Caryospora schokariensis sp. n. (Apicomplexa: Eimeriidae) from Forskal's Sand Snake, *Psammophis schokari* (Serpentes: Colubridae) in Saudi Arabia

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Abstract. A new coccidan parasite of the genus *Caryospora* is described from the intestinal contents of the colubrid snake, *Psammophis schokari* (Forskal's 1775) collected at Gazan city, southern region, Saudi Arabia. Oocysts of *Caryospora schokariensis* sp.n. are elongate-ellipsoid, 49x33.5 (44.1-52.3X30.0-37.4) µm, with smooth brownish-yellow bilayered wall, 1.5 (1.3-1.8) µm thick. Micropyle, polar granule and oocyst residuum are absent. Sporocysts are ovoid, 21.8X15.1 (20.0-22.6X14.5-16.4) µm., with a prominent Stieda and substieda bodies. Sporocyst residuum is present as a granulated compact mass. Sporozoites are banana-shaped, each with two subspherical refractile bodies.

Key words: *Caryospora schokariensis* sp.n., coccidia, developmental stages, *Psammophis schokari*.

Introduction

Forskal's sand snake, *Psammophis schokari* (Forskal, 1775) is a widely distributed colubrid snake species, occurring in desert and semidesert regions of Middle East (Modry *et al.*, 1999), including Saudi Arabia (Al-Sadoon 1989). Although several species of *Caryospora* have been reported from the members of the family Colubridae (Matuschka 1986, Upton *et al.*, 1990, Modry and Koudela 1998), only *Caryospora maxima* has been described from *Psammophis schokari* (Modry *et al.* 1999). Here with, a new publication dealing with reptilian coccidiosis in Saudi Arabia (Kasim and Al-Shawa 1988; Amoudi 1989; Alyousif and Al-Shawa 1997, 1998; Alyousif, *et al.* 1997; Al-Rasheid and Alyousif 1998; Alyousif and Al-Rasheid 2001). A detailed description of a new species of *Caryospora* infecting *P. schokari* from Gazan, Saudi Arabia is represented.

Materials and Methods

During August 2001, eight specimens of adult *Psammophis schokari* were cuptured alive from Gazan city, southern region of Saudi Arabia. These snakes were placed individually in plastic cages and their faecal samples were collected separately and mixed with 2.5% (w/v) aqueous potassium dichromate solution to inhibit bacterial growth. The suspension was then spread in a thin layer in petri dishes and incubated at 25 ± 2 °C for 1 week and examined periodically to determine the sporulation time. Sporulated oocysts were concentrated by flotation with Sheather's sugar solution technique. Thirty sporulated oocysts and 30 sporocysts were examined and measured using

Zeiss photomicroscope fitted with a 100X apochromatic oil immersion objective and a 10X ocular micrometer. The number of layers of the oocyst wall, its thickness and the detailed structure of the sporocysts were examined after crushing oocysts by applying pressure to the coverslip. All measurements are in micrometers with the ranges in parentheses following the means.

One infected snake was sacrificed and tissue samples of liver, gall bladder, kidney, small and large intestine were fixed in 10% neutral buffered formalin and processed for light microscopy using standard histological method. Paraffin sections were stained with haematoxylin and eosin (H&E) and examined for the presence of the different endogenous stages of coccidia.

