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Klossiella cobayae associated with Chronic Interstitial Nephritis in Guinea Pigs

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INTRODUCTION

The protozoan parasites *Klossiella* belong to the class *Telosporea*, subclass *Coccidia*, order *Eucoccidiae*, Suborder *Adeleina*, family *Klossiellidae* (*Honigber et al.* 1964). Members of this family have been described in guinea pigs (*K. cobayae*), mice (*K. muris*), horses (*K. equi*), (*Vetterling* 1975) and laboratory rats (*Shadduck* and *Pakes* 1978). *K. cobayae*, the kidney coccidium of the guinea pig, was earlier considered ubiquitous among guinea pig colonies in various parts of the world, but reports on this parasite are nowadays rare (*Boniciu et al.* 1957, *Manning et al.* 1984). The life cycle of K. cobayae is direct. According to *Vetterling* (1976), the first schizogony occurs in the capillary endothelial cells of the kidney and other organs. Mature schizonts contain merozoites that are released into the blood when the host cell ruptures. Merozoites could invade other endothelial cells repeating the schizogonic phase, or could reach the kidney, where a second generation schizogony occurs in the epithelium of the proximal convoluted tubules. Large schizonts containing numerous merozoites distend the tubular cells, until they rupture. The merozoites are released and enter the epithelium of the thick limb of Henle's loop. There gametogony starts. Differentiation of macro- and microgametes is followed by fertilization of the macrogametes. The resultant zygote undergoes sporogony. The sporont produces sporoblasts that become sporocysts. Each sporocyst gives rise to numerous sporozoites. The host cell membrane ruptures releasing sporulated sporocysts that leave the kidney carried by the urine. When ingested by a new host, the sporozoites excyst in the intestinal lumen. Once they reach the capillaries they invade endothelial cells, starting a new cycle.

We communicate herein the finding of *K. cobayae* in convertionally kept, clinically healthy guinea pigs submitted for health monitoring at the National Veterinary Institute, Uppsala, Sweden.

MATERIAL AND METHODS

Animals: Six conventionally kept, female, Dunkin-Hartley guinea pigs of body weights ranging from 730 g to 1070 g were selected from the breeding stock of a research unit, and submitted for health monitoring. After euthanasia by CO_2 inhalation, necropsies were immediately performed.

Histology: Samples from different organs were fixed in 10 % buffered formaline for 24 hours. After routine histological processing, the material was embedded in paraffin, cut 4 µm thick sections and stained with haematoxylin and eosin. Additional sections from kidney were stained according to the following methods: Azan, Gram, PAS, and van Gieson.

Parasitology: Routine parasitological examination was performed. Lung samples were used for detection of antibodies against *Toxoplasma gondii* and *Encephalitozoon cuniculi*, by means of india-ink immunoassay (*Waller* 1977).

RESULTS

Kidney

Macroscopical examination: Two guinea pigs exhibited bilateral granular appear-

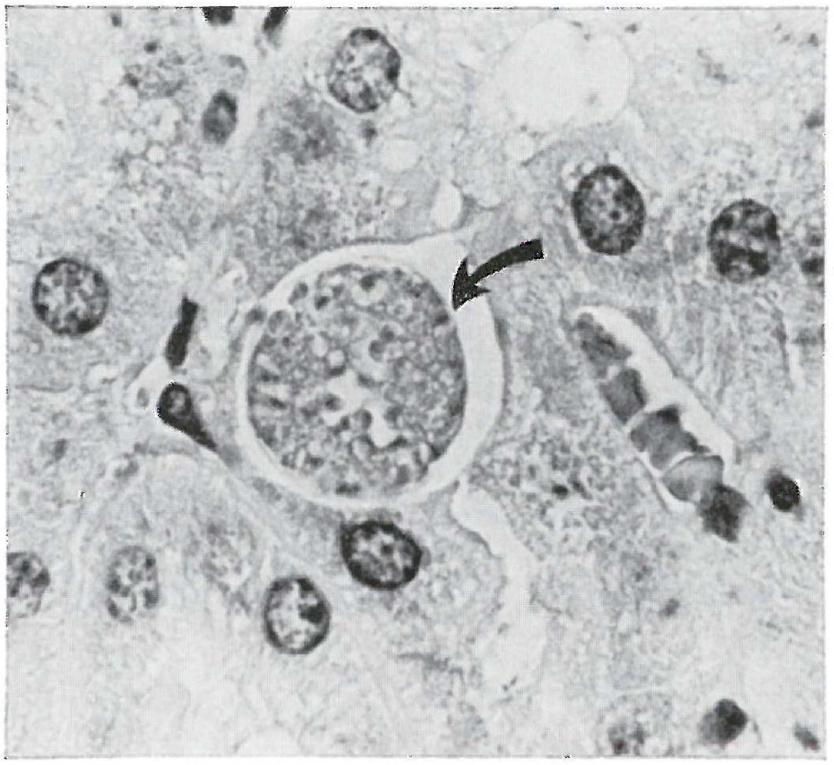

Fig. 1. Kidney. Schizont containing numerous merozoites, filling the lumen of a proximal convoluted tubule (arrow). H. & E. × 1 000.

ence of the subcapsular surface. Four guinea pigs did not present any macroscopical change.

