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Full Length Research Paper

Use of thermal imaging for the early detection of signs of disease in pigs challenged orally with *Salmonella* typhimurium and *Escherichia coli*

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A total of 27 piglets were randomly assigned to three treatment groups (control, infection with *Salmonella enterica* serover Typhimurium KCTC 2515 (ST) and infection with *Escherichia coli* KCTC 2571 (EC)) in a completely randomized design to early detection of signs of disease based on body temperature. Totally three inoculations were done consisting of 14 days each and then thermal images were captured to calculate body temperature of pigs at 0, 2, 6, 12, and 24 h and then every 24 h up to 14 days post inoculation. A reduced average daily gain (ADG) was observed in the first week of all three post inoculation ($P < 0.05$), while the gain:feed ratio was decreased at first week during first inoculation in both ST and EC group compared to control ($P < 0.05$). Body temperature was elevated in ST infected piglets at 24 h, peaked at 72 h ($P < 0.05$) and remained elevated, however, the EC induced piglets showed a subnormal body temperature throughout the experimental period relative to the control ($P < 0.05$). Taken together, the results indicate that signs of disease following experimentally induced bacterial infection in pigs can be detected quickly and easily using thermal images.

Key words: Piglets, growth performance, thermal image, body temperature, disease diagonosis.

INTRODUCTION

Infections with enteric pathogens such as *Salmonella* or *Escherichia coli* are common during intensive pig production and are associated with poor performance and animal welfare (Pijpers et al., 1991; Greiner et al., 2000). Such infections may also pose a risk to human health and lead to expensive veterinarian intervention costs. Delays in detection may result in the outbreak of diseases and even more expenses. Detection of animal illness and providing individual and group-by-group mass therapy is not effective. Since infection is primarily spread via horizontal transmission from carrier animals to others in close contact (Schwartz, 1991), the best way to handle an illness is early detection of diseases followed by application of treatment before the disease can spread.

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Healthy animals are important to profit and performance, as well as animal welfare. Timely interventions may interrupt the lifetime carriage of *Salmonella* and *E. coli* and improve the health and growth of weaned pigs.

Swine have an unusual means of regulating core temperature. Due to the low number of sweat glands (Moritz and Henriques, 1947; Montagna and Yun, 1964), pigs reduce their core temperature by increasing the rate of peripheral blood flow through the skin, panting, or moistening the skin with water. However, panting and evaporation are less efficient regulation systems under some environmental conditions and are not always possible in confined pig housing. Fever is the earliest and one of the main clinical signs of many diseases (Plonait, 2004). Previous investigations showed that *S. typhimurium* challenged piglets showed increased body temperature (Balaji et al., 2000), while *E. coli* endotoxin inoculation resulted in subnormal body temperature (Halloy et al., 2004) compared to controls. Based on the way heat is transmitted through the skin of swine, infrared images have the potential for use in the detection of body temperature (Ingram and Weaver, 1969; Godynicki et al., 1985).

Traulsen et al. (2010) concluded that infrared thermography allows routine measurements of body surface temperature that can be used for early disease detection. Röhlinger et al. (1979) reported the possibility of using infrared cameras (IRCs) to measure body surface temperature for early disease detection in various animals, including swine. Scolari et al. (2011) used an IRC to detect the rise and fall of vulvar skin temperature of sows during estrus. Therefore, the present study was conducted to measure the growth performance and potential for early detection of sign of disease based on body temperature patterns measured using a thermal camera as an early disease diagnostic tool in *Salmonella enterica* serover Typhimurium and *E. coli* infected piglets.

MATERIALS AND METHODS

All experimental procedures used in this study were approved by the Animal Care and Use Committee of Sunchon National University, South Korea.

