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Proceedings & Abstracts

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INVITED SPEAKERS PROCEEDINGS
CONTROL OF PARATUBERCULOSIS IN SOME EUROPEAN COUNTRIES

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Abstract

The prevalence of paratuberculosis (Johne's disease) in livestock varies in European countries, as are the efforts to control the disease on national levels. Control programmes for paratuberculosis are expensive, work intensive and tedious. As the outcome of these intensive programmes often is unrewarding, participation is limited. Therefore a basic “minimal programme” for the control of paratuberculosis in cattle is suggested.

Introduction

Paratuberculosis is caused by Mycobacterium avium subspecies paratuberculosis (MAP) and occurs in ruminants worldwide (Turenne and Alexander, 2010).

Infections with MAP mostly take place at or immediately after birth, although adult cattle can become infected too (Whitlock, 1996). The infection usually takes place by oral ingestion of the organism (Whitlock, 1996), a detailed description of the disease is given by Fecteau and Whitlock (2010).

The reported prevalence of paratuberculosis in European cattle varies up to 84.7% MAP positive dairy herds in parts of Germany (Hacker et al., 2004) and almost freedom of the disease in Sweden (Holmström and Stenlund, 2005). A review of the incidence of paratuberculosis in Europe was published by Nielsen and Toft (2009).

Many different control programmes for paratuberculosis have been suggested, most of them based on “test and cull”, combined with hygienic precautions. The aim of these programmes is to identify and remove animals with a MAP-infection from the herd and to prevent further spreading of the disease within the herd.

Examples for National Control Programmes for Paratuberculosis in Europe

In Sweden, the disease is notifiable and the country has established the most rigorous control programme for paratuberculosis in cattle (Sternberg and Viske, 2003). All live cattle imported to Sweden have to be tested for MAP (Holmström and Stenlund, 2005) and if a MAP-positive herd is detected in the country, a stamping out policy is applied (Sternberg et al., 2007).

In Austria clinical paratuberculosis in cattle and other domestic ruminants also is a notifiable disease since 2006. Animals with confirmed clinical paratuberculosis have to be culled and
hygienic precautions to prevent further spreading of the disease have to be applied (Khol et al., 2007).

A new voluntary control programme, with the aim to reduce the MAP-level in milk, was implemented in the Netherlands in 2006 (Bakker, 2010). Participating farms are assigned status A, B or C, based on the results of the regularly testing of milk or serum by ELISA. (Weber and Van Shaik, 2009).

In Denmark a voluntary risk-based control programme for paratuberculosis also was established in 2006. In this programme, cows are either categorized as high-risk or a low-risk animals, based on the results of repeated milk ELIA results. High-risk animals require hygienic precautions and slaughtering of cows with repeated positive ELISA-results is recommended (Nielsen and Toft, 2011).

**Minimal Control Programme**

A basic “minimal control programme” with well-defined minimum standards should be considered as an alternative to the often rejected intensive control programmes for paratuberculosis. Such a “minimal programme” in cattle involves only 3 simple steps only: 1. Diagnostic evaluation of diarrhoea in adult cattle, culling of animals with clinical paratuberculosis. 2. Establishing of basic management measures to prevent new infections within the herd, limited to a few and realizable precautions according to the possibilities of the farm. 3. Regularly evaluation of the MAP-herd status with the focus on MAP-shedding, including the use of environmental faecal samples (Khol et al., 2009).

This “minimal control programme” might not be able to fully replace more intensive programmes for paratuberculosis in cattle, but can be implemented with reasonable costs and work load in most farms. These 3 rather simple steps can help to reduce new infections within the herd as well as between herds.

**References**


IDENTIFICATION AND PROTECTION OF PARATUBERCULOSIS-NEGATIVE CATTLE HERDS

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Abstract

Paratuberculosis is caused by Mycobacterium avium subspecies paratuberculosis (MAP) and can cause significant economic losses in affected herds. Many different control programs for MAP-positive herds have been suggested, but beside these eradication programs prevention of spreading of the disease to negative herds is crucial for the control of MAP. Environmental fecal samples could be used as a cheap and reliable method to assess the MAP-herd level and to identify and protect free herds.

Introduction

Paratuberculosis is caused by Mycobacterium avium subspecies paratuberculosis (MAP) and occurs in ruminants worldwide (Turenne and Alexander, 2010). Infections with MAP mostly take place at, or immediately after birth, although adult cattle can become infected too (Whitlock, 1996). Animals usually get infected by oral ingestion of the organism (Whitlock, 1996). A detailed description of paratuberculosis in cattle is given by Fecteau and Whitlock (2010).

The reported prevalence of paratuberculosis in European cattle varies and reaches up to 84.7% MAP positive dairy herds in parts of Germany (Hacker et al., 2004). A detailed review of the incidence of paratuberculosis was published by Nielsen and Toft (2009).

Many different control programs for paratuberculosis have been suggested, most of them focused on identification and culling (“test and cull”) of animals with MAP-infections in paratuberculosis positive herds (Bakker et al., 2010). Beside these eradication programs the prevention of spreading of the disease to negative herds and regions is essential to protect MAP-free livestock.

Environmental Fecal Samples

Environmental fecal samples were used as a cheap and reliable tool to assess the paratuberculosis herd status in large cattle farms (Lombard et al., 2006). In 2 studies the use of environmental fecal samples for assessing the MAP-herd status in small structured agricultural systems was tested (Khol et al., 2009; 2010). Both studies showed that a high percentage of cattle farms could be assigned the correct MAP-status by the use of consecutive environmental sampling. Furthermore, all farms with an acute history of paratuberculosis during, or before the study where correctly identified by environmental fecal samples (Khol et
al., 2010). Samples collected from manure channels, alleyways, manure storage sites, and around water troughs where most likely positive for MAP in farms with paratuberculosis (Khol et al., 2009; 2010). While bacteriological culture revealed more accurate results than PCR (Polymerase chain reaction) in the first study (Khol et al., 2009), both methods showed comparable results in the second investigation (Khol et al., 2010).

**Environmental Fecal Sampling in MAP-Free Herds**

It has been shown that environmental fecal samples can serve as a reliable tool to assess the paratuberculosis status of cattle herds. Samples should be taken repeatedly, for example every 6 months, to increase the sensitivity of this sampling scheme. As the testing of environmental samples for MAP is cheap and easy to perform, it could be used in a large scale to assess the paratuberculosis herd level. The results of these tests can be used to identify MAP-unsuspicious (or free) herds and to introduce trade restrictions between herds with and without MAP-positive environmental samples. Furthermore, environmental sampling could be used to assess the MAP-herd level before animals have contact with other herds (pasture, exhibitions, etc.) as a replacement or an addition to the often applied single animal testing. Thereby environmental fecal samples could become a major tool for the abatement of paratuberculosis in the future and help to identify and protect MAP-free herds.

**References**


CONTROL OF PARATUBERCULOSIS IN CATTLE HERDS IN THE
CZECH REPUBLIC

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Paratuberculosis is a disease, which causes considerable economic loses primarily in dairy cattle herds. The animals with paratuberculosis shed viable *Mycobacterium avium* subspecies *paratuberculosis* (MAP) in their milk and faeces.

In the Czech Republic the control of paratuberculosis began in the ‘90s and was based on serological (RVK) and cultivation methods. The control of disease was based on the removal of infected animals from the herd and ban on animals’ export and transfer. This approach was applied worldwide, but was found to be ineffective especially due to the low sensitivity of previously used serological methods and long time required for growth of MAP in *vitro*. Therefore a quantitative real time PCR (qPCR) based on the detection of MAP-specific targets (*F57* or *IS900*) with internal amplification control was developed at our institute in 2008. This method was subsequently implemented to the diagnostic of paratuberculosis. On selected farms, qPCR on faecal samples was processed simultaneously with solid cultivation and serum ELISA examination. Monitoring of 620 individual cows revealed that the animals can be divided on three/four groups depended on the intensity of shedding (determined by qPCR). Concordance of the qPCR, solid culture and ELISA higher than 70 % was recorded in group of high shedders.

Based on the acquired data from the farms, the logistic regression analysis of probability of ELISA positivity and absolute number of MAP determined by qPCR was performed (Figure 1). This model served for the estimation of probability that the individual will be ELISA positive by the determination of MAP number in faeces. The interpretation of results is therefore based on assumption that e.g. if there will be $10^4$ MAP organisms in a sample, the probability of this animal being ELISA positive will be 55%. The odds ratio is equal to 1.66, which is interpreted that if the number of MAP in faeces increases of 1 log, the odd that this animal will be ELISA positive will be of 1.66 times higher.

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### Table 1

<table>
<thead>
<tr>
<th>Absolute numbers of MAP organisms determined by qPCR</th>
<th>Number of positive samples by:</th>
<th>qPCR</th>
<th>Solid cultivation</th>
<th>ELISA</th>
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<tbody>
<tr>
<td>non shedders negative</td>
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<td></td>
<td></td>
<td></td>
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<tr>
<td>low/moderate shedders</td>
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<td></td>
</tr>
<tr>
<td>$10^0$</td>
<td>345*</td>
<td>4.6 %</td>
<td>11.9 %</td>
<td></td>
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<tr>
<td>$10^1$</td>
<td>13</td>
<td>0</td>
<td>23.1 %</td>
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<tr>
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<tr>
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<tr>
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<tr>
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<td>Total</td>
<td>620</td>
<td>107</td>
<td>131</td>
<td></td>
</tr>
</tbody>
</table>

* This value refers to the number of samples negative by qPCR.

### Figure 1

**ELISA vs. qPCR**

- **Probability of ELISA positivity (%)**
- **Order of magnitude qPCR**
- **Model**
- **Empirical data**
VARIABILITY OF VIRULENCE PROPERTIES OF CLOSTRIDIUM CHAUVOEI AS THE BASIS OF SELF-CONTROL OF EPIZOOTIC AND INFECTIOUS PROCESSES OF BLACK LEG

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It was found that in permanently disadvantaged areas concerning Black leg two kinds of Cl. chauvoei’s population are floating, which are different in the level of virulence.

The interaction of heterologous populations of the pathogen of infection (according to virulence) and horned cattle (according to a level of susceptibility) is the material basis of self-control of an epizootic process of Black leg

Keywords: infectious diseases, soil infection, klostrydiozes, evolution, Black leg, pathogenicity, infectious process, epizootic process, self-control of epizootic process of Black leg.

Analysis of the literature shows that there are no exhaustive answers to the most important questions about the nature, the biological significance and the evolutinal origins of the properties that are responsible for a specific, exceptional in its expression, pathogenicity of Cl. chauvoei, the ascertainment of which will give the ability to understand how this kind of microbes causes diseases, and on base of this knowledge to justify the measures of specific and general prevention.

It is considered that the agent of Black leg is the obligate parasite. That’s why, the biological feasibility of the agent’s pathogenic properties, its importance in ensuring of the continuation of species is not put into question by anyone, and the ability of this organism to lead a saprophytic mode of existence in the external environment and the ability to form spores and in this way to ensure the continuity of the species’ existence is not taken into account by anyone, in fact.

Wide and uneven spread of infection Cl. chauvoei in Ukraine and in certain regions and administrative areas tells about the presence appropirate conditions for natural circulation of the pathogen in certain regions and their absence in others.

The aim of our studies was to examine the causes of Black leg’s locality, the ecology of the pathogen of this infection, to establish ways of the existence of Cl. chauvoei in soil and their role in maintaining the epizootic trouble areas on Black leg.

By studying of 64 soil samples selected from four cattle cemeteries, the culture of Cl. chauvoei is detected in one case from a well-preserved corpse bull hip bone of bull’s carcass, which was in a depth 140–160 cm. The culture was highly virulent for guinea pigs.
The study of 78 soil samples from pastures identified 12 isolates of Cl. chauvoei—all were typical cultures by morphological and biochemical properties for Cl. chauvoei, but were not virulent.

In our previous experiments in an epizootic seat of Black leg a wide circulation of infectious agents in the environment was found-on the surface of farm fences, carts, on the surface of scalp and into feces of horned cattle.

Our studies as to the duration of preservation of Cl. chauvoei in different depths in soil of cattle cemeteries, pastures, farms, where outbreaks of Black leg were observed, indicate the presence of two kinds of Cl. chauvoei—one is highly virulent and the other (grazing) - avirulent.

We believe that the long-term preservation of viability of spores of the pathogen of Black leg and its virulence properties were observed due to the fact that the cattle cemetery, where virulent isolate of Cl. chauvoei was detected, is located in thick layers of yellow clay, that by influence of rainfall conserved tissue of the body, including the pathogen of Black leg, out of the full ichorization. In these conditions the spores of Cl. Chauvoei could not vegetate and that’s why they preserved original morphological features, biochemical and virulence properties for a long time (in our case 17 years). Thus, dead animals buried on the cattle cemeteries, which are located on large clay layers, may be examined as a potential threat of Black leg appearing during decades.

The study of the degree of the loss of pathogenicity of pasture isolates showed that being in the surface layers of soil of pastures, Cl. chauvoei significantly reduces its virulence properties, but as we found, they are not completely lost. We might think that in the surface layers of soil of pastures where these isolates were detected another mechanism of preserving agent is acting, which leads to a decrease in its virulence properties. We found this sequence when virulent museum strains of Cl. chauvoei were sifted from one environment to other for a long time and they were not carried through the body of susceptible animals (guinea pigs). A similar trend is reported by several researchers.

We assumed that the change in virulence properties of Cl. chauvoei in the abiotic environment is only possible with its reproduction when the spore form goes into a vegetative form and the last - in the spore form.

To confirm this fact we conducted an experiment on three varieties of meadow black soils. Germination of spores of Cl. chauvoei was observed under the influence of two types of black soils—dark-podzolic and gray-podzolic, and in peat soil the germination of spores did not occur. Spores of all four studied strains were vegetating. Thus, black soils of hydromorphic origin of natural grassland pastures with undamaged top layer can act as a medium of reproduction for the Black leg pathogen.

So, in the abiotic environment the population of Cl. chauvoei according to virulence is heterogeneous and consists of two kinds—one is highly virulent, but relatively not numerous, and the second is more common in certain natural geographic zones, but little or not virulent.
We found the longer period from the outbreak of Black leg, the virulence of the pathogen is the weaker. Thus, the activity of stationary epizootic seats of Black leg is the highest during the first three -four years and represents 80.5% of total recurrent outbreaks of a disease, the next – only 12.2%.

We believe that the feature of length in display of epizootic process in Black leg, despite precautions held in disadvantaged areas (disinfection, disposal of dead bodies, active immunization of susceptible livestock) is the stationarity, which, in our opinion, is primarily determined by the vegetation of spores of Cl. chauvoei in grassland soils.

Due to this mechanism some level of populations of the pathogen of infection is supported in the disadvantaged areas. Herewith at first this population is highly virulent, such as if it has just passed through the body of susceptible animals. This is evidenced by the high frequency of outbreaks in the first years after a display of infection. Reducing of the number of outbreaks in the more distant times is a reflection of the fact that every year the population becomes less virulent. However, complete loss of pathogenicity, as determined by our experiments, does not occur. This indicates that the pathogenicity of the pathogen of Black leg is a stable genotypic trait that emerged in the evolution of the relationship between populations of Cl. chauvoei and horned cattle. This is the reason to attribute this microorganism to parasites of electives, not sapronosis as previously it was thought.

On the other hand, the manifestation of infection through significant periods of time (19, 27 and 59 years) is primarily determined by a high resistance of spores of the pathogen to unfavorable environmental conditions and by special conditions of its storage that can be created in nature, as in the case described by us.

Due to the existence of two kinds of Cl. chauvoei a bad situation is formed, maintained and irregulated on infected areas. The role of each of these kinds in the “parasite – host” scheme is not the same, and therefore the value of each of them in support of epizootic process is not equivalent.

Taking into consideration the spread of the pathogen of Black leg through all continents of the world, its same antigenic structure, similar pathogenesis, clinical manifestations and pathological changes in different species of animals, we can assume that the evolution of pathogenic properties of Cl. chauvoei occurred simultaneously according to one scenario on all continents.

At the same time on particular disadvantaged territory the heterogeneity in populations of microorganisms is observed on such an important feature as the pathogenicity.

Thus, along with relatively stable species traits such as obligatory of anaerobiosis, fastidiousness to growth needs, uniformity of antigenic structure and biochemical activity we mark a variable constant of virulence, which is labile and is determined by the activity of the exotoxin’s enzymes (deoxyribonuclease, hyaluronidase, hemolysins, lethal toxin and some others).

We can affirm that permanent features of physiology and biochemistry of Cl. chauvoei were created in the early stages of evolution, while variable features, including virulence and
factors causing it – at a later stage, in the period when the population of horned cattle was formed, and the relationships of microorganisms with this type of warm-blooded animals have become permanent.
The 5 main treatment goals in calf diarrhea are to: 1) replace free water, electrolyte, and base deficits; 2) provide nutritional support; 3) facilitate repair of injured intestinal epithelium; 4) eliminate *E coli* bacteremia; 5) decrease *E coli* concentrations in the proximal small intestine.

