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Seasonal changes in fecal testosterone concentrations and their relationship to the reproductive behavior, antler cycle and grouping patterns in free-ranging male Pampas deer (*Ozotoceros bezoarticus bezoarticus*)

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Abstract

The purpose of this study was to validate noninvasive endocrine monitoring techniques for Pampas deer and to evaluate seasonal changes in testicular steroidogenic activity and their correlation to reproductive behavior, antler cycle and group size. Thus, fecal samples, behavioral data and observations of antler status were collected at monthly intervals during 1 year from free-ranging Pampas deer stags (three radio-collared individuals and 15 random individuals) living in Emas National Park, Brazil (18°S latitude). Fecal steroids were extracted using 80% methanol and steroid concentrations were quantified by a commercial enzyme immunoassay (EIA). Fecal testosterone concentrations peaked in December–January (summer), March (early autumn) and in August–September (winter–spring), with minimal values from April–July. Reproductive behavior had two

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peaks, the first in December–January, characterized by predominately anogenital sniffing, flehmen, urine sniffing, chasing and mounting behavior, and the second peak in July–September (behavior primarily related to gland marking). There were significant correlations between fecal testosterone and reproductive behavior ($r = 0.490$), and between fecal testosterone and antler phases ($r = 0.239$). Antler casting and regrowth occurred under low testosterone concentrations, whereas velvet shedding was associated with high concentrations of testosterone. We inferred that Pampas deer stags exhibited a seasonal cycle that modulated sexual behavior and the antler cycle, and we concluded that fecal steroid analysis was a practical and reliable non-invasive method for the evaluation of the endocrine status of free-ranging Pampas deer.

Keywords: Pampas deer; Reproductive cycles; Testosterone; Feces; Antler cycle

1. Introduction

In most seasonally breeding mammals, several environmental cues (e.g., nutritional, physical and social factors) modulate the reproductive performance of an individual and act as indicators of the onset and cessation of sexual activity [1,2]. For the family Cervidae, almost all detailed reproductive knowledge in males has been derived from studies of temperate species that exhibit a distinct seasonal cycle of reproductive behavior and antler growth [3,4]. Photoperiod is responsible for the adaptation of these deer species to high latitudes, therefore breeding occurs during decreasing daylengths [3]. Although many species of deer live in tropical and subtropical regions (with comparatively minor annual changes in photoperiod), information about their reproductive biology and antler cycles is comparatively rare.

The Pampas deer (*Ozotoceros bezoarticus*) is a medium-sized cervid originally distributed throughout the grasslands of eastern South America (between 5° and 40°S latitude). Due to human activities, these ungulates are currently listed as endagered in Appendix 1 of the CITES and exist in small, isolated populations in Argentina, Bolivia, Brazil, Paraguay and Uruguay [5–7]. Information regarding Pampas deer reproduction has been reported mainly on females, which are polyoestrous, with estrous cycles approximately 21 days long and a 7 months gestation [7,8]. Studies in South America have shown that births in Ozotoceros are not strictly seasonal, but occurred more frequently from September to November in Argentina and Uruguay [6], and from August to November in Brazil [7]. In additional, Frädrich [9] reported that captive Pampas deer in the West Berlin zoo had no fixed rutting season. Nevertheless, males of this species exhibit a well-defined antler cycle, similar to temperate stags, with antler casting from April to May in the Brazilian Pantanal [10], June to August in Uruguay [6] and August to September in Argentina [11]. Therefore, studies involving assessment of hormones, behavior and antler cycle in stags are important to compare seasonal reproductive changes of this tropical species to temperate cervids.

The development of assisted reproductive techniques such as artificial insemination, in vitro fertilization and embryo transfer for non-domestic animals depend on the knowledge of their basic reproductive physiology, which normally requires the collection of repeated blood samples for hormonal evaluations. However, in many cervid species, these endocrine studies are not practical and even dangerous due to excessive stress caused by capture and