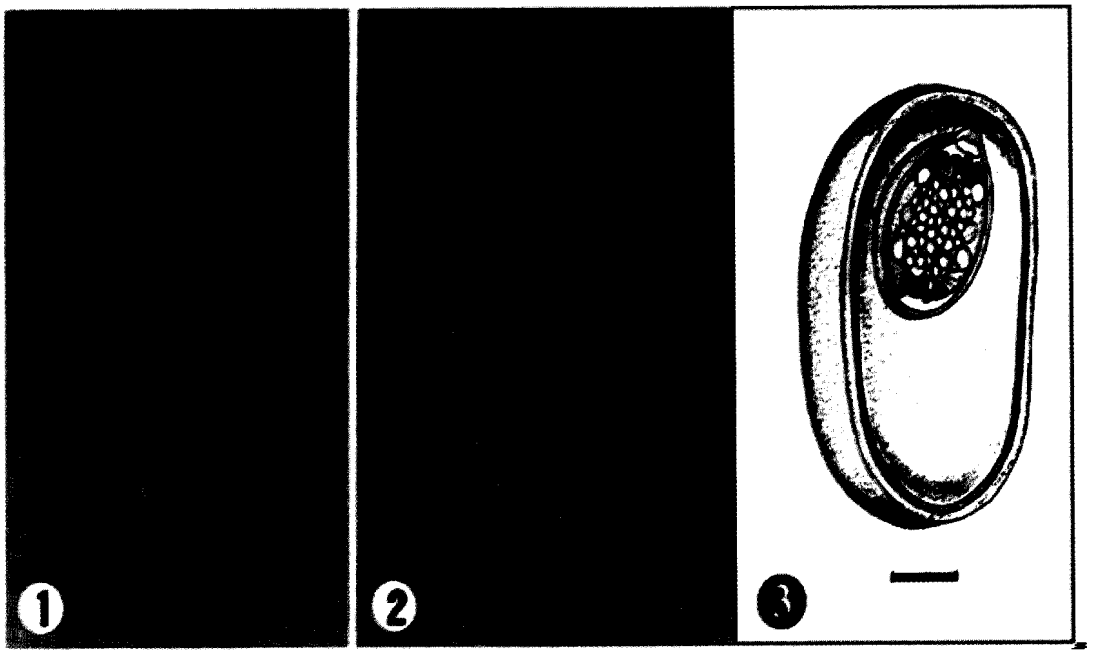
Results

During a survey of the parasitic fauna of the southern region of Saudi Arabia a total of eight adult forskal's sand snake, *Psammophis schokari* were examined for caryosporan infection, two of them had large numbers of oocysts in their faecal contents, and there is no doubt that these oocysts representing a new species of *Caryospora* which we describe below.

***Caryospora schokariensis*. sp.n. (Figs. 1-3).**

Description:

Sporulated oocysts were elongate-ellipsoid in shape with smooth surface, 30 oocysts from the infected snake were measured 49.4 X33.5 (44.1-52.3X30.0-37.4), shape index (length /width ratio) 1.48 (1.24-1.74). The oocyst wall is brownish-yellow, measured 1.5 (1.4-1.6) thick, and bilayered by light microscopy. Micropyle, oocyst residuum and polar granule are absent.



Figs. 1-2. Photomicrographs of sporulated oocysts of *Caryospora schokariensis* sp. n. from naturally infected *Psammophis schokari*. X 1000. **Fig. 3.** Composite line drawing of sporulated oocysts of *C. schokariensis* sp. n. Scale bar = 10 μm.

Single ovoid sporocyst is present, 21.8 X15.1 (20.0-22.6 X14.5-16.4) with a smooth single layered wall, shape index, 1.44 (1.23-1.55). A dome-like Stieda body is present of 1.8 height X3.5 wide (1.5-2.1 X3.2-3.8) and a substieda body of 1.5 high X 4.1 wide (1.3-1.7 X 3.5-4.4). A sporocyst residuum is present, consisting of many granules in a compact mass. Sporozoites are banana-shaped measuring, 16.6 X3.7 (14.7-17.5 X3.2-4.0) and are arranged head to tail within the sporocyst. Four sporozoites lie in one direction, whereas the other four lie in the opposite direction. Each sporozoite contains a prominent paranuclear spherical to subspherical refractile bodies. The single nucleus is located between the two refractile bodies.

Histological examination of the intestine from the infected snakes revealed various endogenous development stages (schizogony and gamogony) in the epithelial cells of the small intestine (Figs. 4-8).

