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Research article

Application of infrared thermography as a determinant of estrous conditions of Sapera dairy goats

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Abstract

Importance of the work: Detecting a goat's estrous usually involves invasive methods that are less accurate because they are subjective, require significant time and resources and may cause problems with animal welfare. Infrared thermography (IRT) is one alternative solution because this method is modern, non-invasive and safe.

Objectives: To investigate the capabilities of IRT to predict and detect estrous in goats. Materials & Methods: Twelve Sapera dairy goats were synchronized using CIDR® containing 0.3 g of progesterone for 18 d and an injection of serum gonadotropin at a dose of 400 international units intramuscularly 48 h before CIDR® withdrawal. Visualization of the estrous response, vaginal (T_{vaginal}) and rectal (T_{rectal}) temperatures, Draminski® score and IRT sensing results on the vulva (IRT_{vulva}) and rectum (IRT_{anal}) were measured six times daily starting at the time of the CIDR® withdrawal.

Results: All goats showed the onset of estrous at 24–32 h after CIDR® withdrawal. T_{vaginal} (mean \pm SD, 39.20 \pm 0.13°C), T_{rectal} (39.20 \pm 0.15°C), IRT_{vulva} (37.03 \pm 0.17 °C), and IRT_{anal} (37.04 ± 0.10°C) during estrous were higher than normal conditions. The Draminski® score was in the range 356–373 at the time of estrous. IRT_{vagina} and IRT_{anal} were positively correlated with T_{vaginal} and T_{rectal}, but not with the Draminski[®] score.

Main finding: IRT is a promising non-invasive tool for reproductive management in goats to identify superficial temperature variations during estrous in the vulva or vagina. IRT sensing alone or in combination with estrous observation would increase the probability of estrus detection in goats.

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Introduction

Many methods can be used to accurately detect goats undergoing estrous, for example, through visual observation, changes in vaginal temperature and mucus resistance and silent behavior when riding a male (Silva et al., 2017). Other methods have also been used to measure changes in body temperature during estrous (Anggriawan et al., 2017; Tian et al., 2021), LH (Luteinizing hormone) surge, and ovulation (Fisher et al., 2008; Knox, 2019), as well as the use of commercial estrous detection devices, such as Draminski® (Murtaza et al., 2020). However, handling animals when measuring temperature, taking blood samples or inserting probes for estrous detection devices can cause discomfort and stress to the goats, eventually affecting the interpretation of the actual results. Conventional strategies for estrus detection are regularly utilized by observing the behavior of the female that are impacted by length and frequency of perceptions (Banuvalli et al., 2015). Furthermore, expanded group size and the requirement for enhancement work have expanded the reliance on estrus detection tools. One alternative solution is to use non-invasive diagnostic tools, such as infrared thermography (IRT). With this tool, body surface temperature can be measured quickly and precisely while minimizing discomfort and stress conditions in the target goats.

In this context, at the exploratory level, infrared thermography plays an important role as a projected technique that is non-invasive and can be utilized for examination purposes in various animal production fields, particularly in the identification of thermal levels (Sykes et al., 2012). In the livestock industry, IRT has been utilized for applications in the conclusion of mastitis in dairy cows (Martins et al., 2013; Sathiyabarathi et al., 2016), assessing the scrotal temperature as an indication of fertility in cattle and goats (Weschenfelder et al., 2013) and for the assessment of heat stress in dairy cattle (Montanholi et al., 2008). In addition, it has been utilized as a helpful and proficient procedure for estrous detection in dairy ruminants (Talukder et al., 2014), particularly in recognizing an increase of the surface temperature in the vulva region to distinguish between the periods of estrous and diestrus (Siregar et al., 2016). Stelletta et al. (2017) revealed a connection between the thermal pattern of the vulva after hormonal estrous synchronization and fertility after artificial insemination. This was an outcome of changes in body temperature during the estrous cycle, because of expanded blood flow, which increases with ovulation (Ola et al., 2006).

