Noninvasive Monitoring of Physiological Metrics in Mammals

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Abstract

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Pulse rate and changes in blood flow in animals are indicators of illness, injury, and physiological responses to environmental changes. Noncontact monitoring of these metrics via thermal imaging is a critical step forward in assessing captive and wild animal health. In this study, I investigated whether the FLIR One Pro infrared thermal camera extension for smartphones could reliably detect pulse rate in semi-free-ranging domestic pigs. I recorded the base of the ears of pigs at rest, extracted mean pixel intensity from video frames, and converted the timestamps of spikes in intensity values to heart rate data. My findings suggest that there is a limited possibility that a cost-effective thermal camera extension can be used to accurately assess pulse rate in domestic pigs. Additionally, my findings confirm that low humidity and indirect sunlight are optimal atmospheric and weather conditions to maximize the precision of pulse rates detected in domestic pigs using infrared thermal technology.

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INTRODUCTION

A rapid pulse rate and redistribution of blood are indicators of acute stress in mammals (e.g. Bartolomé et al., 2013; Travain et al., 2015; Barrault et al., 2022). Monitoring acute stress is critical, especially during a potential medical emergency. Long-term stress induces biochemical changes that may have long-term health consequences, such as compromised immune responses (Sapolsky, 2021). Unfortunately, typical methods of taking these measurements in nonhuman animals involve stress-inducing physical contact including capture, restraint, anesthesia, and continual restriction of movement for study durations (Stukelj et al., 2022). For instance, the pulses of domestic pigs are generally monitored by a human or a computerized belt that they wear for up to 24 hours — both of which are invasive approaches (e.g. Olsson et al., 1999; de Jong et al., 2000; Kuwahara et al., 2004; Lyhne et al., 2022). Data collected with these methods don't reflect an animal's physiological responses to stimuli independent of human interaction.

Recently, noninvasive techniques have become more popular for monitoring animal physiology. For example, concentrations of hormones correlated with stress responses can be extracted from urine and fecal samples (Sapolsky, 2021). However, these hormones are involved in regulatory functions beyond stress. Data gathered in these studies aren't definitive indicators of stress. Incorporating novel methodology analyzing other physiological attributes into studies such as these will better illustrate the biological underpinnings of

physiological changes and behavioral correlates. In tandem with commonly used methodologies, new approaches to monitoring animal physiology will offer a more definitive assessment of mammalian health.

A pulse is a temporary change in blood volume that pushes oxygen-rich blood out of the heart. Recent research has confirmed that pulses can be detected remotely with cameras. Mean pixel color values in regions of interest in the human face captured by RGB (red-greed-blue) cameras, which capture videos with light in the visual spectrum, yield a pulse signal (Qiu et al., 2021). Pixel intensity values of video frames captured by near-infrared cameras can be used to calculate human heart rate variability metrics (Zhang et al., 2018; Selvaraju et al., 2022). Despite these advancements in the application of noncontact video technology to physiological monitoring, however, few studies have investigated the use of this technology with nonhumans, and fewer still have made use of robust infrared thermal technology to do so (e.g. Barbosa Pereira et al., 2019). However, those that have have demonstrated the broad applicability of thermal technology to the welfare of animals raised for agriculture, competitions, zoo housing, and human companionship (e.g. Bartolomé et al., 2013; Travain et al., 2015; Dezecache et al., 2017). Expanding on this research is a step toward increasing attention to the welfare of nonhuman animals in wild and captive settings.

The present study investigates whether using an infrared thermal camera to extract pulse rate from active pigs outside of a controlled laboratory setting is

feasible. The design of this study imitates realistic situations in which nonhuman animals that might benefit from these noninvasive monitoring techniques live. I hypothesize that an infrared thermal camera will register small changes in temperature in a well-vascularized region of pigs' ears when warm blood pulses through veins in that area. I expected to find that the camera registered minute increases in heat radiation from this well-vascularized region of interest when warm blood flowed through the pigs' veins in pulses. Because infrared thermal cameras convert heat radiation to "false colors" to represent heat distributions, I hypothesized I would find that once converted to grayscale images to remove confounding hue interference, variations in mean pixel intensity across video frames corresponded to fluctuations in heat radiation (Lavers et al., 2005; Cilulko et al., 2012). Thus, I expected to find that they correspond to pulses from which heart rate variability metrics can be calculated.

My hypotheses build on the framework for near-infrared and RGB video studies in humans in a novel way that accounts for differences between humans and pigs, including skin thickness and light conditions available during regular activity patterns. In addition to emphasizing the removal of invasive techniques from welfare evaluations, the design of this study prioritizes affordability. The high price for quality thermal imaging technology poses a significant barrier to thermal research (Cilulko et al., 2013). Therefore, I selected a lower cost model of thermal camera that integrates with a smartphone for this research. Overall, less than $2,000 was spent on equipment needed to conduct this study — a significant

reduction from the tens of thousands of dollars that some high-end thermal cameras cost.

Stress

Throughout their lives, animals interact with dynamic environments. They constantly adjust their physiological balance to interact optimally with these environments. An idyllic autonomic state is one in which internal organic processes are running efficiently such that the organism they support thrives — this is described by the term homeostasis (Porges,1995). Disruptions to this state occur as animals go about their regular activities. This indicates a healthy redirection of energy to meet animals' changing needs, such as a new demand for muscular support during locomotion. More substantial deviations from a homeostatic state are present when animals interact with stressors in their environment (Bartolomé et al., 2013). Stressors may be defined as perceived external challenges to an animal or the animal's anticipation preceding such challenges (Sapolsky, 2021). Regardless of whether a stimulus poses a genuine threat to an animal's survival, a stress response takes place to prepare the animal to react to that stimulus (Porges, 1995; Goldstein, 2013).

The mammalian autonomic nervous system consists of two branches: the sympathetic nervous system and the parasympathetic nervous system. The sympathetic nervous system is responsible for initiating physiological arousal, while the parasympathetic nervous system is responsible for re-establishing

physiological homeostasis (von Borell et al., 2007; Goldstein, 2013). Generally, sympathetic nervous system activity is correlated with an increase in heart rate. However, the sympathetic nervous system is not isolated from the parasympathetic nervous system; instead, many of the physiological processes the sympathetic nervous system initiates are also affected by parasympathetic activity (Porges, 1995; von Borell et al., 2007; Mohr et al., 2002). It is a challenge to distinguish between sympathetic and parasympathetic activity when analyzing mammalian physiological metrics. However, the Poly-Vagal Theory offers a framework that facilitates more precise classification of which branch of the autonomic nervous system is most active at a given point in time (Ioannou et al., 2014).

Mammals' vagus nerve directs responses in cardiac and respiratory organs of the body. The vagus nerve is capable of inhibiting the activity of the sympathetic nervous system and thereby preparing the body of a mammal to recover from arousal to a relatively homeostatic state (Routledge et al., 2002; Ioannou et al., 2014). Vagus nerve activity implicated in a stimulus response is referred to broadly as vagal tone. Under the Poly-Vagal Theory, fluctuations in vagal tone are considered indicators of stress reactions (Mohr et al., 2002). Therefore, cardiac output is a product of interactions between the sympathetic and vagus-moderated parasympathetic branches of the nervous system, often referred to as sympathovagal balance (Franchini and Cowley, 2004; Salomão et al., 2015; Krause et al., 2017).

Using cardiac metrics to analyze a mammal's basal autonomic state (i.e. under rest conditions) is an emerging method of gauging that mammal's susceptibility to stress. Individuals exposed to stressful situations early in life show reduced basal vagal tone. Without a strong parasympathetic nervous system, they may be more vulnerable to significant fluctuations in physiological homeostasis when they encounter even small stressors (Geverink et al., 2002; Zupan et al., 2016; Krause et al., 2017). Of course, this reactivity is individually variable. Additionally, no observations of consistent trends across mammalian taxa have been established due to the novelty of studies investigating correlations between basal vagal tone and susceptibility to stressor-induced arousal. Gathering physiological data when mammals are at rest may nonetheless be valuable in that it may reveal how susceptible they are to stress in their lifetimes.

