Quantifying the autonomic response to stressors – one way to expand the definition of "stress" in animals

Matt Gaidica[1], Ben Dantzer[1,2],

[1]Department of Psychology, University of Michigan, Ann Arbor, MI, USA

[2]Department of Ecology and Evolutionary Biology, University of Michigan, Ann Arbor, MI, USA

*Corresponding author: dantzer@umich.edu

Abstract

Quantifying the impact of changes or stimuli in the external and internal environment that are challenging (“stressors”) to whole organisms is difficult. To date, physiological ecologists and ecological physiologists have mostly used measures of glucocorticoids (GCs) to assess the impact of stressors on animals. This is of course too simplistic as Hans Seyle himself characterized the response of organisms to “noxious stimuli” using multiple physiological responses. Possible solutions include increasing the number of biomarkers to more accurately characterize the “stress state” of animal or just measuring different biomarkers to more accurately characterize the degree of acute or chronic stressors an animal is experiencing. We focus on the latter and discuss how heart rate (HR) heart rate variability (HRV) may be better predictors of the degree of activation of the sympathetic-adrenal-medullary system and complement or even replace measures of GCs as indicators of animal health, welfare, fitness, or their level of exposure to stressors. The miniaturization of biological sensor technology (“bio-sensors” or “bio-loggers”) presents an opportunity to reassess measures of stress and develop new approaches. We describe some modern approaches to gathering these HR and HRV data in free-living animals with the aim that heart dynamics will be more integrated with measures of GCs as bio-markers of stress and predictors of fitness in free-living animals.

Introduction

There is a growing appreciation that measures of “stress” are problematic. In vertebrate physiological ecology and ecological physiology, researchers have almost exclusively relied on glucocorticoids (GCs) for assessing how stressors impact animals (Romero et al. 2015; MacDougall-Shackleton et al. 2019). For example, in a review of how to measure “stress” in wildlife using measures of GCs, Sheriff et al. (2011) stated that “*Measuring GC levels does not equate to measuring ‘‘stress’’, but they are a critical component of the stress response and, when taken together with other indices of stress (e.g., measures of immune function, metabolism, nitrogen balance), they offer considerable insight into how animals perceive and adapt to their environment.*”. Measuring GCs is nonetheless useful because they seem to play a major role in facilitating organismal resilience through environmental challenges (Sapolsky et al., 2000; Vitousek et al., 2018). However, this is problematic because of its simplicity as the stress response is multifaceted, composed of autonomic, neuroendocrine, and behavioral responses. In fact, glucocorticoids are not necessarily symptomatic of stress, nor do they reliably predict individual animal fitness in nature (Breuner 2008; Bonier et al. 2009; Crespi et al. 2013).

Other than altering our terminology and verbiage so that we are more careful to not equate “an increase in GCs” with “an increase in stress levels”, where do we go from here? One solution is to make our measures of “stress” more multifaceted than they have been in the past. Interestingly, this is similar to what Seyle (Selye 1936; Selye 1943; Selye 1956) described when he characterized the stress response as the "general adaptation syndrome" or "general alarm reaction". Seyle measured not just one feature of the stress response but the "syndrome" that was composed of multiple physiological responses and endpoints indicative of exposure to noxious stimuli. The suggestion to expand how we quantify the stress response has been called for recently. For example, Breuner et al. (2013) highlighted the need for other metrics to quantify exposure to chronic stress such as glucose or free-fatty acid levels or the production of heat shock proteins. Dickens and Romero (2013) emphasized that there is not one single physiological variable that one could use to characterize individuals as being "under chronic stress". Recent studies (e.g., Romero et al. (2009)) have also endeavored to quantify the multitude of effects stressors have on behavior, the hypothalamic-pituitary-adrenal (HPA) axis, and the sympathetic-adrenal-medullary system.

Those of us that study free-living animals have been historically constrained by the field environment to the extent that concurrent toolsets are difficult to implement. Thus, the reliance on GCs as an accessible and proximate measure of stress has become prominent, though flawed. The miniaturization of biological sensor technology ("bio-sensors" or "bio-loggers") presents an opportunity to reassess measures of stress and develop new approaches—potentially animal-borne—that can be united with the vast work on measures of GCs in free-living animals. One such route of investigation focuses on the brain-heart axis as they are bi-directionally connected to the sympathetic-adrenal-medullary system. Here, the electrical activity of the central nervous system modulates the catecholaminergic tone onto the heart, causing changes in heart rate (HR) and heart rate variability (HRV, Fig. 1). This link between the brain and the rest of the body is potentially significant with respect to stress. We aim to describe the physiological mechanisms involved in the sympathetic stress response, their significance in animal physiological ecology and ecological physiology, and present modern approaches to gathering these data in free-living animals. In doing so, we hope to present a balanced comparison, and perhaps, integration of heart dynamics with GCs as bio-markers of stress and predictors of fitness in free-living animals.

The Autonomic Stress Response

The common caricature of the immediate response to adversity or environmental challenges (“fight or flight”) is accompanied by a cascade of physiological changes. In mammals, the limbic system is highly conserved and serves as a neural substrate for fears and emotions (Jänig 1985), controlling sympathetic outflow systems to eventually activate the cardiac muscle and adrenal glands (Porges 1995; Chapleau and Abboud; Jansen et al. 1995). This is how and why GCs remain a valid surrogate measure for stress, and although the effect of GCs on the body are relatively slow, they play an important role in responding and adapting by regulating glucose production and temporarily suppressing the immune system (Padgett and Glaser 2003).

More immediate are the effects catecholamines, namely, epinephrine and norepinephrine, which agonize ß1-receptors on the heart enhancing contractility while increasing heart rate (Cyr and Romero 2009; Lacombe and Jones 1990). Meanwhile, the parasympathetic pathway (i.e. “rest and digest”) is inhibited, which is why the stress response is said to be “sympathetic-dominant”. The heart also naturally accelerates during inhalation due to the inhibition of vagal outflow, which is restored via the release of acetylcholine following exhalation (Eckberg and Eckberg 1982).

Prolonged activation of the sympathetic system can be due to emotional and neural dysregulation, environmental uncertainty, consistent threats, lingering noxious stimuli, or irregular breathing, and should ultimately be detrimental and maladaptive. For example, one of the best cardiac risk factors in humans remains an elevated heart rate (greater than 90 beats per minute) due to sympathetic over-activation (Zhang et al. 2016; Curtis and O'Keefe Jr 2002) and similar, negative implications of an elevated heart rate has been found in other animals (Umana et al. 2003). ß-blockers are competitive antagonists that compete with endogenous catecholamines on ß-receptors and can reduce heart rate and normalize blood pressure (Amer 1977). Importantly, significant positive outcomes relating to cardiac events and mortality from the use of ß-blockers suggest heart dynamics as the crucial physiological measure (Arnold et al. 2008) rather than systemic catecholamines.

Heart Rate

HR has historical significance as one of the best measures to assess the health and behavioral status across taxa (Levine 1997). HR is often used to characterize the autonomic response to stress as it can reflect the balance between the sympathetic and parasympathetic systems that elevate and depress heart rate, respectively. The autonomic influence over HR has been directly tested by co-administering a betablocker (propranolol) and anticholinergic (atropine), therefore exposing the spontaneous HR generated intrinsically by the sinoatrial node (Jose and Collison 1970). Somewhat surprisingly, the heart beats faster when the autonomic inputs are blocked in this fashion, suggesting that the parasympathetic branch, which depresses heart rate through the vagal nerve, is normally dominating. In contrast to sympathetic activation, these findings have led researchers to alternatively focus on vagal "tone" as a health indicator (Levy and Schwartz 1994).

