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Incidence of hypocalcemia and its changes of biochemical parameters in periparturient cows

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Abstract : In this study, we investigate the status of calcium (Ca) homeostasis at parturition in three dairy farms (I, II, and III), Heilongjiang, China. Twenty multiparous Holstein cows from each farm were randomly assigned to this experiment. The dietary cation-anion difference (DCAD) was 91 mEq/kg of DM for farm I, 152 mEq/kg of DM for farm II, and 85 mEq/kg of DM for farm III. Incidence of hypocalcemia was above 75% and urine pH was above 7.25 at calving in each farm. Compared to other farms, cows in farm II that fed the greatest positive DCAD had the lowest concentration of serum Ca, the highest concentration of serum PTH, and the greatest urine pH at calving ($p < 0.05$). However, there was not significant difference in serum 1,25-dihydroxy-vitamin D and hydroxyproline concentration of the cows among three farms. This is the first study to confirm that hypocalcemia is very prevalent at calving in Chinese dairy farms, and the high positive DCAD is a major risk factor that results in hypocalcemia at calving, which may reduce ability of the cow to maintain Ca homeostasis.

Keywords : Ca homeostasis, cation-anion difference, dairy cows, hypocalcemia, urine pH

Milk fever is a major metabolic disease that calcium (Ca) homeostatic mechanisms fail to maintain normal blood Ca levels around calving. Evaluation of Ca homeostatic mechanisms has increasingly contributed to the understanding of the fundamental mechanisms involved in milk fever [1]. However, there have been few reports about the Ca homeostatic capacity of dairy cows with hypocalcemia when calving in China. This study is a survey to status of hypocalcemia and levels of dietary cation anion difference (DCAD), urine pH and serum Ca, hydroxyproline (HYP), alkaline phosphatase (ALP), parathyroid hormone (PTH), calcitonin (CT), 1,25-dihydroxy-vitamin D (DHVD) in three dairy farms, Heilongjiang province, China.

Tail venous blood and urine were collected using 10 mL tube and 50 mL cup respectively from twenty healthy cows within 12 h after calving in each dairy farm. The cows were Holstein breed with three to six years of age, fed the total mixed ration, and had the similar milk yield. The blood was centrifuged immediately after collection at 4,000 g for 10 min and serum was harvested. Serum Ca and HYP were determined using commercial kits (Chang Chun HuiLi Bioengineering, China). Serum concentration of PTH was measured at Nucleo-Radiology Department of Harbin Medical University in China using a commercially available radioimmunoassay kit (Beijing Atomic High Technology, China). Serum concentrations of DHVD were measured using a high performance liquid chromatography at Product Quality Monitoring Institute in Changchun, China. Serum Ca concentrations less than 2.20 mmol/L were considered to hypocalcemia [5]. Urine pH was detected using a precise pH test paper (Uritest 8A; Youlite Medical Electronic, China).

Feed from each dairy farm was collected at prepartum 10 days by quatation method. The DCAD was calculated using the formula DCAD (mEq/kg of DM) = [(Na/23 + K/39)-(Cl/35.5 + S/16)] [3].

Student's *t* test was used for comparison of means. The data were expressed as mean ± SD and statistical significance was established at $p < 0.05$.

In the three farms, survey showed incidence of hypocalcemia, DCAD and urine pH were above 75%, 85 mEq/kg of DM and 7.25 at calving, respectively.

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Table 1. Status of hypocalcemia and DCAD & changes of seven parameters in dairy cows at 12 h after calving

Parameter	Farm I	Farm II	Farm III
Ca (mmol/L)	1.91 (0.22)[b]	1.82 (0.23)[a]	2.01 (0.18)[b]
ALP (U/L)	73 (31)	58 (20)	79 (26)
HYP (μg/mL)	2.63 (0.21)	2.31 (0.23)	2.52 (0.24)
PTH (*p*mol/L)	192 (35)[b]	260(40)[a]	220 (33)[b]
CT (*p*mol/L)	36 (10)	39 (8)	34 (9)
DHVD (ng/mL)	26 (8)	27 (7)	25 (6)
pH	7.50 (0.25)[b]	8.00 (0.25)[a]	7.25 (0.25)[b]
DCAD (meq/kg of DM)	91[b]	152[a]	85[b]
Incidence of hypocalcemia (%)	80	80	75

[a,b]Values superscripted by the different lower-case letters ($p < 0.05$) between three farms. ALP: alkaline phosphatase, Ca: calcium, CT: calcitonin, DCAD: dietary cation-anion difference, DHVD: 1,25-dihydroxyvitamin D, HYP: hydroxyproline, PTH: parathormone.

Dairy cows in farm II had the highest DCAD ($p < 0.05$), urine pH and serum PTH concentration ($p < 0.05$), still had the lowest serum Ca concentration ($p < 0.05$). Level of serum CT, DHVD, ALP, and HYP was not significantly different among three farms (Table 1).

Some studies suggested that DCAD before calving should be under 40 mEq/kg of DM to reduce incidence of hypocalcemia (less than 50%), maintain normal urine pH (6.5~7.0) and blood Ca (more than 2.20 mmol/L) [2, 4]. The high positive DCAD before parturition, resulted from high content of dietary K or Na, is an important etiologic factor of milk fever [1, 5]. The high dietary Na or K result in a state of metabolic alkalosis, which reduces the responsiveness of bone and kidney to PTH [2, 6]. It has been known that PTH is the major factor that controls bone osteoclast activity. When cows have a negative Ca balance which stimulates secretion of PTH, the secreted PTH enhances bone Ca resorption, and activates renal tubules to produce DHVD during transition period resulting in elevated serum Ca, HYP and ALP usually ought to increase [3, 7]. In the present study, cows have a lower concentration of serum Ca and increased secretion of PTH, especially in farm II, which suggests that response of PTH to low Ca is active to prevent a severe decline of serum Ca. However, there is no difference in serum ALP, HYP, and DHVD among three farms, which implies that the bones and kidney of the cows fed the high positive DCAD may be refractory to PTH stimulation. Thus, further research is needed to elucidate the detailed reason for so high incidence of hypocalcemia.

In conclusion, the high incidence of hypocalcemia was a serious problem in three farms, which may be associated with high positive DCAD. The high DCAD is a major risk factor for hypocalcemia before calving that may induce metabolic alkalosis and then reduce the ability of the cow to maintain Ca homeostasis.

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