Effect of the first and second postpartum partial milking on blood serum calcium concentration in dairy cows

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ABSTRACT: Parturient paresis and subclinical hypocalcemia are frequent metabolic disorders in dairy cows postpartum. The aim of this study was to determine the effect of postpartum partial milking in the first two milkings on blood serum calcium concentration in dairy cows. Twenty multiparous Holstein dairy cows were randomized into two groups. Cows of group 1 (n = 10) were partially milked at the first and second milking postpartum. Cows of group 2 (n = 10) were completely milked. Blood samples were collected from all animals 5–7 days before calving, within 30 min after calving, and 4, 8, 12, 16, 20, 24, 28, and 32 h after calving for determination of serum calcium (Ca), phosphorus (P), and magnesium (Mg) concentrations. Colostrum production was registered and sampled in the first and second milking. Concentration of Ca in colostrum was determined by atomic absorption spectrophotometry. Serum Ca and P concentrations decreased in both groups after parturition ($P < 0.05$) and remained low during 32 h postpartum with no difference observed between groups ($P > 0.05$). Serum concentrations of Mg were stable in all samples and no statistical difference was observed between groups ($P > 0.05$). Colostrum production was higher in completely milked cows only in the first postpartum milking ($P < 0.05$), but there was no difference between groups at the second milking. Total Ca secretion in colostrum was higher in the complete milking group at the first and second postpartum milking. Colostrum Ca secretion increased at the second milking with respect to the first one in both groups ($P < 0.05$). There was no correlation between serum Ca and colostrum Ca ($P > 0.05$). In this study, the partial milking of colostrum in the first and second milking postpartum did not prevent subclinical hypocalcemia in dairy cows.

Keywords: incomplete colostrum milking; subclinical hypocalcemia; milk fever; magnesium; postpartum cow

Parturient paresis and subclinical hypocalcemia are frequent metabolic disorders in the first days postpartum which predispose dairy cows to other diseases (Goff, 2008; Aparicio-Cecilio et al., 2012). The prevalence of parturient paresis in dairy cows in the United States was 4–14%, averaging 9.0% (Curtis et al., 1985). The direct annual cost for treating parturient paresis in this country makes 15 million dollars and its secondary complications generate an additional 120 million cost (Curtis et al., 1983).

Subclinical hypocalcemia is more frequent than the clinical form and occurs in over 60% of cows during the first 24 h after calving since the beginning of the parturition process coincides with the secretion of colostrum, which contains 2.3 g of calcium/kg on average. A high-production cow produces 10–20 kg of colostrum in the first 24 h after calving with 23–46 g of calcium being secreted through it, which represents an amount eight times higher than that of the entire plasma pool (Goff et al., 2000). This mineral mobilization is performed by the mechanisms that regulate serum Ca which, when they are not efficient, cause parturient paresis. Air inflation into the mammary

Supported by the National Autonomous University of Mexico (Project PAPIIT IN216409).

gland of cows with parturient paresis increases intramammary pressure decreasing colostrum production and Ca secretion and leads the cow to regain its muscle ability (Niedermeier and Smith, 1950). However, this measure is not practical, since it is difficult to perform and, moreover, it increases the risk of intramammary infections. Experimentally, cows that have undergone mastectomy do not present hypocalcemia at calving, indicating that the decrease in serum Ca concentration is caused by the production of colostrum rather than by the endocrine changes associated with calving (Goff et al., 2002). It has been observed in previous studies of our research that when the group of cows (n = 10) was partially milked only in the first milking (3.0 l of colostrum), serum Ca concentration was not different from that of completely milked cows. In another study, it was observed that primiparous cows secrete the same amount of Ca through colostrum as multiparous cows and this was slightly positively correlated with serum Ca (Salgado et al., 2013).

