

Evaluation of Neutrophil Function in Periparturient Dairy Cattle

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Introduction

The periparturient period in dairy cattle is associated with increased incidence of several metabolic diseases. It has been hypothesized that reduced immune function may play an important role in the increased risk of certain diseases during this time period. This study examined neutrophil function of periparturient cows.

Materials and Methods

The investigation was conducted on Holstein cows from the Elora Dairy Research Center in the summer of 2001. Thirteen primiparous and fourteen multiparous cows were included in the study. Nine animals had been vaccinated with a commercially available *Escherichia coli* vaccine within the past month. Whole blood and serum samples were collected from each animal 1 to 6 days prior to calving and then again at 1 to 6 days after calving. Blood was collected from two mid-lactation control cows each day to control for day-to-day test variation. Glucose, beta-hydroxy butyrate (BHBA) and non-esterified fatty acids (NEFA) were measured in each serum sample. Neutrophils were purified from whole blood and their function tested using phagocytosis, chemotaxis and oxidative burst assays. Neutrophil phagocytosis was determined using flow cytometry to quantify ingestion of FITC-labeled *Staphylococcus aureus*. Chemotaxis was measured in a 96-well plate system (ChemoTx, Neuroprobe), using zymosan-activated serum as the chemoattractant. To measure the oxidative burst, neutrophils were loaded with H2DCF_A, and the

change in fluorescence following exposure to PMA was measured using flow cytometry. Data were expressed as a percentage of the two control cows' values for that day, to control for day-to-day variation in the assays. Associations between serum energy indicators, parity, and vaccination status precalving with neutrophil function were tested with analysis of variance.

Results

Post partum heifers had significantly reduced oxidative burst and phagocytosis function ($p=0.03$ and $p=0.04$, respectively) when compared to multiparous animals. Neutrophil function was not significantly different between pre- and post-calving samples, and NEFA and BHBA concentrations did not significantly impact neutrophil function. Chemotaxis was negatively correlated with phagocytosis and oxidative burst, and phagocytosis and oxidative burst were positively correlated. Glucose levels post-calving were positively correlated with chemotaxis ($p=0.08$). Vaccination status was positively correlated with pre-calving phagocytosis ($p=0.03$).

Conclusions

Neutrophil function was significantly different in primiparous compared to multiparous cows, perhaps related to differing metabolic states. The lack of correlation between indicators of negative energy balance, especially BHBA and neutrophil function, was surprising, given that BHBA impairs neutrophil oxidative burst *in vitro*. Consistent changes in neutrophil function at the time of calving were not detected in this study. In particular, suppression of neutrophil function in the immediate postpartum period was not identified.