The utilization of IRT in goats has been effectively described and it is generally connected with understanding thermoregulation of changes in the surface temperature and the effect of environmental condition on animal welfare (Nääs et al., 2014). The applicability of this method in reproductive analysis presents important assumptions. In any case, a possible limitation of this innovation is the expense of the infrared cameras utilized (Rainwater-Lovett et al., 2009). Other limiting factors are the distance, point of estimation, daylight, wind, drafts, soil, dampness or unfamiliar material on the hair coat. The impact of weather conditions, circadian and ultradian rhythms, season, draining, laying and rumination are additional factors to consider (Johnson et al., 2011).

To date there appears to be no detailed information regarding changes in vaginal and vulvar temperatures, visualization of estrous response, and the scores of estrous detection devices from the beginning to the end of estrous. Therefore, the current study aimed to evaluate the visualization of the estrous response, measurement of vaginal and rectal temperatures, the value of the Draminski estrous detector score and the potential results of IRT sensing (vaginal and anal) for detection of estrous in dairy goats.

Materials and Methods

This research was conducted at the Indonesian Research Institute for Animal Production (IRIAP). The animals used in this study were 12 Sapera dairy goats of parity 2-3 with a live weight range of 30-40 kg that were kept in individual cages measuring 1.6 m ×1.0 m. The Sapera goat is one of the dairy goats in Indonesia, which is a crossbreed between the Saanen goat from Switzerland and the Etawah crossbreed goat from Indonesia. Before the study, all goats had received oral administration of albendazole (5 mg/kg body weight). The feed given was in commercial concentrate and elephant grass (Pennisetum purpureum Schum.) silages as much as 1,400 g/d (based on pre-research results on the maximum amount of concentrate and silage consumed by a Sapera dairy goat). The concentrate was in the form of C-Prolac produced by PT. Citra Ina Feedmill, Jakarta, Indonesia. The nutrient composition of the concentrate was: moisture 12%; protein, 18-20%; fat, 7%; fiber, 21%; ash, 10%; calcium, 0.8-1.0% and phosphorous 0.6-1.0%. Feed was offered twice daily at 0700 hours and 1500 hours in West Indonesia Time. Drinking water was provided ad *libitum* from nipples attached to each cage.

Experimental design

The Sapera dairy goats were synchronized with a CIDR® insert (InterAg; Hamilton, Australia) and pregnant mare's serum gonadotrophin (PMSG) (Heibei Yunfeng Animal Pharmaceutical Co. Ltd; Hebei, China). The schematic of the research procedure, estrous synchronization protocol and parameter observation schedule are presented in Fig. 1. Intravaginal insertion of the CIDR® containing 0.3 g of progesterone for 18 d was performed using an applicator that had been smeared with a lubricant at the tip. The application was carried out by holding the goat in a standing position and in a calm state. The CIDR® removal was carried out 18 d after insertion, simply by pulling the CIDR® cord slowly. Injection of serum Gonadotropin 400 international units (IUs) intramuscularly was conducted 48 h before CIDR® removal. Injection using a 3 mL disposable syringe was conducted by first cleaning the hair and applying an antiseptic. After the injection, the goats were returned to the individual pens, ensuring no excessive bleeding or swelling.

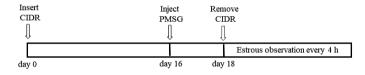


Fig. 1 Schematic diagram of research procedure, estrous synchronization protocol and parameter observation schedule

The goats were observed six times daily day starting when the CIDR® was removed, for as long as the goats were in heat and until they showed no signs of estrous. The parameters observed were: vaginal and rectal temperatures, behavior and visualization of the estrous response, the value of the Draminski® estrous detector score and the IRT values (on the vulva and anally). The IRT observations were carried out before other parameters. To control bias among the observers, the observation process was only carried out by one observer who was assisted by one person in charge of handling goats.

Vaginal and rectal temperature

The vaginal temperature (T_{vagina}) was measured by inserting a digital thermometer (model MC-245; Omron Healthcare Co. Ltd.; Kyoto, Japan) into the vagina as deep as \pm 5 cm. Likewise, the rectal (T_{rectal}) temperature was measured by inserting a different digital thermometer) into the rectum \pm 5 cm deep.

