Influence of Body Condition Score on Relationships Between Metabolic Status and Oxidative Stress in Periparturient Dairy Cows*

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ABSTRACT

Twenty-four dairy cows were monitored during the transition period. We observed changes of oxidative status and relationships between oxidative and metabolic status. Body condition score (BCS) of the 24 animals at the beginning of the trial $(30.4 \pm 2 \text{ d before expected})$ calving) was between 2.0 and 3.6. The BCS was recorded and blood samples were collected weekly during the last 30 d of pregnancy and the first 30 DIM. Plasma samples were analyzed to determine indices of oxidative status: reactive oxygen metabolites (ROM); thiobarbituric acid-reactive substances (TBARS); thiol groups (SH); glutathione peroxidase (GSH-Px), and indices of energy metabolism: glucose, β -hydroxybutyrate, and nonesterified fatty acids. In erythrocytes we determined indices of oxidative status: GSH-Px, superoxide dismutase (SOD), and intracellular SH. Before calving, cows showed an increase of plasma SH, SOD, and GSH-Px, a decrease of erythrocyte GSH-Px and plasma ROM, and no changes in erythrocyte SH. After calving, cows showed a decrease of plasma and erythrocyte SH and SOD, and an increase of ROM, TBARS, and plasma GSH-Px. Cows with higher BCS at the beginning of the trial and greater loss of BCS after calving, had higher plasma ROM, TBARS, and SH, and lower SOD and erythrocyte SH in the postpartum period. Oxidative status of dairy cows was related to energy status. Cows with higher BHBA and NEFA showed higher ROM and TBARS and lower levels of antioxidants. Results of the present study demonstrated that cows can experience oxidative stress during the peripartum period, and cows with higher BCS and greater BCS losses are more sensitive to oxidative stress.

(**Key words:** dairy cow, transition period, oxidative status, metabolic status)

Abbreviation key: GSH-Px = Se-glutathione peroxidase, **HBCS** = high BCS, **LBCS** = low BCS, **MBCS** =

medium BCS, **PCV** = packed cell volume, **ROM** = reactive oxygen metabolites, **SH** = thiol groups, **SOD** = CuZn-superoxide dismutase, **TBARS** = thiobarbituric acid-reactive substances.

INTRODUCTION

Oxidative stress in a living organism is a result of an imbalance between reactive oxygen metabolites (**ROM**) production and neutralizing capacity of antioxidant mechanisms (Sies, 1991). Oxidative stress leads to peroxidative damage of lipids and other macromolecules, with consequent alteration of cell membranes and other cellular components (Toyokuni, 1999). Oxidative stress can lead to the modification of important physiological and metabolic functions. Some researchers (Miller et al., 1993; 1994) reported that oxidative stress could alter the physiology and could cause pathologies.

The transition period is particularly important for health and subsequent performance of dairy cows, which are exposed to drastic physiological changes and metabolic stress (Grummer, 1993; Goff and Horst, 1997; Drackley, 1999). Relationships between BCS and incidence of metabolic diseases have been exhaustively reported (Gearhart et al., 1990; Studer, 1998). It has been hypothesized that an involvement of oxidative stress during transition period is the etiology of some diseases and disorders in dairy cows (Harrison et al., 1984; Smith et al., 1984; Gröhn et al., 1989; Lomba, 1996).

In humans, recent studies showed a close relationship between high body mass index, body weight loss, and oxidative stress (Ozata et al., 2002; Higdon and Frei, 2003; Keaney et al., 2003), and researchers hypothesized a possible relationship between oxidative stress and incidence of some metabolic diseases (Higdon and Frei, 2003; Morrow, 2003).

Our hypothesis is that oxidative status might also be related to metabolic disorders in dairy cows. To date, there is no information available concerning the relationship between changes in oxidative status indices and metabolic status in periparturient dairy cows. In the present study, we evaluated oxidative status and assessed the possible relationships between oxidative and metabolic status in periparturient dairy cows.

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Table 1. Ingredient and nutrient compositions of diets administered during the experimental period.

| Ingredient (% of DM) | Far-off dry cows | Close-up dry cows | Lactating cows |
|------------------------------------|---------------------|----------------------|-------------------|
| Corn silage | 11.90 | 18.80 | 26.40 |
| Rye-grass silage | 3.20 | 5.00 | 7.00 |
| Alfalfa hay | 4.30 | 6.80 | 9.60 |
| Rye-grass hay | 58.50 | 34.10 | 7.70 |
| Commercial mixed feed ¹ | 6.10 | 9.60 | 13.50 |
| Corn, ground | 6.20 | 9.80 | 13.70 |
| Oat, ground | 4.00 | 6.20 | 8.70 |
| Wheat middlings flour | 2.10 | 3.30 | 4.70 |
| Cotton seed | 2.90 | 4.70 | 6.60 |
| Mineral premix ² | 1.00 | 1.60 | 2.20 |
| Nutrient composition on DM basis | | | |
| NE _L , Mcal/kg | 1.30 | 1.40 | 1.55 |
| Crude proteins, % | 12.30 | 14.20 | 16.60 |
| NDF, $\hat{\%}$ | 42.10 | 38.20 | 34.10 |

