Associations of behavior-based measurements and clinical disease in preweaned, group-housed dairy calves

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ABSTRACT

Producers report bovine respiratory disease and neonatal calf diarrhea as the 2 most common diseases in preweaned calves, both of which can affect calf performance and welfare. Housing calves in groups during the preweaning period has increased in popularity and has the potential to improve calf welfare, but only if producers can detect and treat disease efficiently. A health-screening tool is needed that allows producers to identify suspect animals but minimize the time spent examining healthy animals. The objective of this study was to determine if disease in preweaned, group-housed dairy calves was associated with behavioral measures of illness. This cross-sectional study included the evaluation of preweaned, group-housed calves ($n = 206$) on 4 farms in Wisconsin, United States. Farm visits included the scoring of key behaviors (abnormal posture when lying or standing, isolation from the group, lethargy, and 2 approach tests that tested the willingness of calves to approach a stationary person) that were hypothesized to be components of sickness behavior. Following the behavior scoring, calves underwent an individual health assessment for bovine respiratory disease, neonatal calf diarrhea, and umbilical infections. Each behavior category was scored as normal (0 points) or abnormal (1 point), and then categories were summed to obtain a total behavior score for each calf. Behavior scores ranged from 0 (normal) to 5 (severely abnormal). A total behavior score of 3 or greater was considered positive for disease. The outcome of a positive or negative test on the behavior score were analyzed using PROC LOGISTIC (SAS Institute Inc., Cary, NC) and the model controlled for calf age and farm. Bovine respiratory disease was significantly associated with a behavior score of 3 or greater. The use of a behavior score shows promise as a screening tool for disease in preweaned calves.

Key words: calf, behavior, bovine respiratory disease, neonatal calf diarrhea

INTRODUCTION

Neonatal calf diarrhea (**NCD**) and bovine respiratory disease (**BRD**) result in increased costs for the farm and are reported to affect 24 and 12% of preweaned calves in the United States, respectively (USDA, 2010). These diseases are also the 2 most common causes of mortality in preweaned calves (USDA, 2010).

Bovine respiratory disease has been associated with reduced growth (Virtala et al., 1996a; Stanton et al., 2012), decreased calving ease and survival to first calving (Stanton et al., 2012), increased age at first calving (Waltner-Toews et al., 1986; Stanton et al., 2012), and increased risk of not completing first lactation (Bach, 2011). Neonatal calf diarrhea has been associated with increased age at first calving (Waltner-Toews et al., 1986) and reduced average daily gain (Windeyer et al., 2014). Furthermore, sickness and the feelings of malaise that accompany illness can result in poor animal welfare (Millman, 2007).

To improve health and animal welfare, producers must be able to reliably identify sick animals, but many use measures that have poor sensitivity for the overall identification of illness in calves (Sivula et al., 1996). Multiple methods for disease identification are available, including assessment of individual clinical signs (McGuirk, 2008), and changes in feeding behavior data using both automated milk feeders (Svensson and Jensen, 2007; Borderas et al., 2009b) and radio frequency technology (Quimby et al., 2001). However, automated monitoring technologies are expensive, they require further validation, and the data from these technologies may be difficult for producers to interpret and use.

The observation of particular behaviors related to the sickness response may be a helpful low-tech method of disease identification. Behavioral changes that accompany illness, collectively termed sickness behavior, are thought to be an adaptive response that increases an animal's probability of survival (Hart, 1988). Changes

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include lethargy, increased time spent lying down, self-isolation, and a decrease in grooming, appetite, and exploratory behavior (Hart, 1988; Haba et al., 2012; Proudfoot et al., 2014; Cramer and Stanton, 2015).

We hypothesized that postural changes displayed by sick calves would include short lying, lateral lying, and standing with a hunched posture. Short lying, defined as a calf lying with its head and legs gathered close to its body, may reduce the amount of exposed surface area and aid in heat conservation. A lateral lying posture may be adopted in sick calves to alleviate abdominal discomfort or in animals that are too weak to maintain sternal recumbence. A hunched standing posture has been reported in mice injected with LPS, which serves as a model for sickness behavior (Sparkman et al., 2005). Abdominal discomfort and pain has been reported in humans during intestinal tract inflammation (Al-Chaer and Traub, 2002); a hunched standing posture in calves may be indicative of abdominal discomfort that is associated with enteric disease.

