Impact of relocation on dairy cows

Crafton Oliver Wilkes

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Committee:

Dr. Frank C. Gwazdauskas, Chair
Dr. Michael L. McGilliard
Dr. Ray L. Nebel
Dr. R. Michael Akers, Department Head

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ABSTRACT

Several indicators of animal welfare were measured to determine the impact of relocation on lactating dairy cows. In experiment one, locomotion was scored on a 4-point scale where 1 = normal and 4 = lame. Cleanliness was scored on a 4-point scale where 1 = little or no manure visible and 4 = heavy plaques of manure on 3 body regions: udder, lower leg, and flank/upper leg. In experiment two, milking parlor behaviors observed were cow reactivity (REACT), latency to enter the parlor (LAT), and plasma cortisol (CORT). REACT was evaluated on a numeric scale (0 = ideal milker to 3 = steps and kicks frequently) to define behavior during udder preparation, claw fitting, and milking. LAT was the time necessary for each cow to enter the milking parlor. In these studies, the effects of treatment or breed on MY, lameness, parlor behaviors, stress, and cow cleanliness were monitored. In experiment one, cows with access to a rubber mat in front of the feed bunk had lower locomotion scores and cleaner lower legs. There were no breed effects on locomotion, but Jerseys had cleaner lower legs than Holsteins. The effect of the new facility on locomotion occurred gradually. Cows with higher locomotion scores had decreased milk production, but the results were not significant. In experiment two, relocation caused an increase in plasma cortisol and LAT. Milk yield decreased the p.m. on the day of relocation, but overall milk yield was not affected. Jerseys had lower plasma cortisol levels and latency to enter the parlor, but had a greater decline in p.m. milk yield the day of relocation. In conclusion, alternative flooring may alleviate some locomotion problems caused by relocation to a new facility. Additionally, moving to a new facility may have an effect on behavior and stress, but these effects are short-lived and effects on overall milk production minimal.

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Introduction

The Virginia Tech Dairy Center updated its facilities in the summer of 2004, and with the opening a unique opportunity was made available. Studies have investigated the effect of transport and distance traveled (Dixit et al., 2001) and stress induced by being milked in an unfamiliar setting (Rushen et al., 2001). And while different flooring systems have been examined to determine their effect on locomotion and lameness (Vaarst et al., 1998), there is little research regarding the effect a new dairy facility has on the welfare of cows and the duration of these effects.

The cause of lameness is multifaceted: diet (Donovan et al., 2004), housing (Cook et al., 2004), and exposure to moisture (Borderas et al., 2004) have been linked to diminished foot health. Before the current study, the hooves of all cows in the herd were examined and trimmed by a veterinarian and treated for any disorders. Post-trimming, cattle locomotion was observed regularly using a quantitative scale described by Sprecher et al. (1997). In addition to locomotion scoring, cow cleanliness was scored (Cook, 2002) and both were used to determine how the new facility affected cow welfare.

Stress can be measured numerous ways. In lactating cows, increased stereotypies such as tongue rolling, exploration, and agonistic behaviors (Schrader, 2002), or increased movement in the milking parlor (Hillerton et al., 2002) indicate increased stress. Physiologically, increased plasma cortisol (González et al., 2003) and blood leukocyte counts (Kent and Ewbank, 1986) are correlated with stressful situations. The current experiment looked to compare physiological and behavioral stress indicators and their effect on milk production.

The objectives of this research were to:

1) To determine the effect relocation to a new facility had on locomotion and hoof health in dairy cows, and if a rubber mat placed in front of the feed bunk altered these effects.
2) To determine the effect relocation to a new facility had on stress and milking parlor behaviors in dairy cows.

Chapter 1: Literature Review

1.1 Overview of Locomotion, Lameness, and Cleanliness

Lameness in dairy cows is a costly health problem for both the individual producer and the dairy industry. Lame cows contribute to monetary losses through treatment costs, decreased performance, increased culling, and reduced cull value (Hernandez et al., 2001). Relocation to new facilities can increase locomotion score and incidence of lameness. This may be associated with more textured flooring (Phillips and Morris, 2001), diet (Manson and Leaver, 1988), or activity changes in free stalls (Cook et al., 2004).

The effect of surface conditions on cow slippage and locomotion was examined by Phillips and Morris (2000). Dry, wet, and shallow (5 cm) or deep (12.5 cm) slurry from cattle excreta were used to cover a 20 x 3 m concrete aisle that cows were taught to walk down for a food reward. A switchback design was used to account for the effects of learning. Each cow was monitored 3 times on each surface by human observation and video recorder placed ¾ down the last 7 m of the aisle. Stride length, frequency, and various joint angles were recorded. Spots of blue paint were placed on the elbow, carpal, metacarpo-phalangeal joints of the forelimb and the stifle, tarsal, and metatarso-phalangeal joints for ease of identification. Wet concrete had no effect on the walking rate, stepping rate, or stride length of the cows compared to dry concrete. However, it did decrease the angle of the forefoot to the floor and reduce the arcs made by the hind limb joints, suggesting that cows found the surface more slippery. Slurry reduced walking rate and stepping rate, but cows in deep slurry increased step length. Arcs in hind- and forelimb joints were reduced in both shallow and deep slurry, but this was attributed to difficulty in removing the limbs from the deep slurry. Slurry reduced slippage and altered the walking pattern of the cows compared to dry or wet concrete.

The walking behavior of dairy cows on surfaces with different friction coefficients was examined to determine the optimal roughness for barn flooring (Phillips and Morris, 2001). The coefficients were measured with a weighted platform with four hooves underneath to simulate cow movement. Five non-lactating British-Friesian cows were used with four different floor treatments. The treatments used were: epoxy resin with no aggregate, and 0.5, 1.2, and 2.5 mm aggregates imbedded in the epoxy. Each cow was recorded 15 times on each surface; the cows were tested on 3 occasions and each occasion involved 5 circuits. The cows walked down an aisle approximately 15 m long and 1.3 m wide. The data recording method followed closely the procedure used by Phillips and Morris (2000) previously. Phillips and Morris (2001) concluded that floor friction has an effect on cattle walking patterns. Stepping rate increased for lower friction levels, while stride length increased for larger aggregates. The stifle, tarsal, and metatarso-phalangeal joints were more vertical on the floor with no aggregate, and least vertical on the floor with 0.5 or 1.2 mm aggregate. This indicates that the cows had a higher confidence in foot traction with higher friction flooring. The results of this study, along with previous studies (Phillips et al., 1998; 2000), suggest that the optimal friction coefficient for cattle floors is between 0.4 and 0.5.

Cow lying behavior, dirtiness, and locomotion were examined to determine the effect of four different flooring surfaces commonly used during stand off periods (Fisher et al., 2003). Thirty-two non-lactating Friesian cows were given access to a specially designed wood chip pad, a concrete yard, a gravel-surfaced farm laneway, and small paddock in a replicated 4 x 4 Latin square design. Cows were on each treatment for 4 d of each week, with a 3-d recovery period on pasture, for 5 wk per treatment. Heavy rainfall was simulated using high-pressure sprinkler systems placed around the stand off areas. Lying behavior was examined by video recorder while on the treatment and human observation was used during the daily 3-h grazing period. Frequency of lying, duration of each lying bout, and total lying time were recorded. Dirtiness was recorded from 35 different areas on the cow based on a 4-point scale established by Scott and Kelly (1989). Locomotion was measured by calculating gait length as cows walked individually down an 18.1 m farm laneway. Duration of lying bout was not affected by any treatment; however, the number of lying bouts was significantly higher for the wood chip pad than the other groups, with a mean of 8.1 ± 0.83 lying bouts at stand off. Cows on the wood chip pad also experienced greater total lying time and lower dirtiness scores. Gait length for the wood chip pad was not significantly different from the concrete yard, with scores of 0.66 and 0.64 ± 0.019 m, respectively. Gait length for both the farm laneway and the small paddock was 0.68 ± 0.019 m. It was concluded that the wood chip pad was best suited for extended stand off periods, but the laneway and paddock could have similar results with better drainage of the areas. The concrete yard was the least suitable because it shortened total lying time and decreased gait length, suggesting that hoof problems could arise if the cows spent more than the allotted 4 d on the surface.

In addition to stand off areas, freestall base material has been examined to determine its effect on lameness (Cook et al., 2004). Twelve Wisconsin dairy herds participated in the study; 6 herds had freestall barns with sand bedding vs. 6 herds with geotextile mattress bedding. Lying behavior, including total lying time, lying duration, perching, and time standing up, was recorded via four video cameras placed across the feed alley in the adjacent pen to get a full view of the treatment group. During the filming period, cows were given a locomotion score during a single milking. A 4-point scale was used, where 1 = nonlame, 2 = slightly lame, 3 = moderately lame, and 4 = severely lame (Cook, 2002a). Cows with a score of 3 spent less time lying in the stall and greater time standing in the stall than cows with a score of 1 or 2 with respect to both treatments. Cows exposed to mattress bedding spent more time standing in the stalls, less time feeding, and had fewer stall use sessions than cows with sand bedding. Additionally, cows on sand were less likely to alter their behavior due to lameness compared to cows on mattresses.

Aside from the variables studied by Cook et al. (2004), agonistic behaviors and displacements were examined to determine their effect on lameness (Galindo et al., 2000). Forty Holstein-Friesian cows were observed by human and video recorders daily for 8 d to determine displacement indices and weekly for 6 mo to record locomotion scores, clinical lameness cases, and freestall behaviors. Agonistic behaviors included butts to head, neck, and ribs; dominant tendencies such as chasing or threatening other cows; and submissive behavior such as active avoidance of other cows with no previous interaction. An index of displacement using the agonistic behaviors was created to determine high-, middle-, and low-ranking cows. Hoof scores for each cow were recorded initially, and individually if any clinical lameness cases arose during weekly visits. Locomotion scores were recorded as cows entered the milking parlor during weekly visits. Low-ranking cows spent more time standing still and standing halfway in the stall than the other cows. Cows diagnosed as lame were more likely to be lower ranking and spent more time standing halfway in a cubicle. It was concluded that cows that spent more time standing still were more predisposed to lameness, and low-ranking cows were more likely to stand still. Also, because lameness is caused by many variables, behavior and social dominance should be examined as possible sources.

While freestall surfaces and social rank have been linked to lameness, this may be a result of lying time budgeting, or more importantly, time standing on the concrete alleyway. The effect of rubber flooring in front of the feed bunk compared to grooved concrete on time allocation in cows was studied (Fregenosi et al., 2004). A herd of 48 dairy cows was divided into 4 groups of 12 and given access to either a 2.5 cm grooved rubber mat or grooved concrete in front of the feed bunk. Observations were recorded by video camera for 48 h/wk for 6 wk. Lying and standing behavior, social interactions, and time spent eating were recorded. Cows with access to rubber flooring did not spend more time eating than cows on concrete; however, they spent more time standing at the feed bunk, with a mean of 5.5% of their time, than the cows with concrete flooring that had a mean of 4.8% of their daily time. Cows with rubber flooring allotted 11.0% of their time to standing elsewhere, compared to 9.0% for cows with concrete flooring. The cows with concrete flooring spent more time lying in their stall. It was concluded that rubber flooring was slightly more comfortable to dairy cows, and further testing should be pursued to determine the effect on hoof-related injuries.

The interaction between alley flooring systems and stall bedding also has an impact on claw and leg health (Vokey et al., 2001). Alleyways were either grooved concrete or fitted 1.9-cm thick interlocking rubber mats covering all contact points. The stalls were either a concrete base with sawdust, a rubber mattress dusted with sawdust, or sand. Hoof scores were recorded on the hind legs at wk 1 and 15, and locomotion scores were recorded at wk 1, 5, 10, and 14. The locomotion score was on a 1 to 4 scale, where 1 = normal gait and 4 = one deliberate step at a time, abnormal gait (Manson and Leaver, 1988; Wells et al., 1993). Additionally, hock lesion and swelling scores were recorded during milking. Rubber alleys did not prevent clinical lameness or hock lesions, but did show potential for claw development when paired with sand stalls. In combination with cement stalls, rubber alleys were problematic because cows preferred lying in the alley, leading to increased incidence of mastitis. A rubber alleyway combined with an abrasive stall surface such as sand or concrete may alleviate some claw deformation, resulting in better hoof health. Others have also found improved claw health and reduced lameness with softer surfaces. Cows housed in deep litter straw-based systems show fewer incidences of claw disorders than cows housed on either slatted or grooved concrete (Somers et al., 2003). Similarly, deep litter systems resulted in fewer hoof and hock disorders in cows than tie-stall housing (Vaarst et al., 1998). However, all of the studies noted great variation among individual cows.

1.2 Overview of Behavioral Stress

Relocation to an unfamiliar environment presents a stressful situation that can impact dairy cow behavior. While the impact may not be permanent, this acute stress can decrease milk yields, increase defecation, urination, and vocalization (Rushen et al., 2001). Other changes that may occur during milking include reluctance to enter the parlor, an alteration in entry order into the parlor (Ceballos and Weary, 2002), and increased agitation indicated by kicking or stepping during pre-milk stripping or milking (Dickson et al., 1970; Hillerton et al., 2001).

A milking temperament score was used in a study as one parameter to determine social dominance among large groups of cows (Dickson et al., 1970). The 4-point scale ranged from a score of 1, a very quiet cow that was extremely docile during milking and the "ideal" milker; to 4, a cow that was restless during milking, appeared agitated, kicked at handler, and flinched when a hand was placed on her. In addition to the milking temperament score, observations of aggressive interactions in the barn during feeding, milk production, stage of lactation, age, height, and weight were used to establish dominance values in 27 Wisconsin dairy herds. Although no relationship was found between the temperament score and the other variables, many subsequent studies have used this scoring system to evaluate variables such as stress.

Entry order and reactivity were used as two measures of minimum requirements for the International Standards Organization in British milking parlors (Hillerton et al., 2001). The focus of this study was to determine the effect of two differently weighted and sized milking clusters on teat and udder health and cow behavior in the milking parlor. Eighteen dairies were evaluated twice each, however, only the second observations were reported. The reactivity scale used was similar to the milking temperament score used by Dickson et al. (1970). The differences between the two scoring systems were the inclusion of defecation and urination in the former study, a whole herd score, as opposed to scoring individual cows, and a 0 to 3 scale instead of the 1 to 4 scale used by Dickson et al. (1970). The heavier milking clusters resulted in higher, less satisfactory scores with a mean of 1.43 ± 0.37, compared to the 0.33 ± 0.37 mean of the common clusters. It was concluded that the increased score indicated discomfort and elevated stress.

Paranhos da Costa and Broom (2001) conducted an experiment to determine whether or not being milked on the non-preferred side of the milking parlor caused more stress or discomfort to cows. Side preference data were collected from 70 Holstein-Freisian cows during 40 milking sessions in a two-sided tandem parlor. Cows were grouped into three categories: high consistency in side choice (> 86%), some consistency (72 < SC < 85%), or low consistency

(< 71%). Cow reactivity in the parlor, fitting time of the milking cluster, milking duration, and milk yield were measured. Cow reactivity was observed continuously from the time of entry into the parlor until the milking had started, and fitting time was recorded using a stopwatch. The study found that there were individual differences in consistency of side choice and in cow reactivity, fitting time, milking duration, and milk yield. It also found that being milked on the non-preferred side of the parlor did not significantly affect the cow's welfare. Variability in side choice and consistency were attributed to a lack of symmetry in the milking parlor, natural laterality, and animal temperament.

Small amounts of concentrate were fed to cows in the milking parlor to determine the effects on entrance behavior (Ceballos and Weary, 2002). Groups of 17 cows were fed 0, 0.3, or 0.6 kg concentrate using a computer controlled feeding system. Entry order, latency to fully enter parlor, and defecation/urination in the parlor were recorded before and after treatment began. All data were recorded by one observer standing in the milking parlor. Cows were fed ad libitum while in the barn. Before treatment began, there were no significant differences between the three groups for any of the variables measured. After treatment, the cows that were fed concentrate while in the parlor had a lower mean entrance order and a reduced latency to enter the parlor. Cows receiving no concentrate in the parlor were more likely to require pushing into the parlor once treatment began. There was no effect of feeding on defecation or urination in the parlor. The amount of concentrate fed in the parlor had no effect on any of the variables. While stress was not one of the factors specifically examined in this study, it was noted that feeding concentrate in the parlor reduced the need for pushing or forcefully moving cows into the parlor, potentially leading to a less stressful environment.