¹Contained 87% DM, 23.7% CP and 1.89 Mcal/kg on DM basis and 100,000 IU of vitamin A, 8000 IU of vitamin D, 100 mg of vitamin E, 500 mg of niacin, 10.4 mg of vitamin B₁, 14 mg of vitamin B₂, 4 mg of vitamin B₆, 0.1 mg of vitamin B₁₂, 200 mg of Fe, 30 mg of Cu, 160 mg of Mg, 4.4 mg of Co, 10.8 mg of I, 360 mg of Zn, and 0.6 mg of Se per kilogram.

²A mixture of 33.3% CaCO₃, 31.7 Ca₃(PO₄)₂, 16.7% MgO, 16.6% NaHCO₃, and 1.7% ZnSO₄.

MATERIALS AND METHODS

Animals, Housing, and Feeding

The trial was carried out in a commercial dairy herd consisting of 290 lactating cows. The average milk yield per lactation (305-DIM) of the herd was more than 9000 kg. Twenty-four pregnant Holstein cows (10 primiparous, 7 at second calving, and 7 at third calving) were selected based on their BCS, parity, and expected calving date. In particular, cows with BCS < 2.5, BCS from 2.6 to 3.0, and BCS > 3.0 were assigned to low (LBCS), medium (MBCS), and high (HBCS) BCS groups, respectively. Mean values of BCS at 30.4 ± 2 d before calving were 2.3 ± 0.2 , 2.8 ± 0.1 , and 3.2 ± 0.2 in LBCS, MBCS, and HBCS, respectively. Body condition score was statistically different between the 3 groups (P <0.01). The 3 groups of 8 healthy cows were balanced across expected calving date and parity. The animals were monitored during the last 30 d of pregnancy and the first 30 DIM. The average $(\pm SD)$ parity was: 1.9 \pm 0.8, 1.9 \pm 0.9, and 1.9 \pm 0.8 in LBCS, MBCS, and HBCS, respectively.

The diets administered during the trial consisted of a base ration given ad libitum to achieve 5 to 10% feed refusals as a daily TMR (at ~0930 h). The composition of the diet used during the experiment (far-off dry, closeup, and lactation diet) is reported in Table 1. The closeup diet was offered during the last 10 d before the expected calving.

Measurements and Sampling

Dry matter of feedstuffs was determined by forcedair oven drying at 65°C to constant weight. Crude protein was determined by macro-Kjeldahl method (AOAC, 1984). Ether extract and ash were determined according to AOAC methods (AOAC, 1984). The NDF was analyzed according to the method described by Goering and Van Soest (1970).

During the trial, BCS was recorded and blood samples were taken weekly from each animal. Body condition was scored by the same person adopting a 5-point scale method (ADAS, 1986). Bleeding was carried out using evacuated tubes containing Li-heparin as anticoagulant. Body condition score was recorded and blood samples were taken from the jugular vein at d 30.4 \pm 2.0, 24.7 \pm 2.4, 17.6 \pm 2.2, 10.8 \pm 2.2, and 3.9 \pm 1.6 prepartum and at 4, 11, 18, 25, and 30 d postpartum. Whole blood was used to determine packed cell volume (PCV) by microhemocytometer. Blood samples were centrifuged at $2700 \times g$ for 10 min at 4°C and plasma was separated. Plasma samples were analyzed to determine glucose (Instrumentation Laboratory, Lexington, MA), BHBA (Barnouin et al., 1986), and NEFA (NEFA-C kit, Wako Fine Chemical Industries Inc., Dallas, TX). The following indices of oxidative status were analyzed in plasma samples: Se-glutathione peroxidase (GSH-Px) activity, thiol groups (SH) concentration, ROM, and thiobarbituric acid-reactive substances (TBARS) concentration.

Erythrocytes were obtained by centrifuging 0.5 mL of blood at $2200 \times g$ for 10 min at 4°C. Erythrocytes were then washed 4 times with 3 mL of 0.9% NaCl solution for 10 min at $2200 \times g$ at 4°C. After the final wash, red blood cells were lysed by hypotonic shock using 2.0 mL of cold ultrapure water. The hemolysate was mixed and left at 4°C for 15 min. Erythrocyte lysate was analyzed to determine intracellular SH concentra-

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