THE JAPANENSE QUAIL (*Corturnix japonica*): A NEW INTERMEDIATED HOST FOR *Cystoisospora felis* (WENYON, 1923) FRENKEL, 1977 (APICOMPLEXA: CYSTOISOSPORINAE)*

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ABSTRACT. Rodrigues J. da S., Meireles G.S. de, Flausino W. & Lopes C.W.G. **The japanense quail (*Corturnix japonica*): a new intermediated host for *Cystoisospora felis* (Wenyon, 1923) Frenkel, 1977 (Apicomplexa: Cystoisosporinae).** [A codorna japonesa (*Coturnix japonica*): um novo hospedeiro para *Cystoisospora felis* (Wenyon 1923) Frenkel 1977 (Apicomplexa: Cystoisosporinae)]. *Revista Brasileira de Medicina Veterinária, 34(1):14-18, 2012.* Curso de Pós-Graduação em Ciências Veterinárias, Instituto de Veterinária, Universidade Federal Rural do Rio de Janeiro, BR 465 km 7, Seropédica, RJ 23890-000, Brasil. E-mail: lopescwg@ufrrj.br

Cystoisospora felis is an obligatory intracellular parasite that infects several species of felines. Infection is transmitted by either ingesting sporulated oocysts directly, or consuming an intermediated host harboring monozoic cysts. In this study, 10 Japanese quails (*Corturnix japonica*) were infected with a pure inoculum of *C. felis* (10^6 sporulated oocysts/ml); after 60 days post infection, liver, spleen, and cloacal bursa were removed from each quail and were separately fed to 3 kittens. A forth kitten was infected with 10^6 sporulated oocysts/ml orally. Fecal samples were collected from each kitten daily and evaluated for the presence of oocysts; the percent sporulation of resulting oocysts was calculated daily. In addition, 50 sporulated oocysts from each infected individual were measured in µm, and evaluated for length, width and shape index. Kitten fed on liver had an average oocyst length of 44.30 (39.53 – 48.84), width of 31.30 (27.44 – 36.05) and shape index of 1.40 (1.16 – 1.58); while the kitten fed on spleen had an average length of 46.30 (41.40– 50.71), width of 32.90 (29.30 – 37.20) and shape índex of 1.40 (1.22 – 1.71). The kitten fed on cloacal bursa had an average length of 44.80 (40.00 – 49.31), width of 31.20 (27.91 – 36.5) and shape index of 1.40 (1.18 – 1.60); and the kitten infected with sporulated oocysts orally had an average length of 43.60 (40.30 – 47.58), width of 30.80 (26.66 – 34.22) and shape index of 1.40 (1.23 – 1.58). Furthermore the prepatent and patent periods were determined for C. *felis* in the quail that serves as an experimental model for working in experimental conditions.

KEY WORDS. *Cystoisospora felis*, Japanese quail, experimental infection.

RESUMO. *Cystoisospora felis* é um parasito intracelular obrigatório que acomete felídeos de diversas espécies, e podem se contaminar tanto pela ingestão de oocistos esporulados, quanto pela ingestão de tecidos de hospedeiros intermediários infectados previamente com oocistos esporulados. Neste es-

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tudo, 10 codornas japonesas foram infectadas com um inóculo puro de oocistos de *C. felis,* na concentração de 10^6 e, após 60 dias, o baço, o fígado e a bursa cloacal dessas codornas infectadas foram oferecidos a três filhotes de gato livres de infecção. Um quarto gato ainda foi infectado com uma suspensão de 10^6 oocistos viáveis de *C. felis* por via oral. Após o início da eliminação, os oocistos foram colocados para esporular e, em seguida, 50 oocistos oriundos de cada infecção foram mensurados em µm, apresentado em média diâmetro maior (DM), diâmetro menor (dm) e índice morfométrico com os seguintes valores: 44,30 (39,53 – 48,84), 31.30 (27,44 – 36,05) e 1,40 (1,16 – 1,58) para os oriundos do animal infectado com fígado, 46,30±2,20, 32,90±1,90 e 1,40±0,1 para os oocistos do animal infectado com baço; 44,80 (40,00 – 49,31), 31,20 (27,91 – 36,5) e 1,40 (1,.18 – 1,60), para os oocistos oriundos do animal infectado com bursa cloacal; 43,60 (40,30 – 47,58), 30,80 (26,66 – 34,22) e 1,40 para os oocistos do animal que recebeu oocistos direto por via oral. Com estes resultados foi possível concluir que não existem diferenças significativas na morfometria dos oocistos de *C. felis*. Além disso, os períodos, pré-patente e patente foram semelhantes quando gatos foram alimentados com vísceras de codornas infectadas previamente com oocistos esporulados de *C. felis* em comparação com o animal que recebeu 10^6 oocistos esporulados por via oral.

PALAVRAS-CHAVE. *Cystoisospora felis*, codorna japonesa, infecção experimental.