Dehydration is due to increased loss of fluid in the stool and decreased fluid intake (inappetance or intentional milk deprivation). The latter effect is generally overlooked but plays an important role in calf diarrhea. The most accurate methods for assessing dehydration are the extent of eyeball recession into the orbit and skin tent duration in the neck region. All other methods of assessment are inferior to these 2 methods. Eye recession is measured by rolling the lower eyelid out to its normal position and measuring the distance between the cornea and lower eyelid. The recommended formula to estimate hydration status is: % dehydration = 1.7 x (eyeball recession in mm). Eyeball recession is less accurate in calves with chronic weight loss than in calves that have been on a reasonable plane of nutrition because chronic weight loss leads to loss of the periorbital fat and eye recession.

The method used for fluid administration in calves with diarrhea should be based on the presence or absence of a suckle reflex and degree of dehydration. Calves that are able to suckle and are less than 6% dehydrated (eye reccesed < 3 mm into the orbit) should be administered oral rehydration therapy (ORT); some of the fluid can be intubated using oroesophageal intubation if needed. Calves that are not able to suckle, or have a depressed palpebral reflex, or that are 8% or more dehydrated (eye reccesed 4 or more mm into the orbit) should receive intravenous fluids.

Much progress has been made over the last 30 years of the ideal ORT solution for diarrheic calves. The osmolality should range from isotonic (300 mOsm/kg) to hypertonic (700 mOsm/kg). Low osmolality fluids (300 mOsm/kg) have inadequate energy content because they have insufficient glucose. For this reason, if milk is withheld, then hypertonic oral electrolyte solutions (~600 mOsm/kg) should be administered. If milk is fed, then isotonic oral electrolyte solutions (300 mOsm/kg) should be administered, because inadequate energy content is no longer an issue. The sodium concentration should be between 90 and 130 mM/L. The ORT solution should also contain glucose and either acetate, propionate, or glycine to facilitate Na absorption and provide energy. The ORT should contain an alkalinizing agent (preferably acetate or propionate instead of bicarbonate), at a concentration range of 40 to 80 mM/L. Acetate-containing fluids can be fed with milk as acetate does not raise abomasal pH or inhibit milk clotting. Bicarbonate-containing fluids are more effective at rapidly correcting severe acidemia, but the main theoretical disadvantage of bicarbonate-containing oral fluids are that the pH of the abomasum (a natural defense mechanism) is increased. We have
recently found that a theoretical disadvantage that bicarbonate inhibits clotting of milk in the abomasum is not true, at least when low bicarbonate solutions are fed.

Lactated Ringers solution (LRS) or Acetated Ringers solutions can be used to correct mild to moderate acidosis (venous pH >7.20; base deficit >-10 mEq/L). Lactate and acetate must be metabolized before they have an alkalinizing effect. D-lactate is very slowly metabolized by ruminants (LRS contains both D and L-lactate). Bicarbonate should be used to correct severe acidemia (pH <7.20, base excess <-15 mEq/L). A revolutionary approach to fluid administration is provided by rapid intravenous administration of small volume hypertonic saline solution (4-5 ml/kg over 4-5 minutes of 7.2% NaCl solution = 2400 mOsm/L or hypertonic sodium bicarbonate = 8.4% NaHCO3 solution = 2000 mOsm/L). This protocol provides the fastest resuscitation of dehydrated calves and should be considered the treatment of choice for the rapid resuscitation of comatose diarrheic calves. Hypertonic saline solution induces a rapid increase in plasma volume, cardiac output, and mean arterial pressure. The volume expansion that occurs due to osmotically drawing water in the intracellular space and gastrointestinal lumen into the extravascular space is approximately 3 mL for every 1 mL of hypertonic saline infused.

The intravenous administration of hypertonic saline must be combined with oral administration of an isotonic alkalinizing electrolyte solution. Hypertonic saline alone does not correct acidemia due to metabolic acidosis; alkalinizing activity must therefore come from concurrent administration of an oral electrolyte solution. Alternatively, IV administration of hypertonic sodium bicarbonate solution (8.4%, 4-5 ml/kg over 4-5 minutes) can be used instead of hypertonic saline to increase blood pH in calves with metabolic acidosis. Finally, hypertonic solutions should not be administered to calves with hyponatremia, and there are some concerns with the administration of hypertonic sodium solutions to animals with chronic and severe hypernatremia.

References
Introduction

In the European Union, there is an increasing pressure from consumers and policy to reduce the use of antibiotics in farm animals. This requires good farm-management practice and optimizing the environment for the animals, including good hygiene, nutrition and properly designed vaccination programs to avoid or minimize diseases. In no other period, morbidity and mortality in calves is as high as in the first four weeks of life. However, neonates and young calves are so far not a normal target of vaccination (Thiry, 2012). The reason for this is the postulated interference with maternally derived antibodies as well as infections already in the first days of life, e.g. with rotavirus or enterotoxic E. coli. Instead, vaccination of the dam is aimed to improve the transmission of antigen specific antibodies via colostrum. This strategy is performed to protect calves against neonatal diarrhoea, but it is seldom used against pathogens causing respiratory infections (Schelcher, 2012). The emergence of Bovine Neonatal Pancytopenia (BNP) is another reason why the immunology of the newborn calf came into the focus of scientific interest in recent years. BNP is the first described alloimmune disease with increased incidence which could be directly attributed to vaccination (Bridger et al., 2011; Deutskens et al., 2011; Foucras et al., 2011; Sauter-Louis et al., 2012).

Immunology of the newborn calf

It is known from intrauterine infections with BVD virus, that the bovine fetus becomes immunocompetent during the second trimester of pregnancy. Such calves are able to eliminate the infection and are born with neutralizing BVDV-antibodies (Bolin, 1995). According to some authors, the immune system of calves is fully developed at birth, albeit immature (Cortese, 2009). Susceptibility of newborns to pathogens is not attributable to any inherent inability to mount an immune response but is caused by the fact that their immune system is unprimed (Tizard, 1992). Moreover, only particular activities of the phagocytic defense are impaired in the neonatal calf (Menge et al., 1998). The impaired phagocytosis of polymorphonuclear leukocytes (PMNL), compared to 3-9-week-old calves, is compensated by very high leukocyte numbers and a high percentage of PMNL in neonatal calves. This means, that the total capacity for microbial elimination by blood PMNL must be several fold higher than in older calves. Moreover, monocytes of newborn calves showed a higher bacterial uptake than monocytes of 3-9 week-old calves, probably also contributing to the low PMNL activities in the newborn calf (Menge et al., 1998).

Colostrum and passive immunity by maternally derived antibodies

Because the bovine placenta is of epitheliochorial type, there is no transplacental transfer of antibodies or white blood cells to the fetus. Therefore, calves are normally
agammaglobulinaemic at birth. Thus, the health of neonatal calves heavily depends on a rapid and sufficient uptake of colostral immunoglobulins and immune cells (B cells, predominantly CD8\(^+\) cells, macrophages, and neutrophils) which are functional after absorption (Riedel-Caspary and Schmidt, 1990; Menge et al., 1999; Chase et al., 2008; Taylor et al., 1994).

The primary antibody in bovine colostrum is IgG (36-77 g/L, compared to 1.0 to 1.8 g/L in milk; Murphy et al., 1999). In cattle, there is a selective transfer of IgG1 from the serum across the alveolar epithelium of the mammary gland during the last few weeks of pregnancy, which is a function of the Fc fragment in the epithelium (Murphy et al., 1999). Consequently, there is a significant decrease in the IgG1 concentration in maternal blood, with reaches their minimum around parturition (Herr, 2009).

Failure of passive transfer (FPT) is a condition in which neonates do not acquire protective serum levels of maternal antibodies. A principal component of antibody transport through the intestine (by pinocytosis) to reach the circulation of the calf is the neonatal receptor for the Fc portion of immunoglobulin. Haplotype alleles for each gene of the neonatal Fc receptor (FCGRT and b2M) have been evaluated for association with FPT. These alleles reside on different bovine chromosomes and therefore represent genetic risk associated with FPT from different haplotype blocks. With regards to FCGRT, dams with one or more copies of FCGRT haplotype 3 were shown to be 3.8 times more likely to have a calf with FPT (Laegreid et al. 2002; Clawson et al., 2004).

Besides these genetic aspects affecting passive transfer in newborn calves, many other factors may affect passive transfer, including timing of colostrum ingestion, the method and volume of colostrum administration, the immunoglobulin concentration of the colostrum, presence of the dam, and the presence of respiratory acidosis in the calf (Weaver et al., 2000). According to literature, the results regarding application method (bucket or bottle fed vs. force feeding with an esophageal feeder, are inconclusive. Of crucial importance is that calves should receive colostrum already in the first hour after birth. Total amount ingested in the first 24 hours of life should be at least 4 liters. By 6 hours after birth, only approximately 50% of the absorptive capacity remains; by 8 hours, 33%; and by 24 hours, no absorption is typically seen (Rischen, 1981; Cortese, 2009).

The colostral antibodies which are absorbed in the first hours of life are absolutely necessary to protect the calf against systemic infections. To a limited amount they can be resecreted into the gut, but infectious diarrhoea can only be prohibited if continuously high specific antibody concentrations are present in the gut lumen. In farms with diarrhoea problems, the so called “protection feeding” is recommended: Between the 3\(^{th}\) and 14\(^{th}\) day of life, normal milk (or milk replacer) is supplemented with \(\frac{1}{2}\) to 1 liter of high quality colostrum per day. Acidified colostrum (by adding formic acid or sorbic acid) is stable at room temperature in sealed containers up to 2 weeks; untreated colostrum can be kept refrigerated up to 7 days and frozen up to 1 year. Such colostrum can also be given to calves whose mothers have just recently joined the herd to administer herd specific antibodies.

**Conclusion**
Maternally antibodies transferred in colostrum are essential for protection against local and systemic infections during the first weeks of life. Most critical factors are the amount of colostrum available and the delay between birth and first suckling. Maternal immunization has become an important strategy to increase the concentration of antibodies against specific antigens. Even if field studies carried out on this have only modest evidence, it is widely accepted that vaccination of pregnant cows can improve the diarrhoea situation. This strategy is seldom used to protect the calf against respiratory infections, and the results of clinical trials are conflicting (Cortese, 2009). At least, it could be shown that pregnant cows vaccinated against Mannheimia haemolytica had increased specific serum and colostral antibody titre, and the calves of vaccinated dams had significantly higher passive antibody titres than those of non-vaccinates (Van Donkersgoed et al., 1995; Hodgins and Shewen, 1996). For vaccination of young calves against respiratory diseases, mucosal application of live vaccines seems to have advantages. Vaccines against other important pathogens, such as cryptosporidium, chlamydia and mycoplasma, should also be developed in future.

Reference list available from the author
PREVALENCE AND TYPES OF BOVINE CONGENITAL DEFECTS, DISORDERS, MALFORMATIONS AND ANOMALIES IN A DAIRY CALF POPULATION

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Congenital defects (CD) are generally considered uncommon disorders caused by genetic defects or teratogens. Prospective studies of CD in cattle populations are rare. The study objective was to establish the prevalence and types of CD in an Irish cattle population. Thirty dairy herds were monitored over 3 years (2010-2012) in a whole-herd surveillance study design. Herdowners were provided with a carcass collection service for all perinatal mortalities (0-2 days). Necropsy examinations were performed on all calves. In total, 680 calves were examined of which 164 (24%) had at least one CD. Excluding multiple CD, 23 CD were detected. Defects of the digestive system (40% of CD), multiple body systems (23) and the peritoneal cavity (13) were most commonly diagnosed. Lethal CDs were recorded in 16% of calves; diagnosed in the same body systems and order as above. The 5 most common CD recorded (% of CD and % of dead calves) were intestinal atresia (36, 8.7), multiple CD (23, 5.4), thyroid stalk (10, 2.4), omphalocoele (9, 2.1) and hepatocoele (8, 1.9). The majority of intestinal atresia was located in the jejunum (94%); the remainder in the colon. Almost three-quarters of the herds (73%) had at least one case of intestinal atresia and 17% of herds had more than 5 cases. These results demonstrate that congenital defects are more common than is generally recognized and they account for a significant proportion of perinatal mortalities. In this population intestinal atresia was unusually common. These novel findings warrant further research on this defect.
The protective role of maternal antibodies in newborn animals has been a well-known and generally accepted fact for many years. However, this protection also has a seamy side; it is commonly assumed that passively received antibodies suppress the active specific immune response in neonatal animals. During the last fifty years, this fact was confirmed many times and has become an essential part of the "canon" of practical vaccinology. According to this paradigm, it is not recommended to vaccinate newborns if specific maternal antibodies persist in the blood. This opinion persists despite the fact that opposite reports have existed for more than thirty years. They have demonstrated that at least in some cases this opinion has not been in accordance with results of controlled experiments.

There are several explanations of discrepancies between the two opposing views. Firstly, it is different experimental designs and methods for antibody detection. This review is to give a brief survey of existing knowledge related to this field, and determine unclear questions which should be further investigated.

The fact that the suppression of an active immune response does not concern cell-mediated immunity can be considered as clearly demonstrated. Most recent studies, focused on this problem, clearly demonstrated that cell-mediated immunity is not affected by the presence of any quantity of colostrum derived antibodies. This fact was revealed in two last two decades because the role of cell-mediated immunity in newborn animals was not recognised before. Similarly, most of the previous studies were restricted to the determination of antibodies, and solely of the IgG isotype. In addition, these studies often only assessed the intensity of the primary antibody response and only in one term after immunization. These facts led to a false idea that maternal colostrum-derived antibodies affect all types of immune responses.

But when we determined the dynamics of the humoral immune response of the two major immunoglobulin isotypes, we found that the antigen administered to animals with specific colostrum-derived antibodies persisting in their blood, induced only a weak and delayed IgG isotype antibody response, but antibody response linked to IgM isotype was only little affected. This observation points out the fact that priming of a specific antibody response at the level of IgM antibody is not, at least in some cases, affected by the presence of persisting colostrum derived antibodies. The second administration of antigen results in a secondary humoral immune response comparable to the response of animals without any maternal antibodies. The latter fact has been observed several times in the past, but has not so far been widely accepted in clinical practice.

Previous results clearly demonstrated that the concept of a general inhibitory effect of passively received maternal antibodies on the induction of an active immune response...
contains a number of ambiguities. However, the fact remains that the inhibition of primary immune response, particularly with regard to IgG isotype, is indisputable. The practical question therefore arises: How to vaccinate newborn animals with persistent maternal antibodies with one-dose vaccines? In these cases, we recommend to either postpone vaccination until the time when antibodies disappear or, better, to vaccinate twice.

The above described experiments brought sufficient evidence deconstructing the generally accepted idea of active inhibition of immune responses by passively received maternal antibodies, but have not clearly defined the conditions under which the newborn animals can be vaccinated regardless of persisting colostrum-derived antibodies.

The following questions still remain unclear and are worth further investigation:

The effect of using different adjuvants, which may in some cases help to overcome the inhibitory effect of maternal antibodies.

The age of the vaccinated animals, which itself, regardless of maternal antibodies, plays a role in postnatal development of the immune response.

The level of persisting maternal antibodies and their relation to the amount of the antigen administered, or their mutual ratio (antigen/antibody ratio).

The fact that maternal colostrum-derived antibodies permeate from the blood via neonatal Fc receptor in the respiratory tract mucosa during the first hours after birth we consider especially important. These antibodies are of IgG1 isotype and are protective by nature. Protection of respiratory mucosa by passive antibodies is limited to a relatively short time. However, their role in the induction of local specific immune response is unclear and they deserve a more detailed study. Very partial results on this problem are not only insufficient, but also conflicting. Nevertheless, at a time when more and more vaccines are applied by the intranasal route to induce a local immune response, answering of this question is very important. Our very partial results are also somewhat conflicting. Calves with persisting colostrum-derived antibodies in nasal secretions were protected against experimental infection with IBR virus, however, without inducing a local immune response detectable either in the blood or in nasal secretions. On the other hand, in differently arranged experiments carried out on piglets, the inhibitory effect of maternal antibodies persisting on the respiratory tract mucosa was not observed. The latter result is closely related to another of our observations which in the case of infection with *Actinobacillus pleuropneumoniae*. This experiment showed that mild infection in the presence of specific maternal antibodies, persisting in the blood and respiratory mucosa of newborn piglets, induced high level of specific immunity protecting them against subsequent experimental infection with a high infective dose. This experiment was a model of a common situation in herds in which infectious agents are still present. In those herds, the levels of antibodies in maternal colostrum are sufficiently high due to vaccination or subclinical infections of adult animals. In the case that the newborn animals with maternal antibodies in their airway mucosa become infected with a mild infectious dose, the result is a high level of protection in accordance with the above described situation.
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DAIRY HERD HEALTH MANAGEMENT: RATIONALE AND TOOLS
- VETERINARY SKILLS -

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Key words: Tools, veterinary advice, skills, dairy cattle, herd health and production management programs, quality risk management

Abstract

The herd health and production management programs, has become one important issue in the past years, principally after the constitution of the European College Bovine Health Herd Health Management and quality risk management should be a complement of the daily work of the Veterinarians, principally what we considered as Tools to Perform a herd Check using our knowledge in Herd Health Management and Production Programs. With this we can improve the profitability of the Farm as well to contribute to welfare of the dairy cattle.

