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Association of immediate postpartum plasma calcium concentration with early-lactation clinical diseases, culling, reproduction, and milk production in Holstein cows

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ABSTRACT

The objective of this study was to evaluate the association of postpartum plasma Ca concentration with early-lactation disease outcomes, culling within 60 d in milk, pregnancy to first service, and milk production. A total of 1,453 cows from 5 commercial dairy farms in New York State were enrolled in a prospective cohort study from February to November 2015. Blood samples were collected within 12 h of parturition, and plasma was submitted to a diagnostic laboratory for total Ca measurement. Early-lactation disease, reproductive performance, and milk production from Dairy Herd Improvement Association (DHIA) test-day data were compiled from each farm's management software. Multivariable Poisson regression models were built to evaluate the association of plasma Ca with the risks of retained placenta (RP), metritis, displaced abomasum (DA), clinical mastitis, culling within 60 d in milk, and pregnancy to first service. Repeated-measures ANOVA were used to evaluate the association of Ca at parturition with milk production across the first 9 DHIA tests. Herd was considered a random effect in all models. Primiparous cows were modeled separately from multiparous cows if differential responses were observed. Calcium was not associated with the risk of RP, metritis, clinical mastitis, or pregnancy to first service in primiparous or multiparous cows. For multiparous cows only, higher Ca concentration tended to be associated with increased culling within the first 60 d in milk. Multiparous cows with Ca ≤ 1.85 mmol/L had an increased risk of being diagnosed with a DA compared with cows with Ca > 1.85 mmol/L. For the milk production models, Ca was not associated with the amount of milk produced within the first 9 DHIA tests in primiparous cows; however, multiparous cows

with Ca ≤ 1.95 mmol/L produced, on average, 1.1 kg more milk per day across the 9 DHIA tests than their multiparous counterparts with Ca > 1.95 mmol/L. Our results indicate that plasma Ca concentration measured within 12 h of parturition is a poor predictor of early-lactation health outcomes. Reduced Ca concentration in the immediate postpartum period was associated with higher milk production in multiparous cows. From these results, we caution that studies attempting to categorize subclinical hypocalcemia based on a single sample in the immediate postpartum period could misclassify the disorder.

Key words: dairy cow, calcium, subclinical hypocalcemia, milk production

INTRODUCTION

Parturition marks a key period in the productive cycle of a dairy cow as it is characterized by sudden dietary and hormonal changes and social challenges. Consequently, early-lactation cows are at a high risk for developing metabolic diseases that can directly affect their lactation performance. Subclinical hypocalcemia (SCH; low blood Ca concentration without clinical signs of ataxia or paresis) is one of the metabolic disorders in early lactation that can detrimentally affect the productivity of the postparturient cow. Around 50% of recently calved multiparous cows are believed to suffer from the condition (Reinhardt et al., 2011).

Traditionally, SCH has been classified as serum or plasma total Ca concentration ≤ 1.87 mmol/L (7.5 mg/dL; Goff et al., 1996), ≤ 1.95 mmol/L (7.8 mg/dL; Massey et al., 1993), or < 2.0 mmol/L (8.0 mg/dL; Oetzel et al., 1988; Oetzel, 1996). The number of studies in this area is continually growing, with the latest reports using higher classification cut-points (Chapinal et al., 2011, 2012; Martinez et al., 2012). However, some studies have found strong associations of lower Ca concentration in the postpartum period with detrimental health and production outcomes, whereas others have

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shown no associations. In a large study across herds in the United States and Canada, cows with Ca concentrations ≤ 2.1 and ≤ 2.2 mmol/L (sampled within wk 1 of parturition) had decreased milk production and increased odds of displaced abomasum (**DA**), respectively (Chapinal et al., 2011, 2012). Martinez et al. (2012), using a categorization of SCH with Ca ≤ 2.14 mmol/L based on the lowest serum Ca concentration collected within 0 to 3 DIM, demonstrated that SCH was associated with metritis. Chamberlin et al. (2013), using a cut-point for SCH based on ionized Ca < 1.0 mmol/L (approximately equivalent to 2.0 mmol/L of total Ca) on the day of calving, found no associations between the condition and early-lactation disease outcomes. Differences in SCH classification between studies using Ca concentrations in blood samples collected at different time points relative to parturition may affect our ability to establish associations between Ca concentration and early-lactation outcomes. The literature currently lacks standardization of SCH classification, and there is no consensus as to when and how dairy consultants should more objectively assess SCH at the herd and cow levels.