Estrous response (behavior and visualization)

Observation of the behavior of the female goats was carried out using a buck wearing an apron, performed according to the method described by Santoso et al. (2014). The behavior of the female goat was given a score of 3 if it was silent when climbed, a score of 2 if it was quiet and refused to be mounted, and a score of 1 if it rejected the buck. The noise behavior of the female goat was given a score of 3 if it made a frequent sound, a score of 2 if it made an infrequent sound, and a score of 1 if it was silent and there was no reaction. Agitated female goat behavior was given a score of 2 if restless and 1 if calm. The behavior of the female goat wagging her tail was assigned a score of 2 if there was a flick and a score of 1 if there was no flick. The behavior of female goats urinating after mating was given a score of 2 and a score of 1 if there was no urination.

Observation of the visualization of the external reproductive organs of each female goat was carried out simultaneously, performed according to the method described by Santoso et al. (2014) and Murtaza et al. (2020). Measurement changes in the color of the vaginal mucosa were given a score of 3 for red, 2 for pink and 1 for pale pink. Swelling of the vulva was assigned a score of 3 if there was swelling, a score of 2 if the vulva was only slightly swollen and a score of 1 if the vulva was wrinkled. The viscosity of estrous mucus was given a score of 3 if the mucus was viscous, clear hanging or moist around the vulva, a score of 2 if the amount was small and a score of 1 if there was no secretion.

Draminski estrous detector score

Detection of estrous using the Draminski® estrous detector (Draminski's SA; Poland) was performed by inserting a vaginal probe equipped with an electrode at the probe tip of the device to a depth of three-quarters of the vagina, then pressing the ON button three times periodically and waiting for the measurement results for a few seconds until the number that appears was constant on the tool screen.

Infrared thermography

An overview of the IRT sensing results using an infrared camera (One Gen 3 brand; FLIR Systems Co. Ltd.; Australia) with an emissivity coefficient of 0.98. Routine care of the animals and cage was provided by the kennel staff to minimize the dirtiness of the goat as a factor that might directly or indirectly affect the accuracy and precision of the

measurements. Other limitations regarding IRT, such as wind and humidity, were minimized due to the closed cage system. The IRT sensing of each animal was taken at the vulva (IRT_{vulva}) and rectum (IRT_{anal}) with a distance of about 1 m from each point or body part observed and then stored in a memory card, before being transferred to a laptop computer for analysis using the ThermaCAM Professional Researcher 2.10 software (USA).

Statistical analysis

Observational data from the visualization of estrous response, vaginal and rectal temperatures, Draminski® score and IRT sensing results of the female Sapera dairy goats were analyzed using the MIXED procedure from the SAS V.9.1 software (SAS, 2004). Differences between treatments were determined based on analysis of variance using the MIXED procedure of SAS. A significant difference between the levels of a classification variable was considered when p < 0.05. Data were presented as mean \pm SD values, obtained using the MEANS procedure of SAS. The correlation value obtained from the IRT sensing was related to vaginal temperature, rectal temperature and the Draminski® score using the CORR procedure of SAS.

Ethics statement

This study was approved by the Ethics Committee of the Institutional Animal Care and Use Committee, Ministry of Agriculture, the Republic of Indonesia (Approval no. Balitbangtan/Balitnak/Rm/04/2019)

