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SOME NEW SPECIES OF *CARYOSPORA* (APICOMPLEXA: EIMERIIDAE) FROM BRAZILIAN SNAKES, AND A RE-DESCRIPTION OF *C. JARARACAE* CARINI, 1939

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The mature oocysts of six new species of Caryospora *are described from the faeces of Brazilian snakes. They are differentiated from other species previously recorded from reptiles, largely on the size and shape of the oocyst and sporocyst, structure of the oocyst wall, and presence or absence of a polar body.* C. paraensis *n. sp., and* C. carajasensis *n. sp., are from the "false coral",* Oxyrhopus petola digitalis*;* C. pseustesi *n. sp., from the "egg-eater",* Pseustes sulphureus sulphureus; C. epicratesi *n. sp., from the "red boa",* Epicrates cenchria cenchria; *and* C. micruri *n. sp., and* C. constanciae *n. sp., from the "coral snake",* Micrurus spixii spixii. *A re-description is given of* C. jararacae *Carini, 1939, from the "jararaca"* Bothrops atrox, *embodying some additional morphological features.*

Key words: Apicomplexa — Eimeriidae - *Caryospora* · *Caryospora carajasensis* n. sp. - *Caryospora paraensis* n. sp. - *Caryospora pseustesi* n. sp. - *Caryospora epicratesi* n. sp. - *Caryospora micruri* n. sp. - *Caryospora constanciae* n. sp. - *Caryospora jararacae* · *Oxyrhopus petola digitalis* · *Pseustes sulphureus sulphureus* · *Epicrates cenchria cenchria* · *Micrurus spixii spixii* · *Bothrops atrox* - coccidia - oocysts - snakes - Serpentes - Brazil

Oocysts of the coccidian *Caryospora* Léger, 1904 possess a single sporocyst containing 8 sporozoites. Levine (1988) lists 34 species, of which 19 are from snakes, 11 from birds, 2 from lizards and 1 each from a turtle and a rodent (Tables I and II for the reptilian species). There are representatives of the genus in both the Old and the New Worlds: the type species is *C. simplex* Léger, 1904, from the European snake *Vipera aspis*.

An examination of faeces from a number of snakes from Amazonian Brazil has revealed 7 different *Caryospora* species: 2 in *Oxyrhopus petola digitalis* (Reuss); 2 in *Micrurus spixii spixii* Wagler; and 1 each in *Epicrates cenchria cenchria* (Linn.), *Pseustes sulphureus sulphureus* (Wagler) and *Bothrops atrox* (Linn.). Six of these parasites are considered to represent new species of *Caryospora* and are described below. That in *B. atrox* is thought to most probably be *C. jararacae* Carini, 1939, and a redescription includes some additional morphological features not mentioned by that author.

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MATERIALS AND METHODS

Faecal material from *O. p. digitalis* and *P. s. sulphureus* was extracted from the lower part of the intestine on autopsy. That from the other snakes was removed from their cages, at various times during their captivity in the Herpetology Section of the Museu Paraense Emílio Goeldi, Belém, Pará. Gall-bladder contents were examined from the two autopsied snakes.

Faeces were gently triturated in 2% (w/v) aqueous $K_2Cr_2O_7$ solution and maintained as thin layers in covered petri-dishes kept at 24-26°C. The suspensions were examined without prior sieving, and $ZnSO_4$ flotation concentration was resorted to only when paucity of parasites made this necessary. Fifty oocysts and sporocysts of each *Caryospora* species were measured, using an ocular micrometer, x 8 eyepieces and a x 100 neofluar objective. Photomicrographs were prepared using a Zeiss

Microflash II and Ilford Pan F film. Line drawings were made during direct microscope observations, supplemented by details from photomicrographs. Measurements are in micrometers (μm): they are presented as means, with the standard deviations, the range in parentheses, followed by the shape-index (ratio of length/width).

RESULTS

Caryospora paraensis n. sp.
(Figs 1, 2 and 17)

Diagnosis — Oocysts predominantly spherical (76%) to subspherical (24%), 17.3 $\pm$ 0.7 x 17.0 $\pm$ 0.7 (16.2 – 18.7 x 15.0 – 18.7), shape-index 1.0 (1.0 – 1.1). Oocyst wall smooth, colourless, with no striations and of a single layer about 1.0 thick: no micropyle. No oocyst residuum, but with a single polar body of about 2.0 x 1.2, usually adhering to the oocyst wall. Sporocyst broadly ellipsoidal, 14.4 $\pm$ 0.6 x 10.8 $\pm$ 0.6 (13.7 – 16.2 x 10.0 – 12.5), shape-index 1.3 (1.2 – 1.5), with conspicuous Stieda and sub-Stieda bodies giving the composite structure a "stopper-like" appearance. The 8 sporozoites sometimes arranged around a central, compact sporocyst residuum, at other times irregularly disposed and with the residuum scattered. Refractile bodies present, but it remains uncertain if there are 1 or 2 of these in each sporocyst.

Type host — Oxyrhopus petola digitalis (Reuss) (Serpentes: Colubridae).

Location in host — Undetermined. As no oocysts were seen in the gall-bladder contents, the site of infection is most likely the intestinal tract. Oocysts described from the faeces.

Sporulation — No information.

Type material — Oocysts preserved in 10% formol-saline and held in the Department of Parasitology, Instituto Evandro Chagas, Belém, Pará, Brazil. Repository number NM84. In our hands fixation in formol-saline has not proved very suitable for subsequent morphological study.

Type locality — Serra dos Carajás, Pará State, north Brazil (6°S — 50°. 18'W).

Prevalence — No information.

Pathogenicity — No gross pathology was noticable at autopsy, but a histological study of the intestine has still to be made.

Etymology — The specific name is derived from the geographic location of the host, in Pará State.

Caryospora carajasensis n.sp.
(Figs 3, 4 and 18)

Diagnosis — Oocysts predominantly slightly subspherical (87%) to spherical (13%), 22.9 $\pm$ 0.8 x 21.5 $\pm$ 0.8 (20.0 – 25.0 x 18.7 – 23.7), shape-index 1.1 (1.0 – 1.2) Oocyst wall yellowish, about 1.2 thick and of 2 layers: an outer thicker one with a rough surface and bearing coarse striations (pores?), and an inner thin, smooth layer. No micropyle. No oocyst residuum or polar body. Sporocyst broadly ellipsoidal, 17.5 $\pm$ 0.6 x 12.9 $\pm$ 0.6 (16.2 – 18.7 x 11.2 – 13.7), shape index 1.3 (1.3 – 1.5). Stieda and sub-Stieda bodies together forming a "stopper-like" structure. The 8 sporozoites may encircle a central sporocyst residuum, like the staves of a barrel, or both sporozoites and residuum may be irregularly disposed. Refractile bodies present, but it could not be determined if there were one or two of these in each sporozoite.

Type host — Oxyrhopus petola digitalis (Reuss) (Serpentes: Colubridae).

Location in host — Uncertain, but probably the intestinal tract: no oocysts seen in the gall-bladder contents. Oocysts described from the faeces.

Sporulation — No information.

Type material — Oocysts in 10% formol-saline, and held in the Parasitology Department of the Instituto Evandro Chagas, Belém, Pará, Brazil. Repository number NM84a.

Type locality — Serra dos Carajás, Pará State, north Brazil (6° S — 50. 18'W).

Prevalence — No information.

Pathogenicity — No gross pathology discernable in the intestine.

Etymology — Specific name derived from the locality known as the Serra dos Carajás in which

Caryospora pseustesi n.sp.
(Fig. 19*)

Diagnosis — Oocysts slightly subspherical (60%) to spherical (40%), 26.4 ± 0.8 x 25.4 ± 0.7 (25.0 – 27.5 x 23.7 – 27.5), shape-index 1.0 (1.0 – 1.1). Oocyst wall golden yellow in colour, about 1.7 thick and with no micropyle. It is seemingly of 2 layers as viewed by bright-light microscopy: an inner thicker layer with prominent striations, and an outer smooth, fine layer. No oocyst residuum or polar body. Sporocyst broadly ellipsoidal, 18.9 ± 0.7 x 13.8 ± 0.6 (17.5 – 20.0 x 12.5 – 15.0), shape-index 1.4 (1.2 – 1.4), with conspicuous Stieda and sub-Stieda bodies forming a "stopper-like" structure. The 8 sporozoites are irregularly disposed and the sporocyst residuum tends to be dispersed. Refractile bodies present, but it could not be determined if there were 1 or 2 in each sporozoite.

Type host — Pseustes sulphureus sulphureus (Wagler) (Serpentes: Colubridae).

Location in host — As no oocysts were found in the gall-bladder contents, development of the parasite is most likely in the intestinal tract. Oocysts described in the faeces.

Sporulation — No information.

Type material — Oocysts in 10% formol-saline, held in the Department of Parasitology, Instituto Evandro Chagas, Belém, Pará, Brazil.

Type locality — Serra dos Carajás, Pará State, north Brazil (6° S — 50°. 18'W).

Prevalence — No information.

Pathogenicity — No gross pathology seen in the autopsied snake.

Etymology — The specific name is derived from the generic name of the snake, ***Pseustes***, in which the parasite was found.

