Gammaglobulin and selenium status in healthy neonatal dairy calves in Switzerland

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Introduction

Calves are born agammaglobulinemic and depend on the maternal antibodies provided in their dams' colostrum to cope with infectious agents during their first weeks of life. Failure to ingest an adequate volume of colostrum or to absorb adequate amounts of immunoglobulins (Ig), e.g. in case of poor colostral quality, can cause hypogam-

Summary

Serum samples from 142 calves and their dams were analyzed for gammaglobulins (γG, calves) and selenium concentrations (Se, calves and dams). A questionnaire provided information about birth and colostrum management. The calves and their dams were distributed into two groups according to the calves’ γG concentration (< 10 and ≥ 10 g/L). Se concentrations were compared between groups. The correlation between γG and Se concentrations in the calves and their dams was analyzed. Risk factors for failure of passive transfer and Se deficiency were assessed based on the questionnaire. The γG concentration of 42.9 % of the calves was < 10 g/L (median: 10.9). Calves showed significantly higher γG values after optimized colostrum administration than calves with suboptimal colostrum administration (p < 0.004). The median Se concentration was 26.8 and 36.5 μg/L for the calves and dams, respectively. A high correlation was observed between the Se concentration of the dam and her calf (r = 0.72, p < 0.001). The calves' Se and γG concentrations were not significantly correlated. These results demonstrate that further efforts toward better information of farmers regarding colostrum management and Se supply are warranted.

Keywords: selenium, gammaglobulins, immunoglobulins, calf health, colostrum
maglobulinemia in newborn calves, which is associated with a higher risk of infectious diseases (Donovan et al., 1998; Weaver et al., 2000). Hence, failure of passive transfer (FPT) of immunity from the dam to the calf is also associated with higher morbidity and mortality rates in calves (Dewell et al., 2006). Some authors have used cutoff points of < 8 g/L serum gammaglobulin (γG) (Lanz Uhde et al., 2008) or < 8 g/L serum IgG1 (Dewell et al., 2006) to describe FPT. Others considered < 10 g/L serum IgG1 (Barrington and Parish, 2002) or < 10 g/L serum IgG (Beam et al., 2009) as adequate. Recommendations for successful colostrum management include the ingestion of at least 2 L of high quality colostrum (≥ 50 g/L Ig) within 2–3 hours of birth, followed by another 2 L within the next 4 hours (Rademacher, 2007; Godden, 2008). In a study about the prevalence of major pathogens in diarrheic calves in Switzerland, 90.5% of 147 calves showed FPT defined as < 8 g/L γG. Had a cutoff point of < 10 g/L γG been chosen, 96.6% of the diarrheic calves would have been considered to have FPT (Lanz Uhde et al., 2008). No data for healthy calves from the same area were available for comparison.

Selenium (Se) is an essential trace element which contributes to the protection of cells from oxidative damage and therefore plays an important role in health problems related to oxidative stress such as mastitis and retained placenta in dairy cows or white muscle disease (WMD) in calves (Combs, 2001; Spears and Weiss 2008; Maas and Valberg, 2009). The amount of Se ingested by cattle with solid feed depends not only on the Se content in soils, but may also be influenced by factors such as soil pH, moisture, or the use of Se-containing fertilizers (Combs, 2001). Different reference ranges, depending on the material and the analysis method used have been reported for blood Se values in the literature. Reference values for marginal Se concentrations vary between 30 and 70 μg/L serum (Scholz and Stöber, 2002), and 50–120 μg/L whole blood (Radostitis et al., 2007; Guard 2008; Maas and Valberg, 2009). Selenium serum concentrations of > 70 μg/L (Scholz and Stöber, 2002) and whole blood concentrations of > 70–300 μg/L have been described as normal (Radostitis et al., 2007; Guard, 2008; Maas and Valberg, 2009). Two studies on the Se status of cows and calves in Switzerland report low serum and whole blood concentrations of clinically healthy animals: in the study by Mathis et al. (1983), the mean blood Se concentration in cows and calves from farms with a high incidence of WMD was 15.6 μg/L, and was significantly different from the Se concentration in animals from farms without the disease (29.6 μg/L). These authors described the critical limit for WMD as being approximately 20–30 μg Se in one liter of whole blood (Mathis et al., 1983). In a study on serum Se and vitamin E concentrations in cows from farms with and without increased incidence of different diseases related to Se (chronic mastitis in cows, WMD in calves), 90% of the control animals had Se concentrations below 30 μg/L without showing clinical signs of Se deficiency (Braun et al., 1991). Newer data on the Se status of cows and calves in Switzerland are not available.

