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Interaction of 5-hydroxy-L-tryptophan and negative dietary cation-anion difference on calcium homeostasis in multiparous peripartum dairy cows

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ABSTRACT

Hypocalcemia affects almost 50% of all dairy cows. Our laboratory has previously demonstrated that infusions of the serotonin precursor 5-hydroxy-L-tryptophan (5-HTP) increase circulating calcium concentrations in the Holstein transition cow. It is unknown whether feeding a negative DCAD diet alters the relationship between 5-HTP and hypocalcemia. The main objective of this study was to determine whether feeding a negative dietary cation-anion difference (−DCAD) diet before calving in conjunction with 5-HTP treatment could further diminish the magnitude of hypocalcemia at the time of calving. We used a randomized complete block design with a 2 × 2 factorial arrangement. Thirty-one multiparous Holstein cows were fed either a positive (+13 mEq/100 g) or negative (−13 mEq/100 g) DCAD diet 21 d before parturition and were intravenously infused daily with saline or 5-HTP (1 mg/kg) starting 7 d before the estimated date of parturition. Cows were blocked by parity and were randomly assigned to 1 of 4 treatment groups: positive DCAD plus saline, positive DCAD plus 5-HTP, negative DCAD plus saline, and negative DCAD plus 5-HTP, resulting in n = 8 per group. Total calcium (tCa), ionized calcium (iCa), and feed intake were recorded. The iCa was elevated prepartum in the −DCAD/5-HTP group compared with the other treatment groups as well as on d 0 and 1 postpartum. Although differences in tCa were not significant across the pre- or postpartum periods, tCa was numerically higher on d 0 and significantly higher on d 1 in −DCAD/5-HTP cows compared with all other groups. Prepartum the −DCAD/5-HTP treatment group ate less than the other treatment groups; however, postpartum dry matter intake differences were not significant. These findings demonstrate that feeding a −DCAD diet in conjunction with 5-HTP prepartum can increase postpartum circulating iCa concentrations and therefore diminish the magnitude of hypocalcemia at the time of parturition.
Key words: dietary cation-anion difference, serotonin, calcium

INTRODUCTION

The periparturient period is one of the most high-risk periods of the dairy cow's life. During this time, as much as 50% of dairy cows are estimated to experience some degree of hypocalcemia (Reinhardt et al., 2011), with an additional 5% of all dairy cows being diagnosed with clinical hypocalcemia (USDA, 2007). Hypocalcemia and subclinical hypocalcemia have been described as gateway diseases leading to increased rates of ketosis, fatty liver, metritis, retained placenta (**RP**), displaced abomasum, and mastitis as well as negatively affecting potential production and increasing the risk of involuntary culling in the dairy herd (Seifi et al., 2011; Chamberlin et al., 2013; Goff, 2014). It is estimated that more than $550 million is lost annually in milk production alone in the United States dairy industry due to hypocalcemia (Guard, 1996; Hare et al., 2006; USDA, 2017).

At parturition, the dam's rapidly increasing Ca demand exceeds the amount that she can consume to maintain Ca homeostasis. During late gestation, the dairy cow requires approximately 10 to 16 g of Ca per day. On d 1 of lactation, a cow loses approximately 23 g of Ca to colostrum production, 30 to 50 g of Ca per day within the first few days of lactation, and by peak production, Ca loss into the milk increases to roughly 80 g/d (Horst et al., 2005). Total blood calcium (**tCa**) of a healthy adult cow is maintained within the range of 2.1 to 2.5 mM (Goff, 2008) and it is estimated that the cow will deplete her Ca reservoir 7 to 10 times per day to meet the demand of peak milk production (Horst et. al, 2005). With increased Ca demands, the cow is required to increase absorption of Ca from the intes-

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tine, decrease loss of Ca into the urine, and increase mobilization of Ca from bone tissue to maintain Ca homeostasis, while also producing milk.

Increased attention to and control of dietary levels of several key minerals as well as the introduction of anionic salts has had a significant effect on the reduction of hypocalcemia in the dairy industry (Block, 1984; USDA, 2007). However, with almost 50% of all dairy cows second lactation and greater affected by subclinical hypocalcemia, control and mitigation of subclinical hypocalcemia remains a concern for the dairy industry (Rodriguez et al., 2017).