Acute stressors (e.g., an immediate, novel, or unpredictable stimulus) seem to affect humans, and free-living birds and mammals in similar ways, although direct comparisons are difficult and often necessitate captivity. In wild birds, anthropogenic disturbances such as exposure to humans can elevate heart rates (Viblanc et al. 2015; Viblanc et al. 2012a) as can exposure to agonistic interactions between neighbors (Viblanc et al. 2012b). Black bears have a significant increase in HR associated with the perceived threat of road crossings (Ditmer et al. 2018). Similar elevations in HR are found in captive birds and mammals where restraint or noxious stimuli are presented (Nephew et al. 2003; Ellen et al. 2014). The startle response is also associated with an immediate elevation in HR following the disturbance (Young and Leaton 1994; Nephew and Romero 2003; Johnson and Mayers 2001; Laferton et al. 2018).

How chronic stress impacts HR is less clear. For example, 4 weeks of exposure to a chronic stress paradigm causes sustained elevations in baseline HR in laboratory rats (Grippo et al. 2003). In female prairie voles (*Microtus ochrogaster*), social isolation is a type of chronic stressor and voles experiencing 4 weeks of social isolation exhibited substantial increases in resting HR (Grippo et al. 2007; Grippo and Johnson 2009). In captive birds, baseline HR is initially elevated for the first 30 hours but declines to control levels after 10-14 months (Dickens and Romero 2009) Fischer and Romero (2016). Interestingly, the daytime increase in HR during chronic stress exposure (15-16 days) was met with a nighttime decrease during that same period (Romero et al. 2009). However, this counterbalancing effect to a chronic stressor (wounding) is abolished if the birds are moulting (Kostelanetz et al. 2009), which in itself may be a stressful life history stage in birds as it is often associated with elevated GCs.

Heart rhythm is still one of the first vital signs examined by a physician, but as we expect with other species, it is not interpreted without greater context (e.g., did the patient drink a cup of coffee in the waiting room?). Even then, the prognostic quality of HR itself is tenuous in clinical medicine. In humans, a typical standard deviation of heart rate can be up to 10 beats per minute, which is not itself significantly different from some disease conditions (Albanese et al. 2016). HR associations with chronic conditions like depression are mixed, sometimes showing higher resting HR (Krittayaphong et al. 1997) and sometimes lower (Hu et al. 2016). Conditions such as depression are often accompanied by a sedentary lifestyle and metabolic syndrome (Licht et al. 2011; Thayer et al. 2010), which taken together represent a unique, maladaptive condition that is, of course, rare in wild animals. Socioemotional conditions like anxiety are not met with the same physiological adaptations as physical stressors (Watkins et al. 1998), although primates may have specially adapted neural machinery for these situations (Cameron and Schoenfeld 2018). The problem with using HR as a window into the stress state of an animal is challenging from a taxonomic, environmental, circadian, and life history perspective, as they are all factors that affect HR and HR-associated adaptations (Viblanc et al. 2015). Therefore, a cardiac measure that provides more resolution into the underlying physiology is required and may be found in HRV.

Heart Rate Variability

In 1965 it was found that the inter-beat interval of the heart was an earlier predictor of fetal distress than HR itself (Hon and Lee 1963). Overlapping with the rise of accessible computing power, statistical measures of HRV were soon pioneered (Akselrod et al. 1981). By the late 1980s HRV gained clinical relevance as a detector of autonomic neuropathy in diabetic patients (Ewing et al. 1985) and as a strong predictor of mortality following an acute myocardial infarction (Wolf et al. 1978; Bigger Jr et al. 1992; Malik et al. 1989; Kleiger et al. 1987). Since then, there has been a relatively lengthy body of research showing that HRV is an accurate measurement of the activity of the autonomic stress response (Thayer et al. 2012) and signifies a state of heightened vigilance (Thayer and Lane 2000). A reduction in HRV (i.e., a more regular heartbeat) is a result of vagal withdrawal and sympathetic activation (Schiweck et al. 2019) characteristic of exposure to stressors (Stauss 2003; von Borell et al. 2007; Cyr and Romero 2009; Koolhaas et al. 1999; Perini and Veicsteinas 2003), which can result in unfavorable health outcomes. The regularity between heartbeats under sympathetic dominance is likely advantageous for survival, as it guarantees a consistent blood circulation and delivery of nutrients and glucose to peripheral organs. However, maintaining such a mode of operation may be biophysically maladaptive, as it can render the organism impervious to changing circumstances (Thayer and Sternberg 2006). An autonomic blockade not only increases HR but decreases HRV in humans (Camm et al. 1996) and rodents (e.g., Lakin et al. 2018, Cyr et al. 2008) highlighting the important influence of vagal tone on regulating HRV.

Across different types of species, exposure to stress is associated with a reduction in HRV. For example, transportation or acute restraint (e.g., during grooming) of agricultural animals is associated with a reduction in their HRV (Schmidt et al. 2010; Reefmann et al. 2009). Lameness in cows (characterized as abnormalities of the feet that cause pain when moving and may lead to infection and sepsis) is a type of chronic stressor and cows that exhibit lameness had lower HRV than those that did not exhibit these symptoms (Kovacs et al., 2015). In laboratory rats, 4 weeks of exposure to a chronic stress paradigm resulted in reductions in HRV (Grippo et al. 2003). Female prairie voles exposed to social isolation for 4 weeks exhibited reductions in HRV (Grippo et al. 2007; Grippo and Johnson 2009). It is less clear how stress and HRV are related in non-mammals although many findings are consistent (Fischer and Romero 2016; Müller et al. 2017). Acute stress (trauma associated with surgery) in snakes almost eliminates HRV for the first 10 days following surgery (Sanches et al. 2019). In contrast, studies in captive birds exposed to a chronic stress paradigm for 16 or 18 d, HRV was unaffected (Cyr and Romero 2009; Kostelanetz et al. 2009).

The causes and consequences of variation in HRV have been best investigated in humans. In humans, HRV decreases with age (Reardon and Malik 1996; Padgett and Glaser 2003) and many pathophysiological conditions including heart failure, diabetes, and hypertension (Xhyheri et al. 2012) as well as obesity (Mazurak et al. 2016). It is unclear how mental stress manifests in non-humans, however patients with depression have a lower HRV (Krittayaphong et al. 1997) which is unrelated to existing cardiovascular disease (Carney and Freedland 2009) and worsening symptoms further decrease HRV (Kemp et al. 2010; Krittayaphong et al. 1997). HRV is negatively correlated with exposure to stressful experiences (Porges 2003; Stauss 2003; von Borell et al. 2007) and self-reported anxiety (Berntson and Cacioppo 2004) or increased work-related stress (Chandola et al. 2008; Thayer et al. 2010). Experimental application of standardized psychological stress tests (e.g., Stroop or speech task paradigm) to humans also decreases their HRV during wakefulness (Delaney and Brodie 2000) and subsequently during sleep (Hall et al. 2004).

HRV may also be a window into the reactiveness and integration capacity of the central nervous system (CNS) to deal with challenges and coordinate context-specific responses in the periphery (Thayer et al. 2012). However, the central site responsible for these adaptations has been challenged by the fact that the sinoatrial node is also plastic (Stein et al. 2002). The finding that adaptations to HRV in response to physical exercise are abolished during an autonomic block (e.g., propranolol hydrochloride and atropine) support the former hypothesis, that the CNS modulates HRV through the parasympathetic pathway (Lakin et al. 2018).

