IMPACT OF TWO DIFFERENT PREVENTIVE TREATMENTS ON MILK FEVER INCIDENCE IN DRY DAIRY COWS

DJELOVANJE DVAJU RAZLIČITIH PREVENTIVNIH POSTUPAKA NA POJAVU MLIJEČNE GROZNICE U ZASUŠEHIH MLIJEČNIH KRAVA

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Izvorni znanstveni članak

SUMMARY

Various methods for the prevention of milk fever (MF) in dry dairy cows are available. We decided on intramuscular (i/m) administration of high vitamin D₃ doses and oral administration of anionic salts. On three dairy farms with similar dietary treatments these two methods were applied in 30 Black and White cows. The first experimental group of cows (n = 10; DCAD = +95.99 mEq/kg DM) was given 10 ml Duphafral® Vit. D₃ 1000 (i/m) one week before the expected calving. The second group (n = 10; DCDA = −99.00 mEq/kg DMI) was orally administered 300 g of anionic salts (KatAn®) per day two weeks before calving. The third group of cows (n = 10, DCDA = +95.99 mEq/kg DMI) was the control group. During the experimental period we controlled the clinical status of cows and venal blood samples were taken (2 and 1 week before parturition, 1, 2 and 7 days postpartum) for the determination of Ca, iP and Mg values. It was established that best preventive results were achieved by i/m application of vitamin D₃. Only one cow from this group was two days after parturition affected by MF; in serum we found 1.47 mmol/L Ca; 0.71 mmol/L iP; 1.37 mmol/L Mg. In KatAn® group one cow was affected by typical MF (1.65 mmol/L Ca; 1.40 mmol/L iP; 1.25 mmol/L Mg) the first day after calving and one cow showed retained foetal membranes. In the control group the incidence of the diseases was considerably higher. One cow had MF (1.34 mmol/L Ca; 0.83 mmol/L iP; 1.48 mmol/L Mg), one had retained foetal membranes and one cow showed left-side displacement of the abomasums within one week after calving. On the basis of the analyses of clinical-laboratory data we are of the opinion that the best prophylactic measure for the prevention of milk fever and with it related diseases on small and medium large farms in Slovenia is i/m administration of high doses of vitamin D₃.

Key words: dry dairy cows, blood biochemical analysis, Ca, iP, Mg, milk fever, prevention,

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INTRODUCTION

Effective health care of dairy cattle will greatly improve herd’s productivity and keep disease to a minimum. As the milk yield of dairy cows in Slovenia has increased over the past 30 years, so has the overall incidence of milk fever (MF), from around 3% to over 7% (Gregorovič et al., 1967; Gašperlin et al., 2002). In some of the Holstein Frisian herds up to 30% of the animals have contracted MF (Gregorovič et al., 1967; Klinkon and Klinkon, 1994). This has resulted in a need to find an effective inexpensive preventive method which can be especially used on farms where dairy cattle are concentrated.

The reason for post calving hypocalcaemia is that some cows are unable to match their rapidly increasing requirements for calcium for milk secretion by absorbing sufficient calcium from their gut or by mobilizing calcium from their own skeleton. Preventive strategies are aimed at ensuring that the strong homeostatic mechanisms that control blood calcium levels are well prepared. Stimulation of release of parathyroid hormone and the formation of 1,25-dihydroxy vitamin D₃ with the resultant increased absorption of calcium from the bone take at least 24 hours. The timing of preventive procedures is therefore important (Andrews, 2000; Goff, 2004).

MF (hypocalcaemia) generally occurs at near calving. It is caused by a large calcium demand at the onset of milk production. Both subclinical and clinical apparent MF may present a major economic problem due to reduction of both the milk production and the life spans of affected high-producing dairy cows (Oetzel, 1988; Ruegg, 1991). There is also a highly significant relationship between MF and dystocia, retained fetal membranes, ketosis, mastitis, displacement of the abomasums and uterine prolapses (Gröhn, et al., 1990; Massey, et al., 1993; Ziggers, 2004). The after effects of hypocalcaemia conditions are very costly, illustrating the importance of prevention. Compounding the problem are the ever changing nutritional needs of the cow, her lactation/dry period needs, feed quality changes, and producer personal management practices (Gašperlin, et al., 2002; Rice, 2005).