Animals and experimental design

A total of 27 newly weaned piglets (crossbred ((Landrace x Yorkshire) x Duroc), and mean body weight 8 kg and 28 days of age) were used for three consecutive inoculations. The length of each post inoculation period was 14 days and the experiment was conducted for a total of 42 days. At the onset of the study, each animal was tested bacteriologically (fecal) before receiving the *Salmonella* and *E. coli* challenge and were found to be negative for the bacteria. The piglets were assigned to one of the three treatment groups (control, infection with *Salmonella enterica* serover Typhimurium (ST) and infection with *Escherichia coli* (EC)) in each post inoculation period in a completely randomized design based on their initial body weight. Each treatment had three replicate pens with three pigs per pen. The pigs in each group were reared in three isolated pens (slatted floor and a space allowance of 0.75 m^2 per piglet) in an environmentally controlled room and allowed a 1-week adaptation period before commencing the study. Pigs in the ST and EC group were then orally infected with 10 mL mixed suspensions (1:1) of *S. enterica* serover Typhimurium KCTC 2515 (5.5×10^6 cfu/ml) and *E. coli* KCTC 2571 (3.7×10^6 cfu/ml). An ambient temperature of 24 ± 1.49°C was maintained in the pig houses via overhanging electric heaters. During the experimental period, pigs were provided with a commercial diet *ad libitum*. The pens were illuminated by artificial light, and ventilation was provided by ten air changes per hour. The body weight and feed intake were recorded on a weekly basis until the end of the experiment.

Infrared thermography of piglets

Thermal images of piglets were captured using an infrared camera (Thermo Tracer, Type- TH5104R CAT I, NEC San-ei Instruments, Ltd., Tokyo, Japan) at a fixed distance of approximately 1 m from the animal. The emissivity value was set to 0.985 and the thermograph resolution was calibrated to ambient temperature and humidity as per the manufacturerer's recommendations. The piglets were numbered temporarily and allowed to settle before infrared thermographic (IT) images were captured. Multiple images were captured and the most informative image for an individual piglet was used. Thermal images were captured at 0, 2, 6, 12, and 24 h and then every day for 14 days post inoculation (dpi) without handling the piglets. Images were stored in a memory card, then transferred to a computer for analysis using the InfRec Analyzer Lite software system. The temperatures acquired from the IT images were recorded from three locations in each piglet (head, body and tail region), and the average values were used to calculate the standard body temperature of piglets.

Statistical analysis

All recorded data were statistically analysed using the SAS system *v.* 9.1 (SAS Institute. Inc., Cary, NC, USA, 2003). The individual pen was considered as the experimental unit for growth performance and body temperature calculation. Treatment means were computed with the LSMEANs and significant treatment x time interactions were observed for body temperature using thermal camera. Longitudinal significant variations in body temperature due to *Salmonella* and *E. coli* infection in different time periods are denoted by abcde, while treatment effects on specific time period denoted by xyz. Duncan's multiple range tests were used to identify significant differences among treatment groups for growth performances and body temperature. A probability level of $P < 0.05$ was considered statistically significant.

RESULTS

The average daily gain (ADG), average daily feed intake (ADFI) and gain:feed ratio of piglets are shown in Figures 1, 2 and 3. In the present study, we observed a decreased ADG at the first week of each post inoculation period in the ST and EC groups ($P < 0.05$) (Figure 1). A reduced ADFI was found in the second week of first inoculation and in the first week of second inoculation in the ST and EC infection groups relative to the control ($P < 0.05$) (Figure 2). A reduced gain:feed ratio was observed in the first week of first inoculation ($P < 0.05$) (Figure 3). Although the gain:feed ratio was decreased in