Compared to individuals with high resting HRV, those with low resting HRV do not recover as quickly from psychological stressors based on cardiovascular, endocrine, and immune markers (Weber et al. 2010). One meta-analysis showed that poor recovery following laboratory stressors is associated with cardiovascular risk status (e.g., elevated blood pressure, hypertension, clinical cardiac events), although subjects also exhibited heightened reactivity (Chida and Steptoe 2010). Indeed, biological responses to stressors or threats can be exaggerated, leading to anxiety or aggression (Valiente et al. 2003; Carthy et al. 2010). For example, studies on severely depressed individuals are mixed, albeit consistently atypical, showing both higher reactivity or a blunted response to stressors (Hamilton and Alloy 2016; Schiweck et al. 2019) similar to the inverted U performance-arousal curve of the Yerkes-Dodson Law (Yerkes and Dodson 1908; Cohen 2011). Physical exercise is one way to decrease resting HRV while establishing normal/optimal autonomic reactivity (Kiss et al. 2016). Unlike psychological stressors, transient activation of the autonomic system from exercise is followed by an augmentation of vagal tone (Pardo et al. 2000).

Associations between GCs & HRV

Given that most studies to date have focused on measuring “stress” using only GCs, it is useful to briefly look at the associations between GCs and HRV. Some studies that employ captivity as a chronic stressor show that the period following initial captivity is associated with an increase in GCs and HR and a decline in HRV but as the time from initial captivity increases, GCs and HR decline and HRV increases (Dickens and Romero 2009). Following the transportation of agricultural animals (a type of acute stressor), GCs are elevated and HRV is reduced (Schmidt et al. 2010). However, other studies find no association between HRV and measures of GCs or even the opposite association where both GCs and HRV are elevated. For example, lameness in cows (a type of chronic stressor) was associated with reduced HRV but no change in fecal glucocorticoid metabolites compared to non-lame cows (Pacifici et al. 2015). In wild birds brought into captivity, HRV and plasma GCs were reduced at the beginning of captivity compared to 6-7 days after captivity was initiated (Fischer et al. 2016; Fischer et al. 2018). These latter studies suggest that the lag time from the initiation of the stressor (in these studies it was captivity) affects their impact on HRV or GCs; HRV is low immediately after the start of captivity and then increases whereas GCs are low immediately after captivity and then increase.

In humans, HRV does not directly correlate with the cortisol waking response (Stalder et al. 2011) highlighting the potential nuance of HRV. Interpreting HRV may benefit from the context of the three-stage response model originally proposed by Selye (Selye 1956). That is, HRV measured during the alarm, resistance, and exhaustion stages of the stress response should be assumed to communicate different information about the state of an animal.

Tools and Methods to Measure HR and HRV

As a relatively young field of investigation, the tools and methods used to record HR and HRV are actively undergoing standardization, which may account for conflicting results. An electrocardiogram (ECG, sometimes called an EKG from the German word *Elektro-kardiographie*) is the gold standard method for measuring heart rhythms and requires an amplifier and electrodes to be strategically placed near the heart muscle. Ample resolution on an ECG will provide information about atrial and ventricular depolarization and repolarization. Ventricular depolarization is the largest deflection in the signal and can be identified as the R-wave in the QRS-wave complex. The inter-beat interval (IBI) is synonymous with the normal-to-normal R-R interval (NN) and is the basis for calculating time-series measures like mean NN interval or HR, as well as statistical operations that are used in HRV analyses.

HR and HRV measures can be affected by a subject's head or body position, respiration rate or pace, sex, age, and aerobic fitness level, and interrelate with natural biophysical rhythms (e.g., circadian, metabolic, hormonal). Clinicians and researchers alike should be aware of the relatively extensive list of best practices and caveats when approaching HR and HRV measurements (Shaffer and Ginsberg 2017; Camm et al. 1996). Here, we focus our discussion on the potential utility of HRV analyses based on "short-term" 5-minute recordings, as this is a well-documented standard, reasonable to achieve in animals from battery-powered devices, and applicable to 24-hour recordings that are chunked into smaller time windows.

Common time-series HRV measures include the standard deviation of all NN intervals (SDNN, measured in ms), the root mean square of successive differences of the NN interval (RMSSD), and the percentage of adjacent NN intervals that differ by more than 50 ms (pNN50) (Camm et al. 1996). SDNN measurements reflect both sympathetic and parasympathetic activity, but in short-term recordings, the primary source of variation is parasympathetically-mediated respiratory sinus arrhythmia, making this measure extremely sensitive to respiratory status (Shaffer et al. 2014). RMSSD and pNN50 are correlated with each other and closely with parasympathetic activity. Although time-series analyses are conceptually straight forward, they fail to correlate with the same measures over 24 hours (Shaffer and Ginsberg 2017), making their interpretation context-dependent. Studies have also found that short-term recordings are prognostically insufficient (Kleiger et al. 2005).

Analyses in the frequency-domain may offer more insight for short-term recordings. This often begins by subjecting the time-series data (i.e., the entire 5 minutes) to a form of spectral analysis where the power contributions from different frequency bands can be viewed in two dimensions (power × frequency). A generalized approach has been to quantify low-frequency (LF, 0.04 – 0.15 Hz) and high-frequency (HF, 0.15 – 0.4 Hz) power which are correlated with sympathetic and parasympathetic tone, respectively (Xhyheri et al. 2012). Therefore, the LF/HF ratio has been suggested as an index of the interaction between sympathetic and vagal activity (Pagani et al. 1986), but this notion has been challenged, primarily because the LF band is multifaceted. For example, during resting conditions LF power represents baroreflex rhythms, and may only approximate sympathetic tone when subjects are ambulating (Shaffer and Ginsberg 2017).

Bio-loggers to Record HR and HRV

In recent years, the use of miniaturized bio-loggers to make physiological and environmental measurements from free-ranging animals has radically transformed scientific capabilities. Technological strides in battery, computation, memory, and sensor technology continue to support a rich suite of bio-logging tools that are not only becoming smaller and longer-lasting, but providing multi-featured, high-resolution data (Williams et al. 2019).

Natural stressors imposed by natural environments may better approximate the physiological capabilities of an animal to respond and adapt than stressors applied in the laboratory (Williams et al. 2016). However, being multifaceted, accurately measuring the stress response using an attachable, or implantable bio-logger is challenging. Some of the first attempts in this vein used accelerometers to identify behavioral patterns in response to stressors (Kröschel et al. 2017). Accelerometry has also been used to map micro-movements onto specific internal (Wilson et al. 2008) or disease states (Downey et al. 2017; Cancela et al. 2014). The dynamic relationship between HR and respiration has been characterized using bio-loggers in diving birds (Butler and Woakes 1979) and again examined in penguins with a device capable of identifying unique body functions such as defecation (Wilson et al. 2004). Self-contained, automated hemodynamic measurement units have been used to sample blood during a physical challenge to analyze changes in GCs and other hormones (Takei et al. 2016; Landry et al. 2014).