Heart Rate Variability as a Stress Response Metric

The Significance of Heart Rate Variability

Heart rate variability is an umbrella term that encompasses small changes as well as longer-lasting alterations in an organism's heart rate. The cardiac cycle is not constant in duration. The length of interbeat intervals, or the amount of time between heartbeats, is modified so that the cardiac cycle can compensate for any discrepancies or deviations in factors such as blood pressure and blood gas concentrations (Porges, 1995; von Borell et al., 2007).

Because heart rate is directly affected by interactions between the sympathetic and parasympathetic branches of the autonomic nervous system, observed variation in heart rate as a function of time can serve as an indication of a shift in autonomic balance (Uceda et al., 2020). Heart rate variability is expected to be high in healthy animals (Mohr et al., 2002). Indeed, among humans with cardiovascular complications, heart rate variability tends to be low (Routledge et al., 2002). Low heart rate variability indicates a breakdown in the biological circuits monitoring homeostasis, which prevents necessary tweaks being made to heart rate. This in turn inhibits animals' ability to respond to internal and external challenges — their cardiovascular dynamics are restricted when they must dedicate more energy to reacting to a new load on their system (Mohr et al., 2002).

Physiological responses are leveraged to respond to regular occurrences as well as stressors. For instance, the delicate balance between sympathetic and parasympathetic control of short-term variation in heart rate is noticeable in regular breathing patterns. Heart rate typically increases when an animal inhales and decreases when they exhale. This pattern, referred to as respiratory sinus arrhythmia, is expected in mammals and considered an indicator of healthy regulation of physiology by the autonomic nervous system (Mésangeau et al., 1999).

Measuring Heart Rate Variability

Heart rate variability is a metric that can be accurately and precisely extracted from variable durations of monitoring time. Heart rate variability measurements are calculated from variations in heart rate detected during monitoring periods. Because it is impractical to collect heart rate data from animals over exceptionally long periods of time, studies generally consist of data collection over periods lasting up to 24 hours (e.g. Kuwahara et al., 2004; Lyhne et al., 2022). For studies lasting many hours, electrocardiograms are constructed from devices that are strapped to animals for the duration of the study (e.g. Olsson et al., 1999; de Jong et al., 2000). A data collection period of 5 minutes is considered acceptable for extracting precise heart rate variability data when electrocardiographic measurements cannot be taken continuously by a computer (Task Force, 1996). Heart rate variability analysis for both long- and short-duration monitoring studies can be separated into two broad categories: time domain analysis and frequency domain analysis.

Time Domain Heart Rate Variability Analysis

Time domain analyses use interbeat intervals to assess the strength of factors contributing to heart rate variability. Long-term heart rate variability can be extrapolated from time-domain variables because instantaneous heart rate measurements are considered indicators of long-term heart rate variability (von Borrell et al., 2007; García et al., 2014; Salomão et al., 2015).

If multiple data sets are being compared in the time domain, they must be the same length. This standardizes the number of data points (i.e. heartbeats) registered in each data set. Heart rate variability is positively correlated with data collection time. Therefore, datasets of variable lengths will skew calculations to reflect more or less variability in heart rate, respectively (von Borell et al., 2007).

Of time domain parameters extracted from collected data, the root mean square of successive interbeat intervals (RMSSD) is considered the most universally informative. RMSSD is the square root of the squared, summed, and averaged differences between interbeat intervals observed in a data collection period (Uceda et al., 2020). As it reflects heart rate variability in the short term, which is influenced directly by the parasympathetic nervous system (Porges, 1995; Mésangeau et al., 1999), it is accepted as an indicator of parasympathetic activity (Zebunke et al., 2013; Uceda et al., 2020). The number of successive intervals between normal heart beats (i.e. beats that are initiated by the sinoatrial node in the heart and thus reflect a full heart beat) that exceed 50 ms in differences between each other is expressed as a percentage. This is the pNN50 measurement (Shaffer and Ginsberg, 2017). The statistical value of pNN50 is similar to that of RMSSD; however, RMSSD is more closely tied to indications of respiratory sinus arrhythmia and is thus preferred to the pNN50 (Shaffer and Ginsberg, 2017). Additionally, RMSSD can be used in conjunction with other metrics to provide more robust statistical analyses of the autonomic valence of a physiological response.

Interbeat intervals may be used to calculate the standard deviations between pulses within five-minute periods. This measurement, abbreviated as SDNN, is used to calculate total heart rate variability over the period of data collection (Uceda et al., 2020). The ratio between SDNN and RMSSD reflects the overall balance between sympathetic and parasympathetic activity, much like the high-to-low-frequency ratio in the frequency domain discussed below. Whether calculated separately or later combined into this ratio, SDNN and RMSSD are informative about variability in heart rate in response to different conditions. They may reflect baseline autonomic balance in an animal while the animal is resting or autonomic activity in that animal when facing stressors (Mohr et al., 2000).

Thus, time domain heart rate variability metrics illuminate changes in sympathetic and parasympathetic nervous system activity that influence heart rate. There are additional variables that have not been discussed here that provide additional insight into autonomic modulation (e.g. Uceda et al., 2020). The widely used variables described heretofore, however, capture the significance and applicability of time-domain heart rate variability analysis.

Visualizing Time Domain Heart Rate Variability Components

Capturing heart rate variability metrics via videography presents unique challenges when it comes to interpreting and presenting input. Independent Component Analysis isolates independent signals from video sources and is therefore especially useful for breaking down color (RGB) and near-infrared

videos because isolation entails the separation of channels embedded in video input — like red, green, and blue channels (Poh et al., 2010; Zhang et al., 2018). Independent Component Analysis is a practical approach to displaying time- and frequency-domain metrics simultaneously.

Tachograms and Recurrence Quantification Analysis, however, are more specialized than Independent Component Analysis for exclusively displaying time-domain variables. Defined simply, tachograms depict interbeat intervals on a time scale. Recurrence Quantification Analysis operates on the principle that heart rate variability determinants are not always linear. Recurrence plots display each signal datum (e.g. interbeat intervals) as functions of the previous datum. This creates a robust visual display that is buffered from influence from the size or assumed distribution of a dataset (von Borrell et al., 2007).

Frequency Domain Heart Rate Variability Analysis

Frequency domain analyses separate heart rate data into three primary constituents (Salomão et al., 2015). The high frequency (HF) component is a short-term heart rate variability metric and is thus chiefly responsive to parasympathetic influence, rendering it an effective tool with which to gauge vagal activity and the intensity of respiratory sinus arrhythmia (Salomão et al., 2015; Uceda et al., 2020). The low frequency (LF) and very low frequency (VLF) components are modulated by both the sympathetic and parasympathetic branches of the autonomic nervous system; LF fluctuations have widely been considered

indications of the intensity of sympathetic tone but when less variable may also reflect less parasympathetic activity (Voss et al., 2003; Salomão et al., 2015; Uceda et al., 2020). The difference in the determination of the frequency components that make up a heart rate signal allow them to be quantified separately and later combined into ratios that convey the magnitude of autonomic activity during data collection periods. For instance, the LF/HF ratio is widely believed to index overall interactions between the sympathetic and parasympathetic branches of the autonomic nervous system — sympathovagal balance — although basal physiological functions such as thermoregulation may influence this index, making it active when no observable stimuli are present (von Borell et al., 2007; Salomão et al., 2015).

Therefore, frequency domain heart rate variability metrics are as informative as time-domain metrics when assessing autonomic activity in animals; variables in each domain have similar applications for animal health and stress response analyses.