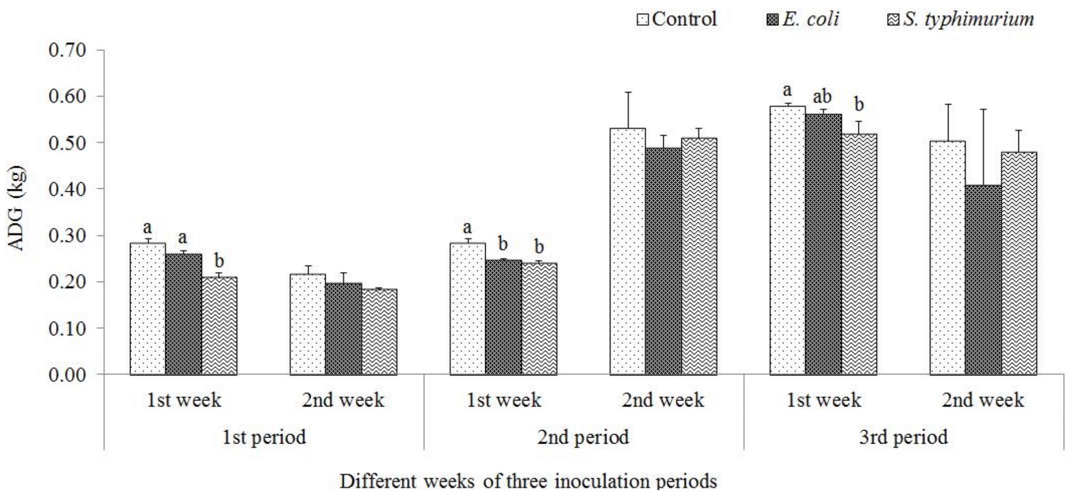


Figure 1. Mean average daily gain (ADG) (kg) of piglets in different weeks during the three inoculation periods**.** Data are presented as the mean ± SE. Bars at a week without a common letter differed significantly (P < 0.05).

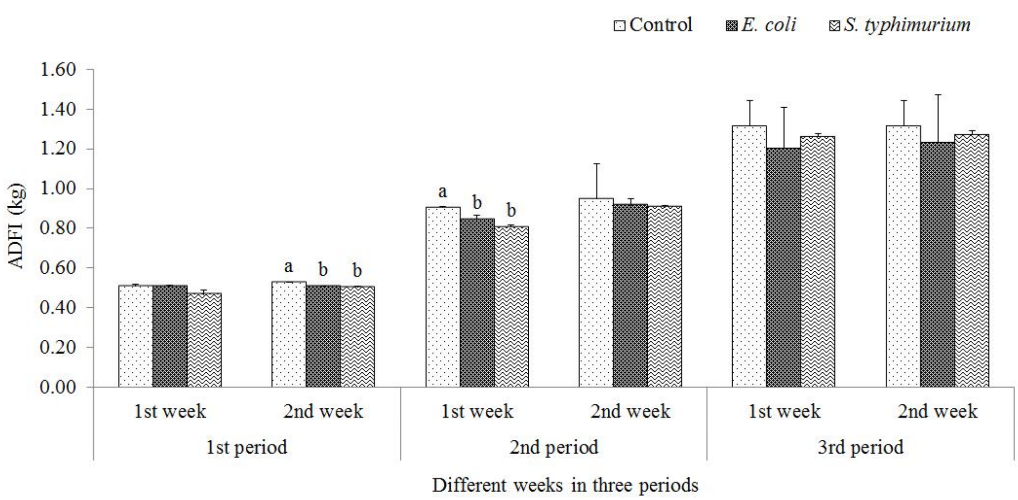


Figure 2. Mean average daily feed intake (ADFI) (kg) in different weeks during the three inoculation periods**.** Data are presented as the mean ± SE. Bars at a week without a common letter differed significantly (P < 0.05).

EC group, it was increased in ST group during second week of first inoculation and second inoculation period compared to control (P < 0.05) (Figure 3). Thermal images of piglets for control, infected with *Salmonella enterica* serover Typhimurium KCTC 2515 and *Escherichia coli* KCTC 257 are shown in Figure 4.

The body temperature patterns of piglets with bacterial infections are shown in Figure 5, 6 and 7. During the first inoculation, no significant variations were observed until 24 h post inoculation, while piglets challenged with ST showed a gradual increase in temperature by 24 h. The maximum temperature increase relative to the control was observed at 72 h (P < 0.05), and this value remained elevated until the end of the study period (Figure 5). However, the EC induced piglets showed a subnormal body temperature pattern throughout the experimental period during all three inoculation periods relative to the control (P < 0.05) (Figures 5, 6 and 7). Average daily body temperature was significantly lower in EC group while it was alleviated in ST group compared to control (P < 0.05) (Figure 8).