Caryospora epicratesi n.sp.
(Figs 5, 6 and 20)

Diagnosis — Predominantly subspherical (82%) to spherical (18%), 22.9 ± 1.5 x 21.4 ± 1.2 (18.7 – 25.0 x 17.5 – 22.5), shape-index 1.1 (1.0 – 1.2). Oocyst wall smooth, colourless, with no micropyle and approximately 1.5 thick. It is composed of 2 layers, as seen by bright light microscopy: an inner thin, non-striated layer of about 0.5, and an outer thicker one which is striated and about 1.0 thick. No oocyst residuum, but a single polar body of approximately 2.3 x 2.0 is a constant feature and most frequently is seen adhering to the wall of the sporocyst (67%) rather than that of the oocyst (36%). Sporocyst broadly ellipsoidal, 17.6 ± 0.7 x 12.5 ± 0.2 (16.2 – 18.7 x 11.2 – 12.5), shape-index 1.4 (1.3 – 1.5). Stieda and sub-Stieda bodies together forming a conspicuous "stopper-like" structure. The 8 sporozoites are usually arranged about a prominent sporocyst residuum, like the staves of a barrel. Anterior and posterior refractile bodies present in each sporozoite.

Type host — Epicrates cenchria cenchria (Linn.) (Serpentes: Boidae).

Location in host — Uncertain. Oocysts described in faeces from the living host.

Sporulation — Exogenous: no other information.

Type material — Oocysts in 10% formol-saline and held in the Department of Parasitology, Instituto Evandro Chagas, Belém, Pará, Brazil. Repository number FN62.

Type locality — Area of the hydroelectric dam "Samuel", Rondônia, Brazil (8° 45'S: 63° 28'W).

Prevalence — No information.

Pathogenicity — The snake appeared to be in good health: faeces normal.

Etymology — The specific name is derived from the generic name of the snake, ***Epicrates***, from which the parasite is described.

Caryospora jararacae Carini, 1939
(Figs 7, 8 and 21)

Diagnosis — Oocysts predominantly spherical (86%), occasionally slightly subspherical (14%), 13.0 ± 0.5 x 13.0 ± 0.4 (12.5 – 14.4 x 12.5 – 14.4), shape-index 1.0 (1.0 – 1.1). Oocyst wall smooth, colourless and of a single layer about 0.7 thick, with no striations: no micropyle. No oocyst residuum: the single polar body usually adheres to

* Photomicrographs regrettably lost: line drawing only.

the oocyst wall, and measures about 1.2 x 1.0. Sporocyst broadly ellipsoidal, 10.0 ± 0.3 x 8.7 ± 0.3 (8.7 – 11.2 x 7.5 – 8.7), shape-index 1.1 (1.1 – 1.3), with a delicate wall bearing an inconspicuous "stopper-like" structure formed by the Stieda and sub-Stieda bodies. Sporocyst residuum of about 20 spherical granules, encompassed by the 8 sporozoites, which are usually arranged longitudinally. Sporozoites with anterior and posterior refractile bodies, and with delicate transverse cytoplasmic corrugations giving a "concertina" effect.

Recorded hosts — Type host is ***Bothrops jararaca*** (Weid): present description is from ***Bothrops atrox*** (Linn.) (Serpentes: Viperidae).

Location in host — Probably the intestinal tract, although Carini (1939) was unable to find developmental stages of the parasite in histological sections of the intestine. In our case, oocysts were described in faeces from the living snake.

Sporulation — Exogenous: no other information.

Neotype material — Oocysts in 10% formol-saline, held in the Parasitology Department of the Instituto Evandro Chagas, Belém, Pará, Brazil. Repository number FN25.

Geographic distribution — Mogi das Cruzes, São Paulo State, Brazil (23°. 31'S: 46°. 11'W) is the type locality: neotype locality is the area of the hydroelectric dam "Balbina", Amazonas State, Brazil (1°.55'S: 59°.28'W).

Prevalence — No information for the infection in *B. jararaca*. One out of 19 *B. atrox* (11%) was infected in the present study.

Pathogenicity — No signs of disease were noted in the infected snake.

Etymology — Carini derived the specific name from that of the type host, *B. jararaca*. Synonym: ***Caryospora jaracae*** (***lapsus calami***, Carini, 1939).

Remarks — The measurements and morphology of the oocysts and sporocysts described by Carini are so similar those of the parasite we have recorded in *B. atrox* that we feel we are most probably dealing with the same organism, namely *C. jararacae*. This is especially likely in that the oocysts are from two species of the same reptilian genus (***Bothrops***).

That Carini did not figure a polar body, sub-Stieda body or the peculiar transverse striations of the sporozoites is not surprising. These structures can be difficult to see in an oocyst as small as.that of *C. jararacae*.

Caryospora micruri n.sp.
(Figs 9, 10, 16 and 22)

Diagnosis — Oocysts predominantly spherical (68%) to slightly subspherical (32%), 16.1 ± 0.6 x 16.0 ± 0.6 (14.4 – 17.5 x 13.7 – 17.5), shape-index 1.0 (1.0 – 1.1). Oocyst wall a single, colourless layer of about 0.6, with no striations and no micropyle. No oocyst residuum, but commonly with a single polar body of about 2.5 x 1.2, usually adhering to the oocyst wall. Sporocyst broadly ellipsoidal, 13.7 ± 0.5 x 10.2 ± 0.3 (12.5 – 15.0 x 9.4 – 11.2), shape-index 1.3 (1.2 – 1.4) with a delicate wall bearing a "stopper-like" structure formed by the Stieda and sub-Stieda bodies. The 8 sporozoites are sometimes arranged longitudinally, to enclose a central residuum, and on other occasions appear to be irregularly disposed: they possess both anterior and posterior refractile bodies.

Type host — Micrurus spixii spixii Wagler (Serpentes: Elapidae).

Location in host — No information.

Sporulation — No information.

Type material — Oocysts in 10% formol-saline, held in the Department of Parasitology, Instituto Evandro Chagas, Belém, Pará, Brazil. Repository number FN60.

Type locality — Area of the hydroelectric dam "Samuel", Rondônia, Brazil (8°45'S: 63°28'W).

Prevalence — No information.

Pathogenicity — The snake showed no signs of illness: faeces normal. Oocysts described in faeces from the living snake.

Etymology — The specific name is derived from the generic name of the host, ***Micrurus***, in which the parasite was found.

Caryospora constanciae n. sp.
(Figs 11, 15 and 23)

Diagnosis — Oocysts predominantly subspherical (88%) to spherical (12%), 20.5 ± 0.7 x 19.2 ± 0.6 (18.7 - 22.5 x 17.5 - 20.0), shape-index 1.1 (1.0 - 1.1). Oocyst wall at first with 2 layers: an outer irregular layer, which is soon lost, and an inner smooth layer. With the two layers the wall is approximately 1.9 - 2.5. The great majority of the oocysts, however, are seen without the outer layer and the smooth, colourless inner wall then measures about 1.2: it has no striations and there is no micropyle. No oocyst residuum, but a single polar body of about 2.5 x 1.2 is attached to the oocyst wall in most specimens. Sporocyst broadly ellipsoidal, 16.1 ± 0.3 x 11.4 ± 0.3 (15.0 - 17.5 x 10.6 - 12.5), shape-index 1.4 (1.3 - 1.5), with a prominent "stopper-like" structure formed by the Stieda and sub-Stieda bodies. The 8 sporozoites are usually disposed longitudinally around the central sporocyst residuum and they possess both anterior and posterior refractile bodies.

Type host — ***Micrurus spixii spixii*** Wagler (Serpentes: Elapidae).

Location in host — No information.

Sporulation — No information.

Type material — Oocysts in 10% formol-saline, held in the Department of Parasitology, Instituto Evandro Chagas, Belém, Pará, Brazil. Repository number FN60a.

Type locality — Area of the hydroelectric dam "Samuel", Rondônia, Brazil (8°45'S: 63°28'W).

Prevalence — No information.

Pathogenicity — The snake showed no signs of disease. Oocysts described from the faeces.

Etymology — The specific name is given as a tribute to Constância M. Franco for her excellent technical assistance.