Swecker et al. (1995) have described the effect of Se supplementation of beef cows with a marginal Se status (blood Se concentration: 50 μg/L) and grazing Se-deficient pastures during the second half of their gestation on colostral IgG concentrations and on the transfer of IgG to their calves. Oral Se supplementation of the cows led to higher colostral IgG concentrations and to higher post suckle serum IgG concentrations in their calves, as compared with parenteral or no supplementation. In another study, direct supplementation of colostrum with Se increased Ig absorption in calves (Kamada et al., 2007). It has been repeatedly observed at the authors’ clinic that calves from farms with a high incidence of neonatal diarrhoea often present with very low to undetectable serum Se concentrations in addition to FPT.

The goal of the present study was to describe the prevalence of FPT in clinically healthy newborn dairy calves in Switzerland and to explore possible correlations between serum Se and γG concentrations of the calves, as well as between the serum Se concentration of the calves and their dams. Finally, risk factors for FTP and Se deficiency in animals originating from farms without known problems related to calf health were assessed based on the questionnaire data regarding the management of newborn calves in Swiss dairy farms.

**Animals, Material and Methods**

**Animals**

The study was conducted in the catchment area of the Clinic for Ruminants in Bern between May 2008 and January 2011. One-hundred-and-forty-two 2 to 5 days old, clinically healthy dairy calves from farms with a minimal herd size of 10 cows were included in the study. The calves belonged to the breeds Red Holstein (n = 85), Holstein-Friesian (n = 29), Brown Swiss (n = 6), Simmental (n = 6), or were crossbreds (n = 16), 72 of them were female and 70 male calves. Inclusion criteria were a good general condition at the time of sampling (according to the findings of a physical examination performed by a veterinarian), especially a good suckling reflex and normal fecal consistency. The birth of calves had to be unassisted (or with minimal assistance by the owner), and calves had to have ingested an adequate volume of colostrum according to the owner’s appreciation. Exclusion criteria included previous treatment with Se (dam and calf) or Ig preparations (calf), and the presence of known problems related to calf health such as diarrhea, pneumonia or BVDV infection on the farm. For twin calves, only the data of the first calf mentioned on the questionnaire was included in the study. Only one calf and its dam were included per farm.
Samples

Blood samples were collected by the local veterinarians from the jugular vein into serum tubes (Sarstedt Monovette®, 01.1601.001). Twenty ml were taken from the calves and from their dams. The blood samples were shipped to the Clinic for Ruminants in Bern within 3 working days. The blood samples were centrifuged (15 min, 10 G), the serum separated and kept frozen (−20 °C) until analysis. The γG fraction in the calves’ serum was quantified by serum electrophoresis (Paragon Serum Protein Electrophoresis-Kit, Beckmann Coulter, USA). Selenium in the serum of the calves and their dams was measured using atomic absorption spectroscopy (Zeenit 650, Analytik Jena AG, Jena, Germany).

Questionnaire

A questionnaire regarding the calf’s birth and colostrum intake was completed by the veterinarian with the owner at the time of blood collection. The registered information included details about the birth (single versus twin calving, spontaneous birth or birth assisted by the owner), the parity of the dam (primiparous versus multiparous) and details about colostrum management (time and volume of the first and the second colostrum feeding).

