



THE USE OF INFRARED THERMOGRAPHY IN THE EARLY DIAGNOSIS OF SEPTIC ARTHRITIS IN CALVES

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Summary

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The aim of this study was to compare the usefulness of thermographic examination of calves with septic arthritis with other diagnostic techniques such as synovial fluid analysis, radiography, ultrasonography and arthroscopy. Thirteen affected joints from eleven calves were used for the trial. The noninfected joints of the trial calves were used as negative controls. Thermography findings showed that heat from 100% of the affected joints was significantly increased. Subchondral osteolysis and new periosteal bone proliferation of articular bones and widening of the intra-articular joint spaces was observed in 10 joints (76.9%) using radiography. Ultrasound examination of all affected joints showed increased effusion with moderate homogeneous echoic structure. Arthroscopic examination showed synovitis and erosion of cartilage in 61.5% and osteophytic formation in 30.7% of affected joints. All diagnostic methods could be used to diagnose joint disease but it was found that thermographic examination in calves with septic arthritis is an easy convenient method to make an earlier diagnosis, which improves treatment success rates.

Key words: arthritis, arthroscopy, calves, thermography, ultrasound

INTRODUCTION

Septic arthritis may occur by direct trauma or contamination of the joint, extension from periarticular infection, or haematogenous extension (Munroe & Gauvin, 1994; Koefler, 1996). In one survey, 24% of stifle disorders were caused by septic arthritis (Ducharme *et al.*, 1985). Neonates seem to be most at risk for septic

arthritis which can be life threatening (Denoble *et al.*, 2010). Multiple joints can be affected in calves (Arican *et al.*, 1998a), because of haematogenous spread of the bacteria from the initial site of infection (Weaver, 1997; Arican *et al.*, 1998b, Desrochers *et al.*, 2001). Diagnosis of septic arthritis is often based on a

combination of clinical signs, radiographic examination and bacterial culture and cytological analysis of synovial fluid (Nilsson & Persson, 1973). Bacteria are isolated in up to 60% of cases of septic arthritis in cattle. Synovial fluid cytological analysis is very useful for the differentiation between infectious and non-infectious arthritis (Ducharme *et al.*, 1985; Rohde *et al.*, 2000). Radiography reveals increased width of the intra-articular space and osteolysis, but it does not allow localisation and exact determination of the character of the soft tissue swelling (Bargai *et al.*, 1989). The first application of diagnostic ultrasound for musculoskeletal disorders in cattle was reported by Kofler (1996). Indications for arthroscopic surgery in cattle include septic arthritis, development of orthopaedic disease or osteochondritis and diagnostic joint exploration (Munroe & Gauvin, 1994; Gaughan, 1996; Brommer *et al.*, 2004; Denoble *et al.*, 2010).

Infrared thermography (IRT) is a modern, noninvasive and safe technique using thermal profile visualisation, with the advantages of time saving and early detection of inflammation. Infection or injuries can activate the animal's natural body resistance mechanism, which is marked by a movement of heat throughout the tissue, higher blood flow and increased surface temperature. Oedema can be detected as pathologically cool zones on the body surface. IRT has been however used less frequently in livestock research to assess such areas, where live organisms can cause changes in vascular circulation, resulting in an increase or decrease of tissue temperature, (Harper, 2000; Knížková, 2002). Earlier detection of septic arthritis in calves would improve their prognosis.

The objective of this study was to compare the detection of septic joints in calves using IRT with clinical examination, radiography, ultrasonography, arthroscopy and laboratory tests.

MATERIALS AND METHODS

This study was performed at the Department of Surgery, Faculty of Veterinary Medicine. Thirteen joints consisting of eight carpals, of which two were bilateral, three stifles, one tarsal and one elbow from eleven calves with septic arthritis were examined. The calves in the study included seven Holstein, three Simmental and one Brown Swiss, between 10 and 60 days of age and weighed on average 51 kg (range 35 to 60 kg). Radiographic, thermographic and ultrasonographic examinations were performed for all calves in the same order. During the arthroscopic examination, synovial fluid was obtained from the joints for routine biochemistry analysis. Blood was collected from the jugular vein of each calf. Blood gases and haematologic variables were investigated. The lameness grade was classified using the locomotion scoring system of Manson & Leaver (1988) (Table 1).

