Utilizing Thermographic Imaging to Detect and Monitor Pregnancy in a Southern White Rhinoceros Erika Defer Safari West edefer@safariwest.com

Part of our responsibility of housing animals under human care is to expand our knowledge to continuously improve their wellbeing, especially for a keystone species such as the southern white rhinoceros. One of the major ways we accomplish this is through scientific research. With the oversight of our research committee, Safari West is conducting research related to behavior and reproduction in southern white rhinos, utilizing thermographic imaging. We have anecdotally seen evidence that supports the ability to detect and monitor pregnancy in our female southern white rhinoceros by monitoring changes in heat signatures on her abdomen, vulva and teats. Images are captured weekly with the FLIR E5 XT and FLIR E76 cameras. We plan to use FLIR Thermal Studio Suite to look at temperature variation over time to determine thermal patterns. The use of this technology to detect and monitor pregnancy is innovative and, to our knowledge, has only been researched in black rhinos and babirusa with limited results. We believe our data will produce significant results that show changes in heat signatures in our female as her fetus developed and during increased periods of fetal activity. The information gathered from this research is invaluable, especially with species and/or individuals that it is unrealistic to collect voluntary samples from and has the potential to translate to in situ research, as it allows for non-invasive data collection. We are continuing this research project postpartum, to ascertain the use of thermographic imaging to monitor estrus cycles in southern white rhinos.

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Abstract

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With the oversight of our research committee, Safari West is conducting research related to behavior and reproduction in southern white rhinos, utilizing thermographic imaging. We have anecdotally seen evidence that supports the ability to detect and monitor pregnancy in our female southern white rhinoceros by monitoring changes in heat signatures on her abdomen, vulva and teats. Images are captured weekly with the FLIR[®] E5 XT and FLIR[®] E76 cameras. We plan to use FLIR[®] Thermal Studio Suite to look at temperature variation over time to determine thermal patterns. The use of this technology to detect and monitor pregnancy is innovative and, to our knowledge, has only been researched in a few species, white rhinos not being one of them. We believe our data will produce significant results that show changes in heat signatures in our female as her fetus developed and during increased periods of fetal activity.

The information gathered from this research is invaluable, especially with species and/or individuals that it is unrealistic to collect voluntary samples from and has the potential to translate to *in situ* research, as it allows for non-invasive data collection. We are continuing this research project postpartum, to ascertain the use of thermographic imaging to monitor estrus cycles in southern white rhinos.

Introduction

The use of infrared thermography (IRT) to study animals has been around for many years, in both domestic and exotic species. However, the variety of applications has expanded greatly in recent years due to the continual advancement of technology and research. Infrared thermographic imaging provides the ability to measure body surface temperature of animals in a non-invasive manner (Speakman and Ward, 1998). IRT has been used in veterinary medicine and research for applications such as identifying inflammation, disease, measuring stress, monitoring thermoregulation, and assessing reproductive measures (Church et al., 2009; Cilulko et al., 2013; McCafferty, 2007; Hilsberg, 1998; Hilsberg, 2002; Rekant et al, 2016). The ability to utilize IRT for diverse purposes, noninvasively and without physical contact, is beneficial to working with a variety of species, including both *in situ* and *ex situ* animals. The noninvasive nature of IRT is especially beneficial when working with potentially dangerous or flighty species (Mota-Rojas et al., 2022).