Additional behavioral changes associated with illness are self-isolation, lethargy, and decreased exploratory behavior. Adult dairy cows that are ill spend more time in a secluded area compared with healthy cows (Proudfoot et al., 2014). Lethargy is recognized as part of the behavioral response to illness (Hart, 1988) and has been included in a clinical illness scoring system for BRD (White et al., 2012). The ability to detect these postural changes in preweaned, group-housed calves has not been evaluated. In previous work, we found that the probability of approaching a person or novel object was lower in calves with clinical signs of BRD compared with healthy calves (Cramer and Stanton, 2015). These findings suggest that animals are less interested in novelty and perform less exploratory behavior during illness.

The objective of this study was to determine if disease in preweaned, group-housed dairy calves was associated with behavioral measures of illness.

MATERIALS AND METHODS

Farm Enrollment

Four farms were enrolled in this study, and farm selection was based on a convenience sample of farm referrals from a veterinarian at the University of Wisconsin-Madison School of Veterinary Medicine (Madison, WI) and dairy extension personnel at the University of Wisconsin-Madison. The number of lactating cows for each farm was as follows: farm 1 had 1,250, farm 2 had 900, farm 3 had 400, and farm 4 had 160. Enrollment criteria included farms that housed preweaned calves in groups for a minimum of 5 weeks during the

preweaning period, the use of computerized automated calf feeders, and recorded treatment and date of birth for calves. Both bull ($n = 20$) and heifer ($n = 186$) calves were included in this study. A single farm visit was conducted at each farm between April and August of 2014.

Farm Staff Interviews

Farm staff involved in calf care and management were asked about the amount of milk fed, treatment protocols, and methods used for detecting illness. Stocking density, bedding type, and the age of calves upon entry to the group pen were also recorded. All farms recorded treatments on paper, and research staff photographed these for later data entry and analysis. Calves were considered to have a treatment history if the calf was treated with an antibiotic or a nonsteroidal antiinflammatory drug in the 7 d before the farm visit or on the day of the visit. Research staff obtained date-of-birth records to calculate calf age on the day of observation. Farm management information was used to describe the study population; only the treatment history of calves and date-of-birth records were used for further analysis.

Behavioral Observations

The observations used in the behavior score were lying and standing posture, isolation from the group, a first human approach test, lethargy score, and a second human approach test (Table 1). Prior to entering each pen, the researcher recorded lying posture, standing posture, and isolation from the group. If a calf's ear tag could not be identified from outside the pen, a picture was taken and the calf was identified during health examinations. The researcher then entered the pen and stood motionless for 60 s for the first approach test. Any calves that were not standing were assigned an encouragement to rise score adapted from Stanton (2011; Table 1) and then the second approach test was performed. Calves that did not stand after the encouragement to rise score was completed were not helped to stand by the researcher. The same researcher performed all behavioral tests on every farm. Each behavior category was scored as 0 (normal) or 1 (abnormal) (Table 1). Scores from every category were summed to determine an overall behavior score. The following behaviors were tested for associations with disease using chi-squared or Fisher's exact test analyses: lying and standing posture, isolation from the group, first approach test, lethargy score, and second approach test; each of these behaviors was associated with at least 1 disease ($P < 0.20$) and was included in the composite

Table 1. Definitions of behavioral measures

Behavioral measure	Score ¹	Description
Lying posture	0	Lying sternal with or without limbs extended; head is extended from the body
	1	Lying lateral with all limbs extended OR lying sternal with all limbs and head tucked toward the body
Standing posture	0	Standing stationary with a straight back, and head is level with or above the shoulders
	1	Standing stationary with a back that is arched up or down, the head is below the shoulders, or both
Isolation	0	Lying within 1 m of at least 1 other calf
	1	Lying at least 1 m or greater from another calf
Approach tests (first and second)	0	At least 1 step in the direction of the stationary person within 60 s
	1	No movement made toward the stationary person within 60 s
Encouragement to rise	0	Calf is already standing; calf rises when researcher walks toward calf; calf rises when researcher is within 1 m and gives single vocal encouragement
	1	Rises after researcher makes contact ² with calf; rises after researcher makes repeated contact with the calf; calf will not stand

¹Score of 0 considered normal; score of 1 considered abnormal.

²Contact was a light tap from the researcher's hand on the calf's back.

behavior score. Sickness behavior can include the expression of many different behaviors, and the behaviors calves express at a given time will depend on variations in both internal and external motivations. As such, a cumulative score was likely to be of greater value than individual responses. Furthermore, observation of the behaviors on-farm entailed a short observation period, and including multiple behaviors in the behavior score was aimed at improving the possibility of identifying a sick animal.