TABLE I

Known species of *Caryospora* from reptilian hosts

Species	Author(s)	Type Host (other recorded hosts)	Recorded Geographic Distribution
C. bengalensis	Mandal (1976)	*Enhydris enhydris* (Serpentes: Colubridae)	India
C. bigenetica	Wacha & Christiansen (1982)	*Crotalus horridus* (*Sistrurus catenatus*) (Serpentes: Viperidae)	North America
C. brasiliensis	Carini (1932) Lainson & Shaw (1973)	*Philodryas aestivus* (*P. olfersi*; *P. nattereri*: *Liophis* (= *Leimadophis*) *poecilogyrus*) (Serpentes: Colubridae)	Brazil
C. carajasensis n.sp.	This paper	*Oxyrhopus petola digitalis* (Colubridae)	Brazil
C. cheloniae	Leibovitz et al. (1978)	*Chelonia m. mydas* (Chelonia: Cheloniidae)	Grand Cayman, W.I.
C. cobrae	Nandi (1985)	*Naja naja* (Serpentes: Elapidae)	India
C. colubris	Matuschka (1984a)	*Coluber viridiflavus* (Colubridae)	Italy

Species	Author(s)	Type Host (other recorded hosts)	Recorded Geographic Distribution
C. constanciae n. sp.	This paper	***Micrurus s. spixii*** (Elapidae)	Brazil
C. corallae	Matuschka (1984b)	*Corallus caninus* (Serpentes: Boidae)	French Guyana
C. demansiae	Cannon (1967)	***Demansia psammophis*** (Elapidae)	Australia
C. dendrelaphis	Cannon & Rzepczyk (1974)	***Dendrelaphis punctulatus*** (Colubridae)	Australia
C. duszynskii	Upton et al., (1984a)	***Elaphe guttata*** (***E.o.obsoleta***) (Colubridae)	North America
C. epicratesi n.sp.	This paper	***Epicrates c. cenchria*** (Boidae)	Brazil
C. ernsti	Upton et al., (1984b)	***Anolis carolinensis*** (Sauria: Iguanidae)	North America
C. gekkonis	Chakravarty & Kar (1947)	*Gekko gecko* (Sauria: Gekkonidae)	India
C. hermae	Bray (1960)	***Psammophis sibilans phillipsi*** (Colubridae)	Liberia
C. japonicum	Matsubayashi (1936)	***Natrix tigrina*** (Colubridae)	Japan
C. jararacae	Carini (1939) This paper	***Bothrops jararaca*** (***B. atrox***) (Viperidae)	***Brazil***
C. lampropeltis	Anderson et al., (1968)	***Lampropeltis c. calligaster*** (Colubridae)	North America
C. legeri	Hoare (1933)	***Psammophis sibilans*** (Colubridae)	Uganda
C. micruri n.sp.	This paper	***Micrurus s. spixii*** (Elapidae)	Brazil
C. najadae	Matuschka (1986a)	***Coluber najadum*** (Colubridae)	Israel
C. najae	Matuschka (1982)	***Naja nigricollis pallida*** (Elapidae)	East Africa
C. paraensis n.sp.	This paper	***Oxyrhopus petola digitalis*** (Colubridae)	Brazil
C. psammophi	Bray (1960)	***Psammophis s. phillipsi*** (Colubridae)	Liberia

Species	Author(s)	Type Host (other recorded hosts)	Recorded Geographic Distribution
C. pseustesi n. sp.	This paper	***Pseustes s. sulphureus*** (Colubridae)	Brazil
C. simplex ***TYPE SPECIES***	Léger (1904, 1911) Upton et al. (1983)	*Vipera aspis* (*V.x. xanthina* (Viperidae)	France
C. telescopis	Matuschka (1986b)	*Telescopus fallax* (Colubridae)	Greece
C. weyerae	Bray (1960)	*Psammophis s. phillipsi* (Colubridae)	Liberia
C. zuckermanae	Bray (1960)	*Coluber ravergieri nummifer* (Colubridae)	Israel