Taxonomic summary

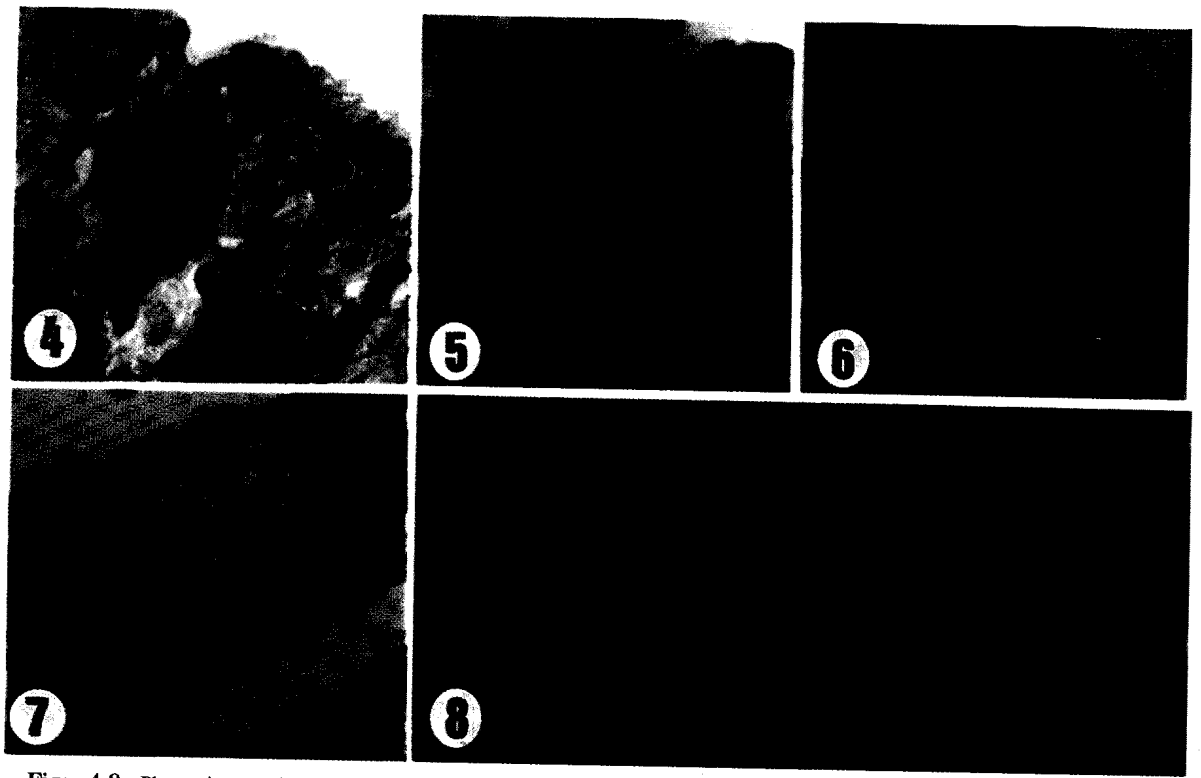
Type host: Forskal's sand snake *Psammophis schokari* (Serpentes: Colubridae).

Type locality: Gazan, southern region, Saudi Arabia.

Prevalence: Found in two eight (25%) infected *Psammophis schokari.*

Site of infection: Histological examination revealed the presence of the endogenous stages within the epithelial cells of the small intestine.

Sporulation time: The majority of oocysts examined had completed sporulation within 24-48 hr. at 25 ± 2 °C.



Figs. 4-8. Photomicrographs of various endogenous stages of *Caryospora schokariensis* sp. n. infecting the small intestine epithelia of *Psammophis schokari* **Fig. 4.** Mature schizont in cross section (arrow) X 1000. **Fig. 5.** Intermediate microgamont, (*) showing random distribution of nuclei. X 1500. **Fig. 6.** Immature macrogamont. (arrow) X 1000. **Fig. 7.** Mature macrogamont (arrow) X 1000. **Fig. 8.** Young oocyst in the host's intestinal cell. X 1200.

Table 1. Comparative data of *Caryospora* species from genus *Psammophis*.

