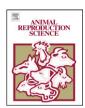


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Determination of semen characteristics and sperm cell ultrastructure of captive coatis (*Nasua nasua*) collected by electroejaculation

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ABSTRACT

Despite the wide geographical distribution of coati (*Nasua nasua*) from the south of Canada to the north of Argentina, studies regarding the reproductive characteristics of this species are extremely limited. The objective of this study was to describe the various characteristics of coati semen by morphometric and ultrastructural analysis. Five mature males were anesthetized and electroejaculated for the collection of semen. Semen was immediately evaluated for color, volume, pH, sperm motility, vigor, morphology, acrosomal integrity, percentage of live cells and hypo-osmotic response by light microscopy. Sperm cell morphometry and ultrastructural analyses were also performed. Observations of seminal characteristics determined by electroejaculation in captive coatis represent a valuable baseline dataset for establishing fertility standards and provide background information that may be useful for assisted breeding programmes in members of the Procyonidae family.

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1. Introduction

The coati (*Nasua nasua*) belongs to the Procyonidae family that originates from *Carnivora* order (Ferri et al., 2008). These animals compose an exclusive group distributed from the south of Canada to the north of Argentina (Guzmán-Lenis, 2004). Despite their wide geographical distribution, relative

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abundance and etiological interest, most of the data concerning this species are focused only on its behavior and ecology (Beisiegel, 2001).

The knowledge with regard to the reproductive characteristics of one species is important for the application of biotechniques seeking its preservation (Silva et al., 2004), or even for the control of its reproduction in the areas where they may be considered as pests (Asano et al., 2003). Although the coati is not an endangered species, information obtained for this animal could be applied to conservation programs for other carnivores such as white-nose coati (*Nasua narica*), South-Brazilian coati (*N. nasua solitaria*) and jupara (*Potos flavus*). The objective of this study is to describe the characteristics of the semen from captive coatis (*N. nasua*), including the morphometric and ultrastructural analysis of the sperm cell.

2. Materials and methods

2.1. Animals

Five mature male coatis (*N. nasua*) were used in this experiment that was conducted from February to October, 2007. They were aged between 2 and 8 years, weighing from 3.5 kg to 6.0 kg, and were kept under natural photoperiod. Four animals were housed in a social cage and belong to the Zoo-botanic Park Onélio Porto that is located in Mossoró, Brazil semi-arid region (5°10′S–37°10′W; temperature range, 27–28° C). The fifth animal is housed in an individual cage on the Ababy Institute of Conservation – EcoPoint, Fortaleza, located in the Brazilian coast (3°43′S–38°30′W; temperature range, 26–27°C). Animals were fed once a day with dog chow pellets and tropical fruits and had free access to water, which was removed from the cage 12 h before electroejaculation.

2.2. Anesthesia

The animals from Zoo-botanic Park were submitted to four sessions for the collection of semen, but the fifth animal was submitted to only two sessions. A total of 18 procedures were conducted and for each animal, 1-month interval was given between sessions. Animals were physically restrained and anesthetized with ketamine (10 mg/kg IM – Ketalar®, Pfizer®, Brazil) plus xylazine (1 mg/kg IM – Rompum®, Bayer®, Brazil) or tiletamine–zolazepam association (8 mg/kg IM – Zoletil®, Virbac®, Brazil). When necessary, 1/4 of the anesthetic dose was applied to keep the animal in a superficial anesthetic plane. Physiologic variables were monitored during all the procedures.

2.3. Collection of semen

Animals were kept in lateral recumbence and semen was collected by using an electroejaculator (Eletrojet®, Eletrovet, São Paulo, Brazil) connected to a 12 V source, following three stimulation cycles with a 5-min interval: the first cycle was composed by 10 stimuli of 20 mA, 30 mA and 40 mA, successively; the second cycle with 10 stimuli of 30 mA, 40 mA and 50 mA, successively; and the third cycle with 10 stimuli of 50 mA and 60 mA (Wildt et al., 1983). The electroejaculator probe measured 12.5 cm \times 1.0 cm; and 8 cm were inserted into the rectum of the coati. Semen was collected into plastic tubes and immediately evaluated.

2.4. Evaluation of semen

The volume was measured by using micropipettes varying from 5 to 200 μ l. Color of the semen was observed and pH was measured by using pH strips (Neutralit®, Merck®, Romania). Percentage of motile sperm and rate of sperm movement (vigor) were assessed immediately using light microscopy under $100\times$ and $400\times$ magnification. Bengal Rose stained smears were prepared with 5 μ l of diluted semen to evaluate sperm morphology and acrosomal integrity using light microscopy ($1000\times$), counting 200 cells per slide. Sperm morphologic abnormalities were classified as primary and secondary. Percentage of live spermatozoa was established by analyzing a slide stained with Brome-phenol Blue under light microscopy ($400\times$), counting 200 cells per slide. Following initial assessment, a $10\,\mu$ l semen aliquot

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