

# Kleinere Mitteilungen.

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(From the Department of Zoology, Calcutta University Calcutta, India.)

*Isospora knowlesi* n. sp. (Coccidia)  
from the intestine of a lizard,  
*Hemidactylus flaviviridis* (RÜPPELL).

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With plate 7.

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## Introduction.

In the course of examining the gut contents of the common wall lizard, *Hemidactylus flaviviridis* (RÜPPELL) we came across a species of coccidia which belongs to the genus *Isospora*. Reference to this sporozoan from India has been made by KNOWLES and DAS-GUPTA (1935) but unfortunately, due to want of material, they could not give its detailed description, except that they noticed the smallness of the size of oocysts. As we have found all the necessary endogenous stages of development in the material at our disposal, we consider ourselves to be in a better position to give it a specific name and prefer to call it *I. knowlesi* n. sp. The species is named after one of the authors (Lieut-Colonel R. KNOWLES, I. M. S.) who first saw it in this lizard from this part of the world.

### Material and Methods.

Examination of faecal matter of lizards made in saline revealed the presence of oocysts of *Isospora* in three out of forty cases. One of these three also contained *Eimeria falviviridis* SETNA and BANA<sup>1)</sup> in its gall bladder and two (?) species of *Eimeria*<sup>2)</sup> in its intestinal epithelium. Faecal matter was at once transferred to 1% chromic acid solution in distilled water in order to study the further development of oocysts. Portions of the infected intestine were also fixed in BOUIN-DUBOSCQ-BRASIL'S fluid and subsequently cut 6  $\mu$  thick and stained in HEIDENHAIN'S iron-alum haematoxylin. These sections were of great value as they showed the endogenous stages of development. The infection was not very rich in any one of these cases and hence it was after a laborious search that we could get all the necessary details. In the case of mixed infection it was however not difficult to distinguish our *Isospora* from *Eimeria* because the former was always found to be intranuclear in habit at all stages.

### Observation on *Isospora knowlesi* n. sp.

As we have not seen a sporozoite entering intestinal epithelium we shall begin our description with the smallest intranuclear stage that we have seen. It measures about 4—5  $\mu$  in diameter and can be clearly seen lying within a vacuole in the nucleus of the epithelial cell (see Pl. 7 Fig. 1). The infected nuclei are almost always found to be dragged away from the general level of the epithelial nuclei and come to lie more towards the lumen of the intestine. With the growth of the parasite the nucleus becomes stretched out while its chromatin is reduced to a ring which borders the growing parasite. We have seen the final products of schizogony in our preparation but, unfortunately earlier stages were not seen by us. Fig. 4 on Pl. 7 is drawn from a transverse section of a group of merozoites lying within the stretched out epithelial nucleus. In Fig. 5 on Pl. 7 we see some of the merozoites in longitudinal section which measure  $8.25 \times 2 \mu$  in dimensions, (as schizogony was not found in smears made from the intestine we had to rely on the measurements taken

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<sup>1)</sup> This is referred to as *Eimeria*, species B by KNOWLES and DAS-GUPTA (1935).

<sup>2)</sup> Referred to as *Eimeria*, species A and C by KNOWLES and DAS-GUPTA (1935).

from the sections). A fully formed merozoite shows one of its end more pointed than the other and possesses a small karyosome in the nucleus. It must be said here that these groups of merozoites in our preparations were to be seen very few and far between. It may be that the process of schizogony takes place very rapidly or, it is quite likely, that schizogony cycle was over when we examined the material and the few groups of merozoites that we have found resulted from a few of the „Slackers“. Keeping this in view therefore, the stage that are pictured here are nothing but different phases in the sporogony cycle of the parasite. This latter argument finds support with us firstly, because of the want of both early stages of schizogony and abundance of group of merozoites, and secondly, the prevalence of the stages in sporogony.

