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The non-invasive and automated detection of bovine respiratory disease onset in receiver calves using infrared thermography

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ABSTRACT

Bovine respiratory disease complex (BRD) causes considerable economic loss and biosecurity cost to the beef industry globally and also results in significant degradation to the welfare of affected animals. The successful treatment of this disease depends on the early, timely and cost effective identification of affected animals. The objective of the present study was to investigate the use of an automated, RFID driven, noninvasive infrared thermography technology to determine BRD in cattle. Sixty-five calves averaging 220 kg were exposed to standard industry practices of transport and auction. The animals were monitored for BRD using conventional biometric signs for clinical scores, core temperatures, haematology, serum cortisol and infrared thermal values over 3 weeks. The data collected demonstrated that true positive animals for BRD based on a gold standard including core temperature, clinical score, white blood cell number and neutrophil/lymphocyte ratio displayed higher peak infrared thermal values of 35.7 ± 0.35 °C compared to true negative animals 34.9 ± 0.22 °C ($P < 0.01$). The study also demonstrated that such biometric data can be non-invasively and automatically collected based on a system developed around the animal's water station. It is concluded that the deployment of such systems in the cattle industry would aid animal managers and practitioners in the identification and management of BRD in cattle populations.

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1. Introduction

Bovine respiratory disease complex (BRD) is common in intensely raised and newly transported calves and refers to the animal displaying an undifferentiated fever in addition to a number of clinical signs, notably respiratory distress (Jericho and Kozub, 2004; Buckham Sporer et al., 2008). BRD is known to be caused by a number or combination of viruses and microorganisms (Jericho and Kozub, 2004; Autio et al., 2007) including *Mycoplasma bovis* (Arcangioli et al., 2008), Coronavirus (Decaro et al., 2008), Bovine Para-Influenza (PI3) (Ellis, 2010), Bovine Respiratory Syncytial Virus (BRSV) (Rola and Polak, 2006), Bovine Viral Diarrhoea Virus (BVD) and Infectious Bovine Rhinotracheitis (IBR) (Jericho and Kozub, 2004).

BRD is a worldwide health problem (Griffin, 2010; Horwood and Mahony, 2011) and one of the major and most economically costly diseases affecting the North American beef industry (Snowder et al., 2006; Taylor et al., 2010). The average incidence of BRD in the USA is calculated to be 20% of receiver calves but can be as high as 40%

(Snowder et al., 2006). The primary cost is reported to be due to both the cost of treatment as well as a reduction in subsequent animal performance and carcass merit (Duff and Galyean, 2007; Gay and Barnouin, 2009). In addition to the direct cost of BRD, the increasing concern regarding the use of antibiotics and the potential impact on the promotion of resistant microbes is also apparent and is a growing focal point for global health organizations (World Health Organization (WHO), 2011) and the cattle industry generally (Jericho and Kozub, 2004; Watts and Sweeney, 2010). This concern has led to the development of antibiotic resistant monitoring systems (Wallmann, 2006) and concern regarding antibiotic resistance has had a significant influence on the national agricultural policy for Research and Development Programs (www.agr.gc.ca).

As discussed by Cusack et al. (2003) and Panousis (2009) the effective management of BRD depends on the early recognition and treatment of the onset of the disease. Unfortunately, the clinical signs of BRD are often not apparent until late in the course of the disease due to the challenge associated with early diagnosis (Poulsen and McGuirk, 2009). Numerous diagnostic approaches have been attempted with varying success including the use of acute phase proteins (Humblet et al., 2004), PCR techniques (Asano et al., 2009; Fulton, 2009) and ELISA (Quinting et al., 2007), breath

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analysis (Burciaga-Robles et al., 2009), analysis of cough or respiratory sounds (Ferrari et al., 2010), assessment of feeding and other animal activity (Basarab et al., 1997; Hanzlicek et al., 2010), analysis of protein profiles (Mitchell et al., 2008), rumen bolus temperature recording (Rose-Dye et al., 2010; Timsit et al., 2010) and various immuno-histochemical techniques (Wallmann, 2006).

The identification of disease in an animal or a population will depend on the information available and on the reliability of that information. Currently, the most accurate methods of diagnosis remain analytical techniques such as serum neutralisation, ELISA or PCR procedures. However, with cattle at risk of BRD the information available is often limited and does not include extensive and expensive serum neutralisation or PCD data. Furthermore, if the assessment of an animal's health necessitates the capture and restraint of that animal in order to collect a biological sample then the stress of the process itself will introduce bias.