DISCUSSION

In most animals, illness is commonly expressed by their

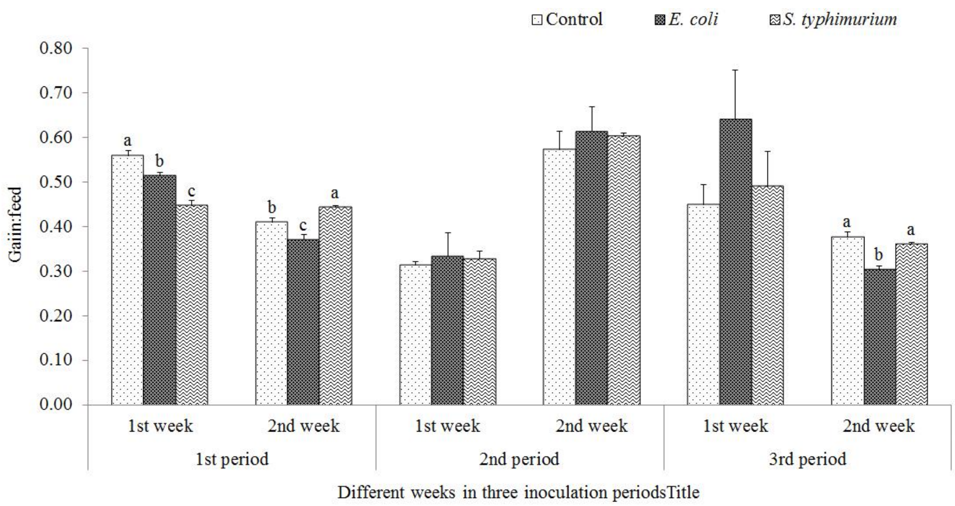
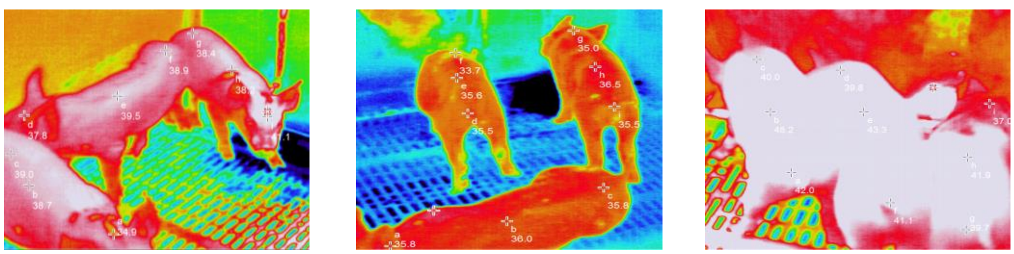


Figure 3. Mean average gain:feed ratio in different weeks during the three inoculation periods**.** Data are presented as the mean ± SE. Bars at a week without a common letter differed significantly (P < 0.05).



Control piglets

E. coli infected piglets

Salmonella infected piglets

Figure 4. Thermal images of piglets for control, infected with *Salmonella enterica* serover Typhimurium KCTC 2515 and *Escherichia coli* KCTC 2571.

