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virulence and epidemiological potential of farm-specific *S. aureus* to highlight particular points of interest of applied milking and hygiene protocols. Immediate recommendations to the dairy producer were to wash and/or change gloves more often especially after milking different groups of cows, stripping the teats before pre-spraying/dipping, reduce infection load by culling or drying-off infected quarters and to create well defined milking groups cows and heifers.

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Inflammation of mammary gland pathogens isolated from cows' milk in Poland

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Objectives: Inflammation of mammary gland is one of the biggest health problems in dairy cows. It is a physiological, protective response and reaction to any kind of injury or mammary tissue destruction. The aim of this study was to determine the pathogens isolated from the milk of cows with clinical mastitis.

Materials and Methods: 403 milk samples were taken from January 2010 to October 2010 and sent to our laboratory using bacteriological culture. Clinical mastitis was identified by clinical examination of cows, including the mammary gland examination, evaluation of macroscopic changes of mammary gland secretion, and somatic cells count (SCC) in milk using the initial method - California Mastitis Test and Schalm reagent plate and Mastirapid. SCC was determined also by microscopic method. The isolation and identification of microorganisms was performed using bacteriological methods (according to rules developed by Malicki, Binek-2004 and Malinowski, Klosowska-2002). The immune and oxidative factors associated with mammary tissues and secretion can play an important role in protecting the udder from infectious diseases, such as mastitis.

Results: Results showed that the most frequently isolated pathogens for clinical mastitis were: Streptococcus species, CNS, Staphylococcus aureus, Streptococcus agalactiae, Gram-negative bacilli and other bacteria. More-over 95 samples of milk showed no growth. Data suggest that early lactation has an important role in developing mastitis. The ratio of Staphylococcus aureus and Streptococcus species were highest in autumn and winter. The ratio of gram-negative bacteria was higher in summer. In cows in which mastitis was associated with increased internal temperature or other clinical signs, the ratio of clinically affected quarters that were infected by Staphylococcus aureus was bigger than that in cows without systemic reaction.

Conclusions: Interaction of oxidative stress and inflammation response is assumed to be possible in the inflammation of mammary gland and the pathogens depends on the season

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Superoxide dismutase and ceruloplasmin blood activity of cows affected with inflammation of mammary gland using Cefquinom

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Objectives: The aim of the study was to evaluate superoxide dismutase (SOD) and ceruloplasmin (CP) in the blood of cows affected with clinical mastitis during the treatment with Cefquinom.

Materials and Methods: The study was conducted on 22 cows. All cows were subjected to an initial clinical examination, including the mammary gland palpation, evaluation of macroscopic changes in milk and evaluation of somatic cell count in milk using the California Mastitis Test. The milk samples were taken to identify the pathogens. On that basis the cows were divided into two groups. First with mastitis where Streptococcus agalactiae was isolated from their milk, and the control group of healthy cows. Blood samples from all cows were collected 9 times from the external jugular vein before, during, and after treatment with cefquinom. The SOD activity in the erythrocytes of

cows was conducted using Ransod test and CP serum concentration was measured by the method described by Sundermann and Nomoto (1970) and Grys (1988).

Results: The serum activity of SOD and CP concentration noted in cows with mastitis was higher than in healthy cows but it was going down during the treatment.

Conclusions: Although not all of the mechanism of the infections of the mammary glands are known our trial confirmed that the infection stimulates the increase of SOD activity and CP concentration in serum and it decreases during the treatment.

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Systematic sanitation of dairy herds with problems caused by Staphylococcus aureus

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Objectives: Several strategies are known for sanitizing dairy herds problems caused by Staphylococcus (*S.*) aureus. They mostly consist of general management measures but specific decision-making at an individual animal level has not been described.

Materials and Methods: A sanitation program in the form of a process chart developed by the Bern Clinic for Ruminants was undertaken in 10 dairy herds with this problem. In an affected herd the cows are divided into three groups: healthy, suspect, infected. Three milk samples (MS), taken at two-week intervals were cultivated. The cows were grouped according to the culture results. Infected cows were treated and sampled again three times or culled. To measure the success of the sanitation program, the key figures "herd somatic cell count" (target <150,000 SCC/ml) and "percentage of cows over limit" (limit: 150'000 SCC/ml, target <20 %) were used. These were compared with the corresponding key figures from dairy herds, which were followed-up by the Bern Clinic for Ruminants

Results: The problem herd sanitation program lasted between 2 and 21 months. A total of 1598 MS were analyzed, of which 241 were *S. aureus* positive (15 %). At the end of the sanitation the "herd somatic cell count" in 9 herds was lower than at the beginning and in 6 of them under the limit of 150'000 SCC/ml. The "percentage of cows over limit" also decreased in 9 herds and this key figure reached in 5 herds the target. This improvement of both key figures in the problem herds was significant. At the beginning of the sanitation both key figures of the problem herds were significant different from those of the herds in the follow-up program (19 herds). At the end, there was no significant difference in the udder health between the problem and the control herds (23 herds).

