

## TWO NEW SPECIES OF *ISOSPORA* (APICOMPLEXA: EIMERIIDAE) FROM SKINKS, *EMOIA* SPP. (SAURIA: SCINCIDAE), FROM FIJI AND PAPUA NEW GUINEA

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**ABSTRACT:** Between September and October 1991 and again during September 1992, skinks (*Emoia* spp.) were collected from various localities on Fiji and Papua New Guinea (PNG) and examined for coccidians. One of 4 (25%) De Vis' emo skinks (*Emoia pallidiceps*) from PNG harbored an undescribed species of *Isospora* in its feces. Oocysts of *Isospora grimbikpelapalai* n. sp. were ellipsoidal to subspheroidal,  $18.1 \times 14.9$  ( $17\text{--}20 \times 14\text{--}16$ )  $\mu\text{m}$ , with a bilayered wall and a length/width index (L/W) of 1.2. Both micropyle and oocyst residuum were absent, but a prominent polar granule was present. Sporocysts were ovoidal,  $10.7 \times 7.6$  ( $10\text{--}11 \times 7\text{--}8$ )  $\mu\text{m}$ , with a L/W index of 1.4. Stieda and sub-Stieda bodies were present, but para-Stieda bodies were absent. The sporocyst residuum consisted of large scattered globules dispersed between sporozoites. Sporozoites were elongate with spheroidal anterior and posterior refractile bodies. *Isospora grimbikpelapalai* was also found in 1 of 2 (50%) Pope's emo skinks (*Emoia popei*) