FEEDING BEHAVIOUR IDENTIFIES DAIRY COWS AT RISK FOR METRITIS

by

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B.Sc. (Agriculture), The University of British Columbia, 2000

A THESIS SUBMITTED IN PARTIAL FULFILMENT OF
THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF SCIENCE

in

THE FACULTY OF GRADUATE STUDIES

(ANIMAL SCIENCE)

THE UNIVERSITY OF BRITISH COLUMBIA

June 2005

ABSTRACT

Dairy cows experience a high incidence of disease in the weeks immediately after calving, but early and accurate diagnosis remains a challenge. Cows suffering from metritis, one common disease after calving, exhibit reduced milk yield and reproductive performance. However, afflicted cows show few overt signs of illness and frequently go unnoticed in the absence of veterinary examination. To determine if changes in feeding behaviour could be used in the identification of animals at risk for metritis, attendance at the feed alley was monitored continuously for 26 Holstein cows during the transition period, beginning 2 weeks before and ending 3 weeks after calving. Every 3±1 d cows were diagnosed for metritis based on rectal body temperature and condition of vaginal discharge. Over the 3 weeks of observations after calving, 69% of cows showed some signs of metritis. These cows spent on average 22 min/d less time at the feed alley during the transition period than did non-metritic cows. For every 10-min decrease in average daily feeding time, cows were twice as likely to be diagnosed with metritis. A threshold of 75 min of average daily feeding time was 89% sensitive and 62% specific for detection of acute metritis. In conclusion, reduced time at the feeder can be used to identify dairy cows at risk for metritis. More research is required to determine how soon before calving at-risk cows can be identified, and if these behavioural differences can also be used in the early diagnosis of other diseases.

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ADF = acid detergent fibre

BCS = body condition score

BT = rectal body temperature

CP = crude protein

DIM = days in milk (number of days since the onset of lactation)

DM = dry matter

DMI = dry matter intake

NDF = neutral detergent fibre

R^2 = coefficient of determination

SD = standard deviation

SED = standard error of the difference between the means

TMR = total mixed ration

VD = vaginal discharge

ACKNOWLDEGEMENTS

I gratefully acknowledge the support of staff and students at the University of British Columbia's Dairy Education and Research Center, and the insightful comments of members of the UBC Animal Welfare Program, in particular members of my supervisory committee, Dr. Marina von Keyserlingk, Dr. Dan Weary, Dr. David Fraser, and Dr. Jim Love. I am especially grateful to Dr. Andreia Vieira for her help in a preliminary trial and in developing the criteria used in this study. I also thank Dr. Karen Beauchemin from Agriculture and Agri-Food Canada for providing the feeding behaviour monitoring equipment and Drs. Brian Upper and Raja Rajamahendran for the diagnostic training they provided. Financial support for was provided by the Natural Sciences and Engineering Research Council of Canada, the Dairy Farmers of Canada, the BC Dairy Foundation, the BC SPCA, members of the BC Veterinary Medical Association and many others listed at www.agsci.ubc.ca/animalwelfare. I owe great thanks to my family and friends for their personal support and to Amy Waterhouse for her immeasurable patience.

CO-AUTHORSHIP STATEMENT

The study was designed collaboratively by Geoff Urton and Drs. Marina von Keyserlingk and Dan Weary. Geoff installed the data collection system and monitored animals with the help of research assistants. He analysed all data and prepared the manuscript under the guidance of Drs. von Keyserlingk and Weary.

CHAPTER 1: INTRODUCTION

Over the last 50 years, dramatic increases in the productivity of animal agriculture have been realized in both the individual animal and the farm as a whole. The intensification of animal management, the power of selective breeding, and improvements in nutrition have all contributed to this increase. Concurrent with this growth we have seen a dramatic decrease in the total number of farms (Bragg and Dalton, 2004). These trends have been particularly noticeable in the dairy industry: by 2002, the population of dairy farms in the US had shrunk to one fifth of the number reported in 1974 and the number of cows/farm had grown nearly four fold (USDA, 2002). Over the same period, the annual productivity/cow has increased by 81% (USDA 2002). In recent years, US productivity/cow has been increasing at a rate of 1.7% per year (USDA 2002), with approximately 33-40% of this growth attributed to improvement in genetics, the remainder being accounted for by improved nutrition and management (Hutjens, 1996).

In Canada, the dairy industry is the fourth most productive agricultural sector, generating $11 billion in processor sales annually (Canadian Dairy Information Centre (CDIC), 2004). The industry employs 64,000 people on the nation's 17,890 dairy farms and 444 primary processing plants. The national dairy herd of 1.1 million cows produced 74 million hectolitres of milk in 2003. The majority of Canadian dairy farms are concentrated in Quebec and Ontario, with 72% of the national herd residing in these two provinces. National milk production is generally increasing at a rate of 2.5% per cow per year (CDIC, 2004) and the average Canadian dairy cow now produces 31 kg of milk per day. This represents an increase of 200% from 1953 (Statistics Canada, 2005).

This dramatic increase in milk yield per cow is attributed primarily to two developments: (1) almost exclusive use (93%) of the high-producing Holstein-Friesian breed and, (2) a selective breeding effort aimed at increasing milk yield per cow (see review by Rauw et al., 1998). Other factors that have played a role include: 1) the development of assisted reproductive technologies (e.g. artificial insemination) (Foote, 1996), 2) a better understanding of nutritional needs at different stages of lactation (e.g. Bines, 1976), and 3) more intensive management systems and infrastructure designed to facilitate efficient management of large numbers of animals (Fox, 1983). Unfortunately, these highly productive dairy systems may place substantial physical and physiological demands on the animals. These demands have consequences for animal welfare that have been largely overlooked by the industry.

Welfare of the Dairy Cow

The concept that humans are responsible for ensuring the welfare of the animals we raise for our use is becoming a widespread social ethic (Rollin, 2004). However, considerable disagreement exists as to what constitutes good or poor welfare. Duncan and Fraser (1997) have identified three schools of thought that have emerged amid attempts to define animal welfare: the naturalness school, the biological fitness school, and the feelings school. These concepts are not mutually exclusive and many stakeholders occupy a position that bridges more than one school – e.g. Webster's (2005) "fit and feeling good" definition of animal welfare.

Those that subscribe to the naturalness school believe that animals should be able to live in a natural way – that is, to experience a natural environment and have the opportunity to express natural behaviours. This concept has perhaps been best described by Rollin (1981) in his application of the Aristotelian concept of *telos* to animals. To Aristotle

everything had an essential nature, or *telos*, defined by both its form and its function: e.g. the “knifeness” of the knife, of which its sharpness and ability to cut are intrinsic features. Similarly, each species can be said to have its own *telos*, genetically programmed through evolution, and expressed with some variation according to environment. The ability of a bird to fly or a cow to graze could be said to be intrinsic features of the *telos* of each of these animals. Ensuring that animals are able to express natural or normal behaviours has been identified as a key criterion of animal welfare by the Farm Animal Welfare Council of the UK (1997) and has even been incorporated into legislation in Sweden.