An important factor that must be considered in parturient paresis is the sudden rate of Ca secretion to colostrum. Presumably, because colostrum synthesis begins about a week before calving, the amount of Ca that is secreted through the first milking postpartum passes to the mammary gland at a slower rate compared to the amount of Ca that passes to the gland for the synthesis of the second milking colostrum; this could be a major factor involved in postpartum hypocalcemia. Our hypothesis was that the partial milking could decrease calcium secretion to colostrum and may reduce cases of postpartum hypocalcemia. The aim of this study was to determine the effect of the first and second postpartum partial milking of colostrum on serum Ca concentrations in dairy cows.

MATERIAL AND METHODS

Animals and treatments. This study was realized in a commercial farm (1800 cows) in La Laguna, near the city of Torreón, northern region of Mexico. Twenty multiparous Holstein cows, clinically healthy, with body condition score between 3.50 and 3.75 prior to calving, were randomized into two groups (n = 10). Total milk production in the previous lactation of these cows (9824 ± 698 kg vs. 9864 ± 782 kg), (P = 0.9) and dry period duration (60.3 ± 3 vs. 60.7 ± 3.2 days), (P = 0.77) were similar in groups 1 and 2, respectively. The cows before calving were maintained in a pen, where they received a total mixed ration twice a day and water *ad libitum* (National Research Council, 2001) (Table 1). All the animals were observed daily during seven days before calving and only cows with normal calving were included in the study. After calving, the animals were transferred to another pen for the fresh cows. Between 3–4 h after calving, cows of group 1 were partially milked at the first milking postpartum, the second partial milking was performed 12 h after the first one. Partial milking was realized by removing the milking machine when the colostrum volume reached approximately 3 l at the first milking and 4 l at the second milking. At the same time the cows of group 2 were completely milked. The cows

Table 1. Ingredients and chemical composition of prepartum and postpartum diets in cows

Ingredient	Prepartum	Postpartum
Alfalfa hay (%)	8.35	6.12
Ground corn (%)	13.14	18.50
Soybean meal (%)	0.55	–
Oats hay (%)	5.11	–
Corn silage (%)	71.2	43.00
Water (%)	–	10.00
Alfalfa silage (%)	–	5.62
Cottonseed meal (%)	–	4.05
Sugarcane molasse (%)	–	3.52
Cotton seed (%)	–	2.51
Canola meal (%)	–	4.68
Vitamin and mineral premix (%)	1.65	2.00
Chemical composition		
Moisture (%)	55.30	51.20
CP (%)	12.02	16.42
NDF (%)	35.81	34.32
NE_L (Mcal/kg)	1.48	1.56
Ca (%)	0.75	0.82
P (%)	0.35	0.37
Mg (%)	0.22	0.25
K (%)	1.30	1.52
Na (%)	0.10	0.29
S (%)	0.48	0.35
Cl (%)	0.35	0.37
DCAD (mEq/kg DM)	98	279

CP = crude protein, NDF = neutral detergent fibre, NE_L = net energy for lactation, DCAD = dietary cation-anion difference calculated using the formula: (Na + K) – (Cl + 0.6 S) (in milliequivalents per kg dry matter) were observed three times daily during 3 days after calving to detect the presence of retained fetal membranes, clinical signs of hypocalcemia, and other health problems; the temperature was measured by rectal thermometer once a day during the same period.

Sampling and analyses. Blood samples were taken from all the cows by tail vein puncture in 7 ml vacuum tubes without anticoagulant (Monoject®; Agtech, Inc., Manhattan, USA) 5–7 days before the estimated calving date, within 30 min after calving, and at 4, 8, 12, 16, 20, 24, 28, and 32 h after calving. The separation of clot from serum was performed within 1 h after sampling by centrifuging at 1200 *g* for 10 min. Serum samples from each cow were preserved in plastic vials (Eppendorf, Hamburg, Germany) at –20°C until analysis.

