

EVALUATING SEX DIFFERENCES IN BEEF CATTLE LUXURY BEHAVIORS
AND THE USE OF ENVIRONMENTAL ENRICHMENT AS A NOVEL
BEHAVIORAL PHENOTYPE

A Thesis

by

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ABSTRACT

Efficient sampling strategies expedite behavioral data collection. While multiple studies have evaluated sampling strategies for core behaviors in cattle, few have focused on social interactions or stereotypic behavior. Therefore, for the first chapter, to identify sampling strategies that accurately capture feedlot cattle social behavior and stereotypic behaviors such as brush use, steers ($n = 3$ pens; 9 steers/pen) were observed from 8:00 to 17:00. Differences among sampling strategies were evaluated using a non-parametric one-way ANOVA Kruskal-Wallis Test. Pearson correlation evaluated the strength of association between a specific sampling strategy and continuous observations. Observing cattle for 15 minutes every 30 minutes yielded the highest accuracy for all behavioral metrics and was considered the most effective strategy for comprehensively evaluating cattle social behavior ($r^2 > .75$; $P < 0.05$). For the second chapter, because an empirical examination of how sex influences cattle social behavior has been absent, our objective was to evaluate the impact of sex on feeding behavior and the performance of social behaviors. Social behaviors and brush use were decoded using continuous observation for 15 minutes of every 30 minutes on d 1, 2, 4, 8, 16, 32, and 64. A Generalized Linear Mixed Model (PROC GLIMMIX) with a Poisson distribution and log transform link (to normalize the data) was used to evaluate differences between sexes for each behavior. The model included sex, sex within pen, and research day as fixed effects, and pen as the random effect. Exit velocity was evaluated in a separate GLIMMIX model that included sex within pen and research day as fixed effect and the animal's individual ID as random effect. Lastly, productivity and feeding behavior traits were evaluated using a GLIMMIX model that included sex as fixed effect and sex within pen as random effect. Heifers showed higher temperament scores than steers ($P < 0.05$). Steers performed on

average and total duration/d more social ($P < 0.001$) and stereotypic behaviors ($P < 0.04$) and used the brush ($P < 0.04$) more than heifers. In bout frequency/d, steers performed more allogrooming ($P = 0.002$), brush displacement ($P = 0.02$), bunk displacement ($P < 0.001$), head butt ($P < 0.001$), and brush use ($P < 0.001$). Lastly, the third chapter address the gap of understanding regarding the relationships among environmental enrichment use, temperament, productivity, and feeding behavior for *Bos indicus* cattle. To examine the effect of divergent behavioral phenotypes for brush usage on temperament, productivity, and feeding behavior patterns, four pens of Brahman yearling cattle (Pen 1 & 2: 15 heifers/pen, and Pen 3 & 4: 19 steers/pen) were housed in dry lots. Ten animals from each pen were marked with a colored ear tag for individual identification. Animals were video recorded for the same time and days as chapter 2, as well as EV and body weight. Animals were categorized into one of three phenotypes (high, medium, low brush use). A PROC GLIMMIX evaluated the interaction of brush usage phenotype and gender as fixed effect, and pen as random effect on final BW (kg), average daily gain (kg/d), dry matter intake (kg/d), feed to gain ratio, bunk visit frequency (events/d), bunk visit duration (min/d), bunk visit eating rate (g/min), head down duration (min/d), head down duration per bunk visit duration, and exit velocity. High brush use steers showed to be the most productive ones, presenting higher FBW ($P = 0.0005$), higher ADG ($P = 0.0002$), and higher DMI ($P = 0.0003$) than low brush use steers, and as expected, steers were more productive than heifers while also being less temperamental than heifers ($P = 0.0008$). These results provide insight into accurate and efficient sampling strategies that expedite social behavior data collection in cattle. Also, the study presents novel empirical information regarding the impact of sex on cattle social behavior, environmental enrichment, productivity, and feeding behavior, suggesting the need for sex-specific statistics in research and management practices and

highlighting the importance of incorporating sex-specific behavioral profiles into sensor technology and algorithm development.

DEDICATION

I dedicate this dissertation to all of my family and friends for all their support, for always believing in me and encouraging me to be better every day and accomplish all my dreams. I also want to thank Carlos Lucena for always being there, day and night, to support me and teach me all the tools that I needed to accomplish my dream of having a master's in animal welfare and behavior. I am also very thankful for ST genetics, especially for Gustavo Toro for allowing me to do my research at ST genetics, and for believing in me since the first day we meet. I also want to thank all the students that did research hours at the animal welfare and behavior lab at Texas A&M University for helping me decode so many hours of cattle video; this research would not be possible without them. I also have deep appreciation for Dr. Riley, who so willingly since the first day that we meet taught me so much, not only in statistics and cattle but life in general, because after all that time we spent looking at statistical models and me practicing my English and him practicing his Spanish I was always happy to leave his office with a lot of new knowledge. I also want to thank Dr. Carstens because his area of expertise was completely different to mine, and he was so willing to pass his knowledge to me, which made so interesting all our meetings and I was able to add feeding behavior data to my dissertation. Also, thanks to Keara, who was always there for me day and night, willing to help and support me in anything that I needed. Lastly to my incredible mentor and professor, Dr. Daigle, who through all these years, has been there to teach me and guide me not only in the academic world but also in life, because she was not only my professor, but more than that, the best friend that my master's could have given me. Thank you for always being there for me. Thank you for supporting me in the best years of my life.

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INTRODUCTION

Cattle are social animals, and as such, the performance of social behaviors (or lack thereof) can provide insight into an animal's welfare state (Poole, 1997). Animals housed in groups perform affiliative behaviors (e.g., allogrooming) that are associated with positive emotions (Boissy et al., 2007), have calming effects (Laister et al., 2011), facilitate the formation of social bonds (Wasilewski, 2003), and can result in improved coat hygiene (Boissy et al., 2007). However, living in groups is also accompanied with inherent costs, including competition for resources (Zobel et al., 2011) such as food, water, and mechanical brushes (Foris et al., 2019). If resources are scarce, highly valued, or defended, animals may engage in agonistic interactions, which could result in injury or death. Also, these animals are normally housed in counter-evolutionary single-sex groups. Feed and nutrition requirements vary with age and sex, so separating the cattle increases the efficiency with which these needs can be met and makes animal management easier (Lalman & Richards, 2017). Therefore, while housing cattle in single-sex social groups may be counter-evolutionary, this management practice can yield welfare benefits. Even so, the frequency, duration, and circadian pattern of social behaviors can provide feedback to producers regarding resource availability, herd synchrony, and level of psychosocial stress.

Social bonding enhances individual fitness (Kulik et al., 2015). The behavior of conspecifics (Schuett, 2009) is likely to influence the individual behavior of gregarious animals, including their physiological fear response (Jones et al., 1995; Perkins, 2016) as well as behavioral foraging efficiency and efficacy (Galef & Giraldeau, 2001). Luxury behaviors (e.g., brush use, mounting, head butting, allogrooming, and bunk displacement) may be relevant to current social status and reproduction but are not essential for health and survival. The strength

of influence that social interactions have on the performance of luxury behaviors by specific individuals will vary due to differences in personality (Marchetti & Drent, 2000), experience, social status, and group size (Gygax et al., 2010).

When cattle are housed in counter-evolutionary single-sexed groups, consideration must be made for sex-specific behavioral differences. Social behaviors are crucial to the development and maintenance of a social structure. However, cattle exhibit sex-specific behavioral differences based on social stability. Dominance in a male herd is relatively unstable and involves more aggression compared with female hierarchies (Sowell et al., 1999), which could impact the amount of mounting and head butting that would be expected in a group of animals. Male Holstein Friesian calves from 4 to 15 months of age are more likely to initiate social interactions than females of the same age among individuals of the same sex (Freslon et al., 2019), suggesting that sex may influence social interactions in cattle. Understanding these differences can facilitate the implementation of best management practices.

Living in captivity occasionally results in the development of behaviors that differ from the behavioral repertoire of the species' wild counterpart (Redbo & Nordblad, 1997). In some cases, these behaviors manifest as stereotypies and, as such, consist of repeated movements that seem to lack any function in the context in which they are performed (Redbo & Nordblad, 1997). While the development of stereotypic behaviors can be indicative of an individual having difficulty coping with their current conditions, the perseverance of these behaviors may be indicative of a positive welfare state. Some of the common stereotypies observed in cattle are bar licking and tongue rolling in response to limited opportunities to engage in orally-centered activities. Cattle will typically stop performing oral stereotypies when allowed to graze and will resume high levels of stereotypies after re-tethering post grazing (Redbo, 1992). Monitoring

these stereotypic behaviors can provide insight into whether the animal is experiencing frustration or has developed a coping strategy when the environment is not in alignment with the species' natural, historical environment.

Assessment of beef cattle temperament has received greater interest in recent years due to the connection between excitability, animal physiology, and carcass composition and quality (King et al., 2006; Struthers, et al., 1997). In *Bos indicus*-derived cattle, temperament, measured as flight speed, has been shown to correlate with weight gain (Fordyce & Goddard, 1984; Struthers et al., 1997) and is reflective of feeding behavior. Individual feeding behavior can be used to evaluate feed efficiency (Lancaster et al., 2009) and reflect health status (Quimby et al., 2001). Technological advancements have made collecting individual animal feeding data possible. Electronic radio frequency identification systems such as GrowSafe System, Ltd., have been designed to measure feeding behavior traits in beef cattle such as dry matter intake (DMI), bunk visit frequency (BVF), bunk visit duration (BVD), bunk visit eating rate (BV eating rate), and head down duration (HDD) with a high degree of accuracy (Mendes, 2011). However, feeding behavior meets a primal need for the animal and may not be as responsive to changes in health status as social interactions. Thus, there are known and novel behavioral phenotypes that can be collected efficiently and measured non-invasively that provide insight into the animal's potential to thrive and be productive and profitable.

One management strategy that is designed to provide animals with the ability to interact with their environment and reduce overall stress is the provision of species-specific, biologically-appropriate environmental enrichment. Environmental enrichment (EE) alters a captive animal's environment with the goal of enhancing the quality of life of the individual by providing opportunities to express natural behaviors and meet behavioral and physical needs (Newberry,

1995; Shepherdson, 1999). Because feedlots provide limited environmental complexity to cattle, adding biologically-relevant stimulation such as mechanical brushes has the potential to enhance welfare by providing environmental complexity, stimulating the performance of pleasurable behaviors, and engaging cattle in natural behaviors.

The type of data collection strategy used to measure the frequency, duration, and total time cattle perform specific behaviors will be influenced by research objectives. Two primary sampling strategies are typically used to capture animal behavior: continuous recording and scan sampling (Altmann, 1974). Continuous sampling is a true record of the animal's behavior, as relevant behaviors are decoded for the entire duration of time. Continuous sampling provides the most accurate representation of a group or individual's behavioral repertoire (Park et al., 2020). However, this method is labor intensive and time consuming (Madruga et al., 2017). Thus, while social behavior is typically dynamic and of short duration, using continuous observation throughout the entire observation period is impractical. Yet, behavior is an objective measurement that can inform welfare assessment and cattle management. Multiple studies have evaluated sampling strategies designed to expedite data collection regarding lying, standing, and brush use behaviors (Main et al., 2010; Mitlöhner et al., 2001) However, little has been published regarding a sampling strategy for capturing the social and stereotypical behavior of cattle housed in feedlots and an efficient method to measure the relationships among feeding behavior, social behavior, temperament, and productivity.

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EVALUATING ACCURATE AND EFFICIENT SAMPLING STRATEGIES DESIGNED TO MEASURE SOCIAL BEHAVIOR AND BRUSH USE IN DYLOT HOUSED CATTLE

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Introduction

Cattle are social animals, and as such, the performance of social behaviors (or lack thereof) can provide insight into an animal's welfare state (Poole, 1997). Cattle are intelligent and curious creatures; thus, social interactions are a critical component of their ethos. Social interactions are a requirement of group living and can have a positive or negative valence. Animals housed in groups perform affiliative behaviors (e.g., allogrooming) that are associated with positive emotions (Boissy et al., 2007), have calming effects (Laister et al., 2011), facilitate the formation of social bonds (Wasilewski, 2003), and can result in improved coat hygiene (Boissy et al., 2007). However, living in groups is also accompanied with inherent costs, including competition for resources (Zobel et al., 2011) such as food, water, and mechanical brushes (Foris et al., 2019). If resources are scarce, highly valued, or defended, animals may engage in agonistic interactions, which could result in injury or death. For example, in overstocked pens, cattle tend to perform more agonistic interactions, and the cattle that are less successful at displacing spend more time lying down, which affects the pen dynamic (Winckler et al., 2015). Thus, the frequency, duration, and circadian pattern of social behaviors can provide feedback to producers regarding resource availability, herd synchrony, and level of psychosocial

stress. An animal's response to a stressor is related to the characteristics of the stressor, such as predictability and controllability, in addition to characteristics of the individual experiencing the situation, such as coping style, genetics, sex, and life experiences (Anisman, 2005).

