

***Eimeria auritanensis* N. SP. AND *E. gambai* CARINI, 1938 (APICOMPLEXA: EIMERIIDAE) FROM THE OPOSSUM *Didelphis aurita* WIED-NEWIED, 1826 (MARSUPIALIA: DIDELPHIDAE) FROM SOUTHEASTERN BRAZIL\***

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**ABSTRACT:-** TEIXEIRA, M.; RAUTA, P.D.; ALBUQUERQUE, G.R.; LOPES, C.W.G. *Eimeria auritanensis* n. sp. and *E. gambai* Carini, 1938 (Apicomplexa: Eimeriidae) from the opossum *Didelphis aurita* Wied-Newied, 1826 (Marsupialia: Didelphidae) from southeastern Brazil. [*Eimeria auritanensis* n. sp. e *Eimeria gambai* Carini, 1938 (Apicomplexa: Eimeriidae) do gambá *Didelphis aurita* Wied-Newied, 1826 (Marsupialia: Didelphidae) do sudeste brasileiro]. *Revista Brasileira de Parasitologia Veterinária*, v. 16, n. 2, p. 83-86, 2007. Curso de Pós-Graduação em Ciências Veterinárias, Universidade Federal Rural do Rio de Janeiro, Km 07 da BR-465, Seropédica, RJ 23890-000, Brasil. E-mail: lopescw@ufrj.br

Two eimerid species are described from the opossum *Didelphis aurita* from southeastern Brazil. *Eimeria auritanensis* n. sp. sporulated oocysts spherical to subspherical ( $31.55 \pm 1.56$  by  $29.55 \pm 1.40\mu\text{m}$ ), shape index 1.07; oocyst wall double layered  $2.10 \pm 0.27\mu\text{m}$  thick, outer yellowish and strongly ornamented having a prominently mammillated surface; inner layer smooth and brownish. There is no micropyle or oocyst residuum, but one or two polar granules are present. Sporocysts ovoid ( $13.20 \pm 1.64$  by  $10.41 \pm 1.10\mu\text{m}$ ) with a faint Stieda's body and residuum composed of granules and spherules. *Eimeria gambai* Carini, 1938 sporulated oocysts ovoid to subspherical ( $26.54 \pm 1.7$  by  $24.82 \pm 1.85\mu\text{m}$ ), shape index 1.07; oocyst wall double layered  $2.10 \pm 0.27\mu\text{m}$  thick, outer colorless to pale yellow entirely pitted, while inner layer smooth dark yellow to pale brown. Micropyle and residual bodies absent, polar granules present. Sporocysts ovoid  $12.49 \pm 1.75$  by  $9.32 \pm 1.01\mu\text{m}$ . Stieda body present and round. Sporocysts residuum composed of many granules and spherules.

**KEY WORDS:** *Didelphis aurita*, *Eimeria auritanensis* n. sp., *Eimeria gambai*, black-eared opossum.

**RESUMO**

Duas espécies do gênero *Eimeria* são descritas no gambá *Didelphis aurita* da Região Sudeste do Brasil. *Eimeria auritanensis* tem oocistos esporulados esféricos a subesféricos ( $31,55 \pm 1,56$  por  $29,55 \pm 1,40\mu\text{m}$ ), índice morfométrico 1,07; parede dupla com  $2,10 \pm 0,27\mu\text{m}$  de espessura, a mais externa amarelada e bastante ornamentada tendo uma superfície proeminentemente mamilar; camada interna lisa e acastanhada. Não apresenta micrópila ou resíduo do oocisto, mas um ou dois grânulos polares estão presentes. Os esporocistos são ovóides ( $13,20 \pm 1,64$  por  $10,41 \pm 1,10\mu\text{m}$ ) com um corpo de Stieda proeminente e resíduo composto de grânulos e esferas. *Eimeria gambai* Carini, 1938 tem oocistos esporulados ovóides a subesféricos ( $26,54 \pm 1,7$  por  $24,82 \pm 1,85\mu\text{m}$ ), com índice morfométrico de 1,07; parede dupla com  $2,10 \pm 0,27\mu\text{m}$  de espessura, externamente incolor ou um pouco amarelada e coberta de ondulações, enquanto internamente é lisa de coloração amarela escura a marrom. A micrópila e o corpo residual estão ausentes, mas grânulos polares estão presentes. Os esporocistos são ovóides com  $12,49 \pm 1,75$  por  $9,32 \pm 1,01\mu\text{m}$ . Um corpo de Stieda arredondado está presente. O resíduo do esporocisto é composto de vários grânulos e esferas.