Health Assessments and Disease Definitions

After the completion of all behavioral observations, standardized health assessments were performed on ev-

ery preweaned calf in group housing (Table 2). Calves were examined for clinical signs of NCD and umbilical infections, and for clinical signs of BRD using a BRD screening tool (McGuirk, 2008; Table 2).

A total BRD score was obtained by summing scores for the following: nasal discharge, cough, rectal temperature, and the highest score between the eye or ear (McGuirk, 2008). Bovine respiratory disease was defined as 2 or more abnormal clinical signs (score of 2 or greater in a category). The clustering of at least 2 abnormal clinical signs is indicative of respiratory disease (McGuirk, 2008). Three classifications for BRD type were generated based on the total BRD score: total BRD score of 4 or 5 (**BRDmild**), total BRD score of 6 or greater (**BRDsevere**), and no BRD. This classi-

Table 2. Standardized health scoring system

Variable	Score 0	Score 1	Score 2	Score 3
BRD ¹ score				
Rectal temperature, °C	37.8–38.2	38.3–38.8	38.9–39.3	≥39.4
Cough	None	Single induced cough	Induced repeated or occasional spontaneous cough	Repeated spontaneous cough
Nasal discharge	Normal serous discharge	Small amount of unilateral cloudy discharge	Bilateral, cloudy, or excessive mucous discharge	Copious bilateral mucopurulent discharge
Eyes	Normal, clear	Small amount of discharge	Moderate amount of bilateral discharge	Heavy ocular discharge
Ears	Normal	Ear flick or head shake	Slight unilateral droop	Head tilt or bilateral droop
NCD ² score	Normal: fecal consistency was not extremely watery	Diarrhea: feces extremely watery	—	—
Umbilical score	Cord cannot be found; umbilical opening is completely flush with the abdomen of the calf	Cord can be palpated, but is soft and approximately the size of a pencil	Cord is larger than a pencil, but the calf does not react when palpated	Umbilical is very swollen, hard, and hot to the touch, and calf reacts when palpated

¹Bovine respiratory disease; scoring adapted from McGuirk (2008).

²Neonatal calf diarrhea.

fication was based on recommendations from the BRD screening tool, which advises that calves with scores of 4 or 5 should be watched for the development of severe signs of BRD (McGuirk, 2008) and calves with scores of 6 or greater should be treated (Lago et al., 2006).

Defecation was stimulated in every calf, and any extremely watery feces were defined as NCD (Cramer and Stanton, 2015). The umbilicus of every calf was palpated and scored from 0 to 3, with a score of 3 considered infected (Table 2). Febrile calves were identified by a rectal temperature $\geq 39.4^{\circ}\text{C}$. Disease categories were not mutually exclusive, and some calves defined as ill had multiple diseases.

Sensitivity and Specificity

A cut point was defined as the total points in the behavior score that determined the delineation between a positive behavior score and a negative behavior score (e.g., at a cut point of 3, calves with total behavior scores of 3 or greater would be considered a positive test). Sensitivity (**Se**) and specificity (**Sp**) were calculated as described by Dohoo et al. (2003) for each possible cut point in the behavior score for the detection of BRD, NCD, and fever. The Se and Sp of the behavior scores for cut points of 1 or greater, 2 or greater, 3 or greater, 4 or greater, and 5 were determined for the identification of each disease category; the cut point that yielded the highest combination of Se and Sp was further analyzed in the multivariable model. This cut point determined the behavior score cut point for a positive and negative behavior score, which was the outcome analyzed in the multivariable model.

The Se of the farm to identify disease was calculated as the proportion of calves that research staff identified with a particular disease treated by the farm within 7 d before the farm visit. To determine the precision of the Se and Sp estimates for test characteristics of the behavior score, positive predictive value (**PV+**) and negative predictive value (**PV−**) were calculated as described by Dohoo et al. (2003). For all calculations

of Se, Sp, PV−, and PV+, disease-negative was defined as calves without a specified disease, rather than completely healthy calves.

Statistical Analysis

We performed univariable analysis using chi-squared or Fisher's exact tests with PROC FREQ (SAS Institute Inc., Cary, NC) and tested the associations between a positive behavior score and the following variables: age, treatment history, farm, BRD type (BRDmild, BRDsevere, or no BRD), NCD, umbilical score, sex, and fever. Variables significantly associated ($P < 0.20$) with the behavior score were offered to the multivariable model for further analysis. We used a multivariable logistic model (PROC LOGISTIC, SAS Institute Inc.) to analyze associations between disease and the probability of a calf testing positive for illness based on the behavior score, adjusting for significant covariates. The multivariable model was constructed using stepwise backward removal of nonsignificant variables ($P > 0.05$). Odds ratios were calculated based on the probability of a positive test. We performed a power analysis (PROC POWER, SAS Institute Inc.) for nonsignificant findings to determine if inadequate sample size was an issue.