Proponents of the biological fitness school of thought generally look to physiological signs of welfare in the individual animal, including disease, injury and stress. Broom’s (1986) assertion that “the welfare of an individual is its state as regards its attempts to cope with its environment” typifies this approach; no explicit reference is made to the necessity of an animal to experience what is natural or to feel good. Incidentally, this definition enables animal welfare to be assessed in a relatively direct manner, due to the availability of objective, diagnostic tools employed regularly by veterinarians and animal physiologists.

Those who subscribe to the feelings school of animal welfare believe that the subjective experience of an animal is what is most relevant to its welfare (e.g. Dawkins, 1985; Duncan, 1993). Relevant to this topic is the concept of sentience, a term which is commonly used to denote either the rudimentary ability to sense (i.e. perceive one’s environment) or the more sophisticated ability to experience feelings and emotions. Much debate has focussed on the issue of animal sentience and on the question of which animals are capable of experiencing unpleasant feelings such as pain.

In consideration of all of these concepts, poor welfare may be said to come about as a result of a number of conditions, including disease, injury, pain, stress, discomfort, frustration, and unnatural environments. On modern dairy farms, cows may experience any of the above conditions as a result of housing and management practices. Tethering of cows (Müller et al., 1989) and calf weaning practices (Flower and Weary, 2003) may prevent animals from expressing natural behaviours and therefore cause stress or frustration. Concrete barn flooring and bed-stalls may contribute to injuries such as hock and hoof lesions (Weary and Taszkun, 2000; Wechsler et al., 2000; Vokey et al., 2001), causing pain or discomfort, and management procedures such as dehorning may cause considerable pain (Faulkner and Weary, 2000; Vickers et al., 2005). Inadequate nutrition can dramatically increase the risk of disease (Radostits et al., 1994). It has also been suggested that dairy cows provided with inadequate access to feed experience hunger, constituting a considerable welfare concern (e.g. Webster, 1995).

Metabolic Demands of the Modern Dairy Cow

As mentioned previously, the selective breeding strategies of the last few decades have focused almost exclusively on increasing milk production. Not surprisingly, this approach may have consequences for welfare. The increased capability for lactation has brought with it the challenge of meeting the increased nutritional needs of the high producing dairy cow. The average modern dairy cow that produces 31 kg of milk per day has substantially different nutritional requirements than did the 1964 cow with a 10 kg per day yield (Statistics Canada, 2005). The modern dairy cow at peak lactation requires approximately 25.0 Mcal of energy, 1.3 kg of protein, 1.5 kg of fat, and 500 g of calcium daily for milk production alone (Bell, 1995).

Supplying a diet adequate for this rate of metabolic output is challenging and recommendations for dietary management of a dairy herd are complex (e.g. Hutjens, 1996). As a result, the knowledge needed to manage the diet of a modern dairy herd is highly specialized and dynamic. Herd managers and nutritionists must coordinate the diets of a diverse herd, all at various stages of lactation. An individual cow may undergo five changes in ration over the course of a single cycle of gestation and lactation. This is particularly relevant to the welfare of the animals, as meeting a dairy cow's nutritional needs is key to maintaining her health (Van Saun and Sniffen, 1996). Moreover, cows that are not provided with adequate nutrition are vulnerable to both metabolic and infectious disease, predominantly during the period of transition from gestation to lactation (e.g. Rukkwamsuk et al., 1999).

The Transition Period

The transition period, as defined by Grummer (1995), is the phase of the dairy cow's gestation-lactation cycle that begins three weeks before calving and lasts until three weeks after calving. Over these six weeks the dairy cow is exposed to dramatic physiological, dietary, and social transitions, including the abrupt transition from pregnancy to lactation, the shift from a low energy to a high-energy diet, and the displacement from one social group to another. The transition period is typified by several characteristic physiological and behavioural changes related to the changes noted above. They include: 1) a decline in feed intake, beginning 3 weeks before calving (e.g. Bertics et al., 1992, Rabelo et al., 2003), 2) depression of certain immune functions both before and after calving (e.g. Kehrli and Goff, 1989; Mallard et al., 1998), 3) a negative energy balance (Zamet et al., 1979; Bertics et al., 1992) and decline in serum calcium and glucose at the onset of lactation (Houe et al., 2001) and, 4) an increase in feed intake beginning immediately after calving (e.g. Rabelo et al., 2003). Aspects of the nutrition, health and management of transition

cows have been discussed extensively in several recent reviews (Drackley, 1999; Goff and Horst, 1997; Van Saun and Sniffen, 1996; Hutjens, 1996; Bell, 1995; Grummer, 1995; Grant and Albright, 1995).

Increased physiological demands during the transition period place an inordinate metabolic demand on the body (Bell, 1995). During this time the cow is unable to ingest sufficient nutrients to meet the metabolic demand of lactation; animals typically enter an energy deficit (Zamet et al., 1979, Bertics et al., 1992) and many cows become hypocalcemic (Houe et al., 2001). The result is an elevated risk for one or more of several common post-parturient metabolic or infectious diseases such as ketosis, displaced abomasum, milk fever, retained placenta, metritis, and mastitis (Rukkwamsuk et al., 1999). These nutrient demands cannot be supported by a traditional grazing diet. In fact, in a recent trial, Holsteins raised on pasture produced 16% less milk than those raised in freestalls and dealt less well with transition (greater weight loss and lower plasma glucose), despite a dietary supplement of grain concentrate (Fontaneli et al., 2005).

Social factors may place further demands on the animals as they must re-establish dominance hierarchies when moved to a new group (Friend and Polan, 1974). Cows may be regrouped once at the beginning of the transition period and again after calving; the stress induced by these social changes may compromise the ability of the cow to cope with the physiological stressors present at this time (Wierenga, 1990). Submissive cows have been shown to regularly surrender their access to valuable resources such as preferred bed stalls and space at the feed bunk to dominant cows (Hasegawa et al., 1997). The needs of these subordinates may be further marginalized in the unstable social environment that exists during transition. Regrouping results in increased competitive

interactions between animals, increasing the risk that submissive cows will face inadequate nutrition and rest (Boe and Faerevik, 2003).

Health of the Transitional Cow

The metabolic and social stressors that prevail during the transition period contribute to the prevalence of disease around the time of calving (Drackley, 1999; Collard et al., 1999). Metabolic diseases such as ketosis and clinical hypocalcaemia (milk fever) are common, occurring with incidences of 3 to 39% and 6 to 12%, respectively (Rajala-Shultz et al, 1999; Zwald et al., 2004; Hardeng and Edge, 2001). As metabolic diseases arise directly from nutritional inadequacies (e.g. Rukkwamsuk et al., 1999), the majority of work to date has focused on nutrition at this time.

