THESIS

THE PERSPECTIVES OF ANIMAL CARETAKERS ON UDDER EDEMA IN DAIRY CATTLE AND THE EFFECTS OF UDDER EDEMA ON PARLOR BEHAVIOR IN FIRST AND SECOND LACTATION DAIRY CATTLE

Submitted by

Cora Annette Okkema

Department of Animal Sciences

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Master's Committee:

Advisor: Temple Grandin

Lily Edwards-Callaway
Noa Román-Muñiz
Kirsten Eilertson

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ABSTRACT

THE PERSPECTIVES OF ANIMAL CARETAKERS ON UDDER EDEMA IN DAIRY CATTLE AND THE EFFECTS OF UDDER EDEMA ON PARLOR BEHAVIOR IN FIRST AND SECOND LACTATION DAIRY CATTLE

Udder edema is a metabolic disorder in dairy cattle that results in lymphatic fluid being trapped in mammary tissues. Severe cases of udder edema can present in the brisket, navel, upper rear legs, and vulva. Known factors associated with udder edema include genetics, nutrition, oxidative stress, and high body condition score. Furthermore, as heifers are developing the mammary system during late-gestation, changes to their physiology are also occurring that influence the presentation of udder edema. Udder edema has been shown to negatively affect milk production, damage support structures of the mammary gland, and increase the risk of secondary diseases, such as mastitis and udder cleft dermatitis. With the concentration of udder edema, two studies were conducted examining dairy caretaker perspectives on udder edema and the effect udder edema has on behavior during the milk procedure. The objectives of the dairy caretaker survey were to 1) capture and evaluate current perspectives on udder edema from dairy caretakers, 2) assess caretakers' knowledge about factors that influence the development of udder edema, and 3) understand what the current monitoring practices of udder edema are on farms. The survey was distributed through an industry magazine and in a dairy exposition's electronic newsletter. A total of thirty caretakers completed the survey. The majority of dairy animal caretakers within the study agreed with the following statements: udder edema is a part of udder health, udder edema is more commonly seen in heifers (93.3%), an industry-standard rating scale would be useful to monitor udder edema

(70%), it is important to be trained to identify udder edema (93.3%) , udder edema increases the risk for mastitis (73.3%), udder edema can be managed through nutrition (73.3%), udder edema negatively affects milk production (80%), high genetic milk production potential increases the risk of udder edema (70%), udder edema affects the attachment of the milking unit (90%), udder edema is painful (90%), and that udder edema is an animal welfare issue (86.7%). Currently, there is no validated scoring tool for udder edema. Based on these findings it can be concluded that the caretakers in the study feel that udder edema has a negative impact on the overall wellbeing and productive ability of their cattle. A second study was conducted to investigate the hypothesis that udder edema causes discomfort to dairy cattle during the milking session. Increased sensitivity of the teats due to udder edema may cause cows to show discomfort-related behaviors, such as stepping and kicking. First and second lactation cows (n=376) on two large Colorado farms (>500 head) were observed in the milking parlor during the udder preparation and active milking phase. All cows were visually examined and scored for udder edema at the end of the milking session. The stepping and kicking behaviors were counted to possibly identify if, at any time during the udder preparation or milking phase, cows with edematous udders showed heightened levels of rear leg behavior. First lactation dairy cattle showed an increase in stepping behavior during the udder preparation phase as compared to first lactation cows without udder edema when the milker is physically manipulating the udder ($p = 0.0168$). Second lactation cows with udder edema had greater step rate than first lactation cows without udder edema during udder prep when the milker was in contact with the udder ($p = 0.0215$). First lactation cows with udder edema have greater frequency of kick behavior compared to second lactation cows with udder edema during the milking session ($p=0.0092$). First lactation cows with udder edema kicked off the milking unit more often compared to first lactation cows without udder edema ($p=0.0500$) and second lactation

cows with udder edema ($p=0.0017$). It was concluded that udder edema can influence a change in step and kick behavior in the milking parlor.

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DEDICATION

To all the farm kids who were seen as just that & the misfits who never can be put in a box - this work is for you. We see the world differently and it is that which makes us a powerful force to be reckoned with. Never stop discovering who you are. Let your passions guide you in this world. Be proud of everything that you are and look forward to who you will become.

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INTRODUCTION

The welfare of a dairy cow has a direct effect on her production ability (von Keyserlingk et al., 2009). Udder edema is common in Holstein dairy operations, with 66% of cows having udder edema at least once (Morrison et al., 2018. Recent research investigating the welfare of cows with udder edema is sparse. Validated measuring methods for accurately assessing the severity of udder edema are also lacking. This review will cover the information that is currently available. The occurrence of udder edema has increased between the late 1970s and late 1990s (Gröhn et al., 1989; Van Dorp et al., 1998; Morrison et al., 2018). Gröhn et al. (1989) found that 0.004% (214) of all cows sampled (61,124) were reported to have udder edema. Van Dorp et al. (1998) reported an incidence rate of 0.01% (1,954 cows) from herds reporting udder edema. Both of these studies were dependent on farms reporting udder edema, which can vary from the actual occurrence rate. Morrison et al. (2018) scored 912 Holstein cows in 3 herds and established prevalence of udder edema based on when edema was present in relation to calving. Edema prepartum affected 12.0% (109) of cows, edema postpartum affected 10.9% (99) of cows, and edema prepartum and on at least 1 occasion postpartum affected 47.5% (433) of cows in the study. Udder edema is the accumulation of lymphatic fluid in and around the interstitial spaces of the mammary gland (Al-Ani, 1984; Kojouri et al., 2015). Physiological edema is not the result of an infectious condition such as mastitis (Moroni et al., 2018). Nevertheless, dairy cattle with udder edema exhibit negative behaviors similar to those observed in mastitis cases, such as decreased lying time, frequent stepping in the parlor (Willis, 1983), and udders and teats that are sensitive to the touch (Vestweber and Al-Ani, 1983). Udder edema can have detrimental effects on the structural integrity of the udder and teats (Moroni et al., 2018), which then increases the risk of mastitis (Slettbakk et al., 1995; Ivemeyer et al., 2011; Morrison et al., 2018) and early culling (Gussmann et al., 2019). The Welfare Quality Assessment Protocol for Cattle (Welfare Quality Consortium, 2009) contains no assessment for evaluating the occurrence or severity of swollen udders. Emphasis is placed on injury to the udder and milk SCC. The dairy industry has focused on mastitis, and noninfectious udder disorders may have not received the attention that was needed. A vast majority of research on udder edema is at minimum 30 yr old, adding to the concern that this disorder has been neglected. A need for more research exists. These noninfectious problems have a great potential to impact both a dairy cow's health and wellness and a farm's profit. Edema negatively affects the longevity of the cow and milk production. Determination of the relationship between udder edema and measurable animal-based outcomes and health records will allow producers to promptly detect risk factors and mitigate the negative effects of edema. Some examples of measurable indicators that may be associated with udder edema include swelling severity, udder cleft dermatitis, milk production, early culling, and restless behavior during milking. Udder edema should be investigated as a possible emerging welfare issue in dairy cows.

OVERVIEW OF UDDER EDEMA

Udder edema is the result of lymphatic fluid accumulating in the interstitial space of the mammary gland and surrounding tissues (Al-Ani, 1984; Tucker et al., 1992; Kojouri et al., 2015; Morrison et al., 2018). In a study on 3 commercial Holstein dairy farms, 30% of the dairy cows never had udder edema, whereas 48% had edema both prepartum and postpartum. Furthermore, 12% had only prepartum edema, whereas 11% had only postpartum edema (Morrison et al., 2018). Parity is negatively associated with prevalence of edema (Morrison et al., 2018). During pregnancy the increase of fetal pressure in the pelvic area causes blood and lymphatic fluid circulation to be impaired, resulting in fluid buildup in the udder tissues (Al-Ani and Vestweber, 1986). This fluid leads to deterioration of the support structures in the udder (Dentine and McDaniel, 1984), misshapen quarters and teats, increased risk of udder cleft dermatitis, and increased risk of mastitis (Morrison et al., 2018). Tenderness of the udder and malformed teats disrupt milk letdown, increase difficulty of attaching the milking machine due to kicking, and negatively affect production potential throughout an animal's lifetime (Vestweber and Al-Ani, 1983; Melendez et al., 2006; Medrano-Galarza et al., 2012).

Designated as a metabolic disorder (Kojouri et al., 2015), physiologic udder edema has been associated with excessive salt in the diet, greater age at first calving (Malven et al., 1983), above-ideal BCS, genetic traits (Ruegg, 2015), physiological changes during maturation and udder development (Dyce and Wensing, 1971; Robbins and Cotran, 1979; Tyler and Ensminger, 2006), oxidative stress (Mueller et al., 1989), increased stress from pen movements when taking out or bringing in new cows to the group, overcrowding (Fustini et al., 2017), and heat stress (Tao and Dahl, 2013). Melendez et al. (2006) found that heifers calving in winter were 3.68 times more likely to develop udder edema than in summer. Sites of fluid accumulation include the brisket, udder, navel, and, in extreme cases, the legs and vulva (Tucker et al., 1992). Edema became less severe and decreased in prevalence as cows increased in parity (Emery et al., 1969; Hayes and Albright, 1976; Morrison et al., 2018). Nevertheless, the presentation of the disorder is the same, causing further damage to the suspensory ligaments and attachments (Vestweber and Al-Ani, 1983). Dairy cows with swollen udders from mastitis exhibit behaviors such as spending less total time lying, less time lying on the affected quarter or quarters, increased standing time, and increased stepping behavior (Siivonen et al., 2011). When milking was performed by the first author, cows with udder edema exhibited greater kicking behavior during milking. Physiological changes such as tenderness of the teats and swelling of udder tissues usually present as painful. Pain has been associated with mastitic udders (Fitzpatrick et al., 2013). The swollen udder tissues caused by edema have not been investigated for painfulness or as a welfare concern on dairy operations.