The serum concentrations of Ca, P, and Mg were determined colourimetrically (Randox Laboratories Ltd., Crumlin, UK) in a semi-automatic analyzer (model Junior; Vital Scientific, Spankeren, the Netherlands). Analyses were performed in the laboratory of Clinical Pathology, Faculty of Veterinary Medicine and Husbandry (FVMH) of the National Autonomous University of Mexico (UNAM). The volumes of colostrum from each cow at the first and second milking were registered and Ca was determined in samples by atomic absorption PerkinElmer spectrometer, model AAnalyst 200 (PerkinElmer, Waltham, USA), at the Animal Nutrition and Biochemistry Laboratory, FVMH, UNAM.

Statistical analysis. Serum Ca, P, and Mg were evaluated separately by the Multivariate Analysis of Variance (MANOVA) repeated measures design using SPSS software (Version 10.0.1, 1999). The model included the effect of type of milking and interaction of time × type of milking. Serum Ca, P, and Mg were the dependent variables, type of milking was the independent variable, and times of sampling were the repeated measures. Data normality was previously determined with Shapiro-Wilk test and covariance homogeneity by Mauchly's criterion (Myers, 1972). A pairwise comparison adjusted by Bonferroni was realized to determine main effects. Colostrum production, colostrum Ca concentration, and total colostrum Ca secretion were evaluated by the Analysis of Variance (ANOVA) for a 2 × 2 factorial design; this model included the effects of interaction of the order of milking × type of milking, order of milking, and type of milking to assure differences in partial and complete and the first and second milking.

RESULTS AND DISCUSSION

Neither the effect of time × type of milking interaction (P = 0.76) nor the effect of the type of milking (P = 0.86) on serum Ca concentration was detected. The effect of time (P = 0.04) was manifested in both groups, serum Ca concentration decreased at partum and remained low postpartum with respect to prepartum value. No cases of clinical parturient paresis were observed. In recent study, realized by Reinhardt et al. (2011), prevalence of subclinical hypocalcemia was 41, 49, and 51% for cows of the 2nd, 3rd, and 4th lactation, respectively. In that study, blood samples were taken in the first 48 h post calving. In our study, the prevalence of subclinical hypocalcemia was 70–80%, the samples were taken at 4-hour intervals until 32 h post calving and probably some

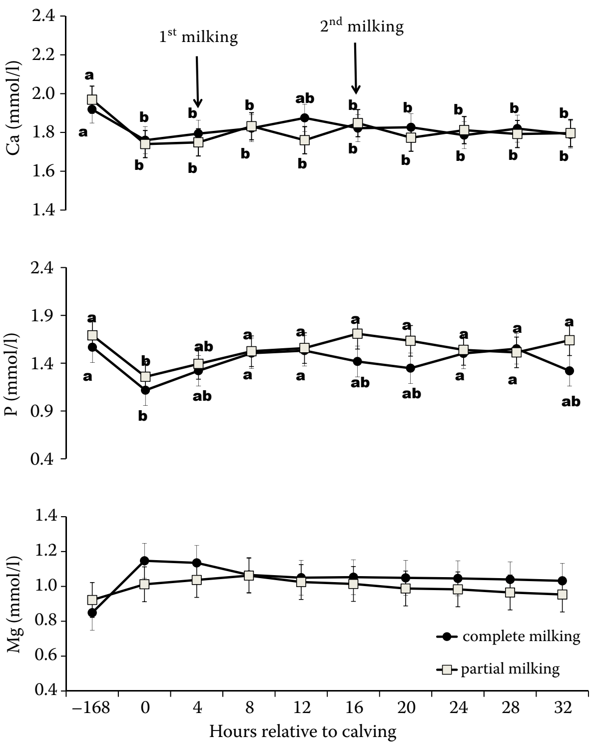

Figure 1. Serum concentration of Ca, P, and Mg (mmol/l) before and after calving in Holstein cows completely or partially milked during the first and second postpartum milking