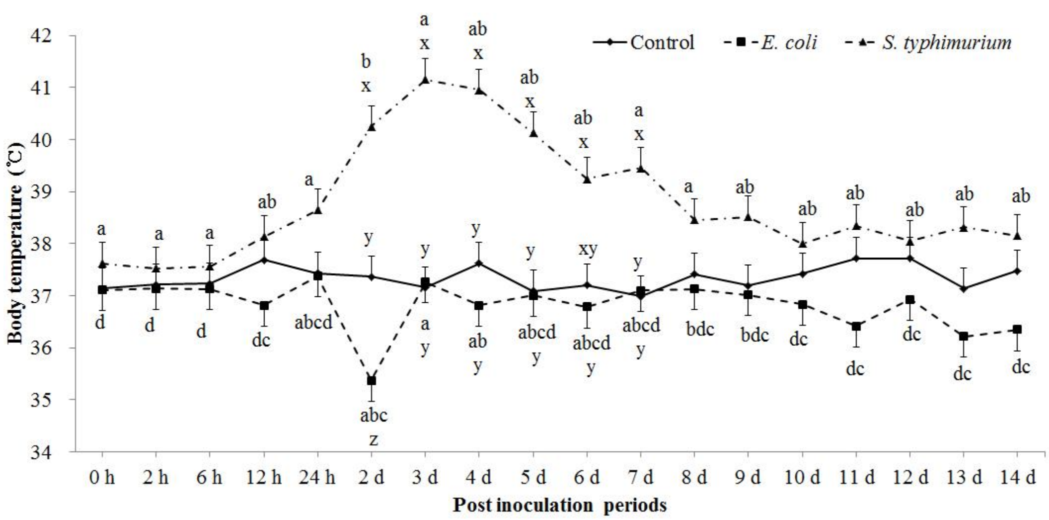


Figure 5. Body temperature pattern of piglets during the first inoculation period (14 days)**.** Data are presented as the mean ± SE. Lines at a particular time period without a common letter differed significantly (P < 0.05).

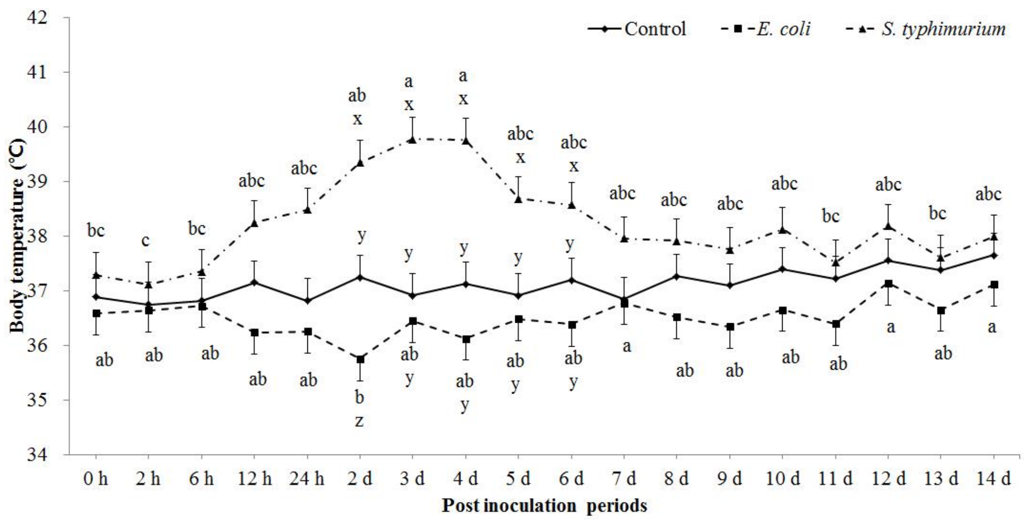


Figure 6. Body temperature pattern of piglets during the second inoculation period (14 days)**.** Data are presented as the mean ± SE. Lines at a particular time period without a common letter differed significantly (P < 0.05).

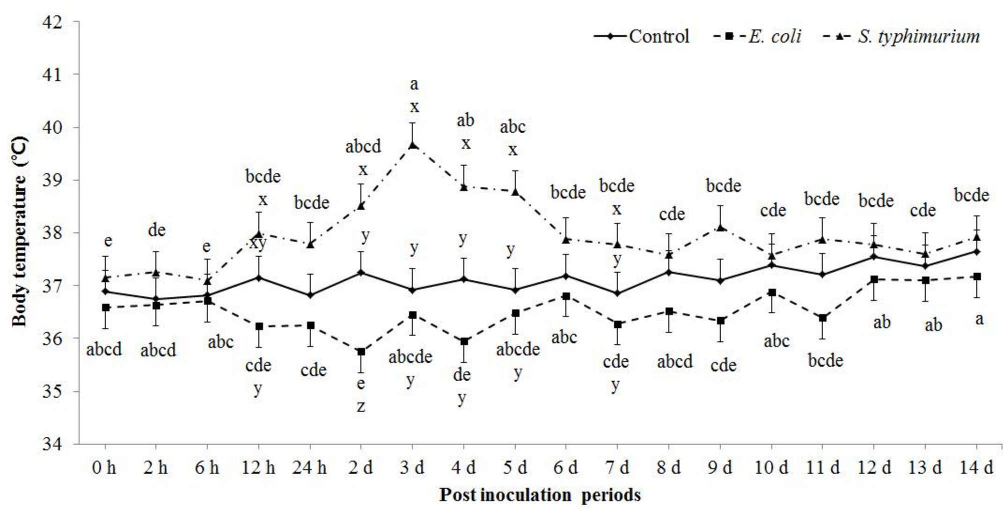


Figure 7. Body temperature pattern of piglets during the third inoculation period (14 days)**.** Data are presented as the mean ± SE. Lines at a particular time period without a common letter differed significantly (P < 0.05).

