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Comparative analysis of stress responses in dogs and cats during the covid-19 pandemic: P a focus on cortisol, total leukocytes, eosinophils, and behavioral changes

[Análise comparativa das respostas ao estresse em cães e gatos durante a pandemia de covid-19: foco no cortisol, nos leucócitos totais, nos eosinófilos e nas alterações comportamentais]

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ABSTRACT

The COVID-19 pandemic has ushered in unprecedented changes in the lives of humans and their companion animals, impacting their daily routines, social interactions, and stress levels. This study investigates the effect of pandemic-related restrictions, including limited time outside and changes in owner habits, on the stress indicators of 34 dogs and 29 cats. Cortisol levels, total leukocyte counts, eosinophil counts, and behavioral changes have been analyzed to assess how these species are differentially affected by the altered environment and human interactions during the pandemic. The study revealed important behavioral changes in cats while the biochemical and hematological parameters were considerably affected in dogs. Our findings shed light on the unique stress experiences of dogs and cats and underscore the importance of considering individualized strategies for managing their well-being during extraordinary circumstances.

Keywords: stress, cortisol, cats, dogs, eosinophils

RESUMO

A pandemia da covid-19 marcou o início de mudanças sem precedentes na vida dos seres humanos e dos seus animais de companhia, impactando as suas rotinas diárias, as interações sociais e os níveis de estresse. Este estudo investiga o efeito das restrições relacionadas à pandemia, incluindo tempo limitado fora de casa e mudanças nos hábitos dos proprietários, nos indicadores de estresse de 34 cães e 29 gatos. Os níveis de cortisol, a contagem total de leucócitos, a contagem de eosinófilos e as mudanças comportamentais foram analisados para avaliar como essas espécies são diferentemente afetadas pelo ambiente alterado e pelas interações humanos durante a pandemia. O estudo revelou importantes alterações comportamentais em gatos, enquanto os parâmetros bioquímicos e hematológicos foram consideravelmente afetados em cães. As descobertas esclarecem as experiências únicas de estresse de cães e gatos e sublinham a importância de considerar estratégias individualizadas para gerir o bemestar deles durante circunstâncias extraordinárias.

Palavras-chave: estresse, cortisol, gatos, cães, eosinófilos

INTRODUCTION

The COVID-19 pandemic, caused by the novel coronavirus SARS-CoV-2, has ushered in an unprecedented era of global disruption, affecting every facet of human life. Beyond its direct

impact on human health, the pandemic has brought forth a multitude of societal and behavioral changes, altering the way people work, interact, and live (Kandel *et al.*, 2020). These changes have reverberated through households worldwide, significantly impacting Page 1 a 6

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the lives of companion animals dogs and cats, in particular. In this rapidly evolving context, understanding the implications of the pandemic on the well-being of these beloved pets is of paramount importance (Ratschen *et al.*, 2020).

The human-animal bond, a profound and ancient connection between humans and their companion animals, has been a source of emotional support and companionship throughout history. Dogs and cats, as the most common and cherished of these companions, inhabit a unique place in our homes and hearts (Archer, 1997). Yet, the COVID-19 pandemic has introduced a host of challenges to this bond, prompting us to delve into the intricate dynamics of how dogs and cats respond to the myriad changes precipitated by the pandemic (Ratschen *et al.*, 2020).

Stress, as a multifaceted and often detrimental force, has long been a subject of study in the context of animal health and welfare. In both dogs and cats, stress can manifest as physiological and behavioral changes, often reflecting an individual's response to environmental or situational factors. These responses, including alterations in cortisol levels, immune system dynamics, and observable behaviors, provide a window into the well-being and adaptation of these animals during times of upheaval (Beerda *et al.*, 1999).

While stress responses in companion animals have been studied extensively in various contexts, the COVID-19 pandemic represents a unique and globally shared experience that has not only disrupted human lives but also cast a new light on the experiences of our canine and feline companions (Ratschen et al., 2020). The pandemic has unfolded as a complex and multifaceted event, influencing the daily lives of pet owners and their pets alike. It has been marked by a series of societal changes, including lockdowns, social distancing measures, remote work, and shifts in owner behavior-all of which have the potential to affect the stress levels of dogs and cats in different ways (Kandel et al., 2020; Xiao et al., 2020).

This article embarks on a comprehensive exploration of the comparative stress responses in dogs and cats during the COVID-19 pandemic, focusing on four key elements: serum cortisol levels, total leukocyte counts, eosinophil counts, and behavioral changes. By examining these aspects, we aim to elucidate how the pandemic-induced limitations, including reduced outdoor activities, altered owner habits, and changes in social dynamics, have shaped the stress experiences of dogs and cats differently.

Cortisol, a steroid hormone produced by the adrenal glands, serves as a well-established marker of stress in both humans and animals. Stressful situations trigger the release of cortisol, often resulting in increased circulating levels (Rusu *et al.*, 2021). By monitoring cortisol levels, the physiological stress response of dogs and cats during the pandemic was assessed.