Isolation from the herd was shown to increase defecation, urination, vocalization, and stepping during milking (Rushen et al., 2001). Eighteen cows were milked under each of three experimental settings: 1) milked in the usual setting; 2) milked alone in an unusual room; and 3) milked in unfamiliar room with human contact. Incidence of stepping was significantly higher in the isolated group, with a mean of 3.89 ± 0.47 min^{-1}, compared to 0.93 ± 0.47 min^{-1} for the control group, and 0.89 ± 0.47 min^{-1} for the human contact group. During the study, only one cow was seen defecating or urinating during milking in the control group, while 13 cows were observed defecating in isolation; human contact curbed this behavior to numbers similar to the control group. Total milk yields for the three groups were not significantly different, but this is due to a higher initial milk yield and lower residual milk yield in the control group compared to the isolation and human contact groups. Although a standardized behavior scale was not utilized for this study, it is clear that the findings correspond to those that did use a milking temperament score.

1.3 Overview of Physiological Stress

Stress has become an increasingly debated issue, as producers feel the negative effects on production from cows and concerns over animal welfare. Corticosteroids have been long been associated with stress; there are increased circulating concentrations during isolation, parturition, and transport (Varner et al., 1983; Moberg, 1991). During transport and subsequent relocation, cows may also experience increased plasma cortisol, lymphocytic ACTH, and heart rate (Boissy and le Neindre, 1997; Dixit et al., 2001).

The objective of a study by Varner et al. (1983) was to determine if relocating a herd would alter corticosteriod secretion or milk production. Two herds were examined; in the first herd, half (18 cows) were moved 100 m to a new facility (merged), while the remaining half stayed in their old tie-stall barn. In the second herd, 50 cows were transported 7 km to a new facility (moved). Blood samples were collected from the caudal blood vessel in the tail 1 h prior to evening milking twice a week for 4 mo before relocation and daily for 2 d before relocation. Samples were also taken immediately before the move, daily for 4 d post-relocation, and twice weekly for 1 mo after relocation. Milk yield, milk fat percentage, SCC, incidence of clinical mastitis, and heat detection were examined also. Corticosteroid concentrations in the moved group increased from 9.4 ± 1.0 ng/mL to 22 ± 2.1 ng/mL before and after relocation; merged cow corticosteroid concentrations went from 8.8 ± 1.6 ng/mL to 21.3 ± 2.0 ng/mL. Production traits did not vary significantly upon relocation; however, p.m. milk yield was reduced initially post-relocation (8.2 ± 0.4 kg) but returned to baseline by the third day (10.8 ± 0.3 kg). It was concluded that relocation had no lasting effects on cows, but factors such as isolation could extend the short-term effects such as increased corticosteroid levels and decreased milk yields.

Transport stress was studied to determine its effect on ACTH secretion from lymphocytes (Dixit et al., 2001). Twelve Holstein-Friesian heifers were implanted with jugular catheters 1 d before transport. Cows were placed in groups of seven and five and transported in a commercial truck for 14 h. Blood samples were collected from each animal before transport, at the end of transport, and 24 h after transport. For short-term transport, seven Holstein-Friesian heifers were transported for 30 min. Blood was collected before and immediately after transport. Heart rate and body temperature were monitored in all of the cows. During long-term transport, ACTH release increased significantly from a mean of 4.72 ± 0.48 pg/mL in lymphocytes before travel to 8.24 ± 1.4 pg/mL immediately after travel. Cows that were rested on the truck for 24 h after travel maintained a high ACTH level, while those loaded off the truck returned to baseline levels. Mean heart rate and body temperature also increased during travel, but returned to basal levels after 24 h rest. Short-term travel did not have any influence on lymphocytic ACTH levels.

Cows exposed to transport may have a suppression of an LH surge (Nanda et al., 1990). Ten early postpartum (between 1 and 2 wk) cows were used twice in groups of 4 and 6. During the experiment, each cow was administered 1 mg estradiol benzoate and transported for 30 min 16 to 18 h later. Jugular catheters were used to collect blood for analysis of plasma cortisol, progesterone, and LH. The estradiol benzoate-induced LH surge was suppressed by the increased levels of cortisol because the surge did not occur in the cows until basal cortisol levels returned. Transport also affected the LH surge more in early postpartum cows.

Cows with a preferred side in the milking parlor had higher heart rates than cows with no side preference or cows (Hopster et al., 1998). Differences in latency to enter the parlor and tachycardiac response were found with respect to preferred side. Two groups of 8 Holstein-Friesian cows were selected based on side preference data recorded by a computer identification system. Cows were milked on both sides of the milking parlor for 18 milking sessions; cows showing no side preference were used as controls. Heart rates were recorded at 5 s intervals using non-invasive telemetric heart rate monitors beginning 5 min before milking and ending when the cow returned to its cubicle housing. Cows milked on their preferred side had a heart rate of 86 ± 2.2 bpm during the last minute of milking; when milked on their non-preferred side they had a heart rate of 87 ± 2.2 bpm. Cows without a side preference averaged 80 ± 2.1 bpm during the same time. However, there was a difference in latency to enter the milking parlor and tachycardiac response. Cows milked on the left side of the parlor took longer to enter the parlor by 11 ± 4.2 s compared to cows milked on the right side. Heart rates also increased by 10.1 ± 1.2% when entering the parlor on the left as opposed to 6.2 ± 1.2% when entering on the right side. It was concluded that milking cows on their non-preferred side in the milking parlor does not increase stress levels enough to pose a threat to animal welfare.

Isolation from the herd during milking results in increased heart rates and plasma cortisol, as well as a depression in oxytocin secretion in cows (Rushen et al., 2001). Cows that were milked alone in unfamiliar surroundings had significantly higher heart rates than those milked in a familiar room. Plasma cortisol area under the curve was also higher in isolation groups, with the control group having 6.4 ± 2.4 ng/mL/min, the isolated group having 15.2 ± 2.4 ng/mL/min, and the human contact group having 13.2 ± 2.5 ng/mL/min. Normal milking also resulted in higher oxytocin levels, increasing by as much as 16.9 ng/mL/min from the other milking groups. It was noted that previous studies had correlated increased heart rate with a higher incidence of behavioral responses such as kicking or stepping (van Reenen et al., 1996), but the increased heart rate could simply be a result of increased movement (de Pasillé et al., 1995).

Heifers denied social interaction and forced into isolation had increased heart rates and plasma cortisol concentrations (Boissy and le Neindre, 1997). Twenty-four heifers, 12 Friesians and 12 Aubrac, were divided into two groups per breed and housed in nonadjacent pens to avoid contact. During trials, each group of heifers was moved from their home pen to a test area. Each heifer had to pass through a bail crush in order to reach the area, and the last heifer through was forced to remain in the bail crush while maintaining visual and limited physical contact. After 16 min, the other heifers were led out of the test area by an experimenter and returned to the test area, or returned to their home pen and replaced with a group of heifers unfamiliar to the subject. All heifers struggled more and had higher overall activity when exposed to unfamiliar animals; however, heart rates were not significantly different compared to exposure to familiar animals. Heart rates took longer to decrease to basal levels with contact with the unfamiliar animals. Aubrac heifers experienced higher heart rates than Friesians, with an initial mean of 102 ± 18 compared to 83 ± 11 beats/min when exposed to familiar heifers. Plasma cortisol levels were measured at the beginning and end of each session. The difference between initial levels and end levels decreased with after each session and Friesian heifers had significantly lower levels than Aubrac. Although the results show that Aubrac are more socially dependent than Friesians, both experienced an increase in heart rate and plasma cortisol, indicating that there is physiological stress associated with social deprivation.

Calves were exposed to road transport for periods of 6 h or 18 h and compared to calves that were not transported (Kent and Ewbank, 1986). Because calves less than 2 wk old were more susceptible to illness and death when transported than older calves (Staples and Haugse, 1974), blood samples were collected before, during, and after transport to determine the effects of transport on blood constituents. Calves exposed to transport for 6 h or 18 h experienced higher plasma cortisol levels than those exposed to no transport, with levels peaking 36 ± 10 nmol/L and 29 ± 8 nmol/L for 6 h and 18 h, respectively, compared to 8 ± 3 nmol/L for samples collected from non-transported calves during the same time. These peaks occurred soon after loading. Neutrophil to lymphocyte (**N:L**) ratio also increased significantly in calves transported for 18 h compared to non-transported calves, however, some non-transported calves did show an increase in N:L ratio, so it could not be used as an indicator of transport stress in calves. It was concluded that loading and unloading calves was a greater stressor than transport.

Chapter 2: Effect of Relocation on Locomotion and Cleanliness on Dairy Cows

ABSTRACT

The objective of this study was to determine the effect of relocation to a new facility on locomotion and cleanliness in dairy cows. Cows at the Virginia Tech Dairy Center were allocated into four pens: 1) access to a rubber mat in the feed bunk area (MAT; n = 17); 2) cows in the northeast section of the new facility (NE; n = 23); 3) two breeds exposed in the northwest section (NW; n = 22 Holsteins and 22 Jerseys); and 4) a group of cows in the southwest section (SW; n = 21). Parity was balanced across all groups. Trial 1 compared all Holsteins in the herd and Trial 2 compared the breed differences in the NW section. Locomotion was scored on a 4-point scale where 1 = normal and 4 = lame. Cleanliness was scored on a 4-point scale where 1 = little or no manure visible and 4 = heavy plaques of manure on three body regions: udder, lower leg, and flank/upper leg. Data were analyzed using the MIXED procedure of SAS. In Trial 1, MAT cows had lower locomotion scores than other groups with a mean score of 1.6 ± 0.1, compared to 1.8 ± 0.1, 1.9 ± 0.1, and 2.0 ± 0.1 for NE, NW, and SW, respectively. MAT cows also had slightly cleaner lower legs, although this may be a result of having a lower stocking density than the other groups. In Trial 2, Holsteins and Jerseys had no difference in locomotion score, but Jerseys did have cleaner lower legs than Holsteins (2.9 ± 0.1 vs. 3.5 ± 0.1, respectively). In both trials, all cows were cleaner (lower scores) on all body regions after relocation, suggesting that the new facility improved hygiene. Locomotion scores also increased across all groups as time passed, which indicates that the cause of lameness in the new facility was gradual; the rubber mat in front of the feed bunk alleviated the effects of the new facility on locomotion.

Keywords: hygiene, locomotion, relocation

2.1 Introduction

Relocation to a new facility offers many benefits. However, there is the potential for adverse effects initially. New flooring surfaces may be much more textured than older flooring that is worn from years of scraping and cow traffic. This may lead to increased lameness due to irritation (Phillips and Morris, 2001). It has also been shown that new concrete is more alkaline, which may lead to increased lameness compared to more neutral, older concrete previously exposed to slightly acidic urine (Bray et al., 1992).

Considerations should be made for the benefits of the new facility. A milking parlor with increased capacity leads to a larger throughput, and in turn, less time standing in the holding area. While increased texture can be an irritant, worn alleys can also cause increased slippage and lameness (van der Tol et al., 2005). An improved and more frequent method of cleaning alleys results in walkways free of excreta that would also decrease slippage. Alternative flooring surfaces such as rubber mats have been proven to improve lameness (Vokey et al., 2001). In addition, a rubber mat placed in front of the feed bunk may increase cow comfort; cows spent more time standing in front of the feed bunk, but showed no change in eating time (Fregenosi et al., 2004).

Cleanliness is not often considered in the welfare of cattle in the UK (Department of Environment, Food, and Rural Affairs, 2003). However, it can be used as an indicator of time allocation, social hierarchy, and farm management. Additionally, cleaner cows are not exposed to as many environmental pathogens, which can lead to improved health. The objective of this study was to determine the effect a rubber mat placed in front of the feed bunk has on the locomotion of dairy cows after relocation to a new freestall barn and to determine breed effects on relocation responses.

2.2 Materials and Methods

Animals and Treatments

This study took place at the Virginia Tech Dairy Center from March 2004 to October 2004. Before relocation, cows were divided into treatment groups to acclimate them to a new social structure. Treatments were not implemented until after relocation. Groups were balanced for stage of lactation and lactation number. The treatment groups were: 1) Holstein cows (n = 17) exposed to a rubber mat placed in front of the feed bunk (**MAT**); 2) Holstein cows (n = 23) located in the northeast section of the new facility (**NE**); 3) Holstein (n = 22) and Jersey cows (n = 22) in the northwest section (**NW**); and 4) Holstein cows (n = 21) in the southwest section (**SW**) (Figure 2.1).

The rubber mat had dimensions of 22 m x 2 m x 1.5 cm and was a recycled conveyor belt from a mining operation (VA Carolina Belting, Inc; Salem, VA). It was placed in front of the feed bunk area and anchored to the alley with screws on the end of the mat closest to the alley flush system water release point to prevent movement from the feed bunk area. All cows in the NE group were fed through Calan doors, and were acclimated to the Calan door system before relocation (Pence, 2005). They were fed one of three diets for 3 wk before and 9 wk after relocation. The diets were 1) a TMR identical to that fed to the rest of the herd; 2) the same TMR top-dressed with alfalfa hay; and 3) the same TMR top-dressed with grass hay. Individual cow eating rates determined the amount of TMR fed; the hay top-dressing comprised 10% of the feed presented to the cows. The NW group was divided into stocking densities of 0.67, 0.83, 1.00, or 1.17 cows per stall. Each NW sub-group had equal numbers of Holsteins and Jerseys; the stocking density of 0.67 cows per stall utilized 8 cows and 12 stalls; 0.83 cows per stall utilized 10 cows and 12 stalls; 1.00 cow per stall utilized 12 cows; and 1.17 cows per stall utilized 14 cows and 12 stalls. The SW group was comprised of lactating Holsteins not assigned to another treatment. Those cows that were not in the NW section were housed in freestall pens with a stocking density of 1.00 cow per stall pre-relocation; after relocation, MAT cows were stocked at 0.50 cow per stall, NE cows at 1.00, and SW at 1.10 cows per stall.

Before relocation, cows at the old facility were housed in a freestall barn with grooved concrete alleys. Stalls (1.1 m x 2.1 m) were bedded with sawdust and partitioned by wooden dividers. Freestall pens had between 20 and 30 stalls. A farm employee scraped alleys daily at 0300 h. Feeding occurred at 0900 h; feed bunks were located centrally to the freestall pens, and raised 0.68 m from the ground. Automatic watering troughs were located next to feed bunks and at the end of freestall pens. In the AM of relocation, farm employees led cows according to treatment to the new facility, a distance of approximately 131 m. The new facility was freestall housing with grooved concrete alleys. Stalls (1.2 m x 2.3 m) were bedded with canvas mattresses covered lightly with sawdust and partitioned by pipe-loop dividers. Freestall pens had between 12 and 64 stalls. Each alley in the new facility was flushed twice daily by an automatic alley flush system. Feeding occurred at 1000 h and 1400 h. Feed bunks were located in the center of the freestall area and were directly on the ground. With the exception of the NE cows, which used Calan doors, cows were required to pass through head gates to gain access to feed.

The milking parlor at the old facility was a single side-opening parlor with six milking stalls. Milking began at 0100 h and 1300 h. Cows were milked according to treatment group and were required to walk uphill on a gravel walkway a distance of approximately 60 m to reach the holding area if not housed in freestall pens adjacent to the milking parlor. The holding area had dimensions of 20.6 m x 5.3 m when the chain crowd gate was extended to full capacity. Cows entered the milking parlor individually once a milking stall was available. Upon exit from the parlor, cows walked through an alley that was 7.2 m x 0.85 m. Cows remained in a separate holding pen until all other cows in the group were milked and then returned to their freestall pen. The milking parlor at the new facility was a double-eight herringbone design. Milking began at 0100 h and 1300 h. All freestall pens were adjacent to the parlor, so walking distance was minimal for each treatment group (5 to 30 m). The holding area measured 11.7 m x 7.6 m when the vertical lifting crowd gate was extended to full capacity. Cows entered the parlor eight at a time on each side of the parlor. Upon exit from the parlor, cows were required to walk down an alley 15.5 m x 1.1 m and pass over a set of scales, and had access to their freestall pens immediately.