RESULTS

We observed 206 preweaned calves: 68, 59, 47, and 32 calves on farms 1, 2, 3, and 4, respectively (Table 3). Overall, the age of calves ranged from 4 to 64 d, with a mean age of 35 d. On all farms, calves entered the group pen by 10 d of age (Table 3).

To identify sick calves, Farm 1 observed drinking speed and overall daily consumption as recorded by the automated milk feeder, as well as nasal discharge. Farm 2 observed nasal discharge and changes in attitude. Farm 3 was participating in a separate research trial to evaluate milk-feeding programs, and all calves were examined twice a week using the BRD screening tool.

Table 3. Description of age and management of preweaned, group-housed dairy calves (n = 206), by farm

Variable	Farm 1	Farm 2	Farm 3	Farm 4	Overall
No. of calves observed	68	59	47	32	206
No. of group pens	4	4	3	3	—
Mean no. of calves per pen (range)	17 (12–23)	14 (11–17)	15 (12–20)	10 (2–16)	—
Mean age at time of observation, d (IQR ¹)	31.4 (20–42)	39.5 (33–48)	35.8 (19–52)	35.2 (14–50.5)	35.3 (22–48)
Age of calves upon entry to group pen, d	8	10	4	8	—
Volume of milk offered, L (milk type)	8 (milk replacer)	8 (milk replacer)	15 (pasteurized whole milk)	6 (pasteurized whole milk)	—

¹Interquartile range.

Farm 4 used milk intake data generated from the automatic feeder to detect sick calves and observed the following for confirmation of disease: watery eyes, droopy ears, wet tails, and extremely watery feces. Alarm information from automatic feeders was not used for analysis, because complete automated feeder information was not available for 3 out of 4 farms: Farm 1 had incomplete feeder records; Farm 2 did not use feeder alarms to detect sick calves, so this information was not available from the feeder; and the feeder on Farm 4 did not record alarms from the previous day.

Because the number of calves with NCD who also had a fever was low ($n = 6$), NCD could not be classified as mild or severe in the same way as BRD. The overall prevalence of BRDmild, BRDsevere, NCD, umbilical infection, and fever are reported in Table 4. The mean ($\pm$ SD) age of calves with BRDmild, BRDsevere, NCD, umbilical infection, and fever was 35 ($\pm$ 16), 35 ($\pm$ 12), 28 ($\pm$ 15), 45 ($\pm$ 9), and 34 ($\pm$ 13) days, respectively. A NCD score was missing for 1 calf due to a data collection error. Twenty-four percent (50/206) of calves had more than 1 of BRD, NCD, fever, or umbilical infection. Four percent (1/23) of calves with BRDmild and 48.4% (15/31) of calves with BRDsevere also had a fever. Thirteen percent (6/45) of calves with NCD also had a fever, and 3.0% (6/206) of calves had a fever of unknown origin.

Farm staff had treated 19.9% (41/206) of calves in the 7 d before the farm visit. Overall, 17.4% (4/23), 29.0% (9/31), and 26.7% (12/45) of calves identified by research staff with BRDmild, BRDsevere, and NCD,

respectively, had been treated by the farm in the previous 7 d (Table 4). The Se of the farms for disease identification of BRDsevere was 29.0% (Table 4).

Abnormal lying and abnormal standing were observed in 11.7% (24/206) and 0.5% (1/209) of all calves, respectively (Table 5). These were combined into 1 category: abnormal posture. Abnormal posture was observed in 19.4% (6/31) of calves with BRDsevere and 13.5% (12/89) of healthy calves (Table 5).

The highest Se and Sp for the identification of BRDsevere occurred between behavior score cut points of 2 and 3. Preliminary analysis showed stronger association between disease and a behavior score cut point of 3. Therefore, behavior scores of 3 or greater were considered a positive test result for disease and were included in the logistic model.

