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Evaluation of a composite pain scale including facial expressions for detecting orthopedic pain in lame dairy cows

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ABSTRACT

Orthopedic disease, presenting as lameness, is a common cause of pain in dairy cattle, often implying a prolonged course of disease, with significant economic losses and impaired animal welfare. To mitigate these negative effects, early identification of lameness and monitoring of pain levels during recovery are crucial. This study aimed to evaluate whether the Cow Pain Scale (CPS) can be used to detect pain behaviors in stationary dairy cows with mild to moderate lameness or not, and to investigate the association between certain CPS items and orthopedic disease. Data were collected on a research dairy farm with a loose-housing system and included 36 clinical lameness cases from 34 individual cows. Each lameness case consisted of 2 trials: when the cow was found to be lame (initial trial), and after treatment and improvement (follow-up trial). Each trial included an on-site pain assessment with the CPS with simultaneous video recording, followed by Sprecher lameness scoring and clinical examination. Blinded pain assessments using the CPS were performed from the video recordings by 3 trained observers at 2 different occasions, 3 wk apart. Using linear mixed models, a significant positive correlation between CPS total scores and Sprecher lameness scores was identified for both video and on-site pain assessments. Predicted CPS total scores increased with increasing lameness scores; however, there was a large overlap in CI, indicating a complex relationship between pain score and pain intensity. Multiple correspondence analysis identified different CPS item combinations, including facial expressions, which were associated with orthopedic pain. Video scoring showed moderate inter- and good intrarater agreement in CPS total scores, but there was considerable variation in inter- and intrarater agreement for different scale items. The scale items "head position" and "back position" showed an overall strong agreement and "attention toward the surroundings" a moderate agreement. In contrast, intrarater agreement for the items "facial expression" and "ear position" was generally weak, which may indicate that these items are harder to assess. The CPS proved to be a reliable tool for video pain assessment in dairy cows with orthopedic pain. Furthermore, we showed that behavioral patterns varied among lame cows, which is why single items within the CPS should be interpreted with caution.
Key words: cattle, pain assessment, pain face, lameness, pain behavior

INTRODUCTION

Orthopedic disease, often presenting as lameness, is a common cause of pain in dairy cows (Thomsen et al., 2023) and negatively affects animal welfare (Whay and Shearer, 2017) and farmers' economies through reduced milk production, higher veterinary costs, and early culling (Ózsvári, 2017). Lameness in cattle usually stems from claw pathologies (Murray et al., 1996; Fenster et al., 2023), many of which progress over time (Leach et al., 1997; Somers et al., 2005) and may take weeks or months to heal (Van Amstel et al., 2003; Somers et al., 2005), thus causing acute and potentially also chronic pain. Despite early detection of claw lesions being important for the treatment response (Leach et al., 2012; Thomas et al., 2016), the prevalence of lameness is underestimated in many herds (Espejo et al., 2006; Leach et al., 2010). Adding nonsteroidal anti-inflammatory drugs to the corrective trimming of claw lesions may have a positive effect on recovery (Thomas et al., 2015; Sadiq et al., 2022), but analgesia is inconsistently used by veterinarians treating lameness in cattle (Hewson et al., 2007; Johnstone et al., 2021), possibly due to challenges in detecting, and thereby acknowledging, pain in individual cows. Therefore, to increase animal welfare and prevent eco-

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The list of standard abbreviations for JDS is available at adsa.org/jds-abbreviations-25. Nonstandard abbreviations are available in the Notes.

nomic losses, refined methods for early detection and assessment of orthopedic pain are needed.

Orthopedic pain in cattle is often assessed by subjective lameness evaluation, commonly using the 5-point Sprecher lameness scale (Sprecher et al., 1997; Schlageter-Tello et al., 2014a). The scale evaluates back position, stride length, and weight bearing. A score above 2 (out of 5) has been suggested to indicate the need for interventions, affecting reproductive performance and culling rates (Sprecher et al., 1997). However, Bicalho et al. (2007) found potentially painful claw lesions in a high proportion of cows with scores of 1/5 (i.e., the lowest score, indicating absence of lameness) and 2/5; 5.6% and 20.1%, respectively. Many painful lesions may therefore go untreated when subjective lameness scoring is employed, which is why other pain assessment methods are required.

Pain may induce species- and injury-specific alterations in behaviors that can be used in pain assessment, as seen in various behavioral-based pain scales designed for specific conditions or general pain in different animal species (Reid et al., 2007; Bussières et al., 2008; Luna et al., 2020). Although there are several bovine pain scales that have been developed for certain painful conditions, for example, postsurgical pain (de Oliveira et al., 2014), lameness (O'Callaghan et al., 2003), and mastitis (Giovannini et al., 2017), the Cow Pain Scale (**CPS**), based on bodily behaviors and facial expressions, was developed for on-site assessment of various painful clinical conditions, including those of more chronic nature (Gleerup et al., 2015). Certain combinations of facial expressions have also been associated with acute pain and are today incorporated in pain scales for many animal species, for example, horses (Dalla Costa et al., 2014; van Loon and Van Dierendonck, 2015), sheep (Häger et al., 2017), piglets (Viscardi et al., 2017), and cattle (Gleerup et al., 2015; Giovannini et al., 2017; Farghal et al., 2024). The "bovine pain face," suggested by Gleerup et al. (2015) includes muscle tensions related to the lips, nostrils, eye region, and the side of the face, and backward or lowered ears and is one of the scale items in the CPS (Gleerup et al., 2015). Recent studies have confirmed changes in facial expressions during acute pain in cattle (Müller et al., 2019; Ginger et al., 2023). However, whether facial expressions change during mild to moderate orthopedic pain in cattle has not yet been investigated, nor has the overall performance of the CPS for assessment of orthopedic pain.

Recently, the CPS and the UNESP-Botucatu Cattle Pain Scale were evaluated, confirming both scales to be valid and reliable for assessing postsurgical pain from video recording (Tomacheuski et al., 2023). However, pain is challenging to assess because individual factors, like personality (Ijichi et al., 2014), age, and sex (Guesgen et al., 2011), as well as pain source (McMeekan et al., 1999; Fogsgaard et al., 2015; Van De Gucht et al., 2017) may affect the pain expression. Whether all behaviors described in the CPS are present in cows with orthopedic pain, or if some behaviors are more frequently present, has not been explored. It is therefore important to evaluate scale performance on each specific pain type and on different group levels.

Both video-based and on-site pain assessment can be applied, as shown in studies of bulls (Tomacheuski et al., 2024) and piglets (Trindade et al., 2023), and they present different advantages and challenges. On-site assessment has been suggested as more practical in a farm setting, not requiring technical equipment and minimizing delays in treatment, but may potentially underestimate pain scores (Trindade et al., 2023; Tomacheuski et al., 2024). Furthermore, the presence of an observer may suppress pain-related behaviors, as demonstrated in horses and rabbits (Torcivia and McDonnell, 2020; Pinho et al., 2023), a challenge that video assessment from surveillance footage could resolve. The time requirement for a pain assessment can affect its feasibility in clinical situations (Reid et al., 2007), not the least in large herds, with limited observation time of individuals (Thomsen et al., 2012). In addition, a pain scale must produce consistent results in repeated measures, that is, it must be reliable (De Vet et al., 2011) and benefit from standardized observer training (Zhang et al., 2019; Dai et al., 2020). It has to the authors' best knowledge not yet been investigated whether the CPS is a reliable tool for orthopedic pain assessment from video recordings of lame dairy cows and how long it takes to assess a cow on a video recording.