Locomotion Scoring

Data collection began in March 2004 with hoof trimming for all cows in the herd. A veterinarian (O. Becvar, oral communication, March, 2004– November, 2004) completed hoof scoring (Greenough et al., 1996) at this time; claw disorders were recorded and treatment applied if necessary. Hoof scores were divided into two categories: diseases of laminitis and diseases with infection (Table 2.1). The diseases of laminitis category had two subcategories of laminitis and sub-clinical laminitis. The diseases of each category and subcategory were given a numerical value of 1 and were added together to create a covariate (laminitis score or disease score) used to determine the effect of pre-relocation hoof scores and locomotion score on milk yields. Locomotion scoring occurred monthly for 2 mo post-trimming to allow time for the observer to become familiar with locomotion scoring (Appendix A1, Table 2.2). Scores were recorded weekly for 12 wk beginning June 19, 2004 and twice monthly for 6 wk, with data collection ending in October 13, 2004. Locomotion scores were on a 1 to 5 scale (Table 2.2) with a lower score denoting greater hoof health (Sprecher et al., 1997; Berry, 2001). Observations were made as each cow exited the milking parlor in the afternoon. The observer stood outside of the parlor exit alley to allow for a less hesitant walking motion. Cows were observed walking a straight line for a distance of 10 m on concrete and while stationary on concrete. Attention was paid to the posture of the back, feet, and head movement in determining locomotion. A level back while walking and standing indicated a normal cow with a locomotion score of 1; a score of 2 indicated mildly lame and is distinguished from a score of 1 only by a slightly arched back when walking. A score of 3 is recognizable by an arched back when walking and standing and noticeably shorter strides with one or more feet; it indicated a moderately lame cow. A score of 4 indicated a lame cow and identified by an arched back when walking and standing and one or more limbs favored but bearing weight. A score of 5 indicated a severely lame cow and was distinguished by an arched back and a refusal to bear weight on one limb.

Cleanliness Scoring

Data collection began in July 5, 2004 and ended October 13, 2004. Scoring was twice monthly. Cleanliness scores were based on a system devised by Cook (2002). Cows were scored on a 1 to 4 scale (Table 2.3) on three regions of their bodies: lower leg; udder; and flank and upper leg. Lower scores were indicative of cleaner body regions. A score of 1 signified little or no manure in the region. A score of 2 meant there was minor splashing of manure in the region. A score of 3 indicated distinct plaques of manure with some hair visible, and a score of 4 denoted confluent plaques of manure covering the area. When making cleanliness scores, the observer examined the left side of the animal because it is the preferred side for resting on a level surface (Albright, 1975). The exception to this was udder scoring. The udder was observed from behind, in addition to the left side, to account for manure splashing caused by the tail.

Statistical Analysis

Main effects of treatment (Trial 1) or breed (Trial 2), lactation number, date, and their interactions were tested with the MIXED procedure of SAS (SAS Institute, Inc., Version 8.2, 1999, Cary, NC). Dependent variables were locomotion score, udder cleanliness score, lower leg cleanliness score, and flank and upper leg cleanliness score. Trial 1 examined the locomotion and cleanliness of Holsteins. Treatments were MAT, NE, NW, or SW, and lactation number was 1, 2, or greater than or equal to 3. Trial 2 examined breed differences between Holsteins and Jerseys in the NW group. A covariate was created by averaging all scores collected 1 mo before relocation to account for discrepancies in scores pre-treatment; it adjusted scores by allowing for a 0.59 point increase in locomotion and cleanliness scores for every 1.0 point increase in the covariate. The following model statement was used for Trial 1: $Y_{ijkl} = \mu + T_i + L_j + TL_{ij} + C_{(ij)k} + D_l + TD_{il} + LD_{jl} + TLD_{ijl} + \beta(S_{(ij)k} - \overline{S}) + E_{ijkl}$

where:

Y_{ijkl} = dependent variable(s)
μ = mean
T_i = fixed effect of treatment (i = MAT, NE, NW, or SW)
L_j = fixed effect of lactation number (j = 1, 2, or $\geq$3)
TL_{ij} = effect of the interaction between treatment and lactation number
C_{ijk} = random effect of cow within treatment and lactation number
D_l = days post-relocation (l = 7-86)
TD_{il} = effect of the interaction between treatment and days post-relocation
LD_{jl} = effect of the interaction between lactation number and days post-relocation
TLD_{ijl} = effect of interaction between treatment, days post-relocation, and lactation number
$\beta(S_{(ij)k} - \overline{S})$ = covariate adjustment for pre-relocation score
E_{ijkl} = residual error

The following model was used for Trial 2: $Y_{ijkl} = \mu + B_i + L_j + BL_{ij} + C_{(ij)k} + D_l + BD_{il} + LD_{jl} + BLD_{ijl} + \beta(S_{(ij)k} - \overline{S}) + E_{ijkl}$

where:

Y_{ijkl} = dependent variable(s)
μ = mean
B_i = fixed effect of breed (i = Holstein or Jersey)
L_j = fixed effect of lactation number (j = 1, 2, or $\geq$3)
BL_{ij} = effect of the interaction between breed and lactation number
C_{ijk} = random effect of cow within treatment and lactation number
D_l = days post-relocation (l = 7-86)
BD_{il} = effect of the interaction between breed and days post-relocation
LD_{jl} = effect of the interaction between lactation number and days post-relocation
BLD_{ijl} = effect of interaction between breed, days post-relocation, and lactation number
$\beta(S_{(ij)k} - \overline{S})$ = covariate adjustment for pre-relocation score
E_{ijkl} = residual error

The following model was used to determine milk yield differences between locomotion scores for Trial 1: $MY_{ijklmno} = \mu + T_i + L_j + TL_{ij} + C_{(ij)k} + D_l + TD_{il} + LD_{jl} + TLD_{ijl} + \beta(S_{(ij)k} - \overline{S}) + M_m + A_n + O_o + E_{ijklmno}$

where:

$MY_{ijklmno}$ = milk yield (kg/d)
μ = mean
T_i = fixed effect of treatment (i = MAT, NE, NW, or SW)
L_j = fixed effect of lactation number (j = 1, 2, or $\geq$3)
TL_{ij} = effect of the interaction between treatment and lactation number
C_{ijk} = random effect of cow within treatment and lactation number

D_l = days post-relocation (l = 7-86)
TD_{il} = effect of the interaction between treatment and day post-relocation
LD_{jl} = effect of the interaction between lactation number and day post-relocation
TLD_{ijl} = effect of interaction between treatment, day post-relocation, and lactation number
$\beta(S_{(ij)k} - \overline{S})$ = covariate adjustment for pre-relocation score
M_m = effect of pre-relocation laminitis score
A_n = effect of pre-relocation disease score
O_o = effect of post-relocation locomotion score
$E_{ijklmno}$ = residual error

The following model was used to determine milk yield differences between locomotion scores for Trial 2: $MY_{ijklmno} = \mu + B_i + L_j + BL_{ij} + C_{(ij)k} + D_l + BD_{il} + LD_{jl} + BLD_{ijl} + \beta(S_{(ij)k} - \overline{S}) + M_m + A_n + O_o + E_{ijklmno}$

where:

$MY_{ijklmno}$ = milk yield (kg/d)
μ = mean
B_i = fixed effect of breed (i = Holstein or Jersey)
L_j = fixed effect of lactation number (j = 1, 2, or $\geq$3)
BL_{ij} = effect of the interaction between breed and lactation number
C_{ijk} = random effect of cow within treatment and lactation number
D_l = days post-relocation (l = 7-86)
BD_{il} = effect of the interaction between breed and days post-relocation
LD_{jl} = effect of the interaction between lactation number and days post-relocation
BLD_{ijl} = effect of interaction between breed, days post-relocation, and lactation number
$\beta(S_{(ij)k} - \overline{S})$ = covariate adjustment for pre-relocation score
M_m = effect of pre-relocation laminitis score
A_n = effect of pre-relocation disease score
O_o = effect of post-relocation locomotion score
$E_{ijklmno}$ = residual error

Results are presented as LS means ± SEM. Differences were considered significant at $P < 0.05$ and trends at $P < 0.10$. Differences were determined with contrasts.

2.3 Results

In Trial 1, treatment had an effect ($P < 0.05$) on locomotion and lower leg cleanliness (Table 2.4); lactation number ($P < 0.01$; Table 2.5), date ($P < 0.01$;

Figure 2.2), and their interaction ($P < 0.05$; Figure 2.3) had an effect on locomotion. There was also a trend for the interaction between treatment and date ($P < 0.10$; Figure 2.4).

The MAT group had significantly lower locomotion scores (less lame) than all other treatments (mean = 1.6; Table 2.4). Additionally, the MAT group had a lower cleanliness score (cleaner) for the lower leg region compared to other groups (mean = 2.7; Figure 2.5C). The treatment by date interaction shows that cows across treatments had similar initial locomotion scores that increased as time progressed (Figure 2.4), but the MAT group did not increase above a score of 2. The SW group had the highest locomotion score (more lame) of any treatment group (mean = 2.0), and increased above 2.5 by d 74.

Locomotion score was higher (more lame) in cows in their third or greater lactation (mean = 2.1; Table 2.5). While cows in all lactation groups showed an increase in locomotion score over time, third or greater lactation cows had the most severe increase (Figure 2.3). Significant differences were not evident until after d 45 post-relocation.

As time progressed, locomotion scores of cows across all treatments increased significantly (Figure 2.2). Locomotion scores increased from a mean of 1.4 on d 14 to 2.3 on d 86. Daily milk yields (kg) decreased in Trial 1 with increased locomotion scores, but the differences were not significant ($P = 0.28$; Figure 2.6). Cleanliness scores across all body regions improved in the new facility ($P < 0.01$; Figure 2.7)

In Trial 2, breed ($P < 0.74$; Table 2.6) and lactation number ($P = 0.14$; Table 2.7) did not affect locomotion score (Figure 2.8). However, date ($P < 0.01$; Figure 2.9) and the interaction between date and lactation number ($P < 0.05$; Figure 2.10) had an impact on locomotion score. Locomotion scores increased from 1.2 on d 14 to 2.4 on d 86 (Figure 2.9). The lactation by date interaction showed cows in third or greater lactations had significantly higher locomotion scores (more lame) by d 86 (Figure 2.10).

Generally, breed (Table 2.6) and lactation number (Table 2.7) did not affect cleanliness. Date and its interactions with breed and lactation number had an effect on cleanliness scores. Cleanliness scores fluctuated from pre-relocation scores and decreased by the end of observation (d 86; Figure 2.11). Breed by date interaction had an effect on lower leg (Figure 2.12) and flank/upper leg cleanliness scores (Figure 2.13). There were breed differences with respect to lower leg cleanliness, with Jerseys (mean = 2.9) having significantly lower scores (cleaner) than Holsteins (mean = 3.5; Figure 2.14A). Lactation number also affected lower leg cleanliness, with scores increasing as lactation number decreased (Figure 2.14B). As in Trial 1, daily milk yields across all cows decreased with increased locomotion scores, but the effect was not significant ($P = 0.27$; Figure 2.15).

2.4 Discussion

In Trial 1, cows in the MAT group had lower locomotion scores (mean = 1.6) than the SW, NE, and NW groups (means = 2.0, 1.8, and 1.9, respectively). Vokey et al. (2001) found that rubber flooring improved hoof health and locomotion, but only in conjugation with an abrasive bedding material such as sand. Additionally, first and second lactation cows experienced lower locomotion scores (means = 1.7) than third or greater lactation cows (mean = 2.1). This agrees with previous findings with respect to multiparous cows and lameness due to metabolic stress from high milk yield (Barkema et al., 1994) and increased age (Eddy and Scott, 1980).

Locomotion score increased in all treatment groups as time in the new facility increased, with locomotion scores increasing from 1.4 on d 7 to 2.3 on d 86 (Figure 2.2). A lactation number by date interaction revealed that third or greater lactation cows developed more severe locomotion scores further into the study. Milk production decreased as locomotion score increased, however, the differences were not significant ($P = 0.28$). These findings are consistent with the results of Juarez et al. (2003), who examined the impact locomotion and walking distance to the milking parlor had on milk production and composition in multiparous Holstein cows. Cows with increased locomotion scores (more lame) experienced decreased milk, fat, and protein production, but there were no significant differences.

There was a significant difference in lower leg cleanliness among treatment groups. The MAT group had a lower cleanliness score (mean = 2.7) than the other groups, which is contradictory to what was expected because cows could potentially use the mat as an alternative to lying in freestalls. However, there was no decline in cleanliness in this group, suggesting that cows did not prefer lying on the mat to lying in the freestalls. It should be noted that there was a decrease across all cleanliness scores over time; the increase in cleanliness score at d 35 can be attributed to a defective water recycling pump that did not allow for normal alley flushing the previous week (Figure 2.7).

In Trial 2, there was no effect of breed on locomotion. Peterse (1985) suggested that Jerseys are less susceptible to lameness problems than Holsteins; however, relocation did not cause significant changes in locomotion score between the two breeds. A lactation number by date interaction revealed that third or greater lactation cows had more severe and sporadic locomotion scores (Figure 2.10). First lactation cows had relatively stable scores until the increase at d 86. Date had the same effect across both breeds as it did in Trial 1, with locomotion scores increasing as time progressed.

Jerseys (mean = 2.9) had lower cleanliness scores for the lower leg region than Holsteins (mean = 3.5), which suggests that the larger freestalls in the new facility allowed them to fully remove themselves from the alley while lying. Older cows were also cleaner in the lower leg region (Figure 2.14B), possibly due to more time spent lying down. The effect of date on cleanliness showed that cows across breeds benefited from relocation; scores from the three body regions improved from pre-relocation scores. Cows in Trial 2 experienced similar results as cows in Trial 1 with respect to the impact of locomotion score on daily milk yield.

2.5 Conclusions

Placing a rubber mat in front of the feed bunk helped protect cows from the effects of new flooring in the new facility. The reason for the worsened locomotion scores as a herd is multi-faceted: cow hooves could be irritated due to the increased aggregate of the new flooring (Phillips and Morris, 2001; Vokey et al., 2001); the increased alkalinity of new concrete may slowly erode hoof health (Bray et al., 1992); or cows could spend more time exploring their surroundings, degrading the hoof over time. Increased locomotion scores were not evident until d 45, so the cause is a gradual aggravation. Others have found that adjustments in walking behavior due to flooring can have detrimental effects on the welfare of cattle (Metz and Bracke, 2003; van der Tol et al., 2005) Holstein and Jersey locomotion scores were affected similarly, suggesting that neither breed is more or less susceptible to lameness issues as a result of relocation to a new facility. Although increased locomotion scores did not significantly affect daily milk yield, there was a decline in production (Warnick et al., 2001), suggesting that more severe conditions in the new facility could have a greater impact on milk yields.

Overall, cow cleanliness benefited from relocation. All regions of the body that were examined had improved cleanliness scores at the end of observation. The most likely cause of this is the improved and more frequent cleaning of alleys and the use of larger freestalls (Nordlund and Cook, 2003). Jerseys benefited more from relocation, showing greater improvement in flank/upper leg and lower leg cleanliness than Holsteins. Another explanation for improved lower leg cleanliness may be that cows stood in the alleys during flushing. While the MAT group had cleaner lower legs (lower scores), it should be noted that the stocking density was much less than the other groups in Trial 1.

2.6 Implications

Alternative flooring in front of the feedbunk can reduce the prevalence of lameness experienced after relocation to a new facility. Furthermore, the new facility caused an increase in locomotion scores, although the effects were not evident until 45 d post-relocation. It may be that larger, more comfortable freestalls and a better method for cleaning alleys led to improved cow cleanliness, which in turn may decrease the risk of environmental mastitis infections. Future studies should examine how long the effects of the new facility last and if locomotion scores and lameness return to basal levels. The impact of improved cow hygiene on mastitis and SCC should be observed. Also, the effect lameness has on reproductive health should be examined.