Living in captivity occasionally results in the development of behaviors that differ from the behavioral repertoire of the species' wild counterpart (Redbo & Nordblad, 1997; Mason, 1991). In some cases, these behaviors manifest as stereotypies and, as such, consist of repeated movements that seem to lack any function in the context in which they are performed (Redbo & Nordblad, 1997). While the development of stereotypic behaviors can be indicative of an individual having difficulty coping with their current conditions, the persistence of these behaviors may be indicative of a positive welfare state. The performance of stereotypic behaviors can be rewarding and a self-reinforcing strategy to cope with their current scenario (Mason, 1991). Cattle typically stop performing stereotypies when allowed to graze, but they will resume high levels of stereotypies after re-tethering post grazing (Redbo, 1992) due to the fact that diet is a key factor affecting this behaviors (Ridge et al., 2020). Some of the common stereotypies observed in cattle are bar licking and tongue rolling; these commonly observed stereotypies are important to measure.

The type of data collection strategy used to measure the frequency, duration, and total time cattle perform specific behaviors will be influenced by research objectives, yet need to be evaluated to expedite large-scale behavioral observations. Two primary sampling strategies are typically used to capture animal behavior: continuous recording and scan sampling (Altmann, 1974). Continuous sampling is a true record of the animal's behavior, as relevant behaviors are decoded for the entire duration of time. Continuous sampling provides the most accurate representation of a group or individual's behavioral repertoire (Park et al., 2020) and can capture

behaviors that occur infrequently, of short duration, or on a circadian pattern (Daigle & Siegford, 2014). However, this method is labor intensive and time consuming (Madruga et al., 2017). With the onset of precision livestock management, and the accompanying challenges regarding processing large data sets and preserving sensor technology battery life, identifying alternative sampling strategies that provide an accurate representation of the continuous record has ethological and technological implications (Whalin et al., 2016).

Behavior is an objective measurement that can inform welfare assessment and cattle management. Multiple studies have evaluated sampling strategies designed to expedite data collection regarding lying, standing, and brush use behaviors (Mitlöhner et al., 2001; Madruga et al., 2017). However, these studies have varied in their implementation of continuous observations, and ultimate recommendations have included identifying context-specific sampling strategies (Horvath et al., 2020). Consequently, little has been published regarding a sampling strategy that monitors social and stereotypical behavior of cattle housed in feedlots. Thus, the objective of this study was to identify a sampling strategy that could accurately and efficiently record the performance of social, agonistic, and stereotypic behaviors in drylot-housed cattle with access to brushes.

Materials and Methods

Data for the present study were derived from a portion of a larger data set. The methodology was previously outlined in Park et al. (2020) and is briefly described below.

Housing, Diet, and Treatments

Fifty-four predominately British and British-Continental crossbred steers were shipped to the Texas A&M Agrilife Research Feedlot in Bushland, Texas, United States, in the fall of 2017. Cattle were blocked by weight into a light (283.95 ± 13.75 kg) and a heavy block ($320.69 \pm$

12.97 kg) before being allocated to one of six pens. Three of these pens each had a stationary L-shaped brush. For this research, only the pens with brushes were included in the analysis. Each pen was 25.5 m by 7 m (19.83m² per head) with earthen flooring. Shade was provided in the form of a partial roof covering (5 x 7 m; 5m² per head). Each pen provided nine individual Calan head gate feeders and housed nine animals accordingly. Water was provided *ad libitum* from an automatic water trough. A schematic of the pen layout is included in Park et al. (2020).

Behavioral Observations

Cattle behavior in the pen was recorded from 08:00 to 17:00 on d 1, 16, and 64, relative to brush placement implementation using a closed-circuit video camera recording system, which was installed to ensure there were no blind spots within the pen. Behavioral data was decoded from video recordings using the continuous sampling method (Altmann, 1974; Mitlöhner et al., 2001). The data focused on the frequency and duration each steer spent engaged in allogrooming, bar licking, tongue rolling, and utilizing the brush, as well as the frequency of headbutting and mounting. All continuous behavioral data were collected by 23 trained observers utilizing BORIS (Version 6.1.4) (Friard & Gamba, 2016). Interobserver reliability between observers and trainer, as well as among observers, was no less than 95% accuracy.

Sampling Strategies

Eight different sampling strategies were selected (Table 1) to evaluate cattle brush use and social and stereotypical behavior. Each of these sampling strategies was extracted from the continuous observation data.

Statistical Analysis

Data were extracted from the continuous observation data set according to eight different sampling strategies (Table 1) to evaluate the impact of sampling strategy on overall outcome.

Average duration of time per bout (sec/bout), number of bouts per day (count/d), and total duration of time per day (sec/d) spent performing each behavior were calculated for each sampling strategy. Normality was evaluated using the univariate procedure in SAS (SAS University Edition). According to the Anderson-Darling and Kolmogorov-Smirnov tests, behavioral values for allogrooming, headbutt, tongue rolling, bar licking, and mount and brush use were not normal and could not be normalized. To test for significant differences among the sampling strategies for each behavior, a Kruskal-Wallis Test was evaluated with a nonparametric One-Way ANOVA. To evaluate the strength of association between the results generated with a specific sampling strategy against the results generated using continuous observation, a Pearson correlation (PROC CORR) was used to correlate the results from the average duration of bout, number of bouts, and total duration of all bouts for individual steers with the results of continuous observation using PROC CORR in SAS software (SAS University Edition).

Results

Allogrooming

While the average duration of an allogrooming bout did not differ across sampling strategies (Figure 1a), two sampling strategies differed from continuous observations for the frequency (Figure 1b) and total duration of time spent allogrooming per day (Figure 1c). Outcomes from the sampling strategies 5EV60 and 5EV30 differed from continuous observations regarding the frequency of allogrooming bouts ($P > 0.10$) and the total duration ($P > 0.21$) of time spent per day engaged in allogrooming behavior. The sampling strategy 15EV30 had the greatest strength of association with continuous observations ($r^2 > 0.80$; $P < 0.0001$), while the sampling strategy 5EV60 ($r^2 > 0.65$; $P < 0.06$) had the weakest relationship (Figure 2a).

Bar Licking

Sampling strategy did not impact the record of average duration of a bar licking bout (Figure 1d), the frequency of these bar licking bouts (Figure 1e), nor the total duration of time spent per day engaged in bar licking behavior (Figure 1f; $P > 0.08$). Bar licking was most accurately captured using the 15EV30 sampling strategy ($r^2 > 0.95$; $P < 0.0001$), while the least accurate sampling strategy was 5EV60 ($r^2 > 0.65$; $P < 0.08$) (Figure 3b).

Tongue Rolling

Irrespective of sampling strategy, average bout duration for tongue rolling ($P > 0.99$) did not differ from continuous observations (Figure 1g). The sampling strategies 15EV30 ($P > 0.2$), and 8To14 ($P > 0.5$) did not differ from continuous observations for both tongue rolling frequency (Figure 2h) and the total duration of time per day spent tongue rolling (Figure 1i). The most accurate sampling strategy to capture tongue rolling was 8To14 ($r^2 > 0.95$; $P < 0.0001$), while the least accurate sampling strategy was 14To16 ($r^2 > 0.80$; $P < 0.06$) (Figure 3c).

Brush Utilization

Irrespective of sampling strategy, bout duration for brush utilization ($P > 0.99$) did not differ from continuous observations (Figure 1d). For brush use bout frequency, 15EV30 ($P > 0.2$) and 8To14 ($P > 0.1$) did not differ from continuous observation. For total duration of brush use per day, 10EV30 ($P > 0.2$), 15EV30 ($P > 0.7$), 14To16 ($P > 0.08$), and 8To14 ($P > 0.1$) did not differ from continuous observations (Figure 1d). The most accurate sampling strategy to capture brush use was 8To14 ($r^2 > 0.70$; $P < 0.0003$), while the least accurate sampling strategy was 14To16 ($r^2 > 0.80$; $P < 0.001$) (Figure 2d).

Head Butt

The sampling strategies 15EV30 ($P > 0.3$) and 8To14 ($P > 0.6$) did not differ from continuous observation for the total number of head butts performed per day within a pen (Figure 3b). The most accurate sampling strategy to capture head butt was 15EV30 ($r^2 > 0.95$; $P < 0.0001$), while the least accurate sampling strategy was 14To16 ($r^2 > 0.80$; $P < 0.001$) (Figure 4a).

Mounting

Irrespective of sampling strategy, bout frequency for mounting ($P > 0.7$) did not differ from continuous observations (Figure 3a). The most accurate sampling strategy to capture mounting was 15EV30 ($r^2 > 0.75$; $P < 0.05$), while the least accurate sampling strategy was 10EV60 ($r^2 > -0.05$; $P < 0.8$) (Figure 4b).

Discussion

The objective of this study was to determine which sampling strategies could accurately capture social and stereotypic behavior in cattle housed in feedlots with access to a brush. Observing cattle behavior for 15 minutes every 30 minutes was shown to be the optimal sampling strategy for evaluating daily bout frequency, average bout duration (sec), and total duration of all bouts (sec). This sampling strategy provides insight as to what happens throughout the day and is designed to capture behaviors that happen infrequently or in short durations (Daigle & Siegford, 2014). On the other hand, observing the animals from 14:00 to 16:00 was shown to be the least accurate sampling strategy. That could be due to the circadian pattern of cattle behavior. For example, behaviors such as brush use, mounting, and allogrooming tend to be performed at lower frequencies during the morning than during the afternoon (Meneses et al., 2021). If the time of day can influence when the behaviors are

expected to occur, then samplings should be distributed throughout the day; otherwise, the accuracy of the sampling strategy could be negatively impacted.

Contemporary housing systems can result in cattle living in high-density groups where animals may be unable to avoid conflict or may impede upon individual spatial needs. Social animals housed in groups will inevitably develop hierarchies and will engage in social behaviors designed to establish a dominance social structure and dictate resource access (Craig, 1986). The use of the space varies depending on the animal's dominance status; for example, according to Donaldson et al. (1971), low-ranking animals were observed moving constantly to prevent or avoid agonistic behavior. Therefore, being aware of the social structure is crucial to identifying individuals who are experiencing difficulties accessing resources and to identifying any management strategies that can alleviate this social stress and minimize competition for resources.

Mounting was shown to be the most difficult behavior to decode, displaying a negative value in the correlation of 10EV60 with continuous observation, which could be due to the infrequent occurrence of mounting behavior. Feedlot steers housed in pens with a brush perform fewer mounts than feedlot cattle housed without a brush (Park et al., 2020). Mounting is a behavior that has multiple connotations. Mounting is required for copulation, is a behavioral signal that can be indicative of reproductive status, and is involved in both affiliative interactions (e.g., play) and agonistic interactions (e.g., establishing dominance, bullying). Thus, the frequency of this behavior is context specific.

Mounting is the behavior performed during the phenomenon of bullying, a scenario when cattle will perform mounting at an abnormally high rate and a single individual is the recipient of the mounting attempts, which can result in injury and death. Outbreaks of Buller Steer Syndrome

occur in 91% of feedlots with a carrying capacity of over 8,000 and is the third most common health problem (after BRD and digestive problems) for feedlot cattle (NAHMS Feedlot, 2011). In 1981, the monetary losses per buller were estimated to be \$23 (Ulbrich, 1981), excluding any economic losses associated with product quality and feed efficiency of penmates. Thus, monitoring mounting has welfare and economic implications beyond indicating reproductive status.

Mounting is used to establish dominance, as dominant animals have shown to be responsible for initiating 60% of the mounts in a group. This behavior is also influenced by the novelty of the animals in the group. In pens of newly introduced cattle, animals engage in greater mounting events, suggesting as well that this behavior is involved in social dominance (Irwin et al., 1979; Klemm et al., 1983), making this behavior complex and variant and therefore hard to measure. Mounting is a behavior that is not only difficult to measure but also difficult to interpret due because of the multidimensional use of this behavioral movement, which is can be used for play behavior, social dominance, stereotypic behavior, or reproductive signaling.

Non-nutritive oral behaviors such as bar licking and tongue rolling can be informative regarding an animal's welfare state. Cattle are orally-motivated creatures that evolved to spend most of their days engaged in orally-centered behaviors such as grazing and ruminating (Ridge, et al 2020). Modern agriculture can include housing cattle in environments that restrict the performance of or limit the duration of time engaged in these natural behaviors, thus altering the duration of time spent moving their mouths (Ridge, et al 2020). Environmental enrichment, such as a brush, reduces the performance of tongue rolling and bar licking (Park et al., 2020). Cattle have shown sustained interest in engaging with environmental enrichment such as brushes, suggesting that cattle brushes provide mental and physical stimulation. This could be the reason

why it was so hard to measure stereotypic behavior in this research. Each of the pens was equipped with an L-shaped brush, so cattle were not performing stereotypic behaviors very frequently.