Visualizing Frequency Domain Heart Rate Variability Components

The Fast Fourier Transform (FFT) mathematically breaks down the components of a signal into its composite sine and cosine waveforms. Its underlying principle is similar to that of ICA but is applied specifically to frequency-domain heart rate variability parameters rather than time and frequency domain metrics (von Borell et al., 2007; Cygankiewicz and Zareba, 2013). As per

the recommendations of the Task Force of The European Society of Cardiology and The North American Society of Pacing and Electrophysiology, short-term, non-parametric heart rate variability analyses in the frequency domain should sample at least 5 minutes of cardiac activity to ensure validity of results. This recommendation is also in the interest of standardizing short-term heart rate variability studies and is posited alongside a call to use 24-hour recordings for time domain processing where possible (Task Force, 1996).

Infrared Thermography: A Unique Approach to Assessing Heart Rate Variability Metrics

Applications of Videography to Remote Photoplethysmography in Humans

Photoplethysmography applies known principles of photon activity to the detection of respiratory and heart rate signals (Nilsson, 2013). Living tissues both absorb and reflect light. In humans, basal absorption of tissue, blood, and bone is changed by oxygenated blood. Oxygenated blood pushed out by the heart muscle in a pulse absorbs more infrared light than deoxygenated blood. This produces a visible difference in pixel intensity across frames in near-infrared time-of-flight cameras applied to remote photoplethysmography tests (Kveisis-Kipge and Rubins, 2016; Nahler et al., 2018).

Despite the promising applications of remote photoplethysmography among humans, several of its inherent limitations make it difficult to apply to nonhuman animals. Movement changes the orientation of the skin surface relative

to light sources and thus alters skin reflectivity. Skin reflectivity is measured and presented by the RGB camera as a variation in skin color. This confounds remote photoplethysmographic measurements collected with RGB cameras (Qiu et al., 2021). Near-infrared waves penetrate human skin partway through the epidermis layer. This fortifies near-infrared imaging against disturbances such as makeup and movement (Zhang et al., 2018), but does not guarantee the same level of efficacy for this methodology when it is applied to nonhuman animals with skin thicker than that of humans. Although near-infrared imaging can be more optimal than RGB imaging for capturing heart and respiration rate, thermal imaging eliminates confounding light variation entirely.

Infrared imaging facilitates the monitoring of blood vessels without interference from lighting in the environment. A critical caveat, of course, is that blood vessels must be clearly in view of the infrared camera to reduce as much noise as possible (Kim et al., 2018). Regardless, controlling for environmental lighting expands the horizons for the application of noncontact heart rate variability data collection among nonhuman animals, whose movements cannot always be controlled and therefore complicate extraction of such data from RGB videos.

Limitations of Infrared Thermal Imaging

Although infrared thermal imaging is an intriguing alternative to light-dependent cameras for the detection and monitoring of heart and respiration

rate, it is not without its constraints. Weather conditions as confounding variables, atmospheric disruptions at long distances, physical movement of a subject (including the use of muscles to display emotion), foliage, hair or fur on a subject, and physiological responses induced by pharmacological substances and/or environmental stressors are among the most glaring limitations of infrared thermal cameras (Cilulko et al., 2012; Clay-Warner and Robinson, 2014). The high cost of infrared thermal technology also erects barriers for researchers. High-quality infrared thermal cameras often cost hundreds or thousands of dollars.

Variable Applications of Infrared Thermography

Despite the inherent limitations of infrared thermal imaging, infrared thermography cameras offer unique insight into physiological responses that are not visible to the naked eye. For example, nonhuman animal interactions involve components that are masked in front of conspecifics and/or human observers. These include fluctuations in stress levels and emotional responses. Because emotional responses are intricately tied to physiological responses, illuminating physiological responses in interactions between nonhuman animals may broaden human understanding of emotional states among animals (Kuraoka et al., 2011; Barrault et al., 2022; de Vevey et al., 2022).

Infrared thermal cameras depict changes in the distribution of heat across animal subjects in the visual spectrum. Recording the redistribution of bloodflow in the facial regions of primates is made possible by infrared thermal imaging, as

increased bloodflow to a specific region increases the heat of that region. Redistributed bloodflow in primates' faces is correlated with behavioral reactions indicative of positive and negative emotional valence in their reactions to stimuli in controlled and wild environments (e.g. Pavlidis et al., 2002; Braesicke et al., 2005; Grandi and Heinzl, 2016; Kano et al., 2016; Dezecache et al., 2017; Chotard et al., 2018; Ermatinger et al., 2019; Brügger et al., 2021; Ross et al., 2021; Barrault et al., 2022). More generally, infrared thermal imaging has proved valuable with regard to demographic assessments of wild mammals, the detection of infections and diseases in mammals, the assessment of mammalian metabolic expenditure, and the monitoring of some reproductive processes among mammals and birds (e.g. Arenas et al., 2002; Nakayama et al., 2005; Dunbar and MacCarthy, 2006; Durrant et al., 2006; MacCafferty, 2007; Hilsberg-Merz, 2008; Hristov et al., 2008; Dunbar et al., 2009; Dezecache et al., 2017; Burke et al., 2019).

In the present study, I investigated whether infrared thermal cameras may be used for the collection of heart rate variability data. I envisioned thermal cameras being used to execute this methodology being simultaneously applied to the assessment of other behavioral and health metrics in captive and wild nonhuman animals. Ideally, this would minimize the cost of the acquisition of research equipment for monitoring physiological changes in animals.

MATERIALS AND METHODS

Pig Sample

Domestic pigs (*Sus scrofa domesticus*) are the product of a complex history of the habituation and reproduction of wild pigs. In the United States of America, domestic pigs are found in a majority of states; however, they were first introduced by Spanish explorers to land adjacent to the Gulf of Mexico (Mississippi Dept. of Wildlife, Fisheries, and Parks, 2022). This initial population of domestic pigs was used as a food source and marks the first arrival of swine in the Americas (Mississippi Dept. of Wildlife, Fisheries, and Parks, 2022). Across Europe and the United States, domestic pigs encompass a wide range of breeds that are still predominantly used for agricultural exploits (Camerlink, 2017). Continued demand for pig meat has resulted in increased efficiency of domestic pig breeding and housing in the agricultural sector. While these pig breeds would roam in areas with trees and underbrush in the wild, they are kept in close proximity to conspecifics and often provided little to no enrichment (Camerlink, 2017). This has in turn attracted inquiries into the welfare of domestic pigs being raised for meat yield and prompted some legal control of pig housing at the national and international levels (Camerlink, 2017). Domestic pigs are also raised for laboratory testing, in which they are subjected to invasive procedures usually aligned with biomedical hypotheses (Camerlink, 2017). Those that are kept as pets are bred in large groups and deprived of nutrition to stunt their growth. This deprivation introduces significant problems later in their lives, including

neurological disabilities and digestive issues (Central Texas Pig Rescue, 2023). The myriad uses of domestic pigs for agriculture, research, and entertainment has encouraged dialogue about reducing stress levelled on domestic pigs and improving their welfare in captivity. Today, domestic pigs seized by the United States government from breeders subjecting them to abuse are distributed to organizations equipped to care for them, including nonprofit pig rescues across the country (Central Texas Pig Rescue, 2023).

An initial total of 16 *Sus scrofa domesticus* specimens were sampled throughout this study at the Central Texas Pig Rescue in Bastrop County, Texas (centraltexaspigs.org). These pigs are semi-free-ranging and live in groups of approximately 20-30 individuals per enclosure. Enclosures vary in size but are spacious proportional to the number of pigs they house. All enclosures encompass small trees that create a forest for the pigs to engage with. They also include at least one large clearing covered with sandy soil, hay, and low platforms built from wooden planks that their food is served on.

The pigs sampled for this study come from a wide variety of backgrounds. Many pigs at Central Texas Pig Rescue were taken in by the rescue following a history of abuse by humans. Thus, some individuals are reluctant to be approached by humans. The rescue pigs socialize regularly with human volunteers. Twice per day, the pigs are fed nutritional pellets supplemented with frozen fruits and vegetables. They have access to water ad libitum and sleep in variably sized enclosed structures referred to as “cabins”. Hay inside the pigs’

cabins and scattered around the clearings in their enclosures is replenished regularly. Extra hay is added to their cabins and enclosures when cold weather is forecasted.