At a cut point of 3, 48.4% (15/31) of calves with BRDsevere tested positive on the behavior score, compared with 24.7% (22/89) of healthy calves (Table 6). The multivariable logistic model included treatment history, fever, NCD, BRD type, age, and farm. Treatment history, fever, and NCD were not significant and were removed from the model ($P > 0.05$). The results from the final model are summarized in Table 7. We observed no difference in the probability of a positive score between calves with BRDmild and BRDsevere (Table 7, $P > 0.05$). Calves with BRDsevere were 4.2 (95% CI: 1.7–10) times more likely to have a positive behavior score than calves without signs of BRD (Table 7, $P < 0.01$). The probability of a calf having a positive behavior score decreased with age ($P < 0.05$). We

Table 4. Disease prevalence (different disease categories were not always mutually exclusive) and proportion of calves (%; no./total in parentheses) treated by farm staff within 7 d of the farm visit in preweaned, group-housed dairy calves ($n = 206$), by farm

Disease category	Farm 1	Farm 2	Farm 3	Farm 4	Overall
BRDmild ¹					
Prevalence	7.4 (5/68)	15.3 (9/59)	8.5 (4/47)	15.6 (5/32)	11.2 (23/206)
Treated	20.0 (1/5)	22.2 (2/9)	25 (1/4)	0 (0/5)	17.4 (4/23)
BRDsevere ²					
Prevalence	11.8 (8/68)	15.3 (9/59)	10.6 (5/47)	28.1 (9/32)	15.1 (31/206)
Treated	12.5 (1/8)	33.3 (3/9)	100 (5/5)	0 (0/9)	29.0 (9/31)
NCD ³					
Prevalence	26.5 (18/68)	32.7 (16/49)	12.8 (6/47)	15.6 (5/32)	22.0 (45/205)
Treated	27.8 (5/18)	25 (4/16)	0 (0/6)	60 (3/5)	26.7 (12/45)
Umbilical infection ⁴					
Prevalence	1.5 (1/68)	2.0 (1/49)	6.4 (3/47)	9.4 (3/32)	3.9 (8/206)
Treated	0 (0/1)	0 (0/1)	0 (0/3)	33.3 (1/3)	12.5 (1/8)
Fever ⁵					
Prevalence	16.18 (11/68)	10.20 (5/49)	6.38 (3/47)	18.75 (6/32)	12.14 (25/206)
Treated	18.2 (2/11)	40.0 (2/5)	66.7 (2/3)	0 (0/6)	24.0 (6/25)

¹Mild bovine respiratory disease: total BRD score of 4 or 5, with at least 2 categories with a score of 2 or greater.

²Severe bovine respiratory disease: total BRD score of 6 or greater.

³Neonatal calf diarrhea: clinical signs of diarrhea; fecal consistency was extremely watery.

⁴Umbilical score of 3 (infected).

⁵Rectal temperature $\geq 39.4^{\circ}\text{C}$.

Table 5. Associations between behavioral measures and disease categories: proportion of sick and healthy calves (%; no./total in parentheses) that had abnormal scores for each behavioral measure, by disease

Behavioral measure	All calves	Healthy ¹	BRDmild ²	BRDsevere ³	NCD ⁴	Fever ⁵
Abnormal posture	12.14 (25/206)	13.48 (12/89) $P = 0.61^6$	8.7 (2/23) $P = 0.25$	19.35 (6/31) $P = 0.18$	6.67 (3/45) $P = 0.19$	8 (2/25) $P = 0.23$
Isolation	5.34 (11/206)	3.37 (3/89) $P = 0.14$	4.35 (1/23) $P = 0.38$	16.13 (5/31) $P = 0.003$	4.44 (2/45) $P = 0.29$	20 (5/25) $P = 0.004$
First approach test	82.52 (170/206)	82.02 (73/89) $P = 0.87$	86.96 (20/23) $P = 0.21$	87.10 (27/31) $P = 0.47$	84.44 (38/45) $P = 0.69$	88 (22/25) $P = 0.18$
Lethargy	22.33 (46/206)	21.35 (19/89) $P = 0.77$	39.13 (9/23) $P = 0.04$	32.26 (10/31) $P = 0.15$	17.78 (8/45) $P = 0.40$	20 (5/25) $P = 0.77$
Second approach test	31.07 (64/206)	67.42 (60/89) $P = 0.68$	30.43 (7/23) $P = 0.94$	87.10 (27/31) $P = 0.02$	57.78 (26/45) $P = 0.07$	92 (23/25) $P = 0.008$

¹No clinical signs of bovine respiratory disease or neonatal calf diarrhea: no fever; umbilical score less than 3; not treated in previous 7 d.

²Mild bovine respiratory disease: total BRD score of 4 or 5, with at least 2 categories with a score of 2 or greater.

³Severe bovine respiratory disease: total BRD score of 6 or greater.

⁴Neonatal calf diarrhea: clinical signs of diarrhea; fecal consistency was extremely watery.

⁵Rectal temperature $\geq 39.4^\circ\text{C}$; fever and disease are not always mutually exclusive.

⁶ P -values were obtained from chi-squared or Fisher's exact tests to determine associations between each behavioral measure and the disease category.

observed no interaction between farm and BRD type ($P > 0.05$). At a cut point of 3, the behavior score had a Se of 48.4% (15/31) and a Sp of 79.4% (139/175) for the identification of calves with BRDsevere. The behavior score had a PV+ of 29.4% (15/51) and a PV– of 89.7% (139/155) for the identification of BRDsevere.

A power analysis revealed that this study had inadequate power (<80%) to detect a difference in the probability of a positive behavior score between the following disease categories: BRDmild versus BRDsevere, NCD versus healthy, fever versus healthy, and BRDmild versus healthy.

DISCUSSION

In the present study, NCD was defined as extremely watery feces. Because of a small sample size, we were unable to further break this down into NCD with or without fever. A more detailed scoring system that

included fever may have prevented misclassification as abnormal when calves were in fact normal (i.e., had watery feces but were clinically healthy), and this may have contributed to our nonsignificant findings. Further research into this area is warranted, specifically incorporating both duration of loose feces, temperature, and hydration status.

Compared with other methods of intensive evaluation, the behavior score had a lower Se. Using a cut point of 3, the behavior score developed in the present study yielded a Se of 48.4% and a Sp of 79.4% for the identification of BRDsevere. The behavior score was developed for use as a potential screening tool for disease in group-housed calves; it would be ideal to have a high level of Se so that sick animals can be identified and a moderate level of Sp to reduce the time required to rule out false positives. A Canadian study compared the BRD screening tool (used to screen calves for BRD in the present study) to ultrasonography of lungs and

Table 6. Proportion of calves (%; no./total in parentheses) by total behavior score and health status

Total behavior score	Healthy ¹	BRDmild ²	BRDsevere ³	NCD ⁴	Fever ⁵
0	12.36 (11/89)	8.70 (2/23)	3.23 (1/31)	11.11 (5/45)	0
1	19.10 (17/89)	17.39 (4/23)	6.68 (3/31)	28.89 (13/45)	20.00 (5/25)
2	43.82 (39/89)	39.13 (9/23)	38.71 (12/31)	40.00 (18/45)	44.00 (11/25)
3	17.98 (16/89)	30.43 (7/23)	41.94 (13/31)	17.78 (8/45)	28.00 (7/25)
4	6.74 (6/89)	0	3.23 (1/31)	2.22 (1/45)	4.00 (1/25)
5	0	4.35 (1/23)	3.23 (1/31)	0	4.00 (1/25)

¹No clinical signs of bovine respiratory disease or diarrhea: no fever; umbilical score less than 3; not treated in previous 7 d.

²Mild bovine respiratory disease: total BRD score of 4 or 5, with at least 2 categories with a score of 2 or greater.

³Severe bovine respiratory disease: total BRD score of 6 or greater.

⁴Neonatal calf diarrhea: clinical signs of diarrhea; fecal consistency was extremely watery.

⁵Rectal temperature $\geq 39.4^\circ\text{C}$; fever and disease are not always mutually exclusive.

auscultation of 4 different portions of lungs (Buczinski et al., 2014). Using ultrasonography as the reference method, the BRD screening tool had a Se of 55% and a Sp of 58%. The Se and Sp of lung auscultation relative to ultrasonography of lungs ranged from 0 to 16.7% and 97.3 to 100%, respectively (Buczinski et al., 2014). Previous work validating the use of ultrasonography found Se and Sp from 85% and 98% for calves with clinical signs of BRD and using postmortem examinations as the reference method (Rabeling et al., 1998). The behavior score developed in the present study had greater Se, but lower Sp than reported for lung auscultation (Buczinski et al., 2014). The behavior score also had lower Se and Sp compared with ultrasonography (Rabeling et al., 1998). However the BRD screening tool, which was the reference method in the present study, uses clinical signs for diagnosis of BRD (McGuirk, 2008) and can have low Se and Sp when compared with ultrasonography (Buczinski et al., 2014). It is possible for calves to have consolidated lungs on ultrasonography but show no clinical signs of disease (Ollivett, 2014). Therefore, calves with subclinical BRD may not have been identified in the present study, and that would bias results toward the null. The inability of the methods used in the present study to detect subclinical disease could affect the results; with the use of ultrasonography, our results may have been different due to the ability to correctly categorize calves with subclinical disease as sick, affecting the Se and Sp of the test. Furthermore, no gold standard is currently available for the identification of upper respiratory, lower respiratory, clinical, or subclinical BRD.

