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Research paper

Chronic stress and the introduction to captivity: How wild house sparrows (*Passer domesticus*) adjust to laboratory conditions

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ABSTRACT

The conditions of captivity can cause chronic stress in wild animals. Newly-captured animals may experience weight loss, elevated glucocorticoid hormones, increased heart rate, increased resting adrenomedullary activation, and an altered heart rate response to acute stressors. As captivity conditions persist, chronic stress may decrease as animals adjust to the stressors of captivity. In this study, house sparrows (*Passer domesticus*) were captured from the wild, fitted with heart rate transmitters in a minor surgical process, and individually housed in an indoor bird facility. Mass, baseline corticosterone, resting heart rate, resting adrenomedullary activation, and the acute heart rate response to a sudden noise were measured over the course of the first 6 weeks of captivity. Birds lost weight during the first weeks of captivity, which was regained by week 5. Baseline corticosterone peaked at day 7, decreased sharply by day 11, and continued to decrease throughout the 6 weeks. Although heart rate in the first 24 h could not be collected, daytime heart rate decreased from day 1 through day 20, where it reached a stable plateau. Daytime heart rate variability decreased through the entire 6 weeks, which may indicate a gradual shift from sympathetic to parasympathetic nervous system regulation of heart rate. The acute heart rate response to a sudden noise lasted longer at day 6 than earlier or later in captivity. In conclusion, the data indicate that the different physiological systems associated with chronic stress adjust to captivity over different timelines.

1. Introduction

Captivity can be a potent source of stress for newly captured wild animals. The conditions of captivity (e.g. confinement, artificial lighting, changes in diet, and the presence of and handling by humans) are unpredictable, uncontrollable stimuli that can activate the stress response systems (Morgan and Tromborg, 2007). During the initial introduction to captivity, wild birds may experience weight loss (e.g. Ruiz et al., 2002; Lattin et al., 2012), high heart rate (Dickens et al., 2009; Fischer and Romero, 2016), elevated glucocorticoid hormones (e.g. Lattin et al., 2012; Fokidis et al., 2011), and alterations to other systems, such as the immune (e.g. Buehler et al., 2008) and reproductive systems (e.g. Lombardo and Thorpe, 2009). Many animals are able to survive well in captivity long term, however, and the period of chronic stress may eventually by followed by acclimation, as the affected physiological systems return to normal or reach a new level appropriate to their new environment. We monitored newly captured house sparrows (*Passer domesticus*) for 6 weeks to determine the timing of acclimation to captivity with a focus on baseline corticosterone (CORT; the primary glucocorticoid in birds) and the adrenomedullary system. These birds were subjected to regular handling and blood sampling throughout the captivity period. They also experienced a minor surgery shortly after capture to fit them with a backpack-style heart rate transmitter, which they wore throughout the study.

The stress response is adaptive; a mechanism to help vertebrates survive challenges to homeostasis (Romero and Wingfield, 2016). A noxious stimulus, such as a predator attack, stimulates first an adrenomedullary response (an increase in the catecholamine hormones epinephrine and norepinephrine leading to an increase in heart rate) followed shortly by a hypothalamic–pituitaryadrenal (HPA) axis response culminating in an increase in glucocorticoid hormones (Sapolsky et al., 2000). These physiological systems work together to help the animal survive the crisis. However, when stressors are ongoing or repeated, the resulting chronic stress can be harmful (Romero et al., 2009). In captive conditions, wild birds may experience a loss in mass (Fokidis et al., 2011; Lattin et al., 2012), an increase in baseline CORT (Adams et al., 2011; Fokidis et al., 2011; Lattin et al., 2012), changes in behavior (Adams et al., 2011; Fokidis et al., 2011), and changes to heart rate and heart rate variability (HRV; a metric of sympathetic nervous

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system activity; see Section 2.4 [Dickens and Romero, 2009; Fischer and Romero, 2016]). The HPA and adrenomedullary systems are independently regulated (Nephew et al., 2003) and therefore may require different lengths of time to acclimate to captive conditions. However, the two systems do influence one another; glucocorticoids act permissively on the action of epinephrine and norepinephrine and so activation of the HPA axis can result in a more effective adrenomedullary response (Sapolsky et al., 2000).

In a previous study, we found that house sparrows had much higher heart rates during the first week after capture than after 1 month in captivity (Fischer and Romero, 2016). Presumably, sometime between day 7 and day 30 of captivity, house sparrows must experience a decrease in heart rate, but how long this decline lasts and whether it has stabilized by day 30 is unknown. This is in contrast to European starlings (*Sternus vulgaris*), the only other bird species to our knowledge in which heart rate has been monitored during adjustment to captivity. European starlings had high heart rates at capture that decreased over the first 48 h to a steady plateau that was the same as long-term captives (Dickens and Romero, 2009). Newly captured house sparrows had a moderately reduced acute response to startle compared to one month captives (Fischer and Romero, 2016). This pattern was more extreme in European starlings, which had a nearly eliminated startle response for at least the first 10 days of captivity (Dickens and Romero, 2009). House sparrows also had elevated baseline CORT during the first 5–7 days of captivity (Fischer and Romero, 2016; Lattin et al., 2012). This may decline towards wild levels, as in white-crowned sparrows (Wingfield et al., 1982), or it may remain elevated, as in the curve-billed thrasher (Fokidis et al., 2011). In many lab studies that use wild birds, two weeks to one month is considered sufficient time for the animals to adjust to captivity (e.g. Bókony et al., 2014; Lattin and Romero, 2014). However, to our knowledge it is unknown whether acclimation in the HPA system coincides with acclimation in the adrenomedullary system for any species. The timing of acclimation for house sparrows, a species that is frequently used in laboratory studies, is also unknown. To address these outstanding concerns, we repeatedly sampled mass, baseline CORT, activity, heart rate, HRV, and the acute heart rate response to startle in wild house sparrows during the first 6 weeks of captivity.