IRT examination

Thermographic examinations were performed for all animals after routine clinical examination. To adjust for skin temperature (control), the difference between the affected and the control area was calculated. After visual inspection of each joint, repeated IRT images were taken from the dorsal view to monitor the temperatures. All images were scanned using a hand-held portable infrared camera (Wahl, Thermal Imager HSI3000 Series), which was calibrated to ambient temperature (0.10 °C at 23 °C and 25 °C scene

Table 1. Locomotion scoring system (Manson & Leaver, 1988)

Score	Indicator
1	Minimal abduction/adduction, no unevenness of gait, no tenderness
1.5	Slight abduction/adduction, no unevenness of gait, no tenderness
2	Abduction/adduction present, uneven gait, perhaps tender
2.5	Abduction/adduction present, uneven gait, tenderness of feet
3	Slight lameness, not affecting behavior
3.5	Obvious lameness, some difficulty turning, not affecting behaviour
4	Obvious lameness, difficulty in turning, behaviour pattern affected
4.5	Some difficulty in rising, difficulty in walking, behaviour pattern affected
5	Extreme difficulty in rising, difficulty in walking, adverse effects on behaviour pattern

temperature) and absorptive conditions. The emissivity value settled on the camera before conducting scanning was 0.93 (Pezeshki *et al.*, 2011). The humidity was 10% to 90% non-condensing for thermal camera. However, the environmental temperature was 7–10 °C and relative humidity – 73%. The dorsal skin surface was cleaned before images were taken and to reduce the effects of environmental factors on thermal data, all images were scanned at the same distance (1.5 to 2 m) from the subject. The continuous output for each animal in each collection time was saved for 5 to 10 s. The ‘Wahl HSI3000 series imager’ application is used for analysis of images previously saved onto an HSI3000 SD card.

Radiographic examination

Dorsopalmar, dorsoplantar, lateromedial and mediolateral views of joints were taken in free standing calves at the same setting (70 kV and 30 mAs) using a Minolta X ray machine (Sp-HF-4.0 Ralco Spain; Imago, Abbiategrasso, Milano; Regius Model 110 Konica, Minolta).

Ultrasonographic examination

Ultrasound examination was performed on full weight bearing calves. Preparation included clipping and shaving the hairs by

razor, followed by antiseptic washing and rinsing with alcohol and acoustic gel being applied to the area. A diagnostic ultrasound machine (Esaote Piemedical, Model 410477) with a 5 to 7.5 MHz convex transducer was used to image bone and soft tissues in sagittal and transverse scan. Images were printed using a video printer and thermal sensitive paper.

Arthroscopic examination

Arthroscopic examination was carried out after general anaesthesia (xylazine 0.01–0.1 mg/kg IV, ketamine 0.1–0.2 mg/kg IV) and aseptic preparation of the affected joints. The joint was effused with 60 millilitres of isotonic saline and a 4 mm, 30° oblique lens (Lawton GmbH & Co. Germany) was inserted via a stab incision as a single approach at the level of the middle carpal joint or stifle joint (Munroe & Gauvin, 1994). The arthroscopic findings were recorded on a Karl-Storz-Endoscope (Telecam SL II). The purulent fluid and fibrin clots in the affected joints were removed and necrotic areas within the synovial membrane were debrided. Pannus deposits on the cartilage were also removed and the joints were lavaged with sterile Ringer’s solution. Skin incisions were sutured, and the leg bandaged.

The degree of chondroplasty was assessed using the French Society of Arthroscopy scoring and grading system (SFA) (Widmar & Blevins, 1994), where the depth of cartilage lesions was classified on a 0–4 scale. The size of the lesions was estimated as a percentage of the whole articular cartilage that could be seen with the arthroscope.

Synovial fluid analysis

Synovial fluid was obtained by arthrocentesis of 13 septic joints and six normal joints as the control, using sterile needles (18 gauge, 50 mm) and syringes and were transferred to vacutainer tubes. The appearance of synovial fluid and volume of synovial fluid from each joint was recorded at the time of arthrocentesis, noting signs of excessive effusion, manifestations of joint pain such as gait impairment and gross or palpable joint enlargement and evidence of systemic disease. Synovial fluid samples were then centrifuged at 10,000 rpm for five minutes to remove cellular debris. Alkaline phosphatase (AlkP), alanine aminotransferase (ALT), amylase (Amy), aspartate aminotransferase (AST), total cholesterol (Chol), creatine kinase (CK), gamma glutamyl transferase (GGT), lactic acid dehydrogenase (LD), total protein (TP), iron binding capacity (UIBC) and uric acid (Uric) were measured (Beckman Coulter marka, AU5800 model autoanalyser USA). Physical, biochemical, and cytological properties (white blood cell and neutrophil count and increased neutrophil percentage) of synovial fluid from septic arthritic joints were investigated.

