

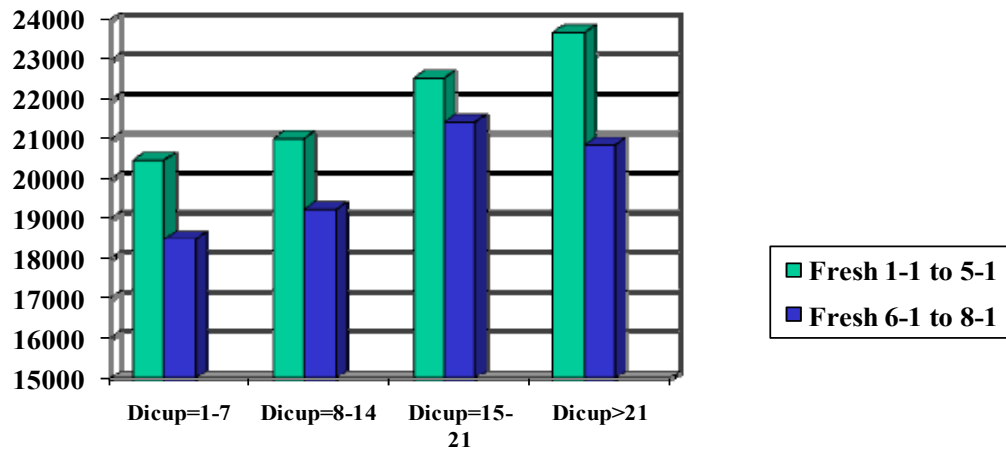
## **MINIMIZING THE EFFECTS OF IMMUNOSUPPRESSION THROUGH MANAGEMENT AND NUTRITION**

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It is a well known fact that the highest number of clinical mastitis cases occurs during the first week of lactation, and that the lactating cow is more likely to develop clinical mastitis during the first three months of lactation than the remainder of the lactating period. There are numerous physical factors that have been suggested to predispose the lactating dairy cow to clinical mastitis. Some of these suggested factors that have been around for many years are cows dripping milk both prior to and after parturition, excessive udder edema resulting in increased incidence of liner slippage during milking, poor letdown and incomplete milkout in first calf heifers, failure of the teat sphincter to close after milking due to edema and hypocalcemia, etc. More recent research has uncovered several nutritional, environmental, physiological and social reasons that directly affect the animal's ability to mount an effective immune response against an invading pathogen. Many of these things that have an adverse effect on the immune system can be minimized through improved nutritional and management procedures on the farm.

During the last ten years there has been much emphasis placed on the importance of managing the transition period from dry cow to lactating cow. A successful transition period determines the overall productivity of the current lactation, including the health of the cow, milk production, and reproductive efficiency. The transition period is also the time that nutritional and management practices can have the biggest impact on the animal's immune system. There is a direct correlation between improved transition period management and a decrease in the probability of acquiring a new clinical case of mastitis, as well as an increased likelihood of the animal overcoming a new intramammary infection because of a better functioning immune system.

One of the most important factors is to make sure that all animals are in the close-up dry pen for a minimum of three weeks. This ration is designed to provide the nutrition necessary to prepare the animal for the upcoming lactation, minimize metabolic disease, and supply the fetus with the nutrients required for the rapid growth it experiences during the last few weeks prior to birth. Most computer programs can be easily set up to monitor the number of days each animal is in the close-up dry pen. If not currently doing this, most dairymen are surprised to find out how many animals do not receive adequate time in this pen. If the dairy does not have a program in place where the heifers are pregnancy tested and accurate conception dates established, a significant number of the first calf heifers freshen with very little time in the close-up pen. The following chart is a summary of data collected from five of my clients representing 13,000 cows:



These dairies were located from Idaho to New Mexico but were all consistent in their results. Animals summarized in this data set gave approximately 1,000 lbs more milk for every additional week they were in the close-up pen.