Table 2.1. Herd hoof score categories and subcategories.[1] (Number of incidences observed in parentheses)

Diseases of laminitis		Diseases with infection
Laminitis	Sub-clinical Laminitis	
Sole ulcer (2)	Double sole (10)	Foot rot (1)
Heel erosion (20)	Corkscrew claw (13)	Digital dermatitis (8)
White line hemorrhage (85)		Interdigital dermatitis (0)
White line separation (40)		Interdigital corn (0)
White line ulcer (2)		
Anterior wall crack (2)		
Sole ulcer hemorrhage (79)		

[1]Adapted from Greenough et al. (1996); Becvar (personal communication)

Table 2.2. Locomotion Scoring Scale[1]

Lameness score	Clinical description	Assessment criteria
1	Normal	The cow stands and walks with a level-back posture. Her gait is normal.
2	Mildly lame	The cow stands with a level-back posture but develops an arched-back posture while walking. Her gait remains normal.
3	Moderately lame	An arched-back posture is evident both while standing and walking. Her gait is affected and is best described as short striding with one or more limbs.
4	Lame	An arched-back posture is always evident and gait is best described as one deliberate step at a time. The cow favors one or more limbs/feet.
5	Severely lame	The cow additionally demonstrates an inability or extreme reluctance to bear weight on one or more of her limbs/feet.

[1]Adapted from Sprecher et al. (1997); Berry (2001)

Table 2.3. Cow Cleanliness Scale[1]

Score	Body Region		
	Lower leg	Udder	Flank/Upper leg
1	Little or no manure above the coronary band.	No manure present.	No manure present.
2	Minor splashing above the coronary band.	Minor splashing of manure near the teats.	Minor splashing of manure.
3	Distinct plaques of manure above the coronary band, but hair is visible.	Distinct Plaques of manure on the lower half of the udder.	Distinct plaques of manure with hair showing through.
4	Solid plaque of manure extending high up the leg.	Confluent plaques of manure encrusted on and around the teats.	Confluent plaques of manure.

[1]Adapted from Cook (2002)

Table 2.4. Locomotion (1-5) and cleanliness (1-4) scores for Holsteins by treatment in Trial 1.

Variable	Treatment				SE	*P* <
	SW	NE	MAT	NW		
Locomotion	2.0[a]	1.8[b]	1.6[c]	1.9[ab]	0.08	0.05
Udder Cleanliness	2.3	2.1	1.9	2.6	0.19	0.09
Lower Leg Cleanliness	3.5[a]	3.1[b]	2.7[c]	3.4[a]	0.12	0.01
Flank/upper leg Cleanliness	2.9	2.5	2.7	2.7	0.15	0.25

[abc] Means within rows with different superscripts differ based on contrasts of each location.

Table 2.5. Locomotion (1-5) and cleanliness (1-4) scores for Holsteins by lactation number in Trial 1. Scores in the same row with different letters are significantly different.

Variable	Lactation Number 1	2	≥3	SE	*P* <
Locomotion	1.7^{b}	1.7^{b}	2.1^{a}	0.08	0.01
Udder Cleanliness	2.4	2.2	2.0	0.11	0.41
Lower Leg Cleanliness	3.4	3.2	3.0	0.10	0.10
Flank/upper leg Cleanliness	2.6	2.7	2.7	0.13	0.83

^{ab} Means within rows with different superscripts differ based on contrasts of each lactation.

Table 2.6. Locomotion (1-5) and cleanliness (1-4) scores for Holsteins and Jerseys in Trial 2.

Variable	Breed Holstein	Jersey	SE	*P* <
Locomotion	1.6	1.7	0.12	0.75
Udder Cleanliness	2.6	2.3	0.13	0.08
Lower Leg Cleanliness	3.5^{a}	2.9^{b}	0.10	0.01
Flank/upper leg Cleanliness	2.7	2.4	0.11	0.05

^{ab} Means within rows with different superscripts differ based on contrasts of each breed.

Table 2.7. Locomotion (1-5) and cleanliness (1-4) scores by lactation number for Holsteins and Jerseys in Trial 2.

Variable	Lactation Number 1	2	≥3	SE	*P* <
Locomotion	1.5	1.6	2.0	0.13	0.74
Udder Cleanliness	2.5	2.4	2.2	0.17	0.53
Lower Leg Cleanliness	3.3	3.1	2.9	0.12	0.08
Flank/upper leg Cleanliness	2.7	2.5	2.5	0.15	0.08

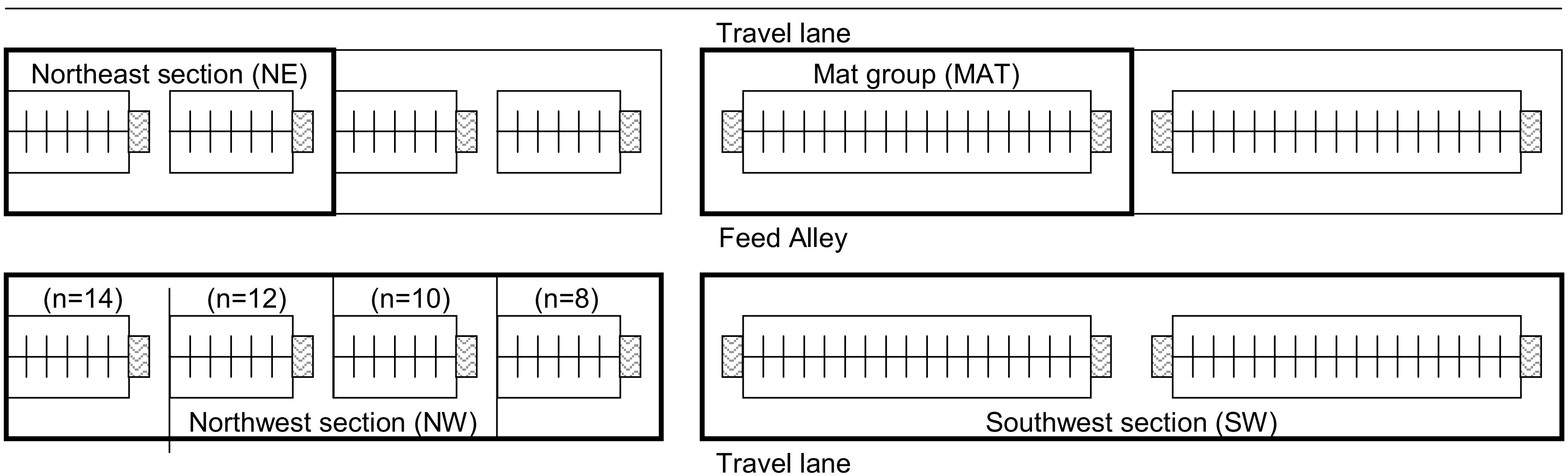


Figure 2.1. Freestall housing assignments in the new facility.

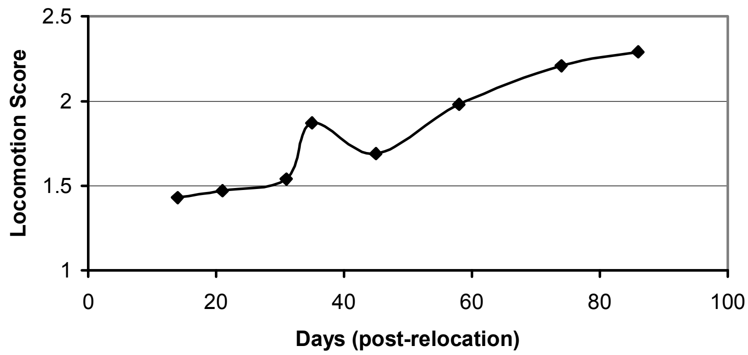


Figure 2.2. Effect of date on locomotion score (1-5) for all treatment groups in Trial 1 (*P* < 0.01).

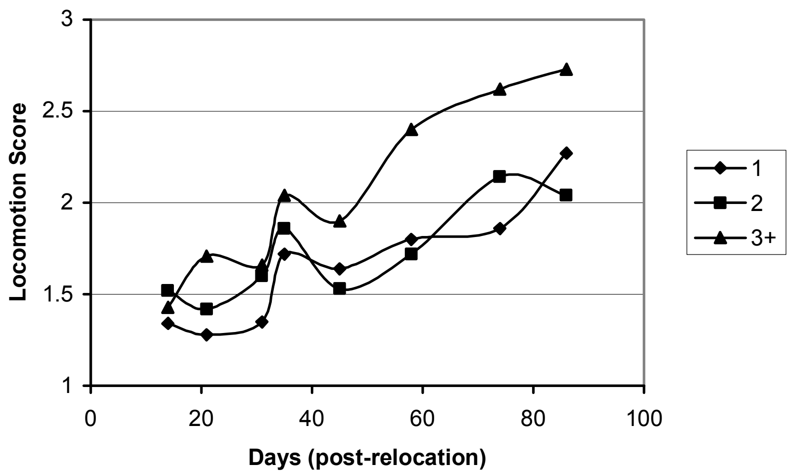


Figure 2.3. Effect of lactation number by date interaction on locomotion score (1-5) for all treatment groups in Trial 1 (*P* < 0.05). Higher scores indicate a greater degree of lameness.

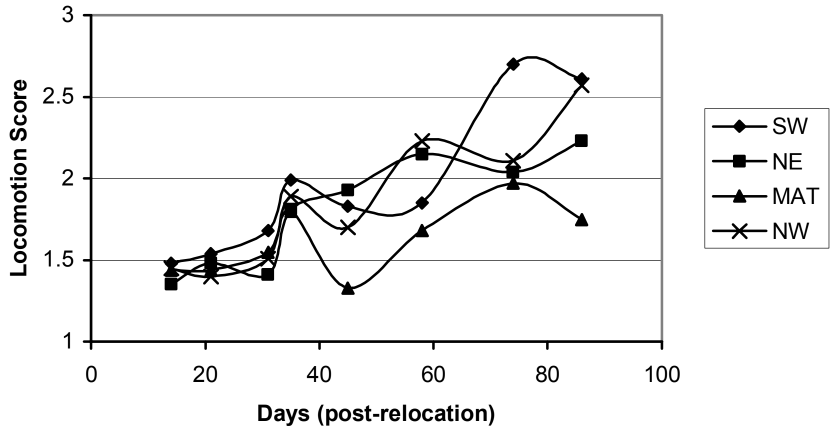
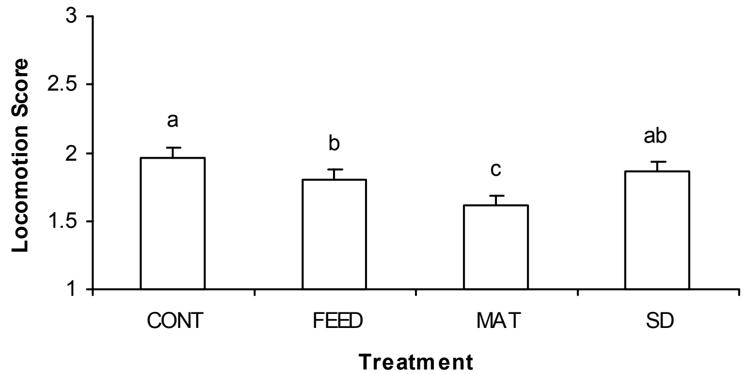
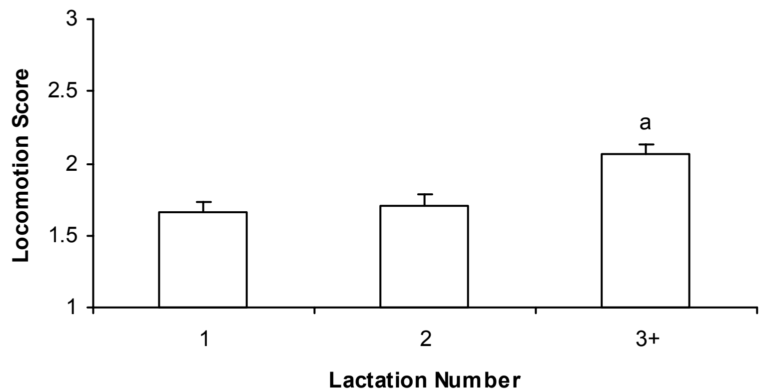


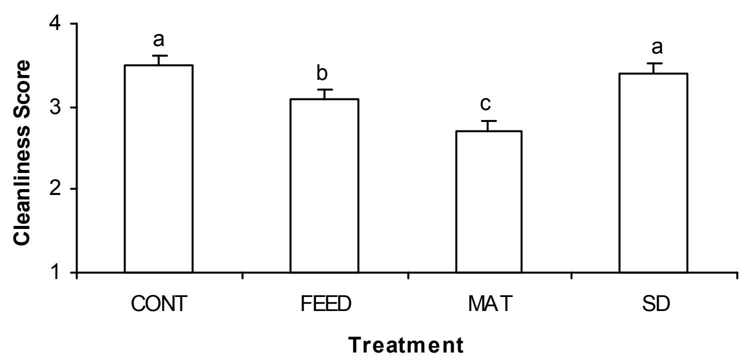
Figure 2.4. Trend of increased locomotion scores (1-5) as time progressed in each treatment group in Trial 1 ($P < 0.10$).



(A)



(B)



(C)

Figure 2.5. Locomotion scores (1-5) by treatment **(A)** and lactation number **(B)**. Lower leg cleanliness scores (1-4) by treatment **(C)**. Values are LS means ± SEM for Holstein cows in Trial 1. Bars with different letters are significantly different ($P < 0.05$).

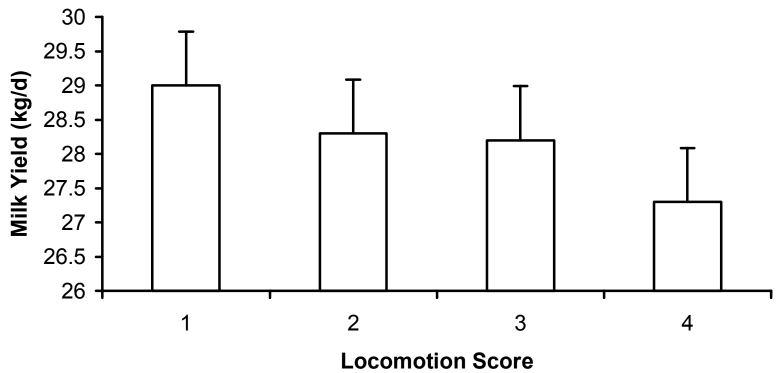


Figure 2.6. Effect of locomotion score on milk yield (kg) for all treatment groups in Trial 1. There were no significant differences (*P* = 0.28).

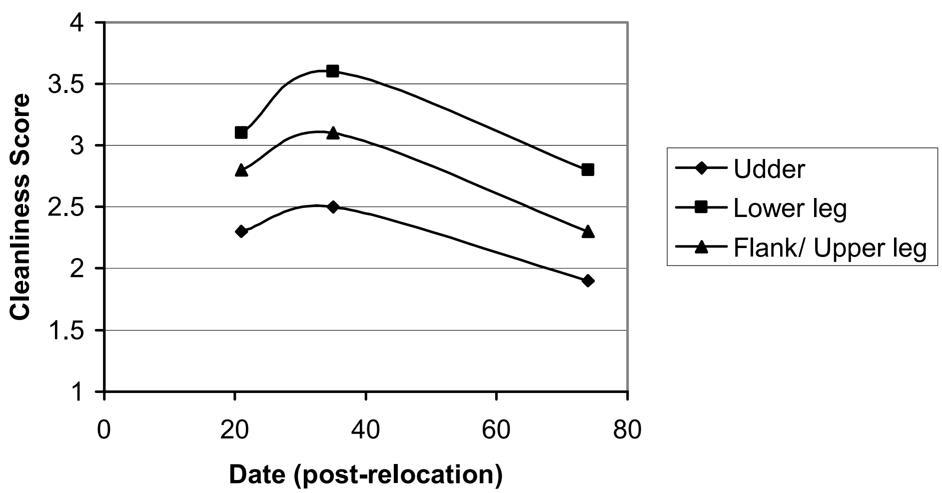
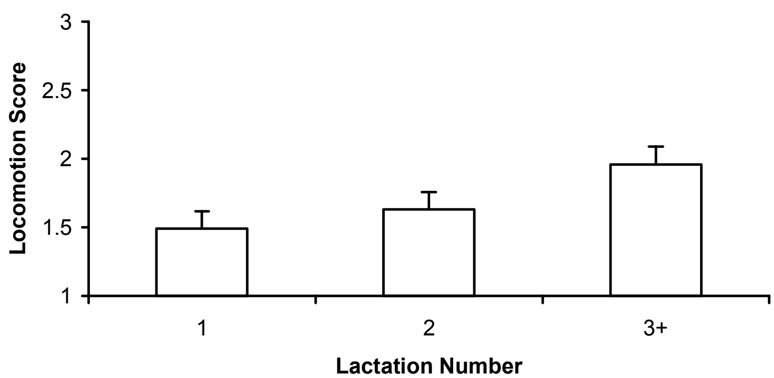


Figure 2.7. Effect of date on cleanliness score (1-4) for all treatment groups in Trial 1 ($P < 0.01$).