Susceptibility to certain infectious diseases also peaks at this time (Smith et al., 1985). Reported incidence rates for infections of the udder (mastitis) and of the uterus (metritis) are 1.7 to 54.6% and 10.1 to 65.5%, respectively (Kelton et al., 1998; Borsberry and Dobson, 1989; Hirvonen et al., 1999). The high prevalence of infectious disease during transition may also be linked to inadequate nutrition, as these diseases commonly occur as a secondary illness to primary metabolic disease (e.g. Reist et al., 2003). Inadequate nutrition may also contribute to the well-documented depression of the immune system that occurs during transition (e.g. Mallard et al., 1998; Kehrli and Goff, 1989). Specifically, infectious agents that the body would eliminate under robust immune conditions have been shown to induce clinical infectious disease in vulnerable areas such as the udder and uterus (Cai et al., 1994).

According to a model for dairy cow response to metabolic stress described by Knight et al. (1999), cows may experience mild, moderate, or severe metabolic stress during transition.

These authors propose that some animals cope well with the metabolic demands of lactation and only experience mild metabolic stress without any dramatic consequences for health or welfare. However, other animals may cope poorly and experience moderate to severe metabolic stress at this time.

Knight et al. (1999) suggested that animals that experience moderate metabolic stress after calving might sacrifice immune function for the sake of maintaining lactation, a considerably more important function at that time. Metabolic resources would not be diverted from lactation until a cow became severely metabolically stressed. Therefore, the common practice of using daily milk yield as a general indicator of animal health is a relatively insensitive method for identifying sick or at-risk animals. For example, milk production responds poorly to mild or subclinical infectious disease. In fact, Rajala-Shultz et al. (1999) found that cows with a fever produced more milk, on average, than did healthy cows. Thus, a more sensitive method of continuously monitoring animal health or risk for disease during the transition period is needed.

As veterinary examination of post-partum cows is relatively infrequent on most dairy farms (commonly once every two weeks), many cases of transition period disease may go unnoticed. Cattle are prey animals and therefore have developed a behavioural stoicism in response to pain and disease. This makes disease detection in the dairy cow particularly difficult. Producers can use urine or milk tests to monitor the health of their animals, but frequent administration of tests on a herd-wide scale can be costly and time-consuming. Moreover, no such tests are available for diagnosing inflammatory uterine disease (metritis or endometritis), one of the most common disorders after calving.

Infectious diseases such as metritis generally receive less attention than the metabolic diseases that are prevalent after calving. This is presumably because ketosis and milk fever have dramatic impacts on milk production and an animal's appearance, while metritis has no overt external symptoms (Lewis, 1997). Interest in prevention and treatment of metritis has increased due to concerns that metritic cows suffer from reduced fertility. Producers who neglect to monitor their animals for infectious disease may also be missing an opportunity to identify animals that are moderately metabolically stressed.

Metritis

Metritis frequently occurs soon after calving and may severely compromise reproductive performance (Fourichon et al*.,* 2000). Metritis and endometritis refer to the inflammation of the uterus and the endometrial lining, respectively. Both diseases are a consequence of sustained infection of the uterus caused by pathogenic bacteria such as *Arcanobarerium pyogenes* (LeBlanc et al., 2002). The distinction between these diseases is of little importance to this thesis so below I refer to both conditions as metritis.

Metritis is most commonly diagnosed within the first 30 days following calving, and is assumed not to be secondary to other disease when diagnosed by the 21st day of lactation (Kaneene and Miller, 1995). Postpartum metritis does not necessarily cause any general signs of illness but can reduce milk yield and diminish fertility (Borsberry and Dobson, 1989; Fourichon et al., 2000; LeBlanc et al., 2002). Many cows also develop a mild uterine inflammation within 14 days of calving and spontaneously recover thereafter without any consequences to reproductive performance (Lowder, 1993).

As indicated above, the reported incidence rate of metritis in dairy cows varies widely (10.1 to 65.5%) (Borsberry and Dobson, 1989; Hirvonen et al., 1999). This variability may

be attributed, in part, to inconsistency in method of diagnosis and the lack of a clear definition of the disease in the literature (LeBlanc et al., 2002). Metritis may also form disease complexes with one or more other diseases common at transition including retained placenta, dystocia, abortion, left displaced abomasum, ketosis, milk fever, and ovarian cysts (Kaneene and Miller, 1995; Curtis et al., 1985) and thus may not be recorded as such. Further, the lack of easily identifiable signs of metritis often results in cases going unnoticed in the absence of a veterinary examination. There is also a lack of research aimed at determining whether specific predisposing factors contribute to metritis (Lewis, 1997).

Transition Period Feeding

Much recent research has focused on nutritional treatments to improve the health of the transition cow (Drackley, 1999). Currently, the National Research Council (2001) recommends increasing the energy content of the diet from the 1.25 Mcal/kg DM recommended during the "far off" dry period to 1.62 Mcal/kg DM during the "close up" dry period (three weeks before calving). This strategy is thought to prepare the cow for the metabolic demands of early lactation and thus minimize the need for body tissue mobilization. However, various factors may prevent cows from actually consuming the diet provided at this time. For instance, feed intake may vary dramatically between animals due to individual differences in body condition (Garnsworthy and Topps, 1982) and social dominance (Friend and Polan, 1974).

Also of relevance is the dramatic decline in feed intake that occurs over the three weeks prior to calving (e.g. Bertics et al.,1992, Rabelo et al., 2003). This phenomenon has been identified as a likely contributing factor to the high incidence of health problems at this time (Van Saun and Sniffen, 1996). While most nutritional-health research in dairy cattle has

focussed on metabolic diseases, nutrition during the transition period may also play a role in infectious disease. Limited feed intake during transition may be partly responsible for sub-optimal immune function, and therefore may increase susceptibility to infectious diseases such as mastitis or metritis (Cai et al., 1994). Alternatively, both appetitive and immune depression during transition may be caused by a third factor, namely endocrinal and metabolic changes, which are discussed below.

The mechanism responsible for the reduction in appetite during the transition period is not well understood, but several possible factors have been identified: 1) increased foetus/uterine size, causing decreased rumen space (Forbes, 1968), 2) increased estrogen secretion (e.g. Bremmer et al, 1999) 3) the presence of accumulated body-fat reserves (Bines and Morant, 1983), and 4) a suite of endocrinal and metabolic factors (Ingvartsen and Boisclair, 2001).

Historically, the theory of decreased available gut space has been well accepted, although the scant available literature yields contradictory conclusions. While one study indicated that the size of the uterus plays a role in depressing appetite in late pregnancy in sheep (Forbes, 1968), other work in dairy cattle has demonstrated that this factor has little effect (Coppock et al, 1972).