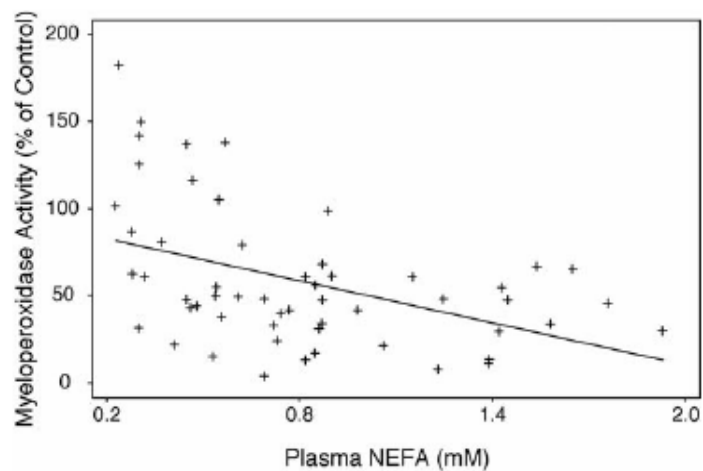
In today's larger dairies, it has become very commonplace for the transition cow to be moved five or more times during this period. Each of these moves requires a social adjustment where the animals have to reestablish the order of dominance. This almost always results in a decrease in dry matter intake in an animal that is already decreasing intake as she approaches parturition. If possible, minimizing pen moves will result in less stress on the animal and help maintain dry matter intake during this critical period. On larger dairies it is also possible to separate the first calf heifers from the multiparous cows. This helps maintain the intakes of the heifers on a more consistent basis.

It has been shown that there is a direct correlation between decreased dry matter intake prior to calving and impaired PMN (neutrophil) function both before and after calving (Hammon et al., 2006). This study showed an increase in uterine health disorders due to impaired neutrophil function. These neutrophils are the white blood cells that are the first line of defense in the mammary gland when pathogens gain access to it. Impaired neutrophil function will increase the chances of a mastitis pathogen becoming well established in the mammary tissue, resulting in a new mastitis infection that is difficult to overcome.

A Canadian study looked the social interactions of animals several weeks prior to calving and its effect on post partum metritis (Huzzy et al., 2007). This is the first published research to show that social behavior may play an important role in transition cow health. Animals that were overcrowded in the close-up pen and/or were less dominant had significant decreases in dry matter intake prior to calving. For every 10-minute decrease in average daily feeding time during the week before calving, the animal was 1.72 times more likely to develop a severe case of metritis after calving. For every 1 kg decrease in dry matter intake, the animals were 3 times more likely to develop severe metritis. In the Hammon study, there was a correlation between impaired neutrophil function and the incidence of metritis. One can assume that the increase in severe metritis in this study was also due to an impaired immune response. Once again, a generalized impairment of the immune system will also have an adverse effect on the ability of the animal to mount an effective immune response against an invading mastitis pathogen.

It is evident then, that not having enough time in the close-up pen, overcrowding, increasing the number of social interactions through multiple pen moves, and mixing older cows with first calf heifers can all have an adverse effect on the animal's immune system, resulting in an increased susceptibility to new mastitis infections.

When animals do not have sufficient energy intake to provide body maintenance requirements, fetal energy requirements, and that required for colostrogenesis and the initiation of lactation, they must mobilize body fat. This is not a very efficient process in the dairy cow, and often results in the deposition of fat in the liver, commonly called fatty liver disease. This conversion of fat into glucose in the liver also results in the production of non-esterified fatty acids (NEFAs). The following graph illustrates that as the plasma NEFA level increases, the ability of the neutrophil to produce myeloperoxidase is decreased. Myeloperoxidase is necessary for the neutrophil to effectively destroy bacteria that it has engulfed. Therefore, NEFAs are directly immunosuppressive to the animal.



It is a fairly common practice for the veterinarian to collect blood samples from close-up dry cows and have them analyzed for NEFA levels. High NEFA levels are indicative of decreased dry matter intakes in the close-up pen and management changes must be implemented to correct this problem. Once again, since neutrophil function is impaired, animals in the late dry period and early lactation will be susceptible to new mastitis infections.