Histopathological examination: Renal microscopical changes were present in all of the guinea pigs, but were more severe and involved larger areas in kidneys with obvious macroscopical lesions. In the advanced cases, affected areas comprised about half of the kidney parenchyma. Large schizonts containing numerous merozoites, characteristic of *K. cobayae*, were observed in tubular lumina (Fig. 1). The number of parasites was low. They were negative for the Gram stain, and neither Azan nor PAS stains evidenced a cuticular structure. The lesions occurred mainly in the cortex. They were severe but focal. Within affected areas almost all tubuli and glomeruli displayed marked disruption. Constant features were tubular degeneration and necrosis and areas of reparative hyperplasia of the epithelium. The basal laminae were thick and hyalinized, and tubular lumina were often filled with homogenous strongly PAS positive material. Mitotic figures were common, both in tubular epithelium and in interstitial fibroblasts. Necrotic tubuli containing mineralized casts were numerous in the medullary zone, often located in areas devoid of inflammatory changes.

The glomeruli displayed thick, hyalinized, strongly PAS positive Bowman's membranes. Severe hypertrophy of the parietal epithelial linning was occasionally observed (Fig. 2). Few glomeruli were atrophic and sclerotic.

The interstitium within affected zones presented marked fibroblastic proliferation and lymphocytic infiltration (Fig. 3). A discrete number of eosinophils was ob-

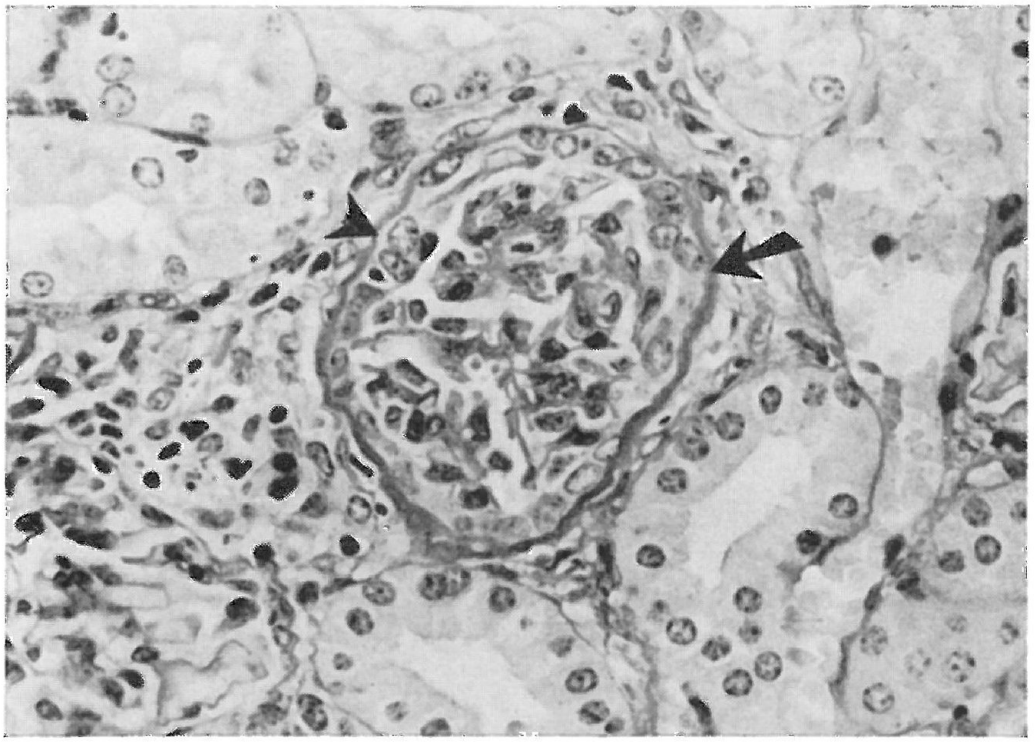

Fig. 2. Kidney. Cortical focus of inflammation. Note marked glomerular changes. The parietal layer of Bowman's capsule exhibits thickened basal lamina (arrow) and hypertrophy of parietal epithelium (arrow head). Atrophic glomerular capillaries display thickened basal laminae. PAS stain × 400.