Data analysis

The non-parametric Spearman’s correlation test was used to explore correlations between the γG and Se concentrations of the calf, and of these concentrations with the time and volume of first colostrum feeding, as well as of the calves’ γG and Se concentration with the Se concentration of their dams (significance level set at p < 0.05). For further analyses, the γG concentrations were re-grouped in a binary outcome variable with < 10 g/L γG (= FPT) and ≥ 10 g/L γG (= adequate), and the Se concentrations were dichotomously grouped in ≤ 35 μg/L Se (Se deficient) and > 35 μg/L Se (Se sufficient) (Guard, 2008). Risk factors (time and volume of first colostrum feeding, parity of the dam, and ease of birth) for deficiency (< 10 g/L γG and ≤ 35 μg/L Se) were evaluated with univariable logistic regression (significance level set at p < 0.05). The explanatory variable ‘ease of birth’ was categorized as i) uncomplicated birth (unassisted birth) and ii) complicated birth (twins calving, and/or mild assistance by the owner). The parameter ‘volume of first colostrum feeding’ was grouped in 2 categories: < 2 L and ≥ 2 L. The ‘time of first colostrum feeding’ was distributed in 3 categories: ≤ 2 h, > 2–≤ 6 h and > 6 h. Reliable data on the volume and time of second colostrum feeding could only be obtained from 10/142 calves and were therefore not further analyzed. A new variable ‘colostrum intake’ was derived from the two variables volume and time of first colostrum feeding with different levels of good practice: i) ≥ 2 L in ≤ 2 h, ii) < 2 L in ≤ 2 h, iii) ≥ 2 L in > 2 h and iv) ≤ 2 L in > 2 h.

A multivariable logistic regression was performed for the binary outcome calf γG concentrations (< 10 g/L γG or ≥ 10 g/L γG) with the explanatory parameters volume of first colostrum feeding, time of first colostrum feeding, parity of the dam and ease of birth. The statistical analyses were done in Stata 10.

Results

Results for γG and Se concentrations were available from all 142 pairs of samples collected. Complete questionnaire data were available for 127 calves. The median γG concentration was 10.9 g/L (Q1 6.7 – Q3 16.3), with 42.9% of the calves showing values < 10 g/L γG and 32% values < 8 g/L γG. The median value for calves with FPT (γG < 10 g/L) was 6.5 g/L (n = 61) and 15.3 g/L for calves with adequate γG concentrations (n = 81; Fig. 1). The median serum Se concentration was 26.8 μg/L (Q1 18.1– Q3 33.3) and 36.5 μg/L (Q1 25.0 – Q3 51.1) in the calves and their dams, respectively. No statistically significant correlation was present between the γG and Se concentrations of the calves (r = 0.16, p = 0.053). No significant difference in Se concentration was observed between calves with γG concentrations < 10 g/L and ≥ 10 g/L and in the Se concentration of their dams (Fig. 2). The Se concentrations between the calves and their dams was highly correlated (r = 0.72, p < 0.001). Weak correlations were observed between the time of first colostrum feeding and the calves’ γG concentration (n = 133, r = −0.18, p = 0.03) and between the volume of first colostrum feeding and γG (n = 134, r = 0.24, p = 0.005). No significant correlations were found between the time (n = 133, r = 0.02, p = 0.81) or the volume (n = 134, r = 0.06, p = 0.49) of first colostrum feeding and the calves’ serum Se concentration.

Univariable analysis revealed that calves receiving their first colostrum feeding more than 6 hours after birth had
a higher risk to be γG deficient than calves receiving their first colostrum within 2 hours after birth. A volume of ≥ 2 L at first colostrum feeding appeared to be a protective factor against γG deficiency, however this effect was not statistically significant. Univariable analysis with the combined variable ‘colostrum intake’ showed that all groups for which best practice of colostrum feeding (> 2 L in < 2h) had not been used had an increased risk for FPT. No significant effects of colostrum management factors on Se status of the calves were observed. Detailed results are given in Table 1. Multivariable analysis revealed an OR for FPT of 7.4 (95% CI 1.4–39.5; p = 0.02) if calves received their first colostrum feeding more than 6 hours after birth when compared to the baseline of ≤ 2 h, whereas no increased risk was observed if the calves received their first colostrum after 2 hours but prior to 6 hours after birth (OR 1.6, 95% CI 0.7–3.4; p = 0.24). A volume of colostrum ≥ 2 L at first feeding still significantly reduced the risk of FPT in comparison to a smaller volume (OR 0.46, 95% CI 0.22–0.99; p = 0.05).