1. Introduction

The animal production has derived from a mixed farming system (family farming) into production systems much more specialized and with a high professional approach. We see in the last years one strong intensification and technological which allow farmers to make profit in an economic situation with diminishing margins between production costs and gross farm income (Huirne et al, 2002; FAO, 2010; Steinfeld et al, 2010).

The intensification of animal production has led to more risks has increased risks for animal and human health. The concentration and intensification has also directly or indirectly lead to the development of vaccines, antibiotics, new technologies and herd health programmes as response (Steinfeld et al, 2010). New diseases outbreaks not seen before in Europe are common, while at the same time the risks and economic impacts are higher. The Herd Health Management preparation, under the umbrella from the ECBHM was one important step because we need as Veterinarians to provide more and more services to Farmers. We discuss in this paper the concept of using all tools that we have available when we deal with Herd Health and Production as well Quality Risk Management (QRM). We should focus our work on the Dairy farm and issues around. The main objective is to increase the farmer income, (Preventing diseases, avoid problems, identify Points of Particular Attention (POPAs) and Control of Critic Points (CCPs). (Noordhuizen et al, 2008)

2. The three dairy farm operations:

We have 3 main units in one Dairy Farm:
1. Young stock rearing

2. Cows in Production

3. Feeding and harvesting

(Figure1). They make part of three economic units, completely interrelated and balanced by management. The Dry cows is another unit and should not be neglected because is a crucial time for the dairy cows. We should avoid Body Condition Scores more than 3.5, which means that we must take attention to the fat cows (high risk of fatty liver syndrome), or too much thin cows (Negative Energy Balance - NEB). If we are able to control the cows during this period, will have less; distokia, retained placenta, endometritis, abomasal displacement, fatty liver syndrome, infertility etc.

Three economic units interrelated and balanced by management, the Dairy Farm operation has several inputs and outputs. Same inputs are Labour, Housing and the invested capital. We can considerer on this item the young stock rearing, cows in production, and the producing cows. The labour and equipment used o the farm as well to prepare the pasturing places, or to achieve harvesting feed make part of the inputs and outputs. The cows in production should produce milk which that goes to primary to the tank and after this will be transported to the Milk Factory. Some farmers can produce enough feed not only for the farm but also for sold some over production to other farms. Each of the three operational units can be characterized by five main features; Production process flow diagrams (It is part of any HACCP application and important to determine de Quality Risk Management (Nordhuizen et al, 2008). We must achieve with our client to find targets and make together some projection pictures. The methodology to achieve the targets according to our plan must be clear explained to the farmer otherwise it will be very difficult to achieve them, another point that we should not forget is to make performance figures, because they allow the team to see if we are in the correct way. Adjustments in all processes and in the targets can be made any time and if necessary. It is clear that when we look to one farm there about 400 processes and function on a dairy farm. Those processes are interactive and subject to changes several times. Some of this processes, are very simple but other ones are much more complex. We deal with living animals, and we have changes every day or at least every week. Otherwise there is also all the
feeding management, and all changes correlated with feeding, for example; silage, hey, straw, concentrate and so on. The risks of contaminations, or even no balanced feeding are always present and we should be able to avoid so far as we can. We must put all of our attention on the dairy farm processes; for example the preparation of the cow to the calving which means good farming codes of practice during the dry period and the final stage of pregnancy (this is very important) in order to have a good calving. Also important is to avoid Negative Energy Balance (NEB) after calving. This also means less stress. In other hand we should take attention to the calf, principally, we should teach the farmer how to conduct a simple Colostrum analysis using for example one Colostrometer. If needed the IGg determination in calves is also possible and will give us precise data about the level of antibodies presented on the calf. This could be the beginning of a process of improving colostrums quality, and allow us to find cows with poor antibodies content in colostrum. This means identify a problem, find solutions (it is possible to keep in fridge colostrums from old dams and rich in antibodies – attention of paratuberculosis (Boersma et al, 2010)

4. The Farm management must be seen as key point to improve the performances of the dairy farm. The farm units as we said above are in principle, which should be screening by any HHMP and then we will be in conditions to one QRM assessment. The herd health management refers to a formal approach of the farm operation so that the farmer can retain his “license to produce and to market”, while at the same time he could be certified when he meets sustainability standards (Nordhuizen et al, 2008). There is a continuous need for establishing proper relationships between dairy farmers. Farmers as well ourselves should, adjust the management practices and farm under the discipline and pressure of the dairy processor and the (consumer) market, and, at the same time, require this license to produce from the citizens (Noordhuizen et al, 2008). There are various choices possible, but at the end it is the economic returns and the relationship with society that count most. Animal health and animal welfare are of utmost importance for both farm income and approval by society (Nordhuizen et al, 2008). Animal health plays a crucial role in the dairy farm: sick cows will show a worse feed conversion and a loss of production, and normally they will have a very low Body Condition Score. To avoid this, we should be aware that feeding correlated with the management, is crucial to have cows in good body condition. For this purpose as veterinarians we must be able to check feeding. This means that we should have a good background on food and cows needs otherwise we will be in a fragile situation to have a profitable discussion with the nutritionist and with the farmer. To avoid this we should have a deep knowledge in nutrition and our skills in this matter will allow us to conduct a good and profitable discussion with our partners of business.

4. What is HHPM

HHPM is a veterinary advisory activity where clinical on a herd level practice is integrated with professional communication. Has already mentioned we need to have deep knowledge in several areas or disciplines to execute a professional HHPM. It is also important, the participation of dairy farmers in veterinary herd health and production management advisory programmes because the management quality is improved. This work should be payed as an
investment by the farmer. These two issues, herd health programmes and quality assurance programmes, and the quality risk management make it quite interesting for veterinarians to participate. Such opportunities to retain a steady income in a shifting market demand should be developed further. Given the fact that curative veterinary service demands will continue to drop, this could be compensated for by preventive (advisory and training) services (Noordhuizen et al, 2012). To avoid this problems it is highly recommended that the Veterinarians should invest much more in Technical as also in Zootechnical knowledge. The communication must be professional and using body language, compassion, perception, dominance, the right tone of voice and transmitting clearly emotions (Kleen, J.L., Rehage, J. (2008). ).This two issues are important because the Technical Veterinary and zootechnical knowledge determines 30% of the advice effect and the professional communication determines 70% of the advice effect. (Kleen, J.L., Rehage, J. (2008).

**4.1. HHPM Protocol**

For this purpose we have 3 components; Routine monitoring, Problem analysis and Prevention. It is crucial to set farm objectives, the visits must be planned and well structured, the applied methodologies should the right ones and never forget the routine monitoring – calves, environment, and record all data. The Farm Inspection Visit (FIV) is a very important part of all this process. After should doing this we can monitoring the results, setup plan of actions, evaluation and risk factors – this point’s will be the basis of any Plan of action, making the evaluation and if needed make adjustments. At least we will be in conditions to make biosecurity planes and one risk management. At each area in the production process we can develop the relevant 5 main features or parameters, and make a protocol also for each area comprising these 5 main features, to address the farm complexity. A set of such protocols compose the Herd Health and Production Management program. Do not forget that those protocols are highly farm specific which means they are unique. (Noordhuizen, et al 2008). In the next picture we present one example of monitoring Herd Health Programme Management:

<table>
<thead>
<tr>
<th>Cows- calves</th>
<th>Environment</th>
<th>Farm data</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body condition score</td>
<td>Housing conditions</td>
<td>Milk recording data</td>
</tr>
<tr>
<td>Rumen fill score</td>
<td>Barn climate factors</td>
<td>Milk quality data</td>
</tr>
<tr>
<td>Faeces consistency score</td>
<td>General hygiene standards</td>
<td>Ration composition</td>
</tr>
<tr>
<td>Faeces digestion score</td>
<td>Milking hygiene</td>
<td>Feed quality analysis results</td>
</tr>
<tr>
<td>Teat end callosity score</td>
<td>Milking method</td>
<td>Water quality analysis results</td>
</tr>
<tr>
<td>Young stock daily growth</td>
<td>Milking machine function</td>
<td>AI &amp; sire information</td>
</tr>
<tr>
<td>Clinical disease cases</td>
<td>Feeding management</td>
<td>Farm economic data</td>
</tr>
<tr>
<td>Reproductive performance</td>
<td>Colostrum management</td>
<td>Yearly performance figures</td>
</tr>
</tbody>
</table>
As mentioned above the Farm Inspection Visits are mean to conduct the monitoring (strong and weak points) in a rapid, cheap and sufficient reliable way. Normally is takes 30 to 45 minutes and it can be used alone or together with HHPM programs. The problem analysis must be discussed with the farmer/manager at the office, and together it will be setup the preventive activities. The strong and weak points are based in observation and interpretation of findings and the Veterinarian must be very well prepared to conduct this. We should look for the cows as also the calves’ performances, as well their environment and management, and never forget to check all the data concerning the farm. If we do this correctly we will be in conditions to formulate a Plan of Action based on the findings, and will allow us to put in practice actions on the short and midlong term.

The participation in veterinary herd health and production management advisory programmes (HHPM) is considered to be a prerequisite for dairy farmers to improve the Dairy Farm profitability. In HHPM the veterinarian can play the coordinator role of the different farm consultants because he visits the farms frequently, he is independent advisor not related to e.g. a feed mill, and he has knowledge and skills in animal/herd associated domains (reproduction, health, production, disease control programmes, welfare). HHPM programmes are being applied since many years in different parts of the world (Brand et al, 2001; Noordhuizen et al, 2008).

4.2 What is needed to execute HHPM?

A- Technical issue

B- Professional Communication

C- Socio-Economic issues

A. Technical Knowledge: This means that Veterinarians should have a good veterinary (clinical) background, and they should know how a dairy farm functions, they must have a deep knowledge in farm facilities. Nutrition is a key investment for the veterinary often neglected by them – they live all in the hands of other specialists for example the Nutritionists – They should make much more investment in this area. Another point is the Farm economics (normally some of us do not take care on this point), because with our work we can improve as said above the profitability of the Farm – Farm Economics. Never forget all the farm comfort issues (not important only for the cows, but also for the Consumer and Media), control the feed, the water quality and availability, the housing and scoring the lameness and participate in formation and all issues related with prevention. Veterinarians are the best partner of the Farmer to identify diseases and setup prevention planes (Biosecurity, Vaccination etc..). The management should be considered part of our preparation and link this with the control of all-on farm processes. The Veterinarians are the unique profession that works with Dairy that, are in conditions to focus on the single, the group and the herd level. Risk identification and Risk Management must be done by the Vet and well explained to the Farmer. Organization, as well planning and structures designs are part of our preparation and must be used in our work. (Cannas et al, Proceedings at the WBC Congress, Nice 2006)

B. Professional Communication:
Communication skills must be used to work in a more satisfying and much more effective way. We should be aware that nonverbal communication is the most important part, and we must take care about this. Sometimes the message that we understand may be quite different from the one intended and vice versa. All the situations may be judge differently by client or vet. Without this we cannot analyze the situations in order to act in the most appropriate way. Veterinarians must be understand that communication skills can be learned (Kleen J.L., Rehage, J. (2008).

C- Socio economic background
We need time which means that we should invest some time to do it. Empathy means that we must learn how to improve our professional communication. We also need all the time Commitment. Trust is one important issue and should be mutual. We need creditability, and something that we need time to achieve that and must be in a high level otherwise the farmer do not trust us. Important when we achieve creditability is to maintain it. If we forget this point we can lose the creditability in seconds or minutes, and we lose the trust from the farmer. The team work need a special approach and the WE level should be part of it. (Kleen J.L., Rehage, J. (2008).

4.3. Before executing a HHPM

We get more insight into management; and this means controlling the relationships with nutrition, milk yield, health status and reproductive performance. We must know how to produce with better benefits and this is part of our job. We should implement together with the farmer Hygiene plans and check and install claw health and udder health plans. We should also control the feed and feeding management and know well the Total Mix Ration (TMR). Young stock rearing is an important issue in any dairy farm, because they will be the future of the farm. Another thing to do is set goals for each area and design protocols to avoid errors. Training farm workers is compulsory in some countries already – at least 35 hours per year in the other countries if needed. We must learn more about farm economics; for example what are the costs to produce 100 liters of milk. (Viegas et al, 2006) We should invest to understand and assess the losses due to diseases. Another area of highly importance is to estimate the reproductive losses and to reduce the production costs and identify what losses are associated with quality deviations and try to make projections on the short and midlong term

5. Executing of the HHPM

The Veterinarians must discuss with the farmer about his farming goals and put some priorities and we should try to listen all of them well. We must ask the farmer to make one inventory of on-farm satisfaction, and if needed help them to conduct scores of all areas between good and poor. After this first step it is important to set HHPM priorities with the farmer. When all is done we are in conditions to do design the Monitoring protocol for the farm visits and start with it. We must put our attention to the problem area indicated by the farmer, and take care about our professional communication. The farms visits must be done every 2 or 4 weeks. (Noordhuizen et al,2008).Basic procedure: is starting discussions with the farmer, making one inventory of satisfaction and setup priorities. Than we can compose
the HHPM and discuss intensively with the farmer and finally we will be in conditions to start the HHPM. The several steps are; Monitoring, address problem areas and put some other problems in the parking lot for later work. After all this it is possible to define preventive measures and put in practice Biosecurity plans. Respect for good dairy farm codes of practice and making work instructions. As already said training the personnel and make a Quality Risk Management (QRM).

6. Concluding remarks

In conclusion, The, existing European College of Bovine Health Management (ECBHM) with about 200 internationally acknowledged veterinary specialists in the different disciplines of bovine herd health has never been invited, in a formal sense, to participate in such networks (www.ecbhm.org). Both the national veterinary associations and the European college ECBHM must take initiatives to propose and set up such networks and debating forums, for the benefit of society, the farming community and animal health and welfare.(Noordhuizen et al 2012).

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Cannas et al–Keynote lecture, WBC, Nice, 2006


EPIDEMIOLOGY OF PRODUCTION DISEASES IN DAIRY COWS ON CZECH DAIRY FARMS

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Institute of Animal Science Praha- Uhříněves

Abstract

In 10 herds of Holstein and 4 herds of Czech Siemental (Fleckvieh) dairy cows the prevalence of production (metabolic) diseases was monitored, throughout the calving-to-calving interval. It was found that health disorders occur the most frequently in the postparturient period, till 30 days in milk. Later on, in peak lactaction, the prevalence of production diseases is lower, and in late lactation, the health status of cows is generally the best. However, dry cows showed various micronutrient deficiencies. The high prevalence of metabolic diseases is associated with fertility disorders and high culling rates.

Introduction

Important prerequisites for breeding efficiency in dairy cows are adequate nutrition, animal welfare and good health status. Without fulfilling these requirements, genetic potential of an animal and the entire herd cannot be expressed and consequently, high direct and indirect economic losses occur. The proportion of culled animals is high and mortality increased. The majority of dairy cows are culled due to disease.

In high-producing dairy herds in the Czech Republic, subclinical and clinical forms of metabolic disorders and associated organ-related diseases, generally referred to as production diseases, have a high prevalence. The most marked body condition loss in cows can be observed in the transition period when the animals undergo large hormonal, metabolic and morphological changes (Goff and Horst 1997). If a cow is not well prepared for parturition, problems are encountered during calving and the postparturient period which continue till the end of the first third of lactation. Hypocalcaemia occurs and the course of puerperium is disturbed by metritis, mastitis, hepatic steatosis or ketosis. Feed intake decreases, energy balance is negative, milk production is reduced under the optimum level, fertility disorders and a number of subclinical and clinical diseases appear.

Material and methods

Analysis of performance and health was carried out in dairy cattle herds. Data was obtained from recordings of the Bohemian-Moravian Society of Breeders.

Prevalence of metabolic disorders and production diseases was monitored in selected 10 herds of Holstein dairy cows and 4 herds of Czech Pied Breed based on continuous inspection of health, production and carrying out repeated metabolic tests in the animals. Blood for
biochemical tests was collected from the tail vein by using a Hemos kit, urine by using catheters and rumen fluid through oesophageal probe. The blood, blood serum, urine and rumen fluid were analysed by standard methods using Hitachi and Cobas Mira analysers, with tests of Randox and BioVendor companies. The assessment of Ca, Mg, Zn, Cu and Mn was conducted by the AAS method and selenium by the hydride method. Samples were also assayed for T4 levels by the chemiluminiscence method on an IMULITE analyser. Volatile fatty acids and lactic acid in the rumen liquid were measured using isotachophoresis.