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**PALAVRAS-CHAVE:** *Didelphis aurita*, *Eimeria auritanensis* n. sp., *Eimeria gambai*, gambá.

**INTRODUCTION**

The black-eared opossum *Didelphis aurita* is one of the commonest marsupials of the eastern Neotropical region of

Brazil with relative wide distribution, living in the tropical rain forest and peridomiliar areas (CERQUEIRA, 1985). They can be affected by a wide variety of parasites including protozoa such as eimerid parasites. The purpose of this study was to investigate the presence of coccidians parasites within the intestine of *D. aurita* from the State of Rio de Janeiro and then identify morphologically all species found.

## MATERIALS AND METHODS

Thirteen adult opossums were captured in three Municipalities of the State of Rio de Janeiro as Rio de Janeiro, Seropédica and Mangaratiba - license n° 96/02-RJ, IBAMA process n° 02022.008098/02-91. Feces were removed directly from the intestine, placed into plastic vials with 2.5% potassium dichromate ( $K_2Cr_2O_7$ ) (w/v) and transported to Laboratório de Coccídios and Coccidioses (PSA-EMBRAPA/UFRRJ), Departamento de Parasitologia Animal, Instituto de Veterinária, Universidade Federal Rural do Rio de Janeiro. Samples were processed and examined following methods described by Duszinsky and Wilber (1997). Thus, fecal materials were passed through double layered gauze and placed into Petri dishes with  $K_2Cr_2O_7$  2.5% at room temperature (20-24°C) permitting aeration. Sporulation time was evaluated daily during a week. Oocysts were recovered from  $K_2Cr_2O_7$  solution by centrifugation in Sheater's solution. Morphological observations, measurements and drawings were performed using a binocular Carl Zeiss microscope Jenapol with apochromatic oil immersion objective and ocular micrometer GF-P16X. A drop of sulfuric acid was added to the lamina to help with the mechanic rupture of membranes that facilitates visualization of inner structures. Photomicrographs were made with a digital camera Sony® MVC-CD250.

Statistical analysis was performed following Sampaio (2002) to calculate mean and standard deviation.

## RESULTS

### *Eimeria auritanensis* n. sp.

**Morphology:** Sporulated oocysts (Figures 1 and 2) are spherical to subspherical measuring  $31.55 \pm 1.56$  by  $29.55 \pm 1.40 \mu m$  (n=73), shape index 1.07. The wall is double layered  $2.10 \pm 0.27 \mu m$  thick, outer membrane is yellowish and strongly ornamented outside and having a prominently mammillated surface; inner layer is smooth and brownish. There is no micropyle or oocyst residuum, but one or two polar granules are often seen. Sporocysts (n=73) are ovoid measuring  $13.20 \pm 1.64$  by  $10.41 \pm 1.10 \mu m$  and they have a faint Stieda's body and residuum composed of granules and spherules.

**Sporulation time:** 8-9 days.

**Type host:** The black eared opossum *Didelphis aurita*.

**Site of infection in the host:** not investigated.

**Type locality:** Mangaratiba, Rio de Janeiro and Seropédica

**Geographic distribution of the host:** From northern Argentina to northeastern Brazil, commonly in the tropical rain forest.



Figure 1. Sporulated oocyst of *Eimeria auritanensis* n. sp. Sheatter's solution. (scale bar =  $10 \mu m$ ).

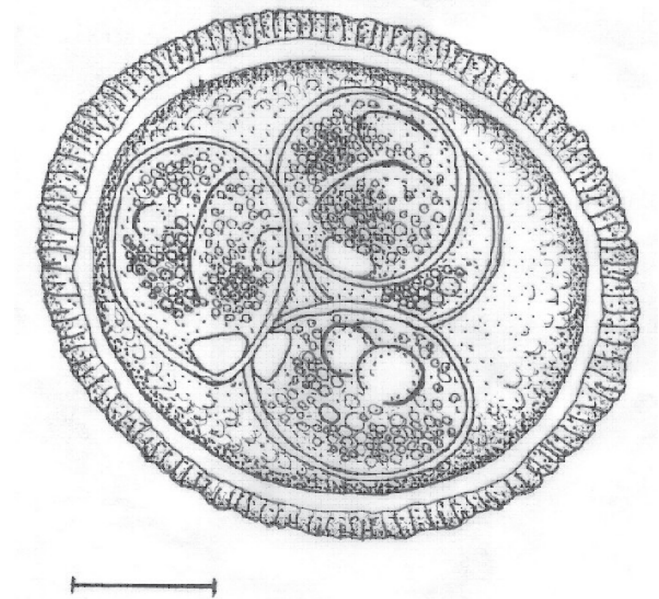


Figure 2. Line drawing of sporulated oocyst of *Eimeria auritanensis* n. sp. (scale bar =  $10 \mu m$ ).