Original stracture	*C. legeri*	*C. hermae*	*C. psammophi*	*C. weyerae*	*C. maxima*	*C. schokariensis*
Oocyst shape	subspherical to ovoid	subspherical	subspherical	subspherical	spherical to subspherical	elongate-ellipsoid
size (µm)	20.8-30.4X 19.2 - 25.6	22.3 X 20.6 21-24 X 20-22	29.1 X 26.8 25-34 X 23-31	16.1 X14.9 14- 18 X 13 - 17	43 X 42.1 40-46 X 40-44	49.4 X 33.5 44.1-52.3X30-37.4
Shape index	no data	1.08	1.09	1.08	1.02	1.48
Wall	smooth	smooth	finely pitted	smooth	smooth	smooth
Micropyle	present	absent	absent	absent	absent	absent
Sporocyst shape	no data	ovoid	ovoid	no data	broadly ellipsoid	ovoid
Size (µm)	16.4-19.2 X 11.2 –13.6	16.5 X 12.6 16-17 X 12-13	20.8 X 14.5 19-23 X 13-16	13.3 X 10 12- 14 X 9-11	21.3 X 16.3 21-22X16-17	21.8 X 15.1 20 -22.6X14.5-16.4
Shape index	no data	1.31	1.44	1.34	1.30	1.44
Sporozoite size (µm)	no data	no data	9 X 2.5	no data	no data	16.6 X 3.7
Host	*Psammophis sibilans sibilans*	*Psammophis sibilans phillipsi*	*Psammophis sibilans phillipsi*	*Psammophis sibilans phillipsi*	*Psammophis schokari*	*Psammophis schokari*
Locality	Uganda	Liberia	Liberia	Liberia	Jordan	Saudi Arabia
Citation	Hoare (1933)	Bray (1960)	Bray (1960)	Bray (1960)	Modry *et.al.* (1999)	Present study

Type specimens: Syntypes (oocysts in 10% formalin) and a phototype are deposited in the parasitological collection, of the Zoology Department Museum, King Saud University, Riyadh, both as (KSUC.-117).

Etymology: The new species name reflects the species name of the host.

Discussion

Only five species of *Caryospora* have been described previously from members of the genus *Psammophis: Caryospora legeri* from *Psammophis sibilans sibilans* from Uganda (Hoare, 1933); *C. hernae; C. Psammophi* and *C. weyerae* from *Psammophis sibilans phillipsi* from Liberia (Bray, 1960) and *C. maxima* from *Psammophis schokari* from Jordan (Modry *et al.* 1999). In this study *Caryospora schokariensis* sp. n. is the second Caryosporian to be described from *Psammophis schokari.* The description of the new *Caryospora* species is mainly based on structure, geographic distribution and type of host (Table 1). *Caryospora schokariensis* sp.n. differs from all previously described species in the shape of the oocyst, *C. schokariensis* has elongate-ellipsoide oocysts, while all the other species have spherical to subspherical oocysts and in having much larger length to width ratios of the oocysts. It also differs from *Caryospora hermae, C. maxima, C. psammophi* and *C. weyerae* in having much larger oocysts and sporocysts. Moreover, *C. schokariensis* differs from *C. legeri* in lacking micropyle. The oocyst of *C. schokariensis* has smooth double-layered wall, whereas the outer layer of *C. psammophi* is finely irregularly pitted. *C. schokariensis* has larger Stieda body rather than reported for *C. maxima* which has larger substieda body. Besides the differences reported for the five *Caryospora* species parasitizing relatively closely related hosts, *C. schokariensis* differs from all other hitherto described caryosporan species from ophidian hosts in size, appearance, as well as in host location and geographical distribution. This indicates that *C. schokariensis* should be considered as a new species.

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References

Amoudi, M.A. 1989. Two new species of *Eimeria:* (Apicomplexa : Eimeriidae from the Saudi Arabian gray monitor (*Varanus griseus*). *J. Sci. Res.*, 11: 72-81.

Al-Rasheid, K.S. and Alyousif, M.S. 1998. *Eimeria yemenensae* n. sp.) Apicomplexa: Eimeriidae) from the rock agama *Agama yemenensis*) in Saudi Arabia. *Parasitol. Inter.*, 47: 127-130.

Al-Sadoon, M.K. 1989. Survey of the reptilian fauna of the Kingdom of Saudi Arabia I. The snake fauna of the central region. *J. Coll. Sci., King Saud Univ.*, 1: 53-69.