Pain is one of the pillars in all welfare assessments, along with health, productivity, and the ability to exhibit natural behaviors (von Keyserlingk et al., 2009). Dairy cows with udder edema need to be objectively evaluated for pain. The first author observed that the same cows with udder edema may have tender teats. Milking becomes an unpleasant experience for both human and cow, further resulting in potential injury, infection, or early culling, greatly affecting the profitability and wellbeing of that animal. The purpose of this review is to present the known factors contributing to the development of udder edema. The welfare of a dairy cow may be affected by udder edema.

PHYSIOLOGICAL CHANGES AND MAMMARY DEVELOPMENT

As a pregnant heifer transitions to the milking string, her body goes through substantial physiological changes. Although the mammary system is already in place, it develops significantly in the last stages of gestation. The animal's internal anatomy begins to change, and, physiologically, the demand of blood for mammary gland development is the precursor to udder edema in primiparous heifers. Mature dairy cows have a subcutaneous abdominal vein, also known as the milk vein, whereas, in heifers, this vein is not present. Young cattle have a cranial epigastric vein, which flows anterior to the mammary gland, and a caudal epigastric vein, which flows posterior of the mammary gland. In the latter stages of gestation, a heifer's cranial epigastric and caudal epigastric veins merge through anastomosis, creating the milk vein (Dyce and Wensing, 1971). Figure 1 in Allen et al. (2008) clearly demonstrates the immense vasculature of the mammary gland and how the caudal and cranial mammary veins come together and form the subcutaneous abdominal vein. The creation of the subcutaneous abdominal vein allows for an increase in the amount of blood flow from the mammary gland.

Two types of edema occur: generalized and local (Moroni et al., 2018). Generalized edema can be seen throughout the body. It is most prominent in the ventral areas, such as the barrel, limbs, and udder. Localized edema occurs because of venous stasis, defined as decreased blood flow in the veins due to an increase in capillary pressure, or because of impaired lymphatic drainage (Robbins and Cotran, 1979). Brisket disease (high-altitude disease) is a prime example of localized edema, where fluid collects in a specific anatomic site instead of involving the whole body (Hecht et al., 1962). Due to the increase of blood needed for mammary gland development, the blood flow will reverse in direction. It is exactly the opposite of the blood flow in a young heifer. With this increase in flow, the veins respond by increasing in size (Linzell, 1960). To distribute vital nutrients and oxygen throughout the body, the blood must travel throughout the body to provide proper oxygen, fluid, and nutrient levels to maintain tissues. Two forces that control fluid movement across blood supply membranes are hydrostatic pressure and osmotic pressure. Osmosis is the diffusion of water across a semipermeable membrane to balance out different concentrations of solutes (salt being a primary one) on the 2 sides of the membrane. Osmotic pressure prevents water from moving across the capillary membrane, whereas hydrostatic pressure pulls water across the membrane (Stillwell, 2013). Proteins in blood plasma draw in water from interstitial spaces. When functioning properly, capillaries bring in extravascular fluid to maintain interstitial fluid levels. Without the ability to drain these tissues due to pressure imbalance, fluid builds up in the interstitial tissues, resulting in udder edema. The same result can occur with the obstruction of lymphatic drainage, but this is more localized.

Another important variable in fluid movement is capillary permeability. Udder edema has a "snowball effect," whereby increased blood flow increases hydrostatic pressure (Tyler and Ensminger, 2006). Greater hydrostatic pressure increases capillary permeability, which then causes leaking of fluid into the interstitial tissues. With more blood protein now in the tissues, more fluid will be drawn to the tissue space. As fluid accumulates, the surrounding tissues become inflamed, which obstructs blood and lymph vessels, disrupting fluid movement into and out of the vessels (University of Idaho, 2018). Histamine, a compound released by cells to respond to inflammatory reactions, has a higher concentration in colostrum and plasma in cows with udder edema (Zarkower, 1967). Higher-than-normal histamine concentration in the udder tissue can lead to self-destructive behavior of cows licking the udder tissue and teats until raw, creating significant lesions that can lead to dermatosis (Eyre and Burka, 1978; Yeruham and Markusfeld, 1996). Another study found periparturient udder edema to be associated with increased risk of clinical mastitis (Slettbakk et al., 1995).

With increased blood flow, decreased fluid movement between the tissues and capillaries, and increased histamine levels, these physiological changes present a challenge for dairy farmers. The best approach is prevention before udder edema becomes severe. Due to the natural physiological, metabolic, and vascular functions throughout the last stage of gestation, the farmer needs to manage nutrition, genetic, and calving-in factors such as body condition and age at calving to provide the most optimal scenario to minimize udder edema. Suggestions such as massaging the udder during milking and the use of diuretics have been shown to have some effects of reducing edema (Alhadrami and Faye, 2016).

GENETIC PREDISPOSITION

Norman et al. (1974) estimated that the genetic correlation of milk yield and edema severity was 0.40. Van Dorp et al. (1998) state that high-yielding cows have increased genetic potential to develop udder edema, due to the substantial genetic correlation found in their study. Shanks et al. (1978) discovered that cows with high production pedigrees had an 11% higher incidence of edema compared with those of low production pedigrees. They also found that the most significant difference between levels of severity (mild, moderate, and severe) in udder edema cases was that high production pedigree cows showed greater edema above the rear udder. Need exists to develop validated scoring tools for accurately assessing udder edema. Additionally, 19% more daughters from high-production sires were diagnosed with udder edema compared with daughters from average sires (Shanks et al., 1978). Increased occurrence is also directly related to increased severity (1–5 score). For detailed score classifications, refer to Table 1 in Dentine and McDaniel (1984). Heritability for edema scores in first lactation was estimated at 0.13 by Dentine and McDaniel (1984). Recall that traits lower than 0.15 are considered to have low heritability (Cassell, 2009). Research on genetic influences and udder edema should be conducted with modern heifers and cows of all dairy breeds.

Malven et al. (1983) discovered that as calf birthweight increases edema decreases, but seasonal fluctuations in ambient temperature had no relation to edema. They had also found age at calving ($P < 0.01$) and increased gestation length ($P < 0.01$) to be significantly correlated with presentation of udder edema. Shanks et al. (1978) found no direct effect between age at calving and edema. Research used in support of a direct association between age at calving and increased occurrence of udder edema include Hays and Albright (1966), Malven et al. (1983), Dentine and McDaniel (1983), and Gröhn et al. (1989). The previously stated studies all had a similar conclusion that age at calving has an association with udder edema. Additionally, Nestor et al. (1988) found that primiparous Jersey cows, scored on a scale from 1 to 5, presented with greater scores of udder edema (3.70) compared with Holstein cows (3.30). It is speculated that edema only appears to be more severe on smaller-framed cows as opposed to taller, wider-framed cows (Wautlet et al., 1990). Melendez et al. (2006) found that for each additional 10 cm of height at calving, the odds of udder edema increased by 23%. Much of this research dates at least 30 yr from our current time. Therefore, this area must be further investigated for updated information and findings.

Conformation traits of the udder are another area of selection that producers focus on for longevity of the animal and herd uniformity. According to Kuczaj and Blicharski (2008), weak udder ligaments as well as udder and nipple defects were most often observed in cows descended from American bulls (6.2%). This may be due to the trend of breeding for high production traits in American dairy operations and sire studs. Udder edema causes deterioration of udder support structures, creating low-hanging udders (Dentine and McDaniel, 1984). Lawstuen et al. (1988, p. 796–797) discovered that “The largest phenotypic correlations were for edema and udder depth

(0.20) and edema and udder support (0.17). Congestion of udders was phenotypically related to deep, low-hanging udders that lacked cleavage. Also, cows with deep udders were more susceptible to mastitis (0.16)." Kuczaj and Blicharski (2008) concluded that the most common reasons behind forced culling are unfavorable characteristics, including body conformation, udder build–udder support, rump setting–ease of calving, and udder and health status of the animal, specifically fertility and udder and limb disorders. Lawstuen et al. (1988) state that both edema and calving ease had significant and positive estimates of genetic correlation with stature (0.42 and 0.61) and strength, which is how well the body shape supports production and longevity (0.28 and 0.85). It must be noted that genetic correlation, how 2 traits influence expression of one another, differs from genetic heritability, the likelihood that a particular trait can be expected to be passed on to the next generation. The reduction of udder edema, with its clear influence on udder support systems and udder health, could potentially have a substantial effect on increasing the longevity of an animal. It may also help reduce early culling due to udder disorders.

PERIPARTURIENT NUTRITION

The transition period for a dairy cow is a time where the cow's endocrine system prepares the body for parturition and lactogenesis. As a dairy cow shifts from late gestation to early lactation, growth hormone increases and plasma insulin decreases. Acute surges of both hormones occur in plasma concentration at parturition (Kunz et al., 1985). These changes in the endocrine system in conjunction with decreased DMI affect metabolism, leading to mobilization of fat from adipose tissue and glycogen from the liver. The same endocrine system changes and DMI decrease occur in dairy heifers nearing parturition (Rabelo et al., 2003). Hayirli et al. (2003) found that heifer DMI decreased from 1.70% of BW at 3 to 1 wk before calving to 1.23% BW in the last week. This is significantly different from multiparous cattle in the same stage of lactation, whose DMI decreased from 2.06% of BW at 3 to 1 wk before calving to 1.36% BW in the last week. Additionally, primiparous cows but not mature cows, with an increased CP requirement of 12.7 to 14.7%, have shown improved lactation performance. Silva et al. (2002) found that heifers had reduced mammary development due to rapid BW gain when fed a high-energy diet. Low DMI during the last week of gestation caused impairment of the liver, resulting in decreased lipid and lipoprotein concentrations. This has been suspected as a causative factor in udder edema. Kojouri et al. (2015) found that serum concentrations of total proteins, triglycerides, cholesterol, and lipoproteins were lower in cows with udder edema.