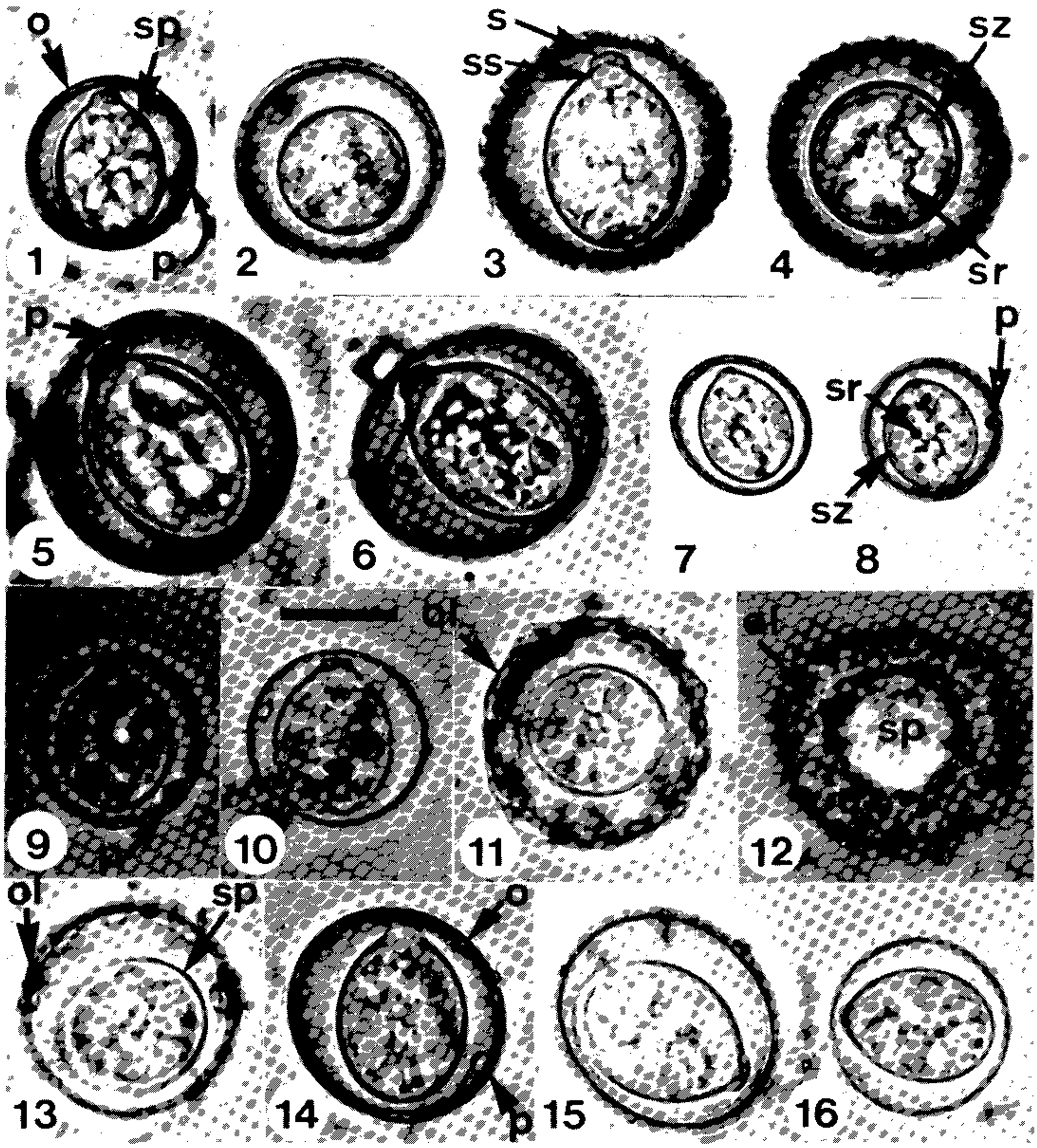
TABLE II

Available descriptions of the known *Caryospora* species of reptiles

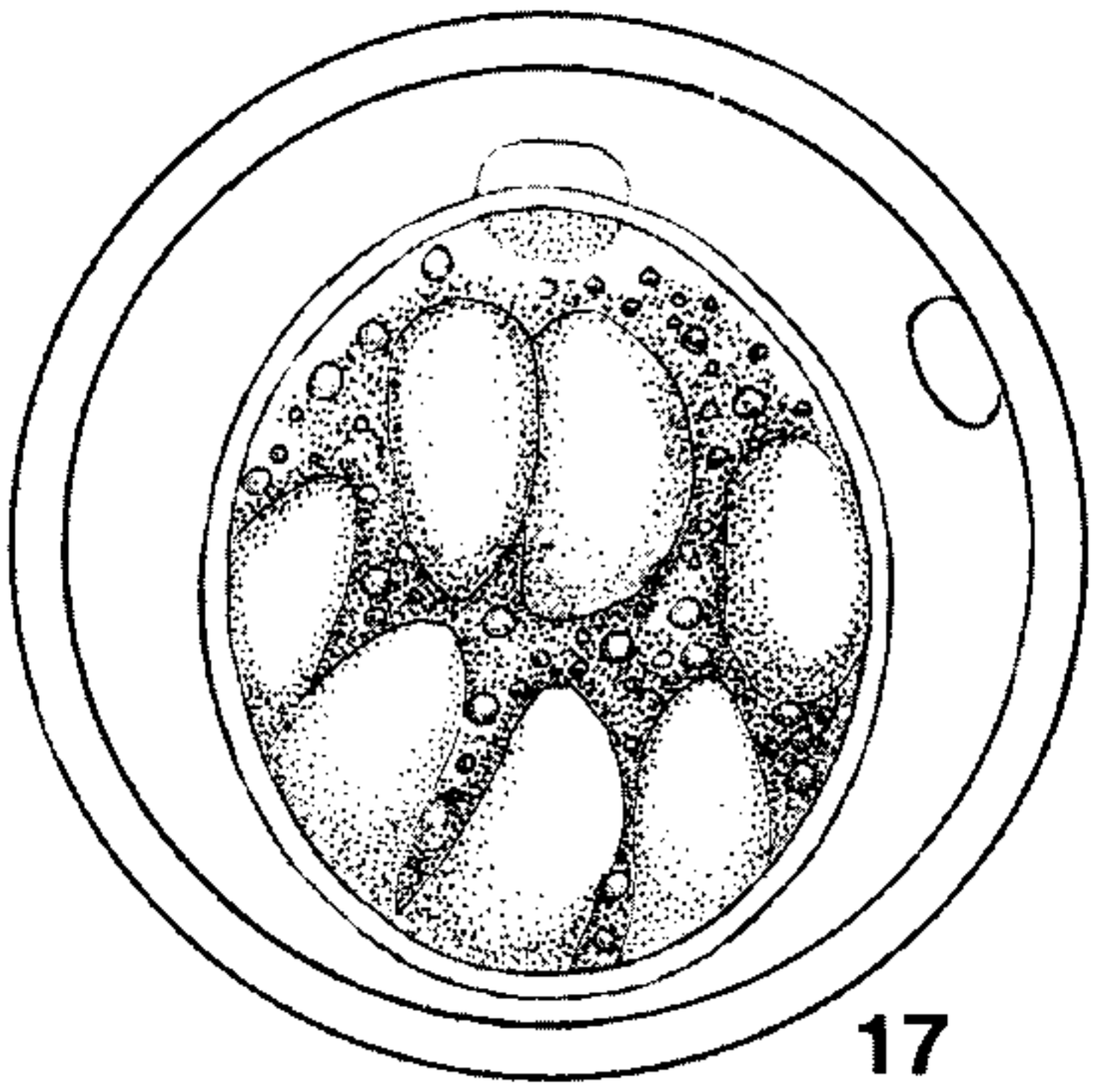
	Oocyst				*Sporocyst*
Species	Shape Size (μm)	Polar Body	Wall (μm); colour; Micropyle (M); no. of Layers (L)	Size (μm)	Stieda Body SubStieda Body (S/SS)
C. bengalensis	Spherical to Sub-spherical; 21.5 (20.0 – 22.5)	0	?; colourless; M; smooth	19.0 x 13.5	S SS?
C. bigenetica	Spherical; 13.2 (10.9 – 15.4)	1	1.0; colourless; 2L outer striated; surface mamillated	10.1 x 7.7 (8.3 – 11.5 x 6.4 – 9.0)	S/SS
C. brasiliensis	Spherical to Sub-spherical; 20.0 – 22.0 (Carini 1932) 21.7 x 20.8 (19.0 – 24.0 x 18.0 – 24.0) Lainson & Shaw, 1973	0	1.0 – 2.0; colourless; smooth; 3L, middle layer striated	17.0 x 13.0 (Carini). 16.1 x 12.2 (16.0 – 19.0 x 12.0 – 18.0) (Lainson & Shaw)	S SS?
C. cheloniae	Highly elongated; 37.4 x 12.8 (33.8 – 40.1 x 11.0 – 14.6)	0	0.3 – 0.6; colourless; Usually ruptures to leave sporocysts free	34.5 x 12.7 (26.2 – 44.1 x 10.6 – 17.3)	S/SS
C. carajasensis n. sp.	Spherical to Sub-spherical; 22.9 x 21.5 (20.0 – 25.0 x 18.7 – 23.7)	0	1.2; yellowish; 2L, outer thicker one with striations	17.5 x 12.9 (16.2 – 18.7 x 11.2 – 13.7)	S/SS

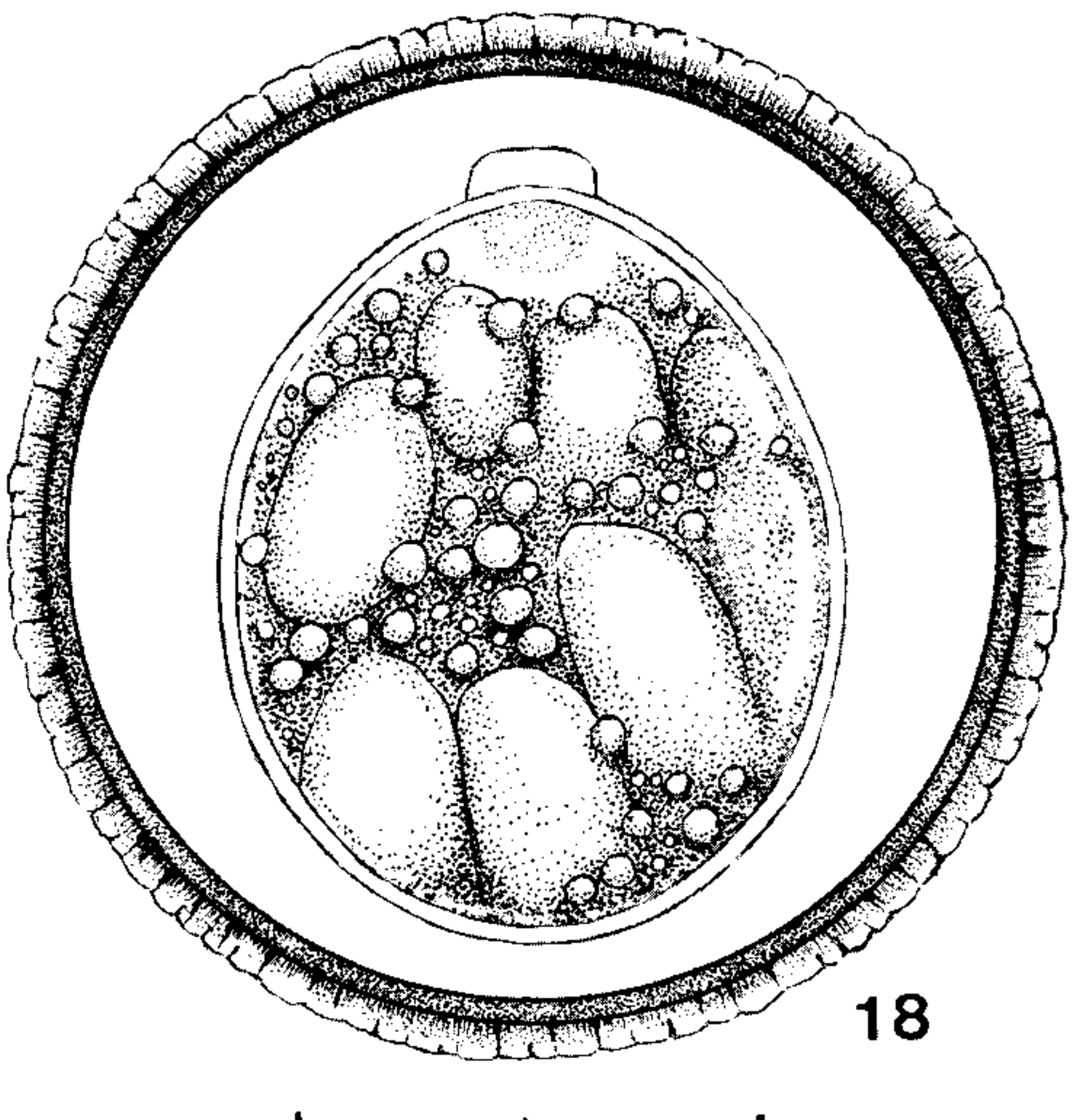
Species	Oocyst: Shape/ Size (μm)	Oocyst: Polar Body	Oocyst: Wall (μm); colour; Micropyle (M); no. of Layers (L)	Sporocyst: Size (μm)	Sporocyst: Stieda Body SubStieda Body (S/SS)
C. cobrae	Spherical to Sub-spherical; 18.5 x 17.2 (16.5 – 19.5 x 16.5 – 18.0)	0	?; colourless; 2L; smooth; M	14.2 x 11.8 (12.0 – 16.5 x 9.0 – 12.8)	S
C. colubris	Spherical to Sub-spherical; 16.2 – 27.9	0	1.0; smooth; 2L; colourless	13.2 – 19.1 x 10.3 – 14.7	S/SS
C. constanciae n. sp.	Spherical to Sub-spherical; 20.5 x 19.2 (18.7 – 22.5 x 17.5 – 20.0)	1	1.9 – 2.5; colourless; 2L, outer one rough and usually shed. Inner one smooth, 1.2	16.1 x 11.4 (15.0 – 17.5 x 10.6 – 12.5)	S/SS
C. corallae	Spherical; 22.4 (18.7 – 24.6);	1	1.0; smooth; 2L colourless	19.1 x 13.1 (17.6 – 20.0 x 11.7 – 14.0)	S
C. desmansiae	Subspherical; 20.4 x 19.8 (16.0 – 27.0 x 14.0 – 27.0)	1	0.6; colourless; 2L	13.1 x 10.8 (11.1 – 15.6 x 8.3 – 12.8)	S/SS
C. dendrelaphis	Spherical to Sub-spherical; 21.4 x 20.7 (12.2 – 25.5 x 12.2 – 24.0)	0	1.0; colourless; smooth; 2L	14.8 x 11.7 (10.3 – 18.0 x 8.5 – 14.1)	S/SS
C. duszynskii	Spherical to Sub-spherical; 25.7 x 24.3 (23.0 – 28.5 x 22.0 – 28.0)	1 or 2	1.5; yellow/orange; 2L, outer layer striated	18.3 x 14.8 (17.0 – 21.5 x 13.5 – 16.5)	S/SS
C. epicratesi n. sp.	Spherical to Sub-spherical; 22.9 x 21.4 (18.7 – 25.0 x 17.5-22.5)	1	1.5; colourless; 2L, outer layer striated	17.6 x 12.5 (16.2 – 18.7 x 11.2 – 12.5)	S/SS
C. ernsti	Spherical; 12.5 (11.0 – 14.5)	1	1.0; colourless; 1L	10.7 x 8.3 (10.0 – 12.5 x 7.5 – 9.0)	S/SS
C. gekkonis	Spherical; 19.8	0	?; 2L; M	"almost spherical" 11.0 in diameter	S SS?
C. hermae	Subspherical 22.3 x 20.6 (21.0 – 24.0 x 20.0 – 22.0)	0	?; colourless; smooth; 2L	16.0 – 17.0 x 12.0 – 13.0	S/SS
C. japonicum	Spherical to Sub-spherical; 18.6 (14.6 – 21.9)	0	?; dark-light brown; 2L	?	S/SS