behaviour (Weary et al., 2009). Farm animals express a wide range of behaviours such as loss of appetite, drinking and social behaviours, many of which may impact their health and welfare directly or indirectly. In the present study, we observed a decreased ADG at the first week of each post inoculation period in the ST and EC groups (Figure 1), which is consistent with the results reported by Van Heugten et al. (1994). Additionally, reduced body weight gain of pigs was observed over 14 days in response to bacterial infection (Balaji et al., 2000), and decreased weight gain, feed intake and efficiency of feed utilization were observed after repeated challenge with non-infectious agents in broilers (Klasing et al., 1987). The reduced ADG of piglets during post inoculation period of piglets might be due to effect of *Salmonella* and *E. coli* infection (Van Heugten et al., 1994; Balaji et al., 2000). ADFI did not affect in the first week, it was decreased in the second week during first inoculation which also extended in the first week of second inoculation (Figure 2). Feed intake was depressed in ST induced pigs in the 120 h period following the challenge, with the maximum decrease being observed 48 h after challenge. In a previous study, intake was returned to control levels at between 120 and

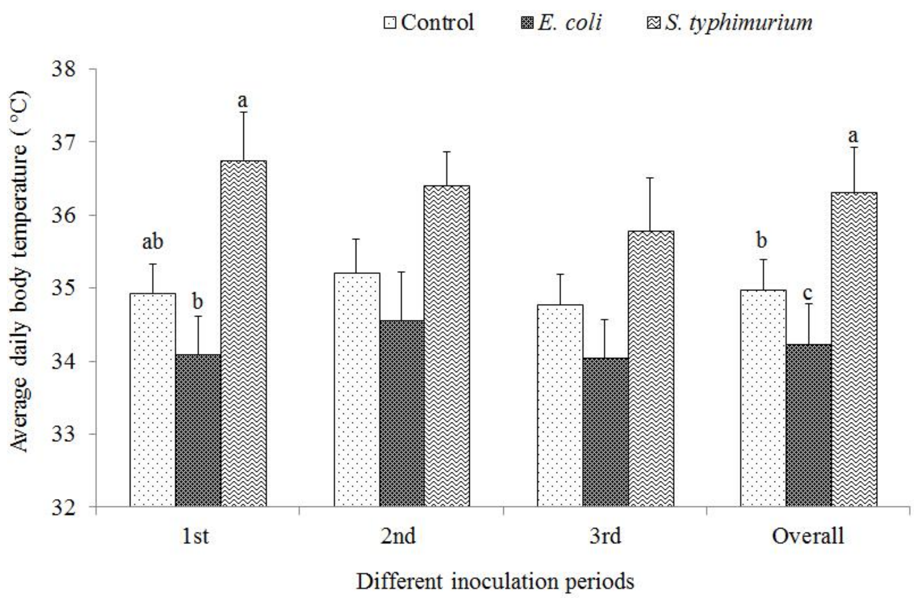


Figure 8. Average daily body temperature pattern of piglets during the three inoculation periods (42 days). Data are presented as the mean ± SE. Bars at a particular inoculation period without a common letter differed significantly (P < 0.05).

144 h of the challenge and was supported by Balaji et al. (2000). A number of infectious diseases of swine are characterized by fever, cachexia, inactivity and anorexia (Hart, 1988). The reduced gain:feed ratio might have been due to the effect of reduced weight gain in the respective periods in each post inoculation period. The overall decreased piglets growth performance of this study can be explained as the effect of bacterial infection which might be the indication of illness in pig (Klasing et al., 1987; Balaji et al., 2000).