When the herd incidence of MF increases to above 5% of high-risk cows (third or later lactations), a specific control program is necessary. Various methods for the control of MF in dairy cows are available. They include dietary management during pre calving period (Goff and Horst, 1993; Block, 1994), oral administration of calcium gels at the time of parturition (Pherson and Jonsson, 1991) and administration of vitamin D₃ or its metabolites immediately before parturition to enhance the mobilization of Ca (Seekles, et al., 1958; Gregorovič, et al., 1967; Yamagishi, et al., 2005).

The aim of our research was to verify the prophylactic effects of i/m application Duphafral® Vit. D₃ 1000 and feeding KatAn® anionic salts on the incidence of apparent MF and with it related diseases.

MATERIAL AND METHODS

The research took place on three dairy farms with a total of 124 Black and White (88%) and Simmental (12%) cows. For the experiment 30 high-yielding Black and White dairy cows that had previously been stricken with parturient paresis were selected. Their age ranged from 4 years 3 months to 10 years 3 months, with average of 6.4 years. All cows were clinically healthy and in good condition (mean BSC = 3.5).

The first experimental group of cows (n = 10; DCAD = +95.99 mEq/kg DM) – Duphafral® group – was given i/m 10 ml Duphafral® Vit. D₃ 1000 (Fort Doge) one week before the expected calving. The second group (n = 10; DCDA = - 99.00 mEq/kg DM) was given 300 g KatAn® anionic salts per day (Kimtec International: 6.5% Mg, 7.3% S and 18.2% Cl; DCAD = [(Na + K) – (Cl + S)] = - 9800 mEq/kg) two weeks before parturition. The third group of cows (n = 10, DCDA = +95.99 mEq/kg DM) was used as a control group.

During the experimental period venal blood samples (v. caudalis mediana) were taken 5 times: 2 and 1 week before parturition, 1, 2 and 7 day postpartum). A Cobas Mira automatic biochemical analyzer (Hoffman-La Roche) was used to determinate calcium (Ca), inorganic phosphorus (IP) and magnesium (Mg) in the blood sera.

During the dry (close-up) period right to the first day after calving the cows were fed with corn and grass silage, hay and concentrates for dry cows with vitamin-mineral supplementation. The average daily feed intake was 11.0 kg of grass silage, 14.0 kg of...
corn silage, 2.0 kg of hay, 1.2 kg of squashed corn grains and 0.8 kg mill prepared concentrates for dry cows. Samples of hay, corn silage, and grass silage, squashed corn grains, concentrates and vitamin-mineral supplementation were analyzed once during the trial. The chemical composition of the fodder was determined by proximal analysis, calcium, sodium, potassium and magnesium were analyzed by an atomic absorption spectrometer and the phosphorus with a molybdovanadate reagent. Analyses on sulfur and chlorine were made by x–ray fluorescence spectrometry.

The formula for calculating the dietary cation-anion difference was as follows: DCAD (mEq/kg DMI) = [(Na⁺ + K⁺) – (Cl⁻ + S⁻)]. The DCAD programs, was written in Excel, ver. 7.0, Windows 95.

The One-Way program from Statistical Package for the Social Science made the statistical evaluations and ANOVA was used to calculate the significance of differences between the test groups of cows and the control group.

**RESULTS**

All cows in the experiment were clinically observed. We focused on periparturient period especially on the day of calving and one week following it. Within this critical period for MF development and accompanying diseases blood samples were taken. We paid close attention to disease symptoms associated with dystocia, prolonged calving time, depression, gait stumbling, recumbency and coma. Cows were clinically observed until one month after calving. Recorded were signs of retained foetal membranes, metritis, dislocation of the abomasum and mastitis. The clinical results and laboratory analyses of affected cows are shown in Table 1 and 2.