Results and Discussion

The estrous responses of the Sapera goats during estrous synchronization are presented in Fig. 2. The 12 female Sapera dairy goats that were synchronized to estrous all showed signs of estrous that was evident from the occurrence of mating processes with males as much as 41.67%, 33.33% and 25.00% at 24 h. 28 h and 32 h after the CIDR® was withdrawn (data not shown in Figure). These results indicated that the onset of estrous occurred at 24-32 h after the insertion of the CIDR®. The same effect was also reported by Khairiyah et al. (2015) in crossbreed Saanen goats where the onset of estrous occurred 23.76 h after the CIDR® was withdrawn, which differed from that reported by Fonseca et al. (2005) in Toggenburg, Saanen, and Alpine dairy goats, where the onset of estrous for the three goat types was more than 48 h. Similarly, Romano et al. (2016) reported that the beginning of estrous in Boer goats occurred at 37–44 h. Therefore, the current study showed that the Sapera dairy goats had a faster estrous response (< 32 h) after the withdrawal of the CIDR®. This proved that the technique of synchronizing estrous with the progesterone was very effective in improving the reproductive performance of the goats, especially in inducing estrous (Silva et al., 2011; Abdelnaby and Abo El-Maaty, 2020). In a study using the CIDR®, the effect of exogenous progesterone on follicular growth showed that high blood concentrations of progesterone could decrease luteinizing hormone (LH) secretion (van Werven et al., 2013; Bartlewski et al., 2015). The decrease in LH secretion causes atresia of the dominant follicle in the ovary and the emergence of new follicle development that can induce estrous and reach the ovulation stage (Menchaca et al.,

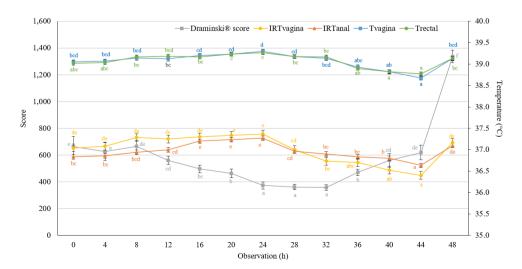


Fig. 2 Oestrous response of female Sapera goat during estrous synchronization, where different lowercase letters on same lines indicate significant (p < 0.05) differences and error bars = \pm SD

2018). In addition, the presence of male goats that are close to female goats that are synchronized to estrous will accelerate the onset of estrous. This result agreed with other research where male goats showed sexual behavior such as making noise and riding female goats, thereby accelerating the onset of estrous, characterized by a surge in LH between 24 h and 48 h after sponge retraction (Panyaboriban et al., 2018).

Although the statistical analysis showed differences in values p < 0.05) at several observation points (vaginal and rectal), in general, they showed the same pattern, namely an increase in temperature from the beginning to 24 h observation, a decrease to 44 h and an eventual return to the initial temperature at 48 h of observation. The mean T_{vagina} during estrous (39.20 ± 0.13 °C) was higher than during normal conditions (39.04±0.20 °C); likewise the mean T_{rectal} during estrous (39.20±0.15 °C) was higher than during normal conditions (39.04±0.17 °C). These results were similar to those reported by Fakruzzaman et al. (2012) in Black Bengal goats where T_{vaginal} and T_{rectal} during estrous (39.60±0.03 and 39.30±0.05 °C) were higher than during normal conditions (38.83±0.11 and 38.85±0.13 °C, respectively). The increase in T_{vaginal} and T_{rectal} during esthrous was related to changes in the environmental conditions around the vagina due to the CIDR® insertion. Manes et al. (2010) reported that the installation of estrous synchronization in ewes intravaginally changed the aerobic conditions of the vagina which affected the thermoregulatory process. T_{rectal} changes could also be caused by endocrine changes due to the release of progesterone from the sponge, which causes an increase in the concentration of the progesterone, affecting the thermoregulation process that changes body temperature (Tansey and Johnson, 2015).

The results of the IRT_{vulva} and IRT_{anal} sensing are presented in Fig. 2. The $IRT_{\mbox{\tiny vulva}}$ and $IRT_{\mbox{\tiny anal}}$ sensing results showed the same pattern as those for T_{vaginal} and T_{rectal} . The mean IRT_{vulva} during estrous (37.03±0.17 °C) was higher than during normal conditions (37.01±0.16 °C). Likewise, the mean IRT_{anal} during estrous (37.04 \pm 0.10 °C) was higher than for normal conditions (36.94±0.10 °C). These results were similar to those described by Khairiyah et al. (2015) where the increases in T_{vagina} and T_{rectal} during estrous were related to changes in environmental conditions around the vagina due to the installation of the CIDR® and an increase in the concentration of progesterone affecting the thermoregulation process. Talukder et al. (2014) reported a rise in temperature of 1.5 °C 24 h before ovulation. Furthermore, Stelletta et al. (2017) noted that the vulvar area is a reference point for using IRT as a non-invasive temperature determination method. An increase in the vulvar temperature occurs during the estrous phase, followed by a decrease in the vulvar temperature as ovulation approaches.