Pigs were included in the initial sample for this study if they met the criteria listed in Table 2. These criteria were established to minimize the interference of novel stimuli in their environments with their pulse rate at the time of sampling. Ultimately, pigs were sampled from 4 enclosures. Recordings of 7 pigs did not meet video duration requirements or contained too many motion artifacts from pig and/or camera movement. These pigs were excluded from the pre-processing sample. The 28 thermal video files included in this study yielded 32 adequately long recordings of appropriate regions of interest from 9 pigs. See Table 1 for descriptions of individuals included in this pre-processing sample.

Design

I gauged the efficacy of the proposed use of infrared thermal imaging as a tool to extract pulse rate from mammals without physical contact. I wrote computer programs to extract timestamps of fluctuations in mean gray levels (i.e. mean pixel brightness) of grayscale images derived from frames of thermal video files. I fed the timestamps of high pixel brightness levels into an open source heart rate variability analysis program that processed them in the time domain.

Procedures

Observational Conditions

I sampled pigs between March 3 and March 18, 2023, when weather conditions in Bastrop County were highly variable. Therefore, I sampled pigs across a wide differential of temperature and humidity conditions (Table 3).

I approached pigs when they were resting on their stomachs or sides. If potential sample pigs shifted to assess my approach, I waited to continue approaching until they had settled back down for 10 or more seconds. Once potential sample pigs met the criteria included in Table 2, I kneeled or sat on the ground beside them. I recorded whichever surface of their ear that the base of their ear was fully visible from. Distances between myself and sample pigs at times of filming varied with the size of the pig as well as their degree of shifting as I approached them. With a few exceptions, if a pig was large (> 600 lbs), I maintained a larger distance between myself and the pig (1-4 feet) as opposed to a medium-sized pig (75-250 lbs, 0.25-4 feet).

Infrared Thermal Video Procedure

Once I approached a pig for filming, I secured the FLIR ONE Pro thermal camera extension from Teledyne Flir to my smartphone. I measured the distance between the camera and a point approximately halfway between the lens and the

pig being filmed. Doubling this distance gave me an estimate of how far away the camera was from each subject.

I proceeded to record the base of one of the pig's ears with the thermal camera extension. The base of a pig's ear is a well-vascularized region (Zezula-Szpyra et al., 2000; Duisit et al., 2017). Thus, it is an ideal place to record minute changes in temperature that are more likely to be associated with pulsations of blood through the pig's veins as opposed to atmospheric conditions or other confounding variables. The front and back of the ear are both appropriate sampling locations. I recorded whichever side of the ear was most accessible to me in terms of my proximity to the pig and the angle of the pig's ear.

Although the nature of this study necessitates recordings 5 minutes in length, I recorded pigs for intervals between 5 and 6 minutes (Task Force, 1996). This accounted for any initial physiological adjustments pigs made to myself and the camera, two stressors in their environment, at the beginning of each recording. When a recording was complete, I took a truecolor image of the pig that had been recorded. This facilitated pig identification and the assessment of potential confounding variables such as behavioral thermoregulation and heat retention of pelage at a later date.

Assessment of Atmospheric Conditions

While filming each pig included in the sample, I laid a General Tools digital temperature and humidity pen (PTH8708) between 1 and 1.5 feet away

from the pig. At the end of each filming period, I recorded the minimum and maximum temperature and humidity to reflect a range of atmospheric variation at the time of filming. The temperature and humidity pen registered variations in atmospheric conditions close to each pig in my sample. This facilitated the analysis of confounding atmospheric variables by creating a record of those conditions in pigs' immediate vicinities, which often differed from local atmospheric conditions due to the interference of soil moisture, heat retention in hay and grass, and other ground-level variables that influenced atmospheric conditions where pigs were laying. This was particularly important on cold or hot days, when pigs exhibited behavioral thermoregulation.

On cold days, pigs laid on and around hay as a form of behavioral thermoregulation. Temperature and humidity recorded by the pen were universally higher near the hay pigs laid on when the outside local temperature was low. On hot days, pigs laid in shade and/or dug small ovular trenches to lay in. Temperature recorded by the pen tended to be lower in the shade and beside the trenches pigs dug when the outside local temperature was high. The data collected from the temperature and humidity pen allowed me to assess the degree of behavioral regulation pigs exhibited in response to weather conditions. This granted me a greater degree of accuracy in recording atmospheric conditions that reflected the environments pigs were engaging directly with while they were being recorded. Table 3 details atmospheric conditions associated with each piece of thermal video data.

Data Analysis

To extract a potential pulse signal from noisy thermal videos, I cropped each video file and pre-processed them using MatLab and shell scripts so that I could input timestamps assumed to indicate an increase in bloodflow (i.e. a pulse) into the open source R package RHRV for robust heart rate analysis (rhrv.r-forge.r-project.org).

I cropped each video file in iMovie such that each frame displayed only the region of interest captured in the original file (Fig. 1). My first MatLab script, GILT (Grayscale Intensity Level Tool; modified from Image Analyst, 2023), iterates through every frame of a video file. GILT converts each frame to a grayscale image to standardize the "false color" values representing variations in temperature that the camera outputs in the visual spectrum. It outputs a graph containing the mean pixel intensity of the grayscale image files as a function of video framerate (Fig. 2).

I used a shell script to calculate timestamps for the intensity values contained in the output files from GILT. I input the modified file created by the shell script into my second MatLab program, PIGS (Peak Intensity Graph Script). PIGS graphs fluctuations in mean pixel intensity from each video frame as a function of time (Fig. 3). It then extracts the timestamps of peaks in mean pixel intensity. These timestamps are stored as a sequence in an Excel sheet that I

converted to a TSV file suitable for use with analytical packages in the R programming language.

Under my hypothesis that fluctuations in mean pixel intensity in an infrared thermal video will yield insight into a mammal's pulse, I assumed that a peak in mean pixel intensity indicated a rise in blood volume underneath the skin. Therefore, I made the assumption that each timestamp of peak mean pixel intensity values represented a pulse of blood through the pig's arteries and veins — thus, they were immediately preceded by a heartbeat. RHRV interprets timestamps in a sequence as a series of heartbeats. I imported peak intensity timestamps into RHRV as a sequence so they would be registered as such.

I applied only the time domain analysis functions in RHRV to my data. I constructed non-interpolated (i.e. not modified) instantaneous heart rate graphs from intensity peak timestamps. These graphs represent changes in the frequency of heartbeats as a function of time (Fig. 4, Fig. 5).

Where necessary, I manually removed extraordinarily large or small instantaneous heart rate values. These were likely the result of motion artifacts or atmospheric conditions influencing intensity values in grayscale video frames. The PIGS script may have registered artificially high frequencies of intensity spikes due to interference from these factors. Otherwise, heart rate graphs remained non-interpolated.

I examined each instantaneous heart rate graph to assess the likelihood that genuine pulses (and thus normal heart beats) were being represented. I did so

by analyzing signal consistency, signal precision, and signal clarity. I deemed a signal precise when high and low instantaneous heart rate values interchanged with a regular midpoint on the y-axis. If this midpoint hovered at 60 beats per minute and above, I considered it likely that the graph registered at least part of a true pulse signal consistent with resting heart rate expectations for healthy adult pigs (Fig. 6, Fig. 7). The heart rate of healthy weaned pigs hovers between 90 and 100 beats per minute — this is a decrease from their average heart rate in earlier life stages and is part of a larger trend in average resting heart rate decline that lasts into adulthood (Barbosa Pereira et al., 2019). Therefore, I expected the average resting heart rate of the pigs included in my final sample to be less than 90 beats per minute but did not reasonably expect it to hold constant below 60 beats per minute.