**Etymology:** the specific name is derivate from the specific name of the vertebrate host.

**Type material:** oocysts in 10% formaldehyde-saline solution deposited at the Parasitology Collection, in the Department of Animal Parasitology, UFRRJ, Seropédica, Rio de Janeiro, Brazil. Repository number is P-012/2006, including phototypes and line drawings.

**Differential diagnosis:** Oocysts described above are different from oocysts of all species of *Eimeria* in the Didelphidae (Table 1). First, considering those species found

Table 1. Characteristics of oocysts from species of the genus *Eimeria*, parasites of Didelphidae.

Species	Characteristics									
	Host	Shape	Oocyst Size (µm)	Wallthick (µm)	Polar granule	Shape	Size (µm)	Stieda body	Refractile globule	Sporulation time (days)
<i>E. didelphydis</i> Carini, 1936	<i>Didelphis aurita</i>	spherical	16 x 16	smooth, colourless	absent	ovoid	12 x 10	present	absent	8
<i>E. haberfield</i> Carini, 1937	<i>Caluromys philander</i>	ovoid	30 x 20	brown-yellow, striated	absent	ovoid	13 x 8	present	absent	6
<i>E. gambai</i> Carini, 1938	<i>Didelphis aurita</i>	ovoid	23-28 x 18-22	~2, brown-yellow, striated	absent	ovoid	12 x 10	present	absent	6-7
<i>E. indianensis</i> Joseph, 1974	<i>Didelphis virginiana</i>	spherical to subspherical	16.3 x 16.3 or 17.6 x 16.4	~1.5, double layered, yellow, striated, rough and pitted	present	ovoid	9.1 x 6.1	present	absent	10
<i>E. caluromydis</i> Laison & Shaw, 1989	<i>Caluromys philander</i>	spherical to subspherical	31.83 x 31.15	~3.21, one layer, brown-yellow, striated	absent	ovoid	14.75 x 9.69	present	-	14
<i>E. philanderi</i> Laison & Shaw, 1989	<i>Philander opossum</i>	spherical to subspherical	23.5 x 22.31	~1.88, double layered, inner brown-yellow, striated, outer colourless, striated	present	ellipsoidal	11.35 x 8.13	present	absent	5
<i>E. cochabambensis</i> Heckscher, Wickesberg, Duzynski, and Gardner, 1999	<i>Marmosops dorothea</i>	subspherical	21.6 x 20.2	~2.0, double layered, yellow, slightly striated,	present	fusiform	11.0 x 7.2	present	present	-
<i>E. marmosopos</i> Heckscher, Wickesberg, Duzynski, and Gardner, 1999	<i>Marmosops dorothea</i>	subspherical	22.2 x 19.9	~2.2, one layer, rough, thickstriated	present	ovoid	11.1 x 6.8	present	present	-
<i>E. micouri</i> Heckscher, Wickesberg, Duzynski, and Gardner, 1999	<i>Micoureou constantaiae</i>	ellipsoidal	24.6 x 18.2	~1.6, double layered, outer pitted, inner transparent	present	fusiform	11.5 x 6.7	present	present	-
<i>E. rugosa</i> Present Work	<i>Didelphis aurita</i>	subspherical	31.55 x 29.55	double layered, striated, brown-yellow, outer pitted	present	ovoid	13.2 x 10.41	present	-	6-8
<i>E. gambai</i> Present Work	<i>Didelphis aurita aurita</i>	ovoid	26.46 x 24.58	double layered, yellow, slightly striated	present	ovoid	12.15 x 9.18	present	-	8-10