(A)



(B)

Figure 2.8. Locomotion scores (1-5) by breed **(A)** and lactation number **(B)** in Trial 2. Values are LS means ± SEM for Holsteins and Jerseys. There were no significant differences (*P* = 0.75).

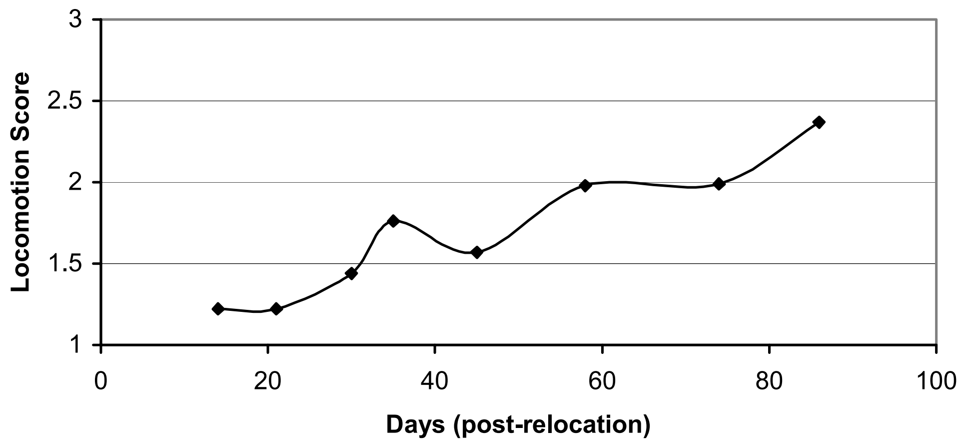


Figure 2.9. Effect of date on locomotion score for all cows in Trial 2 ($P < 0.01$).

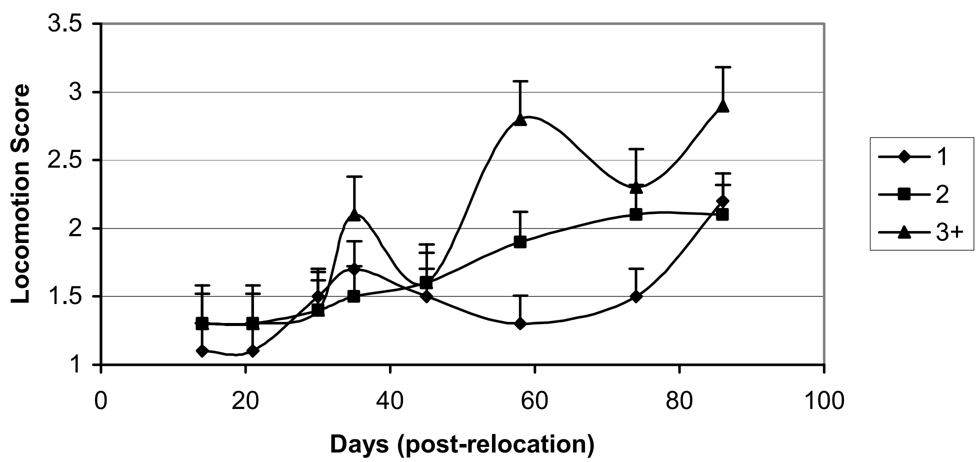


Figure 2.10. Effect of lactation number by date interaction on locomotion score for all breeds in Trial 2 ($P < 0.05$).

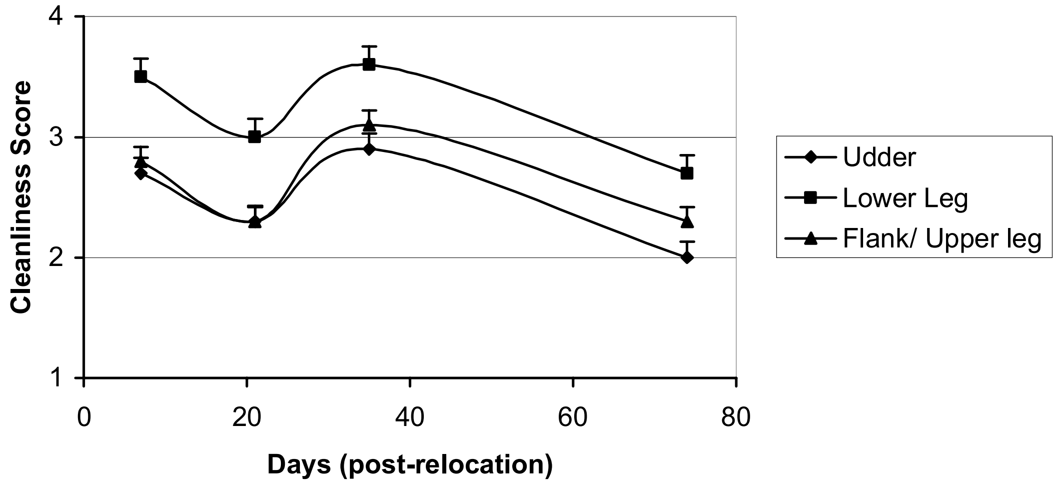


Figure 2.11. Effect of date on cleanliness score for cows in Trial 2 ($P < 0.01$).

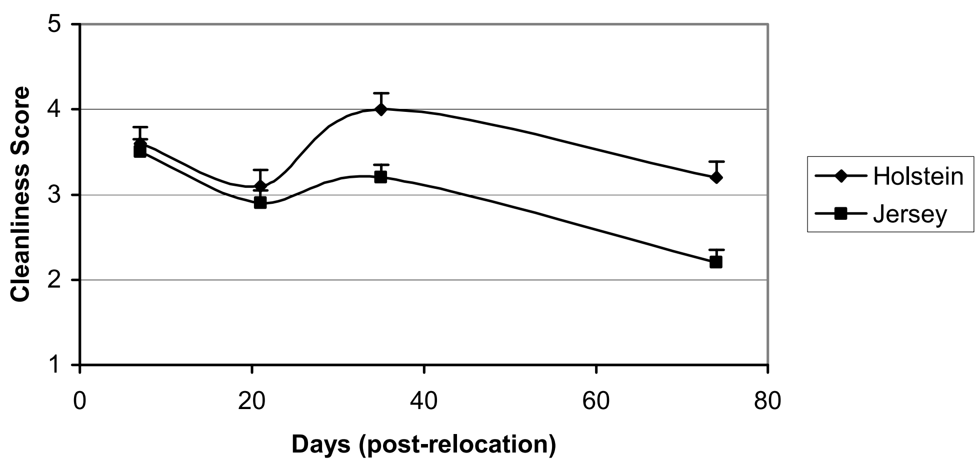


Figure 2.12. Effect of breed by date interaction on lower leg cleanliness in Trial 2 ($P < 0.05$).

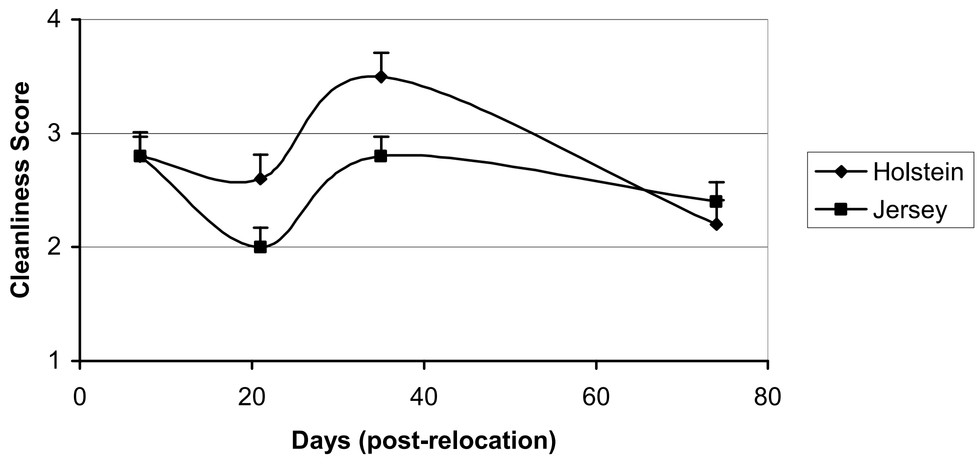
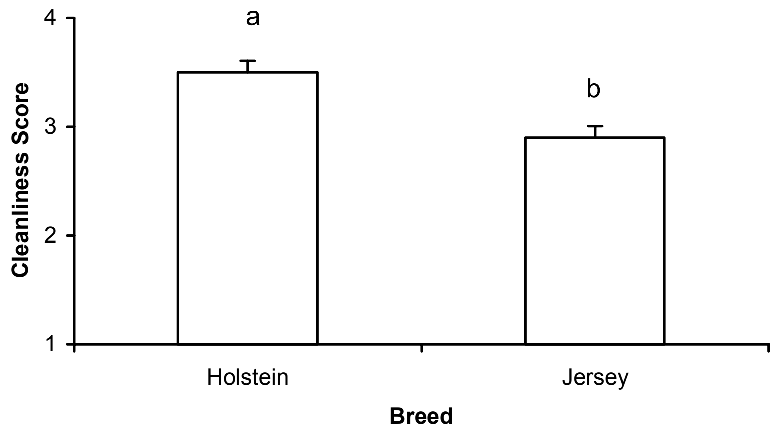
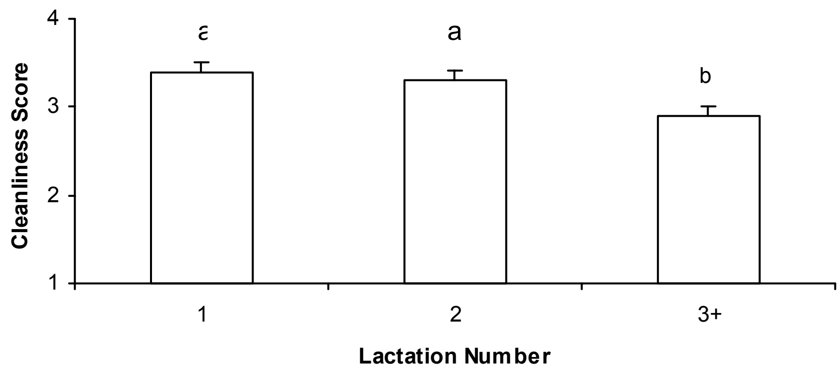


Figure 2.13. Effect of breed by date interaction on flank/upper leg cleanliness in Trial 2 ($P < 0.05$).



(A)



(B)

Figure 2.14. Effect of breed **(A)** and lactation number **(B)** on lower leg cleanliness for Holsteins and Jerseys in Trial 2. Bars with different letters are significantly different ($P < 0.01$).

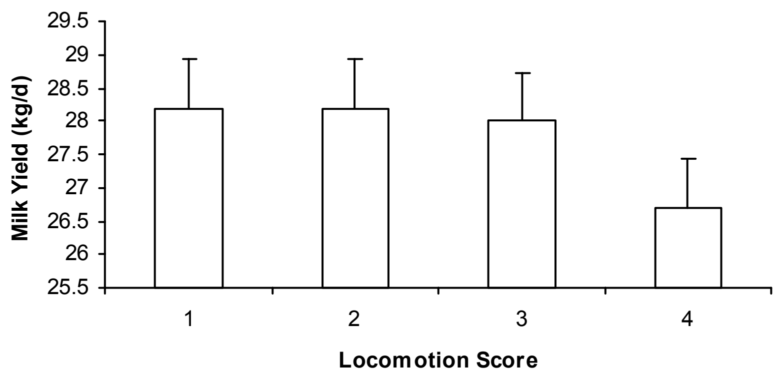


Figure 2.15. Effect of locomotion score on daily milk yield (kg) for Holsteins and Jerseys in Trial 2. There were no significant differences (*P* = 0.27).

Chapter 3: Effect of Relocation on Parlor Behaviors and Stress

ABSTRACT

The objective of this study was to examine how relocation affects measures of cow behavior and stress in the milking parlor. In an attempt to assess stress, cows at the Virginia Tech Dairy Center were allocated to three groups in separate pens: 1) access to a rubber mat in the feed bunk area (MAT; n = 18); 2) cows in the northeast section of the new facility (NE; n = 23); and 3) two breeds in the northwest section (NW; n = 22 Holsteins and 22 Jerseys). Trial 1 compared all Holsteins in the herd and Trial 2 compared the breed differences in the NW section. Parity was balanced across groups. Milking parlor behaviors observed were cow reactivity (REACT), latency to enter the parlor (LAT), and plasma cortisol (CORT). Cow reactivity was scored on a numeric scale (0 = ideal milker to 3 = steps and kicks frequently) to define behavior during udder preparation, claw fitting, and milking. Latency to enter the parlor was the time necessary for each cow to enter the milking parlor. CORT was assessed by RIA. Data were grouped by period with d –14 and –4 as PER 1; d 0 (relocation date) as PER 2; d 1 and 2 as PER 3; and d 7, 14, and 21 as PER 4. Data were analyzed using the Mixed Model procedure of SAS. In Trial 1, relocation caused an increase ($P < 0.05$) in CORT, monocytes, and LAT across all groups. There was a decrease in lymphocytes and p.m. milk yield across all groups during PER 2, but daily milk yields were not affected until PER 4. In Trial 2, LAT was 8.9 ± 0.7 for Holsteins vs. 5.6 ± 0.8 s for Jerseys ($P < 0.05$). Holsteins averaged 6.9 ± 0.7 vs. 4.8 ± 0.6 ng/mL CORT for Jerseys ($P < 0.05$). However, Jersey p.m. milk production during PER 2 decreased more dramatically than Holstein milk production. Treatment did not have an effect on prolonged stress, but overall, relocation caused an acute stress and cows quickly adapted to the new facility. Jerseys had lower CORT and LAT, but milk yields were more affected than Holsteins, suggesting measures of stress do not reflect decreases in milk yield.

Key words: relocation, parlor reactivity, plasma cortisol, stress

3.1 Introduction

There is a wide variety of stressful situations that can affect dairy cows, and multiple responses are possible. Isolation from peers, aversive handling, and transport are several common stressors in cattle. Reactions can range from behavioral changes such as increased vocalization, kicking, and defecation, (Watts and Stookey, 2000; Rushen et al., 1999a,b) or physiological responses including increased plasma cortisol, heart rate, or body temperature (Dixit et al., 2001; Boissy and le Neindre, 1997). Relocation to a new facility offers the opportunity to examine many different stressors and the responses provoked.

Varner et al. (1983) examined the differences in response when cows were moved a distance of 100 m compared to cows moved a distance of 7 km. It was determined that the stress of moving cows such short distances had minimal effect on stress and responses typically lasted less than 12 h. Similarly, calves transported for periods of 6 h or 18 h showed most symptoms of stress (increased plasma corticosteroid concentration and neutrophil to lymphocyte (**N:L**) ratio) after loading or unloading, suggesting that transport was not as stressful as preparing to be transported (Kent and Ewbank, 1986).

Changes in the milking parlor can also affect cow behavior. Heavier milking claws caused cows to be more temperamental during milking than lighter claws (Hillerton et al., 2001). Being milked in an unfamiliar environment can cause the inhibition of milk ejection (Bruckmaier et al., 1997). The parlor in the old Virginia Tech Dairy Center facility allowed cows to be milked from only one side, compared to being milked from either side in the new parlor. Being milked on the non-preferred side has been shown to increase latency to enter the parlor and heart rate in cows (Hopster et al., 1994).

Relocation to a new facility did not involve long transport periods or novelty milking in isolation or constantly changing surroundings. Therefore, it was expected that there would be an increase in behavioral and physiological indicators of stress, but these responses would be short-lived as the cows became acclimated to their new habitat.