The IRT $_{\rm vulva}$ and IRT $_{\rm anal}$ sensing results had lower values than T $_{\rm vagina}$ and T $_{\rm rectal}$, by approximately 2.1 °C on average. This difference was possible due to the locations of the body parts measured. The method of vaginal and rectal temperatures measurement with a thermometer takes time and requires direct contact with goats. In addition, this method allows penetration into the deeper vaginal and rectal areas so that higher values of temperatures are obtained compared those from the use of IRT which only measures the surface temperature of the vulva. In contrast, with measurement using a rectal thermometer, both the depth of penetration and the type of thermometer can induce stress in the measured animals that eventually increase the temperatures obtained.

Detection of estrous using the Draminski® tool showed a value of 662.86 ± 97.24 at the hour 0 observation that decreased to 356.67 ± 15.27 at 32 h and then increased at 48 h to 1,336.67 \pm 47.25. Estrous in female goats was characterized by the mating process with male goats that occurred at 24-32 h after the removal of the CIDR®, with values in the range 356–373. This protocol followed the operating instructions for the Draminski® tool, where if the estrous detector shows a value of 400 or less, it means that the goat is in estrous. In contrast, if it the reading is more than 400, then the goat's condition equivalent to being before or after estrous (Draminski, 2021). The working principle of the device for estrous detection is based on the presence of the resistance of an electric current in the vaginal mucus as an indicator (Yasa et al., 2018). According to Verma et al. (2014), an increase in the volume of cervical mucus during estrous occurs when the high estrogen concentration affects vaginal vasodilatation. The cervical mucus has relatively high conductivity, so the more cervical mucus, the less the electrical resistance. The value displayed by the Draminski® tool in this study was in the range 356–373, indicating that the mucus produced had a relatively large volume because the level of estrogen had increased, indicating that the goat was in the estrous phase.

The behavior and visualization of the external reproductive organs of the female Sapera goats during estrous synchronization are presented in Figs. 3 and 4. The results showed that the behavior of wagging tails began after 4 h of observation, while the behavior of making noise, restlessness, and still refusing to be mounted only began to be seen after 16 h observation; even urination was only seen during the mating process, namely after 24–32 h observation. Furthermore, the female goats did not

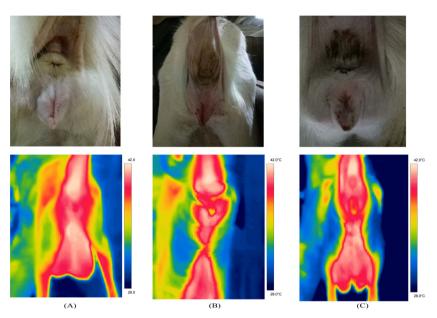


Fig. 3 Visualization of external reproductive organs and results of infrared thermography sensing of female Sapera goats: (A) vaginal mucosa pale pink with wrinkled vulva; (B) pink vaginal mucosa with only slight vulval swelling; (C) red vaginal mucosa with swollen vulva and viscous mucus moistening around vulva

