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With 5 figures in the text. *www.sudanjvr.net*

Camel Experimentally infected with Sarcocystis

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ملخص البحث

في هذا البحث تم تجريع اثنين من حيران الإبل الفطيمة بنوعين من جر يبات المتكيسة اللحمية (*Sarcocystis sporocysts*) والتي جمعت من أمعاء كلاب صغيرة سبق وان أطعمت لحم ابل .أحد الحيران تمت معالجته بالامبروليوم كمضاد للكوكسيديا ، اما الحوار الآخر والذي لم يعالج ، نفق في اليوم السادس والعشرون بعد العدوى ، لم تظهر أي أعراض للمرض على الحوار المعالج والذى ذبح في اليوم الخامس والتسعون بعد العدوى . اظهر التشخيص النسيجي وجود نوعين من طفيلى المتكيسة اللحمية ، أحدي هذه المتكيسات (72.5 –264 x 9.9 – 29.5 ميكرومتر) لها جدار خلوى رقيق (0.5 –1 ميكرومتر) اما الاخري (73 – 155 x 23 – 29.5 ميكرومتر) فلها جدار خلوى سميك (2 – 3 ميكرومتر) . تم الكشف على اجزاء مهضومة اصطناعياً من الحوار المعالج (اللسان ، القلب ، البلعوم ، المرئ ، الحجاب الحاجز والعضلات) اثبت وجود مقوسات (Bradyzoites) تتراوح ابعادها بين 10- 16 ميكرومتر × 3 – 4 ميكرومتر.

Summary

Two weaned camels calves were experimentally infected with two types of sarccystis sporocysts harvested from intestinal mucosae of puppies that had been fed on cameline meat. One camel calf was medicated with Amprolium as anticoccidial agent. The unmedicated calf camel died on day 26 post infection (Pi) while the medicated one remained clinically normal till day 95 when it was killed. Histological findings revealed the presence of two type of Sarcocystis tissue cysts. One form measured 72.5-264µm x 9.9-29.5µm with a thin wall (0.5-1um in width) while the other cyst measured 73-155µm x 23-29.5µm and had a thick cyst wall (2-3um in width). Portions of the tongue, heart, oesophagus, diaphragm and skeletal muscles from the medicated camel calf were artificially digested. They revealed the presence of bradyzoites that measured 10.0-16.0µm x 3-4 µm.

Introduction

Sarcocystis had an obligatory two-host life cycle; the intermediate hosts (Herbivores) acquire infection by ingesting sporocysts that are shed out in the faeces of infected definitive hosts (carnivores). Carnivores become infected by ingesting the encysted form of the parasite in the musculature of intermediate hosts. However, the life cycle of this parasite was unknown until 1972 when Fayer (1972) described gametogony in cell culture. Sarcocystis tissue

cysts of camels were first described by Mason (1910). Hilali and Mohamed (1980) described the dog as a definitive host for *Sarcocystis cameli* in the dromedary. The prevalence of cameline sarcocystosis was 81% in the Sudan (Hussein and Warrag 1985) compared to a prevalence of 36.5% in Egypt (Hillali and Mohamed, 1980). Although camel sarcocystosis is wide spread in the world, little is known regarding the species involved their developmental cycle, pathogenicity and economic significance. The aim of this investigation was to determine the effect of *Sarcocystis* infection in camels and speciate *Sarcocystis* of camels depending upon descriptions of sporocysts isolated from dogs fed camel meat and cysts detected in histosections.

Materials and Methods

Experimental camels:

Two four week-old weaned camel calves (No. 1 and 2) were purchased from Omdurman Camel Market and kept for two weeks prior to infection.

Preparation of inoculum:

Sarcocystis sporocysts were obtained from the intestines of six naive puppies fed with raw camel's meat. Twenty nine days post feeding, the puppies were killed and the intestinal mucosae were artificially digested as described by Box and Smith (1982). The sporocysts obtained were stored at 4^{o}C in 2.5% potassium dichromate until use for infection.

Experimental infection:

Each animal was inoculated orally with $1x10^{6}$ *Sarcocystis* sporocysts. Calf No. 2 was medicated with Amprolium at 100 mg/kg body weight mixed with the food. The medication began on the day of inoculation and continued daily for 30 consecutive days according to the procedure described by Leek and Fayer (1978). The camels were kept under observation during the whole time of the experiment (110 days) in clean pens at the premises of the Department of parasitology, faculty of Veterinary Science, university of Khartoum. During the experimental period, the camels were fed on reboiled milk until the sixth week of age and thereafter on alfalfa hay and concentrates.

Haematological methods:

Blood samples were taken once a week from the jugular vein, using heparinized vacutainer tubes for haematological studies.

Histopathological methods:

Sections were prepared from lung heart, tongue, diaphragm, oesophagus, spleen, kidney, liver, rumen, reticulum, omasum, abomasum, small and large intestines, mesenteric and gastric lymph nodes, brain, eyes, aorta, coronary arteries and skeletal muscles, and stained with H & E and PAS.