**Table 1. Milk fever (MF) incidence and related diseases retained foetal membranes (RFM) and left-side displaced of the abomasum (LDA) in three groups of cows**

<table>
<thead>
<tr>
<th>Group of cows - Skupina krava</th>
<th>Healthy Zdrave</th>
<th>MF</th>
<th>RFM</th>
<th>LDA</th>
<th>Total - Ukupno</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control - Kontrolna</td>
<td>7 (70%)</td>
<td>1 (10%)</td>
<td>1 (10%)</td>
<td>1 (10%)</td>
<td>10 (100%)</td>
</tr>
<tr>
<td>Duphafral® Vit. D₃ 1000</td>
<td>9 (90%)</td>
<td>1 (10%)</td>
<td>-</td>
<td>-</td>
<td>10 (100%)</td>
</tr>
<tr>
<td>KatAn®</td>
<td>8 (90%)</td>
<td>1 (10%)</td>
<td>1 (10%)</td>
<td>-</td>
<td>10 (100%)</td>
</tr>
</tbody>
</table>

**Table 2. Hemato-biochemical* analysis of six cows affected by milk fever (MF), retained foetal membranes (RFM) and left-side displacement of the abomasum (LDA)**

<table>
<thead>
<tr>
<th>Group of cows - Skupina krava</th>
<th>Name of the cow</th>
<th>Diagnosis</th>
<th>Ca mmol/L</th>
<th>IP mmol/L</th>
<th>Mg mmol/L</th>
<th>Na mmol/L</th>
<th>K mmol/L</th>
<th>AST U/L</th>
<th>ALT U/L</th>
<th>GGT U/L</th>
<th>CK U/L</th>
<th>E x10¹²/L</th>
<th>L x10⁹/L</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control Kontrolna</td>
<td>Mili MF</td>
<td>MF</td>
<td>1.34</td>
<td>0.83</td>
<td>1.48</td>
<td>149</td>
<td>5.94</td>
<td>59</td>
<td>10</td>
<td>11</td>
<td>198</td>
<td>6.69</td>
<td>3.4</td>
</tr>
<tr>
<td>Živa RFM</td>
<td>2.15</td>
<td>1.79</td>
<td>0.99</td>
<td>143</td>
<td>6.34</td>
<td>63</td>
<td>12</td>
<td>16</td>
<td>92</td>
<td>6.14</td>
<td>8.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Una LDA</td>
<td>2.34</td>
<td>1.49</td>
<td>0.90</td>
<td>144</td>
<td>5.10</td>
<td>56</td>
<td>9</td>
<td>31</td>
<td>56</td>
<td>5.56</td>
<td>8.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Duphafral® Vit.D₃ 1000</td>
<td>Muta MF</td>
<td>MF</td>
<td>1.47</td>
<td>0.71</td>
<td>1.37</td>
<td>147</td>
<td>5.97</td>
<td>55</td>
<td>13</td>
<td>19</td>
<td>332</td>
<td>6.66</td>
<td>7.0</td>
</tr>
<tr>
<td>KatAn®</td>
<td>Švica RFM</td>
<td>MF</td>
<td>1.65</td>
<td>1.40</td>
<td>1.25</td>
<td>154</td>
<td>6.46</td>
<td>86</td>
<td>16</td>
<td>17</td>
<td>1538</td>
<td>7.19</td>
<td>10.6</td>
</tr>
<tr>
<td>Rika RFM</td>
<td>2.13</td>
<td>1.84</td>
<td>1.00</td>
<td>145</td>
<td>6.01</td>
<td>55</td>
<td>14</td>
<td>13</td>
<td>87</td>
<td>6.23</td>
<td>9.2</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Blood samples were drawn just before making the diagnosis; MF = 1 or 2 days after calving, RFM = 24 hours after calving, LDA = 1 week after calving.*
In our experiment three cows, one from each group, were affected by MF. All cows with MF were recumbent and showed characteristic clinical signs of the disease within 48 hours after calving. The results of the blood analyses taken immediately before therapy also confirmed that cows were affected by typical MF (Table 2).

From Table 2 it is evident that the incidence of postpartum diseases related to MF, and postpartum hypocalcaemia respectively, was highest in the control group of cows. In this group three cows were affected: one by MF, the second had retained foetal membranes and the third left side displacement of the abomasum. In KatAn® group one cow had MF and another cow retained foetal membrane. The analyses of blood serum confirmed that cows were affected by typical signs of MF and related diseases as shown by low Ca and iP values compared to clinically normal cows. The unexpected occurrence of the disease is to our belief associated with deficient Ca concentration in the feed intake after calving when the normal requirements for this essential element are suddenly markedly elevated. During close-up period the feed intake is often reduced. The most severe depression in feed intake takes place just before or at calving when the cow needs energy and especially Ca to expel the foetus and increases colostrum production (Bertics, et al., 1992; Grummer, 1995).

