

Monitor and Evaluate Calves' Health and Welfare

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Precision livestock farming (PLF) research is rapidly increasing and has improved farmers' quality of life, animal welfare, and production efficiency. Automatic milk feeding systems (AMFS) and 3D accelerometers have been the most extensively used technologies in dairy calves.

management

precision livestock farming

automatic milk feeding

1. Introduction

Increasing efficiency in livestock production is needed to ensure that enough food can be produced to meet the demands of a growing population, help reduce the environmental impact, and ensure that food is available and affordable ^{[1][2][3]}. Precision livestock farming (PLF) has the potential to increase efficiency in livestock production by providing farmers with the tools they need to make data-driven decisions and optimise their management practices. This can lead to improved animal welfare, increased productivity, and reduced environmental impact ^{[4][5]}. Besides the importance of the environmental impact, technological development can improve animals' health and welfare and farmers' quality of life ^[6]. PLF employs real-time monitoring technologies to help or automatically intervene in animal management and measure individual data with time variability ^{[7][8][9]}. According to Eckelkamp ^[10], PLF was initially developed in poultry and swine growing operations. Applying these technologies to other farm animals is now very common, especially in more confined production systems.

Dairy cows are considered high-value animals, and thus, applying PLF technologies at an individual level can justify the additional expenses ^[11]. PLF technologies applied to dairy farms have been called precision dairy farming (PDF), employing technology to measure physiological, behavioural, and production indicators to improve management and performance ^[12]. PLF has been more directed towards adult animals; however, increased animal welfare concerns and technology development led to an increased technology expansion for younger animals as well, such as dairy calves. In fact, according to Stygar et al. ^[13], future research should focus on developing and validating PLF technologies dedicated to monitoring calves and heifers' health and welfare. Sun et al. ^[14] reviewed the current automated techniques to monitor calves' health, discussing the possibility of automatically detecting less resilient calves (clinically healthy but prone to disease).

The global trend for dairy farms is to increase in size and decrease in number, so PLF technologies can help in a manner that provides individual information about calf growth and health status. The number of papers addressing the PLF application in calves has been increasing in the last few years; however, there is a lack of agreement in

several results published so far, stressing that more validation studies are still needed [9]. Nonetheless, a review and a confrontation of these results should also highlight which technologies are still in need of proper validation studies and which technologies need more field application studies in order to increase their utilisation at the farm level, where it can actively improve the calves' welfare and hence, their performance.

In dairy calves raising systems, technology can be implemented in health evaluations, behaviour assessments, milk, water, feed intake, and growth measurements. Nonetheless, technologies related to environmental conditions should also be considered [15], despite not being specific to dairy calves. Compared with adult dairy cows, heifers are managed in a less intensive system; however, disease prevalence is considerably high, especially in the preweaning phase [16][17]. Enterogastric, pulmonary, and umbilical disorders are dairy calves' major health problems [18][19][20][21][22]. Although several advances have been made in providing proper treatments to ill calves, the early detection of these diseases and mortality rates are still challenging due to the absence of knowledge and, in some cases, time to monitor the animals. Nevertheless, PLF technologies that can be used as a prognosis for calves' diseases, although lacking in validation studies [9], have been revealing promising results [23][24][25]. According to Costa et al. [9], the most researched PLF technologies for managing the performance and health of pre-weaned dairy calves are triaxial accelerometers, automatic milk feeding systems (AMFS), and technology that measures physiological or physical attributes, such as infrared imaging and 3-dimensional cameras.

2. Technological Applications to Monitor Calves' Health and Welfare

Current technology research to monitor and manage dairy calves is mainly related to health disorders and painful procedures. However, the literature addresses that good animal welfare should not be viewed as merely the absence of pain, discomfort, or hunger but by the quality of life and emotional states. According to Fraser [26], animal welfare should contemplate three views: essential health and functioning, natural adaptations, and affective states. Near future research must also focus on evaluating other welfare standards, such as the absence of fear and the presence of positive emotional states. A good example is a PhD thesis published in 2021, where the positive and negative emotions of calves were studied with infrared thermography by analysing temperature asymmetries in different regions of interest, such as the difference in temperature between the left and right-side hemispheres [27]. Studying farm animals' emotions would allow researchers to better understand how these animals perceive and interact with the environment [27], including their interaction with humans. In this way, a better quality of life could be achieved if the economics and social aspects inherent to animal production could be overcome.

