



J. Dairy Sci. 101:1–10  
<https://doi.org/10.3168/jds.2017-14202>  
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## Association of postpartum hypocalcemia with early-lactation milk yield, reproductive performance, and culling in dairy cows

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### ABSTRACT

Periparturient hypocalcemia is frequently observed and considered as a gateway disease that is associated with various health issues. The objective of this study was to evaluate the association of hypocalcemia with early-lactation milk yield, reproductive performance, and culling across a large number of different managerial systems. A prospective cohort study was conducted based on a convenience sample of 125 dairy herds from 8 federal states of Germany between February 2015 and August 2016. A blood sample was drawn from 1,709 animals within 48 h after parturition and analyzed for serum calcium concentration. After discarding cows ( $n = 283$ ) with missing data, a total of 1,426 cows were considered for final analyses. The median time from calving to sampling was 14.0 h (interquartile range = 5.0–24.9 h). For each herd, a record of the herd management software was requested 150 d after the last cow was sampled. Serum calcium concentration of each cow was associated with early-lactation milk yield (Dairy Herd Improvement Association equivalent test 1 to 3), reproductive performance [days in milk (DIM) at first artificial insemination (AI), pregnancy at first AI, time to pregnancy within 150 DIM], and culling (until 60 DIM) data. Generalized linear mixed models were used to analyze continuous or categorical data. Shared frailty models were used for time to event data. Five different thresholds were used to define hypocalcemia. Thresholds ranged from 1.8 to 2.2 mmol/L using 0.1-mmol/L increments. Clinical hypocalcemia was defined as serum calcium concentration  $<2.0$  mmol/L in combination with clinical signs (e.g., recumbency). The effect of hypocalcemia on milk yield was conditional on parity. In primiparous cows a serum calcium concentration  $<2.0$  mmol/L (6.4% of cows were below this threshold) had no effect on milk production, whereas there was a

tendency for multiparous cows with a serum calcium concentration  $<2.1$  mmol/L (63.2% of cows were below this threshold) to produce 0.80 kg/d more milk compared with multiparous cows at or above the threshold. Multiparous cows suffering from clinical hypocalcemia produced 2.19 kg/d less milk compared with normocalcemic cows in early lactation. Calcium status was not associated with days to first insemination. Cows with a serum calcium concentration  $<1.9$  mmol/L (34.6% of cows below this threshold) had decreased odds (odds ratio = 0.56) of pregnancy at first AI. A serum calcium concentration  $<1.8$  mmol/L (24.1% of cows below this threshold) had a significant effect on time to pregnancy. Compared with animals with a serum calcium concentration  $\geq 1.8$  mmol/L, the hazard of becoming pregnant within 150 DIM was reduced when cows had a serum calcium concentration  $<1.8$  mmol/L (hazard ratio = 0.68). Cows with a serum calcium concentration  $<2.0$  mmol/L (44.3% of cows were below this threshold) had a 1.69 times greater hazard of being culled within the first 60 DIM compared with normocalcemic animals. The present study shows that the association of hypocalcemia with milk yield was conditional on parity and serum calcium concentration measured once within 48 h after calving. Considering reproductive performance and culling in early lactation, a negative effect of postpartum hypocalcemia was demonstrated.

**Key words:** hypocalcemia, milk fever, reproduction, dairy cow

### INTRODUCTION

Transition cows face the challenge of an increased requirement for minerals, especially calcium, to support lactogenesis in early lactation (Goff, 2014). To compensate for this challenge, homeorhetic mechanisms take place to adapt to such increased demands (Martín-Tereso and Martens, 2014). Unsuccessful adaptation leads to hypocalcemia around parturition and has been associated with increased occurrence of diseases (Martinez et al., 2012), decreased milk production (Chapinal

Received November 27, 2017.

Accepted May 22, 2018.

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et al., 2012b), increased culling risk (Seifi et al., 2011; Roberts et al., 2012), and impaired reproductive performance (Martinez et al., 2012; Ribeiro et al., 2013; Caixeta et al., 2017). Megahed et al. (2018) demonstrated that serum calcium concentration decreased 9 h before calving and increased back to the reference range approximately 72 h after calving. Although negative associations of hypocalcemia with production and health were not investigated in this study, it suggests that the major risk period for hypocalcemia might be within this time frame. Hypocalcemia can be either clinical (i.e., recumbency) or subclinical. With a better understanding of the hormonal regulation of calcium mobilization in the periparturient cow (Goff, 2008) and appropriate nutritional management (e.g., DCAD) during the dry period and early lactation, incidence rates of clinical hypocalcemia can be as low as 1% (Oetzel and Miller, 2012). In a recent multisite study comprising 115 herds in Germany, however, the incidence of clinical hypocalcemia averaged 7.2% (Venjakob et al., 2017).