Data and statistical analysis

Non-parametric data were evaluated by Mann-Whitney U test as median (min/max) for each category at each level

to detect any significant difference for synovial fluid analysis. In the thermographic imaging, a positive or negative evaluation was made according to the temperature difference between the hottest and the coldest region over the joint.

RESULTS

Clinical examination

Eleven calves suffering from severe purulent arthritis, with or without concurrent infection of other synovial cavities; were inappetent and dull. Their rectal temperatures, heart and respiratory rates were moderately to markedly increased. They showed lameness grades 4.5 to 5. Swelling and pain in affected joint regions were found and in cases with concurrent infection of adjoining synovial structures in the carpal and digital regions, a diffuse swelling was seen.

Infrared thermography

The computer software of the infrared thermal camera showed an increase in local temperature of between 0.5 and 1.5°C, between the normal and affected joints (elbow, carpal, stifle, and tarsal joints) of affected calves. Thermography findings showed that the temperature increases of all 13 joints (100%) increased in a similar trend to the results of physical examination and joint palpation.

Radiographic results

Soft tissue swelling of the affected joints were revealed in all cases. The width of intra-articular space was assessed in all joints examined. Subchondral osteolysis, new periosteal bone proliferation, osteomyelitis and subluxation of articular bones with or without widening of the intra-articular joint spaces were observed in 10

joints (76.9%). Radiographs of the three affected stifle joints revealed increased width of the intra-articular joint space. Only soft tissue swelling was recorded in the infected elbow joint. Subchondral osteolysis of the proximal sesamoid bones and osteomyelitis changes were revealed in seven out of eight carpal joints. Results of the radiographic examination were distributed: contraction of the joint surface (100%), osteophyte production (72.7%), subchondral sclerosis (63.6%), subluxation (72.7%) and increase of synovial fluid volume (100%).

Ultrasonographic results

Ultrasound was used to image the affected synovial cavities in all patients and to detect the relative degree of effusion compared with synovial thickening. Many of the cases showed joint distension, filled with inflammatory liquid effusion and there were clotted masses of varying echogenicity which meant they were not always clearly demarcated from the periarticular soft tissue. Hypertrophy of the synovial membrane especially villonodular synovitis was recognised as a

moderately echogenic soft tissue mass. It was difficult to examine the articular cartilage as it is hidden by the adjacent bone. Ultrasound examination of all joints showed increasing effusion with thick and moderate homogenous echoic structure. Results of ultrasound examination comprised joint effusion (100%), synovial hypertrophy (72.7%), cartilage lesions (loss of anechoic structure and/or thinning of the cartilage layer, and irregularities of sharpness) (54.5%).

Arthroscopic results

Arthroscopic examination showed that 61.5% of calves had synovitis and cartilage erosion and 30.7% had osteophytic formations. Synovial fluid distension, synovial membrane thickening and proliferation were detected as indications of recent or chronic arthritis. In the majority of clinical cases, synovial lesions were observed with lesions of other components of the joint. Clinical signs and arthroscopic changes of the joint capsule and articular cartilage showed hyperaemia, oedema, petechial haemorrhage, hyper villus and villial thickness of synovial

Table 2. Biochemical markers investigated in the synovial fluid of septic arthritis and control groups. Data are presented as median (min/max).

Biochemical parameters	Septic arthritis	Control	P value
Alkaline phosphatase, U/L	384 (184/720)	87 (30/198)	P<0.02
ALT, U/L	32.23 (8.47/52.07)	12.31 (8.3/18.7)	P<0.05
Amylase U/L	542.3 (126/1046.6)	39.32 (66.2/225.2)	P<0.05
AST U/L	42.54 (19.7/78.9)	19 (13/45)	P<0.01
Cholesterol, mmol/L	13.01(4.4/20.2)	1.01(0.4/1.4)	P<0.01
CK-MB, U/L	6.34 (2.1/10.5)	2.45 (2.4/2.8)	P<0.01
GGT, U/L	342 (131/589)	159 (95.1/218.3)	P<0.01
LDH, U/L	5.4 (3.14/7.54)	1.3 (0.97/1.64)	P<0.001
Total protein, g/L	867 (180/1580)	114 (24/185)	P<0.005
UIBC, mmol/L	1.3 (0.4/2.0)	0.05 (0.04/0.08)	P<0.001
Uric acid, mmol/L	27.8 (10.9/42.7)	5.8 (1.8/11.6)	P<0.03

membrane (100% of all cases), new villial formation, villial atrophy, polyploid villial formation of synovial membrane (100% of all cases) and erosion (63.6%).