The overall aim of this study was to evaluate whether the CPS can be used to detect pain behaviors in mildly to moderately lame dairy cows when they are stationary and to investigate whether cows with orthopedic pain express certain behaviors, and pain-related facial expressions, as described in the pain scale. The specific aims were to (1) investigate the association between total scores of the CPS and lameness scores; (2) investigate the association of specific CPS items and orthopedic pain; (3) investigate whether the pain-related facial expressions described in the CPS are present during orthopedic pain; and (4) assess the reliability of the CPS in video assessments conducted by blinded, trained observers. We hypothesized that CPS total scores would increase with higher lameness scores, and that cows with orthopedic pain would exhibit certain behaviors included in the CPS. We further hypothesized that the CPS would be reliable, but that the scale items "facial expression" and "ear position" would have a lower inter- or intrarater agreement (or both), and that the

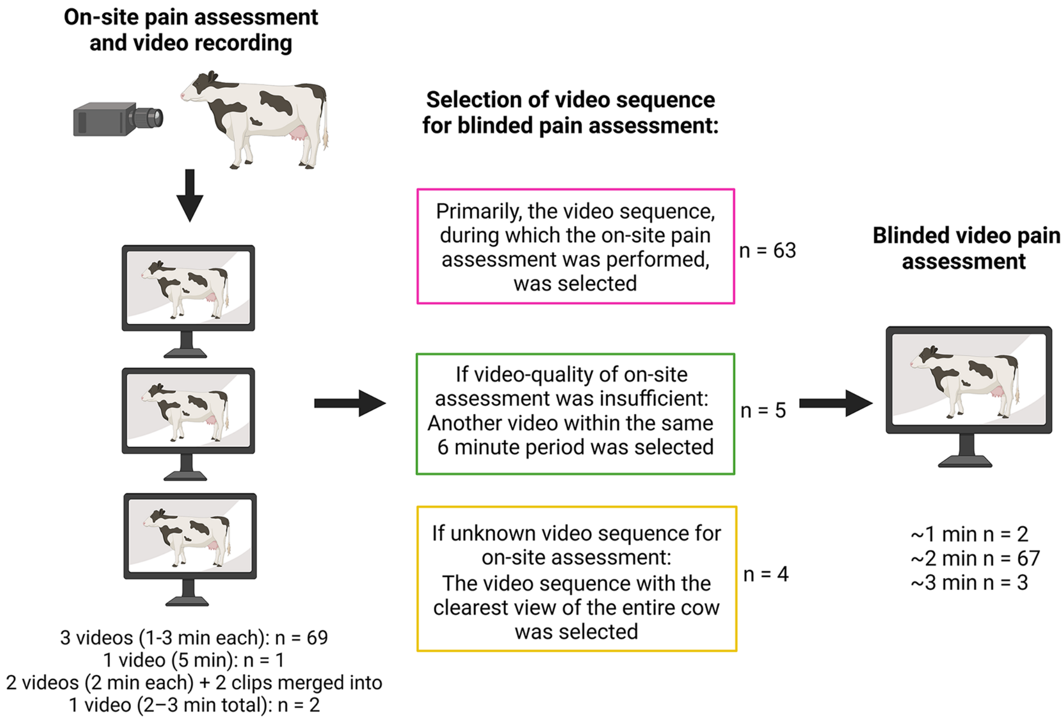

Figure 1. Flowchart showing underlying decisions regarding the selection of video recording for blinded pain assessment of each trial (in total 72 trials). Created by M. Söderlind (2025) in BioRender; https://BioRender.com/i64f407.

assessment time would be higher for initial compared with follow-up trials and during the first compared with the second assessment week.

MATERIALS AND METHODS

Animals and Study Design

This study was designed with consideration for the ARRIVE guidelines (Du Sert et al., 2020) and the 3Rs (Russell and Burch, 1959; Swedish 3Rs Center, 2024). The study protocol was approved by the Swedish Ethic Committee and followed the Swedish legislation of animal research (diary number 5.8.18-10570/2019 and 5.8.18-13069/2021).

The lameness cases were selected from a main dataset collected from 2020 to 2022 of adult dairy cows exhibiting clinical lameness of ≤ 3 on a 0 to 4 modified Sprecher lameness scale (Sprecher et al., 1997; Coetzee et al., 2014) as the only diagnosis. The cows were housed in a loose-housing system at the Swedish Livestock Research Centre, Swedish University of Agricultural Sciences (Uppsala, Sweden), with free access to water and roughage. The main data set included 80 lameness cases, with data from 2 occasions: the first obtained during clinical lameness (initial trial) and

Table 1. Overview of CPS total scores and item scores from video assessments included in inter- and intrarater reliability testing, situations where the reliability testing was applied (for all assessments, for each separate trial, and for each week of assessment), and statistical analyses performed

Reliability	Data	Situation	Statistical method
Interrater	CPS total scores	•All assessments •Initial and follow-up trial •First and second assessment week	ICC (2-way random effects, absolute agreement, single rater)
Intrarater	CPS total scores	•All assessments •Initial and follow-up trials	ICC (2-way random effects, absolute agreement, single rater)
Interrater	CPS item scores	•All assessments •Initial and follow-up trials	Krippendorff's α
Intrarater	CPS item scores	•All assessments •Initial and follow-up trials	Weighted kappa
Intrarater	CPS total scores	•Videos occurring twice during each week	ICC (2-way random effects, absolute agreement, single rater)

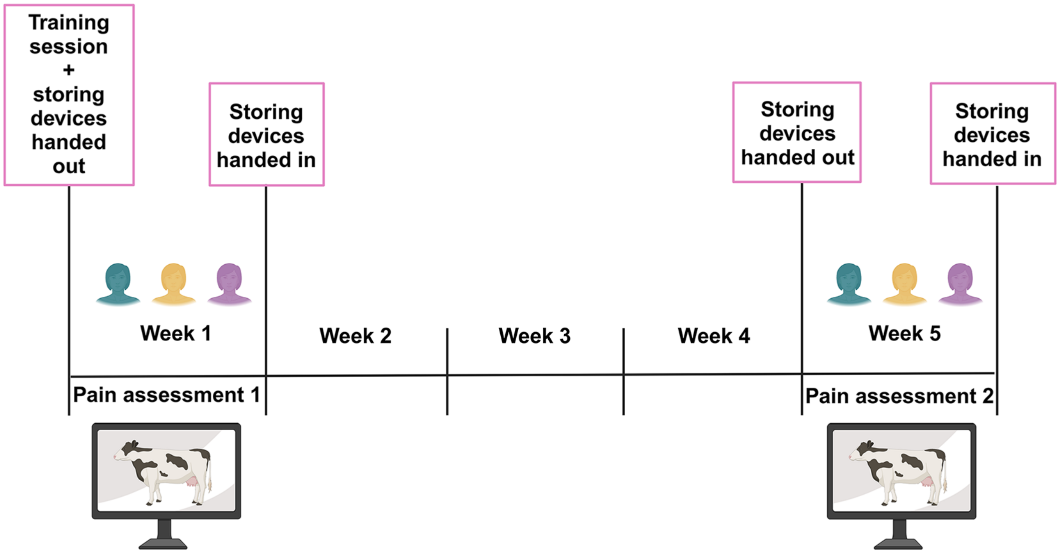

Figure 2. Timeline for blinded video pain assessments by 3 observers. A training session occurred at the start of the first week. Created by M. Söderlind (2025) in BioRender; https://BioRender.com/k92t953.

