**Klossiella equi** in a donkey – a first case report from Iran

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**Abstract.** *Klossiella equi* is the only known and rarely reported coccidian parasite of the renal paranchyma of equids. An aged male donkey (*Equus asinus asinus*) was submitted to necropsy department of veterinary hospital. In histopathological study of renal sections different developmental stages of parasite were observed. These stages were as follow: Trophozoites, microgametes, macrogametes, sporont, budding sporont, sporoblasts, free sporoblasts, mature sporoblast and sporocyst. Parasitic infection with *K. equi* was encountered in the donkey. According to literature review this is the first report of donkey klossiellosis in Iran.

**INTRODUCTION**

Parasites of the genus *Klossiella* have been reported to occur in the kidneys of a wide range of hosts throughout the world. The genus *Klossiella* was established when Smith and Johnson (1902) chose the name *Klossiella muris* for the coccidial parasite which they had observed in the kidney of both wild and laboratory mice (Smith & Johnson, 1902). Since that time, parasite species of several different animals have been added to the genus *Klossiella*. Various developmental stages of these organisms had been observed in affected kidneys. Although the parasite has been recorded in various parts of the world, the prevalence is still unknown. Due to these inconsistent reports, the life cycle and hence the pathogenicity of this parasite has not been fully elucidated.

*Klossiella* infection has usually been considered to be of negligible pathogenicity, and considered incidental findings. Although the disease is rarely clinically apparent in some cases, the non-pathogenic nature of the organisms has been questioned. Rosenmann & Morrison (1975) were of the opinion that *K. muris* could be potentially pathogenic and may impair metabolism under conditions of stress. Ballweber *et al.* (2012) also added that infections with *Klossiella equi* can be chronic in nature and supports the association of increased severity of klossiellosis and impaired immune function. Reports on the occurrence of *Klossiella equi* in the kidneys are infrequent. Therefore, it is necessary to record every case available with the view to reveal the prevalence of this parasite. This paper is intended to report the first donkey klossiellosis in Iran.

**Case Presentation**

In April 2011, an aged (probable 10 year old) donkey (*Equus asinus asinus*) a mixed breed was submitted to the necropsy department of the veterinary hospital for educational necropsy demonstration class. After euthanasia, representative tissues from selected organs (lung, kidney, liver, stomach, intestine and spleen) were sectioned, placed in neutral buffered formalin and processed for histopathologic examination. Samples were embedded in paraffin, sectioned at 5µm, and stained with hematoxylin and eosin (H&E).
Microscopic examination of kidney demonstrated protozoal organisms within the renal tubules. Most of epithelial cells of henle tubules and convoluted tubules of both kidneys were infected with parasite stages. They were vacuolated with numerous halo vacuoles in cytoplasm with different size. Displacement of nucleus and protrusion of cell wall and cytoplasm to lumen were observed. The nomenclatures of parasitic developmental stages were done according to Gardiner \textit{et al.} (1998) (Fig. 1).

These developmental stages which were seen in tubules were as follow: trophozoites, microgametes, macrogametes, sporont, budding sporont, sporoblasts, free sporoblast, mature sporoblast and sporocyst. Trophozoites were detected with halo parasitophorous vacuole in epithelial cells of the thick limb of the loop of henle. Microgametocytes and macrogametocytes were seen in one large parasitophorous vacuole in epithelial cells of the thick limb of the loop of henle. Microgametocytes and macrogametocytes were seen in large parasitophorous vacuoles which were basophilic and eosinophilic respectively (Fig. 2A). Sporonts were seen as basophilic, round and rosette shape structures and approximately 25-30 µm diameters. They were observed within the cytoplasm of the collecting tubules and loop of henle (Fig. 2B). Budding sporont also were basophilic and round with multiple projections from periphery within parasitophorous vacuoles (Fig. 2C). Sizes of budding sporont were same as sporont. Sporoblasts were basophilic, packed and not separated structures in a big parasitophorous vacuole (Fig. 2D). Numerous free mature sporoblasts were late stages and detected by basophilic oval shape structures. Mature sporoblasts had halo capsules around basophilic nuclei with 5-6 µm in width and 12-13 µm in length (Fig. 2E). Free and mature sporoblasts were seen in cyst with delicate wall (Fig. 2D & E). Sporocysts were seen in lumen of renal tubule which was released from parasitophorous vacuoles (Fig. 2F). No other lesion including tubular necrosis and glomerulonephritis were detected.

