Physiology & Behavior 169 (2017) 208–215

Contents lists available at ScienceDirect

Physiology & Behavior

journal homepage: www.elsevier.com/locate/phb

Autonomic, endocrine and behavioural responses to thunder in laboratory and companion dogs

Carla Caroline Franzini de Souza [a], Carolina Elisabetta Martins Maccariello [a], Daniel Penteado Martins Dias [b], Norma Aparecida dos Santos Almeida [a,c], Magda Alves de Medeiros [a,d,*]

[a] Department of Physiological Sciences, Institute of Biology, Federal Rural University of Rio de Janeiro, BR 465, KM 7, 23890-000 Seropédica, RJ, Brazil
[b] Department of Physiology, School of Medicine of Ribeirão Preto, University of São Paulo, 14049-900 Ribeirão Preto, SP, Brazil
[c] Graduate Program in Physiological Sciences, Federal Rural University of Rio de Janeiro, BR 465, KM 7, 23890-000 Seropédica, RJ, Brazil
[d] Graduate Program in Veterinary Medicine and Graduate Program in Physiological Sciences, Federal Rural University of Rio de Janeiro, BR 465, KM 7, 23890-000 Seropédica, RJ, Brazil

HIGHLIGHTS

- Thunder stimulus (103–104 dB) causes physiological and behavioural changes in dogs.
- Sound stimulus induces autonomic imbalance with sympathetic predominance in dogs.
- Companion dogs had higher cortisol levels than Beagles.
- Only companion dogs had significant increase in the cortisol.
- Laboratory dogs had more pronounced behavioural response than companion dogs.

ARTICLE INFO

Article history:
Received 15 August 2016
Received in revised form 23 November 2016
Accepted 2 December 2016
Available online 08 December 2016

ABSTRACT

Dogs are highly sensitive to sound stimuli, especially fireworks, firearms, and thunder, and therefore these sounds are used as models of stress reactivity in dogs. Companion and laboratory dogs may respond differently to stressful stimuli, due to differences in management and their relationship with humans. Therefore, the reactivity of beagle dogs (laboratory) and companion dogs to an acute acoustic stress model was studied by analysing the heart rate variability (HRV; cardiac interval values), serum cortisol levels and various behavioural parameters. Eight beagles and six privately owned dogs with no history of phobia to thunder were used. The sound stimulus consisted of a standardized recording of thunder for 2.5 min with a maximum intensity of 103–104 dB. To evaluate the HRV, cardiac intervals were recorded using a frequency meter (Polar RS800CX model), and later the data were analysed using CardioSeries 2.4.1 software. In both laboratory and companion dogs, thunder promoted an increase in the power of the LF band of the cardiac interval spectrum, in the LF/HF ratio and in the HR, and a decrease in the power of the HF band of the cardiac interval spectrum. Companion dogs showed higher cortisol levels, than beagles, independently of the time point studied and a significant increase in the cortisol levels 15 min after acoustic stress, while beagles did not show any alterations in their cortisol levels in response to the sound. On the other hand, beagles showed higher scores in the trembling, hiding, vigilance, running, salivation, bolting and startle parameters than companion dogs. Our results showed that independently of the sound stimulus, companion dogs had higher cortisol levels than laboratory dogs. Furthermore, the sound stimulus induced a marked autonomic imbalance towards sympathetic predominance in both laboratory and companion dogs. However a significant increase in the cortisol was observed only in companion dogs. On the other hand, in general the behavioural response was more pronounced in laboratory dogs than in companion dogs.

1. Introduction

Animals and humans are constantly subjected to stress in their daily lives. During a stressful situation two neuroendocrine axes are classically activated: the sympathetic adrenomedullary (SAM), which increases the secretion of catecholamines and hence the rapid excitation of the

* Corresponding author at: Department of Physiological Sciences, Institute of Biology, Federal Rural University of Rio de Janeiro, BR 465, KM 7, 23890-000 Seropédica, RJ, Brazil.
E-mail addresses: carlacfranzini@gmail.com (C.C. Franzini de Souza), carolmaccariello@uol.com.br (C.E.M. Maccariello), danielpenteado@gmail.com (D.P.M. Dias), normaalmeida@gmail.com (N.A.S. Almeida), magda.medeiros@gmail.com (M.A. Medeiros).

http://dx.doi.org/10.1016/j.physbeh.2016.12.006

cardiovascular system, contributing to increased heart rate (HR) and arterial blood pressure; and the hypothalamic-pituitary-adrenal (HPA), which leads to an increase in cortisol production, and consequent mobilization of energy [1]. Dysregulation of these stress responses may affect negatively several physiological systems, including the immune and cardiovascular systems, metabolic function and behaviour [2].

Several stimuli can be considered stressors for dogs, for example: transportation [3], social and spatial restrictions [4] and loud noise, [5] among others. Dogs are highly sensitive to sound stimuli, mainly fireworks, firearms and thunder [6]. These stimuli are closely associated with anxiety disorders such as sound phobia in this species [7–9]. Thus, these sound stimuli are used as stress models for dogs [5,6,10, 11] and for studies concerning animal welfare and strategies to reduce stress.

Stress levels can be evaluated by the measurement of physiological and behavioural parameters. The most commonly used parameter for stress assessment in various species is cortisol Also, the analysis of the heart rate variability (HRV) has proved to be a reliable non-invasive tool to evaluate the cardiovascular autonomic modulation in animals undergoing several types of challenging situations [12]. In dogs, monitoring cardiac parameters and serum cortisol level assessments, as well as behavioural analysis have been used to study stress responses [4,6,13–15].