2. Methods

2.1. Experimental design

10 house sparrows (4 females, 6 males) were captured with Potter traps on or near the Tufts University campus in Medford, MA. A group of 5 birds was caught on June 2, 2016 (Group A) and a second group of 5 on June 3 (Group B). On the day of capture (day 0), birds were transported in cloth bags to the laboratory, where they were anesthetized and surgically fitted with a heart rate transmitter harness (see Section 2.2). Birds were kept in individual, darkened cages until they had recovered from surgery. Because of possible anesthesia effects, we began collecting heart rate approximately 24 h after capture. After surgery, birds were moved to individual cages in an indoor bird facility with a 15-h light: 9-h dark cycle to approximate the natural photoperiod at this time of year. They were provided with *ad libitum* food (mixed seed) and water. Temperature in the bird room was maintained around 23 °C. Each cage was fitted with a receiver plate to record heart rate data. Because we had 10 birds and only 7 receiver plates, 3 cages did not have receiver plates. We rotated 3 of the Group A birds and 3 of the Group B birds between cages with and without receiver plates, such that birds were in the cages with receiver plates on all days for which heart rate recording was scheduled. For example, heart rate was recorded for Group A birds from day 1 to day 2 (June 3–4, 2016). At the end of the recording period, we switched cages for 3 Group A birds and 3 Group B birds. Heart rate for Group B birds was then collected for June 4–5 (day 1–2 for that group). A summary of the experimental design is shown in Fig. 1.

Birds were weighed on days 0 (immediately upon capture), 4, 11, 18, 25, 32, and 39 using a spring scale precise to the nearest 0.5 g. On day 0 (immediately upon capture), and every 3–4 days thereafter, a blood sample was taken (see Fig. 1). For each sample, the alar vein was punctured and ~40 μl blood was collected in a heparinized capillary tube. We collected samples within 3 min of capture on day 0, and within 3 min of entering the bird room on all other days. Within this time frame, an acute increase in CORT has not started or has only just begun (Romero and Reed, 2005). Blood samples were kept on ice until processing, when they were centrifuged at ~1200*g* for 8 min (Centrific Model 225, Fisher Scientific, Pittsburgh, PA, USA). Plasma was collected and frozen at -20 °C until CORT concentrations could be analyzed (see Section 2.3). 10–20 μL of plasma was generally recovered.

On days 1, 3, 5, 9, 12, 16, 20, 27, 34, and 41, the birds were caught and their heart rate transmitters were switched on with a magnetic switch. Their resting heart rate was then automatically recorded using Data Science International's Acquisition program for 3 min every 2 h for 24 h. At every sampling interval, the receivers also recorded a unitless activity metric. The receiver plate contains 3 radio receivers. Any change in relative signal strength between the 3 receivers is interpreted as movement within the cage. A higher activity score indicates more movement within the sampling interval. Activity was analyzed on Data Science International's Analysis software. The Acquisition and Analysis programs come bundled with the implantable transmitters.

On days 2, 6, 13, 21, 28, 35, and 42, at the end of the resting heart rate sampling period, an acute heart rate response to startle was recorded. Heart rate was recorded for 5 min. Then the door to the bird room was suddenly opened and closed. Heart rate was recorded for another 10 min. Afterwards, the heart rate transmitters were magnetically switched off to conserve batteries and the 24-h sampling was initiated for the alternate group of birds.

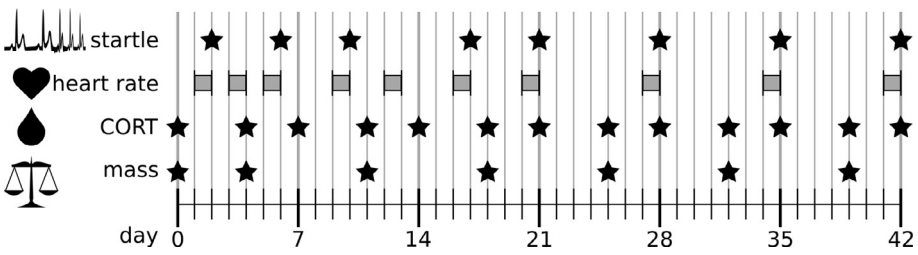

Fig. 1. Timeline of experimental protocols. 5 house sparrows were caught on June 2, 2016 and 5 were caught on June 3. Day 0 refers to the day of capture for both groups. Blood samples for CORT analysis were taken in the morning. Heart rate (as well as HRV and activity) was sampled for 3 min every 2 h beginning in the midafternoon of one day and continuing through midafternoon of the following day. The acute heart rate response to a sudden noise (startle) was recorded after the heart rate samples were completed. Heart rate was recorded for 5 min, the door to the bird facility was opened and suddenly closed, and heart rate was recorded for a further 10 min.