Species	*Oocyst* Shape/Size (μm)	Polar Body	Wall (μm); colour; Micropyle (M); no. of Layers (L)	*Sporocyst* Size (μm)	Stieda Body SubStieda Body (S/SS)
C. jararacae	Spherical; 13.0–14.0 (Carini 1939) 13.0 (12.5–14.4 x 12.5–14.4) (this paper)	1 (This paper)	0.7; colourless; smooth; 1L	9.0–10.0 x 7.0–8.0 (Carini); 10.0 x 8.7 (8.7–11.2 x 7.5–8.7) (this paper)	S/SS
C. lampropeltis	Spherical; 23.3 (20.0–25.0)	1	1.0; yellow/green; M; 2L, outer layer striated	17.1 x 12.3 (15.0–19.0 x 11.0–13.0)	S/SS
C. legeri	Subspherical; 21.0–30.0 x 19.0–26.0	0	"thick"; 2L; M	16.4–19.2 x 11.2–13.6	S/SS
C. micruri n. sp.	Spherical to Subspherical; 16.1 x 16.0 (14.4–17.5 x 13.7–17.5)	1	0.6; colourless; smooth; 1L	13.7 x 10.2 (12.5–15.0 x 9.4–11.2)	S/SS
C. najadae	Spherical; 31.9 (27.9–36.3)	0	1.5–2.0; smooth; 2L	21.1 x 15.2 (19.9–22.2 x 14.0–16.4)	S/SS
C. najae	Spherical; 15.1 (13.8–16.1)	0	1.0; smooth; 2L, inner dark and outer colourless	11.2 x 8.8 (10.7–12.3 x 8.2–9.4)	S/SS
C. paraensis n. sp.	Spherical to Subspherical; 17.3 x 17.0 (16.2 – 18.7 x 15.0 – 18.7)	1	1.0; colourless; smooth; 1L	14.4 x 10.8 (13.7–16.2 x 10.0–12.5)	S/SS
C. psammophi	Subspherical; 29.1 x 26.8 (25.0–34.0 x 23.0–31.0)	0	?; yellow; smooth; 2L, outer layer with striations	20.8 x 14.5 (19.0–23.0 x 13.0–16.0)	S/SS
C. pseustesi n. sp.	Spherical to Subspherical; 26.4 x 25.4 (25.0–27.5 x 23.7–27.5)	0	1.7; Golden yellow 2L, inner one with striations; smooth	18.9 x 13.8 (17.5–20.0 x 12.5–15.0)	S/SS
C. simplex	Spherical; 10.0–15.0 (Léger);	1 or 2	1.4; yellowish; 1L, lightly striated	12.0 x 8.0 (Léger) 11.6 x 8.9 (10.4–	S/SS
TYPE SPECIES	14.9 (13.5–16.2) (Upton et al. 1983)			12.6 x 8.1–9.5) (Upton et al., 1983)	
C. telescopis	Spherical; 21.5 (19.1–23.5)	0	1.0; smooth; 2L	15.0 x 11.4 (14.7–16.2 x 10.3–11.8)	S SS
C. weyerae	Subspherical; 16.1 x 14.9 (14.0–18.0 x 13.0–17.0)	0	?; colourless; smooth; 2L	13.3 x 10.0 (12.0–14.0 x 9.0–11.0)	S SS
C. zuckermanae	Spherical; 15.0–18.0	?	?; Wall with loose, crumpled outer coat	?	?