Neither the parity of the dam nor the ease of birth was associated with the γG or Se status of calves in the univariable or the multivariable analysis.

### Discussion

The low γG and Se concentrations measured in the serum of clinically healthy dairy calves in Switzerland demon-

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**Table 1: Univariable risk factor analysis for γG and Se status in healthy dairy calves.**

<table>
<thead>
<tr>
<th>Category</th>
<th>γG ≥ 10 g/L, n</th>
<th>γG &lt; 10 g/L, n</th>
<th>OR (95% CI)</th>
<th>p</th>
<th>Se &gt; 35 µg/L, n</th>
<th>Se ≤ 35 µg/L, n</th>
<th>OR (95% CI)</th>
<th>p</th>
</tr>
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<tbody>
<tr>
<td>Time of first colostrum feeding</td>
<td></td>
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<tr>
<td>≤ 2 h</td>
<td>47</td>
<td>25</td>
<td>1</td>
<td></td>
<td>14</td>
<td>58</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>&gt; 2 ≤ 6 h</td>
<td>28</td>
<td>24</td>
<td>1.6 (0.8–3.3)</td>
<td>0.2</td>
<td>10</td>
<td>42</td>
<td>1.0 (0.4–2.5)</td>
<td>0.97</td>
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<tr>
<td>&gt; 6 h</td>
<td>2</td>
<td>7</td>
<td>6.6 (1.3–34.1)</td>
<td>0.025</td>
<td>3</td>
<td>6</td>
<td>0.5 (0.1–2.2)</td>
<td>0.34</td>
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<td>Volume of first colostrum feeding</td>
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<tr>
<td>&lt; 2 L</td>
<td>25</td>
<td>28</td>
<td>1</td>
<td></td>
<td>10</td>
<td>43</td>
<td>1</td>
<td></td>
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<tr>
<td>≥ 2 L</td>
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<td>29</td>
<td>0.5 (0.2–1.0)</td>
<td>0.053</td>
<td>17</td>
<td>64</td>
<td>0.9 (0.4–2.1)</td>
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<tr>
<td>≥ 2 L in ≤ 2 h</td>
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<td>12</td>
<td>1</td>
<td></td>
<td>8</td>
<td>32</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>&lt; 2 L in ≤ 2 h</td>
<td>17</td>
<td>20</td>
<td>3.3 (1.3–8.6)</td>
<td>0.016</td>
<td>15</td>
<td>58</td>
<td>1.0 (0.4–2.5)</td>
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<tr>
<td>≥ 2 L in &gt; 2 h</td>
<td>16</td>
<td>16</td>
<td>3.4 (1.2–9.5)</td>
<td>0.017</td>
<td>16</td>
<td>58</td>
<td>1.0 (0.3–3.8)</td>
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<tr>
<td>≤ 2 L in &gt; 2 h</td>
<td>9</td>
<td>11</td>
<td>4.2 (1.3–13.3)</td>
<td>0.014</td>
<td>4</td>
<td>16</td>
<td>1.0 (0.3–3.8)</td>
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<td>Primiparous</td>
<td>17</td>
<td>14</td>
<td>1</td>
<td></td>
<td>11</td>
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<td>Multiparous</td>
<td>63</td>
<td>48</td>
<td>1.1 (0.5–2.5)</td>
<td>0.86</td>
<td>26</td>
<td>85</td>
<td>0.5 (0.2–1.6)</td>
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<tr>
<td>Ease of birth</td>
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<tr>
<td>Uncomplicated</td>
<td>44</td>
<td>34</td>
<td>1</td>
<td></td>
<td>20</td>
<td>58</td>
<td>1</td>
<td></td>
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<tr>
<td>Complicated</td>
<td>36</td>
<td>26</td>
<td>0.9 (0.5–1.8)</td>
<td>0.84</td>
<td>10</td>
<td>52</td>
<td>1.8 (0.8–4.2)</td>
<td>0.18</td>
</tr>
</tbody>
</table>