The male gametocyte ultimately gives rise to biflagellated gametes measuring 4—5  $\mu$  in length and 1—2  $\mu$  in breadth (see Pl. 7 Fig. 7). When fully mature, the nuclear membrane gives way, and these gametes either drop into the lumen of the intestine or penetrate the neighbouring epithelial cells (see Pl. 7 Fig. 6). The female gametocyte with the advancement of age shows a deposition of granules in its peripheral cytoplasm while the karyosome persists for sometime (see Pl. 7 Fig. 3). Later on however the karyosome disappears and a female gamete shows a number of granules in the nucleus. Probably it is a stage prior to fertilization. The next stage that is very common in our preparation is the oocyst in which the karyosome has reappeared (see Pl. 7 Fig. 8). In the oocyst there is a thick inner membrane and a thin outer membrane, and besides these two membranes, while the oocyst is lodged in the cell, there is the widely stretched out membrane of the epithelial nucleus which, however, finally gives way and liberates the oocyst. The remnants of the infected nuclei are not seen round the oocysts picked up from the faecal matter but are left within the cell and degenerate. The infected nuclei in such stages are almost touching the free end of the epithelial cell while the cellular cytoplasm becomes intensely vacuolar in nature (see Pl. 7 Fig. 8). The segmented oocyst seen in the faecal matter measures 18—23  $\mu$  in diameter and contains no oocystic residue. Further development of the oocysts takes place outside the body of the host. Sporocyst measure 12—15  $\mu \times 8—10 \mu$ , and has at one of its poles a 'knob-like' swelling (see Pl. 7 Fig. 10). Usually the two sporocysts lie at right angles to each other within the oocyst and contain sporocystic residue. Oocysts when kept in 1% chromic acid complete their development within four to five days.

List of species of *Isospora* recorded from Lacertilia.

Name of Parasite	Host.	Seat of infection	Dimensions in mucron		Locality
			Oocyst	Sporocyst	
<i>Isospora camillerii</i> HAGENMÜLLER, 1898 (vide HOARE, 1933)	<i>Gongylus ocellatus</i>	intestine	22.0 × 22.0	18.0 × 12.0	—
<i>Isospora mesnili</i> SERGENT, ED., 1902 (vide WENYON, 1926)	<i>Chaemeleo vulgaire</i>	nuclei of intestinal epithelium	30.0 × 30.0	16.0 × 10.0	—
<i>Isospora</i> sp. WENYON, 1926	<i>Agama colonorum</i>	liver and intestine	?	?	Sudan
<i>Isospora</i> sp. ADLER (vide WENYON, 1926)	<i>Varanus griseus</i>	sub-epithelial tissue of small intestine	?	?	Palestine
<i>Isospora ameivae</i> CARINI, 1931 (vide BECKER, 1934)	<i>Ameiva ameiva</i>	—	19.0 × 16.0	?	Brazil
<i>Isospora</i> sp. SETNA, 1933	<i>Calotes versicolor</i>	intestine	?	?	Bombay (India)
<i>Isospora</i> sp. KNOWLES and DAS-GUPTA, 1935	<i>Hemidactylus flaviviridis</i>	intestine	21.4 × 20.0	?	Calcutta (India)
<i>Isospora knowlesi</i> n. sp.	<i>Hemidactylus flaviviridis</i>	nuclei of intestinal epithelium	18.0—23.0 × 18.0—23.0	12.0—15.0 × 8.0—10.0	Calcutta (India)

### Diagnosis.

The tetrazoic disporocystid condition of the oocyst at once places this coccidian in the genus *Isospora*. From the literature extant on the subject it is quite evident that *Isospora* described in this paper can not be identified with any of the Lacertilian coccidia listed in the accompanying table though it approaches *I. camillerii* HAGENMÜLLER in dimensions and *I. mesnili* SERGENT, ED., in being intranuclear in habit.

Systematic position. *Isospora knowlesi* n. sp. (Coccidiida Eimeriidae).

Description. Oocyst thick walled, spherical in shape: 18—23  $\mu$  in diameter; sporocysts, ellipsoidal with a “knob-like” structure at one pole: 12—15  $\mu$   $\times$  8—10  $\mu$ ; sporozoites arranged regularly; large sporocystic residue; unsegmented or segmented (but immature) oocysts discharged from host; sporulation time 4—5 days.

Habitat. Small intestine (intranuclear) of *Hemidactylus flaviviridis* (Lacertilia).

Locality. Calcutta, Bengal, India.

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\* Not seen in the original.