Subclinical hypocalcemia has been defined using thresholds ranging from 2.0 to 2.3 mmol/L (Seifi et al., 2011; Chapinal et al., 2012b; Roberts et al., 2012; Wilhelm et al., 2017). Commonly, the threshold used to categorize a cow as hypocalcemic has been  $\leq 2.0$  mmol/L and has often been applied in studies (DeGaris and Lean, 2008; Reinhardt et al., 2011; Wilhelm et al., 2017). The origin of this threshold, however, was arbitrary, and recent studies have reported higher thresholds. It was shown that hypocalcemia using the thresholds  $\leq 2.1$ ,  $\leq 2.2$ , or  $\leq 2.3$  mmol/L was associated with a negative health outcome such as displaced abomasum and metritis (Chapinal et al., 2011, 2012b; Martinez et al., 2012) or an increased culling risk (Seifi et al., 2011; Roberts et al., 2012). These studies, however, considered a longer risk period postpartum (i.e., 3–7 DIM).

The association of hypocalcemia with milk production is inconsistent in the literature. Evidence exists that hypocalcemia around parturition is associated with decreased milk yield (Chapinal et al., 2012b). Another study does not show an effect of hypocalcemia on milk production (Martinez et al., 2012). In contrast, some studies even suggest that cows suffering from subclinical hypocalcemia produce more milk during early lactation (Jawor et al., 2012; Gild et al., 2015) compared with normocalcemic cows.

It has been shown consistently that the risk of culling during the first 60 d of lactation was greater when serum calcium concentration was  $\leq 2.2$  mmol/L in the first week postpartum [odds ratio (OR) = 1.5; Roberts et al., 2012]. Other authors found a 2.4 and 5.3 times greater risk for culling when serum calcium concentra-

tion was  $\leq 2.2$  and  $\leq 2.3$  mmol/L in the first and second week postpartum, respectively (Seifi et al., 2011).

Some epidemiological multisite studies indicate a negative association of hypocalcemia in the first week postpartum with health, production, and reproductive performance. Although these epidemiological studies showed an association and some evidence for a causal relationship between hypocalcemia and an increased risk for infectious diseases (Martinez et al., 2014), longer risk periods have to be evaluated with caution. It is also possible that reduced feed intake affects serum calcium levels before clinical signs of disease become apparent, as most recently shown by Pinedo et al. (2017) for cows suffering from puerperal metritis. Considering a relatively long risk period of 1 wk might lead to an underestimation of the prevalence and an overestimation of the effect size caused by hypocalcemia. Therefore, the objective of this prospective cohort study was to evaluate the association of hypocalcemia within 48 h after parturition with early-lactation milk yield, culling, and reproductive performance in German dairy herds.

## MATERIALS AND METHODS

### Study Population

A prospective cohort study was conducted based on a convenience sample of 125 dairy herds from 8 federal states of Germany between February 2015 and August 2016. Details and the sample size calculation have been described previously (Venjakob et al., 2017). In brief, inclusion criteria for herds were (1) participation in a federal DHIA equivalent testing system, (2) freestall housing with at least 100 milking cows, (3) feeding of a TMR-based diet, and (4) use of computerized herd management software. Average herd size was 513 and ranged from 112 to 2,607 lactating cows. The average milk production (305-d ECM, 4.0% fat, 3.4% protein) was 9,231 kg (range: 6,257–10,880 kg). Holstein Friesian cows were the predominant breed on 122 farms. Two farms kept Simmental cattle, and 1 farm kept Jersey cattle as the main breed.

### Experimental Procedures

Veterinary practitioners were invited to participate in the study by an information leaflet sent out by regular mail. Participating practices were informed about the nature and duration of the study and received a package containing blood serum collection systems (S-Monovette 9-mL Z, Sarstedt AG & Co, Nürnberg, Germany), cryo-vials (Cryoröhrchen, Carl Roth GmbH & Co. KG, Karlsruhe, Germany) to store serum at  $-20^{\circ}\text{C}$  until analysis, and a written standard operating

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