Results and discussion

The assessment of feed mixtures in the monitored herds did not reveal serious deficiencies, even with respect to stages of the reproductive cycle. In dry cow rations, calcium and potassium content was increased, but zinc and selenium content was insufficient. In feed rations for cows at late-gestation stage, calcium content was also increased and copper and selenium content was at subnormal level. Feed rations for cows at early lactation met requirements in all parameters, with nitrogen concentration ranging between 16.3 and 18%. Feed rations for animals at late lactation stage conformed to the valid nutritional norm.

Standardized lactation yield in the Holstein Breed was 9,137 kg, with fat and protein content being 3.78% and 3.31%, respectively, whereas the lactation yield in the Czech Siemental Breed was 6,764 kg, with fat and protein content being 4% and 3.5%, respectively.

The culling rate of cows was 34.46%. The main reasons of culling were health-related, including infertility (79.7%). Fertility results in herds in the Czech Republic are very unsatisfactory. Pregnancy rate in Holstein heifers after first AI is 59.4, in Czech Siemental heifers 59%. Pregnancy rate in Holstein cows is 34.7 and in Czech Siemental cows 43.8%.

In 10 Holstein Breed herds included in the present study, milk yield per lactation ranged between 7,856 and 12,325 and in 4 Czech Siemental herds between 5,682 and 7,368 kg.

The occurrence of production diseases in dairy cows differed between the investigated herds and was affected by a number of factors, above all by breed, nutrition and management practices. The prevalence of various diseases and metabolic disorders was influenced by the phase of reproductive cycle. The most serious health problems were observed at parturition and during the first 30 or 60 days of lactation, with higher occurrence of some diseases in dairy Holstein cows than in Czech Siemental cows. The best health status was found during late lactation and in dry cows.

Fat cow syndrome was diagnosed in cows prior to calving, being more frequent in Czech Siemental Cattle Breed (up to 60%). Phosphorus and selenium deficiencies were identified in one half of herds of each of the investigated breeds, deficiency in vitamin E and beta carotene was found in all herds. Hepatic steatosis was observed in isolated cases.

Hypocalcemia was diagnosed in cows during delivery - parturient paresis in 2.6 to 6% of multiparous cows after delivery, subclinical hypocalcaemia in 52% and excessive body fat mobilization in 82% of dairy cows. In the postpartum period (up to 30 days of lactation) high prevalence of hepatic steatosis was observed in 62%, subclinical ketosis in 26%, subclinical
hypocalcemia in 38.6%, subclinical hypophosphatemia in 12.8% and subacute ruminal acidosis (SARA) in 28%. In the subsequent period, the above mentioned metabolic disorders persisted, but their occurrence and severity was gradually decreasing. Similarly, the occurrence of copper and selenium deficiencies was decreasing. However, deficiencies in vitamin E and beta-carotene persisted.

The occurrence of the above mentioned metabolic disorders was accompanied by increased occurrence of diseases affecting body organs. Notably, it was metritis with the prevalence of 80% in some herds and mastitis with the prevalence of 18%. Laminitis and abomasal displacement were also observed. Abomasal displacement occurred primarily in Holstein cows with the incidence rates ranging between 2 and 8% in different herds. Fertility levels were very low. Pregnancy rates after first AI were in the range of 24.5 - 36.2% and 31.6 - 42.4% in Holstein and Czech Siemental herds, respectively.

Results of the present study are in accordance with our previous findings (Kováč 2009; Illek 2009, 2010, 2012, 2013) and other authors. Gaal (2007) summarized results from a number of European countries and draw attention to high culling rates, reproductive disorders and high prevalence of metabolic disorders. In the Netherlands, culling of cows ranges between 25 and 35%. In Germany, Lober (2007) reported culling of 27% cows. The culling rate in Slovakia was 26.6% and in Serbia it increased from 20 to 32%. High prevalence of fatty liver syndrome, ketosis, milk fever and subclinical hypocalcaemia appear as very serious. Fatty liver syndrome occurs in the Netherlands in 54% and in Germany in 36 to 45% of dairy cows. The prevalence of ketosis is also high, in Germany from 20 to 35 %. Hadrich (2007) even diagnosed hyperketonemia in 45 to 75% of the examined cows. Abomasal displacement which is also found in different herds from 2 to 7% (Everts 2006) is associated with the above mentioned metabolic disorders. According to Furll, prevalence of milk fever is considerable, being in the range of 2-7% in herds monitored in their study. The health status in herds included in our study is comparable. The occurrence of liver diseases, ketosis and subclinical acidosis in cows in early lactation is serious. The above mentioned metabolic disorders cause immunosuppression, thus predisposing the animals to a series of other diseases which cannot be cured without eliminating their primary causes.

Conclusions

Results following from health monitoring in Czech herds of cows show a high prevalence of metabolic and production-related disorders. The postpartum period and the period of peak milk production can be considered as especially risky provided the health status of dairy cows is unsatisfactory. Fertility disturbances and culling of high proportions of cows are associated with high prevalence of metabolic disorders. The main prerequisite for efficient breeding of dairy cows is maintaining the cows in good condition, which is closely related to adequate nutrition meeting the requirements of the animals during the calving interval, and animal welfare practices. It follows from the present study that the transition period is the most complicated and risky period for cows. It requires good animal care and complex prevention strategies.

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References


HEALTH MANAGEMENT OF EWES DURING PREGNANCY

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ABSTRACT

Objectives of health management of pregnant ewes are successful completion of pregnancy at term, birth of healthy and viable lambs, with optimal birth and potential weaning bodyweight, optimum milk production during subsequent lactation and improved management in relation to drug residues in animal products. Health management of pregnant ewes, initially, includes diagnosis of pregnancy and evaluation of the number of foetuses borne. Nutritional management of ewes depends upon the stage of lactation and aims to prevention of pregnancy toxaemia and other metabolic diseases during the peri-partum period, formation of colostrum in appropriate quantity and quality, production of lambs with normal future birth bodyweight and support of increased milk yield during the subsequent lactation. Udder management of pregnant ewes includes its clinical examination and intramammary administration of antibiotics; objectives are to cure infections which have occurred during previous lactation and prevent development of new mammary infections during the dry period. Management of abortions includes correct and timely diagnosis of causative agent, as well as strategic administrations of chemotherapeutic agents, aiming to prevent abortions in flocks with confirmed infection. During final pregnancy, health management includes administration of appropriate anthelmintic drugs, aiming to eliminate gastrointestinal helminthes and preventing built-up of parasitic burdens at pastures. Vaccinations aim to protect ewes and their offspring, especially against diseases which cause neonatal mortality. Health management also aims to prevent the main metabolic disorders of pregnant ewes. Health management of pregnant ewes is completed with application of husbandry practices before start of lambings.

INTRODUCTION

Objectives of health management of ewes during pregnancy are as follows: i) successful completion of pregnancy at term, ii) birth of healthy and viable lambs, with optimal birth and potential weaning bodyweight, iii) optimum milk production during the subsequent lactation and iv) improved management in relation to drug residues in animal products.

This account presents guidelines, which should be modified accordingly and on farm by farm basis, in order to address health issues according to locally prevailing production systems and health problems. Actually, there are differences in health management of pregnant ewes between various production systems, which are related to the production priorities in the various systems, as well as between flocks within the same production system. Based on this paper, one can adapt details for application in other production systems.
PHYSIOLOGICAL BACKGROUND OF PREGNANCY

Knowledge of physiological background of pregnancy in ewes during pregnancy is important for the overall health management of ewes during pregnancy, among that for pregnancy diagnosis and timely prevention of pathological conditions of ewes. Pre-natal life may be divided into three main periods: i) period of fertilised ovum, which ends with the initial attachment of the blastocyst, ii) embryonic period, from 12th to about 34th day, when rapid growth and differentiation occur, with initial formation of major organs and features of external body and iii) foetal period, characterized by growth and changes in the foetus. Rate of foetal growth depends primarily on feed supply and the ability of the fetus to use the feed, although other factors may also be implicated (Jainudeen and Hafez, 2000a).

Establishment of pregnancy in ewes begins at the blastocyst stage and includes pregnancy recognition signaling, conceptus implantation and placentation. Maternal recognition of pregnancy is mediated by interferon-τ, secreted by the elongating conceptus on the 12th to 13th day of pregnancy (Taverne and Noakes, 2009). Interferon-τ acts on the endometrium to inhibit development of the luteolytic mechanism by inhibiting transcription of the oestrogen receptor-α gene in the luminal and superficial ductal glandular epithelia, which prevents oestrogen-induction of oxytocin receptors and production of luteolytic prostaglandin F\(_2\)a pulses (Spencer et al., 2008). Implantation involves shedding of the zona pellucida, followed by orientation, apposition, attachment and adhesion of the blastocyst to the endometrium. As the embryo develops, the placenta is formed. This is a metabolic and endocrine organ between the developing conceptus and the uterine endometrium. The placenta is also an important source of signaling molecules capable of manipulating the pregnant ewes’ physiology to maintain an environment conducive to a successful pregnancy.

Various hormones participating in pregnancy exhibit fluctuations and interact between them depending on the stage of pregnancy. Principal hormones involved are progesterone, oestrone sulfate, pregnancy specific protein B, pregnancy-associated glucoproteins, prolactin, placental lactogen, prostaglandins and relaxin (Tamasia, 2007; Taverne and Noakes, 2009), as well as ghrelin and leptin.

Pregnancy loss can be the result of one or more of the following events during pregnancy: i) death of embryo or foetus, ii) failure of recognition of pregnancy (interferon-τ insufficiency), iii) inappropriate uterine environment (defects of endometrium or hormonal pattern), iv) placental deficiency or v) decreased progesterone concentration. Hence, abnormal termination of pregnancy may occur at various stages: i) before recognition of pregnancy, which cannot be distinguished from fertilisation failure (early embryonic death), in which case the length of the oestrus cycle would not be affected and the ewe returns to oestrus, as if she had never conceived, ii) after recognition of pregnancy, but before foetal formation (late embryonic death), in which case the ewe returns to oestrus after a longer than normal period of time or iii) during the foetal stage (foetal death), leading to mummification or abortion (section 6) (Jainudeen and Hafez, 2000b). Mortality is more common during the embryonic period. Although fertilisation rates may reach up to 90% to 95%, risk of embryonic death can be as high 20 to 30%. In some cases, particularly in multiple pregnancies, early embryonic deaths may be a process of eliminating unfit genotypes, which would not have survived the entire
length of intrauterine life anyway. In contrast, foetal deaths should always be considered as an abnormal process. Cumulatively, microbial abortions account for a significant proportion of embryonic or foetal deaths in ewes.

In general, factors related to embryonic deaths in ewes may be as follows: i) genetics, ii) hormones, iii) nutrition, iv) age of ewes, v) endometrium, vi) number of ovulations, vii) lactation, viii) infection or ix) environmental factors. Improving knowledge regarding biological and immunological interactions between the pregnant ewes and its embryo(s), as well as the identification of corresponding genes, will provide the potential to identify causes of embryonic deaths, thus improving embryo survival in early pregnancy.

During late pregnancy, increased energy demands of the rapidly developing foetus(es), in combination with hormonal interactions, have an impact on lipid and carbohydrate metabolism of the pregnant animal, putting it at risk to developing pregnancy toxaemia. The disease is the result of negative energy balance, usually secondary to increased energy requirements of the foetuses, and is usually observed in multiparous ewes on limited-energy diets (Andrews, 1997; Brozos et al., 2011). Various nutritional, metabolic, genetic, physiologic, environmental, health and/or management factors and interactions can lead to incomplete glucose synthesis and mobilisation, as well as to fatty acid accumulation to liver, which hampers normal function of the organ with end results increased oxidation of fatty acids and increased production of ketone bodies (Andrews, 1997; Sargison, 2007).

Formation of twin embryos takes place soon after conception, hence they develop in a different environment than singles throughout their embryonic life, from conception until birth, and thereafter. Twin foetuses develop a between-them competition for nutrients, are enveloped by a smaller placenta than single foetuses and live in a restricted physical space. After birth, twins compete for milk supply and attention from the same dam. Twinning results in decreased foetal growth during late pregnancy and it has been concluded that twin foetuses, in late pregnancy, can have features similar to those of single foetuses borne by ewes experiencing undernutrition (Rumball et al., 2008, Fleming, 2012). These authors showed that effects of twinning on late-pregnancy ovine foetuses have many similarities to the effects of undernutrition, which causes a documented decrease in future birth bodyweight of foetuses. Nevertheless, differences between the two situations also exist: in twin pregnancies, the pregnant animal’s metabolic and endocrine environment, the foetal and placental growth and the function of the foetal glucose-insulin axis differ than those in single pregnancies.

PREGNANCY DIAGNOSIS IN EWES

Early and accurate pregnancy diagnosis is important, in order to apply proper and successful management of pregnant ewes. Clearly, the result of pregnancy diagnosis could be only one of the following: i) the ewe is pregnant, ii) the ewe is not pregnant or iii) the findings cannot support an accurate diagnosis, hence the ewe needs to be re-examined. Subsequently to diagnosing pregnancy in a flock/group of ewes, the animals can be grouped and managed according to stage of pregnancy and number of foetuses carried. Moreover, correct pregnancy diagnosis can lead to avoidance of culling pregnant animals, as well as to successful induction of lambing if necessary. Moreover, ewes found not-pregnant can be appropriately managed or else culled.
Several methods are currently in use for pregnancy diagnosis in sheep. These include management approaches, clinical methods, biochemical tests, imaging methods, vaginal biopsy and laparoscopy (Noakes, 2003; Tamasia, 2007; Taverne and Noakes, 2009). In practice, the methods most frequently employed are the evaluation of non-return of mated ewes to oestrus, the transabdominal palpation, the udder examination and the ultrasonographic examination. Currently, diagnostic kits are also commercially available, which can detect concentrations of various hormones, and can be employed for pregnancy diagnosis of ewes. Ultimately, choice of the method for use depends on the operator’s skills, the stage of pregnancy and the availability of facilities and equipment. In any case, the method chosen should satisfy as many as possible of the following criteria: sensitivity, specificity, accuracy, speed, safety, low cost.

The method for early pregnancy diagnosis that meets most of the above criteria is real-time B-mode ultrasonographic examination (Scott and Sargison, 2010; Scott, 2012). This is advantageous, despite the requirement for expensive equipment, because the operator may diagnose viability, growth, size, number, age and sex of foetus(es), as well as placental development. There are two different approaches for ultrasonographic examination of pregnant ewes: transcutaneous and transrectal ultrasonography. Choice of the technique depends on the stage of pregnancy, the available ultrasound probe, the working conditions and the experience of the operator.

Ultrasonographic images characteristic of pregnancy are multiple anechoic luminal sections of the uterus, presence of anechoic fluid and/or ‘C’- or ‘O’-shaped placentomes, as well as presence of embryo(s) or foetus(es) showing heartbeats. Using transrectal examination, embryonic vesicles can be identified 12 to 20 days after mating, while embryo(s) can be recognised 16 to 25 days after mating. Placentomes and the amnion can be seen from about the 25th day of pregnancy. Following transabdominal scanning, pregnancies can be diagnosed on 17th to 30th day of pregnancy, while transrectal examination is more accurate until the 35th day. Between 35th to 70th day, both methods appear to be equally accurate. Taking into account cases of embryonic deaths occurring early in pregnancy, an accurate diagnosis can be performed after the 40th to 50th day of pregnancy (95%-99%). The transcutaneous approach is preferable during the second half of pregnancy. Early estimation of the number of foetuses can be done until the 40th day by using the transrectal technique; the most reliable estimation is possible between 45th to 100th day by using transcutaneous examination, with an accuracy of 90% to 95% (Kähn, 2004; Meinecke-Tillmann and Meinecke, 2007). Estimation of foetal age, when date of mating is not known, can be performed by monitoring embryonic or foetal parameters, with the best period for an accurate diagnosis being between 40th to 80th day of pregnancy.

NUTRITIONAL MANAGEMENT OF PREGNANT EWES

Before the mating period, ewes can be given additional feed with increased energy content (‘flushing’). At the beginning of the mating period, animals should have a body-condition score of ‘3’ to ‘3½’ on the five-point scale. ‘Flushing’ consists of administration of an additional quantity of concentrate feed mixture of 200 to 400 g daily, on top of the ration administered to cover maintenance requirements of the animals. In dairy sheep, it may also be
necessary to sustain lactation, which is progressively approaching its end. Administration of the increased energy feeding should commence at least 35 days before start of the mating period; that interval is equivalent to the length of two full oestrous cycles of sheep. In animals with appropriate body-condition score, this increased energy feeding aims to producing increased ovulation numbers, leading to increased number of lambs born per ewe. In animals in lower body condition score, there is a benefit to other reproductive parameters, but no significant improvement in fecundity of ewes (Heasman et al., 1998).

The same increased energy diet should also be provided during the initial stages of the mating season. This has two benefits: i) animals that had not conceived during the first oestrous cycle of the season, are maintained in a good body condition and ii) animals that had conceived, have a lower risk of early embryonic death (Parr et al., 1982).