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**Figure 2:** Box-plots of the Se concentrations in healthy neonatal dairy calves and their dams according to the calves’ serum γG concentration (γG < 10 g/L, γG ≥ 10 g/L).
strate that further efforts toward better information of farmers regarding the importance of colostrum management and Se for the health of their calves are warranted. Despite a median γG concentration of 10.9 g/L for the 142 calves included in the study, the prevalence of FPT in clinically healthy neonatal calves was almost 43% and remained more than 30% if the cutoff point was set at 8 g/L. Because other factors (e.g., management, infectious agents) than serum Ig concentration may influence the occurrence of disease in a study population, the choice of a cutoff point to describe FPT depends on the goal of the study (Barrington and Parish, 2002). The clinically healthy calves in the present study were expected to have optimal, i.e., high serum γG concentrations, thus the cutoff point at 10 g/L γG was chosen to define FPT. The proportion of healthy calves with a γG concentration below that value was surprisingly high (42.9%). Similar results had been recorded in US dairy heifer calves between 1991–1992 (USDA, 1993), but a more recent study showed a prevalence of FPT reduced to 19.2% in 2007. This positive trend was thought to be the result of educational campaigns for farmers about optimal colostrum management (Beam et al., 2009).

Several management factors play an important role in the achievement of optimal Ig values in calves (Weaver et al., 2000). A delay of first colostrum feeding or the administration of a low volume at first colostrum feeding are known to negatively influence the final Ig serum concentration of the calf (Godden, 2008). According to the information obtained from the questionnaire, 93 of 133 calves (69.9%) had been subjected to insufficient colostrum intake regarding the time or the volume of first colostrum feeding, or different combinations of these two factors. The major risk factor for FPT in the present study was a delay of more than 6 hours between birth and first colostrum feeding, reflecting the progressive decline of intestinal Ig absorption with time (Weaver et al., 2000; Godden, 2008). The volume of first colostrum feeding is also considered to be an important risk factor for FPT (Weaver et al., 2000; Godden 2008), which was confirmed in the present study although the decrease in OR for FPT after the feeding of 2 L colostrum or more at first feeding vs. lower volumes was just over the significance level (p = 0.053). The lesser effect of volume as compared with time of first colostrum feeding may be due to the fact that not only the quantity (volume) of colostrum fed but also its quality (concentration of Ig in the colostrum) influence the final serum Ig concentration of the calves. To minimize the risk of FPT, a calf has to ingest at least 100 g of Ig. In case of poor colostrum quality, the risk of FPT is reduced if a higher volume is fed at first feeding, whereas in case of high colostrum quality the volume of the first feeding may be lower without deleterious effects on the Ig serum concentration of the calf (Godden, 2008). This may explain why prolonged time to the first colostrum feeding appears to be a more important risk factor for FPT than the volume fed.

The parity of the dam has been reported to influence colostrum quality, and significantly lower IgG concentrations have been described in the milk of cows in their first lactation as compared with older cows (Gulliksen et al., 2008). However, factors other than the age of the cow also influence the quality of colostrum, e.g. the volume of the first milking or the duration of the dry period (Godden, 2008). This may explain why the parity of the dam did not strongly influence the risk factor for FPT in the present study. The occurrence of serious birth problems requiring veterinary assistance as they were described as risk factors for FPT in the study of Beam et al., (2009) was an exclusion criteria in the present study because the target population was healthy dairy calves. This may be the reason why the minor birth complications (twin calvings, mild assistance by the owner) taken into consideration in the present study did not significantly influence the risk of FPT.