Artificial digestion:

One kilogram of tissues from oesophagus, tongue, diaphragm, heart and skeletal muscles was obtained from the medicated camel which was killed 95 day post infection. The meat was artificially digested as described by Farooqui *et al*. (1987). The final pellet was examined by light microscope for the presence of tissue-cyst stages.

Results

Clinical findings:

Calf No. 1 remained clinically normal until 20 days after its infection, when it became inappetant and dull. Two days later, it ceased to take food or water, had constipation, became anaemic, lethargic, weak and remained so until it died 26 days PI.

Calf No. 2 remained clinically normal and was killed after 95 day from infection.

Haematological findings:

Infected unmedicated calf No. 1 became anaemic after 16 days from infection. The red blood cell (RBC) counts deceased from 8.1 x1012/L to 3.6x1012/L. The haemoglobin concentration fell from 9.5g/dl to 5.0g/dl. The initial PCV value was 0.290 L/L fell to 0.150 L/L. The haematolgoical value of the infected medicated calf no. 2 remained within the normal level during the experimental period (110 days)

Macroscopic findings:

The following changes were observed in calf No. 1: petechial haemorrhages on the omentum, mesenteric lymph nodes, urinary bladder and serosal surface of visceral organs were observed. Petechial haemorrhages were also found in the brain. The lungs were congested, filled with froth and fluid. Petechial to ecchymotic haemorrhages were also found in the cardiac musculature and in all skeletal muscles examined.

Histological findings:

The most striking microscopic lesions in calf no. 1 were found in mesentric lymph nodes and lungs which revealed thrombi, haemorrhages and intestinal infiltration of lymphoid cells (Fig.1 and 2). Immature cysts containing metrocytes were found in the brain (Fig. 3) in addition, haemorrhages and perivascular lymphoid cell infiltration were also present. The kidneys revealed glomerulonephritis associated with haemorrhage and infiltration of lymphoid cells in the interstitial connective tissue and subcapsulr area. Haemorrhages were also found in the heart, skeletal muscles and intestine. The most striking microscopic finding in calf no. 2 was the presence of Sarcocystis cysts. The sarcocysts were spindle-shaped and located lengthwise between the muscles fibres of heart, oesophagus, tongue, diaphragm and all skeletal muscles examined. There was no evidence of inflammatory reactions at the vicinity where the cysts were located since the parasite has an innocuous effect on the muscle and usually there is no muscle fiber degeneration. Two types of cysts appeared in the sections. The cysts were randomly distributed rather than regularly spaced amongst the muscle fibers.

One type of the cysts measured 72.5-264µm x 9.9-29.5µm and had a thin cyst wall of 0.5-1µm. The cyst wall was formed of two layers; an outer layer composed of radial spines and a smooth inner one. The ground substance extended inwards into the cyst in the form of narrow *septae*, which divided the whole cyst into compartments (Fig. 3). The other sarcocyst type measured 73-155µm x 23-29.5µm. It has a thick striated outer and a smooth inner layers. The cyst wall divided the cyst into compartment (Fig. 4).

Examination of the wet smears of the muscle digest revealed that the bradyzoites obtained from the sarcocysts were banana-shaped with slightly pointed anterior and a rounded posterior ends. The bradyzoites measured 10-16 µm x 3.4 µm (Fig.5).

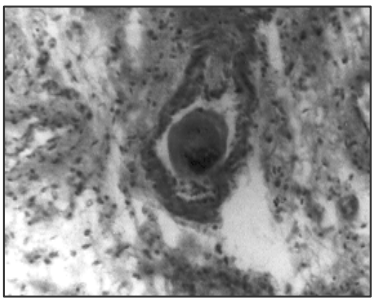

Fig. 1: Mesenteric lymph node of the calf-camel No. 1 showing thrombus in a vein × 400

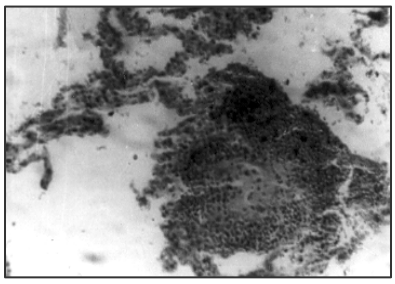

Fig. 2: Lung of the camel calf No.1 showing hemorrhage ×400

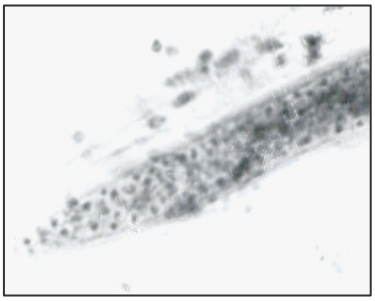

Fig. 3: Type I *Sarcosysts cameli* cyst with thin wall of two layers

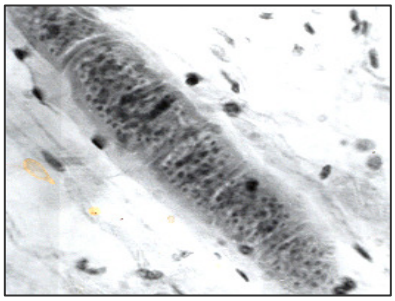

Fig. 4: Type II *Sarcocysts cameli* cyst with thick striated outer and smooth inner layers