The use of infrared thermography (IRT) to diagnose animals with BRD has been established (Schaefer et al., 2004–2010). These findings are also consistent with those recently reported by Hovinen (2009) and Polat et al. (2010) for the determination of mastitis in dairy cattle. Many of the aforementioned technologies including clinical scores, haematology, acute phase proteins, cytokines, antibody response and core temperature monitoring are useful aids to the diagnosis of BRD particularly when its prevalence is high in a population and once the clinical signs of respiratory disease are present. Infrared thermography has been shown to demonstrably diagnose BRD at an earlier stage of the disease than other conventional technologies (Schaefer et al., 2007). However, the early detection of BRD, especially in animal populations where the prevalence of BRD and the virulence is low, is more challenging. More difficult still is the development of any technology that can non-invasively and automatically aid in the diagnoses of BRD. The objective of the present study was to investigate the use of infrared thermography to non-invasively identify animals with BRD in a population with a low prevalence of respiratory disease. The calves used in the present study were exposed to a lower level of stressors for that purpose. Furthermore, it was the objective of the current study to examine the feasibility of automating the collection of infrared thermography data. For infrared thermography to be considered as a practical and feasible analytical tool it is necessary that such a system be demonstrated to be compatible with current RFID tags, be automatable and be more user friendly than carrying a hand held camera around cattle pens.

2. Materials and methods

2.1. Animals and management

Sixty-five receiver calves averaging 220 kg were used. The calves were Herford X Angus from either the Agriculture and Agri-Food Canada, Lacombe Research Centre beef herd ($n = 54$) or the Animal Diseases Research Centre beef herd ($n = 11$) located at Lethbridge, Alberta. The calves were weaned and transported approximately 6 h to a commercial auction mart to simulate transport and handling conditions typical of calves received at feedlots in Canada (receiver calves). The animals were held overnight without feed or water and then returned to the Lacombe Research Centre beef research unit. This protocol simulated a commercial auction sale and exposure. Upon arrival at the Lacombe Research Centre the calves were offloaded, caught in a restraining chute, weighed, blood sampled via a jugular vein venous puncture, sampled for saliva using a cotton swab and placed into outdoor pens measuring approximately 60×60 m with one third of the pen having a roof cover. The calves were fed in a conventional bunk feeder a balanced cereal grain silage ration containing 10% cereal grain which met or exceeded National Research Council (NRC) Recom-

mendations (NRC, 1984). The calves received straw bedding and free access to fresh water via an automatic system shown in Fig. 1. All management and operating procedures met or exceeded the Canadian Council of Animal Care Recommendations (1993) and Codes of Practice for Beef Cattle Guidelines (Agriculture Canada, 1991). In addition, all study protocol was reviewed and approved by the Lacombe Research Centre Animal Care Committee.

While contained in their receiving pens the calves were monitored daily by trained personnel for clinical signs of illness using methods described previously (Schaefer et al., 2004, 2007). Briefly, clinical scores were designed to identify bovine respiratory disease (BRD) and were based on the appearance of four criteria as follows:

Respiratory insult: (0–5): 0 = no insult, normal breath sounds (NBS); 1 = Very Fine Crackle (rale) (VFCR) on auscultation and/or a moderate cough; 2 = Fine Crackle (subcrepitant) (FCR) on auscultation and/or a moderate nasal discharge and moderate cough; 3 = Medium Crackle (crepitant) (MCR) on auscultation and/or a moderate to severe viscous nasal discharge with cough; 4 = Course Crackles (CCR), tachypnoea (>15% of the norm) and/or a severe discharge with respiratory distress and obtunded lung sounds and 5 = CCR with dyspnoea, tachypnoea, marked respiratory distress and/or lung consolidation.

Digestive insult: (0–5): 0 = no insult, normal, eating and drinking; 1 = mild or slight diarrhoea with slight dehydration (<5%) and reduced eating; 2 = moderate diarrhoea with 10% dehydration and reduced feed intake (<50%); 3 = moderate to severe diarrhoea with 10% or less of feed intake and more than 10% dehydration; 4 = severe diarrhoea, and less than 10% of normal feed intake and 5 = severe diarrhoea and not eating, not drinking and dehydrated.

