



Coccidiosis in Japanese Quails (*Coturnix japonica*): Characterization of a Naturally Occurring Infection in a Commercial Rearing Farm¹

■ Author(s)

Teixeira M²
Teixeira Filho WL³
Lopes CWG³

² Curso de Pós-Graduação em Ciências Veterinárias, UFRRJ.

³ Departamento de Parasitologia Animal, Instituto de Veterinária, UFRRJ.

■ Mail Address

Marcel Teixeira
Laboratório de Coccídios e Coccidioses
Projeto Sanidade Animal/Embrapa
Br 465, km 07
23.890-000 - Seropédica, RJ.
Fone 55 21 2682 2940 (ramal 34)

E-mail: teixeira@ufrj.br

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ABSTRACT

A study about coccidiosis in Japanese quails was carried out in order to identify species of the genus *Eimeria* and characterize a naturally occurring infection in a commercial rearing farm. For this purpose, fecal exams, oocyst counting and morphological study were performed, besides necropsy and histopathology to confirm diagnosis. Three species of the genus *Eimeria* were found and identified as *E. tsunodai*, *E. uzura* and *E. bateri*. The natural infection was characterized as subclinical because of the mild and nonspecific clinical signs. Nevertheless, coccidiosis was considered an important disease because endogenous stages of the parasites and a high number of oocysts in feces were associated with intestinal lesions. The results suggest that such infection might represent a limiting factor to this branch of the modern poultry industry.

INTRODUCTION

Quail production can be considered a branch of the modern poultry industry. Similar to the majority of animal production systems, it demands constant improvements on the application of new technologies and sanitary control. However, most studies have been published on Japanese quail nutrition (Garcia *et al.*, 2002) and few have been done concerning quail diseases (Bigland *et al.*, 1964). Among the avian diseases, coccidiosis affect bird development as well as production. Since quails are reared mostly in battery, there is no report in the literature on this disease, especially in this country.

Thus, this study presents an evaluation of coccidiosis in naturally infected Japanese quails from a commercial rearing farm.

MATERIAL AND METHODS

Birds

Japanese quails were reared in a farm located in the municipality of Seropédica, Rio de Janeiro, Brazil. There were approximately 27,000 birds in the farm, including young male and female quails (from 1 to 35 days of age) and mature females (above 35 days of age). Young birds were housed on litter bed and mature birds in cages for egg production.

Fecal samples

Pools of feces were collected from young and mature Japanese quails. A hundred young birds were placed into a plastic box (1.5 x 1.0m) for half an hour and feces samples were collected from the box. In the case of adult birds, feces were collected directly from the cages. Samples were placed into plastic bags and later processed at Laboratório de Coccídios e Coccidioses, Universidade Federal Rural do Rio de Janeiro.



Preparation of oocysts

Samples were diluted into 2.5% aqueous potassium dichromate ($K_2Cr_2O_7$) and kept in Petri dishes for sporulation at room temperature. After sporulation, oocysts were recovered by centrifugation with saturated sugar solution as described by Duszynski & Wilber (1997) and used in subsequent analysis.

Morphological diagnosis and oocyst counting

A Carl Zeiss binocular microscope with immersion objective (100x) and a K-15x PZO (Poland) micrometer was used for oocyst identification. The number of oocysts per gram of feces (OoPg) was determined according to the technique described by Menezes & Lopes (1995).

Necropsy

Two birds were necropsied at the 3rd, 5th, 7th, 14th, 21st, 28th, 35th and 42nd days after birth. Euthanasia was accomplished following Brazilian guidelines (Resolução n.º 714, 20th June, 2002, Conselho Federal de Medicina Veterinária). During necropsy, parts of the small intestine (jejunum and ileum) and caeca were taken for histopathology.

Histological examination

Histological examination was carried out to confirm the presence of developing stages of parasites within the intestine. Sections of 2 cm were excised from the small intestine and caeca of each bird, fixed in 10% formaldehyde solution, processed and stained by hematoxylin-eosin or periodic acid Schiff (PAS), according to Behmer *et al.* (1976). For sequential examinations, a triocular JENAMED, Carl Zeiss-Jena microscope with 40x objective was used.