The use of IRT has been documented in several exotic species under human care such as black rhinos, elephants and suids (Hilsberg, 2008; Krueger et al, 2019; Sykes et al., 2012). Hilsberg stated that rhinos are ideal for utilizing IRT to monitor reproductive events, such as pregnancy, due to their tough skin, that has minimal expansion throughout pregnancy, allowing for clearer edges around the increased-radiation areas (Hilsberg, 1998). This has been supported in research on black rhinos (Diceros bicornis) with thermograms throughout pregnancies (Hilsberg-Merz, 2008). During the third trimester, Hilsberg showed as much as a 6°C difference in the fetal region of the abdomen compared to the rest of the female black rhino, with a significant decrease in temperature difference post calving. In addition, Hilsberg found the female's mammary gland was 4.4°C higher than the rest of her body temperature (1998). Typically, other invasive and non-invasive methods, such as progesterone monitoring via blood draws, cycle monitoring, rectal and transabdominal ultrasounds or fecal hormone testing are needed to confirm and monitor pregnancy in rhino species; however, these methods can be time intensive or require anesthesia (Hilsberg-Merz, 2008). In a more recent study on babirusa (Babyrousa babyrussa), they found the number of weeks of pregnancy positively correlated with the maximum temperature on her sides as well as a shift of the maximum temperature to the female's teats from her thorax and flank areas (Krueger et al., 2019). A study looking at Polish Konik Horses, found that thermal changes on the lateral surface of the abdominal area was different for pregnant and nonpregnant mares (Maśko et al., 2021). Maśko et al. also found that there were differences in the maximum and average temperatures starting in the 6th month of pregnancy and differences in the minimal temperature from the 8th month.

This study focuses on a female southern white rhino (Ceratotherium simum) during her first pregnancy. According to the IUCN red list, white rhinos are listed as near threatened with a decreasing population trend (Emslie, 2020). Furthermore, white rhinos have one of the longest reproductive cycles in terrestrial mammals (Hermes, et al., 2020) and with a decreasing population, it is vital to ensure successful reproduction and offspring. White rhinos typically reach maturity around 6 years of age (Owen-Smith, 1973) with a gestation of approximately 506 days (Hermes et al., 2020). The calf will then stay with the dam for 2-3 years. Southern white rhinos can be challenging to successfully breed under human care for a variety of reasons including diet, uterine pathology, and social organization (Tubbs et al., 2017; Metrione and Eyres, 2014; Hutchins and Kreger, 2006). Breeding issues related to social organization can result from small herd sizes, specifically those that do not include multiple females, which can lead to behavioral influences on unsuccessful breeding, as is the situation we have at Safari West with just one male and one female. Only 3% of dams will successfully get pregnant after 20 years of age (Metrione and Eyres, 2014). The female in this study was 17 years at the time of conception; as she was approaching her late teens, there was a chance that she may have lost her fertility. While she had been housed with other males in the past, copulation was never successful until we introduced a new male in 2021. This male had successfully sired several calves at his previous facilities. The success of her pregnancy was a significant achievement for southern white rhino reproduction which necessitated close monitoring throughout the pregnancy as her advanced age could have led to complications.

While black rhinos (*Diceros bicornis*) have been extensively studied, to our knowledge, no studies have been carried out, until now, to support the use of IRT to detect and monitor pregnancy in a southern white rhino. The objective of this research is to show if changes in heat signatures around the teats, vulva and abdominal area can be detected in our female as her fetus

develops and during increased periods of fetal activity. Changes in heat signatures are expected to be seen in the female's abdominal region as well as in her teats later in gestation. Changes in heat signatures are not expected to be seen on the female's vulva as that is typically associated with estrus. However, as we have continued collecting data postpartum, we expect to see changes in the female's vulva once she starts cycling again to indicate estrus.

Methods

Animals

This study includes 1 male and 1 female southern white rhinoceros. The female arrived at Safari West in 2008 when she was approximately 4 years of age and has been housed at Safari West since. She was 17 years old at the time of conception and 18 years old at the time of parturition. This was the female's first pregnancy. The male arrived at Safari West in 2021 and was 25 years old at the time of conception. He has successfully sired 10 calves prior to arriving at Safari West.

Conception occurred in November 2021. The female was confirmed pregnant via blood tests for progesterone levels as well as through transabdominal ultrasounds. Voluntary blood draws to test for progesterone levels continued, at minimum monthly, and increased when we believed she was close to calving, to monitor a drop in the progesterone. She gave birth to a healthy male calf on April 2nd, 2023.

All participation by the rhinos was voluntary. Animal care staff that the rhinos were comfortable with assisted in the collection of thermograms, blood samples and ultrasounds to ensure low stress and cooperation of the rhinos.