Temperature score: Core temperature (rectal) (0–5): 0 = <37.7 °C; 1 = 37.7–38.2 °C; 2 = 38.3–38.8 °C; 3 = 38.9–39.4 °C; 4 = 39.5–40.0 °C and 5 = >40 °C. Rectal or core temperatures for the calves were collected at the start and end of the study only since only at these times were the animal restrained.

Disposition or lethargy score: (0–5): 0 = no lethargy, normal posture; 1 = mild anorexia or listlessness, depressed appearance; 2 = moderate lethargy and depression, slow to rise, anorectic; 3 = recumbent or abnormal posture, largely depressed; 4 = prostrate, recumbent or abnormal posture and 5 = death.

With respect to laboratory analysis, salivary and serum cortisol was analysed using an enzymatic assay as described by Cook et al. (2009). Haematology analysis and differential counts were conducted on a Cell-Dyne model 3700 haematology analyser (Abbott Labs, Mississauga, Ontario). Serology assessment was conducted by Prairie Diagnostic Services (Saskatoon Saskatchewan) and assessment was carried out for Bovine Viral Diarrhoea (BVD) Virus types 1 and 2 and Infectious Bovine Rhinotracheitis Virus (IBR) via serum neutralisation tests and expressed as a titre or the highest dilution of serum to exert a neutralising effect. Additional assessment for Coronavirus, Bovine Para-influenza (PI3), and Bovine Respiratory Syncytial Virus (BRSV) were conducted by ELISA. Again, these methods have been referenced previously (Schaefer et al., 2007).

The ranking of antibody titre scores was as follows: for BVD and IBR 0–2:1 = negative, 3–13:1 = suspicious, 14–40:1 = low, 41–80:1 = moderate, >80:1 = high. For BRSV, PI3 and Coronavirus <10 = negative, 11–13 = suspicious, 14–50 = low, 51–100 = moderate, >100 = high.

2.2. Determination of true positive (TP) and true negative (TN) animals

The determination of an animal true positive or negative for BRD was based on the comparison to a set of “gold standard” values as per the approach of Humblet et al. (2004) and Schaefer et al. (2007). As described by Galen and Gambino (1975) and Thrusfield (1995) this approach is commonly promoted in both veterinary



Fig. 1. Image of automated, RFID driven, non-invasive IRT scanning station located in a feedlot pen around a water station.

and human medical diagnostic studies. In the current study, the criteria for a true positive animal for BRD was defined as an animal displaying three or more of the following signs; a core temperature of $>40^{\circ}\text{C}$, a white blood cell count of less than 7 or greater than $11 \times 1000/\mu\text{L}$, a clinical score of >3 or a neutrophil/lymphocyte ratio of <0.1 (leucopenia) or >0.8 (neutrophilia). A true negative animal was defined as an animal displaying a score of 0 or 1. These parameters were considered consistent with normal and abnormal ranges suggested by other researchers (Kaneko, 1980; Blood et al., 1983). For laboratory assessments, all calves were monitored at the beginning of the study and again three weeks later.

2.3. Infrared thermal station

With respect to infrared thermal measurements, all calves were monitored for radiated temperatures around the orbital area (eye plus one centimetre surrounding the eye) using a FLIR S60 broadband camera (FLIR Comp., Boston, MA) mounted on a motorised shaft. The scanning camera was interfaced to a control system such that the animal self-collected an infrared image every time it attended the water station. The automatic scanning system used is shown in Fig. 1 and schematically in Fig. 2 and consisted of the following components: the system was built around a conventional two water bowl float design from Ritchie water systems (Ritchie Cattle Fountains, Conrad IA, USA). Panels were installed along the sides of the water bowls with a partition in the centre to position the calves when they accessed water. Extension panels were placed on each side of the water bowls to place the calf's head at

the proper focal distance. This system allowed easy access for the calves from two directions. A panel on one side of the water bowls was modified to facilitate a window measuring approximately 30 cm square in order to view the calves while drinking. Two in-phase loop antennae, specifically designed to read over a defined space where the RFID tags would be located, were mounted on adjacent panels, above and slightly behind each water bowl. Each antenna pair was connected to an Allflex PNL-OEM-MODLE-3 control module (Allflex EID system, Allflex Canada Inc. St-Hyacinthe, P.Q.). Electromagnetic shielding was placed on the side of the panel exposed to the holding pen to prevent reading tags on calves that were not accessing water. The design of the antenna system provided the identification of the calf at the water station as well as the signal to the control system to rotate the camera if necessary and to initiate capturing images when the calf's head was visible through the viewing window in the panel. The camera/motor assembly, enclosed under a protective cover, was located medially between the two viewing windows at a distance that provided a field of view to cover most head positions of the drinking cattle. Rotation of the camera to view a particular window was performed by a geared-head motor, controlled by a set of software commands sent to the motor control circuitry. This design met the good laboratory practice requirements for correct thermography: a fixed focal length and angle with a still image which enabled accurate thermal data collection. Software running on a computer located in an instrumentation cabinet read the tag information from the RFID control module, initiated the camera positioning, acquired the infrared image, performed the thermal analysis of the image