Statistical analysis

Oocysts measurements were analyzed using the software Excel (Microsoft®) for averages and standard deviation.

Photographs

Pictures were made using a digital camera CD Mavica MVC-CD250 Sony®, and a photographic camera f-KAS Automatic-2 with film ISO 100 (21 DIN) (Kodak, Mexico).

RESULTS AND DISCUSSION

Species of the genus *Eimeria*

Three species of the genus *Eimeria* (Figure 1, A, B and C) previously reported affecting Japanese quails

(Teixeira & Lopes, 2002) were identified in the present study, as follows:

a. *Eimeria tsunodai* Tsutsumi, 1972: Sporulated oocysts were ovoid, measuring 20.2 ± 1.5 by 14.88 ± 0.79 μm and 1.36 shape index. Oocyst wall was smooth, double layered, with brownish inner layer and colorless outer layer, and measured 0.99 ± 0.1 μm . Despite usually only one polar granule was present, it could appear in pairs and refractive. Micropyle and residual body of the oocyst were absent. The sporocysts varied from ovoid to ellipsoid and measured 10.41 ± 0.6 by 5.39 ± 0.3 μm . They had a finer end where a small and faint Stieda body projected. The residual body of the sporocyst was present and dispersed among the sporozoites, which were in pairs with globules visible at the enlarged extremity.

b. *Eimeria uzura* Tsunoda & Muraki, 1971: Sporulated oocysts were ovoid, measuring 22.2 ± 6.76 by 16.16 ± 1.13 μm , and 1.32 shape index. Oocyst wall was smooth, double layered, with brownish inner layer and colorless outer layer, measuring 1.08 ± 0.12 μm . Two to five polar granules were observed in the oocysts, sometimes with a massive aspect, but not refractive). The micropyle and residual body of the oocysts were absent. The sporocysts were ovoid measuring 11.76 ± 0.74 by 5.94 ± 0.45 μm , with a finer end, where a piriform Stieda body projected. The residual body of the sporocysts was present and had concentric granules between the sporozoites, which had refractive globules at the enlarged extremity.

c. *Eimeria bateri* Bathia, Pandey & Pande, 1965: Sporulated oocysts were subspherical, ovoid or ellipsoid, measuring 21.50 ± 1.84 by 16.36 ± 1.32 μm and shape index 1.32. Oocyst wall was smooth, double layered, with brownish inner layer and colorless outer layer, measuring 0.99 ± 0.1 μm . A single and refractive polar granule was present, but micropyle and the residual body of the oocyst were absent. Sporocysts were ovoid and measured 10.35 ± 0.73 by 6.66 ± 0.58 μm , with a prominent knob-like Stieda body. The residual body of the sporocyst was dispersed among the sporozoites, which had refractive globules at the enlarged extremity.

Descriptions of *Eimeria* date from the beginning of the last century, and ever since means for an appropriate characterization and identification of the species have been discussed. Several parameters can be used (Long & Joyner, 1984) and new methods have



been developed (Kucera & Reznicky, 1991; Dausgiesch *et al.*, 1999; Gruber & Fernandez, 1999; Pereira *et al.*, 2001). However, morphological differentiation associated to specificity factors is still useful. Duszynski & Wilber (1997) emphasized and encouraged precision in the description of species, and established basic characteristics for an appropriate description of the oocysts. However, accuracy and caution were already recommended by Bandoni & Duszynski (1988). In the present study, species previously described were found, and the final identification was carried out by comparison with the original descriptions of the parasites. Some *Eimeria* species with similar morphology were initially excluded because of the differences in the size of the oocysts. The measurements, in the beginning of the investigation, the measurements allowed to distinguish from closely related species. Initially, *E. coturnicis* (Chakravarty & Kar, 1947), *E. tahamensis* (Amoudi, 1987) and *E. fluminensis* (Teixeira & Lopes, 2002) were excluded. Despite the similar measurements, *E. taldykurganica* (Svambaev & Utebaeva, 1973) did not fit with the descriptions because it has a very different form, essentially ellipsoid with shape index 1.86. Lastly, *E. dispersa* (Tyzzer, 1929) may infect *C. coturnix* but is not infective to *C. japonica* (Tsunoda & Muraki, 1971) and therefore was also dismissed. On the other hand, *E. tsunodai*, *E. uzura* and *E. bateri* were identified in the present study based on the measurements and other features of oocysts, such as polar granules, Stieda body and refractive globules, which were essential to confirm identification.