The behavior score had a PV+ of 29.4%, which would mean that 29% of the calves that tested positive on the behavior score would actually have BRDsevere. The PV− of the behavior score for the identification of BRDsevere was 89.7%, which means that 89.7% of animals that tested negative would actually be healthy. The PV+ is not as desirable, but the PV− is more acceptable because time would not be spent examining a large number of calves that are not sick. Although the Se of the behavior score was lower than other methods of evaluation used in previous studies, it was more sensitive for illness identification than the farm's current detection methods. The Se of the farms for disease identification for BRDsevere was 29.0%. The behavior score had a Se of 48.4% at a cut point of 3, which was greater than the Se of the farm staff and would increase the number of calves with BRDsevere identified on-farm. The ability of farm staff to detect disease in the present study was consistent with previous work, which found that producers generally had poor Se for overall disease detection (Sivula et al., 1996). The Se and Sp of the behavior score may be improved by combining it with other measures of illness, such as changes in feeding behavior. Due to the low Se of the behavior score, it would not be ideal as the sole method of disease detection on a farm. However, it has merit because it is financially feasible for many farms and may improve overall disease detection if no systematic detection is in place. Furthermore, it requires an established amount of time that farm staff would be required to observe calves. In combination with other methods of disease detection, the behavior score could be a useful compo-

Table 7. The probability of preweaned, group-housed dairy calves scoring positive on a behavior score¹ at a cut point of 3 for each disease category

Variable	Estimate	SE	Odds ratio	95% CI	P-value
Intercept	0.19	0.46			
No BRD ² (relative to BRDmild ³)	−0.03	0.35	0.51	0.19–1.42	0.9391
BRDsevere ⁴ (relative to BRDmild)	−0.03	0.35	2.1	0.64–7.1	0.9391
BRDsevere (relative to no BRD)	−0.69	0.25	4.1	1.7–10	0.0064
Age	−0.03	0.01	0.97	0.95–0.99	0.0125
Farm 1 (relative to farm 2)	−0.88	0.33	0.14	0.05–0.36	0.008
Farm 1 (relative to farm 3)	0.58	0.36	0.72	0.23–2.24	0.1249
Farm 4 (relative to farm 1)	0.34	0.35	0.29	0.10–0.88	0.3283
Farm 3 (relative to farm 2)	−0.58	0.36	0.19	0.07–0.54	0.007
Farm 4 (relative to farm 2)	0.34	0.35	0.46	0.17–1.25	0.3283
Farm 3 (relative to farm 4)	−0.56	0.36	0.41	0.13–1.31	0.1249

¹The behavior score consists of abnormal posture, isolation, 2 approach tests, and encouragement to rise. Each of these behaviors was given a score of 0 (normal) or 1 (abnormal), for a total possible score of 6 points. At a cut point of 3, calves scoring 3 or greater were considered positive for disease on this behavior score.

²No clinical signs of bovine respiratory disease.

³Mild bovine respiratory disease: total BRD score of 4 or 5, with at least 2 categories with a score of 2 or greater.

⁴Severe bovine respiratory disease: total BRD score of 6 or greater.

nent of a comprehensive disease detection program. In the future, it would be useful to compare the results from the behavior score in the present study with lung ultrasonography on-farm to confirm BRD diagnosis. This would allow for accurate identification of disease and could help determine which specific behaviors are most useful for identifying calves with BRD.

The total proportion of calves that approached the researcher was 17% for the first approach test and 68% for the second approach test. These findings are consistent with previous work, in which the proportion of calves that approached ranged from 18 to 66% for calves tested at 1 and 6 wk of age, respectively (Cramer and Stanton, 2015). In the present study, 25% of the calves were less than 3 wk of age, which may partially explain the overall low proportion of calves that approached. Cattle are hider species, meaning that calves spend most of their time lying down and hidden from view during the first 3 wk of life, and are therefore relatively inactive during this time (Fraser and Broom, 1997). Because of this, very young calves may exhibit less exploratory behavior. Furthermore, fear may also explain the low response to the approach test. Fear is a competing behavior to inquisitive exploration, which is the intrinsic exploration of an object without the reward of a conventional reinforcer, such as food (Murphy and Wood-Gush, 1978; Wood-Gush and Vestergaard, 1989). Calves had no prior interactions with research staff, which may have led to increased fear and less exploratory behavior. If this behavior score were used routinely on farms, calves would likely be more familiar with research staff and may be more willing to approach.