Fig. 5: A bradyzoite obtained from the artificial digestion of the composite meat of camel calf No 2 ×2000

Discussion

There is considerable confusion in the literature regarding the speciation of *Sarcocystis*. Originally a single species was thought to parasitize each herbivorous species. However, recent studies have shown that, more than one species may parasitize a single intermediate host. In this study, two types of *Sarcocystis* tissue cysts were detected in experimentally induced infection in the camel (*Camelus dromedrius*). It is worth noting that the two different types of sporocysts were detected in the inocula used in this study to induce infection in the camels. Thus, it is probable that at least two different species of *Sarcocystis,* viz. *Sarcocystis cameli* and *Sarcocystis spp*. are transmissible between dogs and camels. Descriptions of sarcocystis tissue cyst given by earlier workers in Egypt indicated the existence of two different types. Mason (1910) in his original report of the occurrence of *Sarcocystis* infection in Egyptian camels described two types of tissue cysts, one with a smooth, non-striated wall and the other with a striated wall. Recently, Abdel Ghafar, *et al*. (1979) described *Sacocystis* tissue cysts in camels in Egypt, that measured 130-180 µm x 60-110 µm and had a smooth wall with little or no striations. On the other hand, Hilali and Mohamed (1980) described a sarcocystis from camels, which measured 33-389 µm x 22-33 µm and had 1-2 µm thick striated walls.

The clinicopathological picture reported in sarcocystosis in camel calves is similar to that described from cattle, sheep and goats (Johnson *et al*., 1975; Leek *et al*. 1977; Dubey *et al,* 1981). Immature cysts found in the brain of calf no. 1 may explain the observed lethargy and unsteady gait. The development of haemorrhages in many organs is common to sarcocystosis in sheep, goats and cattle. First generation meronts develop in arteries and arterioles up to 16 days PI, and they are apparently non-pathogenic. Cattle and sheep inoculated with *Sarcocystis* do survive well during this phase. In this study, calf-camel no. 1 died 26 days PI and this coincided with the development of second generation meronts (Johnson *et al.,* 1975). The release of merozoites or their metabolites in the infected host may act as contributory factor to the development of anaemia (Dubey *et al*., 1981).

Amprolium given in the food at 100 mg/kg for 30 days (beginning on the day of inoculation) was effective in preventing

death in camel calf no. 2, which was, inoculated with $1x10^6$ *Sarcocystis sporocysts*. Amprolium has proved to be effective in reducing the acute effects of *Sarcocystis bovicanis* in experimentally infected calves (Fayer and Johnson, 1975), and *Sarcocystis ovicanis* in experimentally infected lambs (Leek and Fayer, 1978). Amprolium effectively reduced the number of second generation meronts. It is administered prophylactically because it permits the development of immunity while protecting the host from severe disease and rapid death resulting from infection with *Sarcocystis* (Leek and Fayer, 1978).

References

Abdel Ghaffar, F.R., Entzeroth, S.; Chobotar, E.B. and Scholtyseck, E. (1979). *Tropenmed. Parasitol*. **30:** 434-438**.**

Box, E.D. and Smith, J.h. (1982). *J. Parasitol*. **68**(4)**:** 668-673**.**

Dubey, J.P., Weisbrode, S.E.; Speer, C.A. and Sharma S.P. (1981). *J. Am. Vet. Med. Assoc.* **178** (7): 683-699.

Farooqui, A.A., Adams, D.D., Hanson, W.L. and Prest Wood, A.K. (1987). *J. Parasitol*. **73** (4): 681-688.

Fayer, R. (1972). *J. Sci.* **175:** 65-67.

Fayer, R. and Johnson, A.J**.** (1975). *J. Parasitol*. **61**(5)**:** 932-936.

Hillali, M. and Mohamed, H.A. (1980). *Tropenmed. Parasitol*. **31:** 21-214.

Hussein, S.H., Warrag, M. (1985). *Trop. Anim. Hlth. Prod*. **17** (2)**:** 100-110.

Johnson, A.J.; Hildebrandt, P.K. and Fayer, R. (1975). *Am. J. Vet. Res.* **36**: 995-999.

Leek, R.G. and fayer, R. (1978). *Corn. Vet*. **68:** 108-123.

Leek, R.G., Fayer, R. and Johnson, A.J. (1977). *J. Parasitol***. 63:** 242 - 250.

Mason, E.P. (1910). *J. Comp. Path. Therap*. **23:** 168 -176.