These findings provide guidance regarding how to expedite large-scale behavior observations that optimize the collection of cattle social behavior data. The results from this study could be used to design ways to extend the battery life of sensor technology. If a sensor only needs to turn on for 15 minutes every 30 minutes, then the sensor can have a longer battery life and generate smaller yet equally informative datasets, making data management and output easier to use and resulting in increased adoption and greater economic returns for the producer. This research provides several options, depending on the percentage of accuracy desired by the observer, which could be used in the interest of observing the animal's behavior, optimizing social behavior observation, and offering flexibility to the researcher.

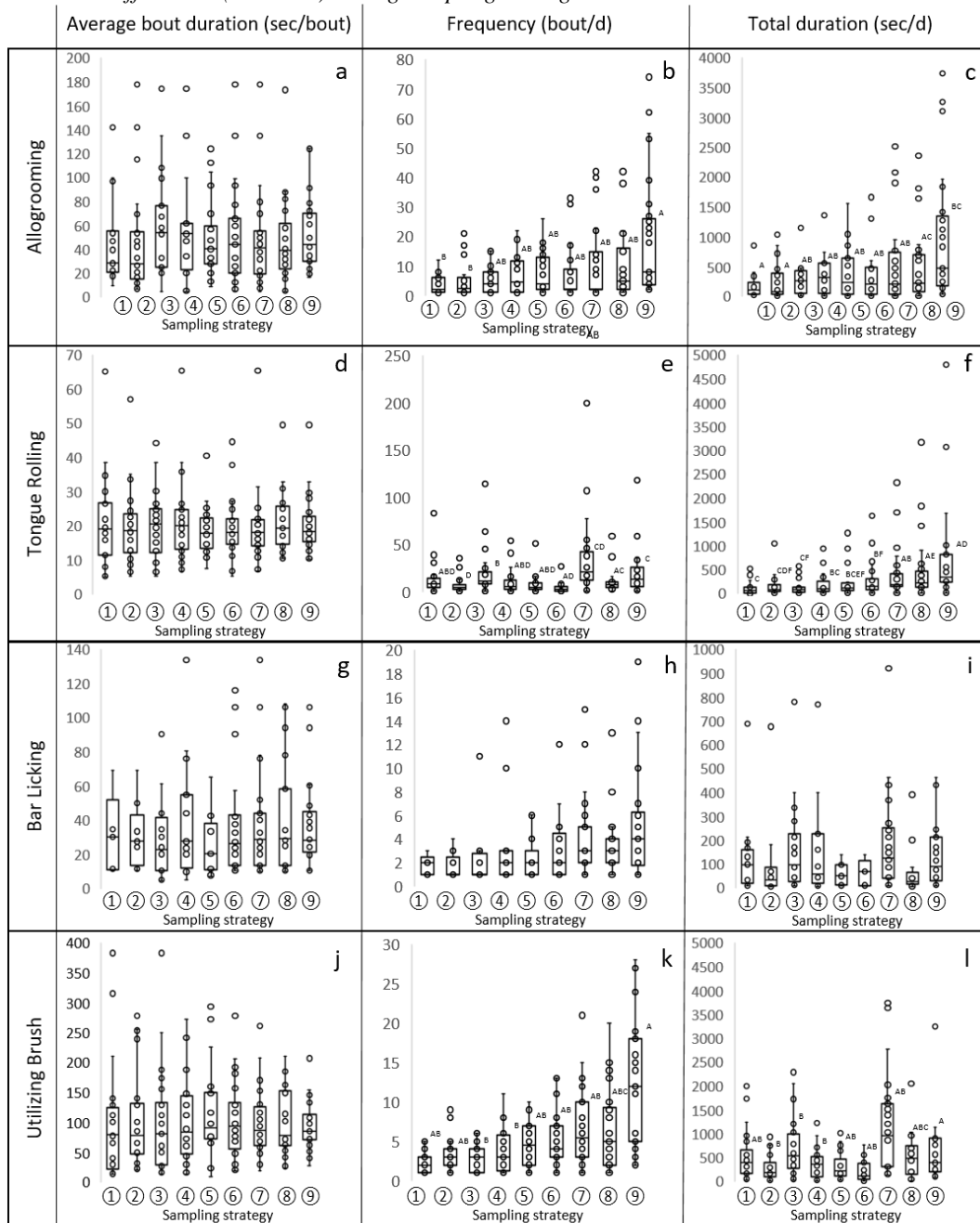
Table 1.

Sampling strategy description, acronym, and total duration of time (min) of video that would need to be evaluated to collect the behavioral data.

Interval ID	Description	Total duration of time/day evaluated (min)
Continuous	Video was decoded from 08:00 to 20:00	720
5EV30	5 minutes of video was decoded every 30 minutes	70
5EV60	5 minutes of video was decoded every 60 minutes	35
10EV30	10 minutes of video was decoded every 30 minutes	140
10EV60	10 minutes of video was decoded every 60 minutes	70
15EV30	15 minutes of video was decoded every 30 minutes	210
15EV60	15 minutes of video was decoded every 60 minutes	105
8to14	Video was decoded from 8:00 to 14:00	360
14to16	Video was decoded from 14:00 to 16:00	120

Figure 1

Impact of sampling strategy on drylot-housed steer: a) allogrooming bout duration (sec/bout), b) allogrooming bout frequency (bout/d), c) allogrooming total duration (sec/d), d) bar licking bout duration (sec/bout), e) bar licking bout frequency (bout/d), f) bar licking total duration (sec/d), g) tongue rolling bout duration (sec/bout), h) tongue rolling bout frequency (bout/d), i) tongue rolling total duration (sec/d), j) utilizing brush bout duration (sec/bout), k) utilizing brush bout frequency (bout/d), and l) utilizing brush total duration (sec/d). Different letters indicate statistical differences ($P < 0.05$) among sampling strategies.



Sampling Strategy Legend: ① = Five minutes decoded every 60 minutes (5EV60); ② = Five minutes decoded every 30 minutes (5EV30); ③ = Ten minutes decoded every 60 minutes (10EV60); ④ = Fifteen minutes decoded every 60 minutes (15EV60); ⑤ = Decoded from 14:00 to 16:00 (14to16); ⑥ = Ten minutes decoded every 30 minutes (10EV30); ⑦ = Fifteen minutes decoded every 30 minutes (15EV30); ⑧ = Decoded from 08:00 to 14:00 (8to14); ⑨ = Continuous

Figure 2

Strength of the relationship (r^2) between continuous observations (from 8:00 to 20:00) and each sampling strategy for a) allogrooming, b) bar licking, c) tongue rolling, and d) brush utilization for average bout duration (sec), frequency of bout per day, and total duration of all bouts (sec/d). The sampling strategies presented (left to right) are organized from shortest to longest duration of video that would need to be observed to collect the data.

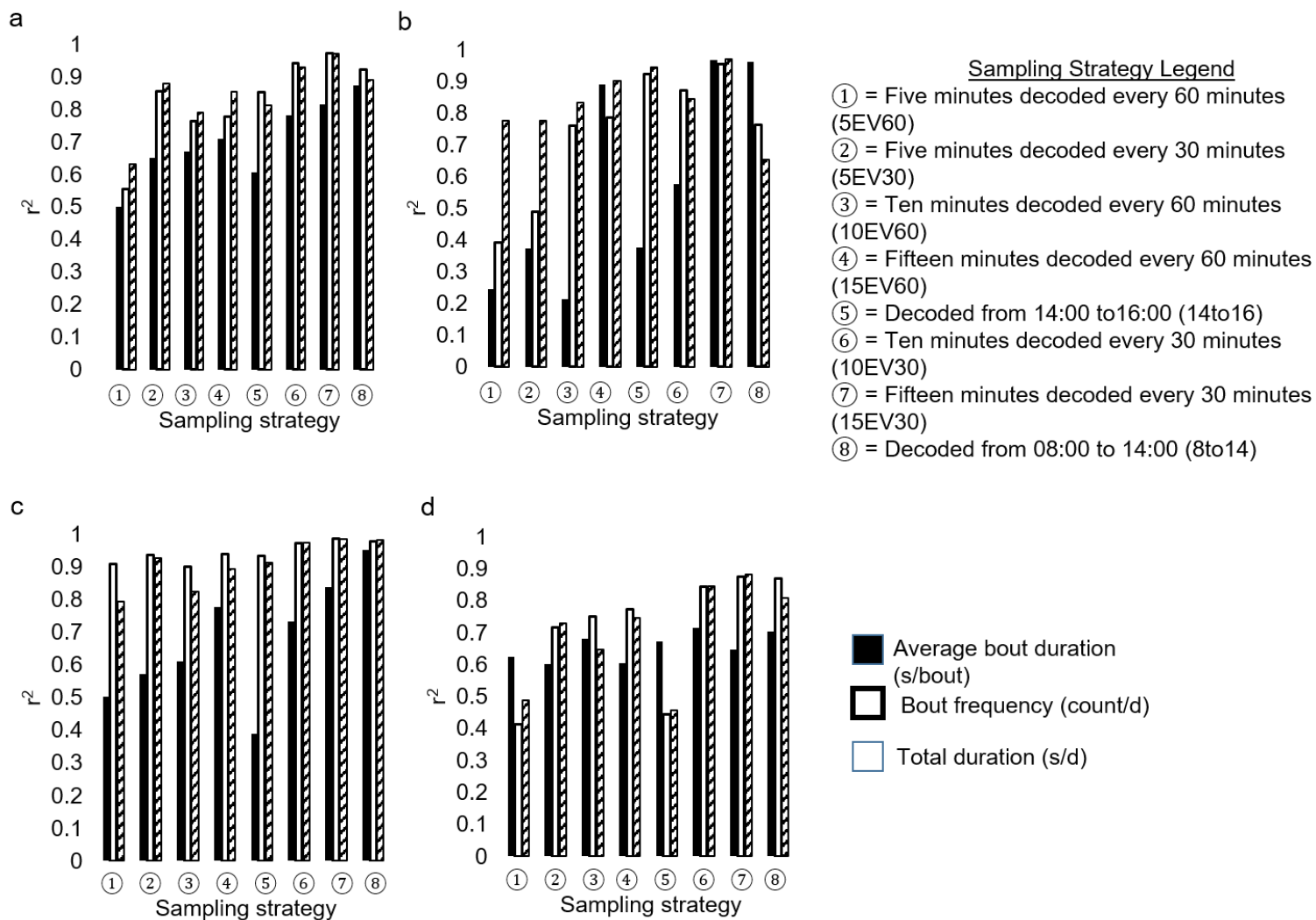


Figure 3

The strength of relationship between continuous observations and each sampling strategy when monitoring the daily frequency of drylot-housed steer engaged in a) mounting and b) head butting. Different letters indicate statistical differences ($P < 0.05$) among sampling strategies.

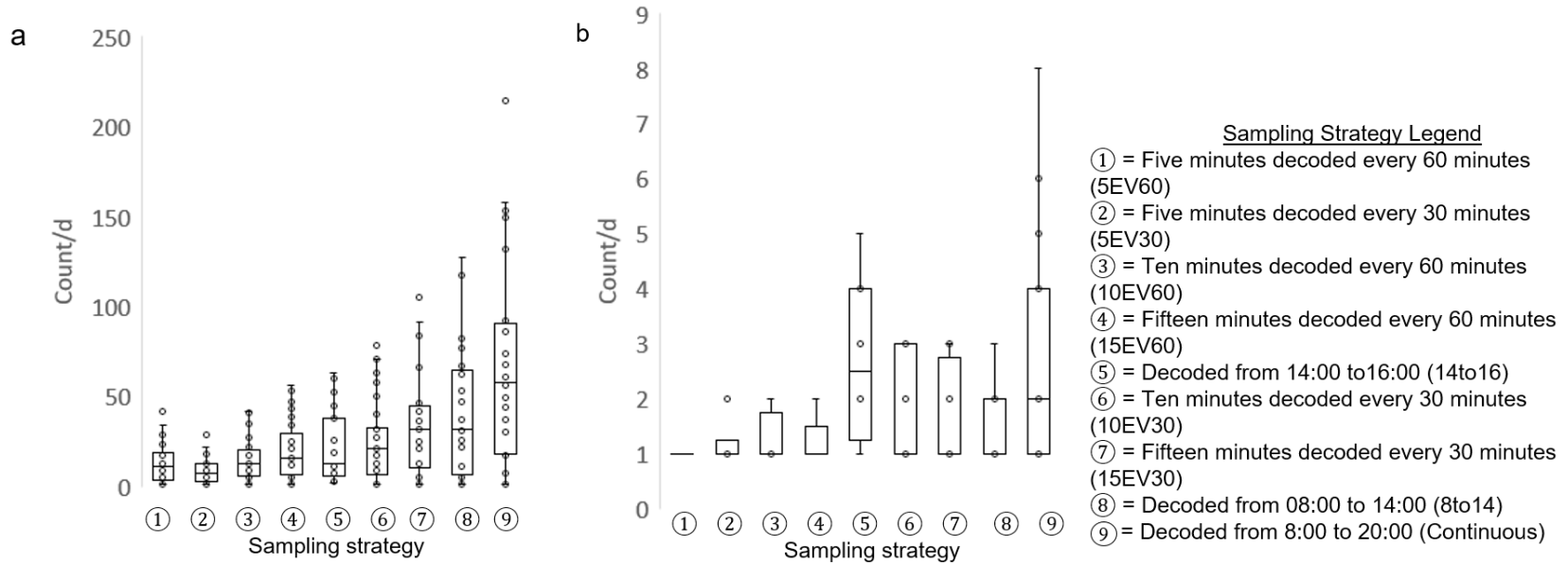
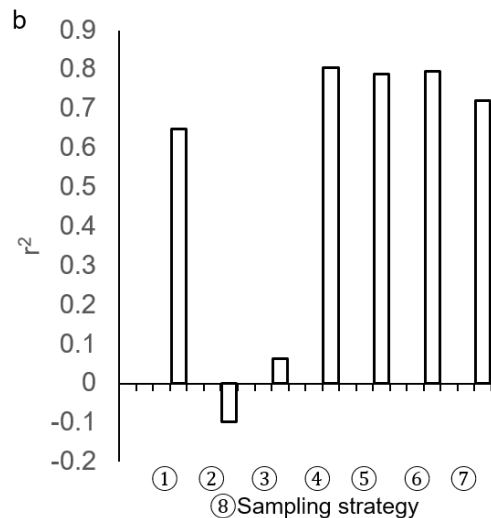
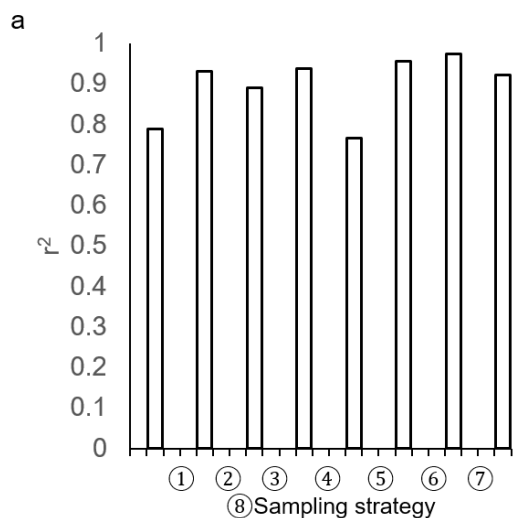