Numerically, more calves approached during the second test than in the first test, except for calves with BRDsevere and fever. The increased period of adaptation to the researcher may have meant that calves become less fearful or more interested in exploration of the stationary human. The proportion of calves with BRDsevere that approached during the first approach test was the same as the proportion that approached during the second test. Severe BRD significantly affected the probability of testing positive on the behavior score, and a large proportion of calves with BRDsevere had a fever. Even with the extra activity in the pen and a longer time to investigate the stationary person, calves with BRDsevere still did not approach. This provides more evidence that BRDsevere affected exploratory behavior, regardless of the time allowed to approach.

This behavior score was intended to be used on-farm in a quick and feasible manner, meaning that animals would be observed only for a short time. By including multiple behaviors in the behavior score, it may have

been more likely that the behavior score would be able to identify sick calves. However, in the present study, a variety of behaviors were chosen that were convenient to measure on-farm. Because this was a pilot study, we did not pursue an all-encompassing approach to determine which behaviors should be included. The addition of other behaviors may strengthen the behavior score, and the authors encourage work in this area.

Calves were less likely to test positive on the behavior score as their age increased. These results agree partially with previous results, which showed that calves were more likely to approach a stationary human and a novel object as they got older (Cramer and Stanton, 2015). Furthermore, young calves seek supplemental heat in colder temperatures (Borderas et al., 2009a), and the adoption of certain lying postures may have been to reduce heat loss, regardless of disease status. Also, calves were identified with NCD at a younger age and with BRD when they were older, both of which are consistent with previous findings (Virtala et al., 1996b; Svensson et al., 2006; USDA, 2010). This clustering of disease and age can make it difficult to separate the effects.

Farm significantly affected the probability of having a behavioral score of 3 or greater, but we observed no interaction between farm and BRD type. Therefore, BRD type was equally likely to affect the probability of scoring positive, regardless of farm. The effect of farm on the probability of scoring positive may have been due to behavioral differences in calves between farms. Social factors, such as the presence or absence of conspecifics, can influence exploratory behaviors (Boissy and Le Neindre, 1990) and would likely vary by farm. Differences in handling by farm staff would also affect the reaction of calves during the behavioral tests, as previous handling experiences can affect the behavior of cattle (Waiblinger et al., 2004). The behavior score should be tested on more farms and modified to develop a score that can identify sick animals on farms with different management practices, such as group size, age of calves, and calf handling.

The overall prevalence of BRD in the present study, regardless of severity, was 26% and defined as 2 or more abnormal clinical signs. Using the same BRD screening tool, Lago et al., (2006) defined BRD as a score of 6 or greater and reported a mean BRD prevalence of 14% (range 0 to 37%), which is consistent with the prevalence of BRDsevere in the present study. Furthermore, the prevalence of BRD in the present study was also consistent with findings from Cramer and Stanton (2015), and both studies used the same BRD screening tool and disease definitions. On a national level, producer-reported BRD prevalence is 12%, which is lower,

but producers may be less sensitive for the detection of disease than methods used in the present study (USDA, 2010). The prevalence of BRD in the present study was consistent with previous literature; it is likely that the association between BRDsevere and a positive behavior score would be valid on other farms.

Severe BRD, as defined here, significantly affected the probability of a calf scoring positive at a cut point of 3. This was likely because these animals expressed sickness behavior, and therefore more abnormal behaviors as indicated on the behavior score. Mild BRD did not significantly affect the probability of a positive behavior score. This behavior score was most useful for the identification of calves with more severe BRD and calves that were healthy, but was not accurate for the identification of calves with mild BRD. This may also have been due to a lack of power to detect a difference between BRDmild and healthy calves for the probability of testing positive on the behavior score. Our results also suggested that behavior alone was not sufficient to accurately identify sick calves; however, behavior may be useful in combination with other methods of disease detection. For these reasons, further test development and research for feasible methods of detecting for disease are important to accurately identify calves at early stages of illness, because early identification is crucial for treatment success (McGuirk, 2008) and to improve animal welfare.

CONCLUSIONS

The use of a behavior score shows promise for the identification of severe BRD in preweaned, group-housed dairy calves. This test had lower-than-desirable Se but moderate Sp. Because of the low Se, a behavior score should not be the only method used for disease detection. However, the behavior score was more sensitive for the identification of BRDsevere than farm staff. The behavior score may be most useful combined with other methods and represents a financially feasible option for detecting sick calves on many farms.

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