3.2 Materials and Methods

Animals and Treatments

This study took place at the Virginia Tech Dairy Center from July 2004 to August 2004. The objective of this study was to determine the effect of relocation on stress in dairy cows by examining parlor behaviors, cortisol, and white blood cell (**WBC**) ratios. Before relocation, cows were divided into treatment groups to acclimate them to a new social structure. However, most treatments were not implemented until after relocation. Grouping was balanced for stage of lactation and lactation number. The treatment groups were as follows: 1) Holstein cows (n = 18) exposed to a rubber mat placed in front of the feed bunk (**MAT**); 2) Holstein cows (n = 23) in the northeast section of the new facility (**NE**); and 3) Holstein (n = 22) and Jersey cows (n = 22) in the northwest section (**NW**) (Figure 3.1).

The rubber mat had dimensions of 22 m x 2 m x 1.5 cm and was a recycled conveyor belt from a mining operation (VA Carolina Belting, Inc.; Salem, VA). It was placed in front of the feed bunk area and anchored to the alley with screws on the end of the mat closest to the alley flush system water release point to prevent movement from the feed bunk area. All cows in the NE group were fed through Calan doors, and were acclimated to the Calan door system before relocation (Pence, 2005). They were fed one of three diets for 3 wk before and 9 wk after relocation. The diets were 1) a TMR identical to that fed to the rest of the herd; 2) TMR top-dressed with alfalfa hay; and 3) TMR top-dressed with grass hay. The amount of TMR fed was determined by individual cow eating rates; the hay top-dressing comprised 10% of the feed presented to the cows daily. The NW group was divided into stocking densities of 0.67, 0.83, 1.00, or 1.17 cows per stall (Pence, 2005). Each NW sub-group had equal numbers of Holsteins and Jerseys; the stocking density of 0.67 cows per stall utilized 8 cows and 12 stalls; 0.83 cows per stall utilized 10 cows and 12 stalls; 1.00 cow per stall utilized 12 cows; and 1.17 cows per stall utilized 14 cows and 12 stalls. Those cows that were not in the NW section were housed in freestall pens with a stocking density of 1.00 cow per stall pre-relocation; after relocation, MAT cows were stocked at 0.50 cows per stall and NE cows at 1.00 cow per stall.

Before relocation, cows at the old facility were housed in a freestall barn with grooved concrete alleys. Stalls (1.07 x 2.13 m) were bedded with sawdust and partitioned by wooden dividers. Freestall pens had between 20 and 30 stalls. A farm employee scraped alleys daily at 0300 h. Feeding occurred at 0900 h; feed bunks were located centrally to the freestall pens, and raised 0.68 m from the ground. Automatic watering troughs were located next to feed bunks and at the end of freestall pens. The morning of relocation, farm employees led cows according to treatment to the new facility, a distance of approximately 131 m. The new facility was freestall housing with grooved concrete alleys. Stalls (1.17 x 2.29 m) were bedded with canvas mattresses covered lightly with sawdust and partitioned by pipe-loop dividers. Freestall pens had between 12 and 64 stalls each. Each alley in the new facility was flushed twice daily by an automatic alley flush system. Feeding occurred at 1000 and 1400 h. Feed bunks were located in the center of the freestall area and were directly on the ground. With the exception of the cows on the feeding study, which used Calan doors, cows were required to pass through head gates to gain access to feed.

The milking parlor at the old facility (Figure 3.2) was a single side-opening parlor with six milking stalls. Milking began at 0100 h and 1300 h daily. Cows were milked according to treatment group and were required to walk uphill on a gravel walkway a distance of approximately 60 m to reach the holding area if not housed in freestall pens adjacent to the milking parlor. The holding area had dimensions of 20.6 m x 5.3 m when the chain crowd gate was extended to full capacity. Cows entered the parlor individually once a milking stall was available. Upon exit from the parlor, cows walked through an alley that was 7.2 m x 0.85 m. Cows remained in a separate holding pen until all other cows in the group were milked and then returned to their freestall pen. The milking parlor at the new facility (Figure 3.3) was a double-eight herringbone design. Milking began at 0100 h and 1300 h. All freestall pens were adjacent to the parlor, so walking distance was minimal (5 to 30 m). The holding area measured 11.7 m x 7.6 m when the vertical lifting crowd gate was extended to full capacity. Cows entered the parlor eight at a time on each side of the parlor. Upon exit from the parlor, cows were required to walk down an alley 15.5 m x 1.1 m and pass over a set of scales, and had access to their freestall pens immediately.

Parlor Behavior Scoring

Parlor behaviors were used as indicators of stress caused by being milked in an unfamiliar environment. Behaviors measured included entry order, latency to enter the parlor, cow reactivity during milking, incidences of defecation and urination, and milk claw fitting time (Paranhos da Costa and Broom, 2001). Data collection occurred on d -14, -4, 0, 1, 2, 5, 7, and 14. Entry order was defined as the order in which each subject entered the milking parlor with respect to its group. Latency to enter the parlor was the time (s) required for each cow to completely pass through the entrance of the milking parlor. If more than one cow entered the parlor at a time, then the time for the second cow was measured from the time the first cow completely passed through the entrance until the second cow completely entered the parlor. Cows were given a voluntary entry period of 60 s before farm employees pushed them into the parlor; the voluntary entry period was added to latency time if pushing was required. Cow reactivity during preparation and fitting was a 4-point scale (Table 3.1) based on a milking temperament scoring system devised by Dickson et al. (1969) and a milking reactivity scale created by Hillerton et al. (2001). A score of zero was indicative of how a docile cow behaved during preparation, claw fitting, and milking. A score of 1 was given to a cow that stood still in the milking stall but shifted her weight or flicked her tail. A score of 2 meant that there were frequent movements, foot lifting, and tail flicking. Cows that received a score of 3 were restless during preparation, kicked at the milker, and quivered at contact with the milker. Incidences of defecation and urination were measured from the time a cow entered the parlor until she exited the parlor. Milk claw fitting time (s) was the time it required the milker to attach the claw to the cow and begin milking. Timing began as soon as the milker removed the claw from its resting position.

Data collection required four observers. In the old facility, two observers stood at the entry point of the parlor; one recorded entry order and the other used a stopwatch to record latency to enter the parlor. The remaining two observers were stationed at opposite ends of the milking pit and recorded cow reactivity, defecation, urination, and milk claw fitting time with a stopwatch (Figure 3.2). In the new facility, an observer was stationed at the edge of each milking platform to record entry order and latency to enter the parlor. Two observers were stationed at the edge of the milking pit to record cow reactivity, defecation, urination, and milk claw fitting time (Figure 3.3). Each observer was responsible for the same measures during each collection date to minimize error.

Blood Smear Preparation

Blood samples were collected after p.m. milking on d -7, -1, 0, 1, 2, 7, 14, and 21. In the old facility, collection took place at head gates adjacent to the milking parlor. In the new facility, collection occurred at the head gates located at the feed bunk in the freestall pens. Samples of 10 mL per cow were collected from jugular veins using 38 mm, 20-gauge Vacutainer (BD; Franklin Lakes, NJ) needles and K_3 EDTA (17.55 mg)-coated evacuated tubes. Samples were stored on ice until all samples had been collected, a time of no greater than 2 hr. Then they were transported to a lab for preparation of blood smears (Gross and Siegel, 1983). One drop of blood from each sample was placed on a microscope slide and smeared by centrifugation with a Larc Spinner (Corning Glass Works, Scientific Instruments Division; Metfield, MA). After smears dried, slides were placed on a staining rack and flood stained with May-Greenwald stain solution (Sigma-Aldrich; St. Louis, MO) for 7 min. Slides were then rinsed with a buffer solution (5.6 g NaH_2PO_4, 21.4 g Na_2HPO_4 per liter dH_2O; 7.2 pH) and air dried with a pipette. Geimsa-Wright stain (Sigma-Aldrich; St. Louis, MO) was diluted 50:50 with dH_2O and used to flood the slides. After 15 min, the Geimsa-Wright stain was poured off; the slides were rinsed in dH_2O, and dried with bibulous paper.

Plasma Cortisol

Once blood drops were placed on microscope slides for smearing, the evacuated tubes were recapped and centrifuged at 4°C at 1851 x g for 20 min to separate plasma. Plasma was transferred to 10 mL centrifuge tubes (Sarstedt; Newton, NC) and stored in a freezer at -20°C for later analysis. Samples were thawed and plasma cortisol concentration was analyzed in duplicate by ^{125}**I** radioimmunoassay (**RIA**) (DiaSorin Inc.; Stillwater, MN). A tracer-buffer reagent was added in 1 mL aliquots to each of the sample tubes. Standards and unknown samples were placed in rabbit anti-cortisol GammaCoat tubes and incubated for 45 min in a 37°C water bath. Plasma cortisol was determined through competitive protein binding. The inter-assay coefficient of variation was 11.4% and the intra-assay coefficient of variation was 13.5%.

WBC Counting

Blood smears were read under the microscope at 40x magnification. WBC counting consisted of counting neutrophils (**N**), eosinophils (**E**), lymphocytes (**L**), and monocytes (**M**) until the total number of leukocytes counted reached 100 cells. The N:L ratio was determined by dividing the number of neutrophils by the number of lymphocytes (Gross and Siegel, 1983). The average percentage of neutrophils in cattle is 15 to 45% of total WBCs; the average percentage of lymphocytes is 45 to 75%. The mean N:L ratio is 0.5:1 (Merck Veterinary Manual, 2003).

Statistical Analysis

Main effects of treatment, lactation number, date, and their interactions were tested with the MIXED procedure of SAS (SAS Institute, Inc.; Cary, NC; Version 8.2, 1999). Collection dates were divided into time periods with respect to relocation in the following manner: **PER 1** = d -7 and -1; **PER 2** = d 0; **PER 3** = d 1 and 2; and **PER 4** = d 7, 14, and 21. Dependent variables were parlor entry order, latency to enter the parlor, cow reactivity to milking, defecation, urination, milking claw fitting time, plasma cortisol, blood leukocytes N, E, L, and M, N:L ratio, and milk yield. Trial 1 examined the parlor behaviors and stress in Holsteins. Treatments were MAT, NE, or NW and lactation number was 1, 2, or greater than or equal to 3. Trial 2 examined breed differences between Holsteins and Jerseys in the NW group. The following model statement was used for Trial 1: $Y_{ijkl} = \mu + T_i + L_j + TL_{ij} + C_{(ij)k} + P_l + TP_{il} + LP_{jl} + TLP_{ijl} + E_{ijkl}$

where:

Y_{ijkl} = dependent variable(s)
μ = mean
T_i = fixed effect of treatment (i = MAT, NE, or NW)
L_j = fixed effect of lactation number (j = 1, 2, or $\geq$3)
TL_{ij} = effect of the interaction between treatment and lactation number
C_{ijk} = random effect of cow within treatment and lactation number
P_l = time period with respect to relocation (l = 1-4)
TP_{il} = effect of the interaction between treatment and time period
LP_{jl} = effect of the interaction between lactation number and time period
TLP_{ijl} = effect of interaction between treatment, time period, and lactation number
E_{ijkl} = residual

The following model was used for Trial 2: $Y_{ijkl} = \mu + B_i + L_j + BL_{ij} + C_{(ij)k} + P_l + BP_{il} + LP_{jl} + BLP_{ijl} + E_{ijkl}$

where:

Y_{ijkl} = dependent variable(s)
μ = mean
B_i = fixed effect of breed (i = Holstein or Jersey)
L_j = fixed effect of lactation number (j = 1, 2, or $\geq$3)
BL_{ij} = effect of the interaction between breed and lactation number
C_{ijk} = random effect of cow within treatment and lactation number
P_l = time period with respect to relocation (l = 1-4)
BP_{il} = effect of the interaction between breed and time period
LP_{jl} = effect of the interaction between lactation number and time period
BLP_{ijl} = effect of interaction between breed, time period, and lactation number
E_{ijkl} = residual

Results are presented as LS means ± SEM. Differences were considered significant at $P < 0.05$ and trends at $P < 0.10$. Differences were determined with contrasts.

3.3 Results

In Trial 1, period had an effect on entry order, latency to enter the parlor, cow reactivity, plasma cortisol, lymphocytes, monocytes, and p.m. milk yield (Table 3.2), with PER 2 negatively affecting all measures except monocytes. Lactation number had an effect on entry order with first lactation cows more likely to enter the parlor last and second lactation cows entering first (Figure 3.4).

Treatment by lactation number interaction had a significant impact on entry order and latency to enter the parlor (Figure 3.5). Third and greater lactation cows in the MAT group entered the parlor later than first and second lactation cows, and were more reluctant to enter compared to older cows in the NE group. Treatment by period interaction had an effect on p.m. milk yield, total milk yield, and plasma cortisol (Figure 3.6). Cortisol was elevated in PER 2 for NE and MAT cows, but not NW cows. However, NW cows had lower cortisol in PER 3 which increased in PER 4. Afternoon milk yield was lower for PER 2, rebounded in PER 3 and was lower in PER 4 except for MAT cows. Total milk yield declined from PER 1 and 2 to PER 4. The interaction of lactation number by time period affected p.m. milk yield (Table 3.3), where second and greater lactation cows had a decline in PER 2, an increase in PER 3, and a decline in PER 4, while first lactation cows had no change.

In Trial 2, breed significantly affected to latency to enter the parlor and plasma cortisol, with Jerseys entering the parlor more willingly than Holsteins (5.7 s compared to 8.6 s) and having lower plasma cortisol (4.8 ng/mL vs. 6.9 ng/mL) ($P < 0.05$; Table 3.4), while eosinophils were lower in Holsteins than Jerseys. Period had an effect on latency to enter the parlor, p.m. milk yield, daily milk yield, plasma cortisol, lymphocytes, monocytes, and N:L ratio (Table 3.5). In PER 2, cows were reluctant to enter the new parlor, had lower lymphocytes, higher monocytes and N:L ratio, and lower p.m. milk yield, showing the effects of acute stress. Cortisol declined from PER 1 and 2 during PER 3 and 4. Daily milk yields decline from PER 1 through PER 4. Lactation number had an effect on latency to enter the parlor, with second and third or greater lactation cows taking longer to enter the parlor (8.8 ± 0.86 s and 7.5 ± 1.11 s, respectively) than first lactation cows (5.4 ± 0.79 s) ($P < 0.05$).

Breed by lactation number interaction had an effect on latency to enter the parlor and N:L ratio (Figure 3.7). Second lactation and older Holsteins were more reluctant to enter the parlor than first lactation Holsteins or any Jersey. Oldest Holsteins and youngest Jerseys had the lowest N:L ratios. Afternoon milk yield was affected by breed by period interaction, as Jersey milk production decreased 45.1% from PER 1 to PER 2, while Holstein milk production decreased by 20.1% during the same time frame ($P < 0.05$; Figure 3.8A). While not significant, decreases were found in total milk yield (Figure 3.7B). Additionally, lactation number by period interaction indicated that second lactation cows decreased most in p.m. milk production from PER 1 to PER 2 (Table 3.6).

3.4 Discussion

In Trial 1, the effects of relocation are evident by an increase in entry order (%), latency to enter the parlor, plasma cortisol, and a decrease in p.m. milk yield during PER 2. The increase in entry order remained high through PER 3 and PER 4, an indication of a permanent change after relocation in which cows that previously entered the parlor earlier during milking were more likely to enter the parlor later with respect to their group. Latency to enter the parlor increased during PER 2, but decreased in each subsequent period, returning to times slightly lower than pre-relocation times (7.1 s vs. 6.6 s). A lower p.m. milk yield and increased cortisol is consistent with the findings of Rushen et al. (2001), who found that cows isolated and milked in an unfamiliar area had lower milk yields than those milked in familiar settings. However, total milk yields did not differ between treatments after collection of residual milk. Plasma cortisol also increased for both treatments during milking, but cows milked in isolation experienced a greater increase in cortisol levels.

Entry order had the greatest variation in results. First lactation cows were more likely to enter the parlor after second lactation cows (57.0% vs. 47.9%).