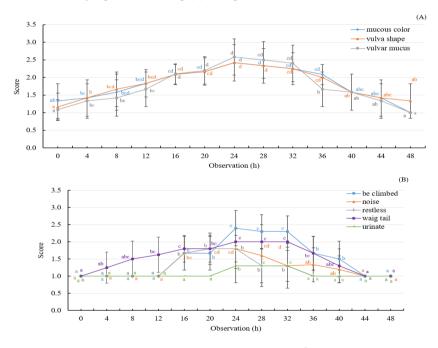


Fig. 4 Female Sapera goats during estrous synchronization: (A) behavior based on Draminski[®] score; (B) visualization of external reproductive organs, where different superscripts on same lines indicate significant (p < 0.05) differences and error bars = \pm SD

show estrous behavior after 44 h of observation. Visualization of the external reproductive organs of the female goats during estrous synchronization showed that the color of the mucosa, the shape of the vulva and vulvar mucus increased until 24 h of observation and decreased until the external reproductive organs returned to normal conditions. Similar results were reported by Khairiyah et al. (2015) in Saanen cross goats where the behavior and visualization of external reproductive organs during estrous, such as red vaginal mucosa with a swollen

vulva, tail wagging, restlessness, sound and silence when being ridden by a male goat occurred 24 h after CIDR® extraction.

In the current study, the behavior and visualization of the external reproductive organs of the female goats during estrous were clearly seen, namely silence when mounting, wagging tail and red vaginal mucosa with swelling of the vulva. Khairiyah et al. (2015) reported variation in the signs of estrous, which was caused by changes in the secretion of estrogen hormone levels between individuals. Furthermore, Abdisa (2018) reported that before reaching

the estrous phase, the estrogen level in the blood increases, increasing uterine turgidity, cervical relaxation, cervical mucus production and the number of endometrial glands cells that cause the initiation of estrous behavior.

Pearson's correlation coefficient (r) between the IRT sensing results with vaginal temperature, rectal temperature and Draminski® values are presented in Table 1. The statistical analysis results showed a significant correlation for all vaginal and anal temperatures evaluated using IRT and digital thermometers, that differed from the Draminski® values. There was a high correlation between IRT_{vulva} and IRT_{anal} (r = +0.716) and between T_{vagina} and T_{rectal} (r = +0.791). These results were not much different from those reported by de Freitas et al. (2018) in Santa Ines sheep in Brazil, where the correlation between IRT_{vulva} and IRT_{anal} was r = +0.62, while between T_{vagina} and T_{rectal} , it was r= +0.80. Furthermore, George et al. (2014) reported a correlation between T_{vagina} and T_{rectal} in sheep and cattle, with values of 0.95 and 0.78, respectively. These results show that anal or rectal temperature measurements can replace vaginal or vulvar temperatures measured using infrared and digital thermometers.

Table 1 Correlation coefficient between infrared thermography (IRT) results with vaginal temperature (IRT_{vagina},), rectal temperature (IRT_{anal}) and Draminski® score

Variable	IRT _{vagina}	IRT _{anal}	T _{vagina}	T _{rectal}
IRT _{anal}	0.716**			
T _{vagina}	0.553**	0.552**		
T _{rectal}	0.545**	0.446*	0.791**	
Draminski® score	0.062^{ns}	$0.007^{\rm ns}$	-0.147ns	-0.123ns

^{*}p < 0.05, **p < 0.01, ns = not significant

The vaginal temperature measured using IRT (IRT $_{vagina}$) had a positive correlation with the digital thermometer measurements of T_{vagina} (0.553) and T_{rectal} (0.545). Likewise, the rectal temperature measured using IRT (IRT $_{anal}$) was positively correlated with the digital thermometer measurements, for both T_{vagina} (0.552) and T_{rectal} (0.446), respectively. In a similar study in sows, IRT was used to identify estrous, with theIRT sensing results in the vulva area being closely related to the estrous phase (Simões et al., 2014). The correlation between the vagina and rectal temperatures was associated with the decreased progesterone levels and LH peaks during estrous. In addition, measurement using a digital thermometer has disadvantages of being time-consuming and invasive (Saranika et al., 2015), as well requiring the tail to be kept raised during measurement (Talukder et al., 2014).

In conclusion, the results showed that IRT was a promising non-invasive tool for reproductive management in goats to identify superficial temperature variations during estrous in the vulva or vagina. IRT sensing alone or in combination with estrous observation of behavior and visualization would increase the probability of estrus detection in goats.

Conflict of Interest

The authors declare that there are no conflicts of interest.

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