Randall et al. (1974) established that reduction of severity of udder edema can be achieved through salt restriction. Severity was rated on a 5-point scale developed by the researchers. Moore et al. (2000) found that heifers respond differently than mature cows to anionic salts in the feed. Different forms of anionic salts include sodium chloride, magnesium sulfate, calcium sulfate, ammonium sulfate, calcium chloride, ammonium chloride, and magnesium chloride. Supplementing with anionic salts into the diet is the only way to achieve a negative DCAD. This is essential to provide the proper dietary needs of a dairy cow. Potassium chloride, which is used as a replacement for sodium chloride, results in the same severity of udder edema as cows supplemented with sodium chloride (Randall et al., 1974). Hutjens (1980) reported that feeding NaCl (227 g), KCl (227 g), or both NaCl and KCl (454 g), each day increased the severity of udder edema over that of control cows and heifers fed no NaCl or KCl. Nestor et al. (1988) stated that cows supplemented with 136 g of NaCl, 136 g of NaCl and 272 g of KHCO3, or 23 g of NaCl and 272 g of KHCO3, compared with 23 g of NaCl with 0g of KHCO3 (control), resulted in first-calf heifers having significantly more severe udder edema than control cows (3.55, 3.20, 3.51, 3.11). Severity was measured using a pitting chart developed by Swett et al. (1938). Cows supplemented with these salts also developed edema sooner, and the edema took more time to clear from the udders. Table 1 in Nestor et al. (1988) has a detailed description of the time points at which edema was scored. Lema et al. (1992) found that feeding diets containing $CaCl_2$ to cows during early lactation may extend the beneficial effects of $CaCl_2$ in moderating edema into the early-lactation interval, due to feeding $CaCl_2$ decreasing severity of edema prepartum. Severity was scored on a 10-point scale developed by Tucker et al. (1992). A dietary program adjusting for changes in DMI and protein needs, and including anionic salts, provides complete nutritional requirements for cows during the transition period to decrease the prevalence and severity of udder edema.

OXIDATIVE STRESS

Oxidative stress occurs when oxygen is not reduced to water during metabolic processes. Partially reduced oxygen becomes a free-radical superoxide (O_2^-; Levine and Kidd, 1985). Increased metabolic rate, rapid growth, high milk production, or extreme aflatoxin exposure could elevate superoxide creation. Superoxide is reduced to hydrogen peroxide (H_2O_2). Both of these occur naturally during the metabolic process and are not harmful when broken down properly. Damage from these is suspected to be the result of converting into more free radicals and the catalytic transition elements, such as iron, which have harmful effects (Gutteridge and Halliwell, 1994). Release of catalytic Fe becomes more likely under conditions of dietary imbalance, trauma, or stress, often accompanying calving (Madsen, 1990). The body has a normal antioxidant capacity, which is responsible for controlling oxygen radicals throughout the body. Enzymes such as catalase, peroxidase, and superoxide dismutase are involved in free-radical scavenging.

Nonenzymatic antioxidants (tocopherols, ascorbic acid, and glutathione) also occur within the body. Exogenous antioxidant molecules, which enter the body through the diet, include vitamin E, vitamin C, carotenoids, and flavonoids (Diplock et al., 1998). When in oxidative stress, reactive oxygen species surpass the defense capability of the antioxidants. Without the control of these radicals by antioxidants and enzymes, free radicals inflict damage on DNA, proteins, and lipids involved in fundamental metabolic processes (Gagné, 2014). These damaged cellular components result in cytotoxicity, genotoxicity, and carcinogenesis when damaged cells proliferate (Gutteridge and Halliwell, 1994). This is a major concern for the stability and functionality of the entire body. Oxidative stress suppresses production of androgens and estrogens, impairing reproduction mechanisms, increasing the occurrence and severity of milk fever, increasing the amount of placental retention, and increasing sodium and water retention, leading to udder edema (Miller et al., 1993). Providing a diet with adequate exogenous antioxidant molecules and amino acids counteracts this disorder (Hou et al., 2015).

Mueller et al. (1989) presented within an abstract the effectiveness of vitamin E, which, as mentioned before, is an antioxidant, in reducing severity of udder edema. It was shown that heifers supplemented with vitamin E (Mueller et al., 1989) and magnesium (Kelly et al., 1990) have less-severe edema. Additionally, Miller et al. (1993) established that daily provision of 1,000 IU of vitamin E paired with a diet containing at least 0.12 ppm of selenium reduced the severity of udder edema in primiparous cows. In diets having less than 0.06 ppm of selenium, udder edema was not reduced when vitamin E was supplemented (Mueller et al., 1989; Miller et al., 1993). This emphasizes the importance that if 1 or more vitamins or minerals are scarce within the ration, the total system remains impaired. During oxidative stress, cows also have decreased immunity, raising the risk of infections such as mastitis. Eliminating oxidative stress reduces the risk of periparturient disorders, reproductive failure, mastitis, and udder edema. This approach is beneficial for longevity of the udder and the animal as a whole.

UDDER EDEMA AND UDDER CLEFT DERMATITIS

Udder edema is associated with mastitis and udder cleft dermatitis (Beattie and Taylor, 2000). Both diseases can cause detrimental effects to the animal's welfare, milk production ability, and longevity on the dairy operation. Udder cleft dermatitis (**UCD**), also known as udder rot or necrotic dermatitis, is a skin lesion that appears in areas of tightly adjacent skin. Such areas include the udder cleft along with the medial (inner) aspect of the thigh and the lateral (outer) aspect of the udder (Ruegg, 2015). Friction between these 2 spaces due to a swollen udder or poor confirmation can lead to chafing, dermatitis, and, if persistent enough, necrosis of the tissues. Persson Waller et al. (2014, p. 310) reported that good udder confirmation with "a strong anterior udder attachment was a protective factor." These udder sores present with pus, thickened and inflamed skin, crust, and wounds that easily bleed. A putrid odor has also been noted in advanced cases. Along with udder edema, other risk factors associated with udder cleft dermatitis on a herd level include high yearly milk production, a mean SCC > 200,000 cells/mL, and an existing prevalence of digital dermatitis (Persson Waller et al., 2014). On an individual cow basis, increasing parity is linked to greater risk of UCD, along with breed, milk production, and previous clinical mastitis cases. Persson Waller et al. (2014) found that cows with UCD had a 3.3× greater risk for clinical mastitis. It is also suggested that skin lesions, hock lesions, and udder lesions can be a source for *Staphylococcus aureus* (Capurro et al., 2010). This was the most isolated pathogen by Naqvi et al. (2018) when surveying which pathogens are most prominent in intramammary infections in dairy heifers. Santos et al. (2004) discovered that a reduction in udder edema is linked to a decrease in SCC. Additional studies have shown that udder edema is a risk factor for mastitis (Gröhn et al., 1989; Slettbakk et al., 1995; Waage et al., 2001), which is the most common udder disease in the dairy industry. Slettbakk et al. (1995) and Waage et al. (2001) found cows with teat and udder edema to have an increased risk of clinical mastitis. Waage et al. (2001) also found that udder edema at calving was positively correlated with teat edema, milk leakage 1 wk prepartum, milk leakage at calving, blood in milk at calving, and skin lesions between udder and thigh, which leads to UCD. Slettbakk et al. (1995) state that proposed reasoning for why edema is a risk factor for mastitis is due to edematous udders being more prone to injury and because these udders have impaired blood circulation. Gröhn et al. (1989, p. 1883) also found that "Mastitis diagnoses increased the odds of concurrent or subsequent diagnosis of udder edema, disorder of the abomasum, ketosis, and nonparturient paresis." Melendez et al. (2006) reported that milk yield at the first DHIA test day was 3.6 kg lower in cows with udder edema. Heise et al. (2016, p. 1253) stated, "Longevity of dairy cows is an economically important trait for farmers and has gained in importance as a global indicator for animal welfare." Heise et al. (2016) further stated, "Longevity results from survival of sequential time periods." Pfeiffer et al. (2015) and Neerhof et al. (2000) found that clinical mastitis does have a negative effect on the longevity of an animal, resulting in early culling. No research has been conducted on the direct impact udder edema has on farm economics. Reduction of udder edema, UCD, and mastitis will not only affect milk production, quality of product, and revenue, they will also increase cow's welfare and longevity potential on the dairy operation.

CONCLUSIONS

This review addressed multiple factors that contribute to udder edema and secondary risk factors associated with udder edema. Scientifically validated udder edema scoring tools need to be developed. We need to acknowledge the potential of udder edema becoming an emerging welfare issue. Diseases of the udder greatly affect the health and wellness of the cow, the quality and quantity of milk being produced, the condition of the udder ligaments, and the longevity of the animal. Impaired lymphatic drainage and blood circulation results in inflamed tissues and tender teats. Improper or lack of supplementation of protein, anionic salts, and minerals increases the risk for udder edema. Udder edema is also associated with udder cleft dermatitis and increased risk of mastitis. Dermatological lesions on the udder and mammary gland infections are likely to compromise animal welfare. Udder edema can be reduced on a short-term basis by adjusting nutrition and on a long-term basis possibly by shifting genetic selection parameters. The area of udder edema has vast potential for research. New and updated research is needed on udder edema because indicators suggest that it may be a welfare issue. The second reason for new research is that most cited material is old and needs updating, including current prevalence of udder edema. Research is needed to fully evaluate the prevalence of this disorder across the modern dairy industry.

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CHAPTER 2: THE PERSPECTIVES OF DAIRY CARETAKERS ON UDDER EDEMA

1. INTRODUCTION

Udder edema is a non-infectious, metabolic disorder that occurs in dairy cattle (Al-Ani, 1984, Tucker et al., 1992, Kojouri et al., 2015, Morrison et al., 2018). This disorder can negatively impact milk production (Melendez et al., 2006), increase the risk for secondary conditions, such as mastitis (Morrison et al., 2018, Nitz et al., 2020), and deteriorate the support structures of the udder (Dentine and McDaniel, 1984). Trends of udder edema prevalence within the United States have not been captured over any period of time. Likewise, an industry-wide validated scoring tool has not been established to monitor and subsequently manage udder edema on dairy facilities. Prior to addressing how dairy caretakers can observe and record udder edema, an assessment of the industry's awareness of udder edema is necessary.

Perspectives of dairy caretakers have been reported in a number of areas, including lameness (Richert et al., 2013), antibiotic use (Fischer et al., 2019), calf welfare (Sumner and von Keyserlingk, 2018) and on-farm euthanasia (Román-Muñiz, et al., 2021). There have been no studies published examining caretaker's or veterinarian's perspectives on udder edema. Caretakers are involved in the daily assessment of an animal's health and wellbeing. Being unaware of the disorder or lack of understanding of it may prevent caretakers from monitoring udder edema. Additionally, the lack of an industry-accepted and validated scoring tool may further inhibit the monitoring of udder edema. The goal of this study was to evaluate current perspectives of dairy caretakers about udder edema, caretaker's knowledge about factors that influence the development of udder edema, and caretaker monitoring practices.