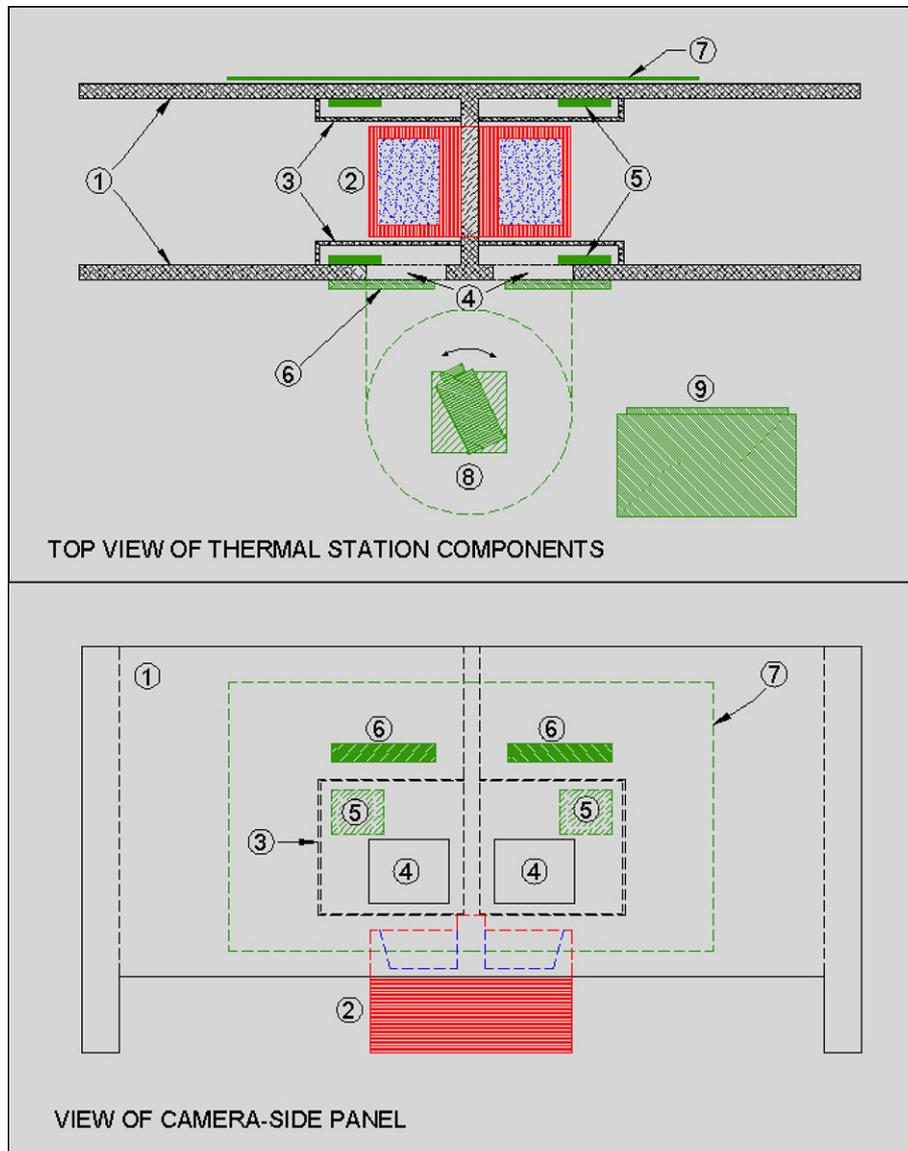


Fig. 2. Drawing of thermal station design. Components: (1) side panels, (2) two bowl water system, (3) extension panels, (4) viewing windows, (5) antennae (6) RFID control modules, (7) electromagnetic shielding (8) infrared camera on motor mount within enclosure, (9) instrumentation cabinet.

and stored the acquired information in a database. The cabinet also housed the power supplies and motor control circuitry used in the system. Instrument integration, and the hardware and software used in this thermal station was designed and developed at the Lacombe Research Centre.