Physiological responses of infection and inflammation in animals are the result of neuroendocrine and immune systems interactions. Adaptive responses include induction of proinflammatory mediators and fever, reduced feed intake and diminished growth performance, as well as development of a regulated, specific, immune response to ward off pathogens. *Salmonella* and *E. coli* are the most successful at colonizing the gastrointestinal tract under stress conditions, when nutrition and immunity are suboptimum, as is the case in newly weaned pigs (Aumaitre et al., 1995). A recent review described changes in peripheral blood flow resulting in alterations of skin temperature of livestock that could be detected by infrared thermography (Stewart et al., 2005). In the present study, piglets challenged with ST and EC showed remarkable temperature variations at different hours and days in all three post inoculation periods. The body temperature patterns of piglets with bacterial infections are shown in Figures 5, 6 and 7. During the first inoculation, no significant variations were observed until 24 h post inoculation, while piglets challenged with ST showed a gradual increase in temperature by 24 h. The maximum temperature increase relative to the control was observed at 72 h, and this value remained elevated until the end of the study period (Figure 5). However, the EC induced piglets showed a subnormal body temperature pattern throughout the experimental period relative to the control (Figures 5, 6 and 7). The increased body temperature of ST challenged piglets observed in the present study is consistent with the results reported by Walsh et al. (2012), who found a tendency for increasing body temperature with increasing time of bacterial inoculation. The febrile conditions in our present study started at 12 h post inoculation, then gradually increased until becoming significantly higher on day 8. These increased levels then continued throughout the experimental period. Balaji et al. (2000) reported that the fever associated with ST was gradual in onset, and was sustained for 5 days following challenge. The infrared thermographies used to detect bacterial infection in pigs in the present study have also been applied in many other studies (Loughmiller et al., 2001; Schaefer et al., 2004; Johnson et al., 2011). Infrared techniques were used by Loughmiller et al. (2001) to measure the body temperature of pigs, and the results showed that it is possible to detect a febrile response using body-surface temperature. Pigs have few sweat glands and are therefore forced to cool down by increasing the rate of blood flow through their skin (Moritz and Henriques, 1947; Montagna and Yun, 1964); accordingly, infrared technology may be a good method for detection of increases in body temperature. Manno et al. (2006) reported that surface temperature increases with increasing ambient temperature. *E. coli* are commonly found in human and animal intestinal tracts and can survive outside the gastrointestinal tract for considerable lengths of time in several environments (Ashbolt, 2004). In the present study, EC induced piglets showed a subnormal body temperature throughout the experimental period in all three post inoculation periods, which is normal and occurs due to starvation and dehydration due to diarrhoea. Feed intake of the EC infected group of this experiment was lower during most of the week in each inoculation period. The body temperature was also lower during the first week, indicating that starvation or lower feed intake can be a cause of subnormal body temperature. Additionally, some pigs infected with EC showed diarrheal symptoms, although we did not record the diarrheal score, which may also have led to subnormal body temperature. The detection of subnormal body temperature in piglets of this study is in agreement with the results of studies conducted by Vianna and Carrive (2005), who investigated changes in temperature at the rat tail in connection with a fear reaction and concluded that it was possible to detect decreased temperature via infrared images. The increased body temperature due to *salmonella* and subnormal body temperature due to *E. coli* infection of this present study could be helpful for the pig farmers to early detection of bacterial infection which is important to take measures for further severe damage. Average daily body temperature was significantly lower in EC group while it was alleviated in ST group compared to control (Figure 8). The overall body temperature in ST group was also alleviated while EC group was decreased compared to control which might be due to the effect of weekly body temperature in pigs. Overall, growth performances and body temperature pattern of challenged piglets could be more beneficial for the farmers in pig monitoring system.

Conclusions

Reduced ADG, ADFI and gain:feed ratio, as well as variations in body temperature are general indicators of bacterial infections in pigs. In the present study, thermal image analysis of heat emissions from pig skin made early detection of ST and EC infection in pigs possible. This method provides the opportunity to measure body temperature continuously without contracting or stressing the animals and with a minimal risk of injuries. Thus, it can be an effective tool to minimize costs and save time while ensuring animal welfare during pig farming.

Conflict of interests

The authors did not declare any conflict of interest.

ACKNOWLEDGEMENTS

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