Cardiac rhythms have been a central focus of bio-logging as HR accurately predicts energy expenditure (Weimerskirch et al. 2002) and metabolic rate (Green 2011), and both HR and HRV are becoming widely appreciated surrogates of stress and resilience (see above). Some bio-loggers perform on-board HR estimation (Pulopulos et al. 2018; Bevan et al. 1997; Chaise et al. 2017) which is more efficient from a power and memory standpoint, but limits HRV analyses. Beat-to-beat signals are useful for time-series analyses, as the periodically measured RMSSD of inter-beat intervals in free-ranging, pregnant horses correlate with changing seasons, which may be due to environmental, pregnancy, or metabolic pressures (Pohlin et al. 2017). However, to perform proper short-term HRV spectral analyses the entire, raw waveform must be analyzed, and to our knowledge has so-far relied on post hoc, rather than on-board computation in battery-powered bio-loggers (although this is not true for consumer ‘wearables’ designed for humans). For example, bar-headed geese have been fitted with bio-loggers that record long-duration, raw ECG patterns during a trans-Himalayan migration (Spivey and Bishop 2014).

It should be recognized that bio-loggers have additional constraints compared to biotelemetry systems, where data is or transmitted rather than directly saved (Fu et al. 2011), and wireless charging might be an option (Young-Ho et al. 2004 Nov). Telemetered heart rhythms have been applied to mammals (Arnold et al. 2004; O'Mara et al. 2017), birds (Cyr et al. 2008), fish (Cooke et al. 2004), and reptiles (Butler et al. 2002). Telemetry systems have outpaced bio-loggers in both capability and use across model systems, although there are notable synergies, and indeed mixed capabilities, as in the form of marine “pop-up” tags (Musyl et al. 2011) and RFID-enabled devices (Williams et al. 2016). Bio-loggers that can record neural data with the resolution to identify single action potentials (> 20 kHz) have been implemented in behaving animals with the option to perform short-term experiments free of a recording tether (Massot et al. 2019). Brain rhythms that are present during sleep can be identified through much slower sampling rates (Aulsebrook et al. 2016; Rattenborg et al. 2008), thereby extending the lifetime and utility of neuro-based bio-loggers. The discovery that great frigatebirds (*Fregrata minor*) sleep mid-flight used bio-loggers that constantly recorded electroencephalography (at 200 Hz) over 10 days (Rattenborg et al. 2016). Similar devices have been implemented in pigeons and represent a powerful toolset when paired with other onboard sensors (Vyssotski et al. 2006). Indications that sleep states affect physical performance and recovery (Shapiro et al. 1981) and that stress has a bidirectional relationship with sleep (Hall et al. 2004; Martire et al. 2019) makes the neural toolset an exciting new direction for bio-logging technology. HR/HRV may be a key marker for the efficiency and effectiveness of autonomic regulatory processes like sleep and can, therefore, be examined in association with physical and reproductive fitness.

The future of bio-logging to quantify how animals respond and recover from exposure to stressors may rely on smarter and more clever recording techniques to mitigate power and memory constraints (Woakes et al. 1995; Clark et al. 2009; Spivey and Bishop 2014; Cox et al. 2018). For example, accelerometry and time-of-day data could augment ECG recording routines, as some cardiac measurements are only relevant following a period of rest or in the morning (Shaffer and Ginsberg 2017). Brain rhythm data not only pairs well with accelerometry data for the ability to distinguish between active, rest, and sleep state but could also coordinate low-power modes on the bio-logger so that the neural recording circuitry is idle when the animal is moving. Although onboard computation is power-intensive, algorithms that perform HRV statistics (Park et al. 2018), detect sleep states (Allocca et al. 2019), extract neuronal spiking rates (Dragas et al. 2013), or discretize any other physiologic variables could be valuable depending on the experimental recording strategy.

The largest challenge to monitoring HR and HRV in free-living animals to obtain an index of their exposure to stress is that it can be more invasive than the collecting of blood samples and other tissues that can be used to measure GCs non-invasively (feces, urine, feathers, hair). However, they may provide much higher quality and informative data than measures of GCs alone (e.g., Aimie-Salleh et al. (2019)). Researchers must also consider whether intervening on either end of an experiment to implant and explant a bio-logger is altogether less disruptive to the population and other species in the community than the consistent or frequent presence of humans, which is often required to collect samples to measure GCs.

Conclusions & Why HRV?

HRV appears to occur in all vertebrate taxa (sensu Sanches et al. 2019) and there are several similarities to studies that use measures of GCs to measure how an animal perceives its world and the degree of environmental challenges it is facing. For example, GCs (Schoenle et al. 2018) and HRV (Muller et al. 2018) both show repeatable individual differences, suggesting that these traits can exhibit an evolutionary response to natural selection. Although there are challenges associated with measuring HRV compare to GCs, we think there are several clear advantages. First, GCs play a role in the mobilization of glucose to fuel behavioral activities and so elevated GCs may be a biomarker of elevated energetic expenditure in wild animals. However, by measuring HRV, measures of HR are also available. It is quite likely that measures of HR better reflect actual energetic expenditure than measures of GCs given that they can be used to estimate oxygen consumption and therefore energetic expenditure (Groscolas et al. 2010; Ellenberg et al. 2013), although they require careful validation (e.g., Hicks et al. (2017)). Second, measures of GCs have been used to investigate how anthropogenic activities impact wildlife (Dantzer et al., 2014) but measures of HR and perhaps HRV in wild animals may provide a more in-depth view of how they affect wildlife. For example, wild animals may not exhibit a behavioral response to anthropogenic activities but still exhibit an increase in HR (Ditmer et al. 2015). Measures of HR may also provide insights into the unexpected impacts of humans on animals such as the presence of wildlife photographers provoking a stronger increase in HR in penguins than did the capture and handling (Ellenberg et al. 2013). Finally, as we noted above, HRV may better reflect the stress state of an animal as well as the amount of acute or chronic stressors the animal has been exposed to compared to measures of GCs. This is reflected by the ability of HRV to predict human health (Thayer and Lane 2000; Lane et al. 2009) and health/welfare in other animals (von Borell et al. 2007). For example, there is a strong interest in understanding how social interactions affect the health and fitness of all animals including humans (“sociality-health-fitness” nexus). Interestingly, social interactions may increase oxytocin (Uvnas-Moberg 1998) and experimental administration of oxytocin increases HRV (Romero et al. 2014). Moreover, social isolation in prairie voles reduces HRV but these effects are abolished if oxytocin is administered during the period of isolation (Grippo and Johnson 2009). These studies suggest one way by which social interactions increase health, well-being, and fitness in animals through its effects on oxytocin and HRV.