Figure 4

The strength of relationship between continuous observations (08:00 to 20:00) and each sampling strategy (indicated in circled numbers) when monitoring the daily frequency of drylot-housed steer a) mounting and b) head butting. The sampling strategies (indicated in circled numbers and described in the legend) presented are organized (left to right) from the shortest to longest duration of time that video recordings would need to be decoded to collect the data.



- Sampling Strategy Legend**
- ① = Five minutes decoded every 60 minutes (5EV60)
 - ② = Five minutes decoded every 30 minutes (5EV30)
 - ③ = Ten minutes decoded every 60 minutes (10EV60)
 - ④ = Fifteen minutes decoded every 60 minutes (15EV60)
 - ⑤ = Decoded from 14:00 to 16:00 (14to16)
 - ⑥ = Ten minutes decoded every 30 minutes (10EV30)
 - ⑦ = Fifteen minutes decoded every 30 minutes (15EV30)
 - ⑧ = Decoded from 08:00 to 14:00 (8to14)

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**SEX-DIFFERENCES IN GROUPS OF YEARLING *BOS INDICUS* SOCIAL,
STEREOTYPIC, BRUSH USE, AND FEEDING BEHAVIOR WHILE HOUSED IN
DRYLOTS WITH ACCESS TO A ROTATING MECHANICAL BRUSH**

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Introduction

Many agricultural animals are socially housed in counter-evolutionary single-sex groups. Cattle are prey animals that, through evolution, have organized themselves into herds of cows and calves with a matriarchal social structure (Hubbard et al., 2021). Bull calves will disperse from the herd once they reach puberty while the females will typically remain with the maternal herd. This sex-biased dispersal reduces the risk of inbreeding (Pusey et al., 2003), results in sex-differentiated social group structures, and facilitates the formation of strong social bonds within the maternal herd. However, this type of natural social dynamic is difficult to maintain and efficiently manage in production agriculture; thus, animal managers will regularly house cattle in non-natural social groupings.

Herd structures under contemporary cattle management systems do not necessarily mirror the natural herd structure of cattle. Upon arrival at the feedlot, cattle are typically divided into single-sex groups of similarly aged individuals to reduce the risk of unexpected pregnancies. Further, feed and nutrition requirements vary with age and sex, so separating the cattle increases the efficiency with which these needs can be met and makes animal management easier (Lalman & Richards, 2017). Therefore, while housing cattle in single-sex social groups may be counter-evolutionary, this management practice can yield welfare benefits.

In species across the animal kingdom, sex influences the prevalence and type of social interactions. Female dogs are more suspicious, excitable, sensitive, and less aggressive than males (Goddard & Beilharz, 1983). Male zebras are less exploratory but are more consistent in their exploratory behavior than females (Schuett & Dall, 2009). Female rhesus macaques engage in more social interactions than males and are more involved in grooming around the time of maturation (Kilik et al., 2015). Sex-based behavioral differences can also be observed in cattle. Dominance in a male herd is relatively unstable and involves more aggression compared with female hierarchies (Sowell et al., 1999). Male Holstein Friesian calves from 4 to 15 months of age are more likely to initiate social interactions than females with individuals of the same sex (Freslon et al., 2019), suggesting that sex may influence social interactions in cattle. Understanding these differences can facilitate the implementation of best management practices.

Male and female cattle differ in physiology and primary and secondary sex characteristics, including behavior. Growth hormone secretion is episodic in bulls and prepubertal heifers but ceases in females once they reach puberty (Campion et al., 1989; Ford & Klindt, 1989), and there have been numerous reports of sex differences in growth and carcass traits (Ray & Marchello, 1969; Tanner, 1970). The use of single-sex groups for research purposes is justifiable, yet this attempt to minimize the confounding factor of sex limits the generalizability of the results and leaves a knowledge gap. Thus, little empirical evidence exists that characterizes sex-specific differences in social behavior (Schuett, 2009).

Social bonding enhances individual fitness (Kulik et al., 2015), is integral to the social behavior of gregarious animals, varies at the individual level, and may be influenced by temperament. The performance of social behaviors can be influenced by the behavior of conspecifics (Schuett, 2009), including the behavioral and physiological fear response (Jones et

al., 1995; Perkins, 2016) and foraging efficiency and efficacy (Galef & Giraldeau, 2001). Group-living ungulates will often coordinate their behavior in time (synchronization) and in space; thus, many behaviors are performed due to social facilitation (Bailey et al., 2000) because most animals in the group will follow one or a few leaders (Sarova & Simecek, 2010). However, the strength of influence that social interactions have on specific individuals will vary due to differences in personality (Marchetti & Drent, 2000) and group size (Gygax et al., 2010). Thus, as groups become larger, the frequency of social interactions will increase, and the duration of each encounter will decrease, resulting in larger groups with many weak social bonds (Gygax et al., 2010). Monitoring these luxury behaviors (e.g., brush use, mounting, head butting, allogrooming, and bunk displacement) provides insight into individual animals' current social and reproductive statuses, but these luxury behaviors are also the first behaviors that are altered when time or energy resources are limited or animals are experiencing stress or discomfort (Mandel, 2013). Luxury behaviors can be influenced by environmental (e.g., infrastructure, stocking density) and managerial (e.g., social mixing, transportation) factors and can give insight into the animal's wellbeing.

Individual temperament may also influence the performance of social behaviors and, in cattle, can be objectively measured using the velocity with which they exit the handling chute (Fell et al., 1999). Temperament is defined as the reactivity of cattle to humans and novel environments (Fordyce et al., 1988) and is influenced by breed, gender, age, previous handling, and genetics (Burrow, 1997; Grandin, 1993). Cattle with faster exit velocities have greater basal concentrations of stress hormones, reduced growth rates, and lower average daily gains (Café et al., 2011; Fordyce et al., 1985), spend less time eating, and have lower dry-matter intake. Steers are typically calmer than heifers (Burrow, 1997, 2001). Therefore, the objective of this study was to characterize sex-

specific differences in social and stereotypic behaviors, brush use, docility, and feeding behavior performed by *Bos indicus* cattle housed in drylots.

Materials and Methods

Animals, Housing, and Management

Brahman yearling cattle (n = 63) were sorted into four single-sex (heifers: n = 2 pens; 15 heifers/pen; steers: n = 2 pens; 19 steers/pen) drylot pens (5 x 1.5 m) that provided 243 m² per heifer and 192 m² per steer, at Sexing Technology's Genetic Development Center (ST Genetics) in Navasota, Texas (Figure 5). Cattle were housed at the facility as part of an 85-day gain and development test during the summer of 2020. Each pen was fitted with a FutureCow ComfortBrush (Figure 5). Cattle were housed in these pens for at least 21 days prior to study commencement.

Diet and Feeding Behavior

Cattle were fed a moderate-energy diet with significant roughage content (Table 2) to promote maximum lean growth. To fulfill the animals' requirements, the diet was composed of a base concentrate mix that contained corn, protein meal, molasses, cottonseed hulls, vitamins, and minerals. The concentrate feed was mixed with silage to provide additional roughage, achieving a final diet of approximately 40% corn and 14% protein. The diet also contained Bovatec (Lasalocid, manufactured by Zoetis US), which reduces the risk of digestive disorders, improves gain and efficiency, and reduces the risk of coccidiosis.

Each pen was equipped with four electronic feed bunks (GrowSafe Systems, Airdrie, AB, Canada) that measured bunk visit duration (BV duration) (min/d), bunk visit frequency (BVF) (events/d), and dry matter intake (DMI) (kg/d).

Behavioral Observations

Social and Luxury Behaviors

Cattle were video recorded continuously for 71 days using a closed-circuit surveillance system (Safevant, Safesky, 1080P Isotect wireless security cameras). Video recordings from d 1, 2, 4, 8, 16, and 64 were decoded using continuous observations for the first 15 minutes of every 30-minute period as described in Lozada et al. (2021), from 8:00 to 20:00. The frequency and duration of luxury and stereotypic behaviors (Table 3) was recorded using BORIS (Version 7.9.15; Friad & Gamba, 2016).

Exit Velocity and Average Daily Gain

Exit velocities (EV), as described by Burrow and Dillon (1997) and Daigle et al. (2020) as the time taken for a heifer to travel 1.8 m, were measured with electronic timers (FarmTek, Inc., Wylie, TX), and body weights (BW) were collected on d -19, 0, 1, 25, 50, and 70. Individual average daily gain (ADG) and average exit velocity (EV) were calculated using the initial (d 0) and final (d 70) BW and EV.

Statistical analysis

Average bout duration (sec/bout), daily bout frequency (bout/day), and total duration of all bouts performed in a single day (sec/day) per behavior were calculated. Normality was evaluated using histograms in SAS version 9.04. A GLIMMIX model with a Poisson distribution and log transform link (to normalize the data) was used to evaluate differences between sexes for each behavior. The model included sex, sex within pen, and research day as fixed effects, and pen as the random effect. Exit velocity was evaluated in a separate GLIMMIX model that included sex, sex within pen, and research day as fixed effect, and the animal's individual ID as random effect.

Lastly, productivity and feeding behavior traits were evaluated using a GLIMMIX model that included sex as fixed effect and sex within pen as random effect.

Results

Steers performed social behaviors more frequently and spent more time engaged in social interactions than heifers (Figure 6). Steers had longer ($P < 0.005$) average bout durations (sec/bout) for allogrooming (heifers: 11.12 ± 1.15 , steers: 31.17 ± 2.05 ; Figure 6a), bar licking (heifers: 12.65 ± 1.2061 , steers: 19.71 ± 1.61 ; Figure 6b), and utilizing the brush (heifers: 114.92 ± 3.72 steers: 125.76 ± 3.85 ; Figure 6c). Further, steers spent more ($P < 0.005$) time per day (sec/d) engaged in allogrooming (heifers: 50.96 ± 3.08 , steers: 124.91 ± 5.69 ; Figure 6d), bar licking (heifers: 24.36 ± 1.49 , steers: 59.47 ± 3.04 ; Figure 6e), and utilizing the brush (heifers: 2439.13 ± 17.17 , steers: 3815.39 ± 21.63 ; Figure 6f). Steers performed allogrooming (heifers: 3.27 ± 0.84 , steers: 3.91 ± 0.93 ; Figure 7a), brush displacements (heifers: 1.08 ± 0.43 , steers: 1.86 ± 0.51 ; Figure 7b), bunk displacements (heifers: 57.87 ± 2.87 , steers: 71.38 ± 3.07 ; Figure 7c), head butts (heifers: 42.72 ± 2.42 , steers: 87.41 ± 3.25 ; Figure 7d), and brush use (heifers: 22.28 ± 1.62 , steers: 31.95 ± 1.97 ; Figure 7e) more frequently (bout/d) than heifers.

