their survival extended in the traditional yoghurt along the storage period (17 days). Reduction percentages of the tested strains in acidophilus yoghurt were 99.93, 99.93 and 99.86%, with log reduction of 3.176, 3.176, and 2.865 cfu/g for Stx O157, Non-Stx O157, and Stx O145 *E. coli,* respectively, in comparison to 91.67, 93.33 and 93.33%, with log reduction of 1.079, 1.176 and 1.176 cfu/g in traditional yoghurt. Statistical analysis showed a significant effect of acidophilus yoghurt in reducing the count of Stx *E. coli* O157 (p=0.001), Non-Stx *E. coli* O157 ($p<0.01$) and Stx *E. coli* O145 ($p<0.01$) compared to the traditional yoghurt. These findings emphasize the potential use of acidophilus yoghurt as a biocontrol alternative method for eliminating pathogenic *E. coli*, as well as other similar applications in the dairy industry.

Evaluation of gamma-aminobutyric acid content in Portuguese cheeses with protected designation of origin status

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Portugal

Abstract

Health-conscious consumers are increasingly paying attention to healthy diets and focusing on natural bioactive compounds in foods and their effects on mental health. This opens new opportunities for the study of artisanal cheeses as biofunctional foods. In the work described in this Research Communication, the gamma-aminobutyric acid (GABA) content of seven different Portuguese cheeses produced from unpasteurized cow, ewe, and goat milk and granted with protected designation of origin (PDO) status was analysed. The PDO cheeses made from cow's milk analysed in this study were São Jorge (3, 4, 7, 12 and 24 months of maturation) and Pico cheeses. PDO cheeses made from ewe's milk were Serra da Estrela, Serpa, Nisa and Azeitão. Cheeses made from ewe and goat milk included Beira Baixa yellow cheese. The GABA content in the Azorean PDO cheeses (made from cow's milk) ranged from 1.23 to 2.64 g/kg of cheese. Higher variations in GABA content were observed in cheeses made from ewe and goat milk (0.73 - 2.31 g/kg). This study provides information on the GABA content in different Portuguese PDO cheeses and shows that hard or semi-hard ripened cheeses are a suitable matrix for GABA production by lactic acid bacteria.

In vitro characterization of anti-inflammatory activities of 3*RS*, 7*R*, 11*R*-phytanic acid

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Abstract

The aim of the research described here was to investigate the *in vitro* immunomodulatory effects of 3*RS*, 7*R*, 11*R*-phytanic acid (3*RS*-PHY) from the perspective of efficacy against autoimmune diseases. 3*RS*-PHY is a milk component with strong agonist activity at the peroxisome proliferator activated receptor (PPAR). As PPAR is a therapeutic target for several human diseases, 3*RS*-PHY intake may have possible health benefits. Recently, we chemically synthesized a preparation of 3*RS*-PHY and demonstrated that 3*RS*-PHY inhibited T-cell production of interferon (IFN)-γ. However, the overall immunomodulatory effects were not evaluated. In this study, mouse splenocytes, purified T-cells and B-cells were stimulated by mitogens and incubated with 3*RS*-PHY, followed by evaluation of cytokine and antibody production. A macrophage-like cell line J774.1 was also incubated with 3*RS*-PHY to evaluate nitric acid production. 3*RS*-PHY decreased mRNA levels not only of IFN-γ but also of interleukin (IL)-2, IL-10 and IL-17A in splenocytes and similar effects were confirmed at the protein level. In addition, 3*RS*-PHY had a direct action on T-cells with preferential inhibitory effects on Th1 and Th17 cytokines such as IFN-γ and IL-17A. Furthermore, 3*RS*-PHY suppressed antibody secretion by B-cells and nitric oxide production by J744.1 almost completely, indicating that 3*RS*-PHY is a bioactive fatty acid with anti-inflammatory properties. These findings encourage further investigations, including *in vivo* experiments, to

evaluate whether 3*RS*-PHY actually shows the potential to prevent autoimmune diseases, and provide basic information to produce milk and dairy products with an increased 3*RS*-PHY concentration.

Cross-sectional determinants of lactose-free food awareness and purchase among adult consumers

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Abstract

This research was conducted to evaluate the knowledge and purchasing behaviour of consumers regarding lactose-free foods. Study data were collected from 1882 individuals with the help of a questionnaire form. Attitudes towards animal welfare, healthy nutrition and gastrointestinal symptoms were also evaluated. The majority (86.6%) of the participants reported that they had heard of the concept of lactose-free foods before, but more than half (67.0%) did not report buying them because of their higher costs compared to regular products and lack of sufficient information. Not causing gas/bloating (32.5%) and being healthier (28.3%) were shown to be the most significant motivational factors in purchasing lactose-free foods ($p<0.05$). The factors affecting purchase of lactose-free products were education, employment status, history of having a food intolerance test and animal welfare perception together with higher scores from the gastrointestinal symptom rating scale, and attitude scale for healthy nutrition ($p<0.05$). Body mass index was not found to be an effective factor determining purcahse intention. The results provide a general overview of consumers, however, further studies on other demographic groups are needed.