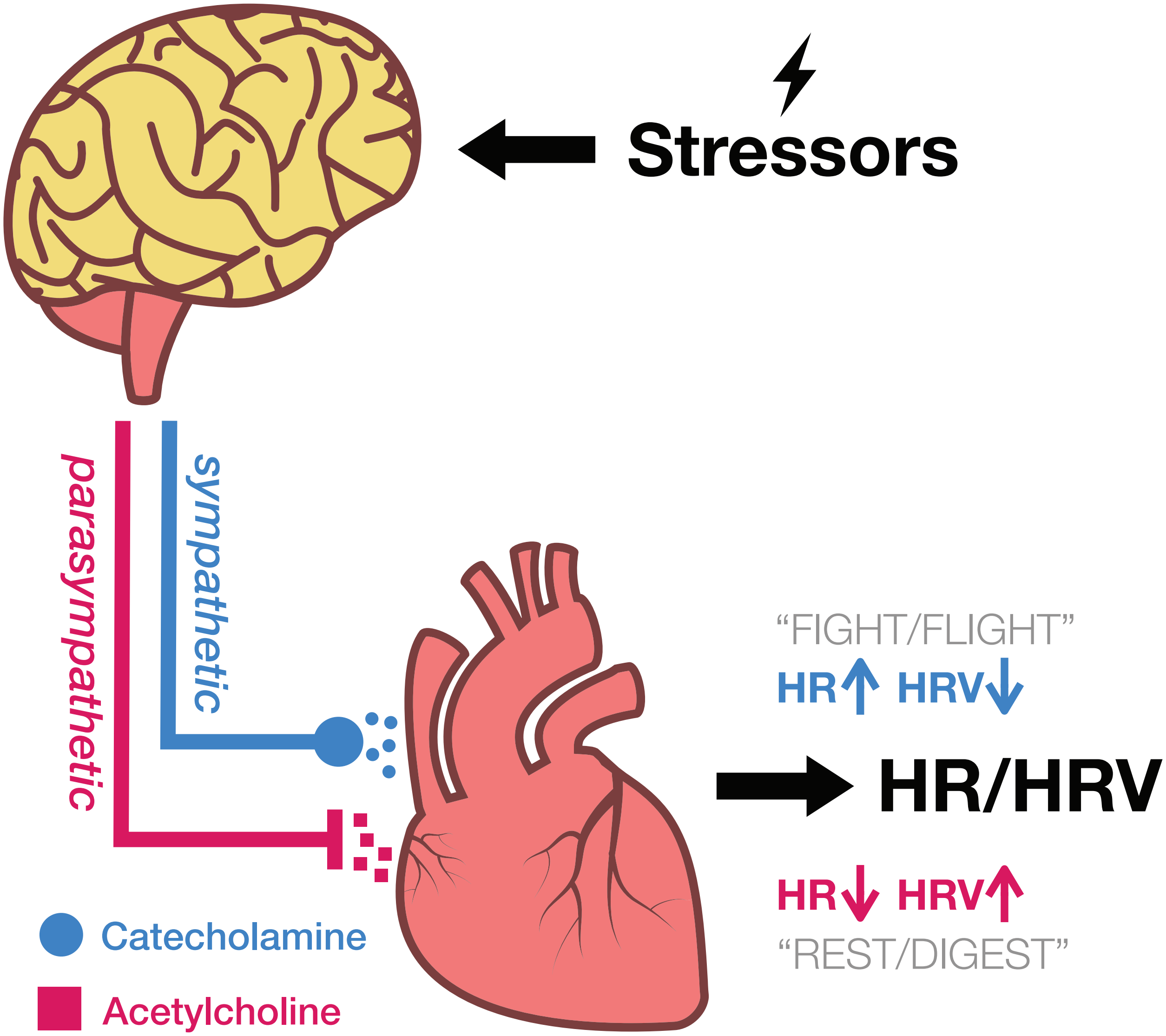
Studies of the relationship between HRV and measures of fitness in wild animals are clearly needed here but these previous studies suggest that HRV may more reliably predict past or current exposure to stress and may more reliably predict fitness. The obvious limitation here is that measuring HR and HRV is still difficult and much more invasive to the individual compared with measures of GCs. However, a comprehensive picture of the stress response to environmental challenges, as Seyle advocated, is going to require data on the autonomic stress response in addition to measures of GCs and HRV may be the most reliable biomarker.

Acknowledgements

Thank you to Elizabeth Addis for inviting us to contribute this manuscript. BD was funded by NSF IOS-1749627.

Figures

Fig 1. Stressors modulate cardiac rhythms through descending autonomic pathways. Sympathetic branch activity is indicative of exposure to stressors and causes an increase in heart rate (HR) and decrease in heart rate variability (HRV), while parasympathetic branch activity decreases HR and increases HRV.



Literature Cited

Aimie-Salleh, N, Malarvili, MB et al. (2019) Fusion of heart rate variability and salivary cortisol for stress response identification based on adverse childhood experience. Med Biol Eng Comput 57:1229–1245.

Akselrod, S, Gordon, D et al. (1981) Power spectrum analysis of heart rate fluctuation: a quantitative probe of beat-to-beat cardiovascular control. American Association for the Advancement of Science,

Albanese, M, Neofytou, M et al. (2016) Evaluation of heart rate measurements in clinical studies: a prospective cohort study in patients with heart disease. Eur J Clin Pharmacol 72:789–795.

Allocca, G, Ma, S et al. (2019) Validation of ‘Somnivore’, a Machine Learning Algorithm for Automated Scoring and Analysis of Polysomnography Data. Front Neurosci 13:207.

Amer, MS (1977) Mechanism of action of beta-blockers in hypertension. Biochemical pharmacology 26:171–175.

Arnold, JM, Fitchett, DH et al. (2008) Resting heart rate: a modifiable prognostic indicator of cardiovascular risk and outcomes. Can J Cardiol 24 Suppl A:3A–8A.

Arnold, W, Ruf, T et al. (2004) Nocturnal hypometabolism as an overwintering strategy of red deer (Cervus elaphus). American Journal of Physiology-Regulatory, Integrative and Comparative Physiology 286:R174–R181.

Aulsebrook, AE, Jones, TM et al. (2016) Sleep Ecophysiology: Integrating Neuroscience and Ecology. Trends Ecol Evol 31:590–599.

Berntson, GG, Cacioppo, JT (2004) Heart rate variability: Stress and psychiatric conditions. Dynamic electrocardiography 57–64.

Bevan, Boyd et al. (1997) Heart rates and abdominal temperatures of free-ranging South Georgian shags, Phalacrocorax georgianus. J Exp Biol 200:661–675.

Bigger Jr, JT, Fleiss, JL et al. (1992) Frequency domain measures of heart period variability and mortality after myocardial infarction. Circulation 85:164–171.

Bonier, F, Martin, PR et al. (2009) Do baseline glucocorticoids predict fitness? Trends in ecology \& Evolution 24:634–642.

Breuner, C (2008) Maternal stress, glucocorticoids, and the maternal/fetal match hypothesis. Hormones and Behavior 54:485.

Breuner, CW, Delehanty, B et al. (2013) Evaluating stress in natural populations of vertebrates: total CORT is not good enough. Functional Ecology 27:24–36.

Butler, PJ, Woakes, AJ (1979) Changes in Heart Rate and Respiratory Frequency During Natural Behaviour of Ducks, with Particular Reference to Diving. The Company of Biologists Ltd,

Butler, PJ, Frappell, PB et al. (2002) The relationship between heart rate and rate of oxygen consumption in Galapagos marine iguanas (Amblyrhynchus cristatus) at two different temperatures. Journal of Experimental Biology 205:1917–1924.

Cameron, HA, Schoenfeld, TJ (2018) Behavioral and structural adaptations to stress. Frontiers in neuroendocrinology 49:106–113.

Camm, AJ, Malik, M et al. (1996) Heart rate variability: standards of measurement, physiological interpretation and clinical use. Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology.

Cancela, J, Pastorino, M et al. (2014) Feasibility study of a wearable system based on a wireless body area network for gait assessment in Parkinson's disease patients. Sensors 14:4618–4633.

Carney, RM, Freedland, KE (2009) Depression and heart rate variability in patients with coronary heart disease. Cleveland Clinic journal of medicine 76:S13.

Carthy, T, Horesh, N et al. (2010) Emotional reactivity and cognitive regulation in anxious children. Behaviour research and therapy 48:384–393.