The immune system also plays a crucial role in the body's response to stress. During stressful events, the immune system can become activated, leading to changes in the counts of various immune cells, including leukocytes. An increase in total leukocyte counts can indicate an immune system response to stress. Eosinophils are involved in the body's immune response and can be affected by stress (Rusu *et al.*, 2021). Stress-induced changes in eosinophil counts can provide insights into potential immune modulation during the pandemic (Viena *et al.*, 2012).

Behavioral manifestations are often the most observable indicators of an animal's emotional well-being (Beerda *et al.*, 1999). Changes in behavior, including restlessness, hiding, vocalization, and altered social interactions, can reflect the emotional and psychological effects of stress (Amat *et al.*, 2015; Strickler and Shull, 2014).

The COVID-19 pandemic has placed dogs and cats at the intersection of a rapidly changing human world, necessitating a closer examination of their experiences and responses (Kandel *et al.*, 2020). It is crucial to recognize that dogs and cats are not only our companions but also sentient beings with unique needs and sensitivities (Archer, 1997). This article seeks to shed light on how these remarkable animals have adapted to the extraordinary circumstances of the pandemic, ultimately enhancing our understanding of the human-animal bond and the importance of safeguarding the well-being of our cherished pets.

MATERIAL AND METHODS

The study was conducted over a period of eight months between March-October 2020 on 34 dogs and 29 cats. The inclusion criteria considered clinically healthy animals, without any expressed pathology. Samples have been collected for biochemistry to determine serum cortisol levels and for hematology to assess any changes related to blood components.

Biochemistry was performed in an accredited laboratory, having been determined the serum cortisol levels. Blood samples were collected from the jugular vein or the saphenous veins (external for dogs and medial for cats) in vacutainers with clot activators inside using a sterile needle and syringe. The animals were handled with care and without causing excessive stress during the collection process. The collected blood samples were allowed to coagulate and then centrifuged to separate the serum from the blood cells.

Hematology was performed using Abaxis Vetscan HM5 analyzer determining total leukocytes and eosinophilic counts, as the main indicators of stress in animals. The blood was collected at the same time as biochemical samples, using EDTA vacutainers.

In addition, each owner filled in a questionnaire noting the main behavioral changes they observed in their pets.

The statistical interpretation was performed using MS Excel 2019, and Fisher's exact test.

RESULTS AND DISCUSSION

Stress is a complex physiological response to a perceived threat or challenge, and it is associated with several negative effects on the pets' physiology and behavior. Stress can suppress the immune system, making the animals more susceptible to disease (Malancus *et al.*, 2022).

One of the central findings of this study is the significant increase in serum cortisol levels observed in both dogs and cats during the COVID-19 pandemic as observed in table 1. For both dogs and cats. The results show very significant statistical changes in terms of serum cortisol values during pandemic for both species, compared to reference data (p<0,05).

Result —	Serum cortisol (µg/dl)				
Kesuit –	Dogs	Reference	Cats	Refernce	
March	6.63±0.27	1.00-4.00	7.11±0.39	1.70-5.00	
April	6.95±0.34	1.00-4.00	6.91±0.41	1.70-5.00	
May	7.01±0.33	1.00-4.00	7.01±0.37	1.70-5.00	
June	6.72±0.43	1.00-4.00	7.64 ± 0.87	1.70-5.00	
July	6.41±0.18	1.00-4.00	7.73±0.56	1.70-5.00	
August	6.55±0.20	1.00-4.00	8.13±0.84	1.70-5.00	
September	5.29±0.23	1.00-4.00	7.87±0.18	1.70-5.00	
October	5.44±0.21	1.00-4.00	7.92±0.65	1.70-5.00	
Average	6.32±0.38	1.00-4.00	7.56±0.53	1.70-5.00	

Table 1. Serum cortisol levels during pandemic in studied dogs and cats

This elevation in cortisol is indicative of heightened stress levels among these animals in response to the pandemic-induced alterations in their daily lives. Cortisol is a well-established biomarker of the stress response in both humans and animals, and its increased secretion is a physiological response to stressors. Our findings align with previous research highlighting the role of cortisol as a sensitive indicator of stress in companion animals (Beerda *et al.*, 1999; Salgirli Demirbas, 2023).

The increase in cortisol levels during the pandemic suggests that dogs and cats perceived the changes in their environment and daily routines as stressful. These changes included reduced outdoor activities, alterations in owner work habits, and shifts in social dynamics due to lockdowns and social distancing measures (Xiao *et al.*, 2020). The fact that both species exhibited this response underscores the universality of the pandemic's impact on companion animals' stress levels (Sipple *et al.*, 2021)

Furthermore, as highlighted in table 1, the temporal dynamics of cortisol secretion in dogs and cats during the pandemic revealed intriguing species-specific patterns. Dogs showed a more immediate cortisol response, with heightened levels observed early in the pandemic (Beerda et al., 1999). In contrast, cats exhibited a delayed cortisol response, with peak levels occurring later. These differences might be attributed to variations in the animals' perception of the stressors or their individual coping mechanisms (Westropp et al., 2006). The delayed cortisol response in cats may also reflect their more independent nature compared to the social and pack-oriented nature of dogs (Ratschen et al., 2020 Viena et al., 2012).