Location

This research was conducted at Safari West Wildlife Preserve which is located in Sonoma County, CA. Safari West is an oak savanna habitat with a Mediterranean climate. Both animals were housed together in a habitat that is 0.96 acres or approximately 42,000 square feet. A couple of weeks prior to calving, the female was moved to a smaller, adjacent habitat that is approximately 7300 square feet. Both habitats have natural dirt as the outside substrate with a variety of natural items such as logs, trees and rocks. Each habitat has a heated boma with a concrete floor and straw bedding.

Thermograms

Thermographic Images were captured weekly with two different models of FLIR® cameras:

FLIR® E5 XT and FLIR® E76 cameras. The E5 XT camera shows a wider field of view and quickly captures the images; however the resolution is slightly lower (160 x 120 resolution). The E76 captures a higher resolution (320 x 240 resolution) and narrower field of view with a more zoomed in photo but is accompanied by some shutter lag. The E76 also has the capability to capture video. Both cameras have relatively accurate temperature sensitivity, which was imperative for this study.

All thermograms were taken in the morning to avoid bias from environmental factors, like the sun, and reduce the chance of high physical activity from the rhinos. Overcast days were preferred. Conception occurred in November of 2021 and we began collecting thermographic images in early 2022. Thermograms of the female's vulva and the male and female abdominal regions, on both the left and right side, were taken weekly starting in March 2022 and continued postpartum. Thermograms of the female's teats were taken weekly starting in August 2022 and continued postpartum. Images were captured at a distance of no more than 15 ft away from each rhino but would typically be captured within a few feet of the rhino.

Opportunistically, video was captured with the E76 camera on overcast days. This video would start when no fetal movement was detected by both watching the female's abdomen with the naked eye and feeling it with a hand. We would record for varied lengths of time to capture the heat signature of her abdominal region as fetal movement increased. Video was also captured on a handheld video camera to compare any visible fetal movement to changes in heat signatures on the E76 camera.

Data Analysis

Images were sorted by date, individual and body location. Image analysis is being conducted using FLIR® Thermal Studio Suite Professional software. This software will be utilized to draw polygon shapes around the various relevant body regions and will allow extraction of specific temperature data. This includes looking at the relative temperature profiles of the abdominal region where the fetus is expected to sit, as well as the teats and vulva.

Changes in average, minimum and maximum temperature overtime will be analyzed for the male and female rhino on the abdominal region, including comparison of postpartum images. Temperature data from both the male and female abdominal regions will be compared to one another, with the male acting as a control for temperature differences associated with pregnancy. Average, minimum and maximum temperatures overtime will be analyzed for the heat signatures on the female's teats and vulva. Temperature data of the relevant body regions will also be compared to the rest of the female's body to determine if there is a difference in temperature.

All data will be exported from the FLIR® Thermal Studio Suite to a CSV file for data analysis.

Results

Data analysis is still currently being conducted. Some preliminary findings are listed below, however full data analysis has not confirmed significant findings at this point.

When analyzing the abdominal thermograms of the female southern white rhino, a change in temperature overtime, throughout her pregnancy, does not seem to be supported (figure 1). However, when comparing maximum and average temperatures of the abdominal region during pregnancy and post pregnancy there appears to be a difference (figure 2). The thermograms also support a difference in temperature readings between the male and female southern white rhino throughout the pregnancy (figure 3). When reviewing the thermograms of the female's teats, temperature changes are evident towards the end of her pregnancy and postpartum (figure 4).

Discussion

This project originally started with the intention of determining estrus in a southern white rhino using IRT. After we determined our female was successfully bred, the goal of the project shifted to pregnancy monitoring. We did not expect thermal changes in the vulva as this is related to estrus and once she was confirmed pregnant, signs of estrus should not be seen. We did expect to see changes in the maximum and average temperatures over time in the thermal images of her abdomen throughout pregnancy and in her teats towards the end of pregnancy and postpartum. We also expected to see differences in abdominal temperatures when comparing the male and female southern white rhinos as well as comparing the female southern white rhino during and post pregnancy.

Several of the thermograms were not suitable for analysis due to the angle the picture was taken or potential temperature bias, mostly from the sun. In order to accurately analyze the differences in the temperatures, we needed to ensure that the images were as consistent as possible and that any apparent environmental bias is eliminated. This should be prioritized in the protocol for taking thermographic images in future related studies.

As we continue to analyze the hundreds of thermograms collected through this study, more knowledge will be attained to compare this research to the studies that have occurred on other species such as the black rhino, babirusa and horses.

We expected to see an increase in temperature over time, however that does not seem to show when analyzing the thermograms. Instead, what we are seeing is a higher temperature in the pregnancy field compared to the rest of the female's body. We also saw a greater temperature difference between the female's pregnancy field and the rest of her body when the female is pregnant compared to postpartum. In addition, there appears to be a difference between the male and female abdominal temperatures supporting that a heat signature is evident along the flank and lower abdomen when the female is pregnant. It may be difficult to determine how far along a female is in her pregnancy with the abdominal thermograms, but it does appear that pregnancy can be detected.

We also monitored the changes in heat signature of the teats over time and there does appear to be an increase in max and average temperature on the teats when the female was getting closer to calving compared to earlier in pregnancy. This was expected and is attributed to the production and secretion of milk from the mammary glands.

As part of this study, we were able to compare the two FLIR® camera models: E5 XT and E76. In situations where the researcher may not be able to approach the animal as closely the E76 may be more beneficial due to the zoom ability and higher resolution. However, the E76 has a shutter lag which can be challenging with an animal that may move quickly or if you are trying to capture a specific image, e.g. the vulva of a rhino while the tail happens to be curled. The quick shutter speed and wide field of view of the E5 XT has many advantages, and considering it is at a lower cost, may be more practical for utilization in the field, as long as the researcher can get relatively close to the animal. While resolution is lower in the E5 XT model, it is still considered a high-quality infrared camera that produces suitable thermograms for temperature pattern

analysis using FLIR® Thermal Studio Suite. Throughout this study we found the benefits for each camera model varied depending on the image we were attempting to capture and cooperation of the rhinos.

The information gathered from this research is invaluable, especially with species and/or individuals that are unrealistic to collect voluntary blood or fecal samples from. Moreover, this research has the potential to translate to *in situ* research, as it allows for non-invasive data collection without physical contact with the animal. We are continuing this research project postpartum, to ascertain the use of thermographic imaging to monitor estrus cycles in southern white rhinos. Continued research on thermograms of the vulva region in nonpregnant white rhinos will provide insight into utilizing IRT to determine estrus cycles via thermal patterns and therefore provide more knowledge to better manage breeding this species under human care.

Further research on this topic will be necessary to confirm the results of this study, ideally including multiple individuals from other facilities and eventually expanding into thermogram collection of wild rhino populations. If specific temperature ranges can be determined that identify a pregnancy field, this would allow for accurate pregnancy diagnosis through a non-invasive and non-contact approach. Changes in heat signature on the teats may assist in predicting time until parturition. This will allow better management and planning for calves of rhinos under human care, as well as closer monitoring and protection of wild rhinos that typically separate themselves from their herd during calving (Owen-Smith, 2004). It would be beneficial to repeat this study in multiparous white rhinos to see if there is a difference in the thermal patterns compared to a primiparous female.

Conclusions

Thermographic imaging is a useful tool for reproductive management of exotic species such as southern white rhinos. There is a lot of evidence that supports IRT can be utilized to determine pregnancy in a variety of species. In addition, monitoring thermograms of teats in southern white rhinos can potentially be a tool to indicate how close a cow is to calving.

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species and has been integral to the analysis of the thermograms in FLIR® Thermal Studio Suite. Kate Fox, Safari West Conservation Coordinator, has also been reliably collecting thermographic images and video throughout the project and continues to work with the research committee on developing projects. Lastly, Lori McNeal, the lead keeper for the rhinos at Safari West, was always ready to assist in asking the rhinos to get into position to allow our research team to take the necessary images. This project was truly a collaborative effort.