There was no effect of interaction of milking type × time for Ca (P = 0.76), P (P = 0.13), and Mg (P = 0.23). There was no effect of milking type for Ca (P = 0.86), P (P = 0.58), and Mg (P = 0.58). There was an effect of time on Ca (P = 0.04) and P (P = 0.01). For Mg there was no effect of time (P = 0.32). Means ± SD
[a,b]statistical difference between means adjusted by Bonferroni at $P < 0.05$

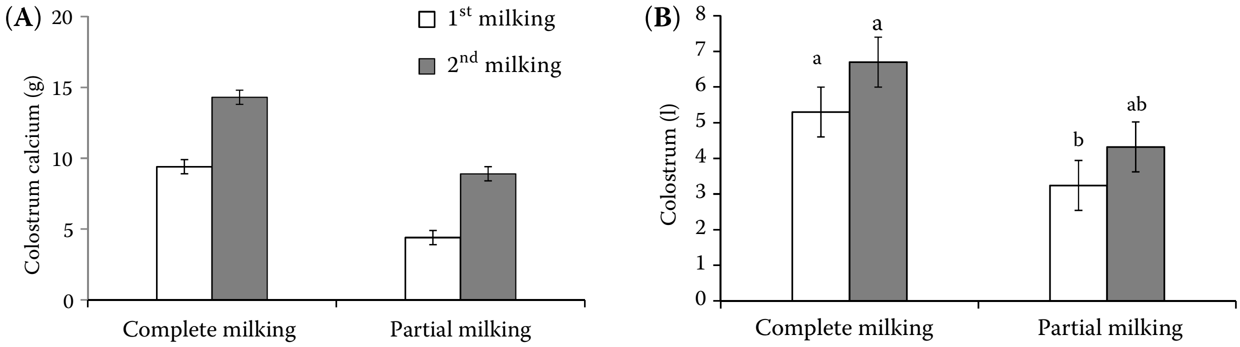

Figure 2. Calcium secretion (**A**) and colostrum production (**B**) in Holstein cows completely or partially milked during the first and second milking postpartum

There was an effect of interaction between milking order and type of milking for colostrum Ca (P = 0.002) and for colostrum volume (P = 0.01). Means ± SD

[a–c]statistical difference between means adjusted by Bonferroni at $P < 0.05$

cows needed more time to normalize calcemia. The prepartum diet in this study was high in Ca and there was no use of anionic salts, these factors could affect the adaptation mechanism to regulate the calcemia after calving. In the study of prevalence of sublinical hypocalcemia (Reinhardt et al., 2011), there is no mention on the composition of the prepartum diet.

In the concentration of serum P, there was neither the effect of time × type of milking (P = 0.13) interaction, nor the effect of type of milking (P = 0.58), but there was the effect of time (P = 0.01). In both groups, the serum P decreased at partum and then increased to the same concentration like prepartum (Figure 1). The decrease in P and Ca concentrations occurred simultaneously. This is because there is an exchange mechanism stimulated by parathyroid hormone, which increases the removal of P and elevates Ca retention by kidney in order to increase serum Ca concentrations when hypocalcemia is detected. In addition, there is secretion of Ca and P to colostrum (Kojouri, 2003). These results agree with the data from Smith et al. (1948) who found that totally milked cows in the first 5 days postpartum and supported by intravenous oxytocin at each milking, had serum Ca similar to that of partially milked cows. However, in their study the amount of Ca that was secreted via colostrum or colostrum produced in each group of cows was not determined. Another study by Aslam and Tucker (1988) found that plasma concentrations of Ca were more stable after each milking in cows that received 1.6 g of Ca by intramammary infusion, compared to those infused with 40 ml of distilled water. In this study, the production of colostrum was higher in cows with more stable plasma Ca concentrations associated with the intramammary Ca infusion. Aslam and Tucker (1988) concluded that the amount of colostrum and the amount of Ca secreted in the colostrum are not the only factors that determine serum Ca concentration postpartum and the same was found in our study.