the second after treatment and clinical improvement (follow-up trial). Each trial included video recording, on-site pain assessments, clinical examination, lameness scoring, and claw examination, described in detail below. Exclusion criteria during the main data collection were severe aversive behaviors during handling or signs of another disease at the clinical examination. However, no cow exhibited such signs, and thus, none were excluded at this stage. A subset of these cases were selected for the current study, based on the following criteria: (1) an initial lameness score of ≤ 3 on a 0 to 4 modified Sprecher lameness scale (Sprecher et al., 1997; Coetzee et al., 2014), and improvement by ≥1 degree at the follow-up trial; (2) available high-quality video recordings from both trials of the cow (at least occasionally) standing with a visible back, head, ears, and face; and (3) available on-site pain scorings from both trials, obtained when the cow was standing. Twenty-four cases could not be included because it could not be ensured that the on-site pain assessment was performed when the cow was standing and 10 cases were not included due to loss of follow-up. Furthermore, 6 cases were excluded due to the lameness score not being improved sufficiently at the follow-up trial, 2 cases missed on-site pain scores, 1 missed video recordings, and 1 lacked visibility of the cow in the video recordings. The final data set included 34 dairy cows (23 Swedish Red and 11 Holstein) of varying parity (first to seventh) and lactation stage (11 to 366 DIM at the initial trial). Two of the cows were lame on 2 separate occasions, with at least 4 mo in between, resulting in 36 lameness cases (front limb: n = 8, hind limb: n = 28). Video recordings from the initial and follow-up trials in the final dataset were used for a subsequent blinded video pain assessment, described in detail below.

On-Site Pain Assessment and Video Recording

Pain assessments were performed using the CPS (Gleerup et al., 2015), which contains the following scale items: attention toward the surroundings, head position, back position, ear position, facial expression, and response to approach. Each item can be scored 0, 1, or, in some cases, 2, resulting in a total score of 0 to 10. Scale item definitions are listed in Supplemental Table S1 (see Notes). Before the first pain assessment, oral instructions on using the CPS and video recording were given by the lead researcher on site. Each cow was recorded with a handheld camera (Canon Legria HF R78) for about 6 subsequent min to obtain footage for the video pain assessment. In most cases (69/72), the recording process was divided into 3 sequences, resulting in three 1- to 3-min long videos (Figure 1). An on-site pain assessment was performed during one of these 1-to 3-min sequences by the person recording (veterinarian, n = 63, or a final-year veterinary student, n = 9). Most recordings (68/72) took place in the cows' normal loose-housing environment, except for one cow that was recorded in a sick box at the initial trial and 3 cows (1 at the initial trial and 2 at the follow-up) that were recorded in an enclosed aisle. In general, no interactions occurred between the observer and the cow before or during recording. However, if the cow was lying down, it was raised (n = 28), and if inaccessible to the camera, it was moved (n = 3). For 7 videos, it was unknown whether the cow had been raised or moved before the filming.

Clinical Examination and Lameness Scoring

After pain assessment and video recording, the cow was herded to a 35-m concrete-floored aisle where a veterinarian performed a full clinical examination to rule out obvious signs of concurrent pathology. An objective gait analysis (which will be described in another paper) was then performed as the cow walked up and down the aisle 1 to 3 times, depending on its motivation and lameness degree, while being recorded from the side using 3 wall-mounted cameras (GoPro HERO3+ and GoPro HERO7) and a handheld video camera (Canon Legria HF R78). For the present study, each trial was retrospectively lameness scored from the recordings by one veterinarian (author MS) using a modified version of the Sprecher lameness scale (0 = no lameness, 4 = severe lameness; Sprecher et al., 1997; Coetzee et al., 2014).

Claw Examination

An experienced claw trimmer conducted claw examinations in a trimming chute at both the initial and follow-up trials. During initial trials, claw examination followed the lameness evaluation. Identified lesions were classified according to the Nordic Claw Atlas (NAV, 2020), and treated according to standard routines. After visual improvement of the lameness (1–5 mo later), a new claw examination was performed by the same claw trimmer to further confirm lesion improvement. If the lesion was improved, a follow-up trial was executed. If the lesion was not improved, the follow-up was postponed and additional treatment was applied if necessary.

Video Pain Assessment

Selection of Video Recordings. A video sequence from each lameness case and trial (initial and follow-up) was selected for blinded video pain assessments. In most cases, the 2-min video recordings during which on-site pain assessments were performed were selected. The detailed selection process is illustrated in Figure 1. To allow for assessment of within-week intrarater agreement, 2 random videos from initial and 2 from follow-up trials were duplicated, resulting in 76 videos for blinded video pain assessment. To ensure blinding, the initial seconds of each video were muted because the person recording stated whether it was an initial or follow-up trial. Files were assigned an anonymized code and randomly ordered by a person not involved in the pain assessments.

Observer Training. From January to March, 2024, 3 veterinarians, experienced with cattle and pain assessments (authors MS, AL, and TÅ), conducted the video pain assessments, 2 of which (MS and AL) also participated in collecting data for the study and performed 13 and 19 of the included on-site pain assessments, respectively. Before video pain assessments, an on-site training session was held by author MS. The session lasted 1 h 20 min and covered pain scoring procedures and scale item definitions through both theoretical and practical training. Observers assessed 10 videos, not part of the assessment dataset, and compared scorings to clarify any issues. The presentation used in the training session is included in Supplemental Material S1 (see Notes).

Pain Assessment Procedure. Each video was assessed twice by all 3 observers during 2 separate weeks, with a 3-week "wash-out" period in between, where observers did not view or discuss the videos. The observers assessed the videos in the same random order and could rewind and rewatch videos if desired. However, once the next video was started, they were not allowed to go back or change previous scorings. Before the second week, videos were randomly reordered and renamed. The timeline for the video pain assessments is illustrated in Figure 2. Because "response to approach" could not be assessed from videos, this scale item was not scored during the video assessments. Instead, the scale item score from the corresponding on-site assessment was added to each video pain assessments' total score. Assessment times and scores were noted. Little guidance is provided in the CPS on duration and frequency of behaviors, both for each scale item and for each score (Gleerup et al., 2015). It leaves for the observer to decide when to score >0, which may affect interrater agreement. In the present study, a definition for scoring duration of behaviors was therefore introduced. Scale items were rated 1 or 2 if a behavior was seen more than once, more than briefly, or both. Briefly in this case meant less than approximately 3 s. Thus, if a behavior was only displayed once and immediately returned to normal after being noted by the observer, it was scored 0. If it was instead seen twice or more, or did not immediately return to normal after being noted by the observer, it was scored 1 or 2 (depending on the CPS item definition; see Supplemental Table S1). Because ears and facial expressions have been reported as dynamic in horses (Rashid et al., 2020; Ask et al., 2024), observers were instructed to estimate durations for facial expressions and ear position according to the following predefined categories: *seldom/short-term* (behavior seen more than once briefly but for a minor part of the observation time); *occasionally/to some extent* (behavior seen for more than a minor part, but less than half of the observation time); *repeatedly/prolonged* (behavior seen for approximately half of the observation time); and *fre-*

Table 2. Range of CPS total scores for video and on-site assessments at initial and follow-up trials, and assessment time (in min) per video for observers A, B and C[1]

Parameter	Situation	Minimum value	Q1	Median	Q3	Maximum value
CPS total scores	Initial trial video	1.00	2.00	4.00	5.25	9.00
	Follow-up trial video	0.00	1.00	2.00	4.00	9.00
	Initial trial on site	0.00	1.75	3.00	4.00	9.00
	Follow-up trial on site	0.00	0.00	0.00	1.00	4.00
Assessment time (min) per video	Observer A	2.00	4.00	5.00	7.00	17.0
	Observer B	2.00	3.00	4.00	5.00	12.0
	Observer C	2.00	3.00	4.00	5.00	10.0

[1]Data include median, minimum and maximum values, and the first (Q1) and third (Q3) quartiles.

quently/continuously (behavior seen for more than half of the observation time).