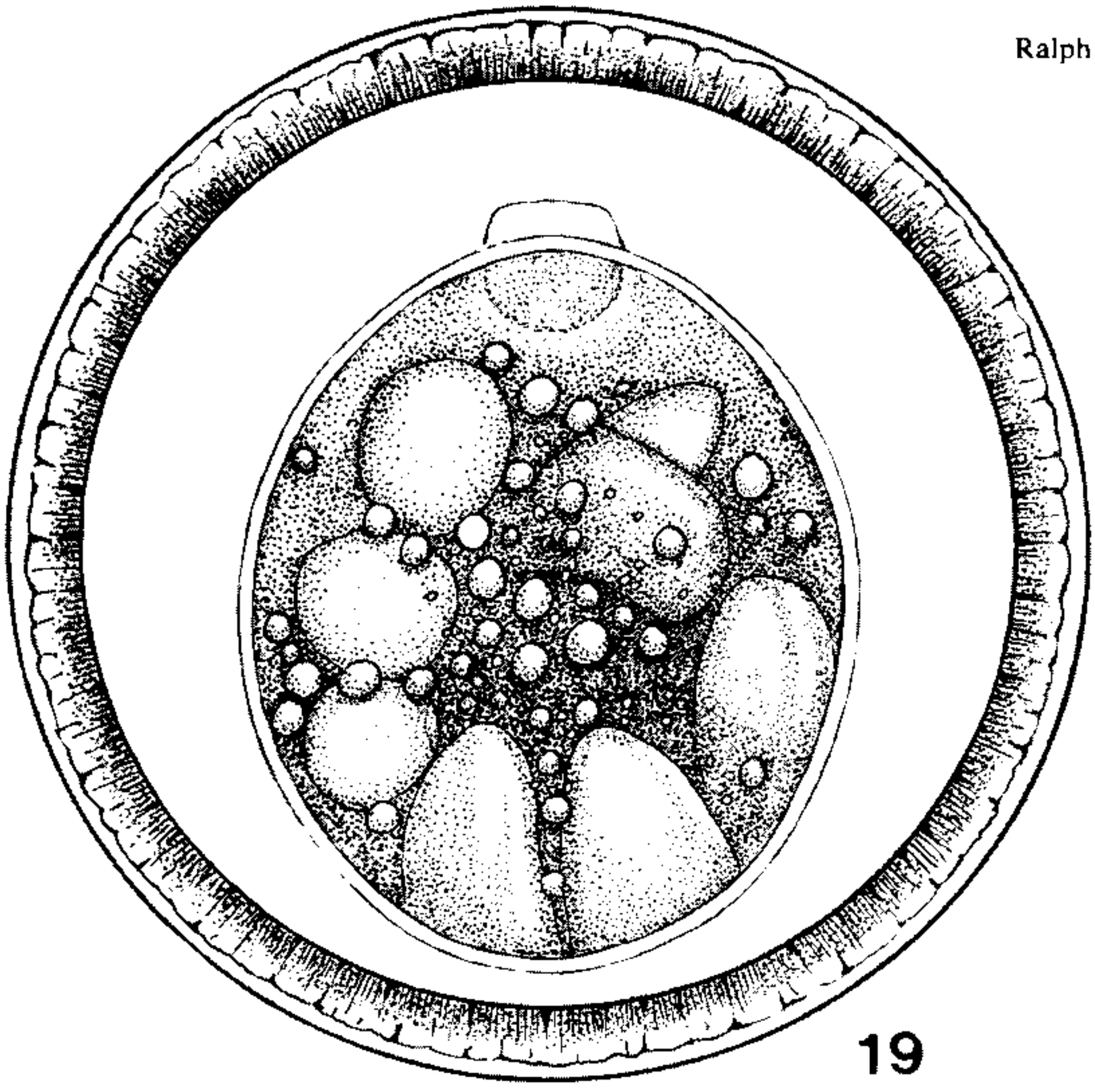


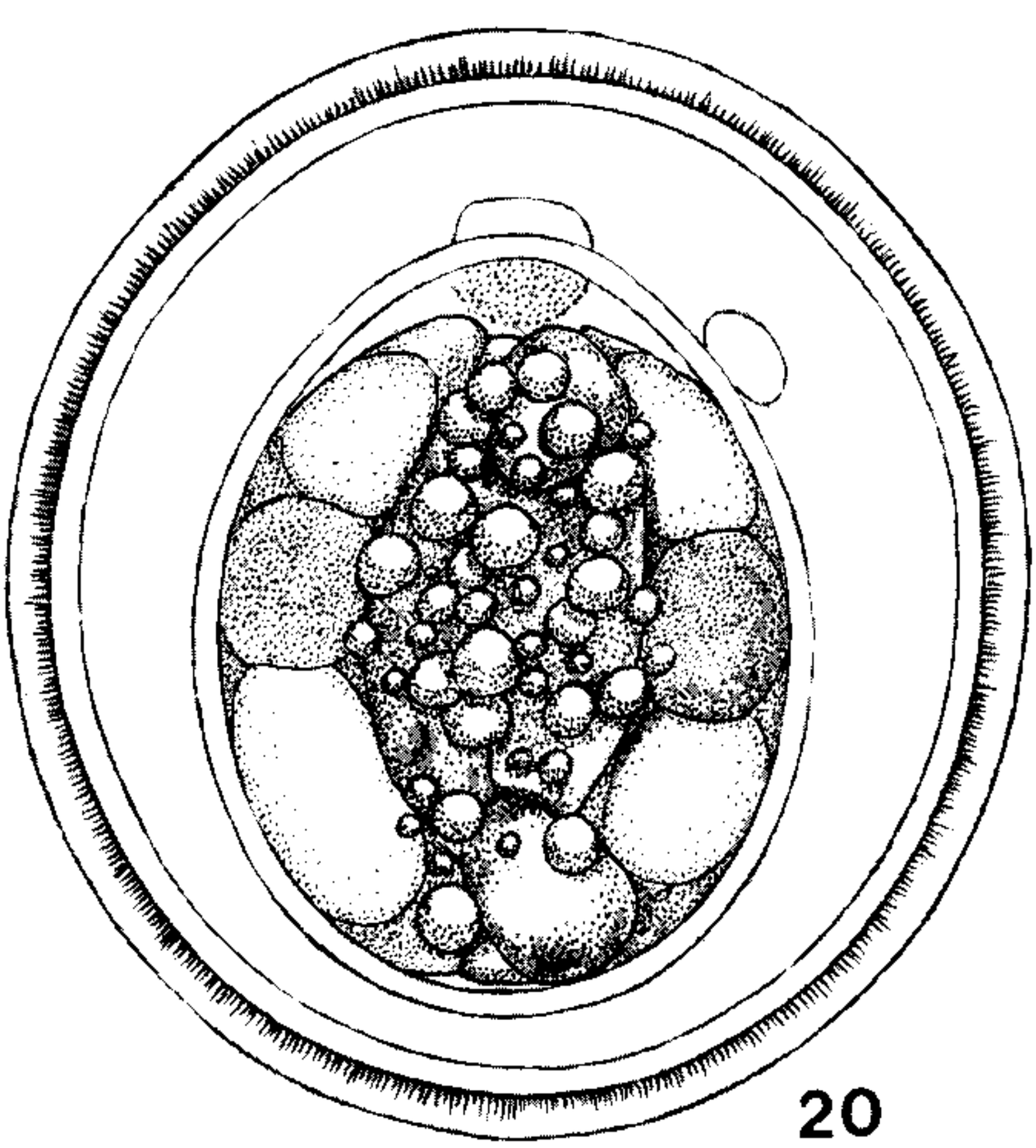
Photomicrographs of mature oocysts of *Caryospora* species from the faeces of Brazilian snakes: bright field microscopy. Figs 1 and 2: *C. paraensis* n.sp., from *Oxyrhopus petola digitalis*. Figs 3 and 4: *C. carajasensis n.sp.*, from the same snake. Figs 5 and 6: *C. pseustesi* n.sp., from *Pseustes s. sulphureus*. Figs 7 and 8: *C. jararacae* Carini, 1939, from *Bothrops atrox*. Figs 9, 10 and 16: *C. micruri* n.sp., from *Micrurus s. spixii*. Figs 11-15: *C. constanciae* n.sp., from the same snake. Figs 11 and 12 show the thick, irregular outer layer (OL) of the oocyst wall of *C. constanciae* in sectional and surface view, respectively: only remnants of it remain in Fig. 13 and it has been completely lost in Figs 14 and 15. In Figs 2, 4, 11 and 13, sporocysts are seen end-on, showing the longitudinal disposition of the sporozoites around the central sporocyst residuum. Bar = 1.0 μm; P = polar body; S = Stieda body; SS = sub-Stieda body; O = oocyst wall; SP = sporocyst; SR = sporocyst residuum; SZ = sporozoites.



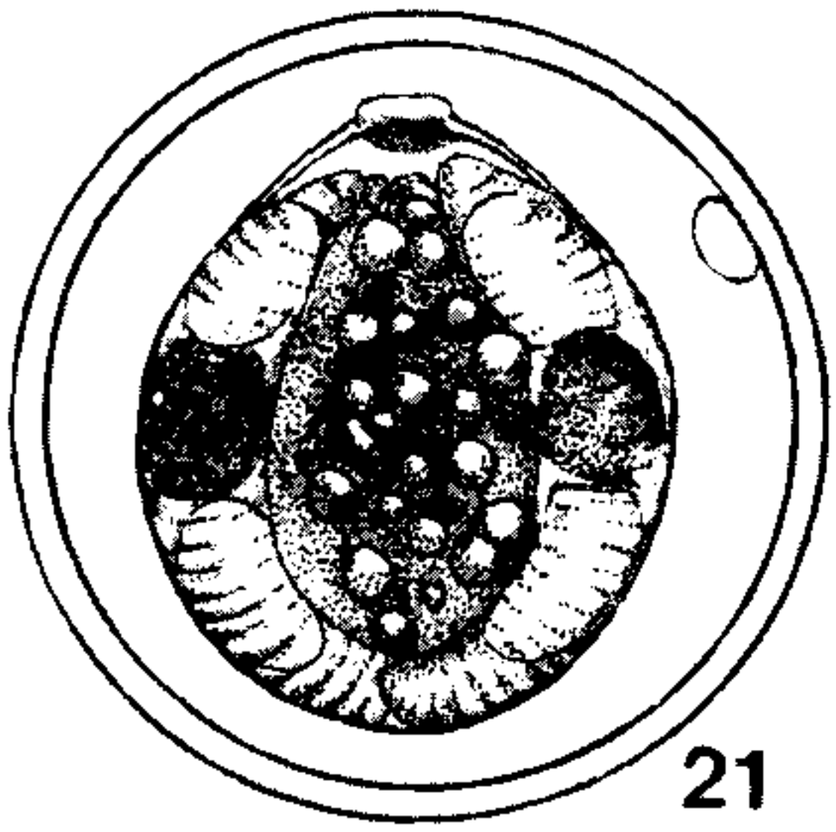


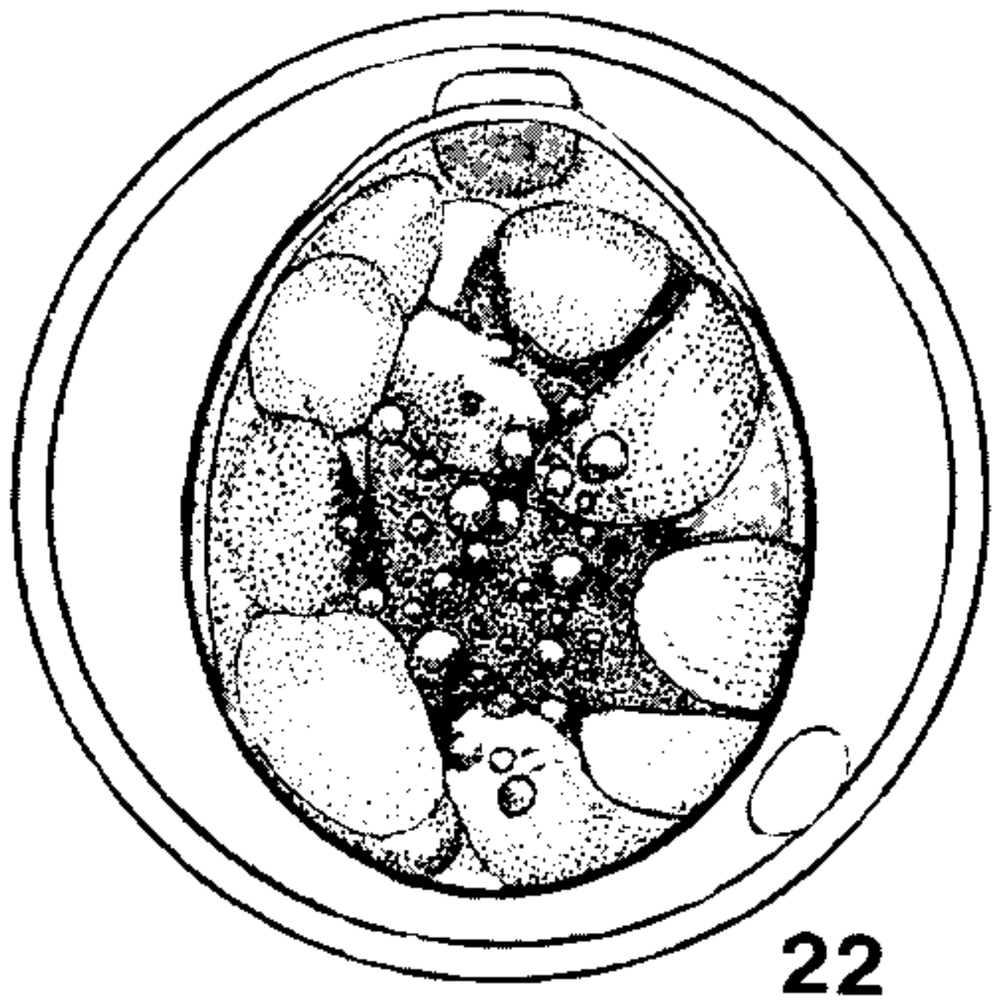
Line drawings of mature oocysts of *Caryospora* species from Brazilian snakes. Fig. 17: *Caryospora paraensis* n.sp. Fig. 18: *Caryospora carajasensis* n.sp.