2. METHODS AND MATERIALS

The study materials and research plan were approved through the Colorado State University Institutional Review Board (20-10161H) prior to project initiation.

2.1 STUDY POPULATION AND RECRUITMENT

The population of interest was dairy animal caretakers, including but not limited to farm owners, herdspeople, farm managers, and herd managers. A herd manager was described as an individual that is only involved in the management of the cattle herd while a farm manager was defined as being responsible for the care of the cattle herd along with being involved in crop production for the farm. Recruitment was conducted through an advertisement, produced by the Progressive Dairy magazine (26,000 subscribers), to participate in an online survey. This advertisement was in the magazine from October 2020 through December 2020. Additionally, The Central Plains Dairy Exposition (1,550 subscribers) included an invitation to participate in the survey in their November 2020 and December 2020 electronic newsletter. No monetary incentive was offered to participants. All responses were anonymous and no identifying information was collected from survey participants. The only forced response question in the survey was the participation consent; all other questions were optional.

2.2 SURVEY DEVELOPMENT AND CONTENT

A twenty-three-question anonymous survey was designed to gain insight on the perspectives of caretakers of udder edema in the dairy industry in the United States. The survey was intended to take less than eight minutes to complete. A variety of question types were utilized in the survey, including Likert Scale, multiple choice, dichotomous, and short answer. The categories of questions included: respondent demographics, farm information and perception statements. The first block of questions in the survey aimed to identify current monitoring practices of udder edema on dairy farms. The second block of questions assessed the perspectives of caretakers regarding udder edema. Perception statements were presented in the form of a Likert scale (strongly disagree – strongly agree) and focused on the respondent's knowledge about udder edema. Statements focused on the risk factors that affect the prevelance and severity of udder edema alongwith udder edema's relationship to other production concerns. Students from the Colorado State University Animal Sciences Department tested the survey for clarity and functionality. The survey was translated into Spanish and tested by native speakers familiar with dairy production terminology. The survey was administered through Qualtrics software (Qualtrics, Provo, UT, USA) and was accessible by either a link or QR code.

2.3 STATISTICAL ANALYSIS

All survey responses were exported from Qualtrics and stored in Excel 2008 (Microsoft Corporation, Redmond, WA). The data were analyzed in version 4.1.1 of R (R Development Core Team, 2021). Summary statistics were completed on the reported demographic information and Likert scale questions. Data was categorized by gender or by caretaker position on farm.

3. RESULTS

A total of sixty-three respondents accessed and filled out at least part of the survey. Thirty-two responses with less than 100% completion were excluded from the analysis. Additionally, one response was excluded as the participant did not reside within the United States, resulting in a total of thirty complete responses. Demographics of the respondents and the farms they represent can be seen in Table 2.1. Sixteen male and fourteen female respondents participated in the survey.

Occupation was categorized into four main categories: Farm manager (n=1), Herd Manager (n=6), Owner (n= 20), and other (n=3). Others included a student, a part-time employee, and an assistant herdsman. All five regions of the United States were represented (West, Southwest, Midwest, Northeast, Southeast). Mean herd size was 526 cows, based on data from twenty-nine of the respondents who reported herd size. Average daily milk production for first lactation cows was 27.76kg and 32.57kg for second plus lactation cows. All completed questionnaires were in English, therefore translation of responses was not necessary. Responses to the perspective questions using a Likert scale can be seen in Table 2.2A and 2.2B.

Table 2.1 Demographics, Herd Size, And Production Levels Per Lactation Group Reported by Survey Respondents

	Overall (n=30)
Position	
Owner	20 (66.7%)
Herd Manager	6 (20.0%)
Farm Manager	1 (3.3%)
Other (specify)	3 (10.0%)
Region in the U.S.*	
Midwest	14 (46.7%)
Northeast	7 (23.3%)
Southeast	2 (6.7%)
Southwest	2 (6.7%)
West	5 (16.7%)
Gender of Participant	
Man	16 (53.3%)
Woman	14 (46.7%)
Milking Herd Size	
Mean (SD)	526 (940)
Median [Min,Max]	150 [20.0, 5000]
No Response	1 (3.3%)
Avg. Pounds Production in 1st Lactation Cows	
Mean (SD)	61.2 (12.3)
Median [Min,Max]	61.5 [39.0, 90.0]
No Response	4 (13.3%)
Avg. Pounds Production in 2nd+ Lactation Cows	
Mean (SD)	71.8 (13.7)
Median [Min,Max]	75.0 [45.0, 100]
No Response	5 (16.7%)

* Midwest: ND, SD, IA, NE, KS, MO, MN, WI, MI, OH, IN, IL, Northeast: PA, DC, MD, DE, NJ, CT, RI, MA, ME, NH, VT, NY, Southeast: WV, NA, KY, TN, NC, SC, GA, AL, MS, AR, LA, FL, Southwest: AZ, NM, TX, OK, West: WA, OR, ID, CA, MT, CO, UT, WY, NV

Table 2.2a Likert Question Responses

	Overall (n=30)
Udder edema is a part of fresh heifer udder health	
Strongly Disagree	1 (3.3%)
Somewhat Disagree	0 (0%)
Neither Agree nor Disagree	5 (16.7%)
Somewhat Agree	10 (33.3%)
Strongly Agree	14 (46.7%)
Training animal caretakers to identify udder edema is important	
Strongly Disagree	0 (0%)
Somewhat Disagree	0 (0%)
Neither Agree nor Disagree	2 (6.7%)
Somewhat Agree	13 (43.3%)
Strongly Agree	15 (50.0%)
An industry-standard rating scale would be useful to monitor udder edema	
Strongly Disagree	0 (0%)
Somewhat Disagree	0 (0%)
Neither Agree nor Disagree	9 (30.0%)
Somewhat Agree	11 (36.7%)
Strongly Agree	10 (33.3%)
Udder edema is common on your farm	
Strongly Disagree	2 (6.7%)
Somewhat Disagree	4 (13.3%)
Neither Agree nor Disagree	2 (6.7%)
Somewhat Agree	20 (66.7%)
Strongly Agree	2 (6.7%)
Udder edema is seen more in heifers versus cows	
Strongly Disagree	0 (0%)
Somewhat Disagree	0 (0%)
Neither Agree nor Disagree	2 (6.7%)
Somewhat Agree	11 (36.7%)
Strongly Agree	17 (56.7%)
Udder edema can be managed through nutrition	
Strongly Disagree	0 (0%)
Somewhat Disagree	1 (3.3%)
Neither Agree nor Disagree	7 (23.3%)
Somewhat Agree	7 (23.3%)
Strongly Agree	15 (50.0%)

Table 2.2b Likert Question Responses

	Overall (n=30)
Udder edema negatively affects milk production	
Strongly Disagree	0 (0%)
Somewhat Disagree	4 (13.3%)
Neither Agree nor Disagree	2 (6.7%)
Somewhat Agree	14 (46.7%)
Strongly Agree	10 (33.3%)
UE affects the attachment of the milking unit	
Strongly Disagree	0 (0%)
Somewhat Disagree	1 (3.3%)
Neither Agree nor Disagree	2 (6.7%)
Somewhat Agree	6 (20.0%)
Strongly Agree	21 (70.0%)
High genetic milk production potential increases the risk of udder edema	
Strongly Disagree	0 (0%)
Somewhat Disagree	0 (0%)
Neither Agree nor Disagree	9 (30.0%)
Somewhat Agree	11 (36.7%)
Strongly Agree	10 (33.3%)
Udder edema is a risk factor for mastitis	
Strongly Disagree	0 (0%)
Somewhat Disagree	4 (13.3%)
Neither Agree nor Disagree	4 (13.3%)
Somewhat Agree	10 (33.3%)
Strongly Agree	12 (40.0%)
Udder edema is a welfare issue	
Strongly Disagree	0 (0%)
Somewhat Disagree	0 (0%)
Neither Agree nor Disagree	4 (13.3%)
Somewhat Agree	18 (60.0%)
Strongly Agree	8 (26.7%)
Udder edema is painful	
Strongly Disagree	0 (0%)
Somewhat Disagree	0 (0%)
Neither Agree nor Disagree	3 (10.0%)
Somewhat Agree	13 (43.3%)
Strongly Agree	14 (46.7%)

3.1 UDDER EDEMA MONITORING

When investigating the udder edema monitoring practices of the respondents, two caretakers (6.7%) stated that they monitor udder edema on their operations. Both respondents said that their protocols are to visually assess the cows for swelling in the udder. One of the two respondents also stated they observe cows for discomfort. This is quite different of a response when examining how many caretakers indicate that udder edema is common on their farm (n=22). Additionally, 93.3% (n=28) of caretakers agreed that udder edema appears more often in heifers than in cows. The majority of respondents (n=28) agreed that that training caretakers to identify udder edema is important. Interestingly though, only twenty-one respondents stated an industry-standard rating scale would be useful to monitor udder edema.

3.2 PERCEPTIONS ABOUT KNOWN FACTORS INFLUENCING UDDER EDEMA PRESENTATION AND SEVERITY

Of the respondents, 73.3% (n=22) were in agreement that udder edema can be mananged through nutrition. Fourteen respondents agreed and eleven respondents neither agreed nor disagreed that high milk production potential does increase the risk of developing udder edema.

3.3 PERCEPTIONS ABOUT KNOWN EFFECTS UDDER EDEMA HAS ON COW HEALTH AND PRODUCTIVITY

There was common agreement among respondents that udder edema negatively affects milk production (n=24). Twenty-one respondents stated they strongly agree that swollen teats due to edema causes difficulty with attaching the milking unit and six respondents selected somewhat agree. Interestingly, 13.3% (n=4) of respondents somewhat disagreed that udder edema increases the risk of mastitis while 73.3% of (n=22) respondents somewhat or strongly agreed with the statement. The subject of pain and welfare in regard to udder edema was presented in the survey. Twenty-seven respondents indicated that udder edema is painful. Twenty-six respondents agreed that udder edema is a welfare issue.