I used RHRV to analyze data that fit my criteria for likely representing normal heart beats in the time domain. This yielded SDNN, RMSSD, and pNN50 values for each graph. In the absence of robust, publicly available heart rate variability data for resting adult domestic pigs, I evaluated the likelihood that these statistics were accurate according to the extremity of the values themselves as well as their variability. I considered values that were especially high or low to be potentially inaccurate. I deemed high variability statistics reflective of a lack of precision in the potential pulse rate my equipment and programs detected. Because pNN50 is expressed as a percentage. I would not expect almost all or almost no adjacent intervals between normal heart beats to exceed 50 ms, so I

would be cautious of the validity of a pNN50 value approaching 0 or 100. I evaluated the validity of SDNN and RMSSD based on how extreme they indicated variability in a heart rate sequence to be. The higher the SDNN and RMSSD values, the less consistent a probable pulse rate signal was and the more likely I was to consider the potential pulse invalid.

RESULTS

Accepted Signals

Out of 32 processed potential heart rate signals, 11 were deemed adequately consistent, precise, and clear so as to be considered accurate reflections of a pulse rate in the pigs being recorded. It was unnecessary to modify any of the signals included in this dataset to exclude extreme values; all instantaneous heart rate sequences remained non-interpolated.

Final Pig Sample

A total of 5 pigs were represented by the data most likely reflecting a heart rate signal. 2 potbelly pigs, 1 American Yorkshire pig, and 2 KuneKune pigs made up this sample. Each breed yielded 3, 1, and 7 pieces of data, respectively. A majority of these pigs — the potbelly and KuneKune pigs — had dark pelage covering the skin of their ears. Only one, the American Yorkshire pig, had a lighter pelage. A description of these pigs can be found in Table 1.

The variable orientation of these pigs' ears necessitated the selection of different regions of interest in thermal recordings of each individual. The tips of the potbelly pigs' ears are oriented dorsally, whereas the American Yorkshire and KuneKune pigs' ears point laterally from their heads at an angle $\leq 45°$. Data was collected from the lateral surface of the potbelly pigs' ears and the posterior surface of the KuneKune pigs' ears. Because the American Yorkshire pig was

stretched out on the ground at the time of recording, the anterior surface of the pig's ear was captured by the thermal camera.

The thermal camera was held at an average distance of 14.65 inches away from the pigs' ears for the recordings included in this final sample, with most recordings being made at distances ranging between 13 and 19 inches.

Time Domain Analysis

SDNN, RMSSD, and pNN50 values were recorded following the conduction of time domain analysis for each piece of data included in the final sample. SDNN values had a mean of 484.178027 ms and a standard deviation of 62.8545191 ms. Values for RMSSD had a mean of 668.216564 ms and a standard deviation of 83.831904 ms. Finally, pNN50 values had a mean of 91.8184727% and a standard deviation of 1.13715631 percentage points. See Table 4 for SDNN, RMSSD, and pNN50 values associated with each piece of data. See Tables 5, 6, and 7 for descriptive statistics spanning all SDNN, RMSSD, and pNN50 values from the final sample.

Atmospheric Conditions for the Final Pig Sample

91% of the data included in the probable heart rate group were collected under conditions in which sunlight was indirect or obscured.

Humidity across the data deemed potentially reflective of pigs' heart rate tended to be lower in the pigs' immediate vicinity than both the humidity levels of

the pig rescue property and the county. Humidity varied between an average range of 57.2% - 62.9% during filming times, while the average humidity of the pig rescue and the county sat at 75% and 77%, respectively.

DISCUSSION

This study explored the feasibility of a cost-effective method of monitoring pig pulse rate noninvasively and without physical contact. I hypothesized that mean pixel intensity values extracted thermal video frames standardized as grayscale images would fluctuate over time in tandem with a pig's pulse. The results of this study did not offer a definitive answer as to whether assessing fluctuations in pixel intensity at the base of pigs' ears using the FLIR One Pro thermal camera extension is adequate for deriving an accurate metric for instantaneous changes in heart rate or extracting time domain measurements from thermal videos. However, they demonstrated that there is a possibility this thermal camera model is capable of detecting a pulse rate that can be used to analyze heart rate variability in semi-free-ranging pigs at rest.

Final Pig Sample Findings

The low number of pieces of data collected that may indicate the detection of a heart rate in this study reduces my confidence recommending the methodology involved in this study as unequivocally effective. However, the presence of this data even within such a small pig sample as that which was included in this study points to the possibility that this method may be effective if it continues to be tested under variable conditions and modified to conform to novel insight regarding its enhancement.

Limitations

The pig sample size involved in this study was limited due to my strict criteria for pigs suitable to approach and film. The pool of study subjects ultimately included as candidates for having potentially accurate instantaneous heart rate graphs was even more restricted. It is possible that the 5 pigs included in the probable heart rate group at the end of my study share a unique anatomical feature that made it easier for the thermal camera to detect heat fluctuations on their ears as opposed to those of other pigs. This might include an especially large artery or vein at the base of their ear or exceptionally thin skin in that area.

The instantaneous heart rate graphs themselves are not consistent between each other. This could indicate an issue with my equipment or the scripts I used to process videos. Regarding equipment, it is impossible for technology to be accurate and precise without fail each time it is used. This principle, coupled with the fact that I selected a more affordable thermal camera model for use that may not be as precise as more costly models, may have introduced variability in my data. Although these factors may have limited my study, testing cost-effective methodological designs is important. Testing modifications to my MatLab scripts (GILT and PIGS) may mitigate more discrepancies in the thermal video files that were not explicitly noticeable in this study. This may also tailor the data extracted from video files to better reflect potential pulse rates such that time domain analyses yield less variable statistics. I designed those scripts; as such, there is an

inevitably high risk of error in the video processing methodology I used as a result of erroneous input. Sophisticated technological models utilizing artificial intelligence and other cutting-edge programs to filter noisy thermal imaging signals have been used in recent studies (e.g. Barbosa Pereira et al., 2019; Niu et al., 2019). My programs may not have been as powerful in terms of removing all motion artifacts and other errors from my dataset before I processed peak intensity values as heart rate sequences.

Atmospheric Influence on Thermal Videos

An overwhelming majority of the pieces of data included in the probable heart rate group were collected under shady or cloudy environmental conditions. Prolonged exposure to sunlight in thermal recordings excluded from this group may have masked small changes in thermal radiation from the pigs registered by the camera, especially since pigs' ears are thin appendages usually covered in a blanket of hair and thus susceptible to quick changes in temperature. This susceptibility, which varies individually by nature of the different colors and thickness of skin and pelage, may in turn influence the amount of thermal radiation the camera detects from their surface. Under the analytical protocol I used, sunny conditions could have created artificially high intensity value readings in grayscale video frames, thus disrupting any components of a potential pulse rate that may have otherwise been picked up.

Lower humidity levels are ideal for thermal imaging. Thermal cameras struggle to register thermal radiation that is distorted by water particles in the air (Lavers et al., 2005). If humidity is higher in the immediate vicinity of an object being monitored, the information that the thermal camera collects will be disrupted by this distortion. It was advantageous in this study that humidity closer to the pigs themselves was lower than the humidity recorded for the rescue property and the county the rescue is located in, as this reduced interference of water particles with the thermal camera. Therefore, the thermal camera was operating at a high rate of precision as it was being used to record pigs. This enhanced its performance in terms of monitoring minute changes in temperature due to pulses.

Accuracy of Time Domain Analysis

The SDNN and RMSSD values yielded by the time domain analysis of the final sample involved in this study suggest high variability within potential heart beat sequences. The mean of SDNN values from the final sample exceeds half a second. Although SDNN values tend to be more accurate over the course of 24-hour sampling periods as opposed to 5-minute sampling windows, such a substantial measure of variability within heart rate sequences indicates an especially notable lack of consistency in the regularity of probable heart beats. Ultimately, high variability within time domain statistical measures is not promising in terms of validating the methodology investigated in this study. The

high mean of RMSSD values across data included in the final sample raises concerns along similar lines. This points to an overall lack of accuracy within sequences of probable heart beats included in the final data sample.