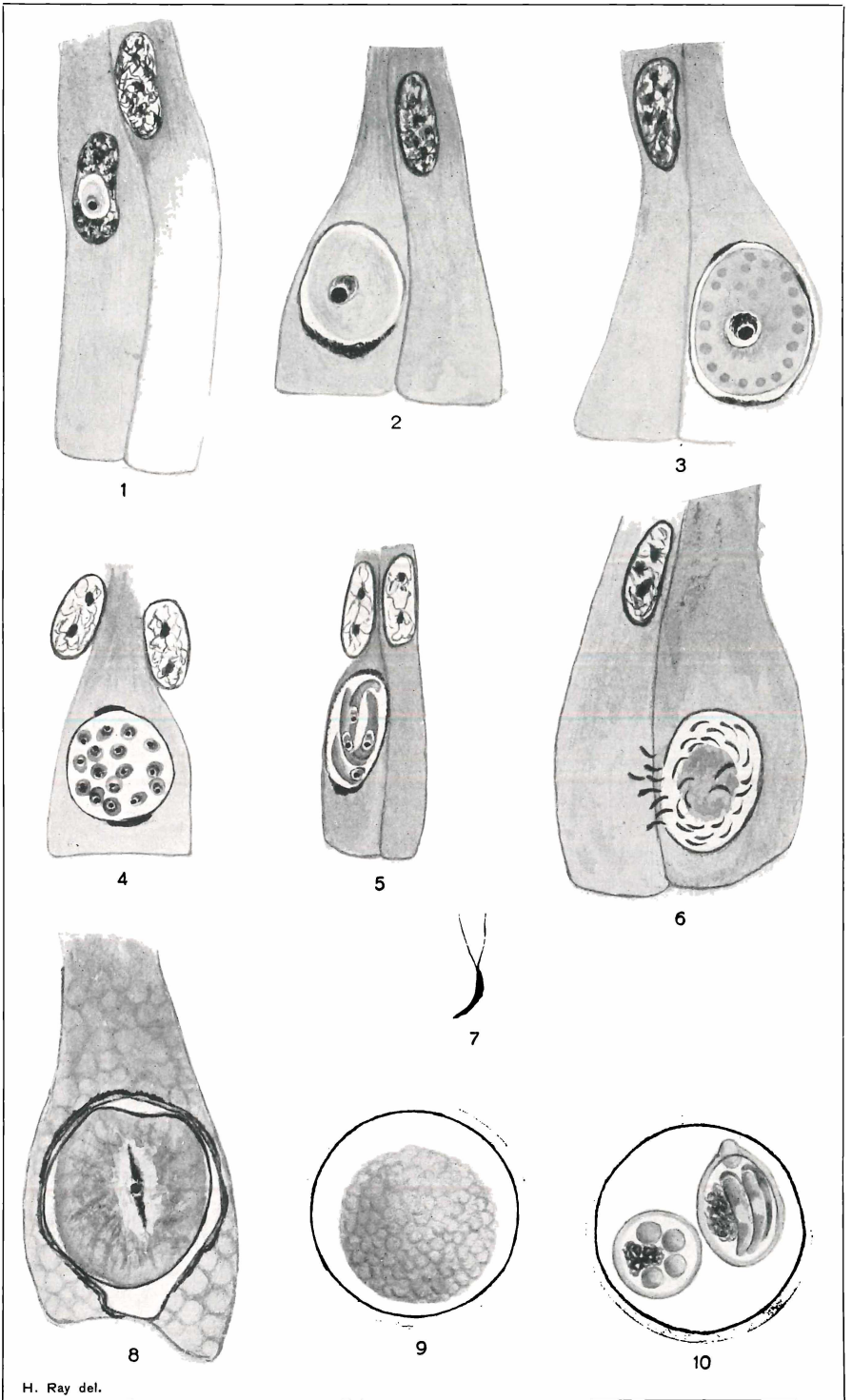
## Explanation of plate.

### Plate 7.

All figures are camera lucida drawings either of living specimens or from sections fixed in BOUIN-DUBOSCQ-BRASIL'S fluid and stained with HEIDENHAIN'S iron haematoxylin.  $\times 1666$ , except Fig. 7 which is magnified 3500 times.

#### *Isospora knowlesi* n. sp.

- Fig. 1. Young intranuclear gametocyte.
  - Fig. 2. Growing female gametocyte.
  - Fig. 3. Full grown female gamete.
  - Fig. 4. Group of merozoites in transverse section.
  - Fig. 5. Same in longitudinal section.
  - Fig. 6. Group of male gametes. Host nucleus ruptured and number of gametes entering neighbouring cell.
  - Fig. 7. Biflagellated male gamete.
  - Fig. 8. Oocyst — still within the host nucleus — note the vacuolar nature of the cell and extensively stretched out nuclear membrane.
  - Fig. 9. Oocyst from the faecal matter.
  - Fig. 10. Mature oocyst after being in 1% chromic acid solution for five days.
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