The serum Se concentrations measured in calves (26.8 μg/L) and cows (36.5 μg/L) in the present study were below reference values cited in the literature from European and North American sources (Scholz and Stöber, 2002; Guard, 2008; Maas and Valberg, 2009), but they were comparable to values reported in earlier studies on cows and calves in Switzerland (Mathis et al., 1983; Braun et al., 1991; Stocker et al., 1993). Mathis et al. (1983) determined a critical limit for the occurrence of WMD at a serum Se concentration below 20–30 μg/L. In the study of Stocker et al. (1993), the healthy control calves presented lower Se values (mean Se concentration 14.5 μg/L) than the diseased calves (mean Se concentration 29.1, 27.5 and 23.0 μg/L in three consecutive years). The reason for these surprising results was attributed to unreported treatment of sick calves with Se prior to referral to the clinic. In a third study, serum Se concentration was 10.4 (chronic mastitis) and 11.7 μg/L (WMD) in herds with higher prevalence of those diseases. In the control herds, the mean Se concentration was 17.7 μg/L, and 90% of the control animals had Se concentrations below 30 μg/L without showing clinical signs of Se deficiency (Braun et al., 1991). Authors in Europe (Zust et al., 1996; Hain et al., 2003, Serdaru et al., 2004), and in North America (Campbell et al., 1990) have described low Se concentrations in clinically healthy cattle. The Se content in the feed may vary considerably between different geographic regions, depending on weathering of Se-containing rocks into the soil (Combs, 2001). This fact may at least partially explain the differences observed between the results of studies conducted in Switzerland and in other countries. The present results support the opinion that Se reference ranges determined in a restricted geographical area are only valid locally (Braun et al., 1991; Hain et al., 2003). However, comparison of the present results with values from earlier studies in Switzerland (Mathis et al., 1983; Braun et al., 1991; Stocker et al., 1993) indicate that no substantial improvement has occurred in the supply of adequate amounts of Se to Swiss cattle in the last 20 years.
The positive correlation between the serum Se concentration of the calves and their dams can be explained by the efficient transfer of Se across the placental membranes from the dam to the calf during late pregnancy which takes place even if the cow is Se deficient (Campbell et al., 1990). Transfer of Se to colostrum and milk also occurs and may influence the calf’s serum Se concentration, but only low effective amounts of Se are ingested with milk, which are not of nutritional importance for a deficient calf (Campbell et al., 1990). This is confirmed by the present results as no significant association was found between the volume of colostrum fed and the serum Se values of the calves. Likewise, no significant correlation was observed between the serum γG and Se concentrations of the calves. This is in apparent contradiction with the results of an experimental study (Swecker et al., 1995) where calves of cows supplemented with Se had higher IgG concentrations than calves of non-supplemented cows. These authors postulated that dams receiving adequate amounts of Se may produce more colostrum or that calves with adequate Se levels may be more active and consume more colostrum (Swecker et al., 1995). In the present study, Ig content of the colostrum was not determined, and the fact that multiple factors beside the Se status of the dam and the calf influence the γG concentration of the calf may explain this discrepancy. The strong correlation between the serum Se concentrations of the calves and their dams underline the importance of an adequate Se supply of the dams, especially during late gestation (Campbell et al., 1990; Enjalbert et al., 1999).

In conclusion, the evaluation of the γG and Se status in healthy neonatal Swiss dairy calves revealed an unexpected high percentage (43%) of calves with FPT. The questionnaire showed that only 30% of the calves received colostrum according to good practice guidelines (≥ 2 L in ≤ 2 h), and most of the farmers seem not to consider the second feeding of the calf as a relevant part of colostrum supply to neonatal calves. Serum Se concentrations observed in the present study were barely higher than those reported in former investigations in Switzerland (Mathis et al., 1983; Braun et al., 1991; Stocker et al., 1993), reflecting continued lack of farmers’ awareness for the importance of Se for the health of their animals. These results show that further efforts are warranted to draw the attention of dairy farmers to the benefits of good colostrum management for newborn calves and adequate Se supply on the health status of their animals.

Acknowledgements

We thank the Swiss Association for Ruminant Health (SVW) for supporting this study with the SVW-Research Grant 2008 and all participating veterinarians for collecting blood samples.

correlazione. I risultati sottolineano l’importanza di informare i proprietari sull’importanza dell’apporto di colostro e di selenio per la salute dei vitelli e delle mucche.

References


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Received: 6 December 2011
Accepted: 23 January 2012