Although the variability of pNN50 values is less than the variability of SDNN and RMSSD values, the proximity of the pNN50 mean to 100% is worth pondering. Such a high mean indicates that intervals between normal heart beats adjacent to one other overwhelmingly differ by 50 ms or more (Shaffer and Ginsberg, 2017). I expected lower pNN50 values across the data in the final sample to be lower. It is possible that these values are high because the PIGS script detected intensity values that are more widely distributed than an actual pulse signal that the infrared thermal camera registered. The PIGS script filters peak mean intensity values extracted from grayscale thermal video frames according to the difference between the peak value and the values immediately preceding it (i.e. the prominence of each peak). It additionally filters out peaks that are especially close together. These criteria were incorporated into the program to filter out motion artifacts and other confounding variables that induced artificial mean intensity spikes. However, the minimum required distance between peaks included in the script may be too large and thus exclude accurate pulse data from the final sequence of probable pulses. This in turn increases the amount of time logged between pulses and drives up the pNN50 values. Modifying the programs used to identify potential pulses in this study or acquiring more robust signal processing applications may improve the precision and

accuracy of time domain statistics for probable pulse rates registered by the FLIR One Pro thermal camera extension.

Behavioral Observations

This study was not conducted in isolation. All videos collected, including those ultimately making up the probable heart rate group, were influenced by fluctuations in physiology due to uncontrollable environmental factors.

Pigs are social animals. In the semi-free-ranging pig groups involved in this study, it was common for pigs to be moving around in their enclosures while I was filming resting individuals. Pigs change their heart and respiration rates when they respond to one another's presence. For instance, they often grunt if they are approached by another pig while laying down. This disrupts their basal respiration rate by quickly forcing air through their snout. Healthy pigs' heart rates increase in response to stimuli such as an approach by another individual; this is tied to a sympathetic response and prepares their bodies to fight or flee.

Pigs also respond to weather conditions. They exhibit behavioral thermoregulation as a response to temperature. When outside temperature is high, they may lay in shade, stretch out in dry dirt trenches, or soak in a pond. When outside temperature is low, they may cluster together, find an enclosed shelter to lay in, or surround themselves with heat-retaining hay. Remnants of dirt, mud, water, or hay on their skin affect a thermal camera's ability to register thermal radiation from the tissue that is obscured by those remnants. Thus, behavioral

thermoregulation may interfere with thermal data collection. This may have contributed to the inconsistent and imprecise potential heart rate signals extracted from the thermal data collected for this study, thereby limiting the number of pieces of data included in the final sample of 11 probable heart rate signals.

Lastly, pigs exhibit reflexive responses to factors dependent on atmospheric changes that may confound thermal data. For example, pigs flick their ears to dispel flies. Ear flicking activates muscles in the ear region, so blood flow to those muscles increases to sustain their movement. Overall pixel intensity of thermal images increases as the thermal camera converts this heat spike into a visual display; more peak pixel intensity values factor into a higher heart rate under my methodology. Flies thrive in damp environments and I observed them to be most prolific in the pigs' enclosures when humidity levels were high. By virtue of attracting flies, damp and rainy weather conditions were correlated with an increase in pig ear flicking that had the potential to disrupt accurate pulse signals extracted from thermal videos.

Behavioral observations in the present study are emphasized more heavily as confounding variables than they are in similar studies conducted in the past (e.g. Olsson et al., 1999; de Jong et al., 2000; Kuwahara et al., 2004; Lyhne et al., 2022). Past studies examining heart rate variability in domestic pigs took place in controlled environments and made use of technology that had direct contact with pigs' skin (Olsson et al., 1999; de Jong et al., 2000; Kuwahara et al., 2004; Lyhne et al., 2022). Under such experimental conditions, the possibility for error

introduced by environmental and behavioral factors is reduced. There are few confounding variables that may intercept a signal from the pigs' skin detected by the monitoring device. Pigs also tend to be kept in enclosures that separate them from free-ranging or semi-free-ranging pigs for the duration of the study, thereby reducing confounding factors introduced by social interactions with conspecifics as described above (Olsson et al., 1999; de Jong et al., 2000; Kuwahara et al., 2004; Lyhne et al., 2022).

CONCLUSION

The results of this study demonstrated that there is a possibility that a cost-effective infrared thermal camera can detect pulse rate at the base of pigs' ears. Once filtered, this pulse rate can be used to analyze heart rate variability in semi-free-ranging pigs at rest. Continuing this and similar explorations into affordability and portability of infrared thermal monitoring technology will advance noninvasive physiological monitoring for mammals. Expanding the use of this technology beyond detecting infection, reproductive status, and emotional valence of surface-level sympathetic nervous system activity will be beneficial for wild and captive nonhuman mammals alike. If a model such as that which this study delved into can be used reliably, a single infrared thermal camera can provide a wealth of information about animal welfare safely, easily, and while causing minimal disruption to animal subjects.

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Table 1

Pigs Included in the Pre-Processing Sample

Pig Number	Breed	Hair Color	Ear Orientation
1†	Potbelly	Black, white	Standing upward, pressed flat against head
2	Potbelly	Black	Pointing laterally at 45° angle from head
3†	Potbelly	Black	Standing upward, pressed flat against head
4	American Yorkshire	Pink	Pointing laterally at > 45° angle from head
5†	KuneKune	Black, white spots	Pointing laterally at > 45° angle from head
6†	American Yorkshire	Pink	Pointing laterally at 45° angle from head
7	Potbelly	Black	Standing upward, pressed flat against head
8†	KuneKune	Black, white spots	Pointing laterally at > 45° angle from head
9	Potbelly	Black and white	Pointing laterally at < 45° angle from head

† Pigs included in the sample yielding the most accurate potential heart rate signals.

Table 2

Criteria for Pigs Included in Initial Sample

Requirement
Does not shy away from human approach within 5 feet
Does not move away from stationary human presence within a 3 foot radius of their space
In a resting position for 2 minutes or more
Is not being physically agitated or displaced by other pigs while being approached, habituated to a stationary human presence, or being filmed
Does not display signs of physical discomfort before, during, or after filming
Does not display signs of extreme agitation in response to movements or sounds produced by other pigs

Table 3

Atmospheric Conditions Recorded for Regions of Interest

Datum	Pig	Weather/ Lighting Notes	Min. Recorded Temp. (°C)	Max. Recorded Temp. (°C)	Min. Recorded Humidity (%)	Max. Recorded Humidity (%)	Pig Rescue Temp. (°C)	Pig Rescue Humidity (%)	County Temp. (°C)	County Humidity (%)
1†	1	Sun	24.06	25.17	49	53.1	17.22	69	17.22	76
2	1	Sun	26.67	28	43.1	43.7	17.22	69	17.22	76
3	2	Clouds, sun	26	37.5	40.1	53.4	17.22	69	17.22	76
4*	3	Sun	34.22	34.67	34.5	39	17.22	69	17.22	76
5	4	Shade	23.33	23.67	48.4	56	17.22	69	17.22	76
6	4	Shade	22.78	23.06	55.8	61.8	17.22	69	17.22	76
7	4	Shade	22.06	23.28	57.8	61.8	17.22	69	17.22	76
8	5	Shade	20.28	22.06	45.5	54.1	20.56	57	20.56	57
9	5	Shade	20.28	22.06	45.5	54.1	20.56	57	20.56	57
10	5	Shade	19.5	20.06	43.5	55.7	20.56	57	20.56	57
11	6	After sunset	17.94	19.28	62.2	65.7	16.67	62	17.78	67
12	6	After sunset	18.05	20.39	62.6	68.7	16.67	62	17.78	67
13†	6	After sunset	17.78	18	68.6	70.9	16.67	62	17.78	67
14†	5	Shade	16.17	19.17	33.3	44.4	11.11	61	11.67	59
15†	5	Shade	15.28	16	47.5	47.8	11.11	61	11.67	59
16†	7	Indirect sun	21.17	26.67	28.8	30.5	11.11	61	11.67	59
17†	7	Indirect sun	21.5	24.06	33.2	33.7	11.11	61	11.67	59
18	7	Clouds	21.67	22.89	30.7	32.5	11.11	61	11.67	59
19	7	Indirect sun	25	26	29.4	30.5	14.44	50	15	48
20	7	Indirect sun	23	29.78	29.5	30.7	14.44	50	15	48
21†	8	Clouds, sun	27.56	30	77.6	85.6	21.67	90	21.67	94
22	5	Clouds, sun	27.56	30	77.6	85.6	21.67	90	21.67	94
23†	8	Clouds	29.5	31.56	69.3	77.9	21.67	90	21.67	94
24†	5	Clouds	29.5	31.56	69.3	77.9	21.67	90	21.67	94