Another common hypothesis involves the role of estradiol, serum concentrations of which begin to increase approximately three weeks before calving and peak dramatically over the five days before calving. Negative dose-dependant relationships between estradiol and feed intake have been reported for lactating goats (Forbes, 1986) and castrated male sheep (Forbes, 1974). Bremmer et al. (1999) also reported a significant drop in dry matter intake of lactating cows over two weeks following injection with estradiol. However, they found no such effect in non-lactating animals and suggested that the observed effect in

lactating animals was contingent upon the drop in milk yield observed after estrogen treatment. Although the supposed effects of estrogen are not sufficiently documented or well understood, it has been hypothesized that estradiol may act on the hypothalamus to depress intake (Forbes, 1986). High concentrations of plasma estradiol decrease adipose storage in rats (Hamosh and Hamosh, 1975) and increase hepatic triglyceride production; the increased availability of these fuel sources could play a role in depressing appetite (Sawchenko and Friedman, 1979).

Other literature suggests that the decline in feed intake depends on the amount of body fat present on an animal at the onset of transition (Bines, 1976; Bines and Morant, 1983; Grummer, 1993). One study (Bines and Morant, 1983) found that body condition score at calving was negatively correlated to feed intake after calving. Fatter cows apparently have a weaker appetite after calving than do thinner cows and subsequently reduce their feed intake accordingly (Bines and Morant, 1983).

Ingvartsen and co-authors discuss at length the factors thought to mediate the mobilization of accumulated body reserves and appetite during the transition period. In a series of papers, they present evidence that each of the following factors plays a role: stress hormones (e.g. corticotropin-releasing factor), metabolism-regulating proteins (e.g. glucagon, leptin, insulin, cholecystokinin, and somatostatin) and immune system proteins (e.g. cytokines) (Ingvartsen and Anderson, 2000; Ingvartsen and Boisclair, 2001; Ingvartsen et al., 2003).

Feeding Behaviour as an Indicator of Disease

While the decline in feed intake that occurs before calving may be partially responsible for the high incidence of metabolic and infectious disease during transition, an active immune

system in animals that become sick may act to further depress appetite (Hart, 1988). For this reason, cows that are already eating poorly during transition may continue to experience depressed appetite into lactation. Veterinarians often use some informal assessment of appetite as part of their examination (e.g. Rosenberger, 1979), and there is considerable evidence that the depression of appetite in sick animals is caused by the secretion of cytokine proteins (specifically, interleukin-1) by immune cells (e.g. Kent et al., 1992). However, little literature exists to demonstrate the relationship between appetite and disease in dairy cattle. Previous work has indicated that cows with lower feed intakes are more likely to be diagnosed with metabolic and infectious disease during transition (Marquardt et al., 1977; Zamet et al., 1979; Hammon et al., 2004). However, changes in feed intake must ultimately result from changes in feeding behaviour. Moreover, feeding behaviour has been shown to predict morbidity in feedlot steers (Sowell et al., 1998; 1999) and may be similarly useful for prediction of disease in transitional dairy cows.

Hypothesis

The aim of this study was to test the prediction that cows that spend relatively little time feeding during the transition period are at increased risk of metritis after calving. We hypothesize that animals that exhibit lower or reduced feeding behaviour over the entire transition period are more likely to be diagnosed with metritis.

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CHAPTER 2: FEEDING BEHAVIOUR IDENTIFIES DAIRY COWS AT RISK FOR METRITIS[1]

Introduction

Early identification or prediction of disease can minimize the duration of ill health and curb any resulting economic losses. The transition period, defined by Grummer (1995) as beginning 3 weeks before and ending 3 weeks after calving, is a time when dairy cattle are at high risk for both metabolic and infectious disease (Drackley, 1999). Therefore an improved ability to identify or predict disease in transition cows may be especially useful.

Veterinary examination is the gold standard of disease detection, but such exams are relatively infrequent on most dairy farms (commonly once every two weeks post-partum) and many cases of disease may go unnoticed. Producers can use changes in milk production as well as urine or milk tests to monitor the health of their animals, but these tools have drawbacks. The amount of milk production corresponds poorly with recognition of mild or sub-clinical infectious disease. In fact, Rajala-Shultz et al. (1999) found that cows with a mild fever produced more milk, on average, than did healthy cows. Frequent administration of tests on a herd-wide scale can also be costly and time-consuming. Moreover, no such tests are available for diagnosing inflammatory uterine disease (i.e. metritis or endometritis), one of the most common disorders after calving. Thus a practical method for continuously monitoring transition cows to assess their health status or risk for disease would be beneficial.

[1] A version of this chapter has been accepted for publication in the Journal of Dairy Science. Authors: G. Urton, M.A.G von Keyserlingk, D.M. Weary.

Inflammatory uterine disease frequently occurs soon after calving and may severely compromise reproductive performance (Fourichon et al*.,* 2000). Metritis and endometritis refer to the inflammation of the uterus and of its endometrial lining, respectively. Both diseases are a consequence of sustained infection of the uterus caused by pathogenic bacteria such as *Arcanobacterium pyogenes* (LeBlanc et al., 2002). The distinction between these diseases is of little importance to this paper so below we refer to both conditions as metritis.

The incidence of metritis ranges from 10.1% to 65.5% in dairy cows (e.g. Borsberry and Dobson, 1989; Hirvonen et al., 1999). This wide range in incidence can be attributed in part to inconsistency in diagnosis and the lack of a clear definition of the disease in the literature (LeBlanc et al., 2002). Metritis may also form disease complexes with one or more other common transition period conditions including retained placenta, dystocia, abortion, left displaced abomasum, ketosis, milk fever, and ovarian cysts (Kaneene and Miller, 1995; Curtis et al., 1985). A lack of easily identifiable signs of metritis may also result in cases going unnoticed.

Much of the research on transition cows has focused on minimizing the incidence of disease in a preventive manner, through improved nutrition. Despite great efforts in this area the incidence of metritis remains high (e.g. Dohmen et al., 1995; Hirvonen et al., 1999). Currently, NRC (2001) recommends increasing the energy content of the pre-calving diet from 1.25 Mcal/kg DM (recommended during the “far-off” dry period) to 1.62 Mcal/kg DM for the 3 weeks before calving. This strategy is thought to prepare the cow for the metabolic demands of early lactation and thereby minimizing the need for body tissue mobilization. However, feed intake at this time may vary dramatically between animals due to individual differences in condition (Garnsworthy and Topps, 1982), social dominance

(Friend and Polan, 1974) and likely other factors. Previous work has indicated that cows with lower feed intakes are more likely to be diagnosed with metabolic and infectious disease during transition (Marquardt et al., 1977; Zamet et al., 1979). However, changes in feed intake must ultimately result from changes in feeding behaviour. Feeding behaviour has been shown to predict morbidity in feedlot steers (Sowell et al., 1998, 1999; Quimby et al., 2001) and may be similarly useful for prediction of disease in transition dairy cows.

The aim of this study was to test the hypothesis that cows exhibiting lower or reduced feeding behaviour during the prepartum transition phase are at increased risk of developing metritis after calving.