INTRODUCTION

Cystoisospora felis (Wenyon, 1923) Frenkel 1977, an obligatory-intracellular parasite of the Sarcocystidae family (Cystoisosporinae subfamily), is one of the coccidium more frequently found in the feces of domestic cats (Amaral et al. 1966). *C. felis* oocysts are easily distinguished from other feline-feces coccidia by their large size (Frenkel & Dubey 1972).

Dubey & Frenkel (1972) identified two possible forms of *Cystoisospora* transmission in felines: first, by directly ingesting sporulated oocysts; or second by consuming an intermediate host infected previously with sporulated oocysts. A wide range of animals have been described as intermediate hosts serving as vectors for *Cystoisospora* species: including, mice, rats, and dogs (Frenkel & Dubey 1972); birds (Lindsay & Blagburn 1994); bovines (Fayer & Frenkel 1979); swine (Carvalho Filho et al. 2003); rabbits (Costa & Lopes 1998); Mongolian gerbils (Carvalho Filho et al. 2004); and chickens (Massad et al. 2003).

The systemic distribution of hipnozoites in the viscera of different intermediate hosts was indicated by an accentuate tropism for mesenteric lymph nodes, spleen, liver and Payers' patches in mammals (Frenkel & Dubey 1972, Brösigke et al. 1982, Freire & Lopes 1996, Costa & Lopes 1997).

The present study evaluates the levels of infection obtained when healthy kittens are exposed to different types Japanese quail viscera which have been infected with *C. felis* and compares it to healthy kittens directly exposed to *C. felis*.

MATERIAL AND METHODS

The current study was carried out at the Laboratório de Coccidios e Coccidioses (LCC) – Projeto Sanidade Animal (Embrapa/UFRRJ), Departamento de Parasitologia Animal, Instituto de Veterinária, UFRRJ.

A healthy cat, in the final stage of gestation was brought to the LCC and it was lodged in individual bay with water and food *ad libitum*. An examination of the feces was performed to determine if the female cat was free from gastrointestinal-parasite infection. In addition, a preventive treatment for coccidia was adopted as suggested by Loss & Lopes (1997).

Kittens were separated from their mother 45 days after parturition; fecal examinations were than preformed for 30 days to ensure that kittens were free from coccidia infection. Kittens were then placed separately in suspended cages, which were previously disinfected with sodium hypochlorite and flame torch to prevent possible contaminations.

To evaluate kitten infection via infected viscera, ten Japanese quails (*Corturnix japonica*) were inoculated, using an orogastric tube, with *C. felis* suspension of 10^6 sporulated oocysts/mL (Figure 1). Sixty days after inoculation the Japanese quails were euthanized (Cobea 2007) and their livers, spleens and cloacal bursa were separated. A pure suspension of *C. felis* sporulated oocysts (10^6 oocysts/mL) in Hank' solution (Andrade 2000) was also prepared to evaluate transmission direct infection. *C. felis* sporulated oocysts were obtained from the purification of fecal samples from naturally infected cats.

A total of 4 kittens were used in this study; each received orally a distinct source of infection: cat I –

Figure 1. *Cystoisospora felis* sporulated oocyst. Sugar saturated solution. 1000X.

ground spleen, cat II – ground liver, cat III – ground cloacal bursa, and cat IV - suspension of 10^6 sporulated oocysts/mL. Beginning the day after inoculation, fecal samples were examined daily for infection as described by Figueiredo et al. (1984). For Oocyst sporulation, fecal samples were collected and diluted separately, and stored in plastic vials containing potassium dichromate 2.5% in water solution in a proportion ratio of 1:9 (v/v), which was placed under forced aeration using a aquarium pump and at ~22^0C.

Oocysts were checked daily for sporulation and classified as either, sporulated or non-sporulated. Once 80% of a sample was determined to have sporulated a morphologic analysis and characterization of the oocysts was carried out; 50 sporulated oocysts from each source of infection were measured separately using a micrometric ocular (K-15X PZO) in a binocular microscope (Carl Zeiss). For each oocyst the length and width were determined, as well as the index shape. Pictures were taken using a digital camera model CD Mavica MVC-CD250 Sony®.

During the experimental infections, prepatent and patent periods were observed for determining if there were differences in the source of hipnozoites.

In addition, the means of sporulated oocysts from different sources of infection was statistically compared according to Sampaio (2002).

RESULTS AND DISCUSSION

By comparing the measurements of the length, width and index shape of oocysts recovered from the various sources of infection (Table 1), significant differences between these values were not observed. These results differ from those observed by Medeiros et al. (2007); they found that oocysts of *C. felis* proceeding from mice viscera were larger than those transmitted naturally. Although, the length and width means observed from each source of infection in this study were similar to those observed by Medeiros et al. (2007). Conversely the measurements observed in the present study for oocysts transmitted via infected quail viscera were larger than those observed after transmission by Mongolian gerbil viscera (Carvalho Filho et al. 2004). However the index shape of the oocysts was similar in both studies.