Clinical signs and Gross lesions

During the first week of life, few young quails presented diarrhea, weakness and small blood spots in the upper small intestine (jejunum and ileum). Later, softening of feces at the 14th day and an increased cecum were seen in quails necropsied at the 21st days. From the 35th to the 42nd days, a significant number of quails had diarrhea, which disappeared soon.

There are few reports about the pathogenicity of *Eimeria* in quails. Mazurkiewiewicz *et al.* (1967) reported clinical signs such as lack of appetite, ruffled feathers, uncoordinated movements, inhibition of laying and loss of weight in naturally infected young and mature quails reared at the laboratory. Norton & Pierce (1971) infected young Japanese quails experimentally with *E. bateri* and observed mild loss of weight and, although anorexia and softening of feces were observed at the third day of infection, the disease was

considered mild and easy to overcome. Tsunoda & Muraki (1971) also reported low pathogenicity in Japanese quails experimentally infected with 1×10^5 oocysts of *E. uzura*, observing diarrhea and anemia from the 5th to 8th day of infection. No mortality was not reported in these researches, and disease was considered similar to coccidiosis caused by *E. acervulina* in chickens. Later, Ruff & Fagan (1984) used pure and mixed cultures of *E. uzura* to infect quails. They reported mortality, lower weight gain and poor reproductive performance. Concerning weight gain, *E. tsunodai* was considered more pathogenic than *E. bateri*. Finally, when pathogenicity was investigated, the authors suggested that young quails are more susceptible to the coccidiosis effects. The results in the present work corroborate such opinion, since clinical signs were seen only in young quails. Finally, coccidiosis was assumed to be characteristically subclinical in this rearing farm with nonspecific and very mild clinical signs when the whole period of study was considered.

Histological Observations

Endogenous stages of the parasites were found in the small intestine. These were usually located in the villi, mainly above the nucleus of apical epithelial cells, or in the medium portion close to the glands (Figure 1, D and E). These observations resemble those described by Tsunoda & Muraki (1971), Norton & Pierce (1971) and Tsutsumi (1972) not only because of the site of infection, but also morphology was similar. Thus, endogenous stages observed in the small intestine were assumed to be developmental stages of *E. bateri* and *E. uzura*, while the species found in the cecae might be *E. tsunodai*.

Pathological changes were also observed in the mucosa of the small intestine. Villous erosion (Figure 1, F and G), frequently concomitant with hyperplasia of the crypts of Lieberkühn, was often observed, as well as inflammatory infiltrate characterized by the presence of granulocytes and mononuclear cells, usually associated with edema.

The colonisation of the gastrointestinal tract causes structural and functional changes, since the presence of parasites induces both general and local disturbances. Detailed information on such pathological effects can be obtained from previous reviews of coccidiosis in poultry (Levine, 1982; Soulsby, 1987). A common feature of the infection is a severe depression of the digestive and absorptive capabilities of the mucosa. Furthermore, the magnitude of all



disturbances and functional changes are usually related to the intensity of the parasitic infection.

CONCLUSION

Three species of the genus *Eimeria* were found and identified as *E. bateri*, *E. tsunodai* and *E. uzura*; and

the natural infection was characterized as subclinical. Despite of causing mild and nonspecific clinical signs, coccidiosis was considered an important disease because endogenous stages of the parasites and a high number of oocysts in feces were associated with intestinal lesions. The results strongly suggest that such infection might represent a limiting factor for quail production.