When the cow approaches the end of her gestation period, the calf secretes a high level of cortisol from its adrenal glands. This initiates the cascade of events that leads to parturition. However, cortisol is a highly immunosuppressive agent, and both the cow and the calf are immunosuppressed at the time of parturition. This is the major reason that fresh cows are more susceptible to new mastitis infections. Cortisol is released from the animals' adrenal glands whenever they undergo stress. Stress could be due to any number of reasons such as heat, cold, pen moves, ration changes, overcrowding, decreased dry matter intake, metabolic disease, etc. Therefore, any time that the animal undergoes any type of stress, its immune system is compromised, and it becomes more susceptible to a new mastitis infection. Any effort that can be made to reduce stress on the dairy cow will result in an improvement in their immune function. It is common to see a reduction in clinical mastitis and/or somatic cell counts when dairies in heat stress areas implement effective heat stress abatement procedures.

Another complicating factor that exists around the time of calving is the difficulty of maintaining a constant level of blood calcium in the cow. The fetus requires a significant amount of calcium for bone development during the last three weeks of gestation. At the same time, the process of colostrumogenesis starts approximately five weeks prior to calving. The cow secretes a very large amount of calcium into the colostrum which will always result in a drop in blood calcium level at the time of calving. Clinical and subclinical hypocalcemia is probably the underlying cause of more problems in the fresh cow than anything else. A high percentage of metabolic disease problems as well as an impaired immune function are directly correlated to low calcium levels.

In order to minimize the drop in blood calcium levels at parturition, many dairies feed a specially formulated ration that is called a negative dietary cation:anion diet (DCAD). The goal in this diet is to reduce the potassium (K<sup>+</sup>) and sodium (Na<sup>+</sup>) levels and increase the chloride (Cl<sup>-</sup>) and sulfur (S<sup>-</sup>) levels. This results in a reduction in blood pH which in turn makes the receptor site for parathyroid hormone more efficient. Parathyroid hormone is necessary for the mobilization of calcium from the bones. Improved calcium mobilization makes calcium stores more available at the time of parturition and minimizes the drop in blood calcium.

Calcium is necessary for all muscle contraction in the body. Animals that have clinical hypocalcemia are usually recumbent and show obvious signs of “milk fever”. These animals will often die without calcium replacement therapy. However, animals that experience subclinical hypocalcemia have lower than the normal range of calcium in the bloodstream, but usually do not exhibit any observable clinical signs of hypocalcemia. Since muscle contraction is impaired, this often results in failure of the uterus to involute normally following parturition, a slowing down of the entire gastro-intestinal tract, and an increase in the incidence of displaced abomasum. It is interesting to note that animals with hypocalcemia have blood cortisol levels that are approximately twice as high as animals with subclinical hypocalcemia, and three times higher than animals that are normal at the time of calving (Horst and Jorgensen, 1982). These higher levels of blood cortisol result in a more severe level of immunosuppression at the time of calving, making the animal much more susceptible to infectious diseases including mastitis.

The slowing down of the gastro-intestinal tract often results in decreased dry matter intake. This causes the animal to be in a more severe state of negative energy balance early in lactation. The immune system requires a great deal of both energy and protein in order to function properly. Animals in severe negative energy balance do not have the ability to respond to invading pathogens as effectively as others, and often experience problems with infectious disease early in lactation. The decreased dry matter intake also predisposes the animal to developing problems either with clinical or subclinical ketosis, which will be discussed later.

More recent research has revealed that calcium is also an integral part of the immune system and is involved in intracellular signaling factors of white blood cells (Kimura et al., 2006). It has been called the “second messenger” of the immune system, and the white blood cells of animals with hypocalcemia have a reduced ability to respond to invading pathogens. Fortunately, this impairment can be rapidly restored with proper calcium therapy. The importance of calcium’s direct involvement in the immune system is not well known and is often not considered as one of the problems resulting from subclinical or clinical hypocalcemia.

It is evident that calcium metabolism during the transition period affects the ability of the animals’ immune system to respond successfully to invading pathogens in numerous ways.

Every effort needs to be made both from a nutritional and management standpoint to minimize the drop of blood calcium levels around the time of parturition. This effort will be well-rewarded with a decrease in both metabolic disease and infectious disease.

Ketosis is another metabolic disease that has an adverse effect on the immune system. Any time that the blood glucose level drops to a point where the necessary energy required for cell metabolism is not met, the body starts mobilizing fat stores for energy. This fat is metabolized into glucose in the liver. During this process, metabolites called ketone bodies are produced. The three most common ketone bodies are acetoacetic acid, acetone, and beta hydroxybutyric acid (BHBA). The most predominant ketone is BHBA and is the one most commonly monitored both in humans and animals that are suspects for ketosis.