Chaise, LL, Paterson, W et al. (2017) Implantation of subcutaneous heart rate data loggers in southern elephant seals (Mirounga leonina). Polar Biology 40:2307–2312.

Chandola, T, Britton, A et al. (2008) Work stress and coronary heart disease: what are the mechanisms? European heart journal 29:640–648.

Chapleau, M, Abboud, F Neuro-Cardiovascular Regulation: From Molecules to Man. Annals New York Academic of Sciences

Chida, Y, Steptoe, A (2010) Greater cardiovascular responses to laboratory mental stress are associated with poor subsequent cardiovascular risk status: a meta-analysis of prospective evidence. Hypertension 55:1026–1032.

Clark, TD, Hinch, SG et al. (2009) Sex differences in circulatory oxygen transport parameters of sockeye salmon (Oncorhynchus nerka) on the spawning ground. Journal of Comparative Physiology B 179:663–671.

Cohen, RA (2011) Yerkes–Dodson Law. Encyclopedia of clinical neuropsychology 2737–2738.

Cooke, SJ, Thorstad, EB et al. (2004) Activity and energetics of free-swimming fish: insights from electromyogram telemetry. Fish and Fisheries 5:21–52.

Cox, SL, Orgeret, F et al. (2018) Processing of acceleration and dive data on-board satellite relay tags to investigate diving and foraging behaviour in free-ranging marine predators. Methods Ecol Evol 9:64–77.

Crespi, EJ, Williams, TD et al. (2013) Life history and the ecology of stress: how do glucocorticoid hormones influence life-history variation in animals? Functional Ecology 27:93–106.

Curtis, BM, O’Keefe Jr, JH (2002) Autonomic tone as a cardiovascular risk factor: the dangers of chronic fight or flight. Mayo Clinic Proceedings 77:45–54.

Cyr, NE, Dickens, MJ et al. (2008) Heart rate and heart-rate variability responses to acute and chronic stress in a wild-caught passerine bird. Physiological and Biochemical Zoology 82:332–344.

Cyr, NE, Romero, LM (2009) Identifying hormonal habituation in field studies of stress. General and comparative endocrinology 161:295–303.

Delaney, JPA, Brodie, DA (2000) Effects of short-term psychological stress on the time and frequency domains of heart-rate variability. Perceptual and motor skills 91:515–524.

Dickens, MJ, Romero, LM (2009) Wild European starlings (Sturnus vulgaris) adjust to captivity with sustained sympathetic nervous system drive and a reduced fight-or-flight response. Physiol Biochem Zool 82:603–610.

Dickens, MJ, Romero, LM (2013) A consensus endocrine profile for chronically stressed wild animals does not exist. General and comparative endocrinology 191:177–189.

Ditmer, MA, Rettler, SJ et al. (2018) American black bears perceive the risks of crossing roads. Behavioral Ecology 29:667–675.

Ditmer, MA, Vincent, JB et al. (2015) Bears show a physiological but limited behavioral response to unmanned aerial vehicles. Current Biology 25:2278–2283.

Downey, LA, Tysse, B et al. (2017) Psychomotor tremor and proprioceptive control problems in current and former stimulant drug users: An accelerometer study of heavy users of amphetamine, MDMA, and other recreational stimulants. The Journal of Clinical Pharmacology 57:1330–1337.

Dragas, J, Jäckel, D et al. (2013) An unsupervised method for on-chip neural spike detection in multi-electrode recording systems. 2013 35th Annual International Conference of the IEEE Engineering in Medicine and Biology Society:2535–2538.

Eckberg, DWAINL, Eckberg, MARYAJ (1982) Human sinus node responses to repetitive, ramped carotid baroreceptor stimuli. American Journal of Physiology-Heart and Circulatory Physiology 242:H638–H644.

Ellen, ED, Rodenburg, TB et al. (2014) The prospects of selection for social genetic effects to improve welfare and productivity in livestock. Frontiers in genetics 5:377.

Ellenberg, U, Mattern, T et al. (2013) Heart rate responses provide an objective evaluation of human disturbance stimuli in breeding birds. Conservation Physiology 1

Ewing, DJ, Martyn, CN et al. (1985) The Value of Cardiovascular Autonomic Function Tests: 10 Years Experience in Diabetes. American Diabetes Association,

Fischer, CP, Franco, LA et al. (2016) Are novel objects perceived as stressful? The effect of novelty on heart rate. Physiology \& behavior 161:7–14.

Fischer, CP, Wright-Lichter, J et al. (2018) Chronic stress and the introduction to captivity: how wild house sparrows (Passer domesticus) adjust to laboratory conditions. General and comparative endocrinology 259:85–92.

Fischer, CP, Romero, LM (2016) The use of α- or β-blockers to ameliorate the chronic stress of captivity in the house sparrow (Passer domesticus). Conserv Physiol 4:cow049.

Fu, X, Chen, W et al. (2011) A wireless implantable sensor network system for in vivo monitoring of physiological signals. IEEE Trans Inf Technol Biomed 15:577–584.

Green, JA (2011) The heart rate method for estimating metabolic rate: review and recommendations. Comp Biochem Physiol A Mol Integr Physiol 158:287–304.

Grippo, AJ, Beltz, TG et al. (2003) Behavioral and cardiovascular changes in the chronic mild stress model of depression. Physiology \& behavior 78:703–710.

Grippo, AJ, Gerena, D et al. (2007) Social isolation induces behavioral and neuroendocrine disturbances relevant to depression in female and male prairie voles. Psychoneuroendocrinology 32:966–980.

Grippo, AJ, Johnson, AK (2009) Stress, depression and cardiovascular dysregulation: a review of neurobiological mechanisms and the integration of research from preclinical disease models. Stress 12:1–21.

Groscolas, R, Viera, V et al. (2010) Heart rate as a predictor of energy expenditure in undisturbed fasting and incubating penguins. Journal of Experimental Biology 213:153–160.

Hall, M, Vasko, R et al. (2004) Acute stress affects heart rate variability during sleep. Psychosomatic medicine 66:56–62.

Hamilton, JL, Alloy, LB (2016) Atypical reactivity of heart rate variability to stress and depression across development: Systematic review of the literature and directions for future research. Clin Psychol Rev 50:67–79.

Hicks, O, Burthe, S et al. (2017) Validating accelerometry estimates of energy expenditure across behaviours using heart rate data in a free-living seabird. Journal of Experimental Biology 220:1875–1881.

Hon, EH, Lee, ST (1963) Electronic evaluation of the fetal heart rate. VIII. Patterns preceding fetal death, further observations. American journal of obstetrics and gynecology 87:814–826.

Hu, W, Jin, X et al. (2016) Deceleration and acceleration capacities of heart rate associated with heart failure with high discriminating performance. Scientific reports 6:23617.

Jänig, W (1985) Das vegetative Nervensystem. (ed) Physiologie des Menschen, Springer, pp 119–157.

Jansen, ASP, Van Nguyen, X et al. (1995) Central command neurons of the sympathetic nervous system: basis of the fight-or-flight response. Science 270:644–646.

Johnson, D, Mayers, I (2001) Multiple organ dysfunction syndrome: a narrative review. Canadian Journal of Anesthesia 48:502–509.

Jose, AD, Collison, D (1970) The normal range and determinants of the intrinsic heart rate in man. Cardiovascular research 4:160–167.