Although more studies are still needed in this area, noninvasive precision techniques to access animals' emotional states can provide the knowledge needed to improve farm animals' welfare. Studies with calves, AMFS, and triaxial accelerometers make up the majority of the literature in this area and have already been properly reviewed [9]. However, there are other emerging technologies and different approaches that have not yet been extensively reviewed, e.g., 3D cameras for body measurements and IRT to evaluate acute and prolonged pain situations. One of these recent approaches, which has been gaining popularity, is the simultaneous integration of different

technologies to monitor and evaluate the calves' health status, performance, and pain since it can provide data from different sources, which, in turn, measures different physical, physiological, and behavioural traits. Usually, this multi-technological approach generates much more data that can be more easily processed using machine learning and deep learning techniques rather than traditional statistics.

2.1. Automatic Milk Feeding System

Automatic milk feeding system (AMFS) is probably the most extensively used technology in dairy calves, which can significantly increase the efficiency of dairy calves' raising systems [28]. AMFS can feed multiple calves several times per day without increasing labour requirements [28]. Calves should be fed several times a day since excessive quantities of milk provided at once can increase the risk of dietary diarrhoea. In the AMFS, the daily dose is distributed throughout the day, so higher milk feeding levels can also be achieved compared to the manual system. It has been demonstrated that increasing milk allowance with this strategy can improve calves' performance [29][30][31]. AMFS also makes it possible to do automated gradual weaning, which contributes to minimising post-weaning stress and thus achieving better performances [32].

To function with the AMFS (**Figure 1**), an electronic identifier must be attributed to each calf. Then, relevant information from the calf can be registered in the AMFS software, such as the birth date and body weight. Then, a program regarding milk allowance per day and days being fed is selected for each animal. During the selected period, the calf can drink a certain amount of milk distributed throughout the day. To provide a more distributed feeding throughout the day, the AMFS blocks consumption between consecutive visits, predefined in the software. The AMFS software records individual information about the drinking behaviour of every calf with an active responder. However, the maximum number of calves per unit depends on the machine's capacity. So, it is crucial to be aware of the machine's functionality, such as calibrating milk powder doses, checking for the thermostat accuracy, water flow, etc.



Figure 1. Automatic milk feeding system: (A) shows the control panel, where all the information relative to the calves and the machine is registered and controllable; (B) shows all the components (except the RFID reader and the teat, which are inside the calves' barn); in (C) shows the milk powder deposit; (D) is the mixer cup, where each portion of milk is prepared with milk powder and heated water; in (E) is shown the circuit in which the milk departures from the mixing cup to the teat inside the calves' barn.

Like other automatic monitoring technologies, feeding calves through a robotised system can decrease human-animal interactions, which may not be favourable [33]. An increase in the physical distance between the farmer and the animal could lead to animals becoming more reactive when handled [33]. Older calves (without an active responder) may usurp the younger calves' portion of milk by means of displacement behaviours when groups are not properly managed. The cost of acquisition and maintenance of the machine, which may not be accessible to every farmer, can also be a downside. Nevertheless, AMFSs are well recognised for providing helpful information to monitor calves' health, performance, and welfare [9].