Conclusions: The sanitation program to control *S. aureus* IMI can be displayed and used like a quality assurance program. Even under field condition it has proved to be practical. The status of the udder health of each cow was always known and so the decision-making an individual level was possible. The detection of *S. aureus* positive cows proved to be reliable and affected animals could be removed from the herd. The udder health of the herd could be significantly improved. But it shows also, that a successful sanitation is very lengthy and time-consuming.

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Efficiency of somatic cell count and california mastitis test in the diagnosis of subclinical mastitis in terrincha ewes

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Objectives: This study aimed to compare the efficiency of microbiological test with Californian Mastitis Test and somatic cell count in the diagnosis of Subclinical Mastitis (SM) in Terrincha sheep.

Materials and Methods: Twenty-seven of a flock of about 200 Terrincha

ewes (local breed) were studied for a period of 9 weeks ($n > 497$ samples). Milk samples were aseptically collected from each half udder once a week. At the same time, another sample was collected from the bulk tank. After being transported to Lab under refrigeration all samples were immediately processed. The tests performed were the total microbial count (PCA), the Californian Mastitis Test (CMT) and the somatic cell count (SCC). After PCA testing, all samples exceeding 500 cfu/ml of milk (10-1 dilution) were considered positive to mastitis. The SCC was performed by a Fossmatic equipment at the Lactogal Lab.

Results: CMT was more accurate to predict Negative (87.1%) than Positive (43.1%) samples (Chi-square = 42.5; $P=0.001$), meaning that 12.9% half udders were classified as negative being positive and 47.7% half udders were classified as positive being negative. PCA Negative and Positive samples were related to different SCC values (Negative: $277,048.9 \pm 571,249.7$ vs. Positive: $800,329.5 \pm 1,444.970$ somatic cells; $P=0.001$), allowing to identify the real Positive (infected) half udders in complement to CMT.

Conclusions: CMT was a better Negative than Positive mastitis predictor. - Negative and Positive PCA results matched significant different SCC values ($277,048.9 \pm 571,249.7$ vs. $800,329.5 \pm 1,444.970$ somatic cells).

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Effectiveness of treatment intramammary whit hydrochloride ceftiofur for drying of producing milk cows

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Objectives: Due to the large economic losses of mastitis in farms engaged in milk production, preventive measures must be implemented. The drying process using intramammary antibiotics is one of these measures, as it seeks to protect the mammary gland from infection in order achieve a plentiful and good quality next lactation. The overall objective of this study, was to evaluate the efficacy of an intramammary suspension, containing hydrochloride ceftiofur 500 mg, administered at the time of drying.

Materials and Methods: Using the following procedures: 1) Assessment of health status of mammary glands by the California Mastitis Test (CMT). 2) Sample taking for bacteriological culture and isolation, both processes at the beginning and at the end of the dry period; comparison between these procedures was made at the end. Visual inspection of the udders was made during this period.

Results: According to CMT, at the beginning of the experiment, 25.8% of the mammary glands (31 total) scored grades 1 and 2, suggestive of subclinical mastitis and bacterial growth was obtained in 9.67% of the samples out of which was isolated *E. Coli* and *Streptococcus* sp. At the end of the study the total of the mammary glands scored negative for the California mastitis test, however, the 6.45% showed growth in the bacteriological culture process. *Streptococcus* sp. and *Staphylococcus* sp. were isolated, species that are consistent with those isolated by other researchers in Mexico and the world.

Conclusions: Therefore we conclude that the efficacy of intramammary use of ceftiofur hydrochloride 500 mg for the prevention of subclinical mastitis in dry period is 93.5% and that the use of antibiotics is the best prevention of mastitis during the dry period.