25†	8	Clouds, rain	23.11	29.72	73.6	85.5	21.67	90	21.67	94
26	5	Clouds, rain	23.11	29.72	73.6	85.5	21.67	90	21.67	94
27†	8	Clouds	26.78	27.28	79.4	84.5	21.67	90	21.67	94
28	9	Clouds, shade	12.78	13.28	64	72.6	9.44	44	10.56	48
29	9	Clouds, shade	14.56	21	43.3	55	9.44	44	10.56	48
30	9	Clouds, shade	11.89	13.28	53.6	58.1	9.44	44	10.56	48
31	9	Clouds, shade	20.5	21.28	44.7	49.3	9.44	44	10.56	48
32	9	Clouds, shade	13.78	17.89	36.2	44.8	9.44	44	10.56	48

†These data are associated with non-interpolated instantaneous heart rate graphs deemed to show likely heart rate signals.

*Data for atmospheric conditions in the immediate vicinity of this pig were not collected. The substituted values were recorded for a thermal video of the same individual taken 7 minutes before this recording was made. Weather observations remained consistent.

Table 4

Heart Rate Variability Statistics in the Time Domain From Final Sample Data

Dataset	SDNN (ms)	RMSSD (ms)	pNN50 (%)
1	423.8334	595.6883	91.1111
2	496.5996	675.0384	91.3495
3	440.6204	628.0917	92.4837
4	414.238	583.2554	94.1538
5	395.124	527.23	90.5956
6	573.5396	781.6096	91.1032
7	487.8694	666.5527	93.4328
8	543.7564	764.2953	92.3077
9	574.0581	780.2551	91.3621
10	513.9745	702.0188	91.25
11	462.3449	646.3469	90.8537

Table 5

Descriptive Statistics for SDNN Values Calculated from Final Sample

SDNN	
Mean	484.178027
Standard Error	18.9513506
Standard Deviation	62.8545191
Sample Variance	3950.69057
Range	178.9341

Table 6

Descriptive Statistics for RMSSD Values Calculated from Final Sample

RMSSD	
Mean	668.216564
Standard Error	25.2762701
Standard Deviation	83.831904
Sample Variance	7027.78813
Range	254.3796

Table 7

Descriptive Statistics for pNN50 Values Calculated from Final Sample

pNN50	
Mean	91.8184727
Standard Error	0.34286553
Standard Deviation	1.13715631
Sample Variance	1.29312448
Range	3.5582

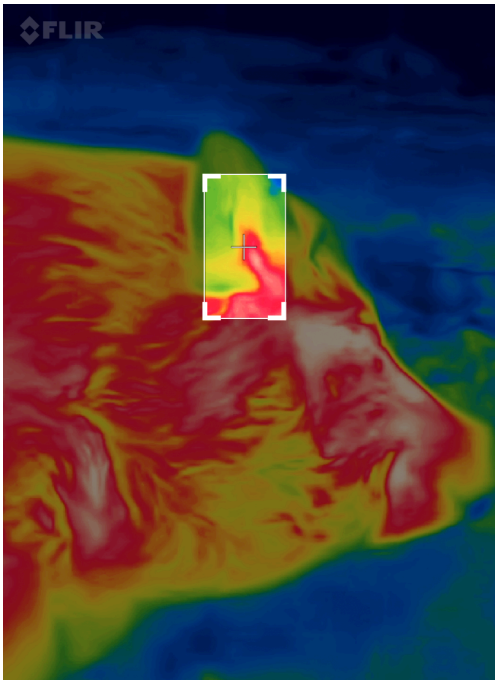

Figure 1. Selecting a region of interest from a thermal video frame in iMovie.

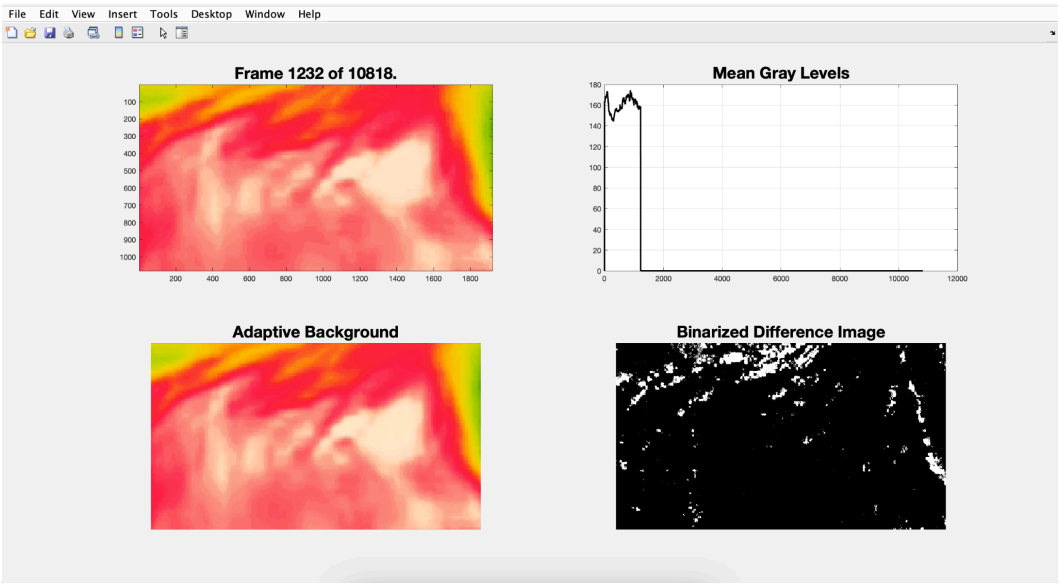

Figure 2. The GILT interface.

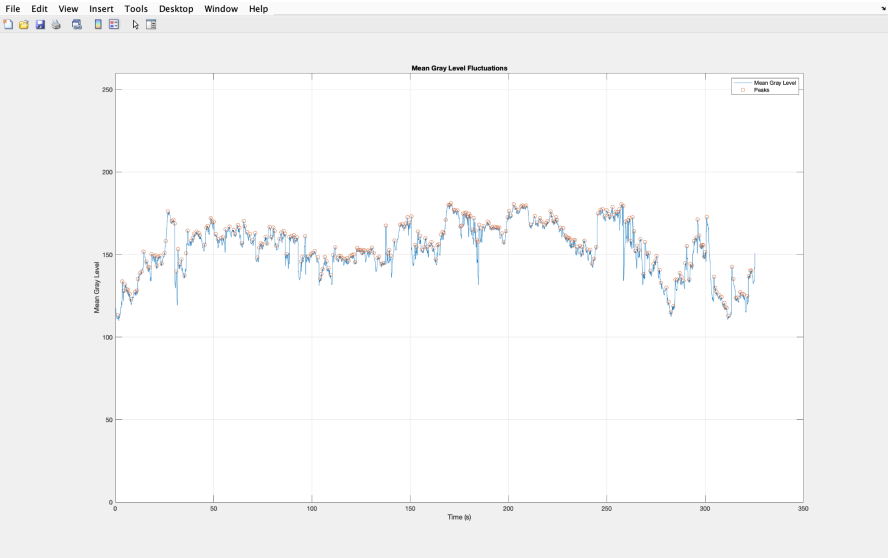

Figure 3. The PIGS graphing interface. The timestamps for each peak intensity value registered by the graphing interface are exported in an Excel file as a sequence of integers.