Alyousif, M.S. and Al-Rasheid, K.S. 2001. *Eimeria auratae* n. sp. (Apicomplexa : Eimeriidae) infecting the lizard *Mabuya aurata* in Saudi Arabia. *Parasitol. Inter.*, 50: 27-32.

Alyousif, M.S., and Al-Shawa, Y.R. 1997. *Isospora acanthodactyli* new species from *Acanthodactylus schmidti* with a new geographical record for *I. deserti* from *Agama pallida* (Finkelman and Paperna, 1994) in Saudi Arabia. *Pakistan. J. Zool.*, 29: 219-223.

Alyousif, M.S., and Al-Shawa, Y.R. 1998. A new coccidian parasite (Apicomplexa: Eimeriidae) from the legless lizard *Diplometopon zarudnyi* (Amphisbaenia: Trogonophidae) in Saudi Arabia. *J. Egypt. Soc. Parasitol.*, 28: 257-261.

Alyousif, M.S., Al-Sadoon, M.K. and Al-Shawa, Y. R. 1997. *Eimeria schmidti* n. sp. (Apicomplexa: Eimeriidae) from the sandy fringe-toed lizard (*Acan-*

thodactylus schmidti) in Saudi Arabia. *J. Egypt. Sco. Parasitol.*, 27: 465-469.

Bray, R.S. 1960. On the parasitic protozoa of Liberia III. Three new species of *Caryospora* Leger, 1904 from the hissing sand snake *Psammophis sibilans phillipsi* and a note on *Caryospora* in *Coluber ravergieri nummifer* from Israel. *J. Protozool.*, 7: 314-320.

Hoare, C.A. 1933. Studies on some new ophidian and avian coccidia from Uganda, with a revision of the classification of the Eimeriidae. *Parasitol.*, 25: 359-388.

Kasim, A.A. and Al-Shawa, Y.R. 1988. *Eimeria sinaitae* n. sp. (Apicomplexa: Eimeriidae) from the rock agama (*Agama sinaita*) in Saudi Arabia. *J. Protozool.*, 35: 388-389.

Matuschka, F.R. 1986. *Caryospora najadae* sp. n. (Apicomplexa: eimeriidae) from Dahl's whip snake. *Coluber najadum* (Serpentes: Colubridae). *Parasitol. Res.*, 72: 181-183.

Modry, D. and Koudela, B. 1998. Coccidia of genus *Caryospora* (Apicomplexa: Eimeriidae) from the long nosed vine snake, *Ahaetulla nasuta* (Serpentes: Colubridae: Boiginae) from southern India, with a description of *C. veselyi* sp. n. *Folia parasitol.*, 45: 167-169.

Modry, D., Koudela, B., Al-Oran, R.M. and Amr, Z. S. 1999. *Caryospora maxima* sp. n. (Apicomplexa: Eimeriidae), new coccidia from Forskal sand snake, *Psammophis schokari* (Serpentes: Colubridae) from Iava desert of Jordan. *Folia Parasitol.* 46: 96-98.

Upton, S.J., Freed, P.S., Burdick, D.A. and McAllister, C.T. 1990. Seven new species of coccidia (Apicomplexa: Eimeriidae) from Reptiles in Madagascar. *Can. J. Zool.*, 68: 2368-2375.

وصف نوع جديد من طفيليات الكاريوسبورا التي تصيب ثعبان ابوالسيور الرملي من عائلة الثعابين الحقيقية Family : Colubridae في المملكة العربية السعودية

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الملخص: تم في هذا البحث دراسة تواجد ووصف نوع جديد من طفيليات الكاريوسبورا وهو *Caryospora schokariensis* sp. n والذي يصيب الأمعاء الدقيقة لثعبان أبو السيور الرملي *Psammophis schokari* و ذلك من منطقة جازان بالمملكة العربية السعودية وقد تم التعرف على شكل وقياسات كل من الحويصلات والأكياس البوغية والأبواغ لهذا الطفيل وقد استنتج من هذه الدراسة ان هذا النوع من طفيليات الكوكسيديا لم يسبق تسجيله من قبل في هذا النوع من الثعابين في المملكة العربية السعودية.