Heifers had greater exit velocities (heifers: 2.05 ± 0.08 , steers: 1.21 ± 0.07 ; Figure 8) than steers; had lower initial body weights ($P = 0.04$), lower final body weights ($P = 0.01$), and lower ADG ($P = 0.008$); were less efficient in converting feed to weight gain ($P = 0.04$); and visited the bunk more frequently than steers (Table 4). Heifers and steers did not differ in their total DMI, bunk visit duration, their eating rate during a bunk visit, nor the duration of time they spent with their head down in the bunk ($P > 0.05$).

Discussion

Compared to heifers, steers performed social and stereotypic behavior for longer periods of time, performed them for longer bout durations, and performed them more frequently, except for bar licking, which was performed at the same frequency by both sexes. Cattle are a mammalian species that practice sex-biased dispersal as part of their natural history. Selection pressures that have resulted in this phenomenon include kin competition, inbreeding avoidance, and spatiotemporal variation in resources or habitat suitability (Li & Kokko, 2018). In cattle, young bull calves will separate from the maternal herd in search of mates; thus, they will develop new dominance hierarchies that reflect their ability to access a mate. Heifers will remain with the maternal herd and will enter existing dominance hierarchies developed to dictate access to resources and spatiotemporal preferences. Thus, there are sex differences in the resources that social hierarchies are centered around, and there are differences in the social dynamics and spatial needs surrounding the development of these social scenarios. This has specific significance for *Bos indicus* cattle because they have been less influenced by human genetic selection pressures (Cooke et al., 2020); thus, the likelihood that they have retained the behavioral repertoire of their wild counterparts is high.

One of the factors that could be affecting the differences between the sexes could be the onset of puberty. The ontogeny of cattle biology dictates that cattle are experiencing puberty when they are approximately one year old (Gregory et al., 1991). Thus, the cattle in this study were undergoing sexual development, during which there are substantial sex-based differences in behavior and motivation, which could have influenced the results of this study. Further, heifers were housed in a more natural group setting compared to steers during this time, which may have contributed to the differences in the performance of stereotypic behaviors and use of the brush.

Stereotypic behaviors are indicative of frustration or an unmet behavioral need. At this age, steers would have been motivated to roam either in solitude or in small groups while maturing to access mates; thus, the reduced ability to roam and the increased intensity of social interactions may have caused steers to experience more frustration than heifers because performing exploratory behavior would promote their overall fitness (Wood-Gush, 1989). Steers may have also performed more social and stereotypic behaviors as part of the fundamental changes in their personalities and normal conditions accompanying the onset of puberty (Neave, et al., 2020), which can be more frustrating to heifers that are less motivated to engage in exploratory behaviors and geographically disperse. These results emphasize the importance of developing sex-specific management strategies and emphasizes the need for sex-specific considerations when housing cattle in captive environments, developing sensor technology, and validating algorithms to increase the accuracy of the data collected.

Steers may have had a stronger intrinsic need to perform exploratory behavior and engage in agonistic interactions, which may have contributed to the increased prevalence of social interactions and brush use. High metabolic rates, structural size, and body mass have been linked with relatively aggressive, bold, and exploratory behavior in cattle (Neave et al., 2020), and since the steers were larger and consumed more feed, they may have also engaged in more animal-environment interactions and increased the probability of interacting with a penmate. Further, the natural history of steer social dynamics suggests that steer social structures may be less stable than heifer social dynamics, which would result in an increased performance of affiliative (e.g., allogrooming) and competitive (e.g., head butt, bunk displacement, brush displacements) behaviors associated with social hierarchy development (Hubbard et al., 2021) thus contributing to the sex-based differences observed in social behaviors and brush use.

Steers performed allogrooming and brush use more frequently and in longer total durations than heifers. Body-care behavior such as grooming is ranked highly among maintenance behaviors in cattle (Kohari et al., 2007). Cattle need environmental amenities to facilitate caring for their bodies by scratching and licking to remove debris and ectoparasites (Fraser & Broom, 1990). Grooming using inanimate objects is part of environmental enrichment, which contributes to animal welfare (Pelley et al., 1995). Under grazing conditions, natural trees play an important role in grooming (Fraser & Broom, 1990), suggesting that cattle use inanimate objects for grooming areas that they are unable to reach during self-grooming (Kohari et al., 2007). Therefore, steers may have performed more allogrooming than heifers to fulfill their social and exploratory needs.

Tongue rolling was observed infrequently and was unable to be statistically analyzed. Throughout the entire research period, tongue rolling was performed eight times by the steers and once by a single heifer. The low prevalence of this specific behavior could have been influenced by diet or the presence of environmental enrichment (EE). Dietary roughage levels can impact the performance of tongue rolling (Ridge et al., 2020), and the presence of a cattle brush has been observed to reduce the prevalence of stereotypic behaviors (Meneses et al., 2021; Park et al., 2020). Thus, the environmental conditions or management strategies implemented during this study may have contributed to the reduced performance of this behavior.

Due to the fact that heifers were shown to be more temperamental and visited the feed bunk more frequently than steers, similar to Curley's (2006) research, EV is positively correlated with serum cortisol concentration in Brahman bulls, suggesting that EV is indicative of stress responsiveness of cattle to human interaction. One aspect to consider for the feed bunk frequency difference between steers and heifers is that heifers were housed in front of an office, and due to

their reactivity to human interaction, if any were close to the feed bunk, which was the closest part to the office, and someone came out of the office, all the animals would immediately go to the other end of the pen, which could have impacted the times they were at the feed bunk, as they had to leave and come back later when there were no humans close to the front of the pen.

Another aspect to consider is that more excitable animals appear to spend more time in a state of nervousness than their calmer counterparts (Burrow & Dillon, 1997), suggesting that they spend more time in a vigilant state and therefore are more likely to stop a feeding bout to examine their surroundings.

Steers were more productive than heifers. As expected, steers began and finished the trial with heavier body weights than heifers; had higher ADG, higher DMI, and lower F:G; and visited the bunk less frequently during the day (Scheiber et al., 1987). Literature focusing on sex differences in automatically measured feeding behavior is scarce, yet an animal's feeding behavior is generally consistent, repeatable, and related to temperament and may be used to predict differences in animal performance and efficiency (Nkruman et al., 2007). Cattle are highly motivated to access brushes (McConnachie et al., 2018), their use follows a circadian rhythm (Meneses et al., 2021), and beef calves with access to brushes have been shown to perform more play and social behaviors compared to calves housed in non-enriched pens (Bulens et al, 2014). Furthermore, second-lactation dairy cows with access to an automated brush had a 3.5% increase in milk yield (Schukken & Young, 2009), which aligns with the results of this research because steers are more productive, use the brush the most, and perform more social behavior than heifers.

Conclusion

Producers have anecdotally reported sex differences in beef cattle behavior, but to our knowledge, no empirical evidence has reported such differences except from cattle sex differences in productivity. In the current study, after evaluating cattle sex differences on temperament, social behavior, stereotypic behavior, feeding behavior and productivity, differences in almost all traits were observed between steers and heifers housed in drylots. These findings show their relevance by providing novel information on how different these creatures are, even though they look so similar on the outside. Consequently, this knowledge can be used to develop sex-specific management strategies that could help the animals better cope with the environment. Also, it shows the importance of adding sex-specific algorithms when developing technology to automatically measure cattle behavior, because if they are compared side-by-side, differences will be observed, but results could bias the trait of interest at the moment. The same happens for research, meaning that sex should always be accounted for irrespective of the trait evaluated. Accounting for sex differences in cattle is also very important due to the increasing population that we try to feed every day, which has made sustainability a popular concept in the past years. If we can find ways to better understand our animals and to improve their welfare on an individual basis and not just based on a specific species, sustainability will be positively impacted. Lastly, other than that, as we see in nature, there are many physical differences in animals between male and female (e. g., peacocks), but in cattle, such differences are not so prominent. Therefore, males have to perform a set of behaviors different from females to be able to stand out from a group and find mates. These traits are very interesting to little by little start to understand, and this quantifiable evidence of some of these behavioral differences will encourage further research to investigate the physiological reasons for the traits to differ.

Table 2.

Dietary ingredient and chemical compositions of the two diets that were fed to both heifers and steers throughout the trial.

	Heifers	Steers
Ingredient composition, % as-fed		
Sorghum Silage	47	40
Premix ¹	43	52.5
Bermuda Hay	10	7.5
Chemical analysis, % DM		
DM %	62.43	66.41
CP %	13.43	14.25
NDF %	44.16	42.82
NEm, Mcal/Kg	71.25	74.68
NEg, Mcal/Kg	43.24	46.41

¹Premix contained minimum 0.75% Ca, 79.09 ppm Zn, 82.72 ppm Mn, 0.33 ppm Se, 29.15 ppm Co.

Table 3.

Ethogram of social and luxury behaviors decoded from video observations of Brahman steers (n = 19) and heifers (n = 15) during a 85-day gain test. Adapted from Park et al., 2020.

Behavior	Definition
Allogrooming	Mouth of one cow is continuously licking the body of another. (The bout is considered finished after the action has stopped for 10 s.)
Head Butt	Head of one cow connects forcefully with the body of another.
Bunk Displacement	One cow pushes another cow out of the feed bunk.
Brush Use	Any part of a heifer's body interacts with the brush. (The bout is considered finished after the action has stopped for 10 s.)
Bar Licking	One cow is continuously licking or biting the bars around the pen. (The bout is considered finished after the action has stopped for 10 s.)
Tongue Rolling	Tongue outside the mouth is continuously moving from one side to the other. (The bout is considered finished after the action has stopped for 10 s.)

Table 4.

Mean (\pm SEM) of Performance and feeding behavior traits that were collected using electronic feedbunks for pens of yearling Bos indicus steers and heifers

Trait	Heifers	Steers	P-value
No. of animals	27	37	
Performance Traits			
Initial BW (Kg)	402.57 \pm 23.37	500.68 \pm 29.93	0.042
Final BW (Kg)	485.96 \pm 21.57	611.99 \pm 27.58	0.011
ADG (Kg/d)	0.96 \pm 0.05	1.28 \pm 0.06	0.008
F:G	11.76 \pm 0.56	9.37 \pm 0.71	0.038
DMI (Kg/d)	10.76 \pm 0.27	11.68 \pm 0.34	0.077
Feeding Behavior Traits			
BV frequency (events/d)	70.98 \pm 3.34	42.29 \pm 4.19	0.002
BV duration (min/d)	112.93 \pm 10.49	97.94 \pm 13.47	0.414
BV eating rate (g/min)	136.88 \pm 12.39	164.39 \pm 15.86	0.221
HD duration (min/d)	65.10 \pm 10.04	55.34 \pm 12.89	0.572
HD duration per BV duration	0.56 \pm 0.05	0.55 \pm 0.07	0.937

¹Individual body weights (BW) were collected on d -19, 0, 1, 25, 50, and 70, and were used to calculate, initial BW, final BW, ADG, F:G, and DMI.

²Each pen was equipped with four electronic feed bunks (GrowSafe Systems, Airdrie, AB, Canada) that measured bunk visit duration (BV duration), bunk visit frequency (BV Frequency), and dry matter intake (DMI) on a daily basis (85 days).

Figure 5.

Diagram of ST Genetics pens, with environmental enrichment provided (CowComfort Cow Brush from FutureCow)

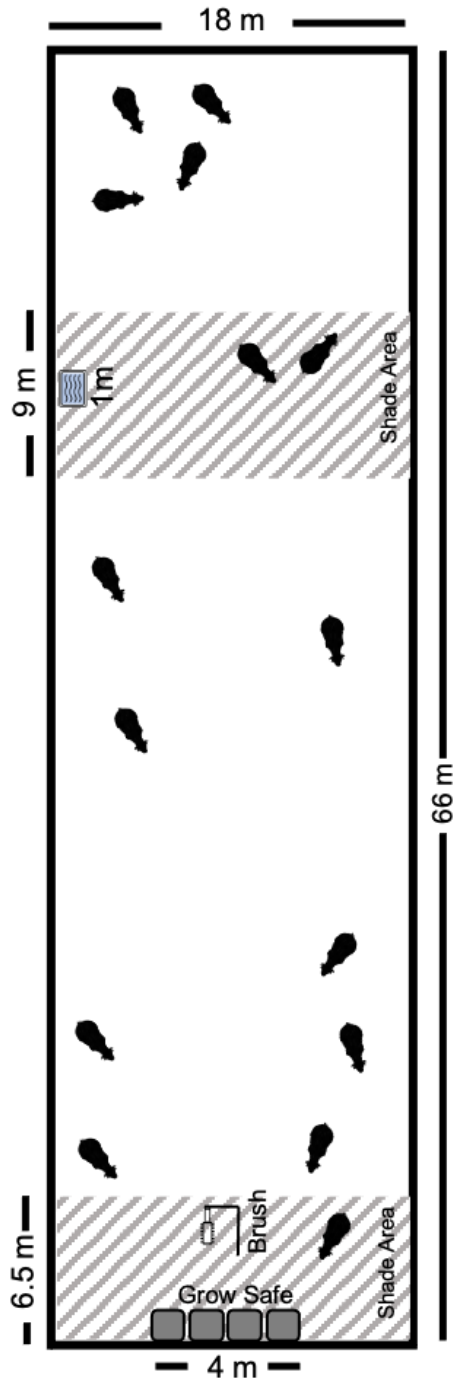


Figure 6.