These results obtained in an objective study of homogeneous groups of cows clearly demonstrate that the i/m administrated Duphafral® Vit. D₃ 1000 one week before calving is a very effective and economical preventive method for MF and accompanying diseases.

**DISCUSSION**

Balancing the cation-anions in the diet is a relatively new method to prevent MF, to improve health and production. Alkalogenic diets (> +200 mEq/kg DMI) tend to cause MF, whereas acidogenic diets tend to prevent it (Dishington, 1975; Oetzel, 1993; Block, 1994). In the literature various approaches for the calculation of DCAD in feed intake are described (Goff, 1992; Tucker, et al., 1992; Sanchez and Blauwickel, 1994). Most authors in their research calculated DCAD on the basis of the difference among the summing up of Na, K, Cl, and S quantities so we used this formula as well. According to this calculation cows from the control group and cows from the group that received the vitamin D₃ injection were fed a diet with mean DCAD +95.99 mEq/kg DMI, and KatAn group mean −99.00 mEq/kg DMI. Many researches report that increased anionic diet contributes only to a lower incidence of clinical form of MF (Ender, et al., 1962; Dishington and Bjørnstad, 1982; Block, 1984), while the others claim that the anionic diet affected also the degree of Ca level in the blood of cows during puerperal period (Vagg and Payne, 1970; Gaynor, et al., 1989; LeClerc and Block, 1989). It turned out that the processes of retention, absorption, and balancing of Ca, iP, 1,25-dihydroxyvitamin D₃ and parathyroid hormone concentration during the puerperal phase of cows were more expressed when pregnant cows were on anionic diet notwithstanding an apparent calcinuria (Verdaris and Evans, 1976; Lomba, et al., 1978; Oetzel, et al., 1991). Thus the anion diet protected the cows from pathological processes of hypocalcaemia because the 1,25-dihydroxyvitamin D₃ level in blood increased.

In our case the control group and the group of cows which was 7 days before the expected calving administered i/m injection of Duphafral® Vit. D₃ 1000, received the same feed intake and thus the same DCAD +95.99 mEq/kg DMI. The results of some research show that feed intakes with DCAD values between +50 to +300 and more mEq/kg DMI generally increase the risk of MF incidence (Block, 1984; Oetzel, 1993; Block, 1994). Many nutritionists are of the opinion that for the prevention of MF it is necessary to add anions salts only when DCAD is higher than +200 mEq/kg DMI (Oetzel, 1993; Block, 1994; Rajčevič, et al, 1999). In our case the addition of 300 g/day of anion salt KatAn® caused the change of DCAD value of feed intake to −99.00 mEq/kg DMI. Sanchez and Blauwickel (1994) claim that only values of DCAD in feed intake of dry cows between −100 to −150 mEq/kg DMI contribute to successful MF prevention. Goff, 1992; Goff and Horst, 1993 reached similar conclusion, namely, only values between −100 and −200 mEq/kg DMI began to efficiently prevent MF.

Several blood analyses in cows with MF were done (Kronfeld, 1971; Ramberg, et al., 1984; Oetzel, 2000). It was established that serum Ca
concentration decreased below 2.0 mmol/L, usually below 1.2 mmol/L and sometimes below 0.5 mmol/L. Clinical picture of MF is sometimes to a certain degree associated with the concentration of the measured Ca in serum, however this is not to be taken for granted. It is important to know that it is a case of physiological occurrence because serum Ca concentration is immediately after calving often decreased below its level, which is from 1.75 to 2.25 mmol/L. A decrease of Ca concentration is observed in all dairy cows to see if they are or not attacked by MF. The line between subclinical hypocalcaemia and clinical form of MF has not been clearly defined yet. Larsen, et al., 2001 report that characteristic clinical signs (muscle weakness, depression of the cardiovascular system, hypothermia, recumbency, depression of consciousness) began to show when Ca concentration in the blood decreased below 1.60 mmol/l. Jazbec, et al., 1970 reports that 84% of cows (n = 120) with clinical form of MF had in the blood ≤1.84 mmol/L Ca, 75% of cows ≤1.13 mmol/L iP and 70% of cows >0.82 mmol/L Mg. Serum iP is usually low, values are between 0.48 to 0.97 mmol/L. Besides hypocalcaemia we can often find the s. c. concurrent with hypophosphataemia. Serum Mg usually slightly increases immediately after calving to 1.65–2.06 mmol/L (Jazbec, et al., 1971; Kronfeld, 1971).