During the first 100 days of pregnancy, there is a slow foetal growth (Blanchart and Sauvant, 1974; Economides, 1981). During the second month of pregnancy, when foetal attachment has been established and placental growth has been completed, foetus(es) can acquire up to 15% to 25% of their future birth bodyweight. During that period, fat deposition in pregnant animals should be limited, but, conversely, excessive bodyweight loss can put ewes at risk to develop pregnancy toxaemia. Protein requirements of animals can be fully covered if proportion of total protein content in ration at that stage of pregnancy is over 8% per dry matter content of feeds provided (Cannas, 2004). At that point, body-condition score of animals should be ‘2’ to 2½’ on the five point scale.

Nutrients are also required for the growth of the placenta, the uterus, the mammary glands and the body reserves of pregnant animals. The placenta has a key role in ensuring that the foetus(es) would receive optimal supplies of nutrients, but overfeeding during mid-pregnancy can lead to restriction in placental size, hence to suboptimal future birth bodyweight of lamb(s) (McDonald et al., 2001).

Restricted nutrition of ewes in early- to mid-pregnancy can inhibit optimal placental growth (McGrab et al., 1992). Few studies have determined whether nutritionally-mediated alterations in placental growth can extend to term or what the impact is on conformation of the newborn. However, where such measurements have been carried out, results obtained at term have been confounded by a compensatory increase in ewes’ nutrition compared with controls over the second half of pregnancy. Clarke et al. (1998) have demonstrated that feeding of ewes bearing a single foetus at close to half their energy requirements between 30th to 80th day of pregnancy significantly reduced mean weight of individual placentomes and total weight of the foetal component of the placenta. Moreover, effects of pregnant ewes’ nutrient restriction were not confined to the placenta, but, also, had a significant influence on their plasma thyroid hormone concentrations, despite no change in plasma concentrations of glucose and free fatty acids (Clarke et al., 1998).

During the final stage of pregnancy, the ovine foetus(es) can develop rapidly, to acquire up to 75% to 80% of their future birth bodyweight. Hence, energy requirements of pregnant ewes increase progressively, as end of pregnancy is approaching. In the final month of pregnancy, protein requirements of pregnant animals also increase, due to foetal requirements and the need to prepare colostrum in the mammary glands. Hence, it is important that pregnant ewes
are given protein-rich, easily-digestible forages; proportion of total protein content of ration can be increased up to 10% (per dry matter content). Ideally, body-condition score of ewes should be ‘2½’ to ‘3½’ one month before lambing and ‘2’ to ‘2½’ at lambing (Cannas et al., 2004).

Stress increases during late pregnancy, especially for ewes bearing multiple foetuses (Economides and Louca, 1981). Foetal energy requirements in the final stage of pregnancy have been calculated as 1.5 MJ ME per kg foetus per day (Hill Farming Research Organisation, 1979). This means that a 50 kg ewe carrying twins would have an energy requirement of about 2.5 to 3 times that of a non-pregnant ewe. However, it is not proposed to cover in full these high requirements, as provision of 25% less energy than above requirements would reduce future birth bodyweight of lambs by only 10%, which is acceptable on biological and economic considerations (Hill Farming Research Organisation, 1979). However, administration of low levels of energy during late pregnancy would lead to pregnancy toxaemia (Morand-Fehr and Sauvant, 1978; Economides and Louca, 1981). On the other hand, high energy of feeding throughout pregnancy can also lead to the same disorder (Orskov, 1982), as well as to dystocia due to excessive foetomaternal disproportion.

Nutritional management during that period aims specifically to i) prevention of pregnancy toxaemia and other metabolic diseases during the peri-partum period (e.g., hypocalcaemia, hypomagnesaemia), ii) formation of colostrum in appropriate quantity and quality, iii) production of lambs with normal future birth bodyweight and iv) support of increased milk yield during the subsequent lactation.

During early- to mid-pregnancy, grazing animals do not have additional requirements to cover. Frequently, these may be covered only through the consumption of a small quantity of concentrates coupled to their grazing intake. Consequently, their body-condition score is usually satisfactory, if grazing possibilities are available. However, in winter months, when herbage availability in pastures is decreased, additional feed should be provided to the animals (McDonald et al., 2010).

In intensive production systems, feeding is formulated based on nutrient requirements of animals and nutritive value of feeds provided, in order to meet daily requirements of the animals. Under these conditions, feed intake of sheep can be measured easily and appropriate feeding can be carried out; for example, ewes may be fed according to the number of foetuses that they carry or according to their body-condition score or age. Bocquier et al. (1995) have considered that allocation of ewes into groups would be the best way for correct nutritional management of pregnant ewes in intensive systems.

In dairy ewes, it has been documented that milk yield of animals could be improved by feeding increased energy quantity during the final stage of pregnancy. This leads to building-up body reserves, which subsequently, during lactation, would be expressed as increased milk production (Morand-Fehr and Sauvant, 1980; Skjevdal, 1982). To note, that under such conditions protein supplementation would also be required.

UDDER HEALTH MANAGEMENT
The principal component of mammary physiology during pregnancy involves the process of involution at the end of lactation preceding pregnancy. The aims of effective udder health management at that period are i) to cure infections which have occurred during the previous lactation and ii) to prevent development of new mammary infections during the dry period. In lactating dairy ewes, Staphylococci and Trueperella (Arcanobacterium) pyogenes are the most frequent causative agents of mastitis in ewes during the dry-period (Saratsis et al., 1998; Spanu et al., 2011).

It is now well documented that incidence risk of mammary infections increases during the first two weeks of the dry-period (Barkema et al., 1998; Saratsis et al., 1998). This increased infection risk has been associated with compromised mammary defences during that period; neutrophil dysfunction, depression of lymphocyte function and decreased cytokine production have been suggested as possible reasons for the increased infection risk of dry mammary glands. Subclinical infection which has occurred during the previous lactation, may also lead to recrudescence of clinical disease at the end of lactation and the start of the dry-period, as a result of the reduced mammary defences of the animal.

The first step to effective udder health management of pregnant ewes is the thorough clinical examination of the mammary glands at the end of lactation; this will help to identify ewes, which should be culled. The udder of all ewes in the flock should be examined by palpation, whilst the animals are run through a race. If mammary abnormalities are suspected, animals can be individually examined; mammary secretion samples may also be collected for microbiological examination (Mavrogianni et al., 2005). Diffuse hardness, abscesses and nodules in the mammary glands are the most common clinical findings during that examination (Saratsis et al., 1998).

The following categories of ewes should be considered for culling based on the results of this examination: i) animals chronically affected, ii) animals which had showed relapsing mastitis during the previous lactation, iii) animals with one mammary gland permanently damaged and iv) animals which had not fully responded to mastitis treatment during the preceding lactation. The benefits of culling such animals include: i) decrease of veterinary expenses for mastitis control in the flock, ii) elimination of sources of potential infection for other animals in the flock and iii) decrease of bulk somatic cell counts in the subsequent lactation (Mavrogianni et al., 2011). Moreover, lambs (especially from multiple births) from ewes with extensive mammary lesions do not thrive as well as those from healthy dams and may require additional feeding, which adds to labour expenses in the flock.

Subsequently, intramammary administration of antimicrobial agents should be carried out to animals, which will be maintained into the subsequent lactation. Administration of intramammary antimicrobial agents at the end of lactation is effective in reducing post-partum mammary infection risk. Although a variety of products is licenced for administration in ewes, ideally, the product for administration should be selected on the results of susceptibility testing of bacteria (Mavrogianni et al., 2011) isolated from the mammary secretion samples of ewes individually examined, as detailed above.
Finally, the results of recent work do not support a hypothesis that the procedure followed for udder drying-off could affect the risk of infection of the mammary glands during the dry period and the immediately post-partum period (Petridis et al., 2011).

MANAGEMENT OF ABORTIONS

In sheep, abortion refers to the expulsion of a foetus before the 135th day of pregnancy, after which a newborn lamb is usually able to survive. Abortion is a significant problem in pregnant ewes and a major source of financial losses. An incidence risk of abortion cases <5% during a season is considered acceptable, whilst a risk <2% is excellent; a repeated, year after year, abortion rate between 2% to 5% suggests presence of endemic diseases in the flock (Menzies, 2007a).

The great majority of abortion cases is of microbial aetiology; various non-infectious agents (e.g., stressors, pharmaceutical agents, nutritional factors) have also been identified to cause abortion in ewes, but are not highly prevalent (Edmondson et al., 2002). Generally, the most common causes of abortion in sheep are the following: *Brucella melitensis*, *Campylobacter fetus subsp. fetus*, *Chlamydophila abortus*, *Coxiella burnetti*, *Toxoplasma gondii* and *Border Disease Virus*; of lesser importance can be various other bacteria (e.g., *Salmonella* spp., *Mycoplasma* spp., *Leptospira* spp.) and viruses. Hence, early control of cases of abortion can prevent a subsequent abortion ‘storm’. In such cases, one faces two problems: i) to establish an accurate diagnosis of the causative agent and ii) to control the disease. Causative diagnosis cannot be achieved in all cases and, frequently, several cases remain undiagnosed (Kirkbride, 1993).

As a general rule, it is recommended that the first 10 cases of abortions occurring in a lambing season should be investigated in detail, whilst, thereafter, only 10% of cases should be investigated (Mavrogianni and Brozos, 2008).

Diagnosis of abortion cases should start with a detailed history, as closely pertaining to the abortion problem, since it is not rare for a stillbirth case to be perceived and reported as abortion. Certain points can offer valuable information and facilitate diagnosis; for example, the incidence of abortion may be higher in certain age groups or in different stages of pregnancy. Other important factors for a successful causative diagnosis include the number of foetuses borne per ewe, possible introduction of replacement animals in the farm, details of vaccinations and nutrition, exposure to toxic plants or drugs and abnormal findings in the animal(s) that had aborted, before, during or after abortion (Menzies, 2011).

Ewes that had aborted, must be isolated and abortion material should be destroyed by burning. Healthy animals must be examined first, whilst securing no contact with animals that had aborted. Farmers of infected flocks should be advised to wear protective clothing that is changed before entering the area of healthy pregnant ewes. If animals that had aborted, need to be milked or mated during the subsequent breeding season, they should remain isolated for at least three weeks. If it is decided to cull them, they should be sent directly to the slaughterhouse, thus avoiding any contact with healthy animals.
Confirmation of the cause of abortion in a flock can be achieved only after appropriate laboratory examinations. Generally, the following material are useful and will support a diagnosis of the causative agent of the problem: i) at least two cotyledons with intercotyledonary placenta from a ewe that had aborted, ii) fresh foetuses from ewes that had aborted, iii) whole blood or serum samples from ewes that had aborted and/or iv) vaginal discharge samples from ewes that had aborted. Material for microbiological examination should be freshly collected and individually packed in sterile containers. Material for histopathologic examination can be fixed in 10% formalin solution. In all cases, deep freezing should be avoided.

In general, abortive disease can be well-controlled by means of vaccination, which is best taking place before start of the breeding season. The possibility of vaccinating pregnant ewes has also been considered (Menzies, 2012).

In flocks with a confirmed abortive disease problem, where vaccination had not been carried out, there may be a need for a strategic administration of chemotherapeutic agents. For example, in flocks with prevailing *C. abortus* problem, starting after the 80th day of pregnancy administration of long-acting oxytetracycline at a dose rate of 20 mg per kg bodyweight and repeated every three weeks, will significantly decrease the abortion risk, although there is some disagreement whether multiple treatments are justified (Menzies, 2011). Although oxytetracycline is effective in the control of an outbreak caused by *C. burnetii*, the literature regarding value of strategic administration of the drug in infected, non-vaccinated flocks is conflicting (Sahin et al., 2008; Angelakis and Raoult, 2010). In unvaccinated flocks, where toxoplasmosis has been diagnosed, control can be instituted through feeding of prophylactic medications during pregnancy; administration of monensin at a dose rate of 16.8 mg per ewe daily has been shown to reduce losses (Buxton et al., 1988), whilst administration of decoquinate at a rate of 2 mg per kg bodyweight daily for the final 14 weeks of pregnancy can also lead to decreasing abortion risk caused by *T. gondii* (Buxton et al., 1996).

MANAGEMENT OF ENDPARASITIC INFECTIONS

A critical timing for anthelmintic treatment in ewes is before mating, i.e. beginning of summer in the para-Mediterranean countries. Any class of anthelmintics may be used at this point. Experimental results have shown that removal of helminthes before mating can improve reproductive efficiency. Garcia-Perez et al. (2002) observed that highest conception rates (>95%) were achieved among animals treated before mating and considered that to be the result of increased fecundity during the first 30 days after ram introduction into the flock; moreover, it seemed that pre-mating treatment had a positive effect on lamb bodyweight per ewe mated. Hence, control of helminthes during the pre-mating period can be considered to have an effect similar to ‘flushing’ (Venter and Greyling, 1994). In case the treatment has to be carried out during or soon after the mating period, the choice of anthelmintics to be used includes levamisole, macrocyclic lactones, amino-acetonitrile derivatives or spiroindoles, if necessary combined with a trematocide. Obviously, at that period, benzimidazoles have to be excluded as a possible choice, as these compounds have documented embryotoxic properties, leading to teratogenesis (Braun, 1997); hence, there is a scope for avoiding their use during
the peri-conception and early pregnancy period. The same precaution should also be maintained in flocks where rams are kept permanently with ewes, as some animals may be already at early pregnancy at the time of benzimidazole administration.

Antiparasitic treatment of pregnant ewes during the last month of pregnancy can be incorporated in flock health management, as animals are particularly susceptible to the effects of parasitism at that time (Coop and Jackson, 2000). The interaction of parasitism and reproductive activity in ewes is expressed with the periparturient rise in worm egg output, which has been attributed to a loss of resistance associated with late pregnancy and early lactation (Dunsmore, 1965). The findings of Houdijk et al. (2003) imply that, in lactating ewes, milk production takes a priority over reduction of helminth numbers. Therefore, treatment of pregnant ewes at this stage eliminates helminthes (thus, increasing production potential for ewes) and prevents the built-up of parasitic burdens at pastures (thus, reducing infection of lambs during the post-parturient period). Effective treatment of pregnant ewes, using drugs with persistent activity, can also result in increased future birth bodyweight of their lambs and increased milk production (up to 44%) during the subsequent lactation (Fthenakis et al. 2005).

The choice of anthelmintics to be used at that time, includes benzimidazoles and macrocyclic lactones, amino-acetonitrile derivatives or spiro-indoles, potentially combined with a trematocide drug. Certainly, choice of the drug to be prescribed, should also take into consideration the rotation of classes of anthelmintics, in order to minimise selection for resistance. Levamisole has been reported to potentially cause abortion if administered in late pregnancy (Braun, 1997), hence, it should better be avoided.

VACCINATIONS

Clostridial infections constitute a significant health problem in sheep and anti-clostridial vaccinations, most often carried out during the last month of pregnancy, will contribute significantly to protection of vaccinated ewes, as well as of their offspring (Lewis, 2011). Internationally, various anti-clostridial vaccines are licenced for use in sheep. Some of these are multivalent, whilst others are composed of few clostridial antigens, mainly Clostridium perfringens. Multivalent vaccines aim to protect, except for Cl. perfringens infections, also against Cl. septicum, Cl. novyi (types B and D), Cl. tetani, Cl. chauvoei and Cl. sordelli (Lewis, 2011). Vaccination of pregnant ewes against clostridiosis should be carried out 20 to 40 days before the expected start of the lambing season, in order to achieve increased antibody titres in the colostrum of the vaccinated animals for protection of the newborn lambs, as the main immune mechanism in clostridial infections is humoral immunity. To note that ewes that had not lambed within three months after the last anti-clostridial vaccination should be revaccinated (Lewis, 2011).

Infections from Manheimia haemolytica can cause mortality in newborn lambs; therefore, vaccination of pregnant ewes can be useful, especially as many risk factors for the disease cannot be avoided. Vaccines incorporating iron-regulated proteins produced by the causative organisms are best for use and they confer cross-protection against all isolates of M. haemolytica and Bibersteinia trehalosi (Scott, 2011). Vaccination for protection against M. haemolytica infection can be combined with anti-clostridial vaccinations (Donachie, 2009;
Sargison, 2009). Nevertheless, in order to maximise protection afforded by vaccination, predisposing factors of the disease need also to be removed.

Contagious ecthyma (‘orf’), caused by *Orf virus*, can also be prevented by vaccination of pregnant ewes, especially in flocks with increased incidence risk of the disease in lambs or increased mortality rate of lambs due to secondary bacterial infections. In general, field studies have confirmed that annual vaccination of pregnant ewes one month before lambing by using an attenuated vaccine, affords adequate protection of newborn lambs (McKeever and Reid, 1987), although vaccination failures may also occur.

Contagious agalactia, caused by *Mycoplasma agalactiae* and other mycoplasmas is a common health problem in the para-Mediterranean countries (De la Fe et al., 2005). Excellent control of the disease can be achieved by combining hygiene, biosecurity measures and effective vaccinations. Vaccination should be carried out during pregnancy, in order to confer immunity to the female animals, as well as to their lambs, as humoral immunity is important for contagious agalactia (Chessa et al., 2009).