*Average bout duration (sec/d) and total duration of time spent per day (sec/d) that steers (S) and heifers (H) spent engaged in allogrooming, bar licking, and utilizing a rotating mechanical brush as measured using continuous observation 15 minutes every 30 minutes from 08:00 to 20:00 h. *P < 0.05 between sexes*

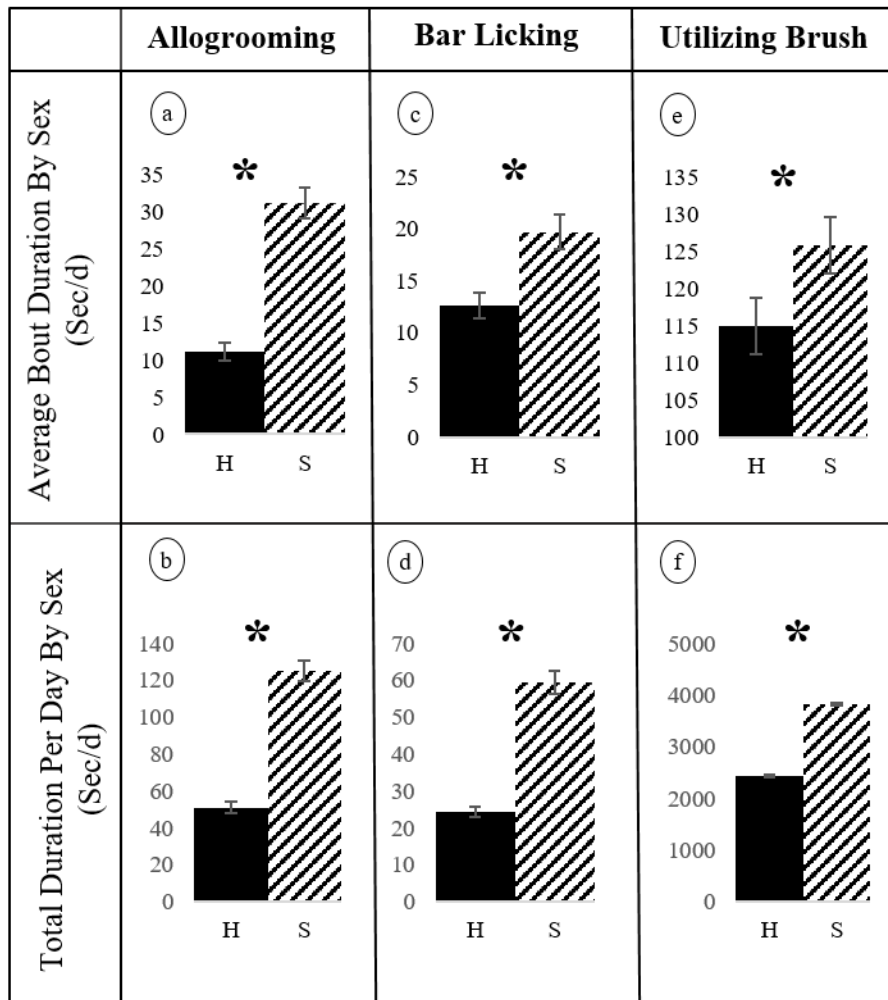


Figure 7.

*Frequency that steers (S) and heifers (H) performed allogrooming, brush displacement, bunk displacement, head butt, utilizing a rotating mechanical brush, and bar licking bouts as measured using continuous observation 15 minutes every 30 minutes from 08:00 to 20:00 h. *P < 0.05 between sexes*

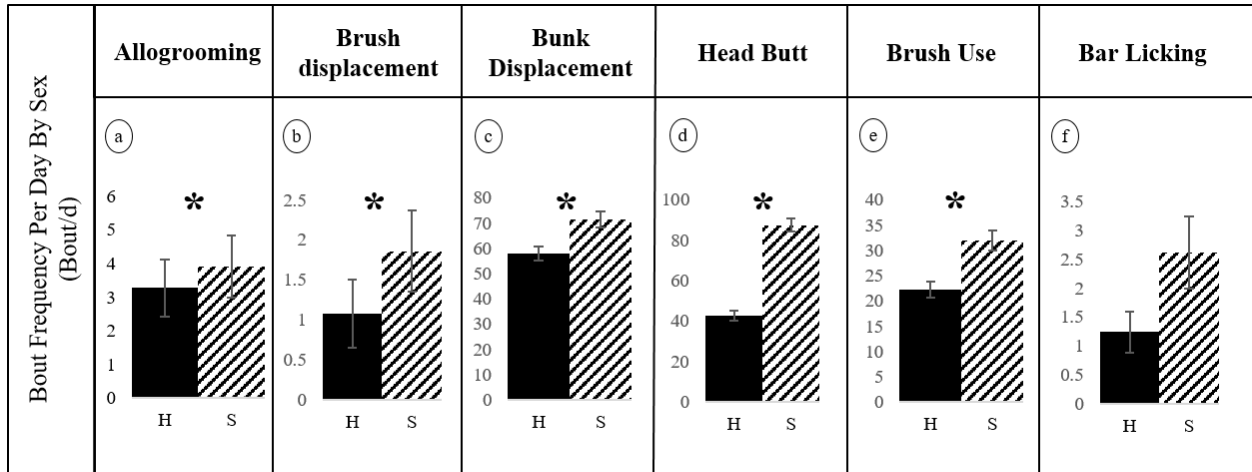
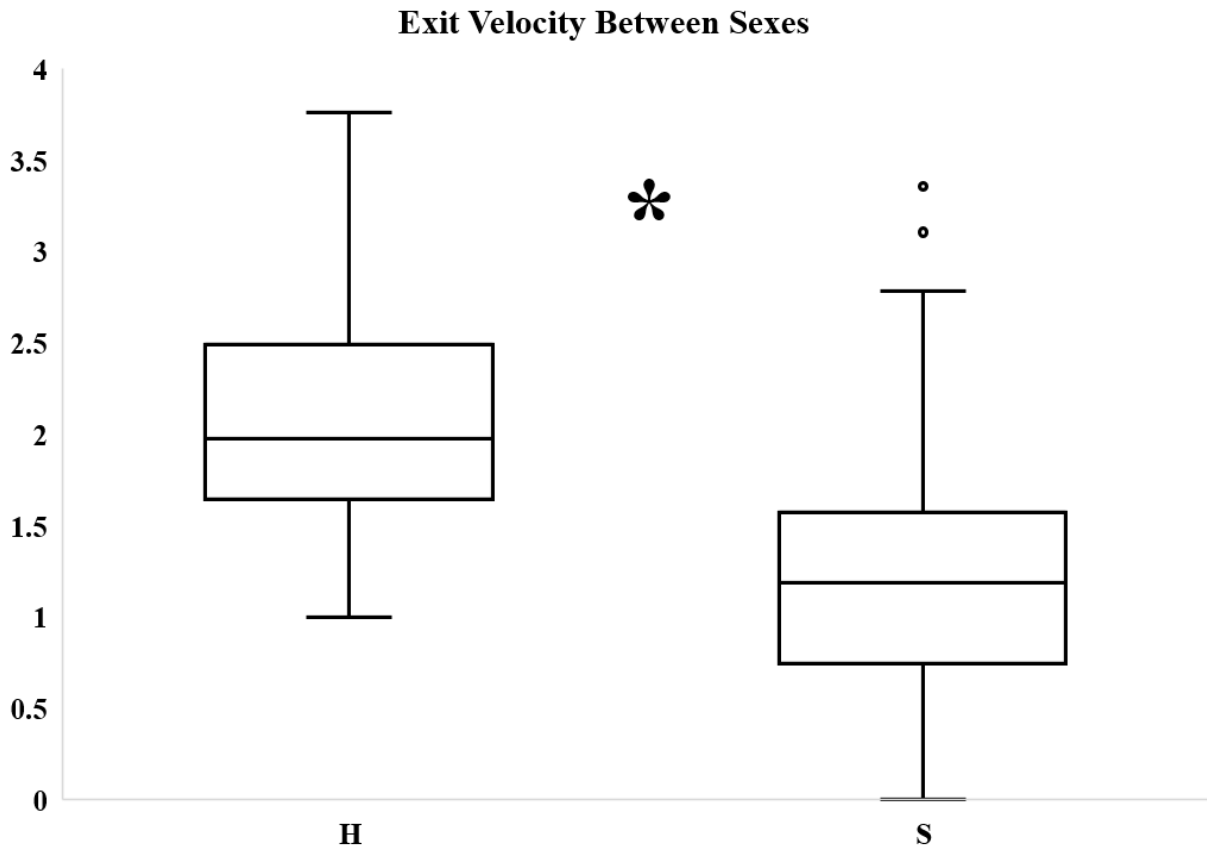


Figure 8.

Average exit velocity (EV^1 , m/s) of steers (S) and heifers (H) that were evaluated on d -19, 0, 1, 25, 50, and 70. * $P < 0.05$.



¹EV, as described by Burrow and Dillion (1997) and Daigle et al. (2020), was calculated using the time it took an individual steer or heifer to traverse 1.8m.

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**EVALUATING THE UTILITY OF INDIVIDUAL CATTLE BRUSH USE BEHAVIOR
AS A NOVEL BEHAVIORAL PHENOTYPE REGARDING PRODUCTIVITY,
TEMPERAMENT, AND FEEDING BEHAVIOR OF BOS INDICUS STEERS AND
HEIFERS HOUSED IN DRY LOTS**

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Introduction

The increasing integration of environmental enrichment (EE), as a measurable resource, into captive animal management presents an opportunity to develop quantifiable behavioral phenotypes regarding the performance of pleasurable behaviors. Brushes are an effective EE for cattle (Park et al., 2020; Miltohner et al., 2001). Cattle are highly motivated to access brushes (McConnachie et al., 2018), their use follows a circadian rhythm (Meneses et al., 2021), and beef calves with access to brushes have been shown to perform more play and social behaviors compared to calves housed in non-enriched pens (Bulens et al, 2014); thus, integrating behavioral phenotypes regarding brush use can increase the robustness of our approach to disease detection and selection for behavioral phenotypes.

Mechanical rotating brushes positively influence cattle welfare by increasing the complexity of the animal's environment and reducing boredom (Park et al., 2020; Bracke et al., 2006). Furthermore, second-lactation dairy cows with access to an automated brush had a 3.5% increase in milk yield (Schukken & Young, 2009). Thus, monitoring the use of this resource, the

brush, presents an opportunity to examine the effect of divergent behavioral phenotypes for brush usage on temperament, productivity, and feeding behavior patterns, thus characterizing behavioral phenotypes of individuals that engage in pleasurable behaviors as associated with a positive affect.

Feeding behavior and temperament are interrelated and can be objectively evaluated as behavioral phenotypes relevant to productivity, product quality, and efficiency (Fordyce and Goddard, 1984; King et al., 2006; Struthers, et al., 1997; Voisinet et al., 1997). Individual feeding behavior can be used to evaluate feed efficiency (Lancaster et al., 2009) and reflect health status (Quimby et al., 2001). Technological advancements have made collecting individual animal feeding data possible. Electronic radio frequency identification-based systems such as GrowSafe System, Ltd., are designed to measure feeding behavior traits in beef cattle such as dry matter intake (DMI), bunk visit frequency (BV frequency), bunk visit duration (BV duration), bunk visit eating rate (BV eating rate), head down duration (HDD) with a high degree of accuracy (Mendes, 2011), thus providing a mechanism for automatically and non-invasively measuring individual animal feeding behavior.

Monitoring a suite of feeding behaviors is one of the most accurate methods of detecting sickness in cattle and can inform genetic selection decisions regarding efficiency (Kelly et al., 2020; Quimby et al., 2001; Svensson & Jensen 2007). While changes in feeding behavior, activity, physiological responses and social interactions have been determined to be useful as an early indication of disease before clinical diagnosis (Buhman et al., 2000; Pillen et al., 2016; Toaff-Rosenstein et al., 2016), evaluating a combination of behavioral traits may be more effective in detecting behavioral repertoire changes than focusing on a single behavioral trait alone (Kayser et al. 2018). However, many cattle may become sick prior to altering their feeding behavior, thus reducing the efficacy of this approach. To further complicate animal management, animals with

excitable temperaments have a stronger anti-predator response, resulting in more excitable animals being better adept at masking injury and disease. This highlights the importance of monitoring behaviors associated with exploration, pleasure, and comfort.

