

HOW TO ASSESS STRESS IN REPTILES

Albert Martínez Silvestre, DVM, MSc, PhD, Dip. ECZM (Herpetology), Accred AVEPA (Animales Exóticos)

Abstract

Stress, as an adaptive response of any animal to a stimulus that presents a threat to homeostasis, can occur in reptiles. Yet, many veterinarians fail to recognize the signs of stress in reptiles. In this article, evaluation of reptile stress has been discussed with a focus on the analysis of behavioral changes, neuroendocrine correlation, and biochemical and physiologic effects in reptiles. If stress can be assessed and treated properly in captive reptiles, their health and well-being can be optimized. Copyright 2014 Elsevier Inc. All rights reserved.

Key words: animal welfare; captivity; neuroendocrine; reptiles; stress

Stress can be defined as a normal biologically adaptive response of an individual to internal or external stimuli representing a threat to homeostasis.¹ The following 4 main factors are found in response to stress: (1) behavioral changes, (2) alterations in the functioning of the autonomic nervous system, (3) neuroendocrine responses, and (4) immune responses. These 4 elements are correlated and coordinated by the hypothalamus and corticotrophin-releasing hormone. Knowledge of all the elements of stress that are delineated earlier can be useful in efforts to measure this disease condition in reptiles. Stress not only induces activation of the sympathetic nervous system but also directs a neuroendocrine response, initially slower but with potentially longer-lasting effects derived from activation of the hypothalamus/pituitary/adrenal (HPA) axis. This neuroendocrine axis is essential in the regulation of physiologic functions (e.g., immune competence, reproduction, metabolism, and behavior). Activation of the HPA axis is initiated when the corticotrophin-releasing hormone produced by the hypothalamus is released following a threat, stimulating the pituitary to secrete adrenocorticotrophic hormone (ACTH). The ACTH acts on the adrenal cortex to stimulate the production of glucocorticoids. The name suprarenal is not used in herpetology, as this gland is not present dorsal to the kidneys in these more primitive vertebrates. The adrenal gland does not undergo embryonic migration and remains very close to the renal vein and the gonad. The adrenal gland consists of 2 tissue types: tissue of mesodermal origin (steroidogenic interrenal cells) and those compounded with cells that are rich in fat. Functionally similar to cells of the mammals' adrenal cortex, secretion from the reptile adrenal gland consists mainly of corticosteroids. Chromatin tissue derived from the neural crest is colored intensely by histologic dyes because the cells have a high secretory component of both adrenaline and noradrenaline. This is the evolutionary equivalent to the adrenal medulla of mammals. However, unlike mammals, these cell types are mixed in a common matrix and do not form distinct regions. Although baseline cortisol levels are identifiable in blood, the main glucocorticoid in reptiles is corticosterone. Capture and events surrounding this process are known to be one of the main stressors in reptile species.² This fact should not be overlooked, but many other conditions affect corticosteroid release. Release of corticosteroids affects different reptile species over different periods; therefore, all undesirable effects of these internally produced compounds must be considered in captive reptile species (Table).

From the Centro de Recuperación de Anfibios y Reptiles de Catalunya (CRARC), Masquefa, Barcelona, Spain.

Address correspondence to: A. Martínez Silvestre, DVM, MSc, PhD, Dip. ECZM (Herpetology), Accred AVEPA (Animales Exóticos), Centro de Recuperación de Anfibios y Reptiles de Catalunya (CRARC), C/Santa Clara, s/n Masquefa 08783, Barcelona, Spain. E-mail: crarc@amasquefa.com.

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TABLE. Time to response (corticosterone release) after stressor

| Stressor | Time |
|------------------------------------|--------------|
| Capture | 3 min to 6 h |
| Exposure to cold and heat | 15 to 25 min |
| Hyperosmotic saline injection | 30 min |
| Intermittent change of environment | 30 min |
| Repeated blood sampling | 1 to 4 h |
| Severe restraint | 4 to 8 h |
| Hypotonic fluid injection | 8 d |
| Exposure to salt water | 1 to 4 wk |
| Hierarchy with dominating males | 10 to 30 d |
| Overcrowding | 10 to 14 d |
| Low relative humidity | 3 wk |

The time to response is deemed the time to stimulation of corticosteroid release.

STRESS BEHAVIORAL MEASUREMENT

Behavioral adaption is considered the optimal response by the animal to an external stressor. Avoiding a threat (i.e., derived from natural behavior of evading predators) is best and biologically requires the least effort to survive or avoid an unfavorable situation. When the individual is able to safely increase the distance from the stressor, the stress response does not usually affect the animal's long-term welfare. However, if the reptile is not able to respond in an appropriate manner, the stressor may cause negative effects on the animal's welfare, defined as *suffering*. For example, escape is not always possible, particularly for captive animals living in a confined area. The behavior shown by animals during the response to stress may provide a measurement of their inner or subjective state and may be useful as a measurement of the stress. If the stressor persists, the individual may show abnormal nonfunctional behaviors. These "displaced" behaviors may help the animal face the psychological aspect of the stressful event and also reduce the full effect of its physiologic response. Specific behavior parameters that are considered useful indicators of chronic stress include the following: aggression, anorexia, redirected activities, stereotypies, and displacement behaviors.³ Variations in the deposition of carotenes have been reported in stressed lizards, with a corresponding change in their normal skin colors.⁴

AUTONOMOUS NERVOUS MEASUREMENT OF STRESS

Once an animal perceives a stimulus as a threat, its brain releases a neuroendocrine response that consists of acute and chronic phases. The acute phase is regulated by the sympathetic nervous

system, which is immediately activated, is short acting, and has a direct effect on most body tissues. Catecholamines are released from the adrenal medulla seconds after the perception of a triggering stimulus, and their catabolism is rapid. The magnitude of this response can be determined by measuring serum catecholamine levels, blood glucose levels, heart rate, and blood pressure. However, determining these measurements may contribute to a stress response. Levels of ACTH, glucocorticoids, and blood glucose are physiologic parameters that can be measured to determine the level of stress in an individual animal. Hyperglycemia, increased hepatic glycogen, protein catabolism, and increased gluconeogenesis enzyme activity have also been identified in animals undergoing stressful events. However, certain studies show differing results as to the relationship of the physiologic parameters delineated earlier and stress. Moreover, there is no reasonable explanation to describe the significant effects of stress on blood glucose or hepatic glycogen levels. In part, these inconsistent results may represent the important correlation of low temperatures and seasonal variation on the reptiles' physiologic response. These variations are an excellent means to assess the magnitude of an acute response to stress but are not reliable when trying to determine a long-term response. This fact, together with the difficulties in measuring parameters such as heart rate under field conditions, makes the response of the autonomous nervous system less valuable in the assessment of stress in reptiles that live in a captive setting.⁵ Reptiles that are progressively and chronically stressed may be predisposed to obesity and hepatic lipidosis. Conversely, acutely or intensely stressed reptile species are predisposed to emaciation, immune depression, and reproductive difficulty.

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