Magnesium concentration was within the reference interval (0.78–1.07 mmol/l), representing adequate magnesium nutritional status. There was no effect of time × type of milking (P = 0.23) interaction and no difference was found between groups (P = 0.58) and time (P = 0.32) effects (Figure 1). According to Goff et al. (2008), if the serum concentration of Mg decreases during hypocalcemia, this is an indicator of hypomagnesemia, which predisposes the cow to hypocalcemia. This was not a problem in our cows and in most cows this disbalance can be easily corrected by adding adequate available Mg to the diet.

Colostrum production at the first milking was greater in cows milked completely (5.3 l) than in partially milked cows (3.2 l) (P = 0.01). At the second milking, there was no difference in the volume of colostrum between complete (6.7 l) and partial (4.3 l) milking (P = 0.12) (Figure 2). Cows that were completely milked during the first two milkings lost via colostrum 24 g of Ca from the body on average. However, partially milked cows reduced Ca loss to 14 g, they did not improve serum Ca concentration significantly. It is possible that Ca was still sequestered from the blood into the mammary gland. Mastectomized cows (Goff et al., 2002) suffer no decline in blood Ca at calving, perhaps because there is no sequestration of Ca in the mammary gland as the mammary gland has been removed.

Total Ca secretion in colostrum was higher in completely milked cows compared with partially milked cows ($P = 0.02$). In both groups, Ca secretion in colostrum increased by the second milking with respect to the first one ($P < 0.03$) (Figure 2). Probably, increased pressure in the mammary gland of the partially milked cows may reduce colostrum production and calcium secretion, but the udder pressure was not measured. Total Ca secretion at the first and second milking did not correlate to serum Ca concentration at any particular time. These results contrast with those obtained by Goff et al. (2002) in which hypocalcemia was not observed in the early days postpartum in mastectomized cows, compared with cows with intact mammary gland producing colostrum normally. In that study, Goff et al. (2002) conclude that it is the onset of colostrum production, rather than endocrine changes associated with calving, which trigger hypocalcemia. In addition, that study did not consider the mobilization of energy needed for milk production. Such changes could negatively affect the copious mechanisms to maintain adequate physiological concentrations of Ca, as suggested by the study of Heuer et al. (1999), which shows that cows with high body condition are at 3.3 times higher relative risk of postpartum hypocalcemia. Another study by Reinhardt et al. (2011) shows that cows with hypocalcemia postpartum have higher serum concentrations of non-esterified fatty acids at the same time but it does not mention which event happens first, so further study is needed. On the other hand, in a study conducted by Wu et al. (2008), cows with serum Ca higher in early postpartum also showed increased secretion of Ca in colostrum. It has been observed that in humans and rats the amount of Ca in colostrum depends on the serum Ca concentration (VanHouten et al., 2004).

In this study, there was a slight positive relationship between Ca secretion in colostrum and serum Ca. In a study in cows milked before calving, a lower incidence of parturient paresis was not observed compared with cows of the control group (Smith et al., 1947), which indicates that reducing the amount of Ca that exits through the mammary gland by partial milking does not determine the degree of hypocalcemia presented in dairy cows.

CONCLUSION

In this study, the serum concentration of Ca did not increase in any of the two groups of cows, and there was no relationship between serum Ca and the Ca secreted via colostrum, so it is concluded that partial milking of colostrum, both at the first and second milking postpartum, does not prevent hypocalcemia in the early hours postpartum.

Acknowledgement. Gabriel Salgado thanks to CONACyT Mexico for scholarship. The authors are indebted to Prof. J.P. Goff, D.V.M., Ph.D. for revision of the manuscript.

REFERENCES

Aparicio-Cecilio A., Bouda J., Salgado-Hernández E.G., Núñez-Ochoa L., Castillo-Mata D.A., Gutiérrez-Chávez A. (2012): Effect of 2-methyl-2-phenoxy propionic acid on serum lipid profile and ovarian activity in dairy cows. Czech Journal of Animal Science, 57, 550–556.