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Characterization of bacterial bovine mastitis in the center region

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Objectives: Bovine mastitis is the most prevalent disease in dairy herds and represents an increasing cost to the dairy industry. Diagnostic laboratories can help the determination of the underlying cause and provide a useful guide towards treatment and control. The aim of this study was to provide information regarding the bacterial agents and antimicrobial sensitivity of bovine mastitis milk samples sent to the Laboratório de Medicina Veterinária

(LMV) diagnostic laboratory.

Materials and Methods: From January 2009 to June 2011, 708 milk samples were received in LMV, from a total of 27 farms in the center region of Portugal. Milk samples from individual quarters ($n=677$), composite quarters ($n=18$) and bulk tank ($n=13$) were cultured in Columbia, MacConkey and Sabouraud agar plates and incubated at 37 °C for 18-48 h. Bacterial isolates were biochemically tested according to LMV standard procedures (Gram, catalase, oxidase, coagulase tests) and identified with the aid of the automatized equipment VITEK® 2 Compact (Biomérieux). Antimicrobial sensitivity testing (AST) was performed by disc diffusion in 101 samples.

Results: Bacterial isolates accounted for 29 % of all samples and 51% of positive samples. Gram positive microorganisms constituted the majority of isolates (79%), with a higher prevalence of *Streptococcus* (Str.) *agalactiae* (28%), followed by *Staphylococcus* (*S.*) *aureus* (18%) and *Str. uberis* (16%). Gram negative bacteria were 21% of bacterial isolates, with 14% of *Escherichia* (*E.*) *coli* and 5% of *Klebsiella* sp. There were 354 samples (50%) with no growth or contaminated. Concerning AST, the highest prevalence of resistant strains was observed for tetracycline (45%), followed by ampicillin (24%) and benzylpenicillin (18%). Multi-drug resistant (MDR) strains were 19% ($n=20$) of total strains tested, and were observed chiefly in Gram positive bacteria (74%). The antimicrobial classes more frequently involved in MDR were the tetracyclines and beta-lactams, with 58% of MDR strains resistant to both beta-lactams and tetracyclines and 21% resistant to quinolones and tetracyclines.

Conclusions: *Str. agalactiae* and *S. aureus* were the most common bacterial infectious agents involved and resistance to antimicrobials was observed predominantly for tetracycline and ampicillin. The occurrence of MDR strains was more prevalent for *Str. agalactiae* and *Str. uberis* and involved mainly tetracyclines and beta-lactams.

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Prevalence of mycoplasma bovis in bulk tank milk in dairy farms of Northern Italy

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Objectives: Recent investigation in Northern Italy showed that *M. bovis* mastitis is an increasing problem on dairy farms. This study reports the prevalence of *M. bovis* from samples of bulk tank milk of farms in a selected area of Northern Italy.

Materials and Methods: All the 351 dairy farms of the Lodi province were subjected to bulk tank milk screening between December and September 2011. *Mycoplasma* culture was performed using 10 µL and 100 µL of sample inoculated onto PPLO agar plates and incubated at 37°C in a 10% CO₂ atmosphere. *Mycoplasma* like colonies were subcultivated, subjected to a specific PCR for *M. bovis*. PCR negative DNA's were amplified/sequenced for the 16SrRNA gene. In three positive farms, individual milk samples were collected from all lactating cows. Strains of *M. bovis* from two farms were tested for their antimicrobial susceptibility by MIC method against: Tylosin (Ty), Tilimicosin (Ti), Lincomycin (L), Chloramphenicol (C), Spectinomycin (S), Oxytetracycline (Ox), Enrofloxacin (E), Danofloxacin (D), Erythromycin (Er), Florphenicol (F), Marbofloxacin (M), Tulathromicin (Tu), Clindamycin (Cl).

Results: *Mycoplasma* like colonies were observed in 11 samples and *M. bovis* were confirmed by PCR in 6 of the 351 bulk milk examined (1,7%). The remaining five cultures showed a DNA sequences related to *Acholeplasma* spp. In the three *M. bovis* positive farms where it was possible to test all lactating animals 105/709 (14.8%), 2/166 (1.2%) and 20/104 (19.23%) were excreting *M. bovis*. Results of the MIC indicated a good correlation between strains within the same farm. High MIC levels were seen in *M. bovis* strains to Ty, Ti, Er, C, F, S, Ox, S and Tu.

Conclusions: The survey showed that *M. bovis* mastitis was present in only a small number of farms, but in two of the three positive farms the mastitis involved a high number of cows causing significant economic losses. Treating *M. bovis* with antimicrobials is not usually effective; it is therefore recom-