Statistical Analysis

All statistical analyses were conducted in R (v 4.3.2; R Core Team, 2023). The dataset consisted of 3 response variables, namely: CPS total scores from video assessments, CPS total scores from on-site assessments, and video scoring time. The Shapiro-Wilk test (Shapiro and Wilk, 1965), a skewness coefficient, and quantile-quantile (Q-Q) and residual plots were used to assess normality and homoscedasticity for each of the response variables. Normality was achieved by square root transforming the variables "CPS total scores video" and "CPS total scores on-site" and logarithmically transforming the variable "video scoring time," before further statistical modeling. Descriptive statistics included computation of the median and first and third quartile for each of the response variables above. Plots were produced using 'ggplot2' (Wickham, 2016). For the inferential statistics (described in the following sections), *P*-values < 0.05 were considered significant.

Cow Pain Scale Total Scores and Assessment Time. Linear mixed models (package lme4, function lmer; Bates et al., 2015) were applied to test the hypotheses that CPS total score increase with higher lameness score during video and on-site pain assessment, and that video pain assessment times differ with week and trial. For the first hypothesis, total scores (video/on site) were set as response variables and lameness score, ranging from 0 to 3, was included as a fixed effect with 4 levels. Whether or not the observer had interacted with the cow (i.e., raised or moved) before the pain assessment and video recording was included as a random effect, as were cow identity and, for the video model, observer also. For the second hypothesis, video assessment time was set as response variable, and observer, trial (initial/follow-up), and assessment week were included as fixed effects. All models were fitted with residual maximum likelihood estimation criterion, and Kenward-Roger approximation for degrees of freedom was applied. Multiple pairwise comparisons were made and estimated marginal means were computed (package emmeans, function emmeans; Lenth et al., 2018). From the fitted models with video total scores as the response variable, CPS total scores were predicted for each lameness grade, and parametric bootstrapping was performed (package lme4, function bootMer) to estimate 2-tailed, 95% CI.

Scale Items. To explore combinations of behaviors during orthopedic pain, associations between scale items and the initial trial (when lameness was present) were analyzed using multiple correspondence analysis (**MCA**; package FactoMineR; Lê et al., 2008). Scale item scores from video assessments were included in the analysis, except for the item "response to approach" where scores from the on-site assessment were included. To explain the variance within scale item scores, the MCA identified 10 dimensions. To test the hypothesis that cows with orthopedic pain would exhibit certain behaviors described in the CPS, 10 logistic mixed effects regression models were built with trial (initial/follow-up) set as the response variable, and dimensions included as fixed effects (e.g., model 1 containing dimension 1, model 2 containing dimension 1 and 2, and so on up to model 10). Interaction with cow before assessment, observer identity, and cow identity were included as random effects. Based on model reduction using Akaike's information criterion, model 9 including dimensions 1 to 9, which together explained 96.2% of the total variance, was selected for further interpretation. To facilitate interpretation, the coordinates of each scale item score were plotted using the package ggplot2. Positive coordinates indicate an association between a scale item score and the initial trial, that is, a behavior associated with orthopedic pain. Negative coordinates indicate a negative association with the initial trial, that is, behaviors more displayed at the follow-up trial. To evaluate whether pain-related facial expressions occurred more often in cows before than after treatment, descriptive statistics were applied.

Table 3. Results from three linear mixed effects regression models[1]

Model	Outcome variable	Effect (random/fixed)	Estimate	SE	*P*-value	Variance
Video assessment	CPS total score	Fixed: Intercept	1.59	0.151	<0.001	—
		Fixed: LS	0.158	0.0220	<0.001	—
		Random: cow identity	—	—	—	0.0860
		Random: observer	—	—	—	0.0180
		Random: cow interaction	—	—	—	0.0370
		Random: residual	—	—	—	0.212
On-site assessment	CPS total score	Fixed: Intercept	0.668	0.132	<0.001	—
		Fixed: LS	0.369	0.0810	<0.001	—
		Random: cow identity	—	—	—	0.000
		Random: cow interaction	—	—	—	0.000
		Random: residual	—	—	—	0.647
Assessment time (video assessment)	Assessment time	Fixed: Intercept	0.712	0.0180	<0.001	—
		Fixed: Second week	−0.0730	0.0150	<0.001	—
		Fixed: Initial trial	0.0540	0.0150	<0.001	—
		Fixed: Observer B	−0.128	0.0180	<0.001	—
		Fixed: Observer C	−0.133	0.0180	<0.001	—
		Random: cow identity	—	—	—	0.0020
		Random: residual	—	—	—	0.0250

[1]Video assessment and on-site assessment test the association between CPS total scores (from video and on-site assessments, respectively) and lameness score (LS). Assessment time tests the association between video assessment time and the first and second assessment week, trial (initial and follow-up trial), and observer (observer A, B, and C). The outcome variable in each model and the included fixed and random effects are stated. The estimate is only given for fixed effects, and the variance is given for random effects. *P*-values <0.05 were considered significant.

Reliability of Pain Assessments. Reliability of the video pain assessments was evaluated using different types of agreement statistics (Intraclass correlation [**ICC**]: package irrNA, function iccna; Brueckl and Heuer, 2022; Krippendorff's α: package irr, function kripp.α; Gamer et al., 2019; and weighted kappa: package irr, function kappa2; McHugh, 2012) described in Table 1. Intraclass correlation is recommended for continuous data (Bédard et al., 2000) and was hence used for the CPS total scores, and Krippendorff's α is more suitable for ordinal ranking data (Hayes and Krippendorff, 2007), and was hence used for the scale item scores. Weighted kappa was used to compute intrarater agreement for scale item scores.

RESULTS

The results showed that CPS total scores increased with lameness severity, and individual variations in behavioral combinations were present. Furthermore, a higher proportion of cows expressed pain-related facial expressions before compared with after treatment. The CPS total scores showed an overall moderate inter- and good intrarater agreement, with some variation across scale items, observers, and trials. In total 72 on-site and 456 video pain assessments were performed. Out of the 456 videos assessments, 24 were duplicates used solely for within-week intrarater agreement analysis of CPS total scores, leaving 432 video assessments, 216 per

Table 4. Estimated marginal means (emmeans) and 95% CI for CPS total scores from video and on-site assessments, and assessment times during the first and second assessment weeks[1]

Analysis	Explanatory variable	Emmeans	SE	Lower CI	Upper CI
Video pain assessment	LS 0	2.51	0.480	1.39	3.97
	LS 1	3.04	0.517	1.78	4.63
	LS 2	3.62	0.564	2.24	5.33
	LS 3	4.24	0.627	2.75	6.06
On-site pain assessment	LS 0	0.45	0.209	0.0211	1.42
	LS 1	1.08	0.223	0.00	4.97
	LS 2	1.98	0.355	0.990	3.30
	LS 3	3.15	0.687	1.84	4.81
Assessment time (video assessment)	Wk 1 follow-up	4.21	0.144	3.94	4.51
	Wk 2 follow-up	3.56	0.122	3.33	3.81
	Wk 1 initial trial	4.77	0.163	4.46	5.10
	Wk 2 initial trial	4.03	0.138	3.77	4.31

[1]Explanatory variables were those set as fixed effect in each linear mixed model respectively, that is, lameness scores (LS = 0–3) on a modified Sprecher scale, and assessment weeks (wk 1 and 2) for follow-up and initial trials.

week and trial and 144 per observer, to be included in the other analyses.