Pregnant ewes cannot be vaccinated with attenuated vaccines, especially vaccines against abortifacient agents, as this would lead to abortion. Therefore, only inactivated vaccines may be used. Inactivated whole cell vaccines are available to protect against *C. abortus* infections and may be administered to pregnant ewes, in view of an outbreak of the infection.

**PREVENTION OF METABOLIC DISORDERS**

Pregnancy toxaemia is a metabolic disorder of pregnant ewes, caused by an abnormal metabolism of carbohydrates and fats, which occurs at the final stage of pregnancy. In order to improve prevention of the disease, especially in farms with prevailing risk factors of the disease, ewes should ideally be allocated into early and late lambing groups on the basis of mating records. Moreover, animals may be subdivided into ewes bearing one foetus and ewes bearing multiple foetuses (Sargison, 2007), as established in pregnancy diagnosis. This allows improved nutritional management during the last stage of pregnancy, as well as administration of anthelmintic treatments and vaccinations at the most appropriate time-point.

Animals at risk of developing pregnancy toxaemia can be identified by measuring β-hydroxybutyrate concentration in their blood during the last six to four weeks of pregnancy, especially as rapid measurement techniques are now available (Panousis et al., 2012). If the number of foetuses carried has not been identified, the value of 0.8 mmol per L should be considered to distinguish animals at risk to develop the disorder. Otherwise, if the number of foetuses carried had been determined, then β-hydroxybutyrate concentration should be measured only in the blood of animals carrying multiple foetuses; in this case, the cut-off value to be used for identifying animals at risk is 1.1 mmol per L (Sargison, 2007; Braun et al., 2010). If financial or labour constraints preclude examination of all animals as above, then examination of around 20% of animals will still provide a valuable overview of the flock situation. If blood measurement is not feasible, semi-quantitive measurement in urine by using dipsticks can be advocated, but results should be considered cautiously. Animals found to have increased concentration of β-hydroxybutyrate in blood or urine should be separated from other animals and monitored closely.
Hypocalcaemia is an acute or subacute pathological condition, which occurs more often shortly before or after parturition. A detailed account of preventive measures for the disease has recently been published by Brozos et al. (2011). In general, an effective preventive strategy against the disease should include i) control of body condition, ii) regulation of calcium, magnesium (and phosphorus content in the feed, iii) monitoring of cation/anion balance in the feed and iv) regular monitoring of calcium concentration in the animals’ blood.

HUSBANDRY PRACTICES BEFORE THE START OF THE LAMBING SEASON

Good management practices require that personnel in charge of peri-parturient ewes are aware of the animals’ needs and have the appropriate knowledge to control and manage potential problems. Moreover, personnel (even the most experienced) should update their skills on latest techniques and management practices.

Allocation of ewes into groups (as discussed above) will reduce workload in the farm, considering the variability in length of pregnancy in ewes. In general, one should focus on avoiding long, spread-out lambing periods, because this results in increased stress for ewes and more workload for the farm staff. Moreover, ewes in different lambing groups should lamb in separate areas of the shed, in order to minimise the risk for build-up of pathogens in the farm environment (Menzies, 2007b).

Regardless of the production system applied in the flock, stress to parturient ewes should be minimal, but still with maximum supervision. Availability of temporary individual lambing pens provides many advantages, such as significant reduction of newborn lamb losses and facilitation of animal marking. Specific measures should be taken to ensure that any purchased replacement ewes will lamb in a separate barn, away from the home flock, in order to avoid potential cross infections (Gelasakis et al., 2010). A similar management should apply to ewe-lambs lambing for the first time. Areas with pregnant ewes should allow a floor space of \( \geq 2 \text{ m}^2 \) per ewe, whereas clean straw bedding should be always available (Gelasakis et al., 2010). Shearing around the perineal area of parturient ewes and dirt removal from the udder and teats of ewes contributes to quick access of lambs to the mammary glands of their dams, as well as decreased incidence risk of post-partum genital infections of ewes and cryptosporidiosis of newborn lambs. Finally, an ‘isolation’ area should be available, where ewes with obstetrical problems or post-partum disorders may be isolated.

CONCLUDING REMARKS

This paper has provided, for the first time, a detailed account of health management of pregnant ewes, which is a multi-faceted very significant task for successful management of the flock within the annual production cycle. The paper provides guidelines for health management of pregnant ewes around the world. Fine-tuning of the procedures will need to be carried out in individual flocks, after taking into account their specific requirements.

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MAINTAINING OPTIMAL RUMEN FUNCTION AND HEALTH IN LACTATING DAIRY COWS

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The ruminant animal is unique in that in order to achieve optimum feed intake and efficiency its ruminal environment must be maintained within certain physiological limits. These limits are required to be maintained to provide a favorable symbiotic relationship between the ruminant host and the ruminal microorganisms. The ruminant should provide the microorganisms an environment of limited oxygen, relatively neutral pH, constant temperature, relatively continuous influx of water and organic matter, constant removal or neutralization of waste products and indigestible matter, and mean retention time greater than microbial generation time. The feeding systems necessary in modern dairy cattle production have made it increasingly difficult to provide a ruminal environment that stays within all of these narrow constraints. The enormous energy requirements of high producing dairy cattle require dairy farmers to feed cattle rations of increasing dry matter intakes and levels of concentrate feeds.

The rumen is a fermentation vat that can hold 40 to 60 gallons of material and is the site of microbial activity. An estimated 150 billion microorganisms per teaspoon are present in its contents. They consist of bacteria, protozoa, and fungi. Bacteria require a warm, moist, oxygen-free environment for optimum growth. This type of environment is naturally maintained in the rumen with a temperature range of 100 to 108°F. If cows are fed a proper balance of forages and grain, the pH should range between 5.8 and 6.4, which allows the growth of many species of bacteria.

The rumen through its strong musculature allows mixing and churning of digesta. The movement of the rumen mixes the contents, promoting turnover and accessibility of the coarser forage particles for regurgitation, cud chewing, size reduction, and microbial digestion. Fine forage particles, dense concentrate particles, and materials which have become hydrated tend to congregate near the bottom. Particles tend to move out from the rumen as they are reduced in size through cud chewing and microbial action. The microbes also pass from the rumen for possible digestion in the lower gastrointestinal tract.

The structuring and composition of rumen contents is influenced by diet. Since the dairy cow consumes such a varied selection of feedstuffs and feed particle sizes, rumen contents do not have a uniform composition, and as a result there is stratification of feed particles. Long-hay diets produce contents with a large, less dense, floating layer beneath the gas dome with relatively liquid contents and suspended fiber beneath. Denser material sinks to the bottom of the rumen. The floating mat is composed of the more recently ingested forage. In diets where forage Adequate forage particle size is necessary to maintain cow and ruminal health through buffering ruminal pH, but varying FPS also has many other effects. Further discussion of forage particle size as it related to rumen function and maintaining adequate cow health is the
subject of much research. Using a proper measuring device is critical to attaining correct values of forage particle size. Defining requirements for physically effective fiber requirements for maintaining proper rumen function is critical. Much research has been devoted to doing this in the past 20 years.
DIAGNOSTIC PROCEDURES TO MEASURE RUMEN ACID BASE STATUS IN DAIRY COWS

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The rumen acid-base status is mainly determined by the synthesis of volatile fatty acids (VFA) and therefore by the diet but it is also influenced by various other processes like emptying rate, buffering, dilution and absorption. The feed and water intake also results in alterations of rumen pH which, however, normally remains within the physiological range. Although numerous papers on rumen metabolism and rumen acid base status have been published there is no general agreement on physiological ranges or thresholds for pathological rumen pH values.

In older literature the acute rumen acidosis was considered the more important condition, however in recent papers the subacute or subclinical rumen acidosis (SARA) is described as the pathological condition of higher importance. Some author state that caused by the high amounts of concentrates in the diet during peak lactation it is impossible to avoid SARA but that it might be mitigated using several management tools. A number of pathological conditions are known for being associated to SARA, e.g. low butter fat concentration, ruminitis-liver abscess-complex, alimentary ketonuria, or laminitis.

There are two general ways to assess rumen acid base status; firstly the direct measurement in rumen fluid and secondarily the indirect estimation of the rumen acid base status using parameters assessing urine or blood acid base status, biochemical parameters, parameters in milk, feeding analysis and clinical signs.

The most commonly used direct method is the pH measurement using pH measurement paper or electronic devices. An alternative method is the titration of the rumen fluid to measure the acid or base excess in the fluid. The advantage of the titration is that this method does not only include the measurement of hydrogen ions but also the effect of weak acids and bases (e.g. bicarbonat, VFA,) and ions, which act as rumen buffers.

The direct measurement of rumen acid base status faces some problems. The rumen fluid must be obtained which can be done using a stomach tube or by rumenocentesis. The insertion of the stomach tube is stressful for the animal and requires two or three people. Additionally, the rumen fluid might be contaminated with saliva which can be avoided using proper equipment and technique. The stomach tube must be long enough to reach the ventral liquid phase in the rumen. The first portion (300 -500 ml) has to be discarded; the second portion can be used for analysis. On the other hand rumenocentesis is not risk free since injuries and hematomas of the rumen wall occur regularly which may results in peritonitis. Additionally, rumenocentesis often requires sedation resulting in subsequent milk withdrawal. In herd diagnostic the rumenocentesis is faster and more efficient; however, if large volumes are required the stomach tube method is the method of choice.
There are studies comparing the measurements obtained using a stomach tube or rumenocentesis. The pH varied considerably between the locations and methods; the associations between the methods and location were weak. It was found that the pH decreased from cranial to caudal by 0.3 to 0.5 units. The rumenocentesis and the stomach tube had a high specificity diagnosing SARA (96%); however, the sensitivity was higher in rumenocentesis (67%) in comparison to the stomach tube method (25%). The average differences between stomach tube method and rumenocentesis varied between 0.26 and 0.76. These differences cannot be explained by saliva contamination as it was described that saliva influences the buffer capacity but the pH was influenced only after massive saliva contamination (> 20%). One conclusion of these studies is that independently which method is used the measurement is only valid for the certain location at the certain time; a single measurement is always a snapshot with limited diagnostic value for the whole rumen. An alternative method which solves a number of the current problems is the usage of submersible rumen device which measure the pH and temperature continuously. The continuous measurement offers a great potential in herd diagnosis and management. The technology spreads increasingly in the practice but currently the high costs and the limited battery life prohibit a wider use in field. It seems reasonable to believe that with decreasing costs and improved technology the method has the potential to improve our ability to evaluate feeding and rumen acid base status.

Caused by the risks and problems associated with the methods obtaining the rumen fluid it has been suggested that indirect measurements can be used as an estimate for rumen acid base status. This concept is based on the experimental findings that rumen acidosis resulted in systemic metabolic acidosis. The systemic metabolic acidosis in blood, tissue and urine could also be found when cattle were fed a low fiber and high concentrate diet. The Net Acid Base Excretion (NABE) which is a parameter characterizing the acid base status in urine was described as the parameter having the highest diagnostic value. In comparison to a control group NABE and urine pH of cows with SARA were significantly decreased under field conditions. Therefore NABE was considered as useful parameter to diagnose SARA. However, the diagnostic value of urine pH and NABE has been assessed and found to be insufficient for single animals but it is possibly an useful tool in herd diagnostics.

Several publications describe low butterfat concentrations, decreased faecal-pH and altered biochemical parameters like fructosamine, betahydroxybutyrate, calcium, lactate, and LDH-activity. In cows suffering from SARA the diagnostic value of the indirect parameters was low caused by various other influences and wide variations of the parameters.

In summary, currently the diagnosis of rumen acid base status requires the sampling of rumen fluid. The definition of SARA is still under discussion, it is very likely that the introduction of intraruminal automatic measurement devices will add knowledge and better understanding. In herd diagnosis indirect measures may provide useful information.
GUIDELINE FOR EVALUATING REPRODUCTIVE PERFORMANCE IN DAIRY CATTLE

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The successful genetic selection for higher milk production in Holstein cows has nearly doubled the average milk production since 1960, to over 11,000 kg/year. Over the same time period, there has been a dramatic decline in the reproductive performance of dairy cows. The average number of days open (interval from calving to conception) and the numbers of services per conception have increased substantially. In order to decrease the longer lactations and the number of cows culled for reproductive reasons it is very important to improve our reproductive management practices (Silva, 2003). Achievement of optimum herd reproductive performance (calving interval of 12 or 13 months with the first calf born at 24 months of age) requires concentrated management activities especially during the first 100 days following calving. Early postpartum breeding of dairy cows results in more calves, and higher milk production per lactation (Britt, 1975). Poor reproductive performance can reduce the number of calves born and milk production and may increase the cost of therapy and semen.

The following management activities are needed to evaluate during the early postpartum period to reach or approach the optimal reproductive performance: careful surveillance and assistance at calving, early diagnosis and treatment of post parturient metabolic and uterine diseases, accurate detection of oestrus, correct timing of insemination and early pregnancy diagnosis.

The most important breeding objective is to reduce the number of calving assistance required. This is even more important, since calving assistance in itself may result in a shift of the calf’s acid-base balance and perinatal mortality. Therefore, the main emphasis should be paid on the prevention of asphyxia of calves at birth, since instruments suitable for a reliable clearing of respiratory passages and maintenance of this state as well as artificial respiration of calves under practical conditions are not yet widely used (Szenci, 2003). While it is not possible to eliminate dystocia, adequate management of heifers during their development (adequate feeding, selection of a sire with a negative expected progeny difference for birth weight) and close observation of cows and heifers during calving are essential for reducing calf losses (Szenci, 2003), birth injuries and infection of the reproductive tract during assistance, which is more likely to occur in cows with dystocia. Dystocia can negatively affect the subsequent pregnancy rate of dams (Radostits et al., 1994).

The rapid increase in energy requirements at the onset of lactation results in negative energy balance (NEBAL) that begins a few days before calving and usually reaches its most negative level (nadir) about 2 to 3 weeks later and used to extend 10 to 12 weeks until the beginning of the usual breeding period (Bell, 1995; Butler, 1989). The NEBAL that develops
spontaneously in dairy cows represents a physiological state of undernutrition. The severity and duration of the NEBAL is primarily related to differences in dry matter intake and its rate of increase during early lactation. Calving in moderate condition (3-3.5) and maintaining feed intake during the periparturient transition period are key factors to reducing NEBAL and avoiding metabolic disorders (milk fever, acidosis, ketosis and fat cow syndrome) that are deleterious to performance. Cows should be challenge-fed during dry-off period and early lactation to prevent the incidence of different metabolic disorders of the puerperal period. These disorders can increase the incidence of reproductive diseases and reduce the reproductive performance. Prevention is more preferable to treatment and requires close attention to nutrition and management (Radostits et al., 1994). The maintenance of good body condition at calving and the provision of a high-density energy diet that does not produce a fatty liver in early lactation are also very important to minimize the detrimental effect of the NEBAL on the return of oestrous cycle after calving.

Cows having dystocia, retained foetal membranes, metabolic disorders (hypocalcaemia, ketosis), or twins are more likely to contract uterine infections than cows calving normally. Postpartum infection of the uterus has long been considered to have a deleterious effect on subsequent fertility (Erbet et al., 1981). Thus, nutrition, population density, management of calving (decrease of stress), sanitation during calving, early diagnosis and treatment of uterine infections are of great importance. It is recommended to treat clinical metritis/endometritis as early and as intensively as possible in order to keep the conception interval short (Drillich et al., 2001). Recently fresh cow programs have been developed based on monitoring cow temperatures each morning for the first 10 days after calving which allow for early treatment (Belschner & Saltman, 2000). However, the results of routine treatment of cows with intrauterine antibiotics (oxytetracycline, ampicillin & cloxacillin), intrauterine antiseptic chemicals (iodine solution, polyvinilpyrrolidone-iodine solution), systemic antibiotics (ampicillin, ceftiofur), or hormones (oxytocin, PGF2a) after calving are greatly variable or their use(s) is/are controversial (Youngquist & Dawn Shore, 1997; Heuwieser et al., 2010).

Oestrus detection is one of the major contributors to low fertility results in the field (Reimers et al., 1985). Simple observation of the herd for 30 minutes in the morning before and after milking, at midday, and late in the evening is recommended to determine oestrus accurately in the field. The use of traditional aids like pressure sensitive mount detectors, tailhead markings, and/or detector animals or recently developed aids like pedometry, electrical resistance measurements, and/or electronic pressure-sensitive mount detectors may improve the accuracy of oestrus detection. The combined use of monitoring of oestrous behaviour and one or more oestrus detection aids may improve its efficiency (Senger, 1994).