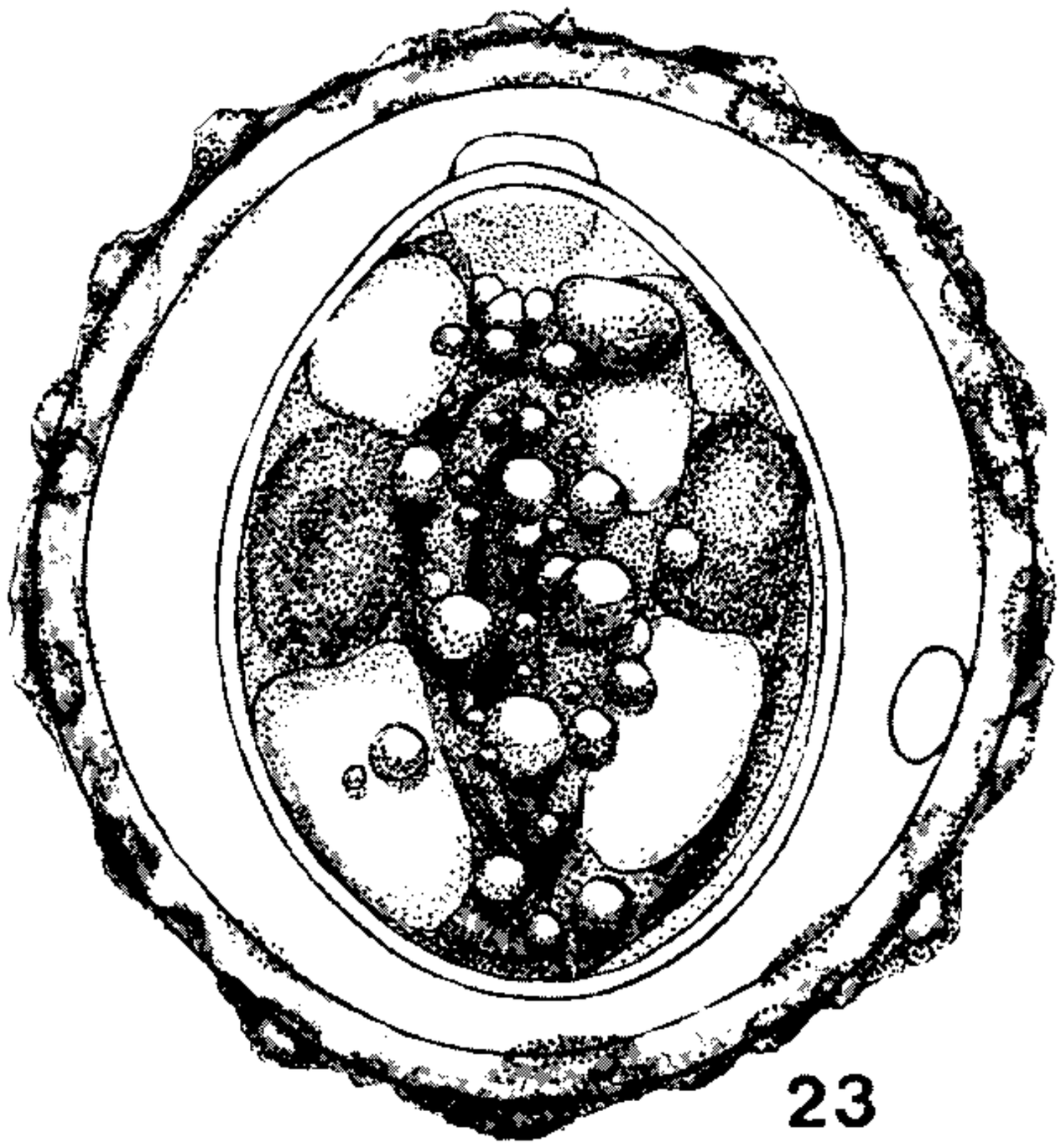




Line drawings of mature oocysts of *Caryospora* species from Brazilian snakes. Fig. 19: ***Caryospora pseustesi*** n.sp. Fig. 20: ***Caryospora epicratesi*** n.sp.







Line drawings of mature oocysts of *Caryospora* species from Brazilian snakes. Fig. 21: *Caryospora jararacae* Carini, 1939. Fig. 22: *Caryospora micruri* n.sp. Fig. 23: *Caryospora constanciae* n.sp.

DISCUSSION

The 6 new species of *Caryospora* described above are separated from each other by the following features.

Neither *C. carajasensis* nor *C. pseustesi* possess a polar body, but they can be separated from each other by their different oocyst and sporocyst measurements (22.9 x 21.5 vs 26.4 x 25.4; and 17.5 x 12.9 vs 18.9 x 13.8, respectively).

Among the 4 species possessing a polar body, *C. constanciae* is notable for the irregular outer layer of its oocyst wall: in size it is closest to *C. epicratesi*, but also separated from that parasite by the absence of oocyst wall striations.

Caryospora micruri most closely resembles *C. paraensis*, but its oocysts and sporocysts are smaller (16.1 x 16.0 vs 17.3 x 17.0; and 13.7 x 10.2 vs 14.4 x 10.8, respectively). The oocyst wall of *C. micruri* is delicate (0.6) and tends to crumple, whereas that of *C. paraensis* is thicker (1.0) and more resistant. The oocysts of both of these coccidia are much smaller than those of *C. constanciae* (20.5 x 19.2) and *C. epicratesi* (22.9 x 21.4).

Of the 24 *Caryospora* species previously described from reptiles (Tables I and II), we can readily differentiate the following from the new species described in this paper.

Caryospora bengalensis, C. cobrae, C. gekkonis, C. lampropeltis and *C. legeri* by their possession of an oocyst micropyle; *C. cheloniae* by its elongated oocysts (37.4 x 12.8); and *C. japonicum* by its dark to light brown oocysts.

Caryospora duszynskii, C. psammophi and C. simplex have oocysts described as "yellow-orange", "yellow" and "yellowish", respectively, and need to be differentiated, therefore, only from *C. carajasensis* and *C. pseustesi*, both of which have a yellow oocyst wall. *C. duszynskii* is distinguished by the possession of 1-2 polar bodies, which are absent in both *C. carajasensis* and *C. pseustesi. C. psammophi* can be separated from *C. pseustesi* by its larger oocyst (29.1 x 26.8 vs 26.4 x 25.4) and sporocyst (20.8 x 14.4 vs 18.9 x 13.8) and lack of a polar body; and from *C. carajasensis* by its much larger oocyst (29.1 x 26.8 vs 22.9 x 21.5). Finally, *C. simplex* differs from both *C. carajasensis* and *C. pseustesi* by its very much smaller, spherical oocyst (10.0–15.0).

Considering those known *Caryospora* species that have spherical-subspherical oocysts with no micropyle, a thick colourless wall of 2 layers and with striations, and a single polar body, we need to compare *C. constanciae* and *C. epicratesi* only with *C. bigenetica*. Oocysts of the latter are much smaller (13.2 vs 20.5 x 19.2 and 22.9 x 21.4).

Among the *Caryospora* species with no micropyle, predominantly spherical oocysts, a polar body and a single-layered, colourless oocyst wall with no striations, *C. paraensis* and *C. micruri* need to be differentiated from *C. ernsti* and *C. jararacae*. The oocysts of the latter two parasites measure only 12.5 and 13.0 respectively, compared with 17.3 x 17.0 and 16.1 x 16.0 for those of the first two. *C. demansiae* has an oocyst similar in many respects to those of *C. paraensis* and *C. micruri*: it is larger, however (20.4 x 19.8), and the wall has 2 layers.

Caryospora najadae and *C. telescopis* both have spherical oocysts, measuring 31.9 and 21.5, respectively. They are differentiated from *C. paraensis* and *C. micruri* by their larger size and lack of a polar body.

Finally, *C. zuckermanae* was described only from non-sporulated oocysts and it remains, therefore, a dubious species. The immature, spherical oocyst was said to have an outer layer resembling "crumpled cellophane". In this respect it might be compared with *C. constanciae*, but the latter has predominantly subspherical oocysts measuring 20.5 x 19.2, compared with the spherical one of *C. zuckermanae*, which is only 15.0–18.0 in diameter.

Interest in the genus *Caryospora* has increased in recent years, following indication of a heteroxenous life-cycle for *C. bubonis*, a parasite of owls (Aves: Strigidae), by Cawthorn & Stockdale (1982). Mice fed 4 weeks previously with the sporulated oocysts were able to serve as a source of infection when fed to clean owls.

Wacha & Christiansen (1982) extended these observations to the *Caryospora* species of snakes when they fed oocysts of *C. bigenetica* to mice and found a variety of developmental stages of the parasite in the connective tissue of the tongue. These included mature micro- and macrogametocytes, fully sporulated sporocysts, "excysted" sporozoites, and "resting" ones contained in cyst-like host cells referred to as "caryocysts". Infected mice were fed to a clean snake, which subsequently passed typical oocysts of *C. bigenetica* in its faeces.

Upton et al. (1984c) found similar developmental stages of *C. simplex* in mice fed with mature oocysts, and Upton & Barnard (1986) showed that not only could infection be induced in clean snakes fed with these mice, but the parasite could be transmitted from snake to snake by the simple ingestion of faecal oocysts, suggesting the heteroxenous life-cycle to be of a faculative nature. It was also shown (Upton et al., 1985) that clean mice could be infected by feeding them with tissues from infected ones.