Aslam M., Tucker W.B. (1998): Influence of intramammary infusion of calcium on the calcium status of periparturient lactating dairy cows. Journal of Dairy Science, 81, 1883–1888.

Curtis C.R., Sniffen C.J., Smith R.D., Powers P.A., Smith C., White M.E., Hillman R.B., Pearson E.J. (1983): Association of parturient hypocalcemia with eight periparturient disorders in Holstein cows. Journal of American Veterinary Medical Association, 183, 559–561.

Curtis C.R., Erb H.N., Sniffen C.J., Smith R.D., Kronfeld D.S.(1985): Path analysis of dry period nutrition, postpartum metabolic and reproductive disorders, and mastitis in Holstein cows. Journal of Dairy Science, 68, 2347–2360.

Goff J.P. (2000): Pathophysiology of calcium and phosphorus disorders. Veterinary Clinics of North America: Food Animal Practice, 16, 319–337.

Goff J.P. (2008): The monitoring, prevention and treatment of milk fever and subclinical hypocalcemia in dairy cows. The Veterinary Journal, 171, 50–57.

Goff J.P., Kimura K., Horst R.L. (2002): Effect of mastectomy on milk fever, energy, and vitamins A, E, and β-carotene status at parturition. Journal of Dairy Science, 5, 1427–1436.

Heuer C., Schukken Y.H., Dobbelaar P. (1999): Postpartum body condition score and results from the first test day milk as predictors of disease, fertility, yield, and culling in commercial dairy herds. Journal of Dairy Science, 82, 295–304.

Kojouri G.H.A. (2003): Parturient paresis and its relationship with hypophosphatemia (abstract). Acta Veterinaria Scandinavica. Supplementum, 98, 303.

Myers J.L. (1972): Fundamental of Experimental Design. 2nd Ed. Allyn and Bacon, Boston, USA.

Niedermeier R.P., Smith V.R. (1950): Parturient paresis IV: The effect of udder inflation upon blood levels of calcium, magnesium and phosphorus in cows with parturient paresis. Journal of Dairy Science, 33, 38–42.

National Research Council (2001): Nutrient Requirements of Dairy Cattle. 7th Ed. National Academies Press, Washington, USA.

Reinhardt T.A., Lippolis J.D., McCluskey B.J., Goff J.P., Horst R.L. (2011): Prevalence of subclinical hypocalcemia in dairy herds. Journal of Dairy Science, 88, 122–124.

Salgado-Hernández E.G., Bouda J., Villa-Godoy A., Romano-Muñoz J.L., Gutiérrez-Chávez J.A., Velásquez-Forero F.H. (2014): Metabolites of vitamin D and minerals in blood and colostrum of primiparous and multiparous dairy cows postpartum. Czech Journal of Animal Science, 59, 11–18.

Smith V.R., Blosser T.H. (1947): Parturient paresis I: The incidence of parturient paresis and changes in the total blood serum calcium at parturition in prepartum milked cows. Journal of Dairy Science, 30, 861–866.

Smith V.R., Niedermeier R.P., Hansen R.G. (1948): Parturient paresis II: The effect of partial versus complete milking upon the total blood serum calcium on dairy cows at parturition. Journal of Dairy Science, 31, 173–177.

VanHouten J., Dann P., McGeoch G., Brown E.M., Krepcho K., Neville M,. Wysolmerski M.M. (2004): The calcium-sensing receptor regulates mammary gland parathyroid hormone-related protein and calcium transport. Journal of Clinical Investigation, 13, 598–608.

Wu W.X., Liu J.X., Xu G.Z.,Ye J.A. (2008): Calcium homeostasis, acid-base balance, and health status in periparturient Holstein cows fed diets with low cation-anion difference. Livestock Science, 117, 7–14.

Received: 2013–07–29
Accepted after corrections: 2013–12–20

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