Since the chances for pregnancy after insemination are much higher when ovulation occurs within the survival time of the semen (Thimberger, 1948; Saacke, 2008) therefore it is important to inseminate the cow about 12 hours after oestrus detection. Due to the fact that 7 to 22% of cows showing oestrus inseminated during the luteal phase and these cows do not conceive or it leads to abortion if they had been pregnant (Appleyard & Cook, 1975; McCaughey & Cooper, 1980; Nebel et al., 1987) therefore determination of correct timing of insemination is of great importance (Van Eerdenburg et al., 2002). When oestrus detection on the farm is good AI at the observed oestrus and PGF2a treatment if needed may be
recommended, when oestrus detection is poor Ov-Synch or Pre-Synch and a fixed time AI are recommended (Mialot et al., 1999).

Accurate early detection of pregnant and non-pregnant cows as well as pregnancy loss mortality plays a key role in the achievement of an optimal calving to conception interval. One of the most recent techniques for diagnosis of early pregnancy in cattle on the farm is B-mode ultrasonography. Under field conditions, acceptable results may be achieved with ultrasonography (using 5 or 7.5 MHz transducers) from Day 26 (heifer) to Day 28 (cow) after AI (Szenci, 2007). The reliability of the test greatly depends on the frequency of the transducer used, the skill of the surgeon (Badtram et al., 1991), the criterion used for a positive pregnancy diagnosis (Szenci et al., 1998) and the position of the uterus in the pelvic inlet (Szenci et al., 1995).
TO PRODUCE AND REPRODUCE, A MAJOR CHALLENGE FOR MODERN HIGH YIELDING DAIRY COWS

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Introduction

At present, dairy cows are able to produce vast amounts of milk mainly due to significant genetic improvements, combined with enhanced nutritional management. A prerequisite for good lactation performance during the cow’s life span is producing a calf at regular intervals. Therefore reproductive efficiency is a worldwide concern in the dairy industry as it influences average daily milk production, average days in milk, number of calves born per year and the generation interval. Many studies have reported a worrisome decrease in the reproductive performance of dairy cows and this problem seems to affect all countries housing high yielding dairy herds. For example, in Belgium the average calving interval increased from 390 to 417 days during the last 12 years. The number of AI per conception rose from 1.43 to 1.80 in the same period and the pregnancy rate at first AI dropped from 56 to 43%.

Negative energy balance

As negative energy balance (NEB) seems to be the ever-returning enemy of good fertility in high yielding dairy herds, the basic strategy to reduce the reproductive decline should definitely focus on keeping the NEB under control. While in modern dairy cows genetic progress in terms of milk yield has outstripped that for intake capacity, a certain degree of NEB is inevitable, certainly in early lactation. The extent of the NEB (both in depth as well as in duration) varies with the magnitude and rate of increase of milk yield compared to energy intake, and can be exacerbated if metabolic conditions, disease, housing or management practices impair nutrient intake. Hence, management strategies by which the effect of a NEB can be limited must be targeted towards increasing nutrient intakes, especially energy. Immediately after calving, the paramount goal should be to maximize energy intake without disturbing rumen fermentation. The first aim of the management of a recently calved dairy cow is to optimize her general health status. Only when optimal health – including an excellent appetite – is achieved, the focus can shift towards achieving an optimal production level. In practice, in their enthusiasm to reach top production levels, farmers often forget this basic principle. To optimize energy intake, the intake of high quality forages in early lactation should be maximized. Once this has been achieved, the energy density of the ration may be increased by gradually raising the amount of concentrates. Generally, under European circumstances, the maximum amount of concentrates given should not exceed 12 kg (9 kg in first lactation animals) and should only be reached at three weeks after calving. Increasing the amount of concentrates too fast may disturb ruminal fermentation, which in turn may give rise to ruminal acidosis and an increased incidence of left abomasal displacement. Currently a lot
of research is going on to study the effect of changing the proportion of the different ingredients of the ration. Increasing the amount of fat to maximize the energy content of the ration and hence the energy intake by the animal, or increasing the amount of glucogenic substances to temper the steep insulin decrease around the moment of calving are excellent illustrations of such measures. For example, the ratio of n6 : n3 fatty acids provided in the diet can influence the synthesis of the 2-series of prostaglandins, which are desirable after calving to speed up uterine involution, but undesirable after insemination as they can contribute to the breakdown of the corpus luteum of pregnancy. Hence, the practical implementation of our current knowledge needs to be a better timing of the introduction of rumen protected fats into the diet in accordance with the reproductive stage of the cow. Although primary results seem to be promising, these studies need further confirmation before definite conclusions can be drawn and the results transferred into practical recommendations. Nutrient or dry matter intake is highly dependent on a lot of factors related both to the cow and to the environment. Among the cow factors, the general health status and body condition score are of major importance. Hence, transition cow programs should focus on maximizing general health and appetite and striving for the ideal body condition score of 3.5 (on 5-point scale) at calving. Aiming for optimal general health includes trimming of the claws at drying off, optimizing rumen health and avoiding metabolic and infectious diseases around calving. Besides this, the veterinarian should provide his herds with a specifically designed standard operating procedure for detecting ill cows as soon as possible and treating them properly. Furthermore, efforts must be made to remove any environmental restrictions to feed intake, as the environment must be conducive to high intake. Cows need time and space for undisturbed feeding and rumination. There is clear evidence now that the design of food passages, barriers, troughs for water supply and cow traffic within the building definitely affect the intake cows will achieve. Intake can vary widely between individuals in a herd with a lot of competition for feeding space. Especially the intake of heifers is easily restricted by competition with older cows. The provision of adequate feeding space reduces this kind of competition largely. The grouping of cows and social behavior also have their implications. A lot of attention should be paid to this point because during the transition period cows are transferred several times from one group to another. Each transfer or relocation implies another challenge for the cows as it brings them in contact with a new group and a new ration. All the energy that is spent in establishing a new social hierarchy is no longer available to produce or reproduce. At the same time, each change in the ration causes a serious drop in dry matter intake and should therefore also be avoided. Although veterinary practitioners are currently not the only advisors on modern dairy herds, they have the advantage that they can use their “clinical eyes” to interpret what is happening in the herd. Besides the use of herd production data which are usually readily available, the use of clinical scoring systems has been proven to provide the veterinarian with an extra tool to evaluate the health status of the animals in relation to their production level. Hence, these scoring systems should be used to evaluate the management system used on the herds at a regular time interval. Furthermore, today’s dairy cows may face a wide variety of environmental stressors. These may include overcrowding, infectious challenges, poor ventilation, poor footing or other forms of chronic or even acute pain, uncomfortable stables, rough handling, and frequent relocation in another group. Most of these stressors affect fertility and should therefore be avoided. Although stress is difficult to define and to show to the herd manager, a lack in cow comfort compromising
the cows’ health and fertility should be noticed and discussed during the regular herd health visits. While top managers have it at their finger-tips and do not need a lot of explanation to adapt their herd to the needs of their modern top producers, others definitely need to be confronted with some eye-openers.

Conclusion

In view of the complex nature of fertility, it is not surprising to find that ideal fertility criteria are extremely difficult to achieve. When infections are involved in a subfertility problem, this can be due either to specific (e.g. BVDV) or non-specific genital infections. The former often strike a whole herd, causing abortions and repeat breeding. The latter are opportunists of unsanitary conditions during calving, dystocia and abnormal puerperium. They often take an insidious course. It is generally agreed, however, that the main negative influence on the fertility of a dairy herd stems not as much from specific or non-specific infections, but rather from the effects of a host of other factors. These factors seldom exert their effects individually but rather interact together, making it difficult to analyze infertility in a given herd. For example, the advancement of animal husbandry practices has increased both herd size and production, but man hours per cow have dwindled. The direct result of this decrease is that less time remains for detecting heats, instituting hygienic measures and trimming claws. Thus the final fertility status of a dairy herd is the result of interactions of a whole range of factors from environmental conditions such as season, herd size and age composition, to pure managerial factors such as breeding policy, nutrition and estrus detection. Breeding efficiency depends almost totally on whether or not the farmer is able to skilfully cope with these factors in his herd. By way of conclusion, subfertility has been proven to be a multifactorial disease and the optimization of herd fertility often requires the optimization of several interfering managerial factors. There is almost never a single solution. Although poor fertility is becoming more and more common in our top dairy herds, there is a wide variation between herds and sometimes between years within the same herd. This latter fact illustrates that the dairy herd acts as a dynamic structure and may need specific adaptations, depending on the specific situations the herd actually has to face. Fertility of a dairy herd is thus a relative phenomenon, expressing what the cows have been able to achieve in the face of a host of interacting factors. To avoid a deterioration of fertility below the accepted standards, the advice given to the farmer should enable him to optimally manage his herd under the given environmental and management conditions. Such advice can best be given by paying regular visits to the farmer (Herd Health and Fertility Control Program) so as to impress upon him the relevant factors of management. Hence, the follow-up of the reproductive performance of a dairy herd should be continuous and not only be restricted to the curative interventions when things are really going wrong. The cornerstone to improving the reproductive performance of lactating dairy cattle also involves the understanding of the biochemical and physiological principles controlling reproductive and lactational processes. The challenge is to integrate this knowledge into nutritional management, production medicine and reproductive management procedures, taking into account the specific obstacles each individual herd has to face, for the purpose of optimizing the fertility of the herd. In the absence of such a holistic approach, the response to traditional veterinary therapies and herd health programs may increasingly diminish.
PUERPERAL DISORDERS IN DAIRY CATTLE – FROM DIAGNOSIS TO SUCCESSFUL THERAPY

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The puerperium may be defined as the period of return to normal reproductive function after pregnancy which includes uterine involution and resumption of ovarian and oestrous activities. The common puerperal disorders in dairy cows are uterine infections (metritis, endometritis, cervicitis, pyometra), ovarian ovulatory dysfunctions (anovulation, cystic ovarian disorders) and dysoestrus (anoestrus, suboestrus, irregular oestrus). The prevalence of these problems varies depending on the herd management systems and predisposing risk factors; malnutrition (pre and post-partum negative energy balance, micronutrient and macronutrient imbalances), parturient problems (dystocia, stillbirth, abortion, premature calving, retained fetal membranes) and disease stressors (mastitis, lameness).

This paper summarises the findings from the latest scientific publications internationally on the diagnosis and therapy of puerperal disorders in dairy cattle.

Diagnosis of puerperal disorders

Uterine infections

Puerperal metritis is diagnosed clinically by the presence of systemic illness/toxaemia (reduced milk yield, dullness, inappetence, pyrexia >39.5°C), an enlarged uterus and fetid vulvar discharge generally within a week postcalving. Reduced feeding time and DMI and hyperhaptoglobinaemia precede metritis and may be used as diagnostic predictors. Clinical endometritis/purulent vaginal discharge is diagnosed by the presence of a purulent vaginal discharge more than three weeks postcalving which may be detected by manual or Metricecheck device removal of vaginal contents, vaginoscopy, or visual observation. Subclinical endometritis(SCE)/cytological endometritis is diagnosed by endometrial cytology (EC) where the presence of >~5% neutrophils in a cytobrush smear collected 5-7 weeks postpartum in the absence of clinical signs indicates infection. Adjunct diagnostic modalities include the urinary strip leukocyte esterase test, ultrasonography (US), and endometrial biopsy. Cervicitis is diagnosed by cervical smear cytology (>~5% neutrophils) approximately four weeks postcalving. Pyometra is diagnosed clinically from the detection of a pus-filled enlarged uterus by transrectal palpation (TP) or US in the presence of a persistent corpus luteum (CL) and a closed cervix more than three weeks postcalving.

Ovulatory dysfunctions

Anovulation is diagnosed clinically by the absence of a CL on TP or US or luteal phase blood (>1ng/ml) or milk (>5ng/ml) progesterone concentrations in serial examinations after approximately 45 days postcalving. Cystic ovarian disorder (COD) is diagnosed clinically by
the presence of a persistent (≥7 days) anovulatory structure identified by TP or US with an internal diameter of >20mm and a wall thickness of >3mm (luteinized cyst) or ≤3mm (follicular cyst) in the absence of a CL and in the presence of anoestrus or irregular cyclicity.

**Dysoestrus**

*Anoestrus* is diagnosed clinically by absence of standing oestrus up to approximately 45 days postcalving in the absence of a CL on repeated examinations 10 to 14 days apart (true anovular anoestrus). *Suboestrus* is diagnosed clinically by reduced expression of behavioural oestrous following ovulation, particularly reduced number of mounts received, diagnosable by visual observation or oestrus mount detectors. *Irregular oestrous* is diagnosed clinically by short (<18 days) or prolonged (>24 days) inter-oestrous intervals.

**Successful therapy of puerperal disorders**

**Uterine infections**

Current evidence from multiple studies indicates that *puerperal metritis* can be successfully treated (including metaphylactically in cows at-risk) with systemic antibiotic, e.g. ceftiofur, penicillin, ampicillin for 3-5 days with supportive therapies for toxaemia as indicated, e.g. fluid therapy, NSAIDs. Treatment of at-risk cows or cows with *clinical endometritis* (CE) with intrauterine (IU) antibiotic, e.g. cephapirin, at one month postcalving has successfully improved conception performance. A recent meta-analysis concluded that PGF2α did not improve reproductive performance in cows diagnosed with CE. Currently there are too few studies published on the treatment (systemic or IU antibiotics, PGF2α or NSAID) of *subclinical endometritis* to draw conclusions about successful therapies. There are no studies on the therapy of cervicitis. *Pyometra* can be successfully treated with one normal dose of PGF2α or if refractory to therapy, two higher dose injections. Efficacy of therapy of uterine disorders depends on the severity of disease, presence of a CL and DIM at treatment. Several novel alternative uterine therapies are undergoing evaluation include IU ozone, proteolytic enzyme, dextrose, neutraceutical, bacteriophage, probiotic and homeopathic therapies.

**Ovulatory dysfunctions**

Treatment of *anovulation* results in earlier conception than no treatment and this can be successfully achieved with a progesterone-supplemented Oysynch (GnRH-PGF2α-GnRH)/timed AI (TAI) programme. While variations of anovular cow treatment regimes are constantly being developed, their success is related to the critical predisposing factors for anovulation, e.g. cow BCS and parity. Current research indicates that successful treatment of *follicular cysts* can best be achieved with a modified Ovsynch/TAI programme. However, where cyst differentiation is not practised, GnRH alone can be successfully be used to treat either follicular or luteal cysts. The high self-cure rate (~50%) during the puerperium must also be recognised.

**Dysoestrus**

Progesterone-supplemented Oysynch/timed AI (TAI) programmes can successfully be used to treat *anoestrus*, *suboestrus* and *irregular oestrus* without the need for oestrous detection.
While veterinary therapies can address these puerperal disorders in the short-term, in the long-term the farmer needs to modify the risk factors predisposing cows to these disorders and improve reproductive management.

References


ASPIRATION OF FOLLICULAR FLUID FOR ACID-BASE BALANCE ANALYSIS IN CATTLE

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Follicular fluid sampling has been performed for many years in the evaluation of the oocyte microenvironment. The most frequently applied method of In Vivo (live testing) aspiration is the use of ultrasound guided transvaginal follicular aspiration (TVFA). However, recovery of follicular fluid for acid-base balance and gas analysis (ABB) from live animals has scarcely been described, perhaps due to the technical difficulties of follicular fluid sampling. Many preanalytical errors in routinely performed blood sampling for ABB analysis caused by the underfilling of syringes, syringe material or the conditions sample storage have been described. Because similar errors should be expected during and after collection of follicular fluid, requirements for anaerobic follicular fluid collection have been reported. In our previous study we have also proven preanalytical changes in O₂, CO₂ and pH values when we used modified ovum pick-up equipment for collection of fluid from ovarian cysts. The samples of cystic fluid were not taken under strictly anaerobic conditions using this equipment and we concluded that the requirements for the development of a new device for follicular fluid collection was necessary.

In this study we evaluated a new TVFA device for ABB analysis of follicular fluid (ABB device) in cattle. A convex ultrasound transducer was placed in a probe holder of the usual shape. A metal guide tube placed in the upper part of the holder was used for insertion and allowed for free movement of the syringe holder. A place for the attachment of an aspiration syringe was located at the front part of the syringe holder. A connecting rod controlling a syringe piston went through the holder’s corpus to its rear. The examiner manipulated the connecting rod through a guide ring that enables the management of aspiration from the outside. Standard disposable needles were used for the aspiration. As a control we used a modified original commercial ovum pick-up set. An original 17-gauge, 60-cm long aspiration needle was connected with a shortened Cook aspiration line (with a length of 20 cm). The total volume of the aspiration needle and aspiration line was 0.9 ml. Aspirations were performed manually into 2 ml syringes.