Kemp, AH, Quintana, DS et al. (2010) Impact of depression and antidepressant treatment on heart rate variability: a review and meta-analysis. Biological psychiatry 67:1067–1074.

Kiss, O, Sydó, N et al. (2016) Detailed heart rate variability analysis in athletes. Clinical Autonomic Research 26:245–252.

Kleiger, RE, Miller, JP et al. (1987) Decreased heart rate variability and its association with increased mortality after acute myocardial infarction. The American journal of cardiology 59:256–262.

Kleiger, RE, Stein, PK et al. (2005) Heart rate variability: measurement and clinical utility. Annals of Noninvasive Electrocardiology 10:88–101.

Koolhaas, JM, Korte, SM et al. (1999) Coping styles in animals: current status in behavior and stress-physiology. Neuroscience \& Biobehavioral Reviews 23:925–935.

Kostelanetz, S, Dickens, MJ et al. (2009) Combined effects of molt and chronic stress on heart rate, heart rate variability, and glucocorticoid physiology in European Starlings. Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology 154:493–501.

Krittayaphong, R, Cascio, WE et al. (1997) Heart rate variability in patients with coronary artery disease: differences in patients with higher and lower depression scores. Psychosom Med 59:231–235.

Kröschel, M, Reineking, B et al. (2017) Remote monitoring of vigilance behavior in large herbivores using acceleration data. Animal Biotelemetry 5:10.

Lacombe, AMA, Jones, DR (1990) The source of circulating catecholamines in forced dived ducks. General and comparative endocrinology 80:41–47.

Laferton, JAC, Stenzel, NM et al. (2018) The Beliefs About Stress Scale (BASS): Development, reliability, and validity. International Journal of Stress Management 25:72.

Lakin, R, Guzman, C et al. (2018) Changes in Heart Rate and Its Regulation by the Autonomic Nervous System Do Not Differ Between Forced and Voluntary Exercise in Mice. Front Physiol 9:841.

Landry, MP, Kruss, S et al. (2014) Experimental tools to study molecular recognition within the nanoparticle corona. Sensors (Basel) 14:16196–16211.

Lane, JE, Boutin, S et al. (2009) Sexually selected behaviour: red squirrel males search for reproductive success. Journal of Animal Ecology 78:296–304.

Lattin, CR, Romero, LM (2015) Seasonal variation in glucocorticoid and mineralocorticoid receptors in metabolic tissues of the house sparrow (Passer domesticus). General and comparative endocrinology 214:95–102.

Levine, HJ (1997) Rest heart rate and life expectancy. J Am Coll Cardiol 30:1104–1106.

Levy, MN, Schwartz, PJ (1994) Vagal control of the heart: Experimental basis and clinical implications. Futura Publishing Company,

Licht, CMM, Penninx, BW et al. (2011) To Include or Not to Include? A Response to the Meta-Analysis of Heart Rate Variability and Depression. Biological Psychiatry 69:e1.

MacDougall-Shackleton, SA, Bonier, F et al. (2019) Glucocorticoids and “Stress” Are Not Synonymous. Integrative Organismal Biology 1

Malik, M, Farrell, T et al. (1989) Heart rate variability in relation to prognosis after myocardial infarction: selection of optimal processing techniques. European heart journal 10:1060–1074.

Martire, VL, Caruso, D et al. (2019) STRESS & SLEEP: A RELATIONSHIP LASTING A LIFETIME. Neuroscience & Biobehavioral Reviews

Massot, B, Arthaud, S et al. (2019) ONEIROS, a new miniature standalone device for recording sleep electrophysiology, physiology, temperatures and behavior in the lab and field. J Neurosci Methods 316:103–116.

Mazurak, N, Sauer, H et al. (2016) Effect of a weight reduction program on baseline and stress-induced heart rate variability in children with obesity. Obesity 24:439–445.

Muller, MS, Vyssotski, AL et al. (2018) Individual differences in heart rate reveal a broad range of autonomic phenotypes in a free-living seabird population. Journal of Experimental Biology 221:jeb182758.

Müller, MS, Vyssotski, AL et al. (2017) Heart rate variability reveals that a decrease in parasympathetic ('rest-and-digest') activity dominates autonomic stress responses in a free-living seabird. Comp Biochem Physiol A Mol Integr Physiol 212:117–126.

Musyl, MK, Domeier, ML et al. (2011) Performance of pop-up satellite archival tags. Marine Ecology Progress Series 433:1–28.

Nephew, BC, Kahn, SA et al. (2003) Heart rate and behavior are regulated independently of corticosterone following diverse acute stressors. General and comparative endocrinology 133:173–180.

Nephew, BC, Romero, LM (2003) Behavioral, physiological, and endocrine responses of starlings to acute increases in density. Hormones and Behavior 44:222–232.

O'Mara, MT, Wikelski, M et al. (2017) Cyclic bouts of extreme bradycardia counteract the high metabolism of frugivorous bats. elife 6:e26686.

Pacifici, M, Foden, WB et al. (2015) Assessing species vulnerability to climate change. Nature climate change 5:215.

Padgett, DA, Glaser, R (2003) How stress influences the immune response. Trends in immunology 24:444–448.

Pagani, M, Lombardi, F et al. (1986) Power spectral analysis of heart rate and arterial pressure variabilities as a marker of sympatho-vagal interaction in man and conscious dog. Circulation research 59:178–193.

Pardo, Y, Noel Merz, CB et al. (2000) Exercise conditioning and heart rate variability: evidence of a threshold effect. Clinical Cardiology 23:615–620.

Park, D, Lee, M et al. (2018) Determination of Optimal Heart Rate Variability Features Based on SVM-Recursive Feature Elimination for Cumulative Stress Monitoring Using ECG Sensor. Sensors (Basel) 18

Perini, R, Veicsteinas, A (2003) Heart rate variability and autonomic activity at rest and during exercise in various physiological conditions. European journal of applied physiology 90:317–325.

Pohlin, F, Brabender, K et al. (2017) Seasonal Variations in Heart Rate Variability as an Indicator of Stress in Free-Ranging Pregnant Przewalski's Horses (E. ferus przewalskii) within the Hortobágy National Park in Hungary. Front Physiol 8:664.

Porges, SW (1995) Orienting in a defensive world: Mammalian modifications of our evolutionary heritage. A polyvagal theory. Psychophysiology 32:301–318.

Porges, SW (2003) The polyvagal theory: Phylogenetic contributions to social behavior. Physiology \& behavior 79:503–513.

Pulopulos, MM, Vanderhasselt, M-A et al. (2018) Association between changes in heart rate variability during the anticipation of a stressful situation and the stress-induced cortisol response. Psychoneuroendocrinology 94:63–71.

Rattenborg, NC, Voirin, B et al. (2016) Evidence that birds sleep in mid-flight. Nat Commun 7:12468.

Rattenborg, NC, Voirin, B et al. (2008) Sleeping outside the box: electroencephalographic measures of sleep in sloths inhabiting a rainforest. Biol Lett 4:402–405.

Reardon, M, Malik, M (1996) Changes in heart rate variability with age. Pacing and clinical electrophysiology 19:1863–1866.

Reefmann, N, B\”utikofer, FK et al. (2009) Physiological expression of emotional reactions in sheep. Physiology \& behavior 98:235–241.

Romero, LM, Platts, SH et al. (2015) Understanding stress in the healthy animal--potential paths for progress. Stress 18:491–497.

Romero, LM, Dickens, MJ et al. (2009) The Reactive Scope Model - a new model integrating homeostasis, allostasis, and stress. Horm Behav 55:375–389.