Prepatent and patent periods were the same among kittens exposed to infected viscera and similar to those exposed directly to sporulated oocysts (Table 2). Analogous results were observed by Dubey & Streitel (1976) and Carvalho Filho et al. (2004). Lindsay & Blagburn (1994) described a patent period from day10-11 for *C. felis*; for cats free of coccidia were infected by 10^4 sporulated oocysts orally. Kittens fed on liver or spleen shed more

Table 1. *Cystoisospora felis* sporulated oocysts from different sources of infection.

Source of infection	Oocysts (µm) (n=50) *		Shape index
	Length	Width	
Liver	44.30 (39.53-48.84)	31.30 (27.44-36.05)	1.40 (1.16-1.58)
Spleen	46.30 (41.40-50.71)	32.90 (29.30-37.20)	1.40 (1.22-1.71)
Cloacal bursa	44.80 (40.00-49.31)	31.20 (27.91-36.5)	1.40 (1.18-1.60)
Sporulated oocysts	43.60 (40.30-47.58)	30.80 (26.66-34.22)	1.40 (1.23-1.58)

* No significant differences were observed.

Table 2. The shedding of *Cystoisospora felis* oocysts by cats infected from different sources.

Origin of infecting material	Source of infection	Period		Number of oocysts counts	
		Pre-patent	Patent	OoPG[a]	Total fecal volume
Japanese quail[b]	Spleen	5	12	3,450	57,740
Liver	5	12	4,880	216,522	
Cloacal Bursa	5	12	445	19,924	
Sporulated oocysts[c]	Fecal oocysts	7	14	31,274	1,565,948

[a] Oocysts per gram of feces
[b] Infected with 10^6 sporulated oocysts/mL previously.
[c] Kittens infected with 10^6 sporulated oocysts/mL orally.

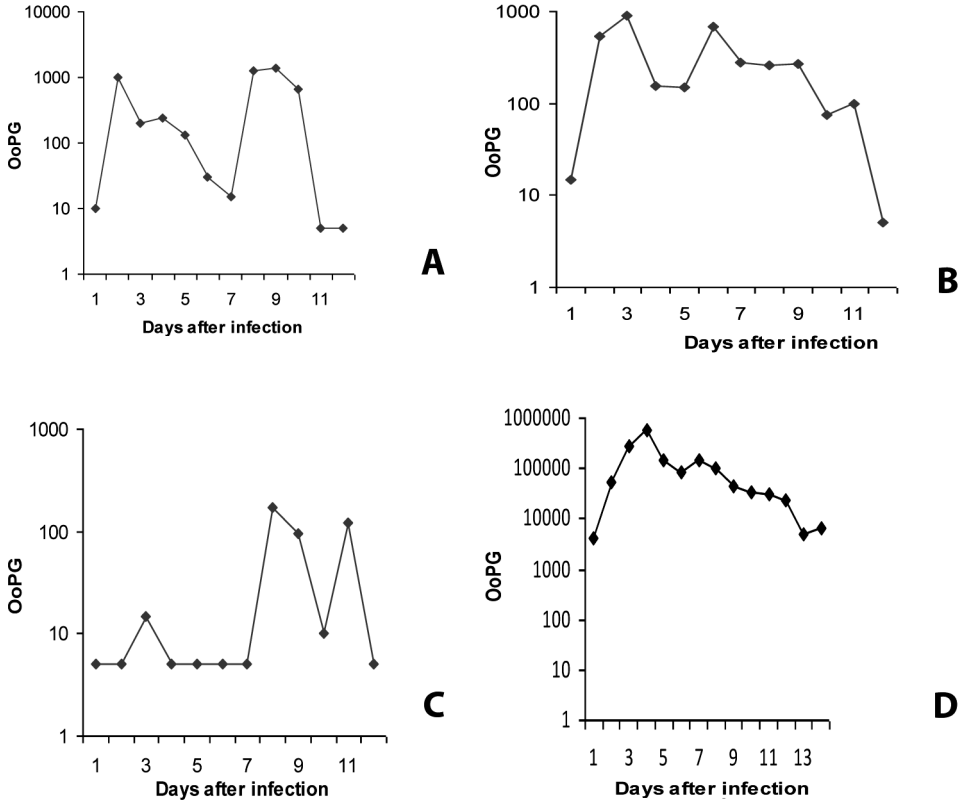

Figure 2. Shedding of *Cystoisospora felis* oocysts by cats fed on, (a) liver, (b) spleen, (c) cloacal bursa of the Japanese quail, and (d) infected with 10^6 sporulated oocysts orally

oocysts than that fed on cloacal bursa, but less than the kitten infected orally (Figure 2).

The data observed in this study was compatible to *Cystoisospora felis* infection, besides it the Japanese quail was considered as a new intermediated host; besides it Japanese quail should be considered as a good intermediated host for biological experimental infection.

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