CONCLUSION

It was established that best results were achieved by application of Duphafra® Vit. D₃ 1000 to dry cows. Only one cow from this group was two days after parturition affected by atypical PH; in serum we found 2.31 mmol/L Ca; 0.94 mmol/L iP; 1.01 mmol/L Mg. In KatAn® group one cow was affected by typical PH (1.47 mmol/L Ca; 0.71 mmol/L iP; 1.37 mmol/L Mg) first day after parturition and two showed retained fetal membranes. In the control group the incidence of the disease was considerably higher. One cow had MF (1.86 mmol/L Ca; 2.36 mmol/L iP; 0.85 mmol/L Mg); three had retained fetal membranes and one cow showed left-side displacement of abomasum. In the control group the incidence of the disease was considerably higher. One cow had MF (1.86 mmol/L Ca; 2.36 mmol/L iP; 0.85 mmol/L Mg); three had retained fetal membranes and one cow showed left-side displacement of abomasum. In the control group the incidence of the disease was considerably higher. One cow had MF (1.86 mmol/L Ca; 2.36 mmol/L iP; 0.85 mmol/L Mg); three had retained fetal membranes and one cow showed left-side displacement of abomasum. On the basis of statistical analyses of clinical-laboratory data we are of the opinion that the best prophylactic measure for the prevention of MF and with it related diseases on small and medium large farms in Slovenia is the i/m administration of high doses of vitamin D₃ one week before calving.

LITERATURE

SAŽETAK

Postoje razne metode za prevenciju mlječne groznice (MF) u zasušenih mlječnih krava. Mi smo se odlučili za intramuskularnu (i/m) primjenu velikih doza vitamina D₃ i oralnu primjenu anionskih soli. Ove su dvije metode primijenjene na 30 crno-bijelih krava na tri mlječne farme sa sličnim postupcima hranjbe. Prva pokusna skupina krava (n=10; DCAD = + 95.99 mEq/kg DMI) dobila je 10 ml Duphaprala vit. D₃ 1000 (i/m) jedan tjedan prije očekivanog telenja. Druga skupina (n=10; DCDA = - 99.00 mEq/kg DMI) dobila je oralno 300 g anionskih soli (KatAn®) na dan dva tjedna prije telenja. Treća skupina krava (n=10; DCDA = + 95.99 mEq/kg DMI) bila je kontrolna skupina. U pokusnom razdoblju kontrolirali smo klinički status krava i uzeli uzorke krvi iz vene (2 i 1 tjedan prije porođaja, 1, 2 i 7 dana postpartum) radi određivanja vrijednosti Ca, iP i Mg. Utvrđeno je da su najbolji preventivni rezultati postignuti i/m primjenom vitamina D₃. Samo je jedna krava iz ove skupine oboljela od MF dva dana nakon porođaja; u serumu smo našli 1.47 mmol/L Ca; 0.71 mmol/L iP; 1.37 mmol/L Mg. U skupini KatAn jedna je krava oboljela od tipične MF (1.65 mmol/L Ca; 1.40 mmol/L iP; 1.25 mmol/L Mg) prvog dana nakon telenja, a kod jedne je krave zadržana posteljica. U kontrolnoj skupini pojava bolesti bila je znatno veća. Jedna je krava imala MF (1.34 mmol/L Ca; 0.83 mmol/L iP; 1.48 mmol/L Mg), jedna je zadržala posteljicu, a kod jedne je došlo do skretanja abomasuma u lijevo u prvom tjednu nakon telenja. Na temelju analiza kliničkih laboratorijskih podataka mišljenja smo da je najbolja profilaktička mjera za prevenciju mlječne groznice i srodnih oboljenja na malim i srednje velikim mlječnim farmama u Sloveniji i/m primjena velikih doza vitamina D₃.

Ključne riječi: zasušene mlječne krave, biokemijska analiza krvi, Ca, iP, Mg, mlječna groznica, prevencija