Daily milk intake, rewarded and unrewarded visits to the feeder, and drinking speed are the three most studied indicators. The use of AMFS to predict the onset of diseases and to evaluate welfare has been successfully tested [24][25][28][34][35][36][37]. Sick calves showed less milk intake, lower drinking speed, and a decrease in unrewarded visits from -2 to -4 days before diagnosis [23][24][34][36][37] that varied with milk allowance level [25][35]. Calves fed with higher milk volumes tend to change their feeding behaviour [25][35]. Cantor et al. [38] used AMFS alarms triggered when the calf reduced 20% of the average milk intake and 30% of average drinking speed based on the past 12 d mean to evaluate the administration of colostrum replacers in health and performance improvement. Conboy et al. [39], in a cross-sectional study involving 523 observations, reported a 63% reduction in daily milk intake of calves with bovine respiratory disease (BRD) and 57% in calves with neonatal calf diarrhoea (NCD). In another study, calves with NCD had a significant decrease in milk intake on the day of diagnosis, 5 days after (observed daily by faecal consistency), and a tendency for -1 d ($p = 0.09$); as for rewarded visits, diarrheic calves presented fewer visits at -1 d and 0 d than healthy calves, but more visits at 3 d and a tendency at 4 d [40]. Moreover, the parallel sensitivity and specificity on the day of diagnosis using milk intake and rewarded visits were not satisfactory (Se of 69% and Sp of 22%). The authors of this study reported that data from the AMFS could not replace the identification of calves with NCD through clinical examination.

One of the animal welfare's "Five Freedoms" is "Freedom from hunger and thirst"; AMFS indicators may help provide information about prolonged hunger in preweaning calves [29][30]. AMFS may also be used to study calves' preferences for milk composition. For example, it was shown that calves had a preference for whole milk over milk replacers [41]. AMFS information was also used to associate temporal between individual differences in feeding behaviours with personality traits, highlighting two distinct groups: calves that visited the machine more often had a superior drinking speed and grew faster, and calves that visited the machine fewer times had an inferior drinking speed and grew slower (sick calves were excluded from the analysis) [42]. Nevertheless, it should be noted that calves can present a distinct individual behavioural rhythmicity throughout the day [43].

2.2. Triaxial Dimension Accelerometers

An accelerometer measures proper acceleration, given by the body's velocity per unit of time in its instantaneous rest frame. Triaxial accelerometers (3D accelerometers) measure proper acceleration simultaneously in three orthogonal directions (x, y, and z) at regular time intervals (10 to 20 Hertz seems to be the most efficient frequency [44]). The generated data can be stored in a data logger or a smartphone or used raw with a commercial system applied [44]. Three-dimensional accelerometers provide information in a three-dimensional plane, unlike unidirectional accelerometers. It records the acceleration forces produced by the animal movement, which are associated with certain behaviours (e.g., lying, standing, suckling, ruminating, and chewing), allowing automated real-time monitoring of the animal's activity, health, and welfare when combined with a proper algorithm. However, certain behaviours are more difficult to predict than others (e.g., standing up and lying down), decreasing the prediction accuracy of these behaviours [44]. In dairy farms, accelerometers are well established as a valuable tool to provide information about the oestrus cycle in cows [45][46][47]. However, practical application in dairy calves is still limited, being used only for research purposes. The battery drainage can also be a limitation, which is related to the sampling frequency and can go from 5 days [48], 15 days [49], to 2 years [50].

The use of 3D accelerometers has been used for a variety of topics within calves' welfare evaluation. Accelerometers have a fundamental basis for behaviour analysis, such as lying and locomotor behaviours. Gait pattern analysis was first successfully achieved with 3D accelerometers by de Passillé et al. [51]. Nevertheless, lying behaviour is regarded as a primary indicator for welfare measurements [24][25][37][52][53][54][55][56][57][58]. The lying time duration and the number and duration of lying bouts can be important indicators of calves' health and welfare status [59]. Manual record of these events has low time-labour efficiency and thus cannot be performed daily. Three-dimensional accelerometers provide a noninvasive measure of calf behaviour, using algorithms to process natural position, speed, and directional data [9]. Data acquired with accelerometers have the potential to provide information for disease detection before illness onset, measure pain behaviours more objectively, and evaluate positive welfare situations.