If SCH classification could be accurately performed shortly following parturition, mitigation strategies could be better implemented and measured in an attempt to lessen the effect of the disorder in early lactation. We hypothesized that reduced Ca concentration in the immediate postpartum period would be associated with detrimental health and production outcomes. The objectives of our study were to evaluate the association of Ca concentration in plasma in the immediate postpartum period in a large, multi-herd observational trial with (1) early-lactation health outcomes, (2) culling within 60 DIM, (3) pregnancy to first service, and (4) milk production.

MATERIALS AND METHODS

A prospective cohort study was performed as part of a larger randomized clinical trial evaluating the effects of an oral Ca bolus supplement given shortly after calving on health and production outcomes. This study was approved by the Cornell University Institutional Animal Care and Use Committee (protocol 2014-0171). Prospective data obtained from cows that calved from February 21 to November 30, 2015, from 5 commercial dairy farms in New York State were used. A sixth farm was included in the randomized clinical trial; however, that farm did not comply with disease definition protocols adopted before the initiation of the study and therefore was not included in this cohort. For the remaining farms, control cows (i.e., no Ca supplementation) with

a blood sample taken within 12 h of parturition were eligible to be included in this study. Blood samples were collected by trained farm personnel via coccygeal venipuncture using a 20-gauge needle attached to a polypropylene syringe, transferred immediately into a 4-mL lithium heparin tube (Greiner Bio-One, Monroe, NC), and stored at 4°C. Samples were shipped to a central processing location twice a week for centrifugation ($1,000 \times g$ for 10 min at 22°C), plasma harvesting, and storage (-20°C) until further analysis. At the end of the trial, plasma samples were submitted to the University of Illinois Veterinary Diagnostic Laboratory (Urbana, IL) for total Ca analysis (intra- and inter-assay CV $< 1.0\%$) using a high-throughput chemistry analyzer (AU680, Beckman Coulter, Brea, CA), with reagents provided by the same manufacturer.

To detect a difference of at least 1.5 kg of milk per test-day with 95% confidence between the SCH and normocalcemic groups, and assuming a SD of 8.0 kg per test-day per group and a 40% prevalence of SCH, 1,000 cows were necessary to have a study with 80% power (OpenEpi version 3.01; OpenEpi, Atlanta, GA).

Total mixed ration samples were collected once weekly from the close-up dry cow group at each farm, composited at the end of the study, and submitted to a commercial laboratory for wet chemistry analysis (Cumberland Valley Analytical Services, Hagerstown, MD), with methods as described by McCarthy et al. (2015). Analyzed DCAD of the close-up diets for each farm are given in Table 1.

Farm personnel recorded disease event data in DairyComp 305 (Valley Agricultural Software, Tulare, CA) using standard disease definition protocols discussed before the start of the study for retained placenta (**RP**; failure to expel fetal membranes within 24 h of parturition), metritis (fetid reddish to brownish uterine discharge often accompanied by systemic signs of illness), DA, and clinical mastitis (presence of visibly abnormal milk). Health, production, and reproduction records were extracted from the herd management software into Microsoft Excel (Microsoft Corp., Redmond, WA) before statistical analyses.

Statistical Analyses and Model-Building Strategies

All statistical analyses were performed in SAS software (version 9.4, SAS Institute Inc., Cary, NC). Descriptive statistics were performed using the **FREQ**, **MEANS**, and **UNIVARIATE** procedures. Parity was categorized into a 4-level variable (first, second, third, and fourth or greater). Calving ease score was dichotomized to represent cows that required small to no assistance or suffered dystocia during calving. A 2-level

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