Over the past several decades, a better understanding of Ca homeostasis during the periparturient period has emerged. Parathyroid hormone (**PTH**) and vitamin D metabolites are critical regulators of calcium balance in nonlactating states; however, lactating mammals require additional lactation specific mechanisms to achieve calcium homeostasis (Wysolmerski, 2012; Kovacs, 2017). In dairy cows, research has demonstrated that the release of PTH by the parathyroid gland reduces loss of urinary Ca, increases bone resorption of Ca, and increases the synthesis of $1{,}25(OH)_2D_3$ to enhance intestinal Ca absorption (Horst et al., 1997). While it was once believed that PTH and vitamin D were the primary modulators of Ca mobilization regardless of the animal's physiological state, more recent work has shown that parathyroid hormone-related protein (**PTHrP**) produced by the mammary tissue is critical in regulating calcium metabolism during lactation (Moseley et al., 1987; Sowers et al., 1996; Mather et al., 1999; VanHouten et al., 2003). However, evidence also indicates that colostral concentrations of PTHrP in cows are not different between healthy and paretic cows (Riond et al., 1996). Because the binding site for PTHrP on the PTH-receptor is homologous to that of PTH, this allows both hormones to induce common cellular signal transduction pathways through interaction with the same receptor (Wysolmerski, 2012). Feeding anionic salts to dairy cows lowers blood pH, thereby altering the physiological structure of the PTH-receptor on osteoblast cells (Block, 1984; Goff, 2008). The conformational change of the PTH-receptor created by feeding the anionic salts is known to increase the ability of PTH to bind to its receptor, leading to increased Ca resorption from bone and increased synthesis of $1{,}25(OH)_2D_3$. It is likely that this may also increase the ability of PTHrP to bind as well.

In recent years, increased attention has been directed toward understanding the role of the monoamine serotonin (5-hydroxytryptamine) on mammary gland and lactation physiology. Specifically, research has demonstrated a role for serotonin in regulating Ca homeostasis during lactation (Hernandez et al., 2012; Weaver et al., 2016). Laporta et al. (2013) and Weaver et at. (2016) have shown an increase in tCa concentrations during lactation in both rodents and cows when serotonin concentrations are increased around the time of parturition through the administration of 5-hydroxy-L-tryptophan (**5-HTP**), the direct precursor for the synthesis of serotonin, which bypasses the use of the rate-limiting enzyme tryptophan hydroxylase 1 (Wang et al., 2002; Laporta et al., 2015; Weaver et al., 2016). Given previous research on serotonin and PTHrP during lactation, it seems necessary to explore this relationship further in the dairy cow, despite conflicting evidence (Laporta et al., 2013; Moore et al., 2015).

This experiment aims to determine if feeding a negative DCAD diet in combination with administration of 5-HTP prepartum increases postpartum blood Ca concentrations compared with feeding a negative DCAD diet alone. Our hypothesis is that feeding a negative DCAD diet in combination with the 5-HTP treatment prepartum will elevate postpartum maternal blood Ca concentrations more than either of these treatments alone.

MATERIALS AND METHODS

The College of Agriculture and Life Sciences Animal Care and Use Committee at the University of Wisconsin–Madison approved all experimental procedures used on animals in this study. Care and use protocol guidelines (A005316) were strictly followed for this experiment.

Animals and Experimental Design

Thirty-one healthy, multiparous Holstein cows (second through fifth lactation), of similar production level and lactation number (average lactation number 2.8 ± 0.3; ±SEM), were used for this experiment. Cows were blocked by parity and average lactation number was not different between treatment groups. Cows enrolled on study did not have a previous history of transition-related disorders. Approximately 4 wk before the expected date of parturition, cows were transported to the Dairy Cattle Center, a tiestall facility, at the University of Wisconsin–Madison, allowing approximately 1 wk for cows to acclimate to the facility. Feed intake was recorded daily by the farm staff. During this acclimation period, all cows were placed on a common herd dry cow diet, which was a low-energy, high-fiber, and low-DCAD diet consisting primarily of corn silage and straw, with no addition of anionic salts.

The experimental design was a randomized complete block design with a 2 × 2 factorial arrangement of treatments resulting in 4 treatment groups: positive DCAD

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