The use of the infrared thermal stations enabled the monitoring of all animals for orbital radiated temperatures every time they attended the water station. The thermal orbital (eye) maximum value for the calves was used in all calculations since this value was found to be most sensitive to stress and disease onset (Schaefer et al., 2004, 2007). All calves were thus monitored for average daily maximum temperatures and for the mean ratio values (MR). The mean ratio which was calculated as the average of daily radiated maximum temperature for a given animal divided by the average daily maximum value for the group of calves.

2.4. Statistics

Verified data was entered into Microsoft Excel (Microsoft Corporation 2007). Basic statistical calculations for data means, standard deviation and two tailed least squares analysis tests (2

tailed *t* tests) were conducted in Microsoft Excel. Response Operant Characteristic curves (ROC) were used to calculate the relationship between true positive (sensitivity) and true negative (100 – specificity) animals for a given biological measurement. These curves were used to calculate the optimal cut off values or values with the greatest efficiency for that parameter. The analysis of data for ROC curves, test specificity and sensitivity, optimal cut off value calculations, positive predictive values and negative predictive values were conducted with MedCalc (2006) software (MedCalc® for Windows. Statistics for Biomedical Research. Version 9. Broekstraat, Mariakerke, Belgium). Infrared thermography data from thermal images were calculated using FLIR Researcher® Software (Version 2.8, Boston, MA).

3. Results

The calves used in the present study were exposed to a lower level of stress compared to typical weaned and receiver calves experiencing a multitude of co-mingling, handling and transport stressors. The verified TP incidence in these calves was thus not unexpectedly comparatively low at 14% (9 out of 65 animals show-

ing a gold standard value of 3 or 4/4). Of interest, however, was the observation that none of these calves were identified as suspect for BRD using subjective clinical signs and none of these animals were deemed to be in need of treatment nor removed for treatment by the animal managers or pen checkers during the study. A total of 44 of the calves displayed TN values (gold standard values of 0 or 1/4) and a further 12 animals displayed intermediate values (gold score value of 2/4) for BRD incidence. The calves typically would visit the water station between 1 and 5 times per day. The amount of time spent at the water station by any individual varied but again typically twenty or more thermal images were captured per animal per day.

The overall average orbital maximum temperature for all calves for the 3 week period was 33.85 ± 0.66 (SD). For this same period, the average for the orbital maximum for the TP calves was 35.44 ± 0.58 and for the TN calves was 34.71 ± 0.57 ($P < 0.01$; Table 1). Since daily thermal values for all animals were monitored, it was possible to follow the radiated temperature rise for the TP calves to the point at which a peak temperature occurred (Table 3 and Fig. 3). The peak thermal response for the TP calves as monitored by the thermal station was $35.7 \text{ }^\circ\text{C}$. The 12 calves displaying intermediate values (a gold standard score of 2 out of 4) showed an average orbital IRT value of $34.9 \pm 0.2 \text{ }^\circ\text{C}$ which was not significantly different from the TN calves but was significantly lower than the TP peak values ($P < 0.01$). Of interest in the present study was the observation that nine of the calves developed BRD signs during the study period and for these animals it was possible to automatically and non-invasively follow the thermal radiated response up to the time of peak thermal response and BRD onset. These data showed that there was over a $1 \text{ }^\circ\text{C}$ elevation in temperature for the TP calves over this time. This contrasts to a flat or basically zero average daily change in temperature for the entire group of animals of $-0.07 \pm 0.27 \text{ }^\circ\text{C/day}$. The data for the mean ratios (MR, Table 1) in general paralleled the orbital maximum values.

In terms of biological values, compared to the TN animals the TP calves (Table 1) displayed higher core temperature values, white blood cell counts, clinical scores and serum cortisol values ($P < 0.01$) with trends towards a higher value in neutrophil/lymphocyte ratios. These values for the intermediate calves were $39.4 \pm 0.73 \text{ }^\circ\text{C}$, $9.589 \pm 2.075 \times 10^3/\mu\text{L}$, 3.5 ± 1.3 , 0.086 ± 0.066 and $135.3 \pm 45.3 \mu\text{mol/L}$ for core temperatures, white blood cell counts, clinical scores, neutrophil/lymphocyte ratio and serum cortisol, respectively. With respect to specific differential counts the TP calves displayed a lower neutrophil count and a higher lymphocyte count, as well as a slight increase in red blood cell numbers ($P < 0.05$) (Table 4). Salivary cortisol displayed a greater variation among animals and thus there were no statistically significant differences in salivary cortisol values among the groups.