Synovial fluid analysis

Visual inspection of the joint fluid revealed that three cases were cloudy, six cases were yellow and four cases were haemorrhagic. Two cases had increased viscosity while eleven cases had purulent and fibrinopurulent exudate. Synovial fluid cytology showed increased white blood cell and neutrophil count and increased neutrophil percentage.

Synovial fluid was pale yellow, clear, free of flocculent material and did not clot in the affected joint. AlkP, Amy, AST, Chol, CK, GGT LD, TP, UIBC and uric levels were increased in arthritic cases compared with normal synovial fluid (Table 2).

DISCUSSION

Infrared thermography has numerous applications not only in industry, but also in human and veterinary medicine, primarily for diagnostic purposes (Harper, 2000; Knížková, 2002; Luzzi *et al.*, 2013). This study showed that thermography is an excellent adjunct to clinical and radiological examination. It has previously been shown that thermographic examination could be used safely for diagnosis of septic joint diseases in calves under some conditions (Harper, 2000; Knížková, 2002; Alsaad *et al.*, 2015). The use of thermography for examination of the joints in calves is particularly informative (Arıcan *et al.*, 1998a). Temperature rise means that thermography can detect anatomical, topographical and functional changes, which are caused by microcirculation changes associated with inflamma-

tory and degenerative processes. As a non-invasive method of diagnosis, infrared thermography is suitable for ongoing monitoring of joint conditions in calves. This study showed that local temperature was increased by 0.5 to 1.5°C in affected joints and thermography data reflects the severity of the inflammatory process. Thermography can detect signs of inflammation in the early stages of joint damage, e.g. this method is useful in sub-clinical or latent forms of joint damage, in the absence of physical signs. Thermographic study displays the increase of local temperature in the projection of the affected joint, which usually occurs before other clinical manifestations and remains for an extended period (Arıcan *et al.*, 1998a). Monitoring of the local temperature can also evaluate the results of treatment. Temperature differentials can be vague on screen but the use of computer software improves the diagnosis and the therapy. Thermography is best used as a complementary method for diagnosing lameness. It has high success rates for the diagnosis and therapy of diseases when it is used in addition to radiography or ultrasonography (Greenough *et al.*, 1997; Kaya *et al.*, 2006), but is not capable of identifying differential diagnostic problems in joint structures such as bone, cartilage and synovial membrane if used alone. In this study, thermography showed that heat of the 13 joints (100%) increased in a similar trend to physical exam.

Radiographic examination can be used to make a diagnosis and establish a prognosis of joint disease. Early radiographic signs of joint diseases with osteomyelitis can appear within five to 10 days and most radiographic signs are present within four to five weeks. Bone tissues must undergo a demineralisation greater than 50% to be visible on radiographs and cartilage

is not visible (Fubini & Ducharme, 2004). In our study, soft tissue swelling with gas and increased articular space were present. Subchondral bone lysis, osteomyelitis, periosteal reaction and bony proliferation were seen in septic chronic cases, but radiography can also be nondiagnostic with concurrent disease (Fubini & Ducharme, 2004).

Ultrasonography was shown to be an effective, fast and non-invasive means of diagnosis for the assessment of joint pathology in cattle (Koeffler, 1996). The most common indications for carpal ultrasound are regional swelling, wounds or distension of carpal synovial structures. Soft tissues are better evaluated with ultrasound examination and synovial fluid and membranes, joint surface and surrounding connective tissues are easily seen within certain limits. In acute septic arthritis, the synovial fluid volume increases, and echogenic fibrin material can be floating in the joint. Subchondral bone is hyperechogenic and lysis or a defect will change its contour (Fubini & Ducharme, 2004). In this study, joint effusions were detected in arthritic cases and hypertrophy of the synovial membrane especially villonodular synovitis was recognised as a moderately echogenic soft tissue mass. It was also noticed during the examination that it was difficult to examine the articular cartilage because it was hidden by the adjacent bone. Abnormal echogenicity of the underlying bone is indicative of periarticular bone lysis. Ultrasonography can provide useful information but interpretation of these images can be difficult.

Arthroscopy is an effective technique for diagnosis and treatment of various orthopaedic disorders in cattle, including septic arthritis and osteochondrosis. Joint disease without associated lameness can-

not be fully evaluated with conventional techniques such as examination or X-ray. The advantages of arthroscopy over other techniques include an excellent exposure of most parts of the joint despite minimal surgical trauma and the possibility of performing a thorough lavage under pressure to remove large amounts of deposited fibrin, making it an excellent alternative for diagnosis and treatment of septic arthritis in calves. Synovial fluid distension and synovial membrane thickening or proliferation was indicative of recent or chronic arthritis.