Diagnosis and Lameness Score

Lameness scores ranged from 1 to 3 out of 4 at the initial trial (score 1: n = 11, score 2: n = 10, score 3: n = 15) and from 0 to1 out of 4 at the follow-up (score 0: n = 30, score 1: n = 6). Sole ulcer was the most common diagnosis (n = 10), followed by claw abscess and foot rot (each n = 6). Details on lameness scores, front- or hindlimb lameness, and diagnoses are included in the Supplemental Table S2 (see Notes).

Pain Assessment Time

Assessment time for each video ranged from 2 to 17 min, with a median of 4 min (Table 2). All 3 observers completed the assessments within each assigned week, but observer A spent significantly more time than each of observers B and C. Across all observers, more time was also spent on initial trials compared with follow-up trials, and during the first compared with the second assessment week (Tables 3 and 4).

Cow Pain Scale Total Score

Video assessments showed a large range of CPS total scores for both initial and follow-up trials, and on-site assessments had a smaller range of CPS total scores for follow-up trials. The median and first and third quartiles are shown in Table 2. Video and on-site CPS total scores were significantly positively associated with lameness scores (Table 3); however, the estimated marginal means were lower and differed more between lameness scores for the on-site compared with the video assessment (Table 4). The unexplained (residual) variance accounted for 21.2% of the total variance for video assessments and 64.7% for on-site assessments (Table 3), Hence, the applied model captured a higher proportion of the total variance in the video dataset compared with the on-site dataset. Furthermore, the variables cow identity and cow interaction (included as random effects) did not explain any of the variance in the on-site dataset, but in the video dataset, they accounted for 8.6% and 3.7% of the variance, respectively. Predicted CPS total scores increased with lameness score; however, the prediction intervals were largely overlapping (Table 5).

Table 5. Predicted CPS total scores (TS) for LS of 0 to 3 on a modified Sprecher lameness scale, with 2.5% and 97.5% representing the lower and upper bounds of the CI

Lameness score	Predicted TS	Lower CI (2.5%)	Upper CI (97.5%)
0	1.59	1.30	1.86
1	1.74	1.49	1.97
2	1.90	1.64	2.15
3	2.06	1.76	2.36

Cow Pain Scale Items

The outcomes of the MCA (i.e., eigenvalues and explained variance for each dimension), and of the logistic mixed effects regression model (i.e., model 9 containing dimensions 1 to 9) are presented in Table 6. Dimensions 1 and 9 were positively associated with the initial trial, that is, when orthopedic pain was present, and dimensions 2 and 8 were negatively associated with the initial trial. Behaviors strongly associated with the initial trial in dimension 1, accounting for most of the variation, included reduced attention toward the surroundings (score 1), a low head position (score 2), pain-related facial expression (score 1), lowered ear position (score 2), reduced response to approach by observer (score 1 or 2), and an arched back position (score 2). Coordinates for each scale item score and dimension are presented in Supplemental Table S3 (see Notes). The level of association between each scale item and orthopedic pain is presented in Figure 3. The different dimensions showed different combinations of behaviors to be associated with orthopedic pain. In dimension 1, most items assigned a score >0 had positive coordinates and were thus associated with orthopedic pain. This illustrates how several behaviors may be present during orthopedic pain. In dimensions 2 and 8, items with positive coordinates, for example, ear position (score 2), instead showed a negative association with orthopedic pain, thus illustrating behaviors that may be less present during orthopedic pain. In dimensions 1 and 9, negative coordinates illustrated negative association with orthopedic pain, that is, behaviors were more displayed at follow-up trials (after improvement of lameness). In dimensions 2 and 8, negative coordinates instead illustrated positive association with orthopedic pain.

Pain-Related Facial Expressions

Observers assessed the CPS item facial expression for all initial and follow-up trials during the 2 assessment weeks (72 trials each in total); however, one observer did not score facial expression for one initial trial. This resulted in 215 initial trials and 216 follow-up trials with facial expression scores across the 2 assessment weeks and 3 observers. About half of the initial trials were scored as showing pain-related facial expression (score 1), compared with a quarter of the follow-up trials. Observer A noted more pain-related facial expressions than observers

Table 6. Dimensions 1 to 10 identified in the MCA[1]

Dimension	Eigenvalue	Variance (%)	Estimate	SE	z Value	*P*-value
Intercept	—	—	0.266	0.444	0.599	0.549
1	0.425	25.5	2.03	0.485	4.19	2.80e-05
2	0.204	12.2	−2.06	0.900	−2.29	0.0223
3	0.196	11.8	−0.466	1.04	−0.448	0.654
4	0.175	10.5	0.281	0.827	0.340	0.734
5	0.169	10.1	−0.217	0.464	−0.469	0.639
6	0.137	8.20	1.81	1.62	1.12	0.265
7	0.116	6.96	0.706	0.433	1.63	0.103
8	0.104	6.24	−3.25	0.785	−4.15	3.39e-05
9	0.0790	4.72	1.13	0.492	2.29	0.0218
10	0.0630	3.76	—	—	—	—

[1]Eigenvalues and variance in percentage (%) explained by each dimension are stated. Dimensions 1 to 9 were included in the final logistic mixed effects regression model. Estimates show associations with trial, where positive values indicate a positive association with orthopedic pain (initial trial), and negative values indicate a negative association. Variance of random effects: cow identity 0.925, interaction 0.303, observer 0.0130. *P*-values <0.05 were considered significant.

B and C, but all observers reported a higher proportion at the initial trial compared with the follow-up trial. The on-site pain assessment resulted in a similar proportion of pain-related facial expressions in initial trials, although it was notably lower in follow-up trials compared with the video assessments (Table 7). The duration of pain-related facial expression and lowered or backward ear position varied across cows in both trials, with each observer noting an even distribution across different time categories (Supplemental Figure S1; see Notes).

Reliability

The CPS total scores overall exhibited moderate inter- and good intrarater agreement between the 2 assessment weeks (Table 8). Intrarater agreement for duplicates within the same week ranged from moderate to excellent, varying by observer. Overall, follow-up trials showed stronger agreement than initial trials (good to excellent and moderate to good, respectively; Table 8). Agreement levels varied by scale item, with head position generally showing the strongest inter- and intrarater agreement (substantial and strong to almost perfect, respectively), whereas ear position showed the weakest (slight and weak respectively, Table 9). Back position showed a substantial interrater agreement and a moderate to strong intrarater agreement, and attention toward the surroundings and facial expression showed a moderate interrater agreement and a weak to moderate intrarater agreement (Table 9).

DISCUSSION

This is the first study to evaluate the CPS for detection of orthopedic pain behaviors in stationary dairy cows with mild to moderate lameness. On-site unblinded pain assessments and blinded pain assessments from video recordings of the same cows were included, and the CPS was found to be reliable in detecting orthopedic pain by video assessment. Behavioral combinations varied among cows, suggesting CPS total scores to be more indicative of orthopedic pain than individual scale items. Furthermore, the pain-related facial expression described in the CPS may indicate orthopedic pain in stationary lame cows, but inconsistencies in this item's display suggests that more research on bovine pain-related facial expressions is needed.