According to Ahloy-Dallaire et al. [60], play behaviour, such as jumping, running, and kicking, are good indicators of improved animal welfare. According to Luu et al. [61], an accelerometer provides reasonable estimates of play behaviour in calves, reducing the sampling rate and measuring acceleration only in the vertical axis. Conversely, Größbacher et al. [62] recorded accelerometer measurements at 1 Hz (in 1-s intervals) and showed that the vertical axis could not be used alone to quantify absolute levels of calves' locomotor play in the home pen. Interestingly, Größbacher et al. [63] found, using accelerometers, that negative play among calves has a contagious effect. Friesian male calves ($n = 325$) were monitored with an activity-monitoring device (Fedometer system, FEDO; ENGS, Rosh Pina, Israel) from 30 to 90 days of life, recording the number of steps, number of lying bouts, and lying time. Frequency and time of visits to the feed bunk were monitored with a proximity sensor placed in the feed bunk; prior to ten days of illness, sick calves' behaviour changed, with fewer steps, less number of visits and lesser time in the feed bunk and increased lying time [58]. However, the best prediction model was only at -1 day prior to disease with a moderate accuracy of 71.5%, Se of 68.8% and an Sp of 72.4%. Gardaloud et al. [64] reported a predictive model for BRD using behaviour data (obtained with an ear-attached 3D accelerometer) from -2 d and -3 d before symptoms with better Se (71.4%) and Sp (95.2%). Moreover, in Goharshahi et al. [65], diarrheic calves had a longer lying time (64.8 min) -1 day prior to clinical identification compared with control calves (ear tag-based

accelerometers; Smartbow GmbH). Similarly, calves with BRD spent more time lying than healthy calves on day -1 [66]; however, the lying bout duration was already greater on day -2 (leg-mounted Axivity AX3). In contrast, in Lowe et al. [24], there were no significant differences in lying time before clinical signs of disease in calves experimentally infected with rotavirus. However, the number of lying bouts decreased, and the duration of the lying bout increased (leg-mounted Hobo Pendant G data loggers).

With an accelerometer placed in the left ear (Smartbow, GmbH, Weibern, Austria), positive results were obtained in detecting lying behaviour, rumination, feed intake, and other activities in calves [56]; however, further development of the algorithm was mentioned as necessary to produce reliable results for milk and water intake. The CowManager Sensor was tested in calves to measure feeding and rumination behaviours against visual observation [67]. The feeding behaviour was well correlated with visual observation ($r = 0.88$), but rumination was not ($r = 0.63$).

Monitoring of negative welfare can also be accessed with accelerometers, for example, in painful procedures such as disbudding [68][69] and castration [70]. The ideal scenario was that these technologies could automatically collect all the behaviours traditionally collected by visual assessment, either in person or with video recordings, and other behaviours that are difficult to measure, such as the number of steps manually. However, the problem seems to lie in a lack of agreement on which indicators are more suitable for pain behaviour analysis. For example, the calves' cautery was disbudded with or without anti-inflammatory administration (flunixin meglumine) and did not show differences in behavioural parameters measured with 3D accelerometers (Hobo Pendant G data logger, Onset Computer Corp.) attached to the right hind leg [71]. Cornual nerve block was performed in both groups, which could explain the absence of statistical differences, although differences in cortisol levels were reported [71]. In a similar study, calves under the effect of anti-inflammatory (meloxicam) were less active than control calves during the first 5 h following dehorning [72]. Nonetheless, lying behaviour (measured with the same 3D accelerometers from the previous study) did not differ between calves' sham disbudded and disbudded using caustic paste with (one group with meloxicam only; another group with lidocaine, and the other with meloxicam + lidocaine) or without pain control; although other indicators of pain presented statistical differences between groups [69]. Dehorned calves without local anaesthesia but with anti-inflammatory injection (meloxicam) or without anti-inflammatory (control) had equal lying times. However, control calves had less lying time post-dehorning than predehorning [73].

Depending on the painful procedure, animals can express a different behavioural response. In a study with beef calves, all of them reduced the number of steps during a 24 h period after castration in comparison with the period prior to castration, regardless of pain control; however, only the calves without pain control presented statistical differences [74]. A dose of 1 mg/kg BW of meloxicam oral suspension was able to reduce the display of painful behaviours and physiological responses after castration (band and surgical methods); calves treated with meloxicam had greater activity and less lying time and number of lying bouts than nontreated calves [75]. The stride length measurement has been used to assess post-castration pain in calves [74], but it seems to not be substantiated [70][76].