Materials and Methods

The experiment was conducted at The University of British Columbia's Dairy Education and Research Centre in Agassiz, BC. In this study, 6 Holstein heifers (BCS at the start of the study = 3.7 ± 0.06; Mean± SD) and 20 Holstein cows (parity 1-3 and BCS 3.6 ± 0.13 at the start of the study) were housed in a freestall barn. Animals were monitored first in a pre-calving pen beginning 3 weeks before the expected date of calving and then in a post-calving pen until 3 weeks after calving. Each of the pens contained 12 stalls deeply bedded with sand and 7.2 m of feed alley space. Stocking density of these pens was maintained at 100% (i.e. 12 cows per pen). When animals demonstrated signs of imminent calving (e.g. swollen vulva, mucus discharge from vulva, enlargement of the udder), they were moved to individual straw-bedded maternity pens; within 24 h after calving, cows were moved into the post-calving pen. Group size was kept constant but group composition was dynamic as animals were moved between pens as they progressed through the transition period.

Animals were fed twice daily and given *ad libitum* access to water. While housed in the maternity pens, animals were fed from a 1m-wide plastic food bin. Starting 3 weeks before expected calving date, cows were fed a TMR consisting of 25.8% alfalfa hay, 47.9% corn silage and 26.3% protein and mineral supplement (Table 1) per cow on a DM basis (DM 41.12%, CP 14.97%, ADF 20.02% and NDF 33.47%). After calving, cows were fed TMR consisting of 7.2% grass hay, 1.8% alfalfa hay, 25.6% corn silage, 11.2% grass silage, 15.6% corn grain, and 38.6% protein and mineral supplement (Table 1) per cow on a DM basis (DM 43.2%, CP 17.53%, ADF 16.59% and NDF 27.87%).

Table 2-1. Ingredient composition of protein and mineral supplements fed to pre-calving and post-calving Holstein cattle.

Pre-calving		Post-calving	
Ingredient	%	Ingredient	%
Corn distillers	20.0	Corn (finely ground)	48.1
Ground Barley	30.6	Megalac[3]	1.2
Canola meal	13.7	Canola meal	8.2
Soybean meal	12.6	Alimet[4]	0.2
Amipro[1]	10.0	Soybean meal	30.0
Ground limestone	3.4	Urea	1.0
Yea-Sacc Farm Pak[2]	3.0	Dicalcium Phosphate	1.1
Magnesium Oxide	1.3	Calcium Carbonate	3.9
Blood meal	1.0	Magnesium Oxide	0.5
Molasses	1.0	Yea-Sacc Farm Pak[2]	0.4
Dicalcium phosphate	0.2	Tallow	1.4
Vitamin/ trace mineral premix	3.2	Salt	1.2
		Sodium Bicarbonate	0.3
		Vitamin/ trace mineral premix	1.5

[1] Unifeed Ltd., Chilliwack, BC, Canada

[2]Alltech, Inc., Nicholasville, KY

[3] Church and Dwight Co., Princeton, NJ

[4] Novus International, Inc., St. Louis, MO

Feeding Behaviour

An electronic system (Growsafe Systems Ltd, Airdrie, AB, Canada) was used to continuously monitor the presence of individual cows at the feed alley in the pre-calving and post-calving pens. This system, previously described and validated by DeVries et al. (2003a; b), scanned for the presence of each animal's transponder when within 0.5 m of the feed alley floor with a read-rate of once every 4.4 s in the pre-calving pen and once every 5.8 s in the post-calving pen. The number of times that each transponder was read was multiplied by the corresponding read-rate to calculate the time each animal spent at the feed alley in a given period.

The day before calving, the day of calving and the two days following calving (d -1, 0, 1, 2) were excluded from analysis due to the absence of a considerable number of animals from the trial pens on these days. The initiation of feeding behaviour data collection varied as a result of the difference between projected and actual calving dates of the animals, but data from all animals was available from d -12.

Individual Animal Factors

The body condition score (BCS; Edmonson et al, 1989; Appendix I) of each cow was evaluated once every 4 ± 1 d by a trained observer. Mean BCS over the pre-calving period was calculated from scores obtained from d -17 to -1 pre-calving and mean BCS over the post-calving period was calculated from scores obtained from d 1 to 21 post-calving. The change in BCS over the transition period was calculated as the difference between these two scores.

Individual milk weights were recorded for every cow at each milking for 21 d post-calving. Protein and fat content of milk was tested at 36 ± 9 d post-calving (DIM ± SD). Parity of

each animal was recorded as either primiparous or multiparous after the experiment. The discrepancy between actual and predicted calving dates also resulted in a range (12-39 d) of time spent on the pre-calving diet (22 ± 6 d, mean ± SD) so this variable was also included in the analysis.

Metritis Diagnosis

Vaginal discharge (**VD**) was observed every 4 ± 1 d after the morning milking (between 08:30 and 10:00 h). Before palpation, the vulva of each cow was cleaned thoroughly with diluted Betadine solution (Purdue Pharma, Pickering, ON, Canada). Discharge was removed by a gloved hand from the opening of the cervix and then examined; the color and smell of the discharge and the relative amount of pus and mucus present were noted. Rectal body temperatures (**BT**) were measured daily immediately after the morning milking with a digital thermometer (GLA M525/550, GLA Agricultural Electronics, San Luis Obispo, CA, USA).

VD was assigned to a category using the following scoring system adapted from Dohmen et al. (1995): no mucus, clear mucus, cloudy mucus or mucus with flecks of pus = 1, mucopurulent and foul smelling = 2, purulent and foul smelling = 3, putrid (reddish brown, watery, foul smelling) = 4. Fever was classified as BT>39.5°C.

As there is disagreement in the literature concerning which diagnostic criteria constitute a case of metritis, we employed two classifications. Animals were classified as Metritic if they showed a VD of 2 or more plus fever (within 3 d before observation of VD ≥2) or Acutely Metritic if they showed a VD of 4 plus fever (within 3 d before observation of VD ≥2).

Statistical analysis

All statistical tests were performed using SAS (The SAS Institute, Inc., Cary, NC, USA) with the individual animal as the observational unit. We designated three different periods for analysis: a pre-calving period (days -9 to -2), a post-calving period (days 2 to 19) and a trial period (pre-calving and post-calving periods combined). A preliminary period (days -12 to -10) was also designated and tested as a covariate. We determined correlations between each of these feeding behaviour variables and our body condition score variables (BCS and change in BCS). Effects of parity group on incidence of Metritis and Acute Metritis were determined using a Fisher Exact test. Differences in feeding behaviour between classes of metritic and non-metritic cows were tested using t-tests. Multivariable logistic regression was performed using the LOGISTIC procedure in SAS to model the effects of feeding behaviour and other factors (parity group, BCS, number of days on the pre-calving diet, and presence or absence of assisted calving) on the presence or absence of Metritis and Acute Metritis. We performed a manual backward stepwise procedure, sequentially removing the least significant variable until only variables with $P < 0.1$ remained.