The CPS total scores from both on-site and video pain assessments were significantly positively associated with lameness scores, and higher CPS total scores were seen for higher lameness severity, confirming the hypothesis that the total score would increase with higher lameness score. However, there was a considerable overlap in confidence intervals, indicating large interindividual variability. Furthermore, the predicted pain score was very similar (approximately 2 out of 10) for all lameness degrees (1 to 3 out of 4). A CPS total score of 3 has previously been suggested as a cut-off value and hence indicative of clinical or postsurgical pain (Gleerup et al., 2015; Tomacheuski et al., 2023), but according to our results, using this threshold would likely lead to failure in detecting animals with mild to moderate orthopedic pain. Similar complexity has been observed in horses with induced orthopedic pain, where movement asymmetry (measured objectively at a trot) and total pain scores (assigned when the horses were resting) showed a nonlinear relationship despite the fact that the source of pain was the same in all horses (Ask et al., 2022). In that study, it was estimated that only during moderate lameness could one anticipate a total pain score above 0. Hence, our results add to the evidence of a highly complex relationship between lameness score and pain intensity while stationary. Furthermore, several cows in the present study had claw lesions in multiple limbs and the orthopedic lesions causing the

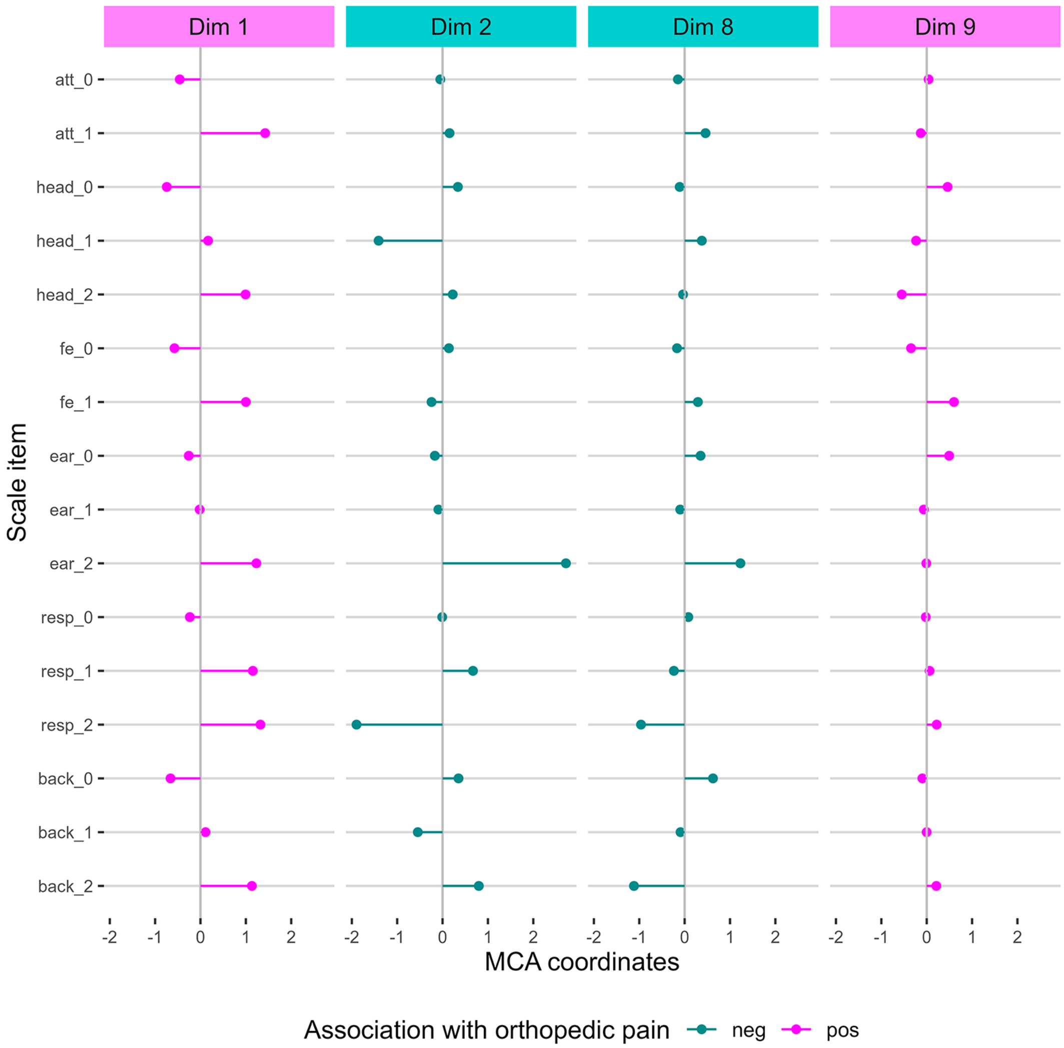

Figure 3. Coordinates from the multiple correspondence analysis (MCA) for scale items in the CPS, distributed across 4 significant dimensions (Dim 1, 2, 8, and 9). Teal-colored dimensions are positively associated with the initial trial where each cow was lame (pos); positive values indicate association with lameness and negative values indicates the opposite. Orchid-colored dimensions are negatively associated with the initial trial (neg); positive values indicate association with the follow-up trial with improved lameness and negative values the opposite. Higher coordinate values indicate a stronger association within the dimension. The following scale items are included: attention toward the surroundings score 0 and 1 (att_0, att_1); head position score 0, 1, and 2 (head_0, head_1, head_2); facial expression score 0 and 1 (fe_0, fe_1); ear position score 0, 1, and 2 (ear_0, ear_1, ear_2); response to approach score 0, 1, and 2 (resp_0, resp_1, resp_2); and back position score 0, 1, and 2 (back_0, back_1, back_2).

lameness were of different types and chronicity. Lesions in multiple claws could imply difficulties in decreasing the load on the sore foot while stationary, potentially causing more pain while stationary compared with being affected by a single claw lesion. In cattle, chronic claw lesions have previously been shown to result in higher posture scores during locomotion compared with acute lesions (O'Callaghan et al., 2003), possibly explained by long-lasting hyperalgesia that can be caused by these lesions (Whay et al., 1998). Chronic orthopedic lesions could potentially also result in increased pain-related behaviors at rest compared with acute lesions. Chronic pain has, in rodents, been shown to induce anxiety- and depression-like behaviors (Cunha et al., 2020; Hsu et al., 2021), which instead could complicate detection of pain-

Table 7. Proportion of trials where cows displayed pain-related facial expressions (score 1 on the item facial expression in the CPS) according to blinded video observers A, B, and C individually and combined (total), as well as the on-site (not blinded) assessment

Assessment	Initial trial	Follow-up trial
Observer A	0.708 (51/72)	0.319 (23/72)
Observer B	0.375 (27/72)	0.194 (14/72)
Observer C	0.437 (31/71)	0.181 (13/72)
Total	0.507 (109/215)	0.231 (50/216)
On-site assessment	0.528 (19/36)	0.0833 (3/36)

Table 8. Inter- and intrarater agreement for CPS total scores, scored by three blinded observers (A, B, C) on video recordings of initial and follow-up trials, and combined (total)[1]

Analysis	Situation	ICC	CI lower limit	CI upper limit	Level of agreement[2]
Interrater agreement video assessment	Total	0.703	0.575	0.792	Moderate
	Initial trial	0.583	0.344	0.740	Moderate
	Follow-up	0.755	0.658	0.831	Good
	First week	0.692	0.488	0.814	Moderate
	Second week	0.720	0.607	0.808	Moderate
Intrarater agreement, observer A	Total	0.841	0.722	0.906	Good
	Initial trial	0.792	0.628	0.888	Good
	Follow-up	0.814	0.591	0.911	Good
	Duplicates	0.836	0.428	0.964	Good
Intrarater agreement, observer B	Total	0.808	0.711	0.876	Good
	Initial trial	0.715	0.513	0.842	Moderate
	Follow-up	0.792	0.628	0.888	Good
	Duplicates	0.647	0.047	0.915	Moderate
Intrarater agreement, observer C	Total	0.889	0.829	0.929	Good
	Initial trial	0.843	0.717	0.916	Good
	Follow-up	0.926	0.860	0.961	Excellent
	Duplicates	0.911	0.653	0.981	Excellent

[1]Scorings from both assessment weeks are included. Interrater agreement during first and second assessment weeks and intrarater agreement from duplicated videos (duplicates) within the same assessment week are also stated.