In order to create a homogenous biological fluid reservoir, enabling the collection of a series of samples to test the ABB device under laboratory conditions, we collected urine from cows in rubber balloons. We collected 15 triads of samples (each triad with two punctures) from reservoirs while the first sample of a triad was taken using the ABB set (ND sample). Then two samples were taken by one puncture using the modified original commercial OPU set from: sample from aerobic phase (AE) with air present in tubing at the start of sampling and a sample from a subsequent anaerobic phase of sampling (AN). Values determined in the second sample from the triad (AE) varied from the values in ND and AN samples (pH 7.685 vs. 7.704 vs. 7.692, pCO₂ 11.13 vs. 10.3 vs. 10.85, pO₂ 6.87 vs. 8.67 vs. 7.02).
Subsequently we performed TVFA in 25 cows bearing ovarian cysts with a diameter of at least 3 cm, 13 aspirations to plastic aspiration syringe (experiment P) and 12 aspirations to glass aspiration syringe (experiment G). We found significantly higher pH in AE in comparison to AN (7.357 vs. 7.348), lower pCO$_2$ (6.85) and higher O$_2$ (14.12) in samples AE in comparison to samples ND and AN (pCO$_2$ 7.36, 7.30; O$_2$ 9.95, 10.63 respectively) in cystic fluid in experiment P. We found significantly higher pH (7.4), lower pCO$_2$ (5.98) and higher pO$_2$ (12.35) in samples AE in comparison to samples ND and AN (pH 7.386, 7.385; pCO$_2$ 6.39, 6.35 and O$_2$ 10.56, 10.65, respectively) in cystic fluid in experiment G.

We concluded that ABB testing was affected by air present in the tubing during AE in comparison to AN and ND. Those preanalytical changes can be prevented by the use of an ABB device because the results obtained with an ABB device were not different from AN samples. Furthermore we confirmed preanalytical changes in the ABB parameters in cystic fluid following storage in plastic aspiration syringes.

We evaluated the feasibility of the ABB device for aspiration of ovarian follicles. Collection of at least 0.6 ml of follicular fluid without visible signs of the presence of blood (pink or red colour of the aspirated fluid) was considered an aspiration success.

The success of single aspiration of individual follicles was 96.2% (50 of 52), individual follicles after FSH stimulation 92.6% (25 of 27), the success of consecutive aspirations of multiple follicles after FSH stimulation was 89.7% (61 of 68).

According to the results of ABB tests from ovarian cysts and the aspiration success from ovarian follicles we concluded that an ABB device equipped with a glass aspiration syringe is the most suitable for use in the sampling of follicular fluid for the analysis of ABB.

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ROLE OF COAGULASE-NEGATIVE STAPHYLOCOCCI IN BOVINE UDDER HEALTH: AN UPDATE

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Introduction

Despite substantial research efforts, mastitis [inflammation of the mammary gland as a result of intramammary infection (IMI)] remains the most costly disease in dairy cattle. As a result of the better control of clinical and subclinical mastitis caused by the major pathogens such as Staphylococcus aureus and Streptococcus agalactiae, the bulk milk somatic cell count (SCC) has decreased over the last decades in many areas of the world. A shift towards a higher incidence of IMI caused by the so-called environmental pathogens (e.g. Streptococcus uberis) and by the coagulase-negative staphylococci (CNS) has been observed over the past few years. Nowadays, CNS have become the principal cause of subclinical mastitis on many dairy farms that have controlled contagious mastitis through adopting the so-called 10 point mastitis and prevention control program.

Diagnostics

The group of CNS consists of over more than 50 different species and subspecies with new species being described regularly. Approximately 10 CNS species are causing IMI in dairy cows. Some of the conflicting data from literature could be explained by species-specific characteristics. Still, in order to study species-specific aspects of CNS, differentiation between species should be performed using accurate tests. This was not the case at the time molecular identification was not available and when commercially available test kits, developed for differentiation of CNS from human origin, were used. The test kits have now been ruled out for further differentiation of CNS associated with cows because they lack sensitivity and positive predictive values are low. A number of molecular tools have now been validated for CNS species identification through comparison with gene-sequencing, considered to be the gold standard. They are currently being used to further elucidate the relevance and epidemiology of CNS species for bovine udder health.

Epidemiology

The distribution of CNS species in milk and in different niches such as teat liners, hands of the milkers, the stall environment and teat apices have recently been described using molecular tools, in search of sources and vectors for the species that are frequently causing IMI. These studies have identified S. chromogenes and S. epidermidis as so-called udder-adapted species (causing the majority of IMI), S. simulans and S. haemolyticus as opportunistic species (able of causing IMI but also surviving in the environment), and e.g. S.
equorum and S. fleuretti as so-called environmental species (typically cultured from the environment of cows). More data are, however, needed to substantiate these results.

One of the striking conclusions is that the distributions of CNS species are highly herd-dependent, indicating the need to identify herd-level factors explaining this variation.

Relevance

The relevance of the CNS group has been scrutinized, showing that CNS and Corynebacterium bovis are associated with a moderate increase in SCC, as compared to the substantial increase in SCC caused by major pathogen IMI. Surprisingly, milk production is higher in CNS infected heifers and cows compared to culture-negative cows, whereas it was strongly reduced in cows with a major pathogen IMI. Also, CNS infected heifers are less likely to develop clinical mastitis throughout first lactation. The percentage contribution of CNS IMI to the bulk milk SCC can still be substantial, but only in well-managed herds with a BMSCC < 200,000 cells/ml.

Only limited studies have quantified the species-specific relevance of CNS using molecular speciation. One of the first to do so concluded that CNS as a group was associated with a quarter SCC in between that of non-infected quarters and quarters infected with major pathogens. The effect of the most prevalent species on the quarter SCC, analysed using a linear mixed model, showed that S. chromogenes, S. simulans, and S. xylosus induced an increase in the SCC that is comparable with that of S. aureus. Also, almost all CNS species were able to cause persistent IMI, with S. chromogenes causing the most. Explanations for the better ability to cause chronic infections could be the capacity to produce biofilms. This was, however, not confirmed as the species causing IMI such as S. chromogenes, S. epidermidis, S. simulans and S. haemolyticus were less likely to carry the biofilm associated protein (bap) and ica (mediating the biosynthesis of the matrix polysaccharide intercellular adhesion) genes when compared with CNS typically surviving in the environment.

Protection

Multiple studies have shown that naturally occurring CNS IMI protect against challenge or field infection with major mastitis pathogens. This was also seen in heifers calving with CNS IMI. Teat apex colonization by CNS has also been suggested to exert a protective effect: heifers with teat apices colonized by S. chromogenes had significantly lower odds of having a quarter milk SCC >200,000 cells/ml the first days after calving whereas teat colonization prior to calving with CNS protected quarters of heifers against IMI caused by major pathogens in early lactation. Potential mechanisms explaining these phenomena could be competitive exclusion of other mastitis pathogens, production of inhibitory substances such as bacteriocins, or stimulation of the innate immunity. The fact that heifers from which the teat apices were colonized by CNS prior to calving had an increased number of viable neutrophils in milk in early lactation could point in the that direction.

Some studies did, however, not confirm the protective aspects of pre-existing CNS IMI or teat colonization or reported that CNS infected quarters were at a higher risk for subsequent IMI.
with major pathogens. The role of CNS IMI and teat colonization remains inconclusive and merits further study.

Conclusions

Lots of exciting CNS research using molecular differentiation of species is currently being undertaken revealing what was hypothesized before: difference between CNS species exist and are relevant. Some species are more adapted to the mammary gland than others, affect udder health differently, are more likely to carry virulence and/or resistance genes, with many more differences between species and within species to be detected.
Objective
The objective of the study was to evaluate on farm culturing system in conditions of Czech dairy farms.

Material and methods
For culturing on farms commercial MicroMast system was used with three plates selective and basic agar media. These examinations were done on five farms with the capacity from 300 to 700 cows on each.

Results and conclusion
In all of farms was markedly improved diagnostic procedure of mastitis and contemporarily was changed the therapeutic protocol for target treatment with antimicrobials according etiological agents. Using the on farm culturing system reduced consumption of antimicrobials was achieved and also higher efficiency of clinical mastitis treatment was achieved. Furthermore reduced risk for antimicrobial residues in milk and reduced risk for the development of antimicrobial resistance in mastitis pathogens was observed.
Effective control of infectious mastitis in cattle herds is one of the main interests of both breeders and veterinary services. It is necessary to find an appropriate solution to this problem not only due to the need for production of high quality and safe food, but also due to economic reasons. The control of animal health, including the mammary gland, is one of the few possibilities how to influence economical herd performance.

The efforts to reduce the prevalence of mastitis to the lowest possible level, similarly as of other infectious diseases, consists of two parts - preventive and therapeutic - which complete each other. Prevention programmes are usually aimed at optimization of measures that prevent transmission of infectious agents between dairy cows within a given herd. In this respect, a number of systems have been developed which more or less fulfil this goal. However, despite their relative success, high prevalence of mastitis still exists, particularly on large farms.

Due to the fact that the majority of etiological agents of infectious mastitis were, and in many cases still are, susceptible to the mainly used antibiotics, the effort to eliminate them was aimed at the excessive treatment of all types of mastitis. This attitude to treatment of all types of mastitis, irrespective of the etiological agent and expression and stage of clinical manifestations, not only decreased their prevalence, but also resulted in excessively increased costs and elevated antibiotic resistance among pathogens. Beside these aspects of a primarily economic character, a special attention should be paid to the aspect of general health. The increasing prevalence of antibiotic resistance among pathogenic organisms is considered as one of the most serious threats to current medicine. The above mentioned facts show that infection mastitis control strategies should be both efficient and connected with the lowest possible use of antibiotics. The system presented here attempts to meet the above mentioned requirements.

The system is based on rapid diagnosis of the main causative agents of infectious mastitis by on-farm milk culture and early treatment with efficient antibiotics. Development of this system was based on a method which has been performed successfully in the U.S.A. for several years (Lago et al. 2011). In the system proposed in the current study, samples are plated on Petri dishes, which are divided into three sectors, containing three different chromogenic media, and thus enabling selective culture of the major mastitis-causing pathogens. The fact that each medium is not only selective, but also chromogenic, allows differentiation of a wide spectrum of microbial genera (in some cases also species) within 18 – 24 h with quite high accuracy. Beside basic differentiation of gram-negative and gram-
positive bacteria, the diagnostic system is able to identify the main groups of infectious mastitis pathogens. Among of gram-negative pathogens they are: E.coli and genera Klebsiella and Candida. The two sectors designed to grow gram-negative bacteria allow differentiation between staphylococci and the other genera of this group. Furthermore, different species of staphylococci - S. aureus, S. haemolyticus and S. chromogenes - can be identified by colony colour. Among other gram-positive bacteria that can be causative agents of infectious mastitis, the following species can be identified in the third sector according to their typical colour: S. agalactiae, S. dysgalactiae, S. uberis and Enterococcus faecalis. Based on the test result, targeted treatment can be initiated promptly. However, mastitis caused by gram-negative bacteria should not be treated, unless it is associated with general clinical symptoms. Selection of the most suitable antibacterial agent for a particular pathogen is based on repeated examination of susceptibility of the isolates.

Indications for the above described microbiological testing are as follows:

- clinical manifestations of mastitis,
- altered mammary gland secretions,
- increased somatic cell count by more than 100% compared to the previous test,
- drying-off,
- monitoring of therapy outcomes

For successful implementation of this system, it should be remembered that the system described here does not provide microbiological diagnosis in the real sense of the word. It is rather a tool suitable for optimal therapy selection. However, a definitive diagnosis can only be made in accredited laboratories that perform the confirmation testing, ascertain difficult-to-identify pathogens and primarily, carry out repeated testing of antibiotic susceptibility. The evaluation of these results leads to selection of the most effective antibiotics for treatment of particular pathogens. Continuous monitoring of antibiotic susceptibility at a herd level allows their prompt use for treatment after the pathogen identification.

In conclusion, it should be pointed out that the described system should become part of the entire herd management tools. Breeders should take into consideration that not individual animal, but the entire herd is the target of treatment, and therefore, organisation measures should be adopted which would allow achieving therapeutic goals.

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Introduction

An unimproved rather high lameness incidence and prevalence has been reported in several studies within the last decades still to nowadays all over the world with a wide range from 10% to 70% (Wells et al, 1993; Green et al, 2002; Chapinal et al, 2013). However, although lameness is recognised as an important animal welfare issue (Shearer et al, 2013) and is ranked as the third most important cause of economic loss on dairy farms (Bruijnis et al, 2010), continuous assessment and monitoring of claw health is still in the early stages (Landmann et al, 2004; Wenz & Giebel, 2012; Kofler et al, 2013).

In contrast, the recording and analysis of dairy cow feeding practices, reproduction and metabolic status and udder health are routine herd health management procedures on many dairy farms today (Khaitsa et al, 2000; Martin et al, 2007; Wenz & Giebel, 2012). The availability of computerised data analysis has been recognised as precondition for efficient herd health monitoring (Etherington et al, 1995).

Computerised claw trimming database programs

In recent years, claw-trimming professionals in Europe and North America have introduced several different programs for digital documentation of claw data. They record client and farm details, cow identification, a pre-determined number of claw lesions, severity scores, and applied treatments. Invoices can be calculated and printed instantly after completion of the visit. They are suitable for automatic analysis of some or all of the following parameters: prevalence of different claw lesions, proportion of lesions in each severity score category, accurate lesion location, and the lameness prevalence. Additionally, most of these programs include interfaces to link with other herd health management programs (Kofler et al, 2011; Wenz & Giebel, 2012; Shearer & Van Amstel, 2013).

The Austrian Klauenmanager program offers twelve defined claw lesions and three severity scores of each lesion (Kofler et al, 2010, 2011), according to recommendations from literature (Greenough & Vermunt, 1991; Leach et al, 1998; Smilie et al, 1999). These authors described methods for geometrical calculation of claw scores using severity scores for the various claw lesions, thus establishing the numerical parameters Cow-Claw-Score (CCS), Farm-Claw-Score (FCS) and Farm-Zone-Score (FZS) (Kofler et al, 2011). The CCS is the sum of the geometrically calculated severity scores of the lesions in all ten zones of all eight claws in one
cow (Greenough & Vermunt, 1991; Leach et al, 1998; Smilie et al, 1999). The FCS is the median of all CCS of a herd (Huber et al, 2004). The FZS is the total of all CCS of all cows in a herd for each particular claw zone, and is helpful to identify the most severely and most frequently affected claw zones in a herd (Kofler et al, 2011, 2013). These numerical parameters reflect the type and severity of claw lesions of one single cow (CCS) and of all the cows in one herd (FCS) with one single number for each (Kofler et al, 2011, 2013). Boxplot graphs of the CCS data illustrate the chronological progression of CCS and FCS in a single herd over subsequent claw trimming visits (Kofler et al, 2013).

Conclusions

Digital recording of claw data at regular claw trimming visits and their immediate computed analysis with automatic calculation of a wide range of parameters and their graphical arrangement provide all the prerequisites for vets to establish a modern claw health monitoring program. Furthermore, such recording programs can be used to review the effects of measures taken to improve lameness in herds (Wenz & Giebel, 2012; Kofler et al, 2013; Shearer & Van Amstel, 2013), and nationwide collected computerised claw data enable the estimation of genetic heritabilities of claw lesions (Aamand, 2006).

In the near future, computerised claw trimming database programs will highly support the monitoring of claw health in dairy herds worldwide.

References


CLAW LENGTH DYNAMICS DURING THE FIRST LACTATION

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Introduction

One of the important attributes defining the claw shape is the length of the dorsal wall. The dorsal wall length is dynamic throughout lactation and if the feet are not trimmed, is determined by horn growth and natural abrasion. The objective of this study was to describe the natural dynamic of the claw dorsal wall length during the first lactation.

Materials and methods

Dorsal wall length (DWL) of both lateral and medial claw of the left rear and left front foot was measured in 24 first lactation dairy heifers in total confinement. Measurements were performed 21 days before calving (time -21), at calving (time 0) and monthly until the end of the lactation (month 1-10). Growth and abrasion were determined with horizontal line grooved 2 cm below the skin-horn junction. Analysis of variance was used to determine the effect of stage of lactation, weight and milk production on DWL, and the difference between medial and lateral claws.

Results

Dorsal wall length was not different at time -21 and 0 (81.8±4.2 mm, 82.2±3.8 mm, respectively). The DWL significantly declined to 74.5±2.4 mm through months 1 to 7 which was followed by increase to 79.1±2.7 mm at month 10 (figure 1, 2). The rate of horn growth and abrasion was different (p<0.05) in the first part of the lactation (1-3 month) (figure 3). The DWL of the rear medial claw was longer than the rear lateral claw at -21 days, at calving and in the first month of the lactation. For the rest of the lactation the medial claw was not significantly longer than the lateral. However, the medial claw of front foot was longer than the lateral claw (p<0.05) for day 21 at calving and the rest of the lactation. Body weight and milk production did not influence horn growth and abrasion rate or length of the dorsal wall at any point of the measurement periods.

Implication

This study revealed that horn growth and abrasion influence the natural shape of the claw during lactation. The change of the dorsal wall length was due to horn growth and abrasion dynamic. Modulating this natural process may lead to more appropriate shape of the claw regardless of hoof trimming and reduce development of claw horn lesions.
Figure 1

Toe length of the rear lateral and medial claw

![Image](image1.png)

Figure 2

Toe length of the front lateral and medial claw

![Image](image2.png)
Dorsal wall growth and abrasion, front and rear, lateral and medial claws

stage of lactation

mm

GR
ABR