in *D. aurita*, measurements of oocysts are long different regarding dimensions of *E. gambai* or *E. didelphydis* and also morphology is very distinct. The outer layer of *E. auritanensis* is darker and much more ornamented than outer layer of *E. gambai*, and *E. didelphydis* has a smooth and single membrane. *Eimeria haberfield* (CARINI, 1937) from *Caluromys philander* has a less striated and single outer layer and also the author related failure in the experimental cross infection in *D. aurita*. Other, *E. indianensis* (JOSEPH, 1974) from the Northern American opossum *D. virginiana* has different dimensions from the present description and different format (spherical to subspherical). Although *E. philanderi* (LAINSON; SHAW, 1989) from *Philander opossum*

has the same oocyst format and double layered wall it has striations in both inner or outer layer plus measurements very distinct. Both, *E. cochabambensis*, *E. micouri* and *E. marmosopos* (HECKSCHER et al., 1999) are different in any aspect of the description above, excepting for the presence of polar granules. Finally, *E. caluromydis* (LAISON; SHAW, 1989) from *C. philander* has much bigger oocysts and thicker membrane, and also *E. auritanensis* has polar granules and a faint Stieda body not present in *E. caluromydis*.

*Eimeria gambai* Carini, 1938

**Morphology:** Sporulated oocysts (Figures 3 and 4) are ovoid measuring  $26.54 \pm 1.7$  by  $24.82 \pm 1.85 \mu\text{m}$  (n=73), shape





Figure 3. Sporulated oocyst of *Eimeria gambai*. Sheatter's solution. (scale bar = 10µm).

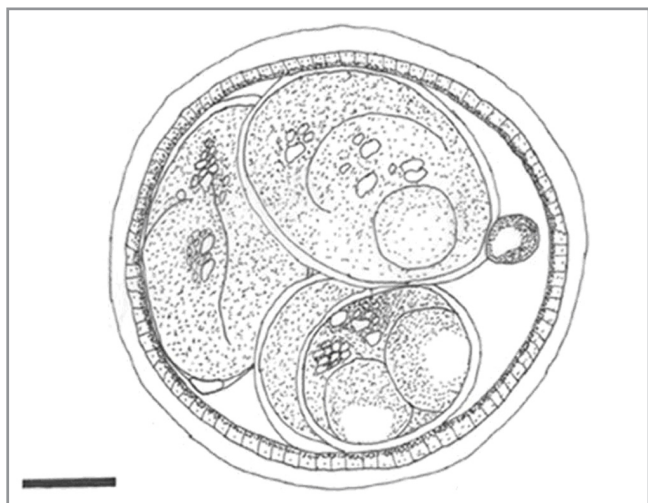


Figure 4. Line drawing of sporulated oocyst of *Eimeria gambai* Carini, 1938 (scale bar = 10µm).

index 1.07. The wall is double layered with  $2.10 \pm 0.27\mu\text{m}$  thick, outer colorless to pale yellow but entirely pitted, while inner layer is smooth with a color ranging from dark yellow to pale brown. Micropyle and residual bodies are absent, whereas polar granules are often present. Sporocysts are ovoid to subspherical ( $n=73$ ) measuring  $12.49 \pm 1.75$  by  $32 \pm 1.01\mu\text{m}$ . The Stieda body is present, round but of difficult visualization. There is a sporocysts residuum composed of many granules and spherules.

*Sporulation time:* 8-10 days.

*Type host:* The black eared opossum *Didelphis aurita*.

*Differential diagnosis:* There are two species of the genus *Eimeria* from the black eared opossum *D. aurita* in the world and both were originally described in Brazil. They are *E.*

*didelphydis* (CARINI, 1936) and *E. gambai* (CARINI, 1938). According to this author oocysts of such species could be differentiated by its measurements and characterization of the wall. Thus, *E. gambai* have larger oocysts with a single layer wall, thick and pitted, while *E. didelphydis* have smaller oocysts with a double layered wall smooth and less thick. Also, the author related possible variations in the color, presence of small dark points (pitted) and thickness of the wall of oocysts in *E. gambai*. In the present study such oocyst with characteristics of *E. didelphydis* were not found. Differently, oocysts of *E. gambai* were widely found in the material of all marsupials. Anyway, after a critical observation in the morphology of oocysts it was found characteristics not featured by Carini (1938). One is the presence of two layers in the wall observed after treatment with  $\text{H}_2\text{SO}_4$ , besides the presence of one or two polar granules not described by the author. Although these differences in the observation of *E. gambai* we do not considered the oocysts found in this study as new species of *Eimeria*.

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