Materials and Methods

Animals, Housing, and Management

Brahman yearling cattle (n = 63) were sorted into four 5 x 1.5 m drylot pens (Heifers: n = 2 pens, 15 heifers/pen; Steers: n = 2 pens, 19 steers/pen) that provided 243 m² per heifer and 192 m² per steer, at Sexing Technology's Genetic Development Center (ST Genetics) in Navasota, Texas (Figure 9). Cattle were housed at the facility for a 85-d gain and development test that was conducted during the summer of 2020. Each pen was fitted with a FutureCow ComfortBrush. Cattle were housed in these pens for at least 21 d prior to study commencement.

Ten animals from each pen were marked with a color ear tag for individual identification and subsequent focal behavioral observation. These animals were selected based upon the exit velocity (EV) and body weights (BW) collected on d -19. Within each pen, animals were divided into three groups (high, medium, low EV) based upon +/- 2 SD from the EV mean. Within each EV group, cattle were sorted by BW. The lightest and the heaviest animals from each group were selected, and an additional animal was randomly selected from the middle. For the medium EV group, four animals were selected evenly across the distribution of animals in each group.

Diet and GrowSafe

Heifers were fed a moderate-energy diet with significant roughage content. This diet was fed with the intention of promoting maximum lean growth and meeting nutritional requirements. The diet was composed of a base concentrate mix (Table 5) that contained corn, protein meal,

molasses, cottonseed hulls, vitamins, and minerals. The concentrate feed was mixed with silage to provide additional roughage, achieving a final diet of approximately 40% corn and 14% protein. The energy level of the diet was intended to achieve a daily gain of 1.5 kg. The diet also contained Bovatec (Zoetis), which reduces the risk of digestive disorders, improves gain and efficiency, and reduces the risk of coccidiosis.

A GrowSafe 4000E system was used for this study to monitor individual animal feeding behavior. Each pen was equipped with four feed bunks that used an antenna to detect the animal's presence and load cells to measure individual feed intake by measuring feed disappearance during each bunk visit. Each bunk was equipped with neck bars to allow only one animal to enter the bunk at a time, and data acquisition software (GrowSafe DAQ; v.6.25), recorded all feeding behavior intake data. The GrowSafe system was designed to monitor feeding behavior by continuously recording the presence of an animal at the bunk through an electronic identification (EID).

Temperament Assessment and Average Daily Gain

Exit velocities (EV), as described by Burrow and Dillon (1997) and Daigle et al. (2020) as the time taken for a heifer to travel 1.8 m, was measured with electronic timers (FarmTek, Inc., Wylie, TX), and body weights (BW) were collected on d -19, 0, 1, 25, 50, and 70. Individual average daily gain (ADG) and average exit velocity (EV) were calculated using the initial (d 0) and final (d 70) BW and EV.

Brush Use

Cattle were video recorded continuously for 85 days using a closed-circuit surveillance system (Safevant, Safesky, 1080P Isotect wireless security cameras). Video recordings from d 1, 2, 4, 8, 16, 32, and 64 were decoded using continuous observations for the first 15 minutes of every

30-minute period beginning at 8:00 and continuing until 20:00. The frequency and duration of brush use for each focal individual was recorded using BORIS (Version 7.9.15; Friad & Gamba, 2016).

Statistical Analysis

Animals were classified into one of three phenotypes based on $\pm 1/2$ SD from the brush usage mean. Using SAS version 9.04, a GLIMMIX model was evaluated with sex, brush use phenotype, and the interaction between brush usage phenotype and sex as fixed effects and pen as random effect. The traits evaluated were initial body weight (IBW, kg), final body weight (FBW, kg), average daily gain (ADG, kg/d), dry matter intake (DMI, kg/d), feed-to-gain ratio (F:G, kg), bunk visit frequency (BVF, events/d), bunk visit duration (BVD, min/d), bunk visit eating rate (BVER, g/min), head down duration (HDD, min/d), head down duration per bunk visit duration ($HDD/BVD = \text{min}$), and exit velocity ($EV = \text{m/sec}$).

Results

Sex had an effect on all the performance traits ($P < 0.05$) and EV ($P = 0.010$), where steers showed to be more productive than heifers, and heifers more temperamental than steers (Table 6). For the feeding behavior traits, sex only had an effect on BV frequency ($P = 0.015$) and BV eating rate ($P = 0.029$). On the other hand, high brush use animals had 24% higher ADG ($P = 0.020$) than low brush use animals (Table 7). Lastly, an interaction between sex and brush use phenotype was observed for DMI ($P = 0.0407$), where high and medium brush use differed from each other, steers showing a higher DMI than heifers (Table 6); but low brush use steers and heifers did not show any differences.

Discussion

Sex differences were observed ($P < 0.05$) in all performance traits and some feeding behavior traits as expected from Chapter 2, which showed that steers used the brush more and were more productive than heifers. The presence of brushes can influence social interactions within the herd and influence the animal-environment interaction (Park et al., 2020). Cows will groom themselves (by licking) and will participate in allogrooming with herd mates, which according to Boissy et al. (2007) could show preferential relationships between group members, reduce tension in groups of animals, and play a major role in achieving a positive mood in animals, providing short- and long-term benefits. This positive effect of the brush on cattle behavior could lead to animals that are less stressed and spend more time engaging with pleasurable behaviors, which is beneficial for productivity. Even though the relationship between brush use and DMI is inconsistent between research studies, where some researchers say that the provision of brush did not affect DMI on Holstein calves (Horvath et al., 2020), in this research we noticed that brush use had a greater impact of steers' DMI than heifers'. The high brush use animals had higher DMI than the low brush use.

The negative effects of behavioral restriction from housing, where animals are deprived from all their natural behaviors, presents a risk of decreasing productivity (Ninomiya, S 2014). Therefore, authors such as Bolt and George (2019) believed that providing EE to cattle benefits animal welfare and productivity and promotes a positive message to the public that the animals are provided with optimum environments. In addition, Keeling et al. (2016) found that in dairy cattle, brush use is associated with higher milk yield and higher roughage intake, while Park et al. (2020) reported that in feedlot cattle, the brush did not compromise productivity. On the other hand, in pigs, Rodarte et al. (2004) found that growing piglets with access to EE had

significantly higher weight gain than piglets with no EE. However, in research done in pigs by Beattie et al. (1995) and Fàbrega et al. (2019), even though EE reduced agonistic behaviors and induced exploratory behavior, EE did not influence productivity. However, in this research, we found that even though only a trend was observed on brush use for F:G, high brush use animals seem to be more productive by showing a higher ADG.

On the other hand, exit velocity, an objective metric of the “fight-or-flight” response, was affected by sex, but not by brush use phenotype.

No effect of brush use was observed in EV, but it did seem like high brush use animals were more productive. This indicates that the exit velocity and brush use are evaluating different components of an animal’s temperament. According to Grandin (1997), temperament constitutes an animal’s excitatory or inhibitory reactions, motor activity levels, habits, emotions, and alertness. More excitable temperaments, with more arousal and fearfulness behaviors in livestock, have been associated with reduced performance, health, and carcass quality (Norris, 2014). Therefore, temperament, as a characteristic that varies at the individual animal level and influences productivity, has increasingly become a focus of many studies hoping to find ways to improve animal welfare (Norris, 2014) and productivity at the same time. A reliable way of measuring temperament is by measuring individual exit velocity (EV). Burrow and Dillon (1997) found that *Bos indicus* crossbred steers with a slow EV gained more weight and had heavier carcasses than the ones with a faster EV. After the United Nations reported that the global population will be approaching 9.7 billion by 2050 (UN, 2015), efficiency is a priority for all food producers. In this study, we showed that high brush use animals are more productive than medium and low brush use animals. This shows that pleasurable behavior can be used as a new phenotype to select for productivity, and if we can consider more factors than just the fear

response for productivity, we can become more accurate and more proactive at achieving the much-needed productivity to feed the increasing population, by making cattle a more cost-effective protein source.

Regarding the feeding behavior traits, sex had an effect on BV frequency, where heifers visited the bunk more frequently than steers, and BV eating rate, where steers eat faster than heifers. Feeding behavior of beef cattle is influenced by a wide variety of external factors, including weather (Schwartzkopf-Genswein et al., 2003), animal temperament (Voisinet et al., 1997), animal health (Wolfger et al., 2015), and animal management practices, such as bunk space and bunk management (Oolson et al., 2019), yet the full effect of beef cattle feeding behavior is not well understood yet (Schwartzkopf-Genswain et al., 2003, 2011). This could be the reason that brush use did not directly affect these traits as much as productivity traits.

Table 5.

Dietary ingredient and chemical compositions of the two diets that were fed to both heifers and steers throughout the trial.

	Heifers	Steers
Ingredient composition, % as-fed		
Sorghum Silage	47	40
Premix ¹	43	52.5
Bermuda Hay	10	7.5
Chemical analysis, % DM		
DM %	62.43	66.41
CP %	13.43	14.25
NDF %	44.16	42.82
NEm, Mcal/Kg	71.25	74.68
NEg, Mcal/Kg	43.24	46.41

¹Premix contained minimum 0.75% Ca, 79.09 ppm Zn, 82.72 ppm Mn, 0.33 ppm Se, 29.15 ppm Co.

Table 6

Mean (\pm SEM) of performance, temperament and feeding behavior in yearling Brahman (*Bos indicus*) steers (S) and heifers (H) as a function of the interaction between sex and brush use phenotype.

Trait	High		Medium		Low		P-value
	H	S	H	S	H	S	
No. of animals	7	5	7	9	4	5	
Total brush use (min/d)	8.97 \pm 0.69	14.21 \pm 1.27	5.59 \pm 0.28	7.28 \pm 0.59	2.09 \pm 0.64	2.01 \pm 0.92	
Performance Traits¹							
Initial BW (Kg)	425.55 \pm 23.94	556.27 \pm 27.83	415.31 \pm 23.95	554.65 \pm 21.83	445.64 \pm 30.26	549.95 \pm 27.83	0.754
Final BW (Kg)	505.98 \pm 27.28	699.09 \pm 30.91	491.27 \pm 27.28	660.79 \pm 25.47	525.38 \pm 33.02	636.12 \pm 30.91	0.268
ADG (Kg/d)	0.91 \pm 0.09	1.60 \pm 0.11	0.88 \pm 0.09	1.22 \pm 0.08	0.87 \pm 0.12	1.02 \pm 0.11	0.056
DMI (Kg/d)	10.50 ^{ab} \pm 0.47	13.59 ^c \pm 0.55	9.99 ^a \pm 0.47	12.01 ^{bc} \pm 0.41	10.52 ^{ab} \pm 0.62	10.68 ^{abc} \pm 0.55	0.041
F:G	12.40 \pm 0.87	8.57 \pm 1.03	11.46 \pm 0.87	10.16 \pm 0.77	12.52 \pm 1.15	10.71 \pm 1.03	0.359
Feeding Behavior Over Trial²							
BV frequency (events/d)	83.85 \pm 10.24	40.49 \pm 11.35	76.54 \pm 10.24	39.78 \pm 9.72	60.94 \pm 11.97	44.19 \pm 11.35	0.278
BV duration (min/d)	94.55 \pm 7.78	98.93 \pm 9.21	90.76 \pm 7.78	102.04 \pm 6.86	117.85 \pm 10.29	88.29 \pm 9.21	0.069
BV eating rate (g/min)	149.82 \pm 12.89	185.42 \pm 15.25	156.92 \pm 12.89	163.02 \pm 11.36	120.23 \pm 17.05	164.48 \pm 15.25	0.346
HD duration (min/d)	48.45 \pm 6.67	47.97 \pm 7.89	54.04 \pm 6.67	48.22 \pm 5.89	47.74 \pm 8.83	33.90 \pm 7.89	0.699
HD duration per BV duration	0.50 \pm 0.05	0.48 \pm 0.06	0.57 \pm 0.05	0.48 \pm 0.04	0.42 \pm 0.06	0.37 \pm 0.05	0.778
Temperament							
Exit Velocity (m/s) ³	1.91 \pm 0.18	1.23 \pm 0.21	1.96 \pm 0.18	1.21 \pm 0.16	1.84 \pm 0.23	0.86 \pm 0.21	0.755

¹Individual body weights (BW) were collected on d -19, 0, 1, 25, 50, and 70, and were used to calculate, initial BW, final BW, ADG, F:G, and DMI.

²Each pen was equipped with four electronic feed bunks (GrowSafe Systems, Airdrie, AB, Canada) that measured bunk visit duration (BV duration), bunk visit frequency (BV Frequency), and dry matter intake (DMI) daily for 85 d.

³EV, as described by Burrow and Dillion (1997) and Daigle et al. (2020), was calculated using the time it took each steer and heifer to traverse 1.8m.

