FAECAL CORTISOL METABOLITES IN RESPONSE TO
MANAGEMENT PRACTICES AND DISTURBANCE IN
DIFFERENT HERBIVORES AND CARNIVORES
(CERATOTHERIUM SIMUM, DICEROS BICORNIS, ELEPHAS
MAXIMUS, LOXODONTA AFRICANA, OKAPIA JOHNSTONI,
RUPICAPRA RUPICAPRA AND LUTRA LUTRA)

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## **Abstract**

Methods for the measurement of cortisol/corticosterone metabolites in faecal samples have been developed during the past years. These methods are potential tools for evaluating changes in cortisol metabolite concentrations during various physiological conditions and in response to various stressors. Preliminary results of these investigations in animals mentioned in the title of this paper are reported.

## Zusammenfassung

In den vergangenen Jahren wurden Methoden für die Messung von Cortisol/Corticosterone Metaboliten in Kotproben entwickelt. Diese Methoden sind potentiell geeignet, Aussagen über Cortisolkonzentrationen sowohl unter physiologischen Bedingungen als auch unter stressbedingten Belastungen zu liefern. Ergebnisse über Untersuchungen bei den im Titel genannten Tierarten werden vorgestellt.

#### Résumé

Des méthodes de mesure des métabolites fécaux de cortisol/corticostérone ont été développées au cours de ces dernières années. Ces méthodes peuvent être utilisées pour évaluer les changements de concentration des métabolites de cortisol lors de différentes conditions physiologiques ainsi que dans des situations de stress. Les résultats préliminaires des analyses faites sur le animaux mentionnés dans le titre sont présentés dans ce rapport.

# Keywords

Stress, glucocorticoids, faeces, cortisol, metabolites, Ceratotherium simum, Diceros bicornis, Elephas maximus, Loxodonta Africana, Okapia johnstoni, Rupicapra rupicapra, Lutra lutra

#### Introduction

Stress has important implications for biological conservation and potential stressors for wild and zoo-animals come from environmental sources, husbandry and veterinary requirements (1,3). Although there is no standard definition and no single physiological measure of 'stress', a working definition is: 'stress is the cumulative response of an animal resulting from interaction with its environment via receptors'. Stress is normally a protective mechanism and an adaptive phenomenon, which in case of chronic stress can reduce fitness of the organism and ultimately can result in diseases (1). Responses to stressors are primarily coping with environmental changes in order to balance internal homeostasis. Responses include biochemical, physiological, mechanical reactions, which may result in numerous physiologic reactions, i.e. changes in heart rate, blood pressure, glucose, lactate, hemogram, haematocrit levels, etc. (1).

Responses are mediated by three different pathways: voluntary motor, the autonomic sympathetic nervous system and adrenal medulla (flight and fight reaction), and the neuroendocrine pathway. Stimulation of the hypothalamus-pituitary-adrenocortical axis (HPA axis) results in the release of glucocorticoids, which are considered good indices of stress in many vertebrates. Responses to stress vary depending on experiences, adaptations, hierarchial status, etc. (1) and plasma cortisol levels (and in turn faecal cortisol metabolites) show individual variability between animals.

#### Material and methods

Methods for the measurement of cortisol/corticosterone metabolites in faecal samples have been developed during the past years at the Institute of Biochemistry, Univ. of Vet. Med., Vienna by colleagues R. Palme and E. Möstl (5-8). This work has shown that cortisol in different species is extensively metabolised and part of these metabolites are excreted into the feces. Depending on the species crossreacting cortisol metabolites can be measured in an 11,17-dioxoandrostane (11-oxoetiocholanolone) enzyme immunoassay (6). In contrast to androgens, cortisol/corticosterone metabolites are characterised by an 11-oxo or 11-OH-group in the steroid molecule; therefore the 11,17-dioxoandrostane enzyme immunoassay shows only low crossreactivities with different androgens and thus is a reliable method for measuring cortisol metabolites. In addition to the 11,17-dioxoandrostane assay, EIAs for cortisol and corticosterone have been used to measure stress related increase of faecal cortisol metabolites in different zoo- and wildlife species (2,11; for detailed literature see reference lists in 2,3). These methods are potential tools for evaluating changes in cortisol metabolite concentrations during various physiological conditions and in response to stressors associated with management practices. Preliminary results of these investigations are reported in this paper.