Third or greater lactation cows in the MAT group, though, had the highest entry order percentage (57.2%) than first and second lactation cows in the MAT group (45.8 and 40.7, respectively). Although previous studies have shown that entry order may be affected by social dominance (Dickson et al., 1969) or novelty feed in the parlor (Ceballos and Weary, 2002), it may not be a good indicator of stress in the milking parlor.

Cows in the NE and MAT groups experienced increases in plasma cortisol during PER 2, suggesting that relocation was a stressful situation. PER 2 levels did not differ from PER 1 in the NW group. There was a decline below basal levels in PER 3 across all treatment groups, and by PER 4 plasma cortisol returned to levels slightly lower than baseline. The steep decline in cortisol levels from PER 2 to PER 3 could be a result of habituation or an exhaustion of the hypothalamic-pituitary-adrenal axis (Gwazdauskas et al., 1980) from the stress of relocation to the new facility.

Cows across all treatment groups produced less milk during p.m. milking during PER 2. These values returned to or near basal levels during PER 3. While p.m. milk yields were lower during PER 2, daily milk yields did not differ from other periods (Figure 3.6). There was a decline in daily milk yields by PER 4, which is likely due to cows reaching later stages of lactation (NRC, 2001).

In Trial 2, Jerseys entered the parlor more quickly than Holsteins and had lower plasma cortisol levels but higher eosinophils. Whereas there were no increases in cortisol during PER 2 as in Trial 1, N:L ratio did increase during PER 2 across both breeds in Trial 2, which suggests that relocation was a stressor. Holsteins and Jerseys responded differently with respect to N:L ratio; third or greater lactation Holsteins had a lower ratio (0.42 N:L), while first lactation Jerseys experienced the same effect (0.52 N:L; Figure 3.7). Latency to enter the parlor was not affected by lactation in Jerseys, but second lactation (12.4 s) and third or greater lactation (8.6 s) Holsteins took longer to enter than first lactation Holsteins (5.6 s; Figure 3.7). Jerseys saw a greater drop in p.m. milk yield (7.6 kg compared to 13.8 kg during PER 1) during PER 2 than Holsteins

(15.4 kg compared to 19.2 kg during PER 1), but daily milk yield was not affected by relocation, similar to Trial 1 (Figure 3.8).

3.5 Conclusions

There were noticeable effects on behavior and stress associated with relocation to the new facility. All changes, physiologically and behaviorally, were most noticeable during PER 2, the day of relocation to the new facility. These changes were short-lived and none were dramatic, so while relocation may have presented a stressful situation, the stress was not great. This could be a result of cows being transported on foot to the new location instead of a greater distance by road (Tarrant, 1990), cows being from a research herd, and therefore more acclimated to stressful situations, or cows transported to a facility that was superior to the old one, which made habituation easy. Holsteins and Jerseys showed different signs of stress, but it is difficult to determine if one breed handled the stress better than the other because stress indicators were lower for Jerseys, but milk yield was more depressed.

3.6 Implications

Relocation to a new facility caused cows to change their behavior and physiological stress indicators were elevated the day of relocation. These changes do not have a great impact on daily milk production. However, milk retained after relocation is ejected during the subsequent milking. Future research to determine the effects of relocation on behavior and stress should examine methods of reducing stress associated with relocation and include a larger number of Jerseys to determine breed differences.

Table 3.1. Cow Reactivity Scoring Scale[1]

Reactivity Score	Assessment Criteria
0	Very quiet; never gives any trouble; extremely docile during milking and preparation; the “ideal” milker.
1	Stands quietly in stall; not bothered by preparation or milking, but may move frequently, shifting weight from side to side; may flick tail occasionally; gives very little trouble.
2	Generally quiet, but moves around a lot; may lift feet occasionally during preparation or milking, but does not kick; flicks tail frequently or appears restless occasionally.
3	Appears very restless during preparation or milking; kicks at handler occasionally; steps from side to side a great deal; quivers when a hand is placed on her.

[1]Adapted from Dickson et al. (1970) and Hillerton et al. (2001)

Table 3.2. Effect of time period on behaviors and measures of stress for Holsteins in Trial 1.

	Period					
	d -14,-4	d 0	d 1,2	d 7,14,21		
Variable	PER 1	PER 2	PER 3	PER 4	SE	*P* <
Entry order (%)	48.5[a]	55.0[b]	54.0[b]	55.0[b]	2.31	0.05
Latency to enter parlor (s)	7.1[a]	13.0[b]	10.2[c]	6.6[a]	1.11	0.01
Cow reactivity (0-3)	0.52[a]	0.58[a]	0.38[b]	0.60[a]	0.08	0.05
Defecations[1]	0.06	0.07	0.13	0.06	0.04	0.15
Urinations[1]	0.07	0.06	0.09	0.06	0.03	0.93
Plasma cortisol (ng/mL)	7.2[a]	10.1[b]	3.0[c]	4.5[d]	0.54	0.01
Neutrophils (%)	29.1	34.3	30.8	30.9	1.77	0.07
Eosinophils (%)	4.3	3.2	4.4	4.5	0.49	0.07
Lymphocytes (%)	57.4[a]	50.2[b]	53.7[abc]	54.9[ac]	1.98	0.01
Monocytes (%)	9.2[a]	13.8[b]	11.1[a]	9.4[a]	1.10	0.01
N:L ratio	0.59	0.73	0.62	0.62	0.05	0.12
p.m. Milk yield (kg)	15.2[a]	11.9[b]	15.0[a]	13.2[c]	0.42	0.01
Milk yield (kg/d)	30.6[a]	29.8[ab]	28.2[b]	24.2[c]	0.82	0.01

[1]Incidences during milking
[abcd] Means within rows with different superscripts differ based on contrasts of each period.

Table 3.3. Effect of lactation number by time period interaction on p.m. milk yield (kg) for Holsteins (181 DIM at d 0) in Trial 1.

	Period				
	d -14,-4	d 0	d 1,2	d 7,14,21	
Lactation Number	PER 1	PER 2	PER 3	PER 4	SE
1	13.9	12.6	13.5	11.8	0.68
2	15.2[a]	11.0[b]	14.7[a]	13.2[c]	0.66
≥3	16.5[a]	12.2[b]	16.9[a]	14.6[c]	0.84

[abc] Means within rows with different superscripts differ at $P < 0.05$ based on contrasts of each period.

Table 3.4. Effect of breed on behaviors and measures of stress for Holsteins and Jerseys in Trial 2.

	Breed			
Variable	Holstein	Jersey	SE	*P* <
Entry Order (%)	54.5	52.8	3.62	0.74
Latency to enter parlor (s)	8.9[a]	5.6[b]	0.76	0.01
Cow reactivity (0-3)	0.51	0.52	0.10	0.95
Defecations[1]	0.10	0.12	0.03	0.70
Urinations[1]	0.76	0.76	0.02	0.99
Plasma cortisol (ng/mL)	6.9[a]	4.8[b]	0.65	0.05
Neutrophils (%)	31.1	30.3	1.54	0.70
Eosinophils (%)	3.0[a]	5.5[b]	0.64	0.01
Lymphocytes (%)	55.0	52.1	1.99	0.27
Monocytes (%)	11.8	12.0	1.04	0.87
N:L ratio (0-1.5)	0.63	0.65	0.05	0.78
p.m. Milk yield (kg)	16.8[a]	11.1[b]	0.54	0.01
Milk yield (kg/d)	32.9[a]	22.2[b]	0.99	0.01

[1]Incidences during milking
[ab] Means within rows with different superscripts differ based on contrasts of each breed.

Table 3.5. Effect of time period on behaviors and measures of stress for Holsteins and Jerseys in Trial 2.

	Period					
	d -14,-4	d 0	d 1,2	d 7,14,21		
Variable	PER 1	PER 2	PER 3	PER 4	SE	*P* <
Entry Order (%)	49.2	55.4	54.0	56.1	3.52	0.22
Latency to enter parlor (s)	6.7[a]	9.7[b]	7.6[ac]	5.0[a]	0.99	0.01
Cow reactivity (0-3)	0.58	0.40	0.44	0.63	0.11	0.22
Defecations[1]	0.09	0.08	0.14	0.12	0.05	0.76
Urinations[1]	0.08	0.06	0.10	0.07	0.03	0.92
Plasma cortisol (ng/mL)	8.6[a]	8.1[a]	2.5[b]	4.2[c]	0.67	0.01
Neutrophils (%)	28.6	34.0	31.0	29.3	1.99	0.09
Eosinophils (%)	4.9	3.6	4.4	4.2	0.67	0.28
Lymphocytes (%)	56.7[a]	49.1[b]	52.6[ab]	55.7[a]	2.16	0.01
Monocytes (%)	9.7[a]	15.6[b]	11.9[c]	10.4[ac]	1.39	0.01
N:L ratio (0-1.5)	0.59[a]	0.77[b]	0.62[a]	0.57[a]	0.05	0.05
p.m. Milk yield (kg)	16.5[a]	11.5[b]	15.0[c]	12.9[d]	0.47	0.01
Milk yield (kg/d)	32.3[a]	28.3[b]	27.9[b]	21.8[c]	1.02	0.01

[1]Incidences during milking

[abcd] Means within rows with different superscripts differ based on contrasts of each period.

Table 3.6. Effect of lactation number by time period interaction on p.m. milk yield (kg) for Holsteins and Jerseys (193 DIM at d 0) in Trial 2.

	Period				
	d -14,-4	d 0	d 1,2	d 7,14,21	
Lactation Number	PER 1	PER 2	PER 3	PER 4	SE
1	14.1[a]	10.9	12.2	10.4	0.68
2	17.4[a]	10.6[b]	14.8[c]	13.0[d]	0.75
$\geq$3	18.1[a]	12.9[b]	17.8[a]	15.2[c]	0.98

[abcd] Means within rows with different superscripts differ at $P < 0.05$ based on contrasts of each period.

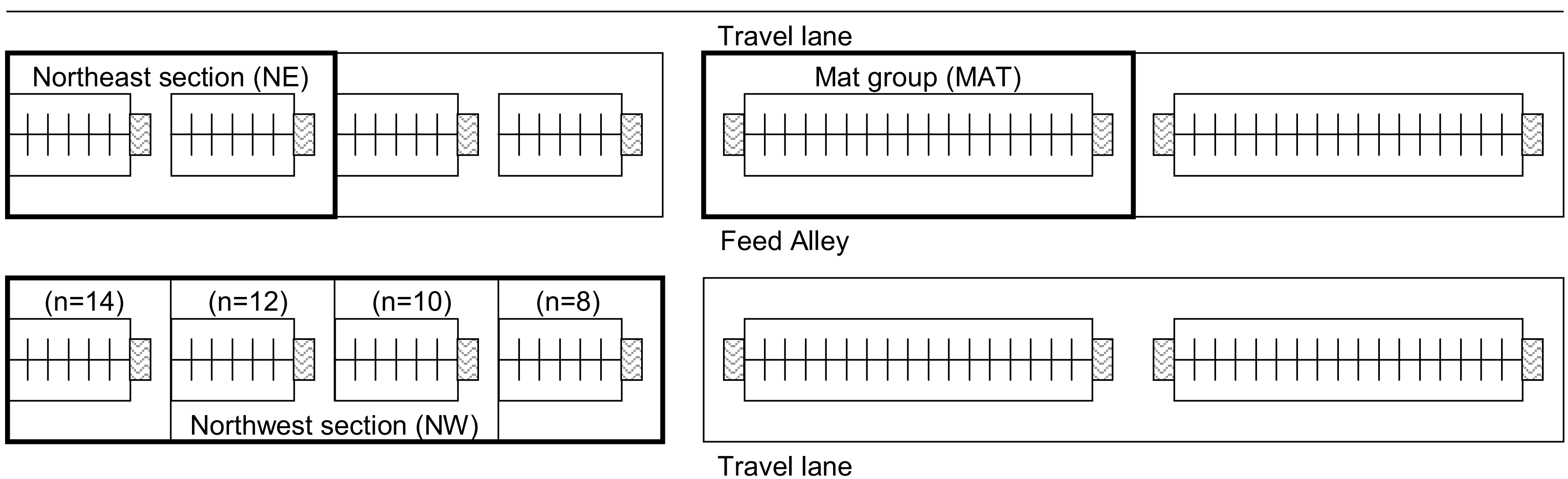


Figure 3.1. Freestall housing assignments in the new facility.

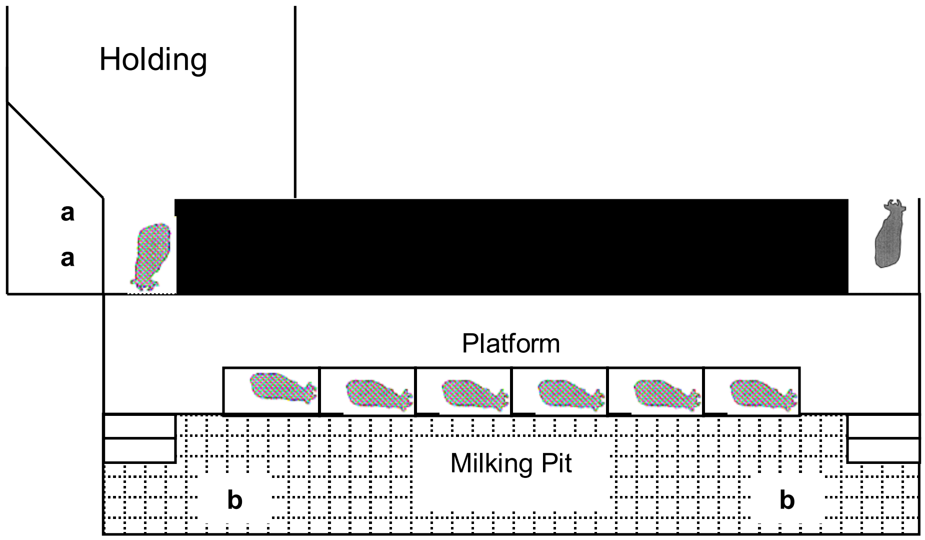


Figure 3.2. Milking parlor in the old facility. Observers recording entry order and latency to enter the parlor (s) are represented by **(a)**. Observers recording cow reactivity (0-3), defecation, urination, and fitting time (s) are represented by **(b)**.

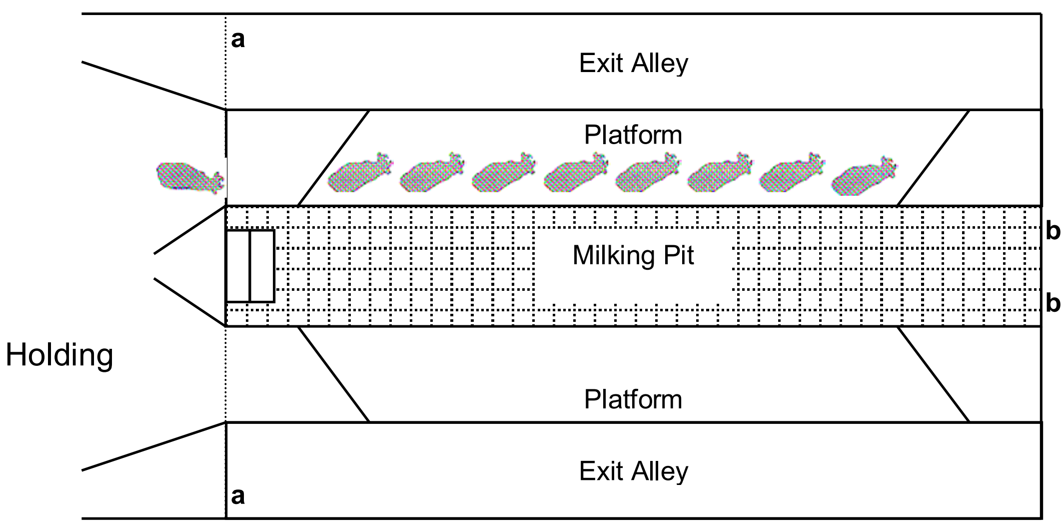


Figure 3.3. Milking parlor in the new facility. Observers recording entry order and latency to enter the parlor (s) are represented by **(a)**. Observers recording cow reactivity (0-3), defecation, urination, and fitting time (s) are represented by **(b)**.