Ketosis may occur towards the end of the dry period or in the early lactation period, but is most commonly seen during the first two weeks of lactation. Negative energy balance is the underlying cause and results most commonly from decreased dry matter intake. As previously discussed, decreased dry matter intake may result from overcrowding, social interaction, pen moves, feed bunk management, metabolic disease, and infectious disease. Decreased nutrient density in the ration may also result in ketosis when dry matter intake is sufficient. High levels of straw in close-up dry cow diets may predispose animals to ketosis. If ketosis is diagnosed during the first week of lactation, it is most likely due to insufficient energy intake during the close-up dry period. If diagnosed during the second week, it is more likely that the insufficient energy intake occurred after freshening. Cows with hypocalcemia are much more likely to develop ketosis due to a decrease in dry matter intake.

A recently published study has confirmed that BHBA has an adverse effect on neutrophil function in several different ways (Grinberg et al., 2008). When bovine neutrophils were incubated with levels of BHBA similar to that of an animal in ketosis, the phagocytosis of *E. coli* P4 was reduced five-fold. When in the presence of *E. coli* P4, bovine neutrophils also are stimulated to produce DNA “nets” that extrude from the neutrophil, entrapping and destroying the bacteria. When incubated with similar levels of BHBA, this ability is reduced ten-fold. Therefore, the initial response of the immune system in the mammary gland to an invading pathogen is severely compromised in the presence of BHBA. This explains why cows with ketosis are much more susceptible to infectious disease and mastitis. The synthesis of poly (3-hydroxybutyrate) and its degradation to BHBA are very common metabolic pathways in many bacteria, including *E. coli* (Grinberg et al., 2008). This may also be a possible role of BHBA as a microbial virulence factor in bovine mastitis.

Another area of current interest in nutrition is the effect of essential fatty acids on the immune system. Essential fatty acids are directly involved in modulating signaling pathways leading to cytokine production and subsequent activation of cytokine receptors (Hwang, 2000). These signaling molecules are important in the normal function of both T and B lymphocytes. One of the essential fatty acids, linoleic acid, is used to make prostaglandin F<sub>2a</sub>, which has a proinflammatory influence on neutrophils, helping them to be more effective in fighting pathogens (Staples et al., 2008). A recent study utilizing calcium salts of safflower oil containing 64% linoleic acid in the diet of dairy cows, showed a significant improvement in neutrophil phagocytosis of *E. coli* (Silvestre et al., 2008). This study also showed a significant increase in the cytokines L-selectin and tumor necrosis factor alpha, as well as an increase in the production of acute phase proteins haptoglobin and fibrinogen in the liver.

The CPM ration formulation software currently contains a fat submodel that allows the nutritionist to utilize information on the amounts of essential fatty acids contained in the ration, and rations can be adjusted to provide these nutrients in the recommended amounts to help optimize the immune system, improve fatty acid metabolism and rumen efficiency, and have a positive effect on reproductive performance.

As previously mentioned, the immunosuppressive hormone cortisol is released during stress. It is very common to see major outbreaks of mastitis during times of severe environmental stress, at any point during the lactation. Cow comfort plays an important role in lowering somatic cell counts and minimizing the incidence of clinical mastitis. Submissive cows in overcrowded pens are also more likely to develop clinical disease. Minimizing pen movements and good feed bunk management will help maintain dry matter intakes in all lactating cows.

The transition period of the dairy cow is by far the most important window of opportunity that the dairyman has to maximize the overall profitability of the dairy cow. Paying close attention to ration formulation, palatability, stocking density, pen movements, feed bunk management, cow comfort, and the calm handling of animals during the transition is paramount in minimizing the amount of immunosuppression that occurs during this critical time period. Close observation of the fresh cow and early detection of both metabolic and infectious disease followed by prompt therapy will minimize the stress on these animals and result in a more effective immune response in fresh cows. The incidence of both clinical and subclinical mastitis can be greatly reduced by combining these nutritional and management procedures with the common mastitis prevention practices recommended by the National Mastitis Council. Improving the ability of the animal to mount an effective immune response against an invading pathogen, is an important part of an effective mastitis control program.

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