The way an individual responds to a stressful stimulus depends on several factors, including their previous experience with stimuli similar or different to the current stimulus. Thus, companion and laboratory dogs may respond differently to stressful stimuli, due to differences in their management and relationship with humans. To the best of our knowledge, no studies have been done to compare autonomic, hormonal, and behavioural responses to acute acoustic stress in companion and laboratory dogs. The purpose of the present study was to evaluate the cardiac autonomic modulation, serum cortisol levels and several behavioural parameters under sound stimuli in beagle dogs (laboratory) and companion dogs (a variety of breeds).

2. Methods

2.1. Animals

All procedures were assessed and approved by the Committee on the use of Human and Animal Subjects in Teaching and Research Ethics of the Federal Rural University of Rio de Janeiro COMEP-UFRRJ (protocol n. 23083.013953/2010-41). Two groups of dogs were studied:

2.1.1. Laboratory dogs

Eight beagles (4 males; 1 to 6 years old; weighing from 9 to 16 kg; in good health) from the Kennel of the Laboratory of Experimental Chemotherapy in Veterinary Parasitology of the Federal Rural University of Rio de Janeiro (Seropédica, Rio de Janeiro, Brazil) were used. At this kennel all dogs were kept in groups of 5 to 8 animals in semi-open kennels (total area of 30 m^2, with 4 m^2 of a sheltered area) with full access to sunlight, and water and food (commercial dog food) ad libitum. The kennels do not provide an isolated environment, so the dogs can hear thunder storms or any other natural events.

2.1.2. Companion dogs

These dogs were recruited through advertisements in the Veterinary Hospital of Small Animals and the community around the Federal Rural University of Rio de Janeiro. Inclusion criteria for the study selection were dogs of either sex, between 2 and 6 years old; weighing from 10 to 30 kg; in good health; and the exclusion criteria were signs or history of neurological or behavioural problems, primarily excessive fear of thunder or any debilitating disease. The recruitment period was from October 2013 until February 2014 and the dog owner answered a questionnaire about the general characteristics of their dogs and their behaviour as suggested by [16]. In total, 22 dog owners expressed interest in taking part in the study. Among these, based on the general physical characteristics, 4 males and 2 females were selected for the study.

None of the animals used in this study came from the same social group (same house or the same group in the kennel).

2.2. Experimental design

All individual experimental procedures started at 0900 h. Initially, with dogs at their locations (houses for companion dogs; kennels for laboratory dogs), a HR monitor designed for human beings (Polar RS800cx, Polar®, Kempele, Finland) was strapped to the chest of the dogs in order to have the HR sampled on a beat-by-beat basis. In order to maximize the contact between the electrodes and the skin, the fur of precordial region skin area was cut using a hair clipper and a conductive gel was applied. Afterwards, the dogs rested undisturbed for 10 min for baseline measurements of HR data (i.e. data classified as basal 1 - house/kennel) and baseline collection of blood samples (Basal 1). Next, the dogs were taken to the test room by car (10 min) in their transit boxes ($87 \times 57 \times 59$cm).

At the test room, the dogs were left undisturbed for 30-minutes followed by another blood sample collection (Basal 2). Animals were allowed to rest quietly for 20 min after which they were placed 1 m away from the sound source. The sound stimulus was turned on for 180 s and blood samples were collected 15 (S15), 30 (S30) and 60 (S60) minutes after the end of the sound stimulus. In order to assess the behavioural responses, the dogs were videotaped before, throughout the whole sound stimulus period and for the 5 min after the sound stimulus ended. Finally, the HR monitors were removed and the dogs were returned to their houses or kennels.

The experiment room was 11m^2 in area, had artificial lighting and the temperature was maintained at 22 ± 1 °C. There were only a few objects in the room: two benches, a veterinary procedure table, a bench with the sound source and a video camera. Two researchers remained in the room during the experiment but without interacting with the dogs at any time in any way. All procedures were performed individually.

2.3. Acoustic stimulus

The sound stimulus consisted of a 180-second-long recording of thunder obtained from two separate sound files (e.g. *very loud thunder*, *a close thunder crack*, and *high quality stereo sound of a major thunder clap during a storm*), purchased from the website http://www.sound-effect.com. The sound file format used (a wave file) has the advantage of preserving all frequency oscillations of the original sound. The sound level was adjusted to 103–104 dB and tested with a decibel meter (Sound Level Meters, Model 732A, BK Precision®, Yorba Linda, CA, USA).

2.4. Heart rate variability

HR was continuously sampled with a heart monitor (RS 800cx, Polar, Kempele, Finland). Following acquisition, the data were transmitted from the heart monitor to the custom computer software (Polar Pro Trainer v5, Polar, Kempele, Finland) through an infrared interface. The recordings were then processed by Polar Pro Trainer software, and the time series of cardiac interval values were generated and loaded into a spreadsheet software (Microsoft Excel v2003) for inspection. A line chart was built from the cardiac interval data points to perform the inspection. Then the erroneous values were visually identified and corrected by calculating the average from two points before and two points after the erroneous data point. A study from Salo et al. [17] tested the effect of correcting different amounts of beats in the cardiac interval series, ranging from 5 to 50% of correction. At 5% of correction, minor changes were seen in the results, as compared to the original non-corrected series [17]. In addition, Peltola [18] indicates that time series with >20% of correction would be rejected. In the current study, taking