2.3. Infrared Thermography

Abnormal body temperature is usually related to illness, and IRT can detect this alteration [77] by measuring radiated electromagnetic energy [78]. IRT measures the animal's surface temperature and thus provides information about some physiological aspects. Usually, thermographic images are taken in highly vascularised anatomical regions, known as thermal windows (e.g., orbital, nasal and perineal regions [79]). However, other regions are also used in IRT validation studies, such as the scapular, the masseteric, the sacral, the umbilical, and the scrotal regions. The orbital region temperature measurement has already been shown to have good intra- and inter-repeatability with IRT in calves under 12 weeks of age [80]. However, there are no similar studies with calves for other anatomical regions. The number of studies with calves using IRT has been increasing in recent years [20][24][25][70][81][82][83][84]. According to Costa et al. [9], the actual temperature of body locations such as eye, shoulder, ear, or side is unknown in calves since IRT measures radiant heat, so IRT is almost exclusively used in illness or pain assessment.

IRT can be used as a noninvasive diagnostic tool providing information to predict infections in calves before clinical symptoms are detected [85]. However, so far, IRT applied to calves has only been used to diagnose enteric diseases, respiratory diseases, and umbilical inflammation.

From the author's knowledge, IRT was first validated in calves as an early indicator of bovine viral diarrhoea [85]. Ten of fifteen heifers were inoculated with bovine viral diarrhoea virus at approximately six months of age, and different body regions were monitored with IRT. The authors have found that the infrared temperature of the orbital region was the most sensitive parameter, with a significant increase one day after inoculation (lower than 1 °C). These changes occurred up to one week before confirmation by laboratory tests, such as haptoglobin levels [85]. Later, Lowe et al. [24] also found an infrared temperature alteration in dairy calves with NCD, infected with *Salmonella*. In this study, the temperature of the lateral region increased; the shoulder (over the trapezius) decreased before clinical signs of disease were detected (up to 4 d before), showing a higher sensitivity than the orbital region, which decreased, but only when the average of the 7 days after and before clinical signs was compared. According to the authors, the temperature increase in the lateral region was probably due to the proximity to the site of infection and localised inflammation of the intestines, and the decrease in temperature of the shoulder could have been due to a restriction of blood flow to the extremities, a response originated from a fever state [24]. Contrary to Schaefer et al. [85], a significant change in the orbital temperature was not observed prior to clinical signs [24]. The metabolic status of the calf influences the heat balance as well as the amount of dissipated heat at the animal's surface so that it can interfere with IRT readings. A significant drop in cheek and ear infrared temperature was observed prior to a diarrhoea bout in calves fed 5 L/d milk but not on calves fed 10 L/d milk [25], showing that milk allowance can affect the IRT capacity as an early predictor of disease. The authors suggested that the drop in temperature may be due to a diminished metabolic rate in the 5 L/d milk group caused by less energy intake, which did not occur in the 10 L/d milk group [25]. The relation between calves' infrared temperature of different body regions and enteric diseases is still not fully understood and needs to be further evaluated. Moreover, it can be suggested that pathogenesis and different etiologic agents can have a significant role in the thermogenesis of the calf and, consequently, in the IRT results.

IRT was studied in calves as an early indicator of BRD [82][86][87]. An increase in the orbital temperature was observed 4–6 days prior to the onset of clinical symptoms in weaned calves [86]. The best sensitivity (68.7%) and specificity (77.4%) were achieved with IRT mean ratio (the mean infrared temperature for the animal divided by the mean maximum temperature for the contemporary group). An automatic system combined with radio-frequency identification (RFID) placed near a water point could identify calves positive or negative to BRD through the infrared temperature of the orbital region [82]. Calves positive to BRD showed a higher peak infrared temperature (35.7 ± 0.35 °C) compared to calves negative to BRD (34.9 ± 0.22 °C) [82]. From the authors' knowledge, there are no further works using IRT as a tool to monitor BRD in calves and to provide validation to these studies. In a narrative review published in 2021 addressing predictive models for BRD in feedlots, IRT was not well placed, mainly due to extra training and costs compared with other methods [88] and a lack of validation studies. Abnormal respiration is a symptom of BRD, so the automatic evaluation of the respiratory frequency could be a valuable tool. Lowe et al. [89] used IRT (T650sc; FLIR Systems AB, Danderyd, Sweden) to monitor the respiratory rate in five dairy calves. A high coefficient of determination ($R^2 = 0.93$) between counting flank movements recorded with a video camera and thermal fluctuations around the nostrils during inhalation and exhalation was obtained [89]. Besides the BRD application, this technique could also be interesting in evaluating tachypnoea associated with heat stress.