The calculation of predictive index values for optimal cut off values, positive predictive values, negative predictive values, test sensitivity and test specificity are shown in Table 2. Again, these values were calculated using ROC analysis. The collected data

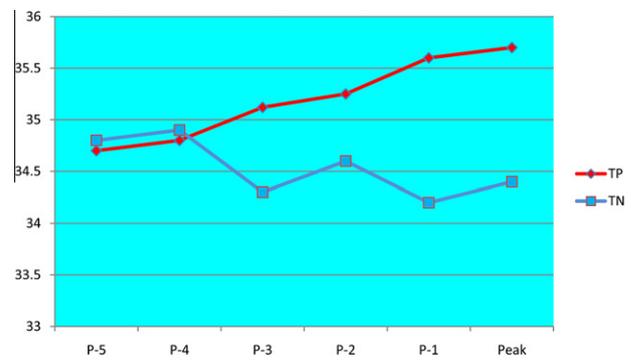


Fig. 3. Example of a true positive animal (3293) displaying rising peak values for orbital maximum temperatures for several days and a comparatively stable orbital maximum temperature for the same several days for a TN calf (1358).

may also be used to estimate test efficiency. In the current study, the test efficiency for the IRT max values was 93% which was as high as any test used.

In terms of seroprevalence the average and range of titres for IBR and BVD were $<1:6$ (range 1:0–1:19) and 1:452 (range $<1:6$ –1:2916), respectively. For the PI3, BRSV and Coronavirus these values were 28 (range 0–68), 19 (range 0–64) and 56 (range 7–95). Most of the calves displayed a low to moderate titre to one or more viruses. In terms of seroconversion, higher values were seen for the true positive (TP) calves. Thirty-three percent of the TP animals seroconverted (>20 units) to BVD virus, 11% to IBR, 0% to BRSV, 11% to PI3 and 11% to corona virus. By contrast in the true negative animals a lower extent of seroconversion was seen. Twenty-two percent seroconverted to BVD, 0% to IBR, 5% to BRSV, 0% to PI3 and 5% to Coronavirus. The data suggest that the BRD viruses measured may have played some significant role in the onset of BRD in the TP animals.

4. Discussion

Identifying animals positive for BRD is not an easy task, particularly if the cattle have mild signs of BRD or the BRD prevalence is low. Part of the reason for this is that there are few gold standards to identify BRD that are absolute (White and Renter, 2009). Again, the most accurate determination of a causative agent for BRD is still likely to be a serum neutralisation, ELISA or PCR technique. However, the gold standards suggested currently in this manuscript have some precedence and support in the literature (Humblet et al., 2004; Blood et al., 1983; Kaneko, 1980; Schaefer et al., 2007) and are reasonable suggested biometric measures for BRD.

The primary haematological response seen in the TP calves compared to the TN animals was the presence of a leucocytosis characterised by lymphophilia and neutropaenia (Tables 1 and 3).

Table 1
Biological values (means \pm SD) for true positive and true negative calves.

| Health status | Core temp ($^\circ\text{C}$) | White blood cells ($\times 10^3/\mu\text{L}^{-1}$) | Clinical score ^a | Neut/lymph ratio ^b | Serum cortisol ($\mu\text{mol/L}$) | IRT ($^\circ\text{C}$) | IRT mean ratio ^c |
|-----------------------------|--------------------------------|--|-----------------------------|-------------------------------|--------------------------------------|--------------------------|-----------------------------|
| True positive (TP) $n = 9$ | 40.2 ± 0.2 | 11.357 ± 2.646 | 5.4 ± 1.1 | 0.103 ± 0.086 | 161.7 ± 32.5 | 35.7 ± 0.35 | 1.018 ± 0.008 |
| True negative (TN) $n = 44$ | 39.0 ± 0.25 | 8.911 ± 1.20 | 2.2 ± 1.3 | 0.167 ± 0.11 | 107.8 ± 50.04 | 34.91 ± 0.22 | 1.000 ± 0.003 |
| Probability ^d | $P < 0.01$ | $P > 0.01$ | $P < 0.01$ | $P = 0.13$ | $P < 0.01$ | $P < 0.01$ | $P < 0.01$ |

^a See Section 2 for a full description of clinical scores.