A range of daily mean feeding times were tested as thresholds for identification of metritic animals. Suitability of each threshold was determined by calculating the sensitivity and specificity of each threshold. Sensitivity was defined as the percentage of metritic animals that demonstrated a sub-threshold daily mean feeding time. Specificity was defined as the percentage of non-metritic animals that demonstrated an above-threshold daily mean feeding time.

Results

Examination of vaginal discharge over the 21 d after calving indicated that 18 cows (69%) experienced some degree of pathological discharge (VD≥ 2). Specifically we noted putrid discharge in 38% of cows, purulent and foul smelling discharge in 27% of cows, and mucopurulent discharge in 4% of cows (one animal). The number of days from calving to the first sign of pathological discharge (VD ≥ 2) ranged from 3 - 15 DIM. The average body temperature (± SD) of all cows over 21 d post-calving was 39.0 ± 0.04°C. The average BCS (± SD) of all cows over the pre-calving and post-calving period were 3.69 ± 0.07 and 3.47 ± 0.07, respectively. Of 6 primiparous cows, 4 were diagnosed with Metritis; statistically, the effect of parity was not significant for Metritis (P = 0.371), but there was a tendency for primiparous cows to be diagnosed with Acute Metritis (P = 0.074).

On average, cattle decreased the time spent at the feed alley over the pre-calving period (by 35% or 32 min; Figure 2-1), and then increased time at the feed alley over the post-calving period (by 99% or 46 min). Mean BCS over the pre-calving period was negatively correlated with trial period feeding time (R^2 = -0. 43, *P* = 0.027) but less so with pre-calving feeding time (R^2 = -0.32, *P* = 0.116). Change in BCS over transition was also negatively correlated with trial period feeding time (R^2 = -0.40, *P* = 0.043) and to a lesser extent with pre-calving feeding time (R^2 = -0.37, P = 0.062). No other animal factors were correlated to feeding behaviour.

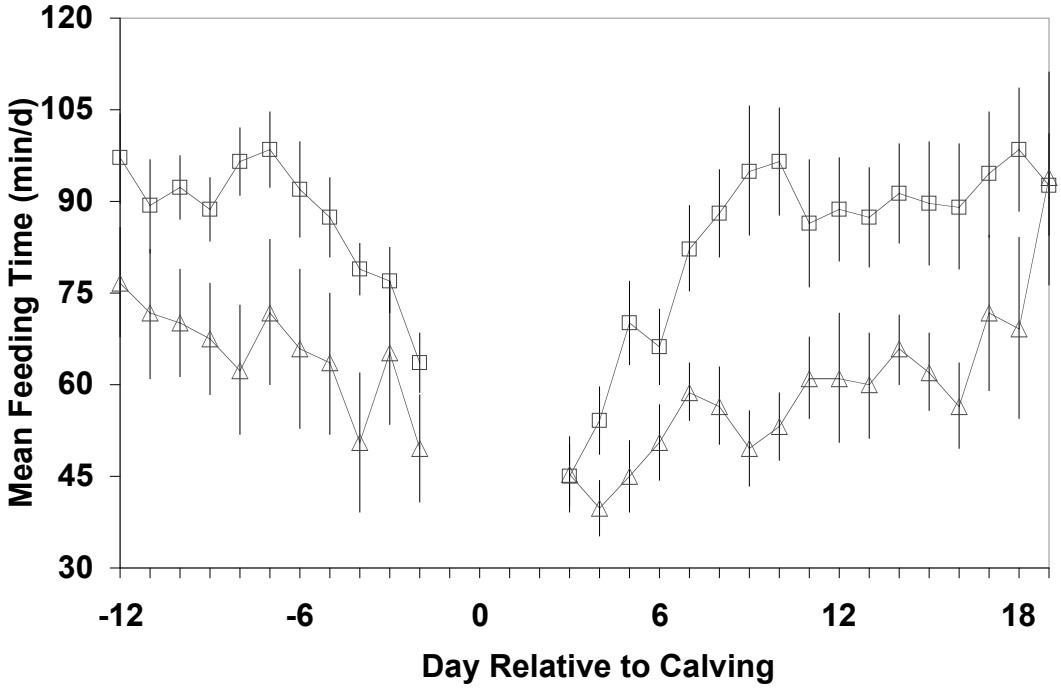

Figure 2-1. Daily mean feeding time (min d^{-1}) of 9 Holstein cows with acute metritis (∆) and 17 Holstein cows without acute metritis (_) ±SE from 12 d before calving until 19 d after calving.

Cows diagnosed with either Metritis or Acute Metritis spent less time feeding over the trial period than non-metritic cows ($P < 0.01$; Table 2-2). These cows also spent significantly less time feeding over the post-calving period than did their healthy counterparts. However, only those cows diagnosed with Acute Metritis showed significantly ($P < 0.05$; Table 2-2) lower feeding time during the pre-calving period.

Table 2-2. Mean and SED[1] feeding times (min d^{-1}) for Holstein cows with and without Metritis and Acute Metritis over four periods[2] relative to calving.

Period	Metritis			Acute Metritis		
	Present (n=14)	Absent (n=12)	SED	Present (n=9)	Absent (n=17)	SED
Preliminary	79.1	93.9	9.60	72.9	92.8	9.73†
Trial	65.4	87.6	7.41**	59.9	83.9	7.66**
Pre-calving	68.9	87.0	3.99†	62.1	85.3	9.16*
Post-calving	73.5	87.9	8.35**	58.8	83.3	8.85*

[1] SED = Standard error of the difference between mean feeding times

[2] Preliminary period = d-12 –d-10; Pre-calving period = d-12-d-2; Post-calving period = d2-d19; Trial Period = Pre-calving and Post-calving periods combined

†$P \leq 0.10$

* $P \leq 0.05$

** $P \leq 0.01$

Logistic regression showed that trial period feeding time accounted for a significant amount of the variation in risk of Metritis and Acute Metritis. Odds of Metritis increased by 1.97 times for every 10-min decrease in average daily trial period feeding time (P_{Wald} = 0.022; 95% confidence interval (**CI_{95}**): 1.97, 4.02). Odds of Acute Metritis increased by 2.08 times with every 10-min decrease in average daily trial period feeding time (P_{Wald} = 0.013; CI_{95}: 1.24, 4.32). None of the covariates were identified as significant for the Metritis model, but for the Acute Metritis model the effect of parity was significant (P_{Wald} = 0.038), with heifers showing a higher risk.

Feeding behaviour would be most useful if measures pre-calving could identify animals at risk for metritis. Pre-calving feeding time was able to account for a significant proportion of variance in Acute Metritis (P_{Wald} = 0.018), with the odds of a positive diagnosis increasing

by 1.57 times for every 10-min decrease in pre-calving feeding time. Parity was again the only significant covariate (P_{Wald} = 0.040; CI_{95}: 1.08, 2.55) in the model.