These investigations have indicated that synovial membrane inflammation and articular degeneration were important factors in many common joint disorders and contributed significantly to joint dysfunction. Information gained through synovial fluid analysis can indicate the nature and extent of intra-articular pathologies and is a valuable adjunct to other diagnostic techniques in establishing diagnoses, selecting treatments, and developing realistic prognoses (Rohde *et al.*, 2000; Francoz *et al.*, 2005). In this study, joint fluid analysis correlated well with disease and all biochemical parameters were increased, a similar finding to previous studies (Nilsson & Persson, 1973). There is often a close correlation among the activities of AlkP, AST and LDH in synovial fluids and the clinical severity of joint disease (Madison *et al.*, 1989). The proportionate increase of enzyme activity to the degree of synovitis has been demonstrated experimentally in the equine intercarpal joint, but the specificity of enzyme levels enabling more specific diagnosis has not been demonstrated. In this study, synovial fluid samples were obtained from clinical patients and represented as a single measurement, like studies by Rohde *et al.* (2000). It is possible

that cellular constituents and TP concentration remain elevated in infected or traumatised joints for different time periods and may not be evident with a single measurement of these variables and the diagnostic classification also relied on an accurate assessment by the attending clinician. Although TP concentration is significantly different between infected and noninfected joints, the difference is insignificant in non-infectious joint diseases. Differences in concentrations and composition of synovial fluid TP may exist between acute and chronic conditions and in acute arthritis, so TP concentration may more accurately reflect joint inflammation (Nilsson & Persson, 1973). In this study where joint diseases were often chronic, the synovial fluid TP concentration may be of limited diagnostic value, as previous studies focussed on infectious arthritis in adult cattle (Madison *et al.*, 1989). Rohde *et al.* (2000) showed that creatinine elevation was not diagnostic in such cases, due to asymptomatic reversible impairment of renal function (Kraus *et al.*, 2002). In this study we observed higher levels of CPK, which can be seen after trauma, injections into a muscle and muscle disease (Billinghurst, 2003); or as disease or injury affecting the heart muscle, skeletal muscle and brain. Urea concentrations in joint fluid were found to be directly proportional to serum ones, despite a wide range of concentrations in normal joints (Billinghurst, 2003). From this relationship, the dilution factor introduced by joint fluid was determined and showed that urea is confirmed as a robust method of quantifying and correcting for the dilution of synovial fluid due to joint fluid or inflammation (Billinghurst, 2003). The levels of GGT were increased in this study and can be due to autoimmune reactions (Rohde *et al.*, 2000).

Elevations in cholesterol levels in serum, cartilage and synovial fluid occurring due to subchondral bone repair can lead to nutritional impairment in joints with OA (Rohde *et al.*, 2000; Kaya *et al.*, 2006). Another study also showed a correlation between hypercholesterolemia and osteoarthritis (Al-Arfaj, 2003). This may explain why cattle with infectious arthritis are often younger (Rohde *et al.*, 2000).

Joint infection in calves may also be associated with failure of passive transfer or umbilical remnant infection (Greenough *et al.*, 1997). In calves, multiple joints are frequently affected, whereas polyarthritis is uncommon in adult cattle. Synovial fluid biomarkers can be divided into synthetic and degradative markers and both are useful in diagnosing early change in cartilage and bone (Greenough *et al.*, 1997). Clinical studies have demonstrated the usefulness of serum biomarkers in diagnosing early joint disease and our study confirms that the results of synovial fluid analysis can be used to improve the diagnosis of infectious arthritis.

CONCLUSION

Although radiographic diagnosis of joint disease was important, radiography of the articular cartilage provided only limited information about the synovial membranes and intraarticular ligaments. Ultrasonographic imaging was able to diagnose synovial effusion. In arthrocentesis, sampling of synovial fluid for cytology, total cell and protein counts and culture and sensitivity testing can be frustrating and nonspecific. For joint capsule lesions, extracapsular and intracapsular lesion information, the synovial membrane is best examined with arthroscopic examination. Diagnostic arthroscopy methods were more reliable and concluded to be

necessary and advantageous than others in calves in the cartilage lesions and synovial hyperplasia and treatment.

All techniques used in this study including synovial fluid analysis, radiography, thermography, ultrasonographic examination could be used for joint disease diagnosis. Thermographic examination was useful in diagnosing joint disease, but heat exchange can be related to many disorders and does not provide specific information about the location of arthritis. Thermographic examination in calves with septic arthritis is a convenient way to detect early changes until a definitive diagnosis is posed and also, can help monitor recovery.

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