4. DISCUSSION

The prevalence of udder edema is not known in the dairy industry nor have any trends been established for the disorder. Dentine & McDaniel (1983) found that 97% of cows had some level of edema around the time of parturition. More recently, Morrison et al. (2018) found in Canada that 70% of cows within that study population had edema. The lack of a prevalence trend is also hindered by not having a validated scoring system for udder edema. Only two respondents within this study stated that udder edema is monitored on their operations, which brings to light the uncommonness of monitoring for udder edema. It can be postulated that training caretakers to recognize udder edema, which twenty-eight respondents in this study agreed was important, may have multiple positive impacts on farm, particularly improved cow health and wellbeing, parlor efficiency, and the safety of the employees while in the milking parlor. Likewise, different treatment protocols could be developed based on the severity of udder edema and development of udder edema can be tracked through the late gestation and early lactation phases. Cows that develop udder edema are at greater risk for teat and udder injury along with diseases such as mastitis and udder cleft dermatitis (Al-Ani & Vestweber, 1986, Sletbakk et al., 1995, Beattie & Taylor, 2000). In this study, twenty-two respondents felt similarly to what has been found within research that cows with udder edema are at greater risk for developing chronic mastitis (Grohn et al., 1989, Slettbakk et al., 1995, Waage et al., 2001, Melendez et al., 2006). Furthermore, udder edema causes deterioration of the support structure in the udder (Dentine and McDaniel, 1984). Low-hanging udders are at an increased risk for mastitis, as found by Lawstuen et al. (1988). Early detection and continued monitoring of udder edema could prevent injuries and secondary diseases from developing, resulting in improved cow health and wellbeing.

First lactation cows are more reactive in the parlor due to unfamiliarity of the parlor and milking procedure (Sutherland and Huddart, 2012). It can be theorized that edema may cause even greater reactivity due to added discomfort from the udder swelling and disturbed milk ejection. It was noted by one of the caretakers that monitor for udder edema also observe cows for discomfort. More than three-quarters of the respondents agreed that udder edema is a welfare issue and is a painful disorder. With the knowledge that first lactation cows are more reactive in the parlor, caretakers can take the time to properly stimulate first lactation cow's udders and be more attentive to milk letdown to promote a positive milking experience during the transition period. Furthermore, being aware of possible increased reactivity in edematous cows due to discomfort can help caretakers be more conscious of cow behavior in the parlor to prevent personal injury from being kicked.

As a result of researching udder edema over decades, multiple scales have been developed to rate severity. The first noted visual tool was documented by Swett et al. (1938) using the pitting technique to confirm if an animal had udder edema. Throughout 1958 to 1983, additional scales were developed for studies investigating suspected factors involved with the development and severity of udder edema (Greenhalgh & Gardner 1958, Schmidt & Schultz 1958, Emery et al. 1968, Randall et al. 1973, Shanks et al. 1978, Dentine & McDaniel 1983). These scales varied from three, four, and five-point scales. Descriptive text identifying the differences between levels of severity lacked in all of these articles. Nestor et al. (1987) established a five point scale (1-5) for rating edema. Each level stated where edema was present on the body and how diffuse the edema was in that specified area. A 10-point scale was developed by Tucker et al. (1992) with similar definitions including diagrams to aid observers in delineating between scores. Morrison et al. (2018) created a four point scale (0-3) with descriptive text for each level and provided photos of animal that correspond with each level. These scales, while having been validated within the studies, do require validation for on-farm use by caretakers to assess the quality and practical application of the scale before the scale can be implemented. This is important to note as 70% of respondents agreed that an industry-standard rating scale would be useful to monitor udder edema. The next step would be to use and test various rating scales on farms for their ease of use and repeatability.

As the exact etiology of udder edema is still unknown, a number of studies have established that one method for managing udder edema is by providing heifers and cows with diets with adequate nutritional levels for their stage of development, stage of lactation, and intake levels. The responses of twenty-two caretakers within this study matched what is found within scientific literature, where the presence and severity of udder edema can be managed through the diet. A negative dietary cation anion difference (DCAD) is essential for multiparous cows less than 30 days away from calving. Anionic salts are the primary tool in creating a negative DCAD to prevent hypocalcemia. Non-lactating cows turnover ~10g of Ca daily, whereas lactating cows turnover ≥30g of Ca daily (Horst et al. 1997). Anionic salts cause blood pH to decrease, inducing metabolic acidosis. This triggers calcium to be released from the bones and increased absorption of calcium by the gastrointestinal tract, raising serum calcium levels in preparation for partuition and lactogenesis. Excessive levels of sodium and potassium have been associated with the development of udder edema (Randall et al., 1973). Morrison et al. (2018) found a conditional association between udder edema and ketosis, where cows with at least one occasion of udder edema in the transition period were at greater likelihood to develop ketosis 2wk postpartum. Late-gestation heifers do not experience hypocalcemia in the transition period, therefore not requiring anionic salts in the diet (Moore et al., 2000). Interestingly, Kojouri et al. (2015) found that heifers with udder edema had metabolic indicators of liver function impairment, paired with low DMI and low serum protein.

In one study, cows that developed udder edema had a decreased milk yield of 3.6 kg/day in comparison to those who did not develop udder edema (Melendez et al., 2006). The caretakers within this study responded in alignment with findings in research, where 80% of caretakers agreed that udder edema negatively affects milk production. Decreasing the number of udder edema cases could prove financially beneficial for farmers, especially if udder edema is prevalent on their operations. According to twenty-two caretakers, udder edema is common on their operations. Cows who developed udder edema are at greater risk for udder edema to reoccur in future lactations (Melendez et al., 2006). Milk production potential has average heritability (0.30) and udder edema has low heritability (0.14) but, the genetic correlation of milk yield and udder edema is 0.40. As average individual milk production has steadily increased from the 1960's, the prevalence of udder edema may have unknowingly increased alongside increased milk yields. Van Dorp et al. (1998) found that phenotypic correlations between milk yield and udder edema are 0.09. Melendez et al. (2006) conjectured that environmental influences such as diet and management could help counteract the negative influences of selecting for milk yield when controlling udder edema. Within the results of the survey, it is shown that 70% of caretakers are aware that cows with a high genetic milk production potential have a higher likelihood of developing udder edema. Investigating the impact environmental and heritable influences has on the presentation and severity of udder edema could provide valuable information on how to manage the disorder. When examining other potential implications udder edema has on the production ability of a dairy cow, teat edema was noted in Melendez et al. (2006). Due to the malformation of the teats, attaching the milking claw has been documented to be difficult with cows presenting with udder edema (Al-Ani and Vestweber, 1984), and was supported by 90% of the respondents in this study. Difficulty of attaching the milking unit due to teat edema could directly affect parlor efficiency because these cows require more labor to milk. This is an area to investigate in further research.

Udder edema and other transition diseases have interactions. Udder edema has been identified as potential precursor for clinical mastitis (Grohn et al., 1989, Slettbakk et al., 1995, Waage et al., 2001, Melendez et al., 2006). Grohn et al. (1989) investigated twenty-three veterinary diagnoses and their relationship to acute and chronic mastitis and teat injury in the records of 41,989 multiparous Finnish Ayrshire dairy cattle and found that udder edema was a risk factor for acute and chronic mastitis. Additionally, retained placenta, teat injury and acute and chronic mastitis were associated with udder edema. Slettbakk et al. (1995) collected data direct from farmers on 565 matched pairs of first and second lactation Norwegian dairy cattle. It was found that periparturient udder edema in both lactations is a risk factor for clinical mastitis ($p = 0.01$). The involvement of first lactation cows within the previously mentioned study is important to note as first lactation cows develop udder edema more commonly and more severely (Dentine and McDaniel, 1983). Twenty-two caretakers agreed that cows with udder edema do have an increased risk for udder edema. These findings raise the importance of managing udder edema to prevent secondary diseases that decreases a cow's welfare and milk production.

Dentine and McDaniel (1983), Kojouri et al. (2015), and Morrison et al. (2018) had similar findings in that udder edema was more prevalent within first lactation cows. This was strongly supported by the caretaker responses, where 93% agreed that udder edema is more common in heifers than in cows. Within the survey, there is a limitation with the statement, "Udder edema is common on your farm." Caretakers may be biased and blind to the actual prevalence of udder edema on their operations. Without accurate monitoring and recording of cases, these responses cannot be confirmed. Additionally, it is not known what the average amount of udder edema is on farms. Morrison et al. (2018) found on three commercial farms in Canada that 48% (n=646) of cows within the study's population presented with edema prepartum and at least on one occasion postpartum. Only 30% (n=403) of the sample population did not develop udder edema either prepartum or postpartum. As stated previously, industry-wide prevalence trends of udder edema have not been established and so, research is needed to determine how wide-spread this disorder is within the industry.

One limitation of this study is the total number of complete responses. During the recruitment phase, the survey was advertised as a questionnaire about udder edema monitoring and management. This study was conducted during the COVID-19 pandemic; therefore, in-person recruitment was not possible. A second limitation to the study is the bias that respondents may have with udder edema. Respondents had to have been familiar with the topic and have a vested interest in udder edema to have the desire to respond to this survey. These respondents are possibly more aware to udder edema's presence on their operations and the impact it has on the cows, which can skew the results of the study. Due to this, along with the small number of responses, the findings cannot be concluded as representative of the United States dairy industry.

5. CONCLUSIONS

Respondents are aware of udder edema on their farms and the negative effects that udder edema has on their cow's production and health. Caretakers stated that training employees to recognize udder is important and that they have an interest in an industry-wide scale to monitor udder edema on farm. Likewise, a majority of caretakers indicated that udder edema is painful and a welfare concern. A larger sample population would provide further insight into industry-wide perceptions of dairy caretakers on udder edema.