Measures most useful for identifying animals at risk for metritis should be both sensitive and specific. The most useful threshold of daily pre-calving and trial period feeding time was 75 min, demonstrating a moderate to high sensitivity and specificity for detection of Acute Metritis and moderate sensitivity and specificity for detection of Metritis (Figure 2-2).

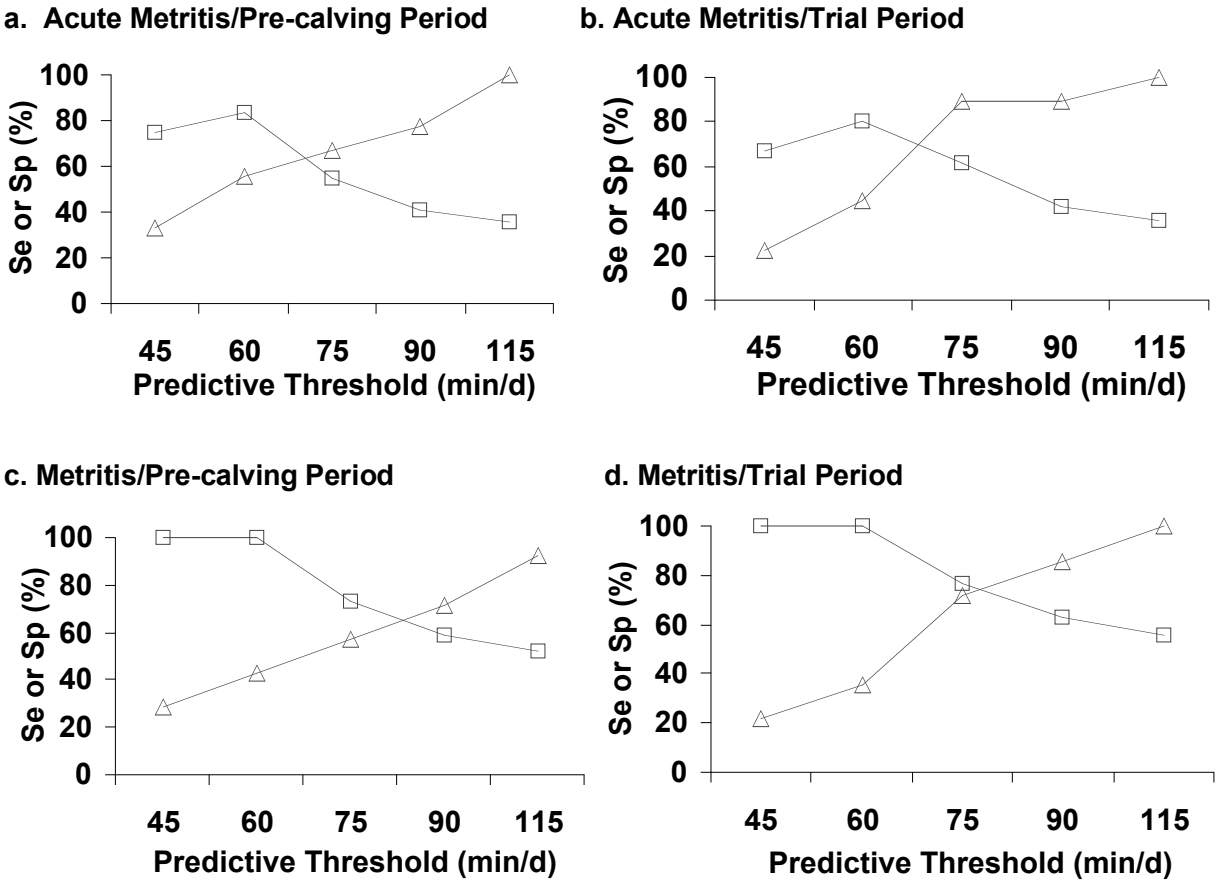

Figure 2-2. Sensitivity (Se; ∆) and specificity (Sp; _) of predictive thresholds of mean daily feeding time (min d^{-1}) in the pre-calving and trial periods to identify Holstein cows with metritis or acute metritis.

Discussion

The proportion of animals showing signs of metritis in the present study was similar to that reported by Dohmen et al. (1995; 80%) and Hirvonen et al. (1999; 66%) who used similar scoring criteria and a comparable day of diagnosis (14 DIM and 4 – 11 DIM, respectively). Le Blanc et al. (2002) conducted a validation of diagnostic criteria for metritis and found that presence of purulent (or worse) discharge after 20 DIM identifies endometritis associated with compromised reproductive function in the dairy cow. For this study, the definitions of metritis were not limited to include only those cases still present after 20 DIM; the interest was in metritis as a systemic infectious disease, not as a risk factor for compromised reproductive function.

Animals decreased their feeding time by 35% over the 2 weeks before calving and increased feeding times by 99% over the 3 weeks after calving. This paper is the first to describe these changes in feeding times over this period, although others have observed a similar pattern in DMI. For example, Zamet et al. (1979) described an unspecified drop in DMI from 28 to 7 d pre-calving, followed by a 30% drop over the week before calving. Bertics et al. (1992) noted a 30% decline over the 3 weeks before calving with the majority of this drop occurring during the last week. Other authors have also documented a doubling in DMI over the 3 weeks after calving (Bertics et al., 1992; Vazquez-Anon et al., 1997).

High BCS before calving has been identified as a factor contributing to the pre-calving decline in feed intake (Bines and Morant, 1983). Our results support this, indicating that over-conditioned cows generally eat less than thinner cows over the transition period. Our data also indicate cows that lose body condition during transition spend less time feeding over this period. Factors thought to mediate the mobilization of accumulated body

reserves and appetite during the transition period are discussed in depth by Ingvartsen and co-authors (Ingvartsen and Anderson, 2000; Ingvartsen and Boisclair, 2001; Ingvartsen et al., 2003).

This study is the first to our knowledge to show how disease relates to feeding behaviour of dairy cows. Conventional feed-intake monitoring systems restrict social interaction between animals while feeding, whereas the system we used allowed cows to interact normally at the feed bunk. Zamet et al. (1979) monitored the health and feed intake of free-stall housed cows over the transition period. Although these researchers noted a 21% lower DMI in metritic cows after calving (4 to 30 DIM), they failed to detect differences in DMI between these two groups before calving. Our work also showed that cows diagnosed with metritis had 29% lower feeding times after calving, but we were also able to show a difference in pre-calving feeding times between healthy and metritic cows. This result suggests that feeding behaviour can be a more sensitive indicator of disease than measures of individual feed intake. Indeed, the same behaviour monitoring system used in the current experiment has been used to successfully identify sick feedlot housed beef steers (Sowell et al., 1998, 1999; Quimby et al., 2001). Sowell et al. (1999) found that time spent feeding was lower (by as much as 35% over 4 d) for steers demonstrating visible signs of bovine respiratory disease than for healthy animals. This figure is comparable to our overall difference in time spent feeding (29%). Quimby et al. (2001) were able to use depressed feeding behaviour to identify morbid steers 4.5 days earlier than were animal handlers. This study used cumulative sums analysis that requires a period of data stability to establish control parameters. This method would likely not be suitable for the relatively unstable feeding behaviour characteristic of the transition period for dairy cows.