^b Neutrophil/lymphocyte ratio.

^c Data represents the peak values for orbital maximum temperatures for the TP and the average orbital maximum values during the same time period for the TN calves.

^d Probability determined by least squares analysis two tailed t -test.

Table 2

Bovine respiratory disease positive predictive and negative predicted values, sensitivity and specificity as determined from Response Operant Characteristic (ROC) analysis.

| Parameter | COV ^a | PPV ^b | NPV ^c | Sensitivity | Specificity | P |
|--|------------------|------------------|------------------|-------------|-------------|-----------------|
| Clinical scores | >3 | 61.2 | 100 | 100 | 89.7 | <i>P</i> < 0.01 |
| Core temp (°C) | 39.55 | 86.3 | 100 | 100 | 97.4 | <i>P</i> < 0.01 |
| White blood cells ($\times 10^3/\mu\text{L}^{-1}$) | 10.2 | 41.2 | 95.8 | 77.8 | 82.1 | <i>P</i> < 0.01 |
| Neutrophil/lymphocyte ratio | 0.074 | 34.4 | 93.7 | 66.7 | 79.5 | <i>P</i> = 0.02 |
| Serum cortisol ($\mu\text{mol/L}$) | 105.9 | 25.9 | 100 | 100 | 53.8 | <i>P</i> < 0.01 |
| IRT absolute value ^d | 35.29 | 86.3 | 100 | 100 | 97.4 | <i>P</i> |
| IRT mean ratio ^e | 1.005 | 55.7 | 100 | 100 | 87.2 | <i>P</i> < 0.01 |

^a Optimal cut off value as determined by ROC analysis.^b Positive predictive value.^c Negative predictive value.^d Infrared thermal value of the TP animals at peak temperature and TN animals at their average maximum infrared thermal value.^e Ratio of the peak infrared values for the TP/ave infrared values for the TN animals.**Table 3**

Peak IRT values for TP animals and for several days prior to peak IRT values.

| | Peak IRT | P-1 | P-2 | P-3 | P-4 | P-5 | P-6 |
|----|----------|-------|-------|-------|-------|-------|------|
| TP | 35.7 | 35.04 | 34.91 | 34.74 | 34.89 | 34.8 | 34.3 |
| TN | 34.87 | 34.58 | 34.74 | 35 | 35.12 | 34.99 | 34.8 |

Mean peak IRT for TP 35.7 ± 0.35 vs. mean values for TN during the same time period 34.87 ± 0.025 (*P* < 0.01).

P-1 equals the day before the peak temperature was evident, P-2 equals 2 days before, etc.

Considering a lymphocytosis is commonly observed during a fever and a neutropaenia during viraemia (Strauss, 1987) these haematological observations are to be expected in TP animals.

In terms of infrared thermography, infrared heat loss is a significant avenue for the dissipation of heat in an animal (Kleiber, 1975). The technology has been demonstrated to be effective in the non-invasive identification of transport and other environmental stressors that alter heat loss (Schaefer et al., 1988; Stewart et al., 2007), as well as pain and fear in cattle (Stewart et al., 2008–2010). An earlier comprehensive review of this subject has been published by McCafferty (2007). Using the gold standards criteria in the current study the data suggest that infrared thermography as a biometric measurement also shows utility in identifying the onset of disease in cattle. This is again consistent with previous findings from our own laboratory (Schaefer et al., 2004–2010; Stewart et al., 2005) and with findings from other laboratories (Polat et al., 2010; Hovinen, 2009; Rainwater-Lovett et al., 2009). Indeed, as demonstrated by the ROC curve calculations, the efficiency of the IRT methods in the current study was equal to or better than any of the other methods (93% efficiency with IRT peak values) and in populations with a higher prevalence of the disease has been demonstrated to be superior to other methods in the early identification of disease onset (Schaefer et al., 2007). In the current study