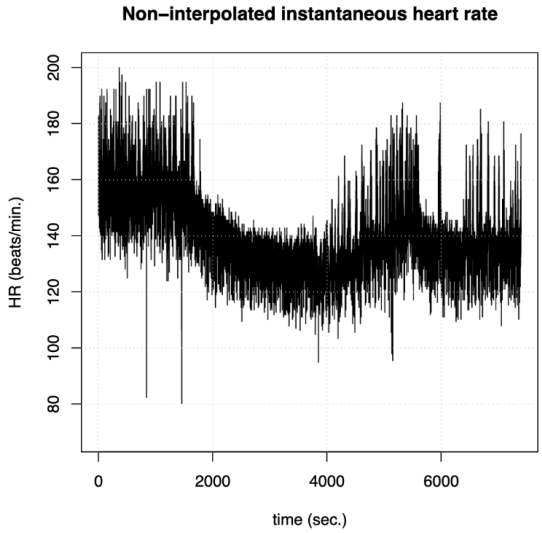

Figure 4. An example of a non-interpolated instantaneous heart rate graph from a human sampled over several hours. Figure from García et al. (2014).

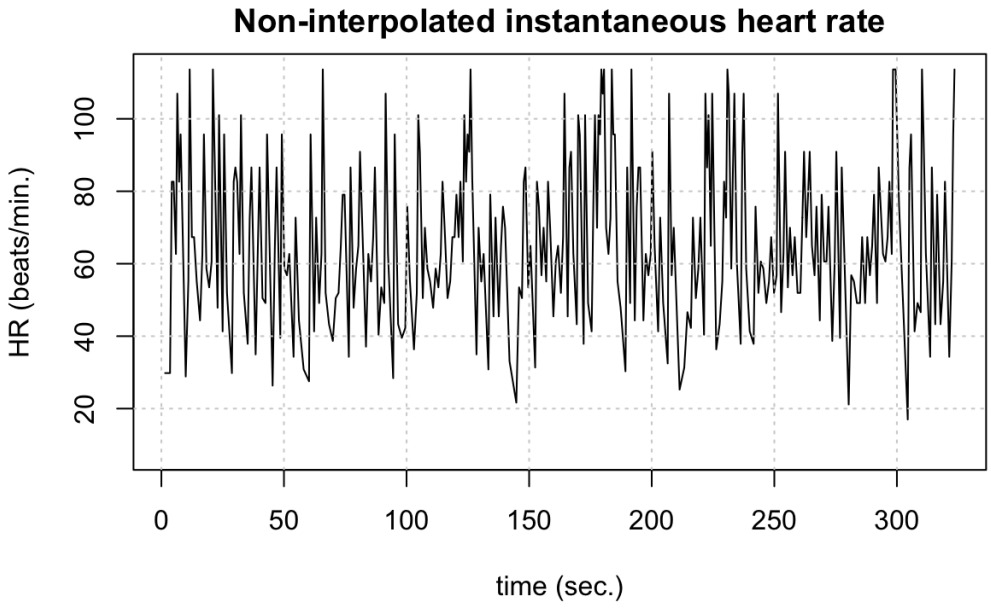

Figure 5. A non-interpolated instantaneous heart rate graph included in my dataset before assessment of probable heart rate took place. This graph represents 300 seconds of peak intensity values extracted from an infrared thermal video of the dorsal surface of a pig's ear.

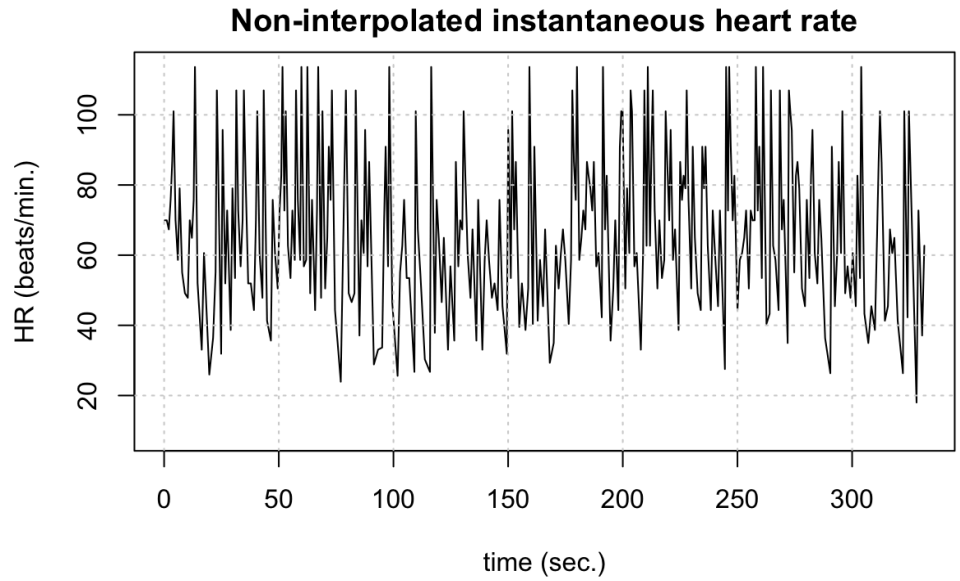

Figure 6. A heart rate signal derived from a thermal video of a pig that is consistent, precise, and clear.

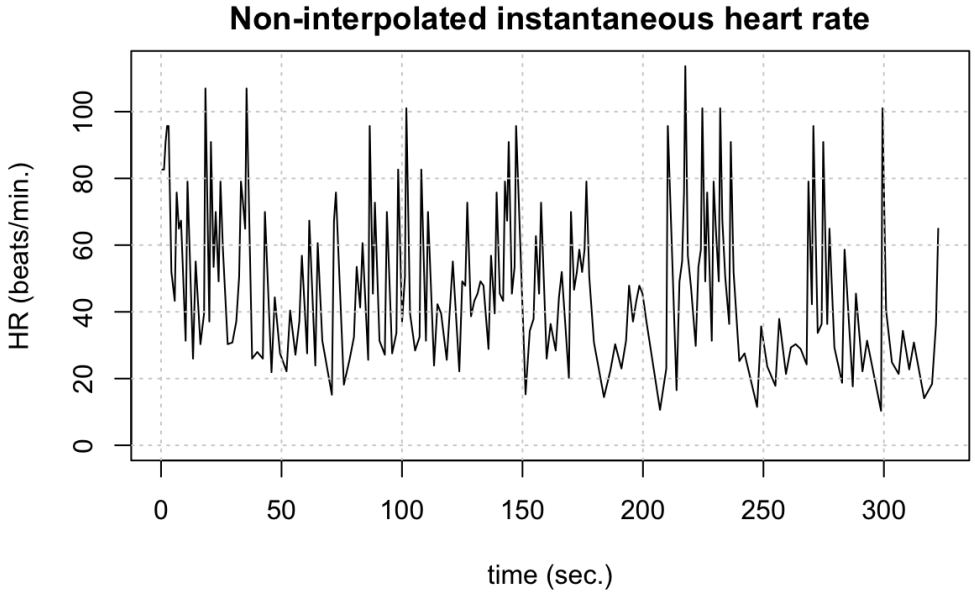

Figure 7. A heart rate signal derived from a thermal video of a pig that is neither consistent, precise, nor clear.