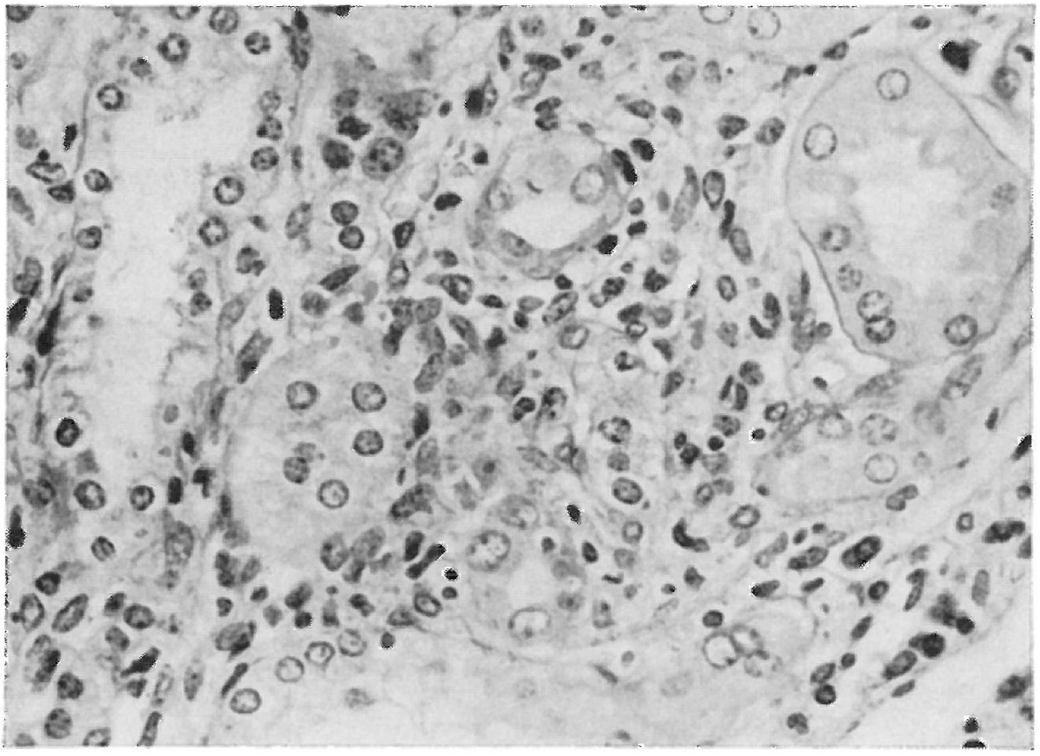

Fig. 3. Kidney. Focus of chronic interstitial nephritis with mild mononuclear infiltration, fibroblastic proliferation and tubular disruption. PAS stain × 400.

served in glomerular and tubular capillaries. Two guinea pigs presented moderate infiltration of lymphocytes and plasma cells in the renal pelvis.

Lungs

Macroscopical changes were not observed. Histologically, all guinea pigs exhibited a marked perivascular infiltration of mononuclear leukocytes and a low number of eosinophils. Mild focal chronic interstitial pneumonia was diagnosed in two guinea pigs. Stages of *K. cobayae* were not observed.

Other visceral organs did not present significant changes.

Parasitological findings: The nasal cavities and the gastrointestinal canal were free of parasites.

The india-ink immunoreaction was negative both for *Toxoplasma gondii* and for *Encephalitozoon cuniculi* antibodies.

Discussion

Schizonts of *K. cobayae* were diagnosed in the kidneys of guinea pigs with lesions of focal chronic interstitial nephritis and chronic pneumonia.

According to *Vetterling* (1976), the most readily identifiable and most frequently seen stages of *K. cobayae* are those occurring in the kidneys. Stages in other locations should be carefully examined since they somewhat resemble *T. gondii* and *E. cuniculi* (*von Cossel* 1958, *Vetterling* 1976). As the kidneys are a common location for *E. cuniculi*, a differential diagnosis must be made also when renal stages of *K. cobayae* are detected.

It has been stated that the parasite is usually nonpathogenic (*Flynn* 1973), produces only slight kidney changes (*Manning et al.* 1984), or that is nonpathogenic exept in very heavy infections (*Shaddock et al.* 1978). However, our findings are in agreement with those of *Hofmann* and *Hänichen* (1970), who observed a pathogenic affect on the kidney, even in a relatively low degree of parasitic infection.