Omphalitis is another common prevalent disease in dairy calves, which is diagnosed based on inflammation signs in the umbilical region [90]. However, some calves can present intra-abdominal inflammation without external signs of inflammation, which makes early detection crucial for the calves' welfare. Few studies address IRT as a tool to diagnose omphalitis in calves, and they have not been reviewed to the author's knowledge. A thermographic camera (FLIR SYSTEMS AB, Sweden; Model: T620 25°) was used to evaluate the efficacy of IRT on the diagnosis of omphalitis in calves up to 30 days of age [20]. Calves diagnosed with omphalitis by physical examination had a significantly higher maximum temperature in the lateral umbilical region than calves clinically healthy (37.0 ± 1.1 °C for the omphalitis group and 35.7 ± 1.8 °C for the control group; $p = 0.002$) [20]. In Robinson et al. [91], a portable infrared thermometer (Dual Laser 50 model 42570, Ex-tech Instruments Corporation, Waltham, MA, USA) was used to measure the surface temperature of the umbilical stump as an indicator of infection, but none of the calves presented signs of infection. Steerforth and Van Winden [90] proposed a scoring system for the diagnosis of omphalitis, which includes temperature measurement of the umbilical region with IRT. In the multilinear regression model for omphalitis, umbilical cord infrared temperature was the smallest contributor to the model ($\beta = 1.3$, $p = 0.003$), which included discharge ($\beta = 4.14$, $p < 0.001$), umbilical hernia ($\beta = 1.73$, $p < 0.024$), and swelling ($\beta = 1.72$, $p < 0.001$). However, only discharge and infrared temperature of the umbilical region were significantly associated with intra-abdominal inflammation in the univariable logistic regression analysis ($p = 0.031$). The authors found that an increase in 0.5 °C of the umbilical stump temperature above a sternal reference temperature had an OR of 2.90 (95% CI: 1.10; 7.63) for intra-abdominal inflammation [90]. These results suggest that IRT may have the potential to detect intra-abdominal omphalitis before the occurrence of topic signs, such as the presence of purulent discharge. Nevertheless, when addressing omphalitis as a general inflammation of the umbilical region, other signs such as discharge, swelling, and umbilical hernia may be enough for the trained professional. Another hypothesis yet to be tested is if the infrared temperature of the umbilical region increases prior to clinical diagnosis.

Furthermore, since pain behaviours are associated with omphalitis, it could be interesting to test if 3D accelerometers can identify and predict calves prone to this disease.

Pyrexia is a common symptom associated with infectious diseases, and its evaluation is integrated into most diagnostic protocols. Pyrexia in calves is commonly assessed through rectal temperature; however, the studies correlating rectal temperature with IRT have poor to moderate results. Bell et al. [92] found a weak correlation between the thermal image temperature of the eye and rectal temperature with a thermal camera in preweaned calves ($r = 0.28$; FLIR SC620, FLIR Comp, Boston, MA, USA). Cossa et al. [93] found a moderate correlation ($r = 0.50$) between the infrared temperature of the eye and the rectal temperature of Holstein's calves. Scoley et al. [80] obtained weak correlations between the calves' core body temperature (rectal temperature) and the infrared temperature of the eye and the rectal area (correlations ranging from 0.16 to 0.47). Cantor et al. [94] also tested the infrared temperature of the orbital region against the rectal temperature in 318 male Holstein calves, but the results substantiated the previous findings. From these results, IRT is not a viable technique to diagnose pyrexia, which can be easily performed by measuring rectal temperature with a thermometer, despite the practical advantages of an automated system that could identify fever in group-housed calves. An accurate but more invasive measure of core body temperature could be accomplished with temperature microchip implants, although its accuracy differed with the implant site [95]. IRT showed promising results in evaluating vaccination's safety and local effects in calves [96]. However, more studies are needed to corroborate these results, as it is the first study using IRT to evaluate the vaccination's local effect on calves.