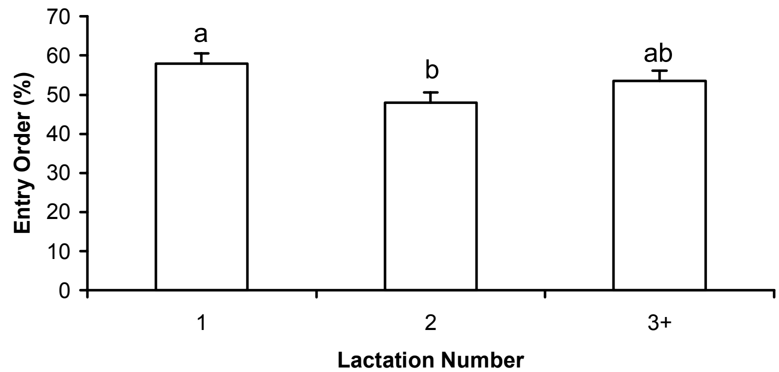
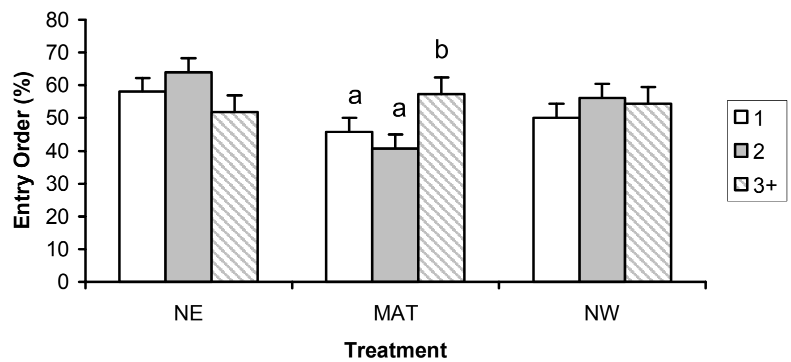
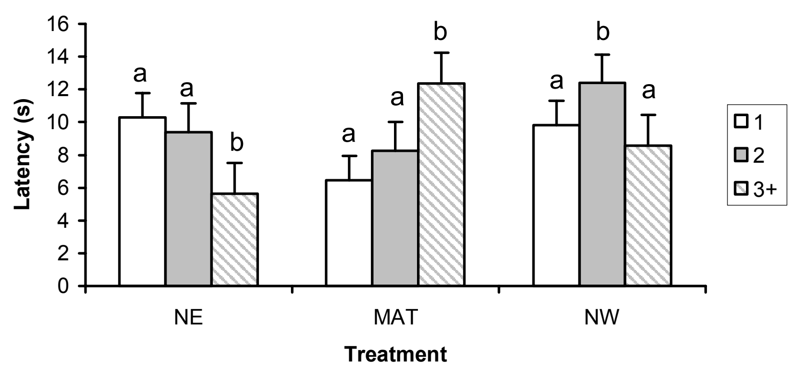


Figure 3.4. Effect of lactation number on entry order (%) for Holsteins in Trial 1. Bars with different letters are significantly different ($P < 0.05$).

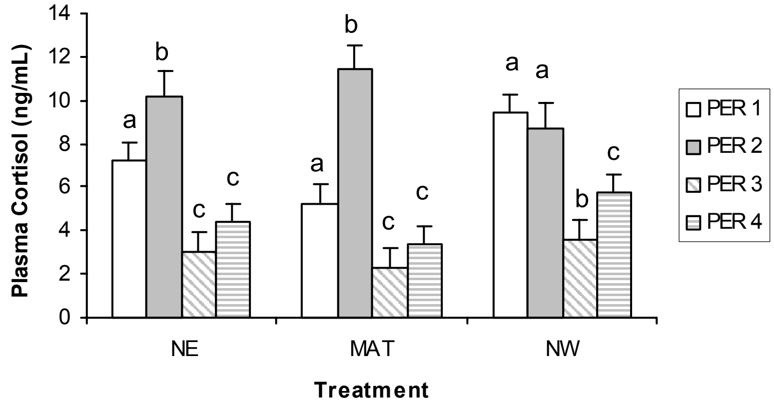


(A)

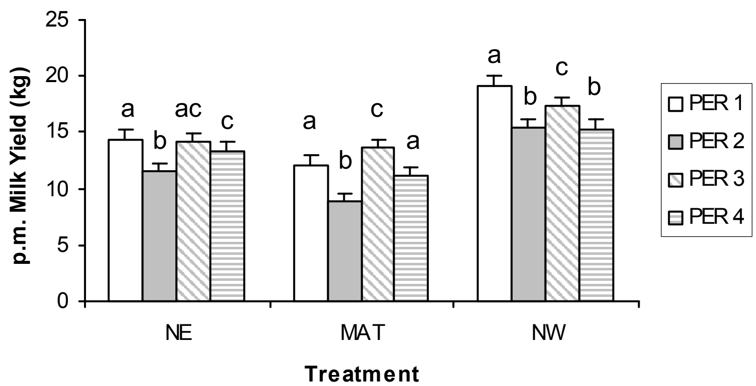


(B)

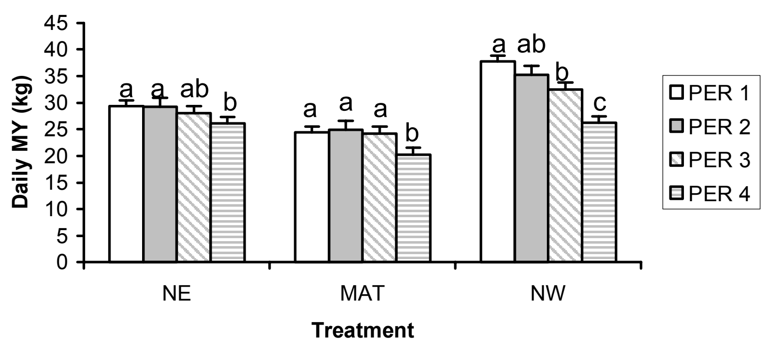
Figure 3.5. Effect of treatment by lactation number interaction on entry order (%) **(A)** and latency to enter the parlor (s) **(B)** for Holsteins in Trial 1. Bars with different letters per treatment are significantly different ($P < 0.05$).



(A)

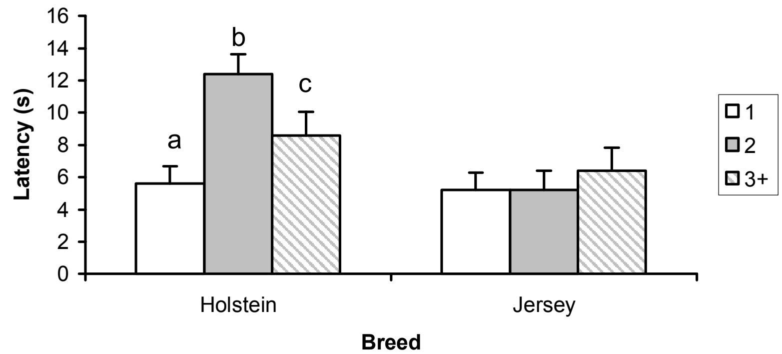


(B)

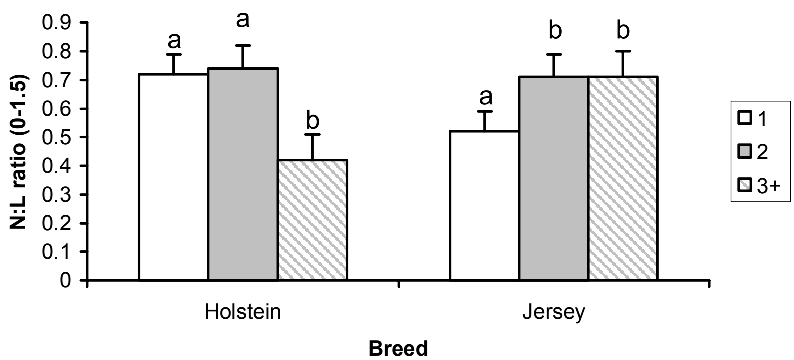


(C)

Figure 3.6. Effect of treatment by time period interaction on plasma cortisol (ng/mL) **(A)**, p.m. MY (kg) **(B)**, and daily MY **(C)** for Holsteins in Trial 1. Bars with different letters per treatment are significantly different ($P < 0.05$).

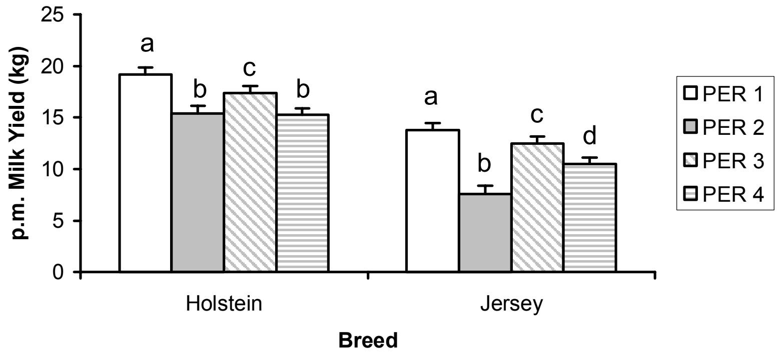


(A)

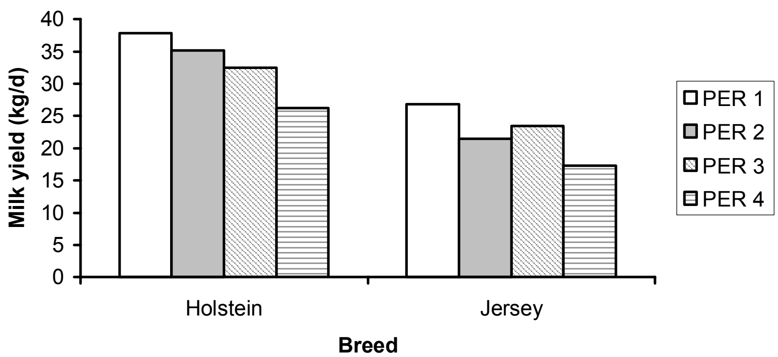


(B)

Figure 3.7. Effect of breed by lactation number interaction on latency to enter the parlor (s) **(A)** and N:L ratio **(B)** for Holsteins and Jerseys in Trial 2. Bars with different letters per breed are significantly different ($P < 0.05$).



(A)



(B)

Figure 3.8. Effect of breed by time period interaction on p.m. milk yield (kg) **(A)** and daily milk yield **(B)** for Holsteins and Jerseys in Trial 2. Bars with different letters per breed are significantly different (P < 0.05). Results in **(B)** are not significant (P = 0.28).

Chapter 4: Conclusions

In these studies, several variables were examined to determine the effect relocation to new dairy facilities has on the welfare of lactating dairy cows. In each of the studies, groups and breeds responded differently to relocation, and while specific observations can be made about some measures, other measures were more ambiguous in results and interpretation.

In experiment one, a rubber mat placed in front of the feed bunk helped prevent increased locomotion and lameness problems that affected cows with no mat. Although the results were not significant, there was a decrease in milk production with higher locomotion scores. The mat was not detrimental to cleanliness, and relocation to the new facility caused overall cleanliness of the herd to improve. Breed was not a factor in locomotion scoring, as Holsteins and Jerseys had similar results throughout observation. Jerseys were cleaner overall, which may be a result of access to larger freestalls in which to rest than they had access to previously.

In experiment two, relocation to the new facility caused cows in the herd to be more reactive during milking and take more time to enter the parlor. There was a decrease in milk production the afternoon after relocation, but residual milk was released in the following milking, so there was no effect on milk production for the day. Plasma cortisol increased the day of relocation, decreased drastically before returning to basal levels. Although there was an increase in cortisol levels, it still fell within the normal range for cattle. Jerseys had lower cortisol levels and were quicker to enter the parlor after relocation than Holsteins, but also had a greater decrease in milk production the afternoon after.

None of the results related to behavior and stress were severe, so relocation to the new facility was either less stressful than expected or cows acclimated more quickly than expected. Greater numbers of Jerseys are necessary to determine breed differences associated with behavior and stress. With proper management and attention to animal welfare, producers should be able to relocate a herd to a new facility without fear of detrimental effects to production.

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APPENDIX

Table A1 Frequency of locomotion score by date in Trial 1; average cows observed was 70/d. (Chapter 2: Effect of Relocation on Locomotion and Cleanliness on Dairy Cows)

	Covariate observation dates				Observation dates								
Score (% observed)	d -29	d -13	d -7	d -1	d 7	d 14	d 21	d 30	d 35	d 45	d 58	d 74	d 86
1	72.5	49.2	62.9	55.7	47.5	59.0	67.9	58.0	43.2	46.4	33.9	29.7	16.7
2	21.3	30.8	24.2	34.4	41.0	34.6	21.0	32.1	34.6	39.1	50.0	37.5	51.7
3	5.0	15.4	8.1	6.6	9.8	6.4	4.9	3.7	13.6	11.6	9.7	18.8	23.3
4	1.3	4.6	4.8	3.3	1.6	0.0	6.2	6.2	8.6	2.9	6.5	14.1	8.3

Table A2 Frequency of locomotion score by date in Trial 2; average cows observed was 42/d. (Chapter 2: Effect of Relocation on Locomotion and Cleanliness on Dairy Cows)

	Covariate observation dates				Observation dates								
Score (% observed)	d -29	d -13	d -7	d -1	d 7	d 14	d 21	d 30	d 35	d 45	d 58	d 74	d 86
1	97.7	68.6	86.0	77.3	74.4	84.1	86.4	70.5	45.5	55.8	36.4	28.6	15.4
2	2.3	22.9	11.6	20.5	20.9	15.9	11.4	22.7	38.6	37.2	45.5	42.9	43.6
3	0.0	8.6	0.0	0.0	0.0	0.0	0.0	0.0	9.1	4.7	6.8	21.4	33.3
4	0.0	0.0	2.3	2.3	4.7	0.0	2.3	6.8	6.8	2.3	11.4	7.1	7.7

Table A3 Frequency of observations by period for Holsteins and Jerseys (NW) in Trial 2. (Chapter 3: Effect of Relocation on Parlor Behaviors and Stress)

Period	Frequency	Percent	Cumulative Frequency	Cumulative Percent
1	88	25.0	88	25.0
2	44	12.5	132	37.5
3	88	25.0	220	62.5
4	132	37.5	352	100

Table A4 Chi-square test for equal proportions for Holsteins and Jerseys (NW) in Trial 2. (Chapter 3: Effect of Relocation on Parlor Behaviors and Stress)

Chi-square	44.0
Degrees of freedom	3
Pr > Chi-square	<0.001
Sample size	352

Table A5 Percentage (%) of laminitis scores observed by treatment group in Trial 1 (Chapter 2: Effect of Relocation on Locomotion and Cleanliness on Dairy Cows).

	Laminitis Score			
Treatment	0	1	2	3
SW	63.3	28.1	5.5	3.1
NE	77.7	15.8	6.0	0.5
MAT	50.7	33.8	11.8	3.7
NW	77.7	17.9	3.6	0.9

Table A6 Percentage (%) of disease scores observed by treatment group in Trial 1 (Chapter 2: Effect of Relocation on Locomotion and Cleanliness on Dairy Cows).

	Disease Score	
Treatment	0	1
SW	99.2	0.8
NE	99.5	0.5
MAT	100.0	0.0
NW	97.3	2.7

Table A7 Percentage (%) of laminitis scores observed by breed in Trial 2 (Chapter 2: Effect of Relocation on Locomotion and Cleanliness on Dairy Cows).

	Laminitis Score			
Treatment	0	1	2	3
Holstein	77.7	17.9	3.6	0.9
Jersey	88.2	8.3	2.1	1.4

Table A8 Percentage (%) of disease scores observed by breed in Trial 2 (Chapter 2: Effect of Relocation on Locomotion and Cleanliness on Dairy Cows).

	Disease Score	
Treatment	0	1
Holstein	97.3	2.7
Jersey	97.9	2.1

VITA

Crafton Oliver Wilkes was born in Roanoke, Virginia on July 12, 1981. He graduated from Franklin County High School in 1999. He began his undergraduate studies at Virginia Tech and was active in Beta Theta Pi, the Chemistry Club, and the Dairy Club. He graduated in May 2003 a Bachelor of Science degree in Dairy Science. In December 2005 he completed a Master of Science degree program in Dairy Science at Virginia Tech.

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