Table 1 - Characteristics of species of the genus *Eimeria* described from *Coturnix* quails.

Species	Author	Host	Oocyst		S.I. ^a	Wall	Polar Granule	Sporocyst		Stieda Body
			shape	size (µm)				shape	size (µm)	
<i>E. uzura</i>	Tsunoda Muraki, 1971	<i>C. japonica</i>	ellipsoidal- ovoid ovoid*	18.75-29.0 x 15.0-22.75 18.86-22.76 x 15.04-17.42*	1.31 1.32*	double double*	0-4 2-4*	? ovoid*	? 11.02-12.5 x 5.49-6.39*	? piriform
<i>E. tsunodai</i>	Tsutsumi, 1972	<i>C. japonica</i>	ovoid ovoid*	15.5-22.5 x 16.5-18.5 18.7-21.7 x 14.09-15.67*	1.30 1.36*	double	0-5 1-2*	? ovoid*	10.3-11.5 x 5.0-6.1 9.81-11.01 x 5.09-5.69*	fainted fainted*
<i>E. bateri</i>	Bathia <i>et al.</i> , 1965	<i>C. coturnix</i>	ellipsoidal, ovoid or subspherical	15.0-28.0 x 14.0-23.0	1.26	double*	1	ovoid	9.0-13.0 x 5.0-8.0	prominent
		<i>C. japonica</i> *	ellipsoidal, ovoid or subspherical	19.66-23.34 x 15.04-17.68*	1.32*		1*	ovoid*	9.66-11.8 x 6.08-7.24*	prominent nipple-like*
<i>E. fluminensis</i>	Teixeira e Lopes, 2002	<i>C. japonica</i>	subspherical	15.44-19.18 x 14.73-18.17	1.05	double	?	ovoid	9.32-11.53 x 5.34-6.5	piriform
<i>E. dispersa</i>	Tyzzer, 1929	<i>C. coturnix</i>	ovoid	17.2-26.4 x 15.4-22.4	?	single	?	ovoid	?	?
<i>E. coturnicis</i>	Chakravarty Kar, 1967	<i>C. coturnix</i>	ovoid	26.4-38.8 x 19.8-26.4	?	double	1	ovoid	13.2-17.2 x 8.8-11.0	knob-like
<i>E. taldykurganica</i>	Svambaev Utebaeva, 1973	<i>C. coturnix</i>	ovoid	21.88-25.40 x 11.9-13.1	1.86	double	1	ovoid	8.1-11.6-3.59-4.8	?
<i>E. tahamensis</i>	Amoudi, 1987	<i>C. delegorguey</i>	ellipsoidal	36.5-42.0 x 25.5-29.0	?	double	1	ovoid	14.0-16.0 x 9.0-11.5	knob-like

(*) present work; (a) shape index; (?) unknown.

REFERENCES

Amoudi MA. *Eimeria tahamensis* n. sp. (Apicomplexa: Eimeriidae) from the Arabian quail (*Coturnix delegorguey* arabica). *Journal of Parasitology* 1987; 34:455-456.

Bandoni SM, Duszynski DW. A plea for improved presentation of type material for coccidia. *Journal of Parasitology* 1988; 74:519-523.

Bathia BB, Pandey TP, Pande BP. *Eimeria bateri* n. sp. from Indian common quail (*Coturnix coturnix*). *Indian Journal of Microbiology* 1965; 5:61-64.

Behmer AO, Tolosa EMC, Neto AGF. Manual de Técnicas para histologia normal e patológica. São Paulo: EDART, Editora da Universidade de São Paulo; 1976.

Bigland CH, Da Massa AJ, Woodard AE. Diseases of Japanese quail (*Coturnix coturnix japonica*) – A flock survey and experimental transmission of selected avian pathogens. *Poultry Science* 1964;13:212-219.

Chakravarty M, Kar AB. A study on the coccidian of Indian birds. *Proceedings of the Royal Society of Edinburgh* 1947; 62:225-233.