From the current studies, IRT can be a viable technique to diagnose respiratory and enteric diseases and local inflammatory processes in calves since it is sensible to radiant heat emitted in locally affected areas but not to variations in core body temperature.

Pain control during pain management procedures such as disbudding, dehorning, and castration is extremely important to secure the calf's welfare. The efficacy of anaesthetic and analgesic drugs administrated during these procedures has been gaining interest in recent years [97]. IRT can be used as a tool to evaluate these pain procedures since alterations in the sympathetic nervous system can be perceived by orbital region temperature fluctuation due to a redirection of blood flow to skeletal muscles [98][99]. Most of the studies that use IRT to measure pain in dairy calves have focused on disbudding [100][101][102][103].

Disbudding is a painful procedure that is executed in most dairy farms, leaving wounds that can take 9 weeks to heal [104]. It is hypothesised that, due to autonomic nervous system response, a decrease in the temperature of the eye occurs upon pain and/or stressful events [105]. Stewart et al. [81] was the first published paper evaluating the capacity of IRT as an index of pain in calves. This work showed that the temperature of the eye abruptly decreased immediately after disbudding (with a gas-powered cautery iron) without local anaesthetic, in contrast to the nonsignificant decrease in calves disbudded with a local anaesthetic (6 mL of 2% lignocaine hydrochloride). A possible reason is that sympathetic vasoconstriction occurs in response to a stressful stimulus [81]. Ten minutes after the procedure, the temperature increased above the baseline in the disbudded groups (with or without anaesthesia). In the control group (sham disbudding), there was no significative alteration in the eye temperature.

Castrated calves also experienced an increase in the maximum eye temperature 5 to 20 min after the procedure (both calves castrated with or without anaesthesia —lignocaine hydrochloride [106]). Kleinhenz et al. [107] observed that castrated calves 2 h after castration with flunixin administration had increased maximum eye temperatures, compared with calves castrated without anaesthetic and sham castrated; however, it was not indicated the level of significance between treatments. The change in temperature of the eye was also tested as a pain biomarker in calves upon disbudding (electric-powered cautery) with or without the administration of an analgesic (flunixin meglumine). However, no differences were found between both groups in any of the pain biomarkers studied, except for cortisol [100]. A decrease in the maximum temperature of the eye was observed in all calves at hours 1 and 2 after the procedure. However, according to the authors, it was not an indication of a stress response, and the time of the first measurement and environmental temperature could have affected the results [100]. In fact, in Stewart et al. [81], the drop in temperature was only within the first minutes after disbudding, with a gradual increase until 15 min above the baseline. Adcock and Tucker [108] also observed a decrease in the temperature of the eye until approximately 3 min after an injection of lidocaine buffered with sodium bicarbonate in the corneal nerve, which was associated with pain-related behaviours. When calves were surgically castrated without local anaesthetic, it was detected a slight decrease in the temperature of the eye within the first 3 min as well, but not in calves castrated with anaesthesia. [106]. Martin et al. [103] did not find a decrease in the orbital temperature right after cautery disbudding, with cornual nerve blocking. Bergamasco et al. [109] observed a drop in the mean temperature of the eye immediately after castration (without pain control) and sham castration. These results suggest that the decrease in the temperature of the ocular region can be related to both stress and pain responses. When calves were disbudded with caustic paste, the difference in the maximum eye temperature was not observed right after the procedure. However, the mean horn bud temperature increased due to local inflammation, but not over 12 h after the paste application [110]. This study investigated the efficacy of an oral analgesic (meloxicam) as a pain mitigator for caustic paste disbudding, but the results suggested that it had a limited influence [110]. Kleinhenz et al. [102] compared a different disbudding technique (carbon dioxide laser scalpel) from the traditional hot-iron disbudding. The goal was to provide a less painful procedure, which was not true in this pilot study. IRT images were taken with a thermography camera (Fluke TiX580, Fluke Corp.) from the surface of the disbudded site to check for differences between treatments; no statistical differences were found in the maximum or minimum temperature [102].

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