[2]Level of agreement according to Koo and Li (2016).

related behaviors. Whether this also occurs in cattle is yet to be explored, and further studies on pain behaviors in cattle with chronic pain are warranted.

In the present study, we performed both on-site and blinded video pain assessments in cattle. In agreement with previous studies of young bulls (Tomacheuski et al., 2024) and piglets (Trindade et al., 2023) undergoing castration, lower pain scores were seen for on-site compared with video assessments. Tomacheuski et al. (2024) suggested that on-site observations might be less suitable for detecting dynamic behaviors, compared with video assessments where it is possible to rewatch sequences. In Trindade et al. (2023), it was proposed that on-site assessments might cause more observer fatigue due to busy surroundings and the need for constant focus throughout the observation. In the present study, a lower on-site CPS total score for lameness degree 0 could indicate an expectation bias. However, one could then also expect a higher score for the higher lameness degrees, which was not the case. Instead, dynamic behaviors such as facial expression and ear position (Rashid et al., 2020; Ask et al., 2024), being more difficult to detect by on-site compared with video assessment, and distractions from the surroundings during the on-site assessment may explain also the difference seen in the present study. However, the fact that video observers in the present study, unlike the on-site observers, received specific training including scoring definitions of behavior duration, and that the video and on-site assessments in many cases were performed by different persons makes direct comparison difficult.

To the authors' best knowledge the time needed for pain assessments on video has not previously been studied, although it is important for implementation of a pain scale in clinical situations (Reid et al., 2007). In the present study, the time spent on video pain assessments varied among observers and decreased from the first to the second assessment week. Factors contributing to this variation may include individual differences in observers' time budgets, thoroughness, or perceived difficulty of assessments. Also, an increased familiarity with the assessment procedure may explain the reduced assessment time during the second week, suggesting observer experience to be important for the feasibility of the pain scale. Furthermore, initial trials required more time than follow-up trials, indicating that assessing behaviors in pain-affected animals is more challenging than in healthy ones.

In the present study, we further explored relationships between CPS items. Each item represents different behaviors, and we tested whether certain combinations of behaviors were more strongly associated with orthopedic pain than others. Several behavioral combinations were associated with the initial trial (when orthopedic pain was present), and all CPS items were relevant for distinguishing between cows with orthopedic pain (initial trial) and after improvement (follow-up trial). This result is in agreement with previous findings of the association between CPS items and postsurgical pain (Tomacheuski et al., 2023).

A reduced response to approach by an observer (score 2) was associated with orthopedic pain, but this item has shown a lack in specificity for surgical pain when assessed from videos (Tomacheuski et al., 2023), suggesting that this behavior may vary by pain type. However, because this item was the only one in the present study that was scored solely by nonblinded observers during on-site assessment, expectation bias may have influenced the results. Although back position has been found not to be sensitive for postsurgical pain (Tomacheuski et al., 2023), we found that a straight back position (score 0) was associated with the follow-up trial (after improvement), and an arched back position (score 2) was associated with orthopedic pain in dimensions 1 and 8. This is not surprising considering the emphasis put on back position in the Sprecher lameness scale used for this study (Sprecher et al., 1997; Schlageter-Tello et al., 2014a), and further confirms the importance of evaluating scale performance for different pain types specifically. On the other hand, dimension 2 displayed an association between an arched back position (score 2) and the follow-up trial (after improvement). Also ear position showed inconsistent associations with the initial or follow-up trials, in line with earlier studies in cattle linking lowered or backward ears to positive low arousal states (Proctor and Carder, 2014) as well as pain (Gleerup et al., 2015; Müller et al., 2019; Farghal et al., 2024) in cattle. A recent study on induced mastitis in cows found backward ears to be less frequent 8 h after, compared with 16 h prior to, mastitis induction (Ginger et al., 2023), further underlining the uncertainty of using ear position as an independent pain indicator. Thus, behaviors expressed by cows with mild to moderate lameness are diverse and complex. Importantly, some pain behaviors may be shown by animals not experiencing pain and vice versa, which is why the CPS total score is likely more indicative of orthopedic pain than the individual scale items.

Similar variation in the association between pain-related facial expression (score 1) and orthopedic pain was identified. To facilitate understanding of this finding, the proportion of scored pain-related facial expressions at the initial and the follow-up trials, respectively, was compared. In approximately half of the initial trials, cows showed a pain-related facial expression, suggesting that although the presence of pain-related facial expressions described in the CPS may indicate orthopedic pain (as seen in dimensions 1 and 9) their absence does not equal the absence of pain.

In contrast, in about a quarter of the follow-up trials, cows showed a pain-related facial expression. This cannot be entirely explained by the fact that some cows (n = 6) were still mildly lame at the follow-up and one may ask why a bovine pain face is present when the cow is most probably free from pain. In a study on horses with acute pain (Rashid et al., 2020), exhaustive registration of all facial activities with the equine facial action coding system was performed. In 10-s video clips of horses without pain, 30% contained combinations of facial activities also seen during pain, illustrating the intricacy of pain-related facial expressions. However, there are several recent studies confirming changes in facial expressions in cattle experiencing acute pain from different sources (Müller et al., 2019; Ginger et al., 2023; Farghal et al., 2024). Although these findings to some degree align with the description in the CPS, notable differences were also observed. In Müller et al. (2019), dilated nostrils, open mouth, and raised inner and outer eyebrows were associated with pain during branding of beef cattle. Similar findings were obtained in a study on calves undergoing castration with the addition of straining of the chewing muscle and orbital tightening (Farghal et al., 2024). In Ginger et al. (2023) dairy cows with induced mastitis were found to show significantly less muzzle movement and less time with open eyes, and a tendency for more dilated nostrils, increased time with a motionless muzzle, decreased blink frequency, and reduced eye movement. Although some of these facial activities may resemble features described in the CPS, this suggests that pain-related facial expressions in cattle are likely more intricate and dynamic than the scale's description conveys. To enhance the applicability of facial expressions as an indicator of pain in cattle, further studies on the dynamics of these expressions in various painful conditions, including chronic pain, are needed.

The scoring threshold introduced in the present study, could potentially have led to overestimation of behaviors, particularly dynamic ones like facial expressions and ear positions. To address this, observers noted the duration of these 2 behaviors, revealing that they varied among cows, being either brief and fluctuating or more constant when scored at both initial and follow-up trial. The definitions chosen were, however, subjectively and somewhat broadly described, which is why the results should be interpreted with caution. Nevertheless, it underscores the need to use a scoring threshold and also implies that different thresholds could yield different results, highlighting the need for further investigation into the dynamic properties of facial expressions and ear positions in cattle.