Romero, T, Nagasawa, M et al. (2014) Oxytocin promotes social bonding in dogs. Proceedings of the National Academy of Sciences 111:9085–9090.

Sanches, PVW, Taylor, EW et al. (2019) Respiratory sinus arrhythmia is a major component of heart rate variability in undisturbed, remotely monitored rattlesnakes, Crotalus durissus. Journal of Experimental Biology 222:jeb197954.

Schiweck, C, Piette, D et al. (2019) Heart rate and high frequency heart rate variability during stress as biomarker for clinical depression. A systematic review. Psychol Med 49:200–211.

Schmidt, A, M\”ostl, E et al. (2010) Cortisol release and heart rate variability in horses during road transport. Hormones and behavior 57:209–215.

Schoenle, LA, Zimmer, C et al. (2018) Understanding context dependence in glucocorticoid--fitness relationships: the role of the nature of the challenge, the intensity and frequency of stressors, and life history. Integrative and comparative biology 58:777–789.

Selye, H (1943) Encyclopedia of endocrinology. AWT Franks Publishing Company,

Selye, H (1956) The stress of life.

Selye, H, others (1936) A syndrome produced by diverse nocuous agents. Nature 138:32.

Shaffer, F, McCraty, R et al. (2014) A healthy heart is not a metronome: an integrative review of the heart’s anatomy and heart rate variability. Front Psychol 5:1040.

Shaffer, F, Ginsberg, JP (2017) An Overview of Heart Rate Variability Metrics and Norms. Frontiers in Public Health 5

Shapiro, CM, Bortz, R et al. (1981) Slow-wave sleep: a recovery period after exercise. Science 214:1253–1254.

Sheriff, MJ, Dantzer, B et al. (2011) Measuring stress in wildlife: techniques for quantifying glucocorticoids. Oecologia 166:869–887.

Spivey, RJ, Bishop, CM (2014) An implantable instrument for studying the long-term flight biology of migratory birds. Rev Sci Instrum 85:014301.

Stalder, T, Evans, P et al. (2011) Associations between the cortisol awakening response and heart rate variability. Psychoneuroendocrinology 36:454–462.

Stauss, HM (2003) Heart rate variability. Am J Physiol Regul Integr Comp Physiol 285:R927–31.

Stein, R, Medeiros, CM et al. (2002) Intrinsic sinus and atrioventricular node electrophysiologic adaptations in endurance athletes. Journal of the American College of Cardiology 39:1033–1038.

Takei, Y, Suzuki, I et al. (2016) Development of an animal-borne blood sample collection device and its deployment for the determination of cardiovascular and stress hormones in phocid seals. American Journal of Physiology-Regulatory, Integrative and Comparative Physiology 311:R788–R796.

Thayer, JF, Lane, RD (2000) A model of neurovisceral integration in emotion regulation and dysregulation. Journal of affective disorders 61:201–216.

Thayer, JF, Sternberg, E (2006) Beyond heart rate variability: vagal regulation of allostatic systems. Annals of the New York Academy of Sciences 1088:361–372.

Thayer, JF, Åhs, F et al. (2012) A meta-analysis of heart rate variability and neuroimaging studies: Implications for heart rate variability as a marker of stress and health. Neuroscience & Biobehavioral Reviews 36:747–756.

Thayer, JF, Yamamoto, SS et al. (2010) The relationship of autonomic imbalance, heart rate variability and cardiovascular disease risk factors. International Journal of Cardiology 141:122–131.

Umana, E, Solares, CA et al. (2003) Tachycardia-induced cardiomyopathy. The American journal of medicine 114:51–55.

Uvnas-Moberg, K (1998) Oxytocin may mediate the benefits of positive social interaction and emotions. Psychoneuroendocrinology 23:819–835.

Valiente, C, Eisenberg, N et al. (2003) The relations of effortful control and reactive control to children’s externalizing problems: A longitudinal assessment. Journal of personality 71:1171–1196.

Viblanc, VA, Smith, AD et al. (2012a) Coping with continuous human disturbance in the wild: insights from penguin heart rate response to various stressors. BMC ecology 12:10.

Viblanc, VA, Smith, AD et al. (2015) Modulation of heart rate response to acute stressors throughout the breeding season in the king penguin Aptenodytes patagonicus. Journal of Experimental Biology 218:1686–1692.

Viblanc, VA, Valette, V et al. (2012b) Coping with social stress: heart rate responses to agonistic interactions in king penguins. Behavioral Ecology 23:1178–1185.

von Borell, E, Langbein, J et al. (2007) Heart rate variability as a measure of autonomic regulation of cardiac activity for assessing stress and welfare in farm animals -- a review. Physiol Behav 92:293–316.

Vyssotski, AL, Serkov, AN et al. (2006) Miniature neurologgers for flying pigeons: multichannel EEG and action and field potentials in combination with GPS recording. Journal of neurophysiology 95:1263–1273.

Watkins, LL, Grossman, P et al. (1998) Anxiety and vagal control of heart rate. Psychosomatic medicine 60:498–502.

Weber, CS, Thayer, JF et al. (2010) Low vagal tone is associated with impaired post stress recovery of cardiovascular, endocrine, and immune markers. Eur J Appl Physiol 109:201–211.

Weimerskirch, H, Shaffer, SA et al. (2002) Heart rate and energy expenditure of incubating wandering albatrosses: basal levels, natural variation, and the effects of human disturbance. J Exp Biol 205:475–483.

Williams, CT, Barnes, BM et al. (2016) Integrating physiology, behavior, and energetics: Biologging in a free-living arctic hibernator. Comp Biochem Physiol A Mol Integr Physiol 202:53–62.

Williams, HJ, Taylor, LA et al. (2019) Optimizing the use of bio-loggers for movement ecology research. J Anim Ecol

Wilson, RP, Scolaro, A et al. (2004) To the bottom of the heart: cloacal movement as an index of cardiac frequency, respiration and digestive evacuation in penguins. Marine Biology 144:813–827.

Wilson, RP, Shepard, ELC et al. (2008) Prying into the intimate details of animal lives: use of a daily diary on animals. Endangered Species Research 4:123–137.

Woakes, AJ, Butler, PJ et al. (1995) Implantable data logging system for heart rate and body temperature: its application to the estimation of field metabolic rates in Antarctic predators. Med Biol Eng Comput 33:145–151.

Wolf, MM, Varigos, GA et al. (1978) Sinus Arrhythmia in Acute Myocardial Infarction. Medical Journal of Australia 2:52–53.

Xhyheri, B, Manfrini, O et al. (2012) Heart Rate Variability Today. Progress in Cardiovascular Diseases 55:321–331.

Yerkes, RM, Dodson, JD (1908) The relation of strength of stimulus to rapidity of habit-formation. Journal of comparative neurology and psychology 18:459–482.

Young-Ho, Y, Eui-Sung, J et al. (2004 Nov) 30th Annual Conference of IEEE Industrial Electronics Society, 2004. IECON 2004 Design of implantable wireless biomedical signals telemetry system. 3:2982–2986 Vol. 3.

Young, BJ, Leaton, RN (1994) Fear potentiation of acoustic startle stimulus-evoked heart rate changes in rats. Behavioral Neuroscience 108:1065.

Zhang, D, Shen, X et al. (2016) Resting heart rate and all-cause and cardiovascular mortality in the general population: a meta-analysis. Cmaj 188:E53–E63.