Dauguschies A, Imarom S, Bollwahn, W. Differentiation of porcine *Eimeria* spp. by morphologic algorithms. *Veterinary Parasitology* 1999; 81:201-210.

Duszynski DW, Wilber PG. A guideline for the preparation of species descriptions in the Eimeriidae. *Journal of Parasitology* 1997; 83: 333-336.

Garcia EA, Mendes AA, Pizzolante CC, Veiga N, Mattos TK. Alimentação de codornas com milho moído e ração de postura no

período pós-jejum durante a muda forçada e seus efeitos sobre o desempenho. *Revista Brasileira de Ciência Avícola* 2002; 4:119-126.

Gruber A, Fernandez S. Recentes avanços na biologia molecular de protozoários do gênero *Eimeria*. In: II Simpósio Internacional sobre coccidiose aviária; 1999; Foz do Iguaçu, PR. Brasil. p. 9-21.

Kucera J, Reznicky M. Differentiation of species of *Eimeria* from the fowl using a computerized image analysis system. *Folia Parasitologica* 1991; 38:107-113.

Levine ND. Protozoan Parasites of Domestic animals and of man. 2 ed. Minneapolis: Burgess Pub. Co; 1982.

Long PL, Joyner LP. Problems in the identification of species of *Eimeria*. *Journal of Protozoology* 1984; 31:535-541.

Mazurkiewicz M, Podlewska D, Wachnik Z. Kokcydioza u przepiórek japonskich. *Medycyna Weterynaryjna* 1967; 23:536-537.

Menezes RCA, Lopes CWG. Epizootiologia da *Eimeria arloingi* em caprinos na microrregião Serrana Fluminense, Rio de Janeiro, Brasil. *Revista da Universidade Federal Rural do Rio de Janeiro* 1995; 17: 5-12.

Norton CC, Pierce MA. The life cycle of *Eimeria bateri* (Protozoa: Eimeriidae) in the Japanese quail *Coturnix japonica*. *Journal of Protozoology* 1971; 18:57-62.

Pereira MJS, Fonseca AH, Lopes CWG. Regressão linear na caracterização de variações morfométricas em coccidia. *Revista Brasileira de Parasitologia Veterinária* 2001; 10:75-78.

Ruff MD; Fagan JM. Pathogenicity of Japanese Quail (*Coturnix japonica*). *Poultry Science*, 1984, 63:55-60.



Soulsby EJL. Parasitologia y enfermedades parasitarias em los animales domésticos. 7 ed. México: Nueva Editorial Interamericana; 1987.

Svambaev SK, Utebaeva MK. Coccidial infections of Phasianus cochicus mongolicus and Coturnix coturnix in Kazakhstan. Izvestiia Akademii Nauk SSSR. Serii biologicheskaja 1973; 6:62-68.

Teixeira M, Lopes CWG. Species of the genus Eimeria (Apicomplexa: Eimeriidae) from Japanese quails (Coturnix japonica) in Brazil and E. fluminensis for the preoccupied E. minima of this quail. Revista Brasileira de Ciências Veterinárias 2002; 9:53-56.

Tsunoda L, Muraki Y. A new coccidium of Japanese quails: Eimeria uzura n. sp. Japanese Journal of Veterinary Science 1971; 33: 227-235.

Tsutsumi Y. Eimeria tsunodai n.sp. (Protozoa:Eimeriidae). A caecal coccidium of Japanese quails (Coturnix japonica). Japanese Journal of Veterinary Science 1972; 34:1-9.

Tyzzer EE. Coccidiosis in gallinaceous birds. American Journal of Hygiene 1929; 10:269-383.