#### Results

# White and black rhinoceroses (Ceratotherium simum and Diceros bicornis)

Translocation of white rhinoceroses is becoming a common management practise within the EEP (10). Samples collected before, during and after transport offer the opportunity for the validation of faecal cortisol metabolite measurements. This physiological approach was tested in 5 white and 1 black rhinoceros. Cortisol metabolites after transport increased only moderately in the 11,17-dioxoandrostane assay, whereas the cortisol or corticosterone assays were unsuitable. Although a small increase in the 11,17-dioxoandrostane assay was observed, these results suggest that for the measurement of faecal cortisol metabolites in rhinoceroses a more appropriate assay needs still to be developed.

# Asian and African elephants (Elephas maximus, Loxodonta africana)

Elephants belong to the most intensively managed animals in zoos. Training, transport and the state of musth in males are conditions, during which elevated cortisol levels are expected. Preliminary results indicate that the 11,17-dioxoandrostane assay is suitable to measure faecal cortisol metabolite concentrations in both species. Cortisol metabolites increased after transport, as well as during physical appearance of musth. A project to test cortisol metabolites in semi-free ranging working elephant bulls in Vietnam is currently under way

# Okapi (Okapia johnstoni)

Results on the measurement of faecal cortisol metabolites in the okapi (*O. johnstoni*) have been reported earlier (9). Adrenocortical activity in this species can by monitored noninvasively in faecal samples using the 11,17-dioxoandrostane assay, whereas both the cortisol and corticosterone assay were unsuitable. After transport of animals between European zoos, concentrations were about 5-10 times higher than initially, but values declined during the days following arrival. One female jumped through a glassed window 3 days after arrival and her cortisol metabolites sharply increased again following the jump through the window. Values then declined to a nadir. Also included in this study was one female with poor acceptance of the male; this animal displays submissive behaviour and lays down in lateral recumbency when a male approaches her. Cortisol metabolites in this female increased about 3-5 fold around oestrus periods when compared to luteal phases. The long acting neuroleptic Trilafon® (perphenazine enanthate) was tested, but so far was unsuccessful in reducing the anxiety related increase in cortisol levels (Bicher, pers. comm.). Since this animal is still at a potential breeding age, development of artificial insemination is considered.

# Chamois (Rupicapra rupicapra)

A group of free-ranging chamois (n=15-20) in the Nationalpark Berchtesgarden were observed during the Bavarian Open in paragliding and faecal samples were collected before, during and after this paragliding contest. Since individual samples could not be collected from these group living animals, a statistical approach was used. Each morning 20 pellets from 10 defaecation piles at the group's night resting site were collected. Intra sample variation between the pellets of a dung heap was low suggesting that samples were from one animal. Mean values of cortisol metabolites from the 10 defaecation piles were calculated each day. Results showed elevated cortisol metabolite values during the two days after the paragliding contest. This method will be used in further experiments in order to correlate cortisol levels with heart rates measured by a subcutanously implanted transmitter.

# European otter (Lutra lutra)

This study is part of a larger relocation program for European otters in Spain (4). Otters were live trapped in the South of Spain or in Portugal and transported to Barcelona zoo. After a period of 3 days of acclimatization animals were examined under anaesthesia and radio transmitters were implanted. Approximately 3 weeks after trapping animals were released into a new environment in the North of Spain. Cortisol metabolites could be measured using the 11,17-dioxoandrostane assay, the corticosterone assay and the cortisol assay; values were highest in the 11,17-dioxoandrostane assay. In general, concentrations showed considerable individual differences. Values increased during the first 3-4 days of sample collection and highest values were seen in response to clinical examination. Thereafter values declined until end of investigation period. Because of the individual variations the effect of Trilafon® on the levels of cortisol could not be determined incontestable.

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