the IRT maximum values and the mean ratio values appeared to show the greatest utility as single measures. Of interest, however, were the observations that the rate of change of an animal's thermal profile as BRD onset occurred and even the degree of variation associated with a given measure such as standard deviation did show promise as valuable indicators. Further evaluation of such parameters on larger data sets and in data sets with perhaps a greater degree of BRD prevalence is merited. In the current study, compared to the industry practice of using clinical signs during pen checking procedures, a lower rate of false negatives was seen. In addition, compared to other procedures using breath analysis (Burgi-Robles et al., 2009), rumen temperature probes (Rose-Dye et al., 2010; Timsit et al., 2010) or any number of biochemical procedures such as PCR, immunohistochemistry, ELISA or acute phase proteins (Decaro et al., 2008; Fulton, 2009; Quinting et al., 2007) the non-invasively collected infrared data is likely to be more cost effective, less labour intense and timely as a diagnostic procedure. Most calves diagnosed with BRD are likely to be treated with one of a few antibiotics regardless of the causative virus. Hence, it may make some practical sense to simply provide early diagnostic information to an animal manager earlier. If specific diagnosis is desired then it may still make sense to conduct a first screen with an automated system such as infrared thermography and subsequently test suspected animals with more precise procedures.

Of interest in the present study was the presentation of a significant number of animals displaying intermediate values for virtually all the biological markers. These animals appeared to be neither TP nor TN. This situation may be a failure of the identification systems to accurately classify animals. Or, conversely, these intermediate animals may well be a population of cattle that are either successfully resisting or slowly succumbing to BRD. In either case, being able to identify such individuals may still be useful from the position of monitoring potential or suspected or emerging BRD cases.

Table 4

Hematology differential count values for TP and TN animals.

| | Neut ^a | Lymph ^b | Mono ^c | Eos ^d | Bas ^e | RBC ^f | Hgb ^g | Crit ^h |
|----------|-------------------|--------------------|-------------------|------------------|------------------|------------------|------------------|-------------------|
| TP mean | 0.564 | 7.790 | 0.960 | 1.440 | 0.065 | 10.6 | 14.4 | 43 |
| SD | 0.152 | 1.651 | 0.413 | 0.785 | 0.021 | 1.36 | 0.78 | 2.6 |
| TN mean | 0.860 | 6.02 | 0.920 | 0.93 | 0.076 | 11.5 | 14.6 | 43 |
| SD | 0.478 | 1.28 | 0.53 | 0.62 | 0.036 | 0.8 | 0.9 | 3 |
| <i>P</i> | 0.07 | 0.01 | 0.8 | 0.04 | 0.36 | 0.01 | 0.5 | 1 |

^a Neutrophil numbers $\times 10^3/\mu\text{L}$.^b Lymphocyte numbers $\times 10^3/\mu\text{L}$.^c Monocyte numbers $\times 10^3/\mu\text{L}$.^d Eosinophil numbers $\times 10^3/\mu\text{L}$.^e Basophil numbers $\times 10^3/\mu\text{L}$.^f Red blood cell numbers $\times 10^6/\mu\text{L}$.^g Haemoglobin g/100 mL.^h Red blood cell haematocrit values (%).

A notable and significant difference of the method used to collect infrared thermal data in the present study to that used in previous studies (Schaefer et al., 2004, 2007, 2009) is that a multiple animal scanning capability of the system has been developed. Mounting the infrared camera on a motor capable of rotating to two different scan windows as signalled from the RFID reader has enabled this system to be placed on two water bowls and hence the capacity to study more animals is significantly increased and can service pens of 100 plus animals. The system was designed to accommodate a second water/thermography station situated parallel to the first station with the camera located centrally between the two stations, hence doubling the animal handling capabilities of the system. Also, compared to other prototypes, the present system is automated and non-invasive by using a RFID reader and lap top computer for data storage. These adaptations are significant from an animal behavioural perspective since the calves can attend the water station voluntarily and without restriction or capture. Hence, both thermal radiated values from the orbital region, as well as watering frequency, are obtained non-invasively. As a result it can be argued that such data are more representative of the animal's normal or steady state values, compared to values collected during animal capture and restraint. Current operation procedures also enable the use of a cell phone communication system to download collected data wirelessly or via the internet. Hence, data and animal responses can be monitored remotely to make management and treatment decisions regarding the emergence of BRD.

Furthermore, the use of a non-invasive, automated, remote sensing system such as the infrared scanning station technology, lends itself to easier collection and oversight for early indicators of animal health aberrations. Such a system would have clear utility in bio-surveillance and bio-security programs.

5. Conclusions

Data collected in the current study demonstrated that the non-invasive, automated collection of infrared thermography data from cattle at risk of BRD is effective at identifying true positive and true negative animals. The study further demonstrated that such technology lends itself to the automated, wireless collection of biometric data useful for bio-security and bio-surveillance purposes.

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