Table 7

*Mean (\pm SEM) of performance, temperament and feeding behavior in yearling Brahman (*Bos indicus*) cattle for brush use phenotype*

Trait	High	Medium	Low	P-value
No. of animals	12	16	9	
Total brush use (min/d)	11.15 \pm 1.00	6.54 \pm 0.41	2.04 \pm 0.55	
Performance Traits¹				
Initial BW (Kg)	489.70 \pm 16.93	486.04 \pm 14.89	496.86 \pm 19.28	0.89
Final BW (Kg)	597.51 \pm 18.08	577.33 \pm 16.01	578.98 \pm 20.48	0.63
ADG (Kg/d)	1.24 ^a \pm 0.08	1.05 ^{ab} \pm 0.07	0.94 ^b \pm 0.09	0.05
DMI (Kg/d)	11.95 \pm 0.34	11.01 \pm 0.33	10.51 \pm 0.44	0.05
F:G	10.62 \pm 0.67	10.87 \pm 0.58	11.64 \pm 0.77	0.60
Feeding Behavior Over Trial²				
BV frequency (events/d)	63.51 \pm 7.01	57.79 \pm 6.64	53.3 \pm 7.73	0.48
BV duration (min/d)	96.29 \pm 6.31	97.16 \pm 5.46	101.49 \pm 7.26	0.85
BV eating rate (g/min)	166.73 \pm 9.92	158.79 \pm 8.58	143.43 \pm 11.42	0.32
HD duration (min/d)	47.74 \pm 5.02	51.14 \pm 4.34	40.39 \pm 5.77	0.34
HD duration per BV duration	0.49 \pm 0.04	0.52 \pm 0.03	0.39 \pm 0.04	0.06
Temperament				
Exit Velocity (m/s) ³	1.56 \pm 0.13	1.59 \pm 0.11	1.34 \pm 0.15	0.394

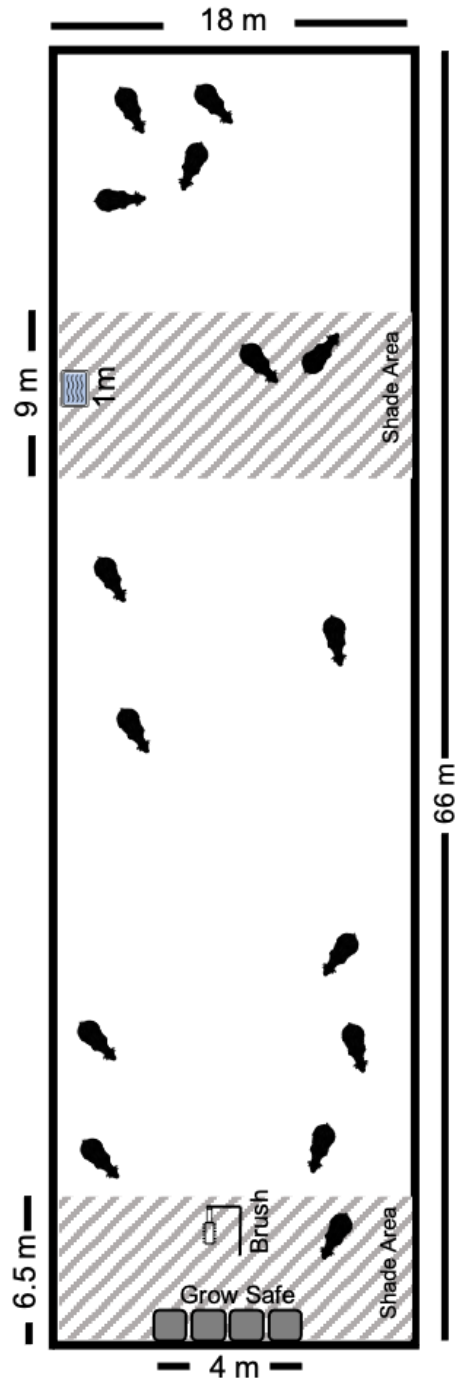
¹Individual body weights (BW) were collected on d -19, 0, 1, 25, 50, and 70, and were used to calculate, initial BW, final BW, ADG, F:G, and DMI.

²Each pen was equipped with four electronic feed bunks (GrowSafe Systems, Airdrie, AB, Canada) that measured bunk visit duration (BV duration), bunk visit frequency (BV Frequency), and dry matter intake (DMI) daily for 85 d.

³EV, as described by Burrow and Dillion (1997) and Daigle et al. (2020), was calculated using the time it took each steer and heifer to traverse 1.8m.

Figure 9.

Diagram of ST Genetics pens, with environmental enrichment provided (CowComfort Cow Brush from FutureCow)



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DISCUSSION

Observing cattle behavior for 15 minutes every 30 minutes was shown to be an optimal sampling strategy for evaluating daily bout frequency, average bout duration (sec), and total duration of all bouts (sec) of social behavior, stereotypic behavior, and brush use in beef cattle housed in drylots. This sampling strategy provides insight as to what happens throughout the day and is designed to capture behaviors that happen infrequently or in short durations (Daigle & Siegford, 2014). On the other hand, observing the animals from 14:00 to 16:00 was shown to be the least accurate sampling strategy. That could be due to the circadian pattern of cattle behavior. For example, behaviors such as brush use, mounting, and allogrooming tend to be performed at lower frequencies during the morning than the afternoon (Meneses et al., 2021). If the time of day can influence when the behaviors are expected to occur, then samplings should be distributed throughout the day; otherwise, the accuracy of the sampling strategy could be negatively impacted.

These findings provide guidance regarding how to expedite large-scale behavior observations that optimize the collection of cattle social behavior data. The results from this study could be used to design ways to extend the battery life of sensor technology. If a sensor only needs to turn on for 15 minutes every 30 minutes, then the sensor can have a longer battery life and generate smaller yet equally informative datasets, making data management and output easier to use and resulting in increased adoption and greater economic returns for the producer. This research provides several options, depending on the percentage of accuracy desired by the observer, which could be used in the interest of observing the animal's behavior, optimizing social behavior observation, and offering flexibility to the researcher.

Consequently after using this sampling strategy, compared to heifers, steers performed social and stereotypic behavior for longer periods of time, longer bout durations, and more frequently, except for bar licking that there was no difference in the frequency of the behavior. Some aspects that can be influencing these results are that cattle are a mammalian species that practices sex-biased dispersal as part of their natural history. Selection pressures that have resulted in this phenomenon include kin competition, inbreeding avoidance, and spatiotemporal variation in resources or habitat suitability (Li and Kokko, 2018). In cattle, young bull calves will separate from the maternal herd in search of mates; thus, they will develop new dominance hierarchies that reflect their ability to access a mate. Heifers will remain with the maternal herd and will enter existing dominance hierarchies developed to dictate access to resources and spatiotemporal preferences. Thus, there are sex-differences in the resource that social hierarchies are centered around, and there are differences in the social dynamics and spatial needs surrounding the development of these social scenarios. In addition, according to Adeyemo & Healt (1982) who evaluated a group of Brown Swiss, Holstein, and White Fulani heifers, body weight was well correlated with dominance rank. Consequently, Haskell et al., (2019) feed intake is affected by dominance, with more dominant steers presenting higher DMI. Which could be affecting behaviors like bunk displacement and brush displacement, and head butt because this behavior are influenced by dominance and social interactions (Raj et al., 1991). Regarding brush use, due that, steers have the intrinsic need of exploratory behavior; this could be one of the factors influencing the higher interaction with the brush from steers than heifers.

Steers performed allogrooming and brush use more frequently and in longer total durations than heifers, aligning with this research, past research showed that brush use is positively linked to self grooming and allo grooming (Horvath et al., 2019). Social bonds between

male-male groups have shown to be tighter than female-female in other species, like dolphins, or raccoons (Randić et al., 2012; Gehrt et al., 2008). Therefore, steers may have performed more allogrooming than heifers in an attempt to fulfill their social and exploratory needs.

One of the factors that could be affecting the differences by sex in stereotypic behavior is that the ontogeny of cattle biology indicates that cattle are experiencing puberty when they are approximately one year old (Gregory et al., 1991). Thus, the cattle in this study were undergoing sexual development, of which there are substantial sex-based differences in behavior and motivation. Stereotypic behaviors are indicative of frustration or an unmet behavioral need. At this age, steers would have been motivated to roam either solitary or in small groups while maturing to access mates, the reduced ability to roam and the intensity of the social interactions (as reflected in stocking density), may have caused steers to experience more frustration than heifers. Due that, they need to exercise their exploratory behavior in order to ensure their fitness (Wood-Gush, 1989).

Productivity also differed between sexes, due that steers were more productive than heifers. As expected steers began and finished the trial with heavier body weights than heifers, had higher ADG, higher DMI, lower F:G, and visited the bunk less frequency during the day (Scheiber et al., 1987). Cattle are highly motivated to access brushes (McConnachie et al., 2018), their use follows a circadian rhythm (Meneses et al., 2021), and beef calves with access to brushes have been shown to perform more play and social behaviors compared to calves housed in non-enriched pens (Bulens et al, 2014). Furthermore, second lactation dairy cows with access to an automated brush had a 3.5% increase in milk yield (Schukken & Young, 2009). Which aligns with the results of this research due that steers are more productive, use the brush the most, and perform more social behavior than heifers.

On the other hand, other than BV frequency, feeding behavior traits did not differ between sexes. Feeding behavior of beef cattle is influenced by a wide variety of external factors, including weather (Schwartzkopf-Genswein et al., 2003), animal temperament (Voisinet et al., 1997), animal health (Wolfger et al., 2015), and animal management practices, such as bunk space, and bunk management (Oolson et al., 2019), yet the full effect of beef cattle feeding behavior are not well understood yet (Schwartzkopf-Genswain et al., 2003, 2011). And could be the reason that it didn't show a difference between sexes.

While heifers presenting higher EV than steers, visited the bunk more frequently than steers, were less efficient, gained less weight, and tended to have lower DMI. Further, none of the remaining automatically recorded feeding behavior traits differed between the sexes. Literature focusing on sex differences in automatically measured feeding behavior is scarce. But, Nkruman et al., (2007) measured EV of Angus, Charolais, and commercial bulls over 3 years, and found no association between EV, feeding frequency, feeding duration, and head down duration, but was negatively correlated with DMI.

Lastly, due to the need to be more productive every day to feed the increasing population that is expected to be 9.7 billion by 2050 (UN, 2015) a new behavioral phenotype to select for productivity was of interest, and finding in this research that brush use can be used to evaluate *Bos Indicus* cattle productivity could be due to several factors. Even though the relationship between brush use and DMI is inconsistent between researches where some researchers say that the provision of brush did not affect DMI on Holstein calves (Horvath et al., 2020), in this research we noticed an interaction between sex and brush use, where high and medium brush use heifers and steers differed between each other. However an effect of brush use on ADG was observed, where higher brush use animals had a higher ADG than medium and low brush use animals, aligning

with previous research in pigs where Rodarte et al. (2004) found that growing piglets with access to EE had significantly higher weight gain than piglets with no EE. Showing that pleasurable behavior can be used as a new phenotype to select for productivity, and if we can consider more factors than just temperament on steer selection for productivity, we can become more accurate and more proactive at achieving the so much needed productivity to feed the increasing population, and become a more cost effective protein source.

CONCLUSION

The objective of this study was to identify a sampling strategy that could accurately and efficiently record the performance of social, agonistic, and stereotypic behaviors in drylot-housed cattle with access to brushes. Consequently, the use of that sampling strategy characterized sex-specific differences in social and stereotypic behaviors, brush use, docility, and feeding behavior performed by *Bos indicus* cattle housed in drylots. Lastly, the productivity, temperament, and feeding behavior differences of cattle with divergent brush use phenotypes were characterized. Observing cattle for 15 minutes every 30 minutes yielded the highest accuracy for all behavioral metrics and was considered the most effective strategy for comprehensively evaluating cattle social behavior. Steers performed all behaviors evaluated (e.g., head butt, allogrooming, mounting, brush use, bunk displacement, brush displacement and tongue rolling) for longer and more frequently than heifers, except bar licking that even though the average bout duration was higher for steers there was no difference on the frequency of the behavior. The steers that used the brush the most, had higher FBW, higher ADG, and higher DMI than low brush use steers, and as expected steers were more productive than heifers while also being less temperamental than heifers.

These results present a validated sampling strategy that can reduce the observation time required to collect social behavior data in cattle in half while retaining a minimum of 85% accuracy for social behaviors (allogrooming, mounting, head butt, and bunk displacement), stereotypic behavior (bar licking, and tongue rolling), and environmental enrichment use (brush use). This knowledge can contribute to the design of sensor technology with and extended battery life, and the development of sex-specific algorithms. Providing insight in to what happens throughout the day, as is designed to capture behaviors that happen infrequently or in short

durations (Daigle & Siegford, 2014). On the other hand, this research highlights the importance of accounting for sex in cattle research, develop sex-specific management practices and develop sex-specific algorithms for technological devices to better understand cattle behavior in a daily basis. Due that, this research demonstrated than other than the external differences that *Bos Indicus* steers and heifers present on the outside, internally, they differ in many aspects as well. Steers showed to be more socially active, use EE like brushes more, be more productive, and less temperamental than heifers. Also suggesting that there is a sex effect on environmental enrichment use, and productivity therefore, monitoring the animal's performance of a pleasurable behavior (brush use), rather than their fear response (EV), may be a useful behavioral phenotype to add when selecting for productivity in *Bos Indicus* steers.

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