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Figures

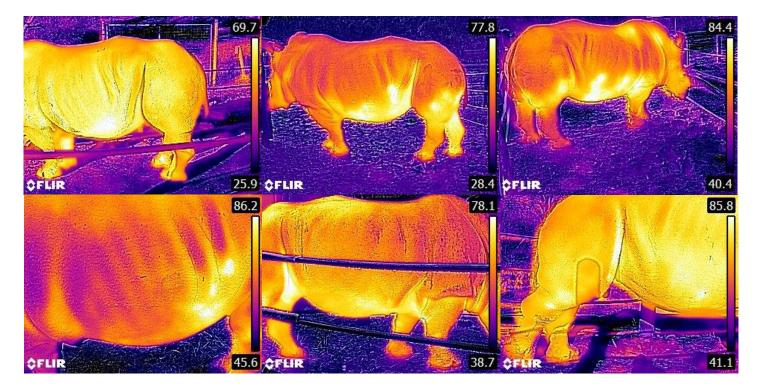


Figure 1. Abdominal thermograms from November (top left), December (top center), January (top right), February (bottom left), March (bottom center) and April - day of calving (bottom right).



Figure 2. Abdominal thermograms from January - 14 months pregnant (left), April - almost 17 months pregnant (center) and April - 14 days postpartum (right).

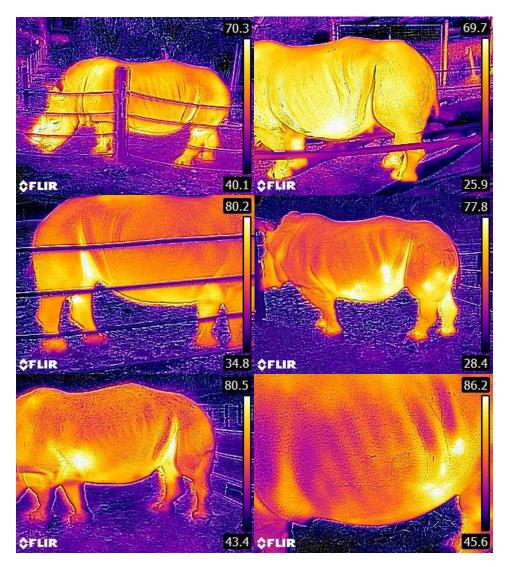


Figure 3. Abdominal thermograms of the male southern white rhino (left) and female southern white rhino (right) from November (top), December (middle) and February (bottom).

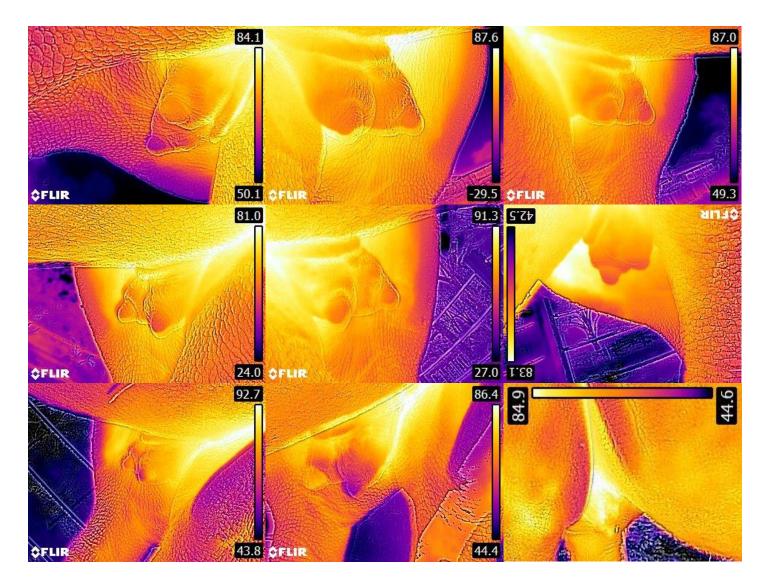


Figure 4. Thermograms of the female southern white rhino's teats from August 2022 (11 months pregnant) to April 2023 (7 days postpartum)