The present study found a moderate interrater agreement, and a moderate to excellent intrarater agreement for CPS total scores, supporting our hypothesis that the CPS is reliable and also aligning with previous findings (Gleerup et al., 2015; Tomacheuski et al., 2023). Agreement was stronger for follow-up trials, indicating that it may be easier to agree on cows showing fewer pain behaviors. We further showed that agreement varies across scale items, suggesting some behaviors are harder to assess. Head position and back position showed the strongest agreement, and attention toward the surroundings, facial expression, and ear position showed lower inter- and intrarater agreement. This is in line with a previous study on horses with orthopedic pain where body behaviors showed higher agreement than

Table 9. Interrater agreement (Krippendorff's α, α_K) and intrarater agreement (weighted kappa, K_w) for each item in the CPS, scored by blinded observers (A, B, C) on video recordings during initial and follow-up trials, and both trials combined (total)

Analysis	Scale item[1]	Situation	Coefficient value	*P*-value	Level of agreement[2]
Interrater agreement video assessments (α_K)	Att	Total	0.519	NA	Moderate
		Initial trial	0.513	NA	Moderate
		Follow-up	0.510	NA	Moderate
	Head	Total	0.643	NA	Substantial
		Initial trial	0.459	NA	Moderate
		Follow-up	0.768	NA	Substantial
	Back	Total	0.667	NA	Substantial
		Initial trial	0.598	NA	Moderate
		Follow-up	0.593	NA	Moderate
	Ear	Total	0.198	NA	Slight
		Initial trial	0.183	NA	Slight
		Follow-up	0.205	NA	Fair
	Face	Total	0.444	NA	Moderate
		Initial trial	0.376	NA	Fair
		Follow-up	0.429	NA	Moderate
Intrarater agreement, observer A (K_w)	Att	Total	0.622	<0.0001	Moderate
		Initial trial	0.625	<0.0001	Moderate
		Follow-up	0.599	0.0003	Weak
	Head	Total	0.822	<0.0001	Strong
		Initial trial	0.849	<0.0001	Strong
		Follow-up	0.707	<0.0001	Moderate
	Back	Total	0.746	<0.0001	Moderate
		Initial trial	0.669	<0.0001	Moderate
		Follow-up	0.755	<0.0001	Moderate
	Ear	Total	0.505	<0.0001	Weak
		Initial trial	0.578	0.0003	Weak
		Follow-up	0.400	0.0024	Weak
	Face	Total	0.669	<0.0001	Moderate
		Initial trial	0.562	0.0004	Weak
		Follow-up	0.664	<0.0001	Moderate
Intrarater agreement, observer B (K_w)	Att	Total	0.467	<0.0001	Weak
		Initial trial	0.721	<0.0001	Moderate
		Follow-up	0.321	0.0253	Minimal
	Head	Total	0.832	<0.0001	Strong
		Initial trial	0.898	<0.0001	Strong
		Follow-up	0.742	<0.0001	Moderate
	Back	Total	0.785	<0.0001	Moderate
		Initial trial	0.748	<0.0001	Moderate
		Follow-up	0.752	<0.0001	Moderate
	Ear	Total	0.417	0.0001	Weak
		Initial trial	0.509	0.0006	Weak
		Follow-up	0.325	0.0354	Minimal
	Face	Total	0.557	<0.0001	Weak
		Initial trial	0.645	0.0001	Moderate
		Follow-up	0.467	0.0049	Weak
Intrarater agreement, observer C (K_w)	Att	Total	0.704	<0.0001	Moderate
		Initial trial	0.748	<0.0001	Moderate
		Follow-up	0.667	<0.0001	Moderate
	Head	Total	0.919	<0.0001	Almost perfect
		Initial trial	0.937	<0.0001	Almost perfect
		Follow-up	0.901	<0.0001	Almost perfect
	Back	Total	0.827	<0.0001	Strong
		Initial trial	0.819	<0.0001	Strong
		Follow-up	0.791	<0.0001	Moderate
	Ear	Total	0.561	<0.0001	Weak
		Initial trial	0.673	<0.0001	Moderate
		Follow-up	0.333	0.0338	Minimal
	Face	Total	0.539	<0.0001	Weak
		Initial trial	0.719	<0.001	Moderate
		Follow-up	0.364	0.0312	Minimal

[1]Scale items: Att = attention toward the surroundings; head = head position; back = back position; ear = ear position; face = facial expression. Both assessment weeks are included. NA = not applicable.

[2]Level of agreement for Krippendorff's α (α_K) according to Landis and Koch (1977); level of agreement for weighted kappa (K_w) according to McHugh (2012).

facial expressions (Ask et al., 2020). The 5 scale items scored during video assessment were likely not assessed simultaneously, but rather one by one throughout the video. Therefore, one reason for the lower agreement could be the more intermittent display of facial and ear behaviors and variation between and within observers in regard to when during the video these scale items were assessed. Additionally, insufficient, or subjective definitions for some scale items, leaving room for the observers' own interpretations, may contribute to lower agreement. For example, ear position lacks a definition for intermediate positions and facial expression definitions (e.g., "worried or strained look," see Supplemental Table S1) allow for subjective interpretation of what the cow is expressing. Furthermore, attention toward the surroundings, head position, and facial expression all include eating in the score of 0, which may lead to varying scores among observers when a cow shows pain-related behaviors while eating.

The present study has several limitations. The study population was limited to adult dairy cows of the breeds Holstein and Swedish Red. Because individual variations in personality (Ijichi et al., 2014), age, and sex (Guesgen et al., 2011) may influence on pain expression, repeated studies including different breeds, ages, and sexes would enhance implementation of the results on a wider population. Parity and lactation status varied across cows and trials in the present study, and to evaluate the influence of these factors on pain behaviors, further studies, preferably with larger datasets, are needed. In the present study, the Sprecher lameness scoring was performed once by a nonblinded observer. Previous studies have shown varying reliability in subjective lameness scoring of cattle (Winckler and Willen, 2001; Channon et al., 2009; Schlageter-Tello et al., 2014b), and thus it is possible that another observer would have assigned the cows different lameness scores. In the future, objective lameness measures would add value in establishing lameness degree. To minimize disturbance to the cow, on-site pain assessments were conducted by a single observer during each trial, and given the extended duration of the data-collection period, observers varied and lacked standardized training. One may also consider a potential observer influence on bovine pain behaviors. Observer presence has been shown to reduce discomfort behaviors in horses (Torcivia and McDonnell, 2020), and to increase pain scores at baseline-measures and decrease pain scores in pain-time points in rabbits (Pinho et al., 2023), both being prey animals like cattle. In the present study, observers were instructed to act calmly, be quiet, stand a few meters away, and not interact with the cow before or during the video recording, to minimize the observer influence. However, some cows had to be raised or moved by the observer, possibly inducing stress, before the pain assessment. Stress may have an effect on pain perception (Butler and Finn, 2009; Jennings et al., 2014), and in horses, stress has shown to induce facial activities similar to those occurring during pain (Lundblad et al., 2021). Future studies are therefore warranted to assess the effect of stress and observer presence on pain behaviors in cattle.

CONCLUSIONS

The present study showed that the CPS is reliable for assessing orthopedic pain in video recordings, and can be used to detect orthopedic pain in stationary lame dairy cows by both video and on-site assessment. However, a large overlap in pain scores across different lameness degrees was identified, indicating a complex relationship between pain score and pain intensity. A distinct behavioral pattern associated with orthopedic pain could not be revealed; instead, cows in orthopedic pain expressed different combinations of behaviors. Therefore, single behaviors within the CPS should be interpreted with caution. Furthermore, a pain-related facial expression described in the CPS was observed during lameness; however, future research on more detailed and dynamic features of facial activities related to orthopedic pain are needed.

NOTES

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Nonstandard abbreviations used: att_0, att_1 = attention toward the surroundings scores of 0 and 1; back_0, back_1, back_2 = back position scores of 0, 1, and 2; CPS = Cow Pain Scale; Dim = dimension; ear_0, ear_1, ear_2 = ear position scores of 0, 1, and 2; emmeans = estimated marginal means; fe_0, fe_1 = facial expression scores of 0 and 1; head_0, head_1, head_2 = head position scores of 0, 1, and 2; ICC = intraclass correlation coefficient; LS = lameness score; MCA = multiple correspondence analysis; NA = not applicable; neg =negative; pos = positive; Q1 = first quartile; Q3 = third quartile; resp_0, resp_1, resp_2 = response to approach scores of 0, 1, and 2; TS = CPS total score.

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