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CHAPTER 3: THE EFFECTS OF UDDER EDEMA ON PARLOR BEHAVIOR IN FIRST AND SECOND LA CTATION DAIRY COWS

1. INTRODUCTION

Udder edema (UE) is a metabolic disorder where lymphatic fluid is trapped in the tissues of the udder around the time of parturition (Bacic et al., 2007). Udder edema has been found to negatively impact milk production (Melendez et al.,2006), increase the risk for udder and teat injuries, chronic mastitis (Bacic et al., 2007), and udder cleft dermatitis (Beattie and Taylor, 2000), and permanently damage mammary tissues (Dentine and McDaniel, 1984). Melendez et al. (2006) theorized that udder edema causes discomfort to cows. It has been documented that edematous udders are more difficult to attach a milking unit because of swollen and malformed teats (Melendez et al., 2006). When examining first lactation cows, edematous udders are a common occurrence in United States dairy cattle (Dentine and McDaniel, 1984). Morrison et al. (2018) also had similar findings on Canadian dairy farms. Furthermore, cows who experience udder edema in their first lactation are at greater risk for its recurrence in future lactations (Melendez et al., 2006). Udder edema is shown to have a negative impact on the health and productive ability of dairy cows and poses a risk for future complications, but it is not known if udder edema affects a cow's behavior while she is in the milking parlor. This could indicate that a cow's welfare may be negatively impacted due to the persistent swelling of the udder tissues pre- and postpartum. Behaviors such as tail flicking, stepping, kicking, shifting, and flinching have been used to indicate if a cow is experiencing pain or discomfort during milking (Willis, 1983, Van Reenan et al., 2002, Sutherland & Huddart, 2012, Doyle and Morran, 2015). Discomfort is defined as an unpleasant feeling, either physical or psychological, that is responded with avoidance behavior (Ashkenazy and DeKeyser Ganz, 2019). Animal pain has been defined by Moloney and Kent

(1997) as a negative physical sensation paired with emotion that reacts to a stimulus to avoid tissue damage or the chance of tissue damage, decrease the risk of damage occurring again, and to promote recovery. By using behavioral observations, caretakers can learn if a management procedure or malady is causing the animal pain or discomfort.

Physiological applications of pain assessment can be sensitive to stress and may not be practical for on-farm use (Molony and Kent, 1997). These limitations are important to note because first lactation cows, when transitioning to the milking herd, experience multiple stressors. The transition period for a first lactation cow includes greater levels of handling, moving into the milking herd, a change in environment, and new sensations during the milking routine. First lactation dairy cows, without prior familiarization to the parlor or milking procedure, may be more reactive during the milking procedure (Wicks et al., 2004). As a result, first lactation cows may present aversive behavior that could cause harm to employees during milking. These behaviors, such as stepping and kicking, could be even greater in cows with udder edema. Behavior observations can begin the systematic evaluation of pain that may be experienced in first lactation cows with udder edema. The aim of this study is to investigate and compare the effects of udder edema on cow behavior in the milking parlor in first and second lactation Holstein dairy cattle.

2. MATERIALS AND METHODS

This study was approved by the Colorado State University's Animal Care and Use Committee (#1681). Cows in the first and second lactation that recently calved were enrolled in the study. Each cow was assigned an edema score and was video recorded in the milking parlor. When watching the recorded videos, observers noted when a cow would step and kick during the udder preparation period along with kick the milking unit off during the milking period. Behavior was separated into timepoints depending on the presentation of the behavior, either udder preparation, attaching of the milking unit, or during the active milking session. The length of each of these timepoints were also noted to determine rates of behavior for each cow.

2.1 DAIRY HERDS & MILKING PROCEDURE

Data was collected from two large (>500 head milking) Colorado dairies. Refer to table 3.1 for udder preparation procedures for Farm A and B. Recommended udder preparation procedure set by the Dairy Practices Council is as follows: apply predip to each teat, massage predip into teat sides and teat end with gloved hand, prestrip three squirts per teat with gloved hand, wipe off teat dip from teat sides and teat end, attach milking unit, and apply postdip to each teat after cessation of milking session.

Table 3.1 Udder preparation practices For Farm A And Farm B

	FARM A	**FARM B**
Step 1	Forestripping	Predip
Step 2	Wash and disinfect teat	Forestrip
Step 3	Dry teat	Wipe teat
Step 4	Attach milking unit	Attach milking unit
Step 5	Automatic take-off	Automatic take-off
Step 6	Post dip	Post dip

Farm A

The milking parlor was a double, twenty herringbone parlor with a rapid exit. The milking claw vacuum system had a vacuum level of 45kPa. Farm A used the FutureCow® brush (GEA Group Aktiengesellschaft, Düsseldorf, Germany) for washing and disinfecting the teats and drying the teats. The teat wash was a non-iodine, non-acid aqueous solution with chlorhexidine gluconate as the active disinfecting ingredient. A 1% iodine-based disinfectant was applied as a post-dip. Farm A had one pen of fresh cows ranging from 1-21DIM. All cows were fed ad libitum total mixed rations with two main ingredients of corn silage and alfalfa silage. None of the first lactation cows had prior training through the milking parlor.

Farm B

The milking parlor was a double, twenty-four stall parallel parlor with a rapid exit. The milking claw vacuum system had a vacuum setting at 41 kPa. Farm B used the Thrifty Dipper T-150 Pro Complete Foamer System™ (Thrifty, Burley, Idaho) for application of the pre-dip. A 0.9% iodine post-dip was applied with a hand-held dip cup after milking was completed. Farm B had two pens of fresh cows ranging from 1-25DIM. Both fresh cow pens at Farm B were fed a similar total mixed ration. All rations included two main ingredients of corn silage and alfalfa silage. All cows were fed ad libitum total mixed rations as well as grazing pasture. None of the first lactation cows had prior training through the milking parlor.

2.2 COWS IN STUDY

All cows in the study were Holsteins (n=376) and were either in their first or second lactation. Farm A had 124 first lactation cows and 74 second lactation cows. Farm B had 124 first lactation cows and 54 second lactation cows. In total there were 248 first lactation cows and 128 second lactation cows. A single observation was conducted per cow between 2-9DIM. Each farm was visited twice a week and cows were observed during the same milking shift each time. Farm A data collection occurred on Monday and Friday at the 8:00 AM milking shift. Farm B data collection occurred on Tuesday and Thursday at the 2:30 PM milking shift.

2.3 UDDER EDEMA SCORING

At the end of the milking session, a cow was indicated as having udder edema present (yes) or being absent of udder edema (no). The presence of udder edema was assigned at the end of the milking session to avoid distortion of the udder from udder fill. A cow was scored to have edema present when the appearance of the medial suspensory ligament was reduced, as defined by Morrison et al. (2018). Within the study, Morrison et al. (2018) categorized udder edema scores into none, mild, moderate, and severe. All cows within this study that presented as mild or greater were scored as having edema present. Scores were determined by the same observer throughout the study. The observer did not come into physical contact with any of the cows. The observer was standing in the milking pit and examined the cow in the milking stall from the rear to assign an udder edema score.

2.4 VIDEO SET UP AND RECORDING

Video recording began as cows entered the parlor. Each cow was identified by their ear tag at the beginning of the milking procedure. Milking sessions were recorded with eight GoPro cameras (GoPro Incorporated, San Mateo, CA). Each camera was fastened onto a secure spot to record cows from the rear while they were in the stalls of the milking parlor.

2.5 BEHAVIOR SCORING

Observational timepoints of the milking procedure were broken down into the nine categories and are defined in Table 3.2. Observations during the milking period were collected during the first five minutes of the milking session to avoid potential aversive behavior from over-milking. Any cows who completed their session before five minutes were noted and the total milking time was recorded. The separation of these behavioral observations matches the different phases of the milking procedure set by the Dairy Practices Council (udder preparation and unit attachment) along with matching the timepoints of udder preparation and active milking used by Kutzer et al. (2015).

Behaviors of stepping, kicking, and kicking off the milking unit were recorded for the rear legs only and are defined in Table 3.3. When a cow shifted her rear legs, each raise of a foot off the floor was counted as a step. All behavior data was recorded with four observers. Each observer received training to identify step and kick behaviors. The observers were tested for reliability using twelve sample videos. All observers, prior to recording of data, reached ninety percent agreement. This was determined by using the Pearson's product-moment correlation test.

Table 3.2 Identified Time Points of The Milking Procedure with Observed Behavior

Observational Time Points	**Definition**
PICS	Steps during udder preparation while the milker was in contact with the udder
PICK	Kicks during udder preparation while the milker was in contact with the udder
PNCS	Steps during udder preparation while the milker was not in contact with the udder
PNCK	Kicks during udder preparation while the milker was not in contact with the udder
SA	Steps during attachment of the milking unit
KA	Kicks during the attachment of the milking unit
SM	Steps during milking after complete attachment
KM	Kicks during milking after complete attachment
KOU	Kicking off the milking unit

Table 3.3 Ethogram of Behaviors Collected in Study

Behavior	Definition
Step	Low, vertical leg lift (Hopster et al., 2002)
Kick	Forceful and rapid movement of hind leg, often paired with direction of leg toward milker or milking unit (Van Reenen et al., 2002)
Kicking Off Milking Unit	A kick resulting in the forcible removal of the milking claw during the milking session.

2.6 STATISTICAL ANALYSIS

Data were analyzed using a linear regression model in version 4.1.1 of R (R Development Core Team, 2021). Explanatory variables and covariates are listed in Table 3.4. During the udder preparation phase, the independent variables in the final model included udder edema score, lactation along with the covariates of day in milk (DIM) and the length of time the milker was in direct contact with the udder during prep. During the attachment and milking phases, the independent variables included udder edema score, lactation and the covariates were day in milk (DIM) and total prep time. Length of time the milker was in direct contact with the udder was removed from the attachment and milking phase final model. The interaction between udder edema score and lactation was examined in all models. Model assumptions for normality and equal variance were evaluated based on residual plots. Our outcomes of interest, listed in Table 3.2, were converted into rate form, as seen in Table 3.5. Our explanatory variables of interest were lactation number and udder edema score (Table 3.4). The relationship between each outcome and the explanatory variables of interest were examined by fitting a separate linear model for each outcome. When examining the rate of steps during udder preparation while the milker was in contact with the udder, a square root transformation was used in the model to address the equal variance assumption. All other models did not use a transformation.