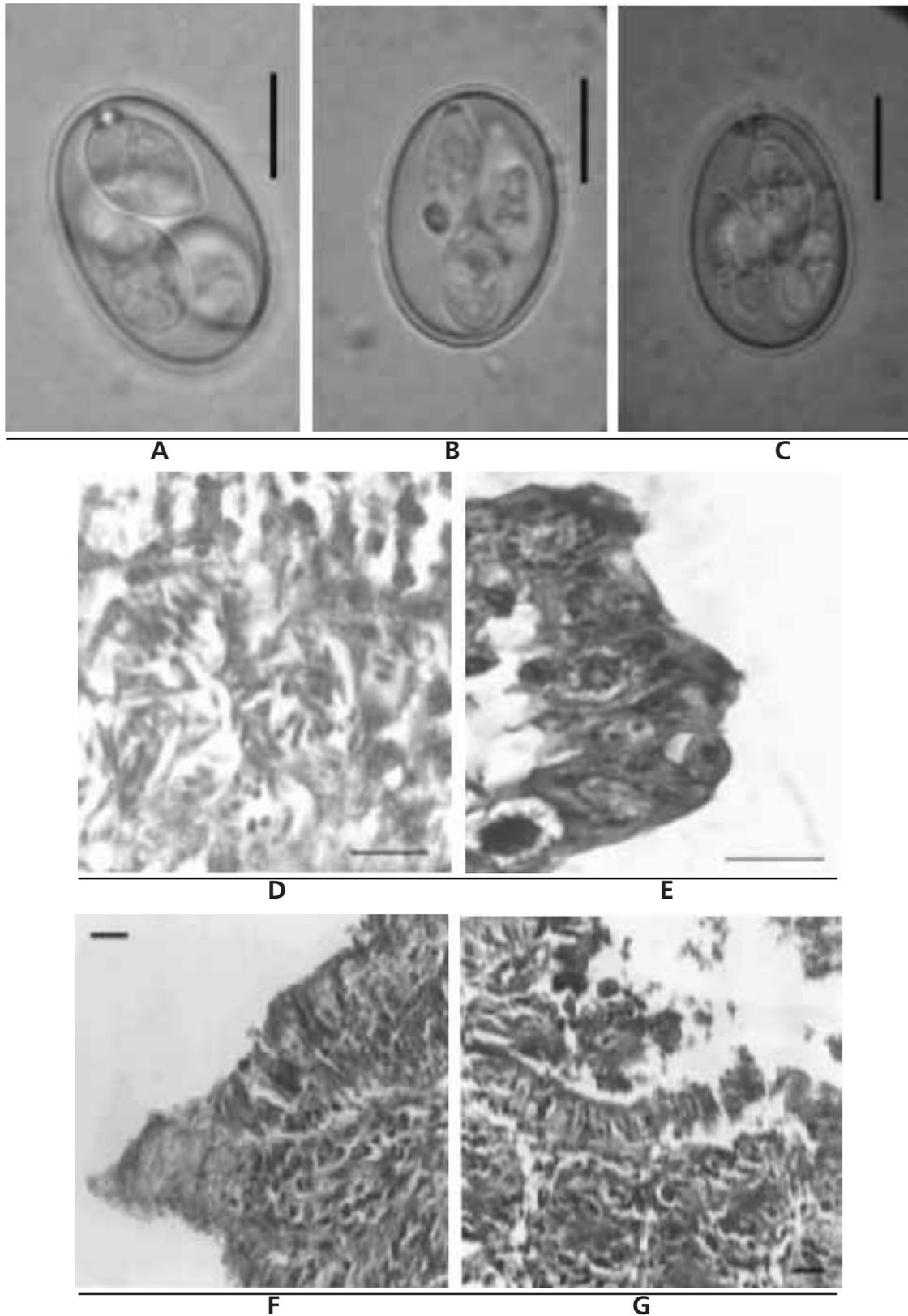


Figure 1 - Sporulated oocysts of (A) *Eimeria bateri*, (B) *E. tsunodai* and (C) *E. uzura* (in saturated sugar solution); Caeca of Japanese quail at the 42nd days, (D) Schizonts with merozoites and ileum at the 14th days, (E) macro and microgamonts, HE; Jejunum of Japanese quail at the 14th days with (F) villous and (G) epithelial erosion, HE; (scale bar = 10µm).