In the present study, stages of *K. cobayae* were not found in the lungs (a systematic search was not attempted). However, lung changes similar to the ones we observed were reported by *Bonciu et al.* (1957) in 90 % of the guinea pigs infected by this parasite. The lesions were produced by parasitic damage to the vascular endothelium. As the bacteriological and parasitological studies from lungs and digestive organs were negative for infections that could induce similar pulmonary changes, we conclude that they are most probably due to extrarenal stages of *K. cobayae.*

Since infected guinea pigs mainly are asymptomatic, the disease usually remains undetected. Guinea pigs are often used for different toxicological studies in which the kidneys or the lungs are target organs (*Hoar* 1976). Lesions caused by *K. cobayae* may thus modify experimental results. Infection with *K. cobayae*, even when asymptomatic, is reported to increase the sensitivity of guinea pigs to different toxic substances (*Bonciu et al.* 1957). The role of *K. cobayae* infection as a source of misinterpretation of experimental results has also been emphasized by *von Kossel* (1958) and *von Hofman* and *Hänichen* (1970).

Reliable methods for identification of live guinea pigs infected with *K. cobayae* are not available, and the authors are not aware of any treatment. However, it has been suggested that this parasite could be controlled by reducing the urine contamination of guinea pigs facilities (*Vetterling* 1976). Cesarean derivation permits the obtention of guinea pigs free from *K. cobayae* (*Flynn* 1973).

The present findings underline that health monitoring including macro- and microscopical examination of different organs is a mandatory procedure for the identification of health problems affecting experimental animals.

References

Bonciu, C., B. Clecner, A. Greceanu et A. Märgineanu: Contribution à l'étude de l'infection naturelle des cobayaes avec des sporozoaires du genre "Klossiella". Arch. Roum. de Pathol. Exper. 1957, 16, 1: 131—143.

von Cossel, L.: Nierenbefunde beim Meerschweinchen bei Klossiellen-infektion (Klossiella cobayae). Schweiz. Z. Path. Bakt. 1958, 21: 62—73.

Flynn, R. J.: Parasites of laboratory animals. The Iowa State University Press, Ames, Iowa 1973.

Hoar, R. M.: Toxicology and teratology. In "The biology of the guinea pig". J. E. Wagner and P. J. Manning, EDS. Academic Press, Inc. Chapter 1976, 18: 269—280.

von Hofmann, H. and *T. Hänichen:* Klossiella cobayae. Nierenkokzidiose bei Meerschweinchen. Berliner und Munchener tierärztliche Wochenschrift 1970, 8: 151—153.

Honigberg, B. M., W. Balamuth, E. C. Bovee, J. O. Corliss, M. Gojdics, R. P. Hall, R. R. Kudo, N. D. Levine, A. R. Loeblich Jr., J. Weiser and *D. H. Wenrich:* A revised classification of the phylum protozoa. J. Protozool. 1964, 11: 7—20.

Manning, P. J., J. E. Wagner and *J. E. Harknes:* Biology and diseases of guinea pigs. In Laboratory Animal Medicine. J. G. Fox, B. J. Cohen, and F. M. Loew, Eds. Academic Press, Inc. Chapter 1984, 6: 149—181.

Shadduck, J. A. and *S. P. Pakes:* Protozoan and metazoan diseases. In Pathology of Laboratory Animals, K. Benirschke, F. M. Garner and T. C. Jones, Eds. Springer Verlag, Chapter 1978, 17: 1587—1696.

Vetterling, J. M.: Protozoan Parasites. In "The Biology of the guinea pig", J. E. Wagner and P. J. Manning. Eds. Academic Press, Inc. Chapter 1976, 12: 163—196.

Waller, T.: The india-ink immunoreaction: A method for the rapid diagnosis of encephalitozoonosis. Lab. Anim. 1977, 11: 93—97.

Summary

Infection with the renal coccidian parasite *K. cobayae* was detected in 6 conventionally kept Dunkin-Hartley guinea pigs, submitted for routine health monitoring at The National Veterinary Intsitute, Uppsala. Chronic inflammatory lesions were microscopically observed in the kidneys and the lungs of all animals.

Abstrakti / K. Pelkonen

Yhteenveto

Kuudestra Statens Veterinärmedicinska Anstalt'iin rutiininomaista terveyden laaduntarkkailua varten lähetetystä konventionaalisesti kasvatetusta Dunkin-Hareley-marsusta löydettiin Klossiella cobayae-munuaiskokkidiparasiitin aiheuttama infektio. Kaikissa eläimissä oli munuaisissa ja keuhkoissa mikroskooppisesti havaittavissa kroonisia tulehdusvaurioita.