Table 3.4 Model Design for Analysis of Parlor Behavior in First and Second Lactation Cows With Edema or Without Edema

Explanatory Variables of Interest	**Description**
Lactation Number	First or second lactation
Edema Code - Yes or No	Binary score for presence of udder edema
Covariates	**Description**
Farm I.D.	Identification of Farm: A or B
Days in Milk (DIM)	Number of days cow has been in current lactation
Duration of Udder Prep in Seconds	Total time elapsed for udder preparation phase
Duration milker was in contact with udder during prep	Total time elapsed during the udder preparation phase that the milker was in direct contact with udder
Duration of Milking Session	Total time elapsed for active milking period in seconds.

Table 3.5 Observational Time Points Converted to Rates

Outcomes of Interest	**Definition**
Prep Time in Contact: Steps	Count of PICS / Total time in seconds milker was in contact with udder during preparation phase
Prep Time in Contact: Kicks	Count of PICK / Total time in seconds milker was in contact with udder during preparation phase
Prep Time Not in Contact: Steps*	Count of PNCS / Total time in seconds milker was not in contact with udder during preparation phase
Prep Time Not in Contact: Kicks*	Count of PNCK / Total time in seconds milker was not in contact with udder during preparation phase
Milking Unit Attachment: Steps	Count of SA / Total time in seconds milker was in contact with udder during milking unit attachment phase
Milking Unit Attachment: Kicks	Count of KA / Total time in seconds milker was in contact with udder during milking unit attachment phase
Active Milking: Steps	Count of SM / Total time in seconds milk unit was completely attached
Active Milking: Kicks	Count of KM / Total time in seconds milk unit was completely attached

* Models analyzing data of rates during udder preparation with no contact were excluded from results as the rate were meaningful above zero.

3. RESULTS

Within this study population, 95.6% (n=237) first lactation cows presented with udder edema and 81.3% (n=104) second lactation cows presented with udder edema (Table 3.6). Overall, 90.7% of cows presented with udder edema. Farm A had 87.4% (n=173) of the cows present with udder edema and Farm B had 94.4% of cows present with udder edema. A Fisher's exact test was applied to determine that udder edema is more prevalent in first lactation cows ($p<0.05$).

Table 3.6 Edema Presence by Lactation Number

	First Lactation (n=248)	Second Lactation (n=128)	Overall (n=376)
Lactation			
Edema Absent	11 (4.4%)	24 (18.8%)	35 (9.3%)
Edema Present	237 (95.6%)	104 (81.3%)	341 (90.7%)

Table 3.7 Lactation Number and Edema Presence by Farm

	Farm A (n=198)	Farm B (n=178)	Overall (n=376)
Lactation			
First Lactation	124 (62.6%)	124 (69.7%)	248 (66.0%)
Second Lactation	74 (37.4%)	54 (30.3%)	128 (34.0%)
Edema Presence			
Edema Absent	25 (12.6%)	10 (5.6%)	35 (9.3%)
Edema Present	173 (87.4%)	168 (94.4%)	341 (90.7%)

All the cows with no edema stepped at least one time, and 98.5% of cows with edema presented stepped at least one time. It was found that 80.6% (n=191) of first lactation cows with udder edema kicked at least one time and 73.1% (n=76) second lactation cows with udder edema kicked at least one time. During the udder preparation phase when the milker was in contact with the udder, first lactation cows with udder edema had a higher rate of stepping at 3.24 steps/session as compared to first lactation cows without udder edema with 1.11 steps/session ($p = 0.0168$). In Table 3.11, second lactation cows with udder edema had greater step rate at 2.93 steps/session than first lactation cows without udder edema during udder prep when the milker was in contact with the udder ($p = 0.0215$) As seen in Table 3.12, first lactation cows with udder edema had greater frequency of kick behavior at 2.68 kicks/session when compared to second lactation cows with udder edema during the active milking session at 1.24 kicks/session ($p=0.0092$). When investigating the behavior of kicking off the milking unit, it was found that first lactation cows with udder edema kicked off the milking unit more often compared to first lactation cows without udder edema ($p=0.0500$) and second lactation cows with udder edema ($p=0.0017$) (Table 3.13). Expected counts per session are presented in Table 3.9 and Table 3.10.

Table 3.8 Summary of Behavior Types Presented in Total Population by Lactation and Presence of Edema

	First Lactation		Second Lactation		Overall	
	Edema Absent (n=11)	**Edema Present (n=237)**	**Edema Absent (n=24)**	**Edema Present (n=104)**	**Edema Absent (n=35)**	**Edema Present (n=341)**
Step Behavior						
No Step	0 (0%)	5 (2.1%)	0 (0%)	0 (0%)	0 (0%)	5 (1.5%)
Step	11 (100%)	232 (97.9%)	24 (100%)	104 (100%)	35 (100%)	336 (98.5%)
Kick Behavior						
No Kick	2 (18.2%)	46 (19.4%)	9 (37.5%)	28 (26.9%)	11 (31.4%)	74 (21.7%)
Kick	9 (81.8%)	191 (80.6%)	15 (62.5%)	76 (73.1%)	24 (68.6%)	267 (78.3%)
Kick Off Behavior						
No Kick Off	10 (90.9%)	196 (82.7%)	24 (100%)	101 (97.1%)	34 (97.1%)	297 (87.1%)
Kick Off	1 (9.1%)	41 (17.3%)	0 (0%)	3 (2.9%)	1 (2.9%)	44 (12.9%)
Number of Steps						
Mean (SD)	11.2 (14.4)	15.0 (11.8)	15.8 (10.5)	15.6 (11.8)	14.3 (11.8)	15.2 (11.8)
Median [Min, Max]	9.00 [1, 53]	12.0 [0, 75]	11.0 [5, 41]	12.0 [1, 65]	10.0 [1, 53]	12.0 [0, 75]
Number of Kicks						
Mean (SD)	3.73 (4.65)	6.20 (8.72)	2.67 (5.13)	3.05 (4.94)	3.00 (4.94)	5.24 (7.69)
Median [Min, Max]	2.00 [0, 16]	3.00 [0, 62]	1.00 [0, 25]	2.00 [0, 21]	2.00 [0, 25]	3.00 [0, 62]
Number of Kick-Offs						
Mean (SD)	0.09 (0.30)	0.21 (0.56)	0 (0)	0.02 (0.16)	0.02 (0.16)	0.16 (0.48)
Median [Min, Max]	0 [0, 1.00]	0 [0, 5.00]	0 [0, 0]	0 [0, 1.00]	0 [0, 1.00]	0 [0, 5.00]

Table 3.9 Rates of Behaviors During Observational Timepoints Converted to Number of Events During Timepoint for First and Second Lactation Cows Categorized by Edema Presence

	Edema Absent 1st Lact			**Edema Present 1st Lact**		
	Behavior Count /Second Rate	**Mean Duration**	**Behavior Counts per Mean Time of Observational Timepoint**	**Behavior Count /Second Rate**	**Mean Duration**	**Behavior Counts per Mean Time of Observational Timepoint**
PICS	0.125 step/sec	8.91s[a,b,d]	1.11 steps/mean time	0.321 step/sec	10.1s[a,c,e]	3.24 step/mean time
PICK	0.137 kick/sec	8.91s	1.22 kick/mean time	0.169 kick/sec	10.1s	1.70 kick/mean time
PNCS	0.011 step/sec	93.1s	1.02 step/mean time	0.019 step/sec	137s	2.63 step/mean time
PNCK	0.001 kick/sec	93.1s	0.09 kick/mean time	0.0004 kick/sec	137s	0.05 kick/mean time
SA	0.095 step/sec	7.45s	0.70 step/mean time	0.275 step/s	10.2s	2.80 steps/mean time
KA	0.124 kick/sec	7.45s	0.92 kick/mean time	0.143 kick/sec	10.2s	1.45 kicks/mean time
SM	0.025 step/sec	278s	7.20 steps/milking session	0.021 step/sec	280s	6.04 step/milking session
KM	0.003 kick/sec	278s	1.25 kicks/milking session	0.009 kick/sec	280s	2.68 kicks/milking session

a: statistical difference PICS, b: statistical difference PICS, c: statistical difference KM, d: statistical difference KOU, e: statistical difference KOU.

Table 3.10 Rates of Behaviors During Observational Timepoints Converted to Number of Events During Timepoint for First and Second Lactation Cows Categorized by Edema Presence

	Edema Absent 2nd Lact			Edema Present 2nd Lact		
	Behavior Count /Second Rate	**Mean Duration**	**Behavior Counts per Mean Time of Observational Timepoint**	**Behavior Count /Second Rate**	**Mean Duration**	**Behavior Counts per Mean Time of Observational Timepoint**
PICS	0.273 step/sec	9.67s	2.63 steps/mean time	0.316 step/sec	9.30s[b,c,e]	2.93 step/mean time
PICK	0.131 kick/sec	9.67s	1.26 kick/mean time	0.138 kick/sec	9.30s	1.28 kick/mean time
PNCS	0.017 step/sec	117s	1.98 step/mean time	0.021 step/sec	127s	2.66 step/mean time
PNCK	0.001 kick/sec	117s	0.11 kick/mean time	0.0006 kick/sec	127s	0.07 kick/mean time
SA	0.271 step/sec	5.71s	1.54 step/mean time	0.270 step/s	4.65s	1.25 steps/mean time
KA	0.085 kick/sec	5.71s	0.48 kick/mean time	0.108 kick/sec	4.65s	0.50 kicks/mean time
SM	0.029 step/sec	288s	8.35 steps/milking session	0.028 step/sec	282s	7.89 step/milking session
KM	0.002 kick/sec	288s	0.83 kick/milking session	0.004 kick/sec	282s	1.24 kicks/milking session

b: statistical difference PICS, c: statistical difference KM, e: statistical difference KOU.