Macroscopic and microscopic inspection of other organs revealed interstitial pneumonia in lung, large gasterophilus infestation in stomach and eosinophilic enteritis. Spleen and liver were normal.

**DISCUSSION**

\textit{Klossiella equi} is the only known coccidian parasite of the equine urinary tract. The parasite has been demonstrated in kidney. It has been rarely documented in equids but appears to have a worldwide distribution. Suedmeyer \textit{et al.} (2006) reported that the definitive life cycle of \textit{K. equi} is unknown. Only various stages in the kidney have been documented, but it is believed to begin with schizont generation within the endothelial cells of Bowman’s capsule. Merozoites then rupture from the host cell and pass distally to the thick limb of henle’s loop where upon they penetrate the epithelial cells (Gardiner \textit{et al.}, 1998). Some merozoites develop into microgametocytes, with each forming 8 to 10 microgametes, and others ultimately develop into macrogametes. Microgametes migrate to the macrogametes and are found in a parasitophorous vacuole. A zygote (i.e., sporont) is formed when the pronuclei of a microgamete and macrogamete fuse. Sporogony then occurs, resulting in the formation of sporoblasts that develop into sporocysts. Within the sporocyst, sporozoites are produced and the sporocyst is released into the tubule and then is passed with the urine into the bladder and subsequently to the ground. Sporocysts are likely ingested while grazing with excystment occurring in
Figure 2. Panel A to F are Photomicrographs of Klossiella equi different stages within renal tubular epithelial cells of donkey. (A). Note trophozoites, macrogamete and microgamete within parasitophorous vacuoles (H & E. Bar: 10µm); (B). Round, basophilic and rosette shape structure within parasitophorous vacuoles is named as sporont (H & E. Bar: 20µm); (C). Budding sporont is late stages after sporont and it is characterized by basophilic, round and multiple projections from periphery within parasitophorous vacuoles (H & E. Bar: 20µm); (D). Three different developmental stages of parasite are obvious within a tubule. Sporont, sporoblast and free sporoblasts are detected (H & E. Bar: 20µm); (E). Mature sporoblasts are located in a parasitophorous vacuole with halo capsule around nuclei (H & E. Bar: 20µm); (F). Free sporocysts are obvious in the lumen (H & E. Bar: 10µm).
the digestive tract. Released sporozoites penetrate the intestinal wall, and enter the blood to travel to the glomeruli where they penetrate the endothelial cells and repeat the cycle (Smales et al., 1996).

Karanja et al. (1995) reported that in a Kenyan donkey, schizonts were not observed and other developmental stages, gametogony and sporogony, were seen and described. Although they named the developmental stages differently, their results are in agreement with observation made in this paper. Not only were schizonts not detected but also gametogonic and sporogonic stages were observed in renal tubules. This finding indicated that schizogony may be occurring in another place in donkey (E. asinus asinus).

Infection with K. equi has been associated with nephrosis and nephritis in immunocompromised equids (Anderson et al., 1988; Ballweber et al., 2012). The absence of any clinical signs relevant to kidney malfunction on K. equi infected donkey in the present study coincide with the finding of many authors such as Karanj et al. (1995) who concluded that many cases of klossiella are not clinically apparent except upon histological examination or in experimental work. In this case report, senility may be the main cause of infestation. Actually electron microscopy is helpful for identification and it was not performed in this case. Further investigation into the life cycle and clinical effects of K. equi are needed to determine the pathogenic nature of the parasite.

According to histopathologic study, different developmental stages of K. equi were described in donkey and the authors believe that this is the first report of klossiellosis in Iran.

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REFERENCES