Our feeding behaviour measures compare favourably in sensitivity and specificity for metritis detection to other tests used for disease detection in dairy cattle. For thresholds that were both suitably sensitive and specific (>60%), our sensitivity ranged from 71% to 89% and our specificity ranged from 62% to 77%. Similarly, suitable thresholds for on-farm milk and urine ketosis tests range in sensitivity from 76% to 80% and in specificity from 76% to 93% (Geishauser et al., 1998). A more sophisticated laboratory test for ketosis using infrared milk analysis achieved high sensitivities of 79% to 100% and specificities of 85 to 100% (Heuer et al., 2001). In comparison, suitable somatic cell count thresholds for mastitis detection were only 61% to 64% sensitive and 65% to 70% specific in one study (Sargeant et al., 2001).

Further work is needed to understand the factors that mediate the link between feeding behaviour and diseases prevalent during transition, including the relationship between feeding behaviour and feed intake. Other behavioural measures may also be helpful in improving our ability to identify animals at risk for metritis and other disease common at transition. Indeed, previous researchers have suggested that a variety of animal factors (e.g. feed intake, milk yield, walking activity) may be useful in the early disease detection for transition cows (Bareille et al., 2003; Edwards and Tozer, 2004), but further empirical work is needed to test these ideas.

Acknowledgements

The authors gratefully acknowledge the support of staff and students at the University of British Columbia's Dairy Education and Research Center, and the insightful comments of members of the UBC Animal Welfare Program at various stages of the project. We are especially grateful to Dr. Andreia Vieira for her help in a preliminary trial and in developing the criteria used in this study. We also thank Agriculture Canada for providing the feeding

behaviour monitoring equipment and Drs. Brian Upper and Raja Rajamahendran for the diagnostic training they provided. Financial support for was provided by the Natural Sciences and Engineering Research Council of Canada, the Dairy Farmers of Canada, the BC Dairy Foundation, the BC Society for the Prevention of Cruelty to Animals, members of the BC Veterinary Medical Association and many others listed at www.agsci.ubc.ca/animalwelfare.

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CHAPTER 3: GENERAL CONCLUSION

Animal welfare science is generally devoted to assessing animals used by humans in various settings (e.g. farming, laboratory, zoo, wild, and companion). These assessments are normally based on physiological or behavioural parameters and allow researchers to determine how environmental conditions, management practices, and procedures affect an animal's well-being. Once problems have been identified, methods of remediation are often proposed and tested. Some animal welfare research also aims to identify and validate new methods of welfare assessment in order to facilitate other research or to provide practical methods of *in situ* assessment. The research conducted for this thesis is of this type. We have proposed a novel method of identifying dairy cows at risk for a known source of poor welfare (metritis) – namely, measuring feeding behaviour over the transition period. This behavioural method of animal assessment may serve as a practical on-farm method of monitoring cows for metritis and possibly other diseases. It also provides a means for identifying animals at risk, which should facilitate disease prevention.

Unfortunately we were not able to gather detailed information on the feeding behaviour of cows immediately around the time of calving, as the animals were moved from the research area to maternity pens. As feed intake at this time may be crucial to a cow's successful transition into lactation, it would be beneficial to develop a research protocol that allows feeding behaviour to be monitored through this period as well. The addition of these data would enable researchers to gain further insight into the changes in feeding behaviour during transition and the use of this measure in identifying animals at risk for disease. Given this additional data, one might find that precalving measures of feeding behaviour bear a more significant relationship with the health parameters we measured.

The relatively few animals studied in this experiment (26) may have hindered our ability to establish with certainty certain relevant relationships between some variables. For example, we identified several non-significant trends in our data, including the relationships between parity and acute metritis, between BCS and precalving feeding behaviour, and between metritis and precalving feeding behaviour. These trends may have proven significant given a larger sample size.

While this thesis provides evidence for a relation between transition period feeding behaviour and metritis, it does not demonstrate cause and effect. As is described in Chapter 1, the depression of appetite, decreased immune function, and the high incidence of infectious disease characteristic of the transition period are likely related. However, the mechanisms at work are complex and are not well understood. Research that improves our understanding of these mechanisms may allow us to prevent disease. Specifically, further work is required to ascertain the relationship between feeding behaviour and feed intake and to understand how these variables relate to metabolic and immune status. Research that incorporates the measurement of immune mediating factors (i.e. cytokines) and by-products of catabolism (i.e. plasma non-esterified fatty acids) with the monitoring of feeding parameters and disease incidence is particularly encouraged.

Lastly, research that investigates the use of alternative management strategies for the transitional cow would also be beneficial. Rastani et al. (2005) recently demonstrated that cows who are not provided with a dry period (continuously milked and fed a high energy diet until calving) coped better with transition than did cows on either a traditional 56-d or a shortened 28-d dry period. The continuously milked cows demonstrated better transition period feeding habits – namely, a less severe drop in prepartum DMI, a lower absolute transition DMI, and a quicker postpartum recovery to peak DMI. They also appeared to

cope better metabolically with the onset of lactation (lower plasma NEFA and liver triglyceride concentrations and a negligible negative energy balance) than did either group of dried-off cows. However, peak milk production of the continuously milked cows was significantly lower than that of the other groups, resulting in a 10 - 15% lower milk yield. Further work investigating the effects of using different dry period lengths on feeding parameters, immune and metabolic status, and disease incidence could identify additional benefits of a shortened dry period.

The findings of Rastani *et al.* (2005) provide a good example of the need for alternative management practices that improve animal welfare without compromising production. Clearly, management practices that improve welfare while maintaining the economic viability of the operation are invaluable as they are far more likely to be adopted. Applied welfare research must take this into consideration in order to have the greatest impact on farm animal welfare in practice.

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APPENDIX I: BODY CONDITION SCORE SCALE

The BCS scale used was that described by Edmonson et al (1989). The scorer assessed the following 8 body regions of each cow:

- spinous processes
- region between spinous and transverse processes
- transverse processes
- overhanging shelf of transverse processes
- tuber coxae (hooks) and tuber ischii (pins) of the pelvis
- region between each pelvic hook and pin
- region between the hooks
- region between the tail-head and pelvic pin

The scorer assigned an overall body condition score to the animal on a scale of 1 to 5, using 0.25 increments, where:

1 = severe underconditioning (emaciated)

2 = definition of the body frame is obvious

3 = body frame and fat covering are well-balanced

4 = body frame is less visible than fat covering

5 = severe overconditioning (obese)