Table 3.11 Step Behavior During Udder Prep While In-Contact with Udder

Contrasts between Lactation Number and Edema Presence			
Contrast	**Estimated Difference in Step Rate**	**p-value, α=0.05**	**Cow Group with Greater Step Rate**
1 No – 2 No**	-0.1693[a]	0.1049	N/A
1 No – 1 Yes	-0.2133	0.0168*	1 Yes
1 No – 2 Yes	-0.2095	0.0215*	2 Yes
2 No – 1 Yes	-0.0439	0.4776	N/A
2 No – 2 Yes	-0.0402	0.5353	N/A
1 Yes – 2 Yes	0.0343[b]	0.9130	N/A

*statistical significance
** 1 stands for first lactation, 2 stands for second lactation, No stands for edema absent, Yes stands for edema present
a: a negative estimated difference in behavior rate indicates that the first animal category has a lower behavior rate than the second animal category.
b: a positive estimated difference in behavior rate indicates that the first animal category has a higher behavior rate than the second animal category

Table 3.12 Kick Behavior During Active Milking

Contrasts between Lactation Number and Edema Presence			
Contrast	**Estimated Difference in Kick Rate**	**p-value, α=0.05**	**Cow Group with Greater Kick Rate**
1 No – 2 No**	0.0010 [b]	0.8628	N/A
1 No – 1 Yes	-0.0058 [a]	0.2639	N/A
1 No – 2 Yes	-0.0005	0.9112	N/A
2 No – 1 Yes	-0.0069	0.0580	N/A
2 No – 2 Yes	-0.0016	0.6640	N/A
1 Yes – 2 Yes	0.0052	0.0092*	1 Yes

*statistical significance
** 1 stands for first lactation, 2 stands for second lactation, No stands for edema absent, Yes stands for edema present
a: a negative estimated difference in behavior rate indicates that the first animal category has a lower behavior rate than the second animal category.
b: a positive estimated difference in behavior rate indicates that the first animal category has a higher behavior rate than the second animal category

Table 3.13 Kicking Off the Milking Unit Behavior

Contrasts between Lactation Number and Edema Presence			
Contrast	**Estimated Difference in Kick-Off Count**	**p-value, α=0.05**	**Cow Group with Higher Kick-Off Rate**
1 No – 2 No**	0.0959 [b]	0.5728	N/A
1 No – 1 Yes	-0.0881 [a]	0.4798	N/A
1 No – 2 Yes	0.0719	0.6218	N/A
2 No – 1 Yes	-0.1841	0.0500*	1 Yes
2 No – 2 Yes	-0.0241	0.8306	N/A
1 Yes – 2 Yes	0.1600	0.0017*	1 Yes

*statistical significance
** 1 stands for first lactation, 2 stands for second lactation, No stands for edema absent, Yes stands for edema present
a: a negative estimated difference in behavior rate indicates that the first animal category has a lower behavior rate than the second animal category.
b: a positive estimated difference in behavior rate indicates that the first animal category has a higher behavior rate than the second animal category

4. DISCUSSION

Within this study, 90.7% of our sample population was identified to have udder edema. Dentine and McDaniel (1983) noted that 97% cows around parturition presented with udder edema. Alternatively, Morrison et al. (2018) identified that in Canada, 70% of their population had udder edema. Both the previously mentioned studies and the current study examined Holstein herds. Hayes and Albright (1976) found that udder edema is more severe in Holstein and Guernsey cattle than Jersey, Ayrshire, or Brown Swiss cattle. Further research could investigate the prevalence of udder edema in the United States dairy industry.

The use of behavioral observations in animal agriculture to identify potentially painful procedures is an accepted practice and has been widely used. Behavior in the milking parlor has been used to identify discomfort in cows with mastitis (Medrano-Galarza et al., 2012), those experiencing overmilking (Cerqueira et al., 2017), and cows exposed to stray voltage while milking (Aneshansley et al., 1992). The behaviors of stepping and kicking during milking have been repeatedly used to indicate whether a cow is experiencing a stressful event in the milking parlor (Willis, 1983, Hopster et al., 2002, Van Reenen et al., 2002, Rousing et al., 2004, Rousing et al., 2006, Sutherland and Huddart, 2012, Cerqueira et al., 2017). Researchers have also used behavioral observations as an indication for cows experiencing pain. Gleerup et al. (2015) developed the Cow Pain Scale, which utilized fifteen behaviors previously identified in literature, to assess initial pain and the progression of pain in dairy cattle. Metz-Stefanowska et al. (1992) reported that greater stepping behavior during milking was associated with discomfort in cows with teat lesions. Medrano-Galarza et al. (2012) found that step and kick behavior of dairy cows did change over time in cows with mastitis, a known painful disease. The use of behavioral observations in animal agriculture to identify potentially painful procedures is an accepted practice and has been widely used. Moreover, it is a minimally invasive method to assess an animal's current affective state. Results indicated that the presence of udder edema influenced the presentation of the stepping behavior during PICS and kicking behavior during KM and KOU. Conclusions about the rates of PNCS and PNCK behavior cannot be made due to low frequency rates (<0.02 behavior/time in seconds). The udder preparation phase is when the milker is in contact with the udder. This physical contact may be a novel experience for first lactation cows or could cause discomfort in cows with udder edema. This is also true during the active milking phase where the sensations of the milking unit may be unfamiliar.

Cows react to a novel stimulus or environment by entering a fear state. Fear is a powerful emotional state that provokes a stress response within an animal (Grandin, 1997). Fear behavior is expressed when a painful stimulus is present that results is defensive behavior and decreased presentation of pain-related behavior. More simply put, fear behaviors are expressed to avoid a possibly painful stimulus. Conversely, pain behavior is triggered by injurious stimulation, prioritizing behaviors that promote healing instead of defensive actions. The activation of the neural fear system is not directly tied to the sensation of pain, but the neural pathway of pain does move through some of the same areas of the brain that are involved with the fear response (Elman and Borsook, 2018). Consequently, an animal's reaction to pain may be altered when the animal is afraid. This may cause conflicting findings when using behavioral observations as the sole determining factor for when cows are experiencing pain from udder edema, particularly in first lactation cows.

With udder edema having greater levels of prevalence in first lactation cows, it is important to note that along with the acts of parturition and lactogenesis, the milking parlor and milking procedure is a novel experience, primarily because of increased handling and unaccustomed sensations (Van Reenen et al., 2002). In the milking parlor, documented fear behaviors of dairy cows include vocalizations, tail switching, stepping, and kicking (Rushen et al., 1999, Munksgaard et al., 2001, Ivemeyer et al., 2011, Kutzer et al., 2015). First lactation dairy cattle are more reactive when adapting to the milking routine when they are not accustomed to the parlor and handling of their udder (Bremner, 1997). When examining both familiarized and non-familiarized cows, Bremner (1997) found that first lactation cows that were familiarized to the milking parlor and milking procedure had a lower frequency of behavior. Hemsworth et al. (1989) also found that first lactation cows who were not handled around the time of calving had more flinch, step, and kick behaviors during the first twenty weeks of lactation. Decreased reactivity and hesitancy to enter the parlor with habituation with first lactation cows that are habituated to the parlor is further supported by Bertenshaw et al. (2008), Sutherland and Huddart (2012), and Kutzer et al. (2015). Decreasing fear responses improves both milk yields and response to udder stimulation for milk letdown (Rushen et al., 1999, Hedlund and Lovlie, 2015, J. Moran and Doyle, 2015). One could postulate these first lactation cows with udder edema are experiencing even greater levels of distress and discomfort than their experienced multiparous counterparts who have been milked before or non-edematous primiparous cows. The decreased reactivity levels of cows that are acclimated to the parlor and milking procedure is important to note as unfamiliarity to touch and sensations in the milking parlor is a potential factor for the change in frequency of behaviors that was seen in this study. To control for the effects non-habituation has on parlor behavior in first lactation cows with udder edema, a sample population of cows should be habituated to the milking parlor and udder preparation and compared to those not habituated.

From this study, it can be postulated that a first lactation cow with an edematous udder may experience discomfort during the milking session while their udder is handled. The discomfort may be due to the pressure within the udder from the trapped lymphatic fluid. When investigating pain in mammalian animals, human models of similar maladies have been used to demonstrate the possibility of sensory effects that animals may experience. In research conducted by Sheikhi Mobarakeh et al. (2018) humans diagnosed with breast cancer-related lymphedema stated that it is a painful morbidity. Self-reported pain levels were reduced when patients received decongestive therapy during the study. Overall, 87.8% of patients reported a significant reduction in pain after the therapy was completed (Sheikhi Mobarakeh et al., 2018). Lymphedema as a side effect of breast cancer is different from udder edema in that lymphedema is a side effect of parts of the lymphatic system being removed from the breast tissue area during surgery or disruption of the lymphatic system due to radiation (Fu, 2014). These findings illude to a potential source of added discomfort for cows with udder edema at the time of lactogenesis: intramammary pressure (IMP) may be at a greater level in cows with edematous udders. As blood flows to the udder in response to udder stimulation, IMP increases (Kitts et al., 1963). The removal of milk relieves pressure within the mammary tissues, but higher levels of pressure may still be present in edematous udders when compared to non-edematous udders. As decongestive therapy was beneficial for the physical and psychological wellbeing of women experiencing lymphedema, further research should investigate the impacts and changes of intra-mammary pressure brought on by udder edema to determine if this disorder is painful. Moreover, exploring the practical relief of this potential added pressure to reduce the negative impacts udder edema has on milk production and udder health should be explored to aid producers in mitigating this disorder. While examining the effect of total udder prep time on the step and kick behavior in cows was not the focus of the study, it may be worthwhile to investigate this variable further when examining parlor behavior in association with udder edema. By providing a standard amount of udder prep time, observations can be made to test if cows with udder edema have ideal milk letdown when compared to cows without udder edema or if edematous udders require longer udder preparation times for adequate milk letdown.

5. CONCLUSIONS

Udder edema was very prevalent (90.7%). Udder edema has a negative impact on parlor behavior during the udder preparation period when the milker is in contact with the udder and during milking. Due to the relationship of udder edema with other transitional diseases, it is paramount to conduct further research to better understand the etiology of this disorder. There is also a need to find practical and economical methods of treatment and conduct an industry-wide epidemiological survey of udder edema to quantify the prevalence of this disorder. The data from this study provide insight on how udder edema in dairy cattle affects both parlor efficiency and animal welfare. There is a need to reduce the prevalence of udder edema because it may negatively impact animal welfare.

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