Finally, the full range of developing stages of a *Caryospora* species has been found in the skin and lymph-nodes of a 2 month-old dog with severe pyogranulomatous dermatitis and which was probably in an immunosuppressed state induced by concurrent distemper virus infection (Dubey et al., 1990).

As a result of these observations, some authors have implied that all species of *Caryospora* have heteroxenous life-cycles, to the extent that the genus was redefined by Wacha & Christiansen (1982) as being "heteroxenous, having two hosts (primary and secondary) with gametogony occurring in each,..... primary hosts mainly birds and reptiles. Secondary hosts mammals, where known". As cautioned by Upton et al., (1984 a), however, "The possibility also exists that some species.... may not be heteroxenous and that some develop directly in a manner similar to most of the Eimeriidae". This certainly might be the case for such species as *C. ernsti* and *C. gekkonis* of lizards, and *C. cheloniae* of a marine turtle, although an alternative is that their secondary hosts are invertebrates (insects?) or non-mammalian vertebrates (fish?)

Apart from the findings of Leibovitz et al. (1978), who recorded pathological effects of *C. cheloniae* in mariculture-reared turtles (*Chelonia mydas mydas*), there is no evidence to suggest that natural infection with *Caryospora* in the primary host is harmful. That in *Ch. m. mydas* might well have become pathological due to the stress of captivity. The suggestion that "Some species of *Caryospora* are pathogenic in their secondary hosts" (Dubey et al., 1990) should also be viewed with caution. It is unwise to assume that the infection in laboratory mice, fed with unusually large numbers of oocysts, is likely to be the same as that in the natural secondary host(s), which is (are) unlikely to be exposed to such massive doses of oocysts. Finally, the severe *Caryospora*-associated dermatitis recorded in a dog was possibly the result of concurrent distemper virus infection (Dubey et al., 1990), which is known to induce an immunosuppressed condition.

As is the case with so many coccidial parasites, we remain sadly ignorant of the full life-cycles of most of the parasites listed in Tables I and II and, in particular, of the degree of specificity they may show in their hosts. Cross-infection experiments with *Caryospora* species from widely separated primary hosts would provide a great deal of important information.

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REFERENCES

ANDERSON, D.R.; DUSZYNSKI, D.W. & MARQUARDT, W.C., 1968. Three new coccidia (Protozoa: Telosporea) from kingsnakes, *Lampropeltis* spp., in Illinois, with a redescription of *Eimeria zamenis* Phisalix, 1921. *J. Parasitol.*, *54*: 577-581.

BRAY, R.S., 1960. On the parasitic Protozoa of Liberia III. Three new species of *Caryospora* Léger, 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.

CANNON, L.R.G., 1967. *Caryospora demansiae* sp.nov. (Sporozoa: Eimeriidae) from the Australian snake *Demansia psammophis* (Elapidae). *Parasitology*, *57*: 221-226.

CANNON, L.R.G. & RZEPCZYK, C.M., 1974. A new species of *Caryospora* Léger, 1904 (Sporozoa: Eimeriidae) from an Australian snake. *Parasitology*, *69*: 197-200.

CARINI, A., 1932. *Caryospora brasiliensis* n.sp. parasita do intestino da cobra verde, *Chlorosoma aestivum*. *Rev. Biol. e Hyg., São Paulo, 3*: 85-87.

CARINI, A., 1939. Sobre uma cariospora da Jararaca. *Arq. Biol., São Paulo, 23*: 41-42.

CHAKRAVARTY, M. & KAR, A.B., 1947. Observations on two reptilian coccida. *J. Roy. Asiat. Soc., Bengal Sci.*, *12*: 3-5.

DUBEY, J.P.; BLACK, S.S.; SANGSTER, L.T.; LINDSAY, D.S.; SUNDERMANN, C.A. & TOPPER, M.J., 1990. *Caryospora*-associated dermatitis in dogs. *J. Parasitol.*, *76*: 552-556.

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

LAINSON, R. & SHAW, J.J., 1973. Coccidia of Brazilian snakes: *Isospora decipiens*, *Eimeria micruri*, *E. liophi* and *E. leimadophi* spp. n., with redescriptions of *Caryospora brasiliensis* Carini, 1932 and *Eimeria poecilogyri* Carini, 1933. *J. Protozool.*, *20*: 358-362.

LÉGER, L., 1904. Protozoaires parasites des vipères. *Bull. Mens. Assoc. Fr. Av. Sci.*, *9*: 208.

LÉGER, L., 1911. *Caryospora simplex*, coccidie monosporée et la classification des coccidies. *Arch. Protistenkd.*, *22*: 71-86.

LEIBOVITZ, L.; REBELL, G. & BOUCHER, G.C., 1978. *Caryospora cheloniae* sp.n., a coccidian pathogen of mariculture-reared green sea turtles (*Chelonia mydas mydas*). *J. Wildl. Dis.*, *14*: 269-275.

LEVINE, N.D., 1988. *The Protozoan Phylum Apicomplexa* Vol. I. CRC Press, Inc., Boca Raton, Florida, U.S.A., 178-179.

MANDAL, R., 1976. *Caryospora bengalensis* sp.n. and *Eimeria fibrilosa* sp.n. new coccidia (Protozoa: Eucoccidia) from a fresh-water snake, *Enhydris enhydris* (Schneider). *Acta Protozool.*, *15*: 405-410.

MATSUBAYASHI, H., 1936. On a new coccidium, *Koidzumiella natrix* n.g., n.sp., parasitic in Japanese snakes; with some remarks on two other ophidian coccidia of the genus *Caryospora* and *Isospora*. *Keio-Igaku*, *16*: 1719-1729.

MATUSCHKA, F-R., 1982. *Caryospora najae* sp.n. (Apicomplexa: Sporozoea: Eimeriidae) from the spitting cobra, *Naja nigricollis pallida* (Serpentes: Elapidae). *J. Parasitol.*, *68*: 1149-1153.

MATUSCHKA, F-R., 1984a. Description of *Caryospora colubris* sp.n. (Apicomplexa: Eimeriidae) from the western whip snake, *Coluber viridiflavus* (Serpentes: Colubridae). *Can. J. Zool.*, *62*: 1525-1527.

MATUSCHKA, F-R., 1984b. *Caryospora corallae* sp.n. (Apicomplexa: Eimeriidae) from the emerald tree boa *Corallus caninus* (Serpentes: Boidae). *J. Protozool.*, *31*: 358-359.

MATUSCHKA, F-R., 1986a. *Caryospora najadae* sp.n. (Apicomplexa: Eimeriidae) from Dahl's whip snake, *Coluber najadum* (Serpentes: Colubridae). *Z. Parasitenkd.*, *72*: 181-183.

MATUSCHKA, F-R., 1986b. *Caryospora telescopis* sp.n. (Apicomplexa: Eimeriidae) from the cat snake, *Telescopus fallax* (Serpentes: Colubridae), and *Caryospora* spec. from the West Caucasian viper, *Vipera kaznakovi* (Serpentes: Viperidae). *Arch. Protistenkd.*, *132*: 93-99.

NANDI, R., 1985. *Caryospora cobrae* n.sp., from the Indian cobra *Naja naja* Linnaeus. *J. Protozool.*, *32*: 204-205.

UPTON, S.J. & BARNARD, S.M., 1986. Experimental transmission of *Caryospora simplex* (Apicomplexa: Eimeriidae) to Palestine vipers, *Vipera xanthina palestinae* (Serpentes: Viperidae). *J. Protozool.*, *33*: 129-130.

UPTON, S.J.; CURRENT, W.L. & BARNARD, S.M., 1984a. A new species of *Caryospora* (Apicomplexa: Eimeriidae) from *Elaphe* spp (Serpentes: Colubridae) of the southeastern and central United States. *Trans Am. Microsc. Soc.*, *103*: 240-244.

UPTON, S.J.; CURRENT, W.L. & BARNARD, S.M., 1984b. A new species of *Caryospora* (Apicomplexa: Eimeriorina) from the green lizard, *Anolis carolinensis*. *Trans. Am. Microsc. Soc.*, *103*: 245-248.

UPTON, S.J.; CURRENT, W.L.; ERNST, J.V. & BARNARD, S.M., 1984c. Extraintestinal development of *Caryospora simplex* (Apicomplexa: Eimeriidae) in experimentally infected mice, *Mus musculus*. *J. Protozool.*, *31*: 392-398.

UPTON, S.J.; ERNST, J.V.; BARNARD, S.M. & CURRENT, W.L., 1983. Redescription of the oocysts of *Caryospora simplex* (Apicomplexa: Eimeriidae) from an Ottoman viper, *Vipera xanthina xanthina*. *Trans. Am. Microsc. Soc.*, *102*: 258-263.

UPTON, S.J.; LINDSAY, D.S.; CURRENT, W.L. & BARNARD, S.M., 1985. Mouse-to-mouse transmission of *Caryospora simplex* (Apicomplexa: Eimeriidae). *J. Parasitol.*, *71*: 395-396.

WACHA, R.S. & CHRISTIANSEN, J.L., 1982. Development of *Caryospora bigenetica* n.sp. (Apicomplexa: Eimeriidae) in rattlesnakes and laboratory mice. *J. Protozool.*, *29*: 272-278.