The study also demonstrated an increase in total leukocyte counts in both dogs and cats during the pandemic, signifying an enhanced immune system response to stressors. The elevation in leukocyte counts is consistent with the notion that stress can trigger an immune system response as the body prepares to defend against potential threats (Beerda *et al.*, 1999). As observed in table 2, the increase is due to elevated neutrophils, an indicator for a non-specific defense response. Dogs and cats both displayed this phenomenon, although the timing of the response varied.

Eosinophil counts, a component of the immune system, showed species-specific variations in response to pandemic-related stressors (Sapolsky, 2002). Dogs exhibited a significant decrease in eosinophil counts, that might indicate immune suppression in response to acute stress. Cats, on the other hand, displayed no significant change in eosinophil counts between the prepandemic and pandemic periods, suggesting a more stable immune response (Seksel, 2012).

The differential eosinophil responses in dogs and cats highlight the complexity of the immune system's reactions to stress. Dogs, as social animals, may experience more pronounced immune modulation in response to stressors, potentially making them more susceptible to fluctuations in immune cell counts (Salgirli Demirbas *et al.*, 2023). Cats, with their more solitary and self-reliant nature, may maintain a more stable immune response under similar circumstances (Westropp *et al.*, 2006; Sable, 1995).

Table 3 reveals that dogs exhibited a rapid increase in total leukocyte counts during the pandemic, suggesting an immediate immune system activation in response to the perceived stressors.

Table 2. Total leukocytes count and WBCs distribution (percentage) during pandemic in studied dogs and cats

Result	Dogs	Reference	Cats	Reference
Total WBCs	17.37±1.79	5-14.1	22.14±3.58	5.5-19.5
$(x10^{3}/mm^{3})$				
Neutrophils (%)	86.4±9.65	61-88	69.82±10.34	47-66
Eosinophils (%)	0.10 ± 0.11	0-9	0.40±0.16	0-4
Basophils (%)	0.20±0.10	0-1	0.14 ± 0.05	0-1
Monocytes (%)	6.3±1.21	2-10	3.87±0.45	0-5
Lymphocytes (%)	6.95±0.84	8-21	25.67±6.04	27-36

Table 3. Total leukocytes count variation during pandemic in studied dogs and cats

Result —	Total Leukocytes (x10 ³ /mm ³)				
	Dogs	Reference	Cats	Reference	
March	19.23±3.43	5-14.1	19.76±4.29	5.5-19.5	
April	20.85±4.15	5-14.1	20.91±3.55	5.5-19.5	
May	21.16±4.98	5-14.1	19.54±3.17	5.5-19.5	
June	19.76±3.17	5-14.1	20.24±2.07	5.5-19.5	
July	19.94±2.88	5-14.1	24.88±3.43	5.5-19.5	
August	15.83±0.98	5-14.1	22.43±2.14	5.5-19.5	
September	15.34±1.43	5-14.1	26.14±2.88	5.5-19.5	
October	15.98±1.31	5-14.1	24.13±3.35	5.5-19.5	
Average	18.22±2.16	5-14.1	22.28±2.93	5.5-19.5	

This observation aligns with the notion that dogs are highly attuned to their owners' emotional states and may respond more dynamically to changes in human behavior (Brooks *et al.*, 2018). In contrast, cats exhibited a delayed leukocyte response, with elevated counts occurring later in the pandemic. This delayed response might reflect the more self-sufficient nature of cats and their ability to cope with stressors more independently (Albrecht and Eichele, 2003; Amat *et al.*, 2015).

Behavioral manifestations are often the most observable indicators of an animal's emotional well-being (Ratschen et al., 2020; Friedmann, 1995). Changes in behavior, including restlessness, hiding, vocalization, and altered social interactions, can reflect the emotional and psychological effects of stress (Alho et al., 2016; Nibblett et al., 2015). The questionnaires filled in by the owners showed several changes during the pandemic with the most frequent ones represented by increased restlessness (55.9%) and attention-seeking behaviors (44.1%) in dogs. On the other hand, cats exhibited more pronounced hiding (55.2%), reduced grooming (48.3%), and increased territorial behavior (41.4%) during the pandemic.

CONCLUSION

The COVID-19 pandemic has brought about unique stress challenges for dogs and cats, with both species exhibiting increased cortisol levels and altered behaviors. The variation in eosinophil counts between the two species highlight their distinct immune responses to stressors during the pandemic. This study underscores the importance of recognizing the specific needs of dogs and cats during extraordinary circumstances and tailoring interventions to mitigate stress and promote their well-being. Further research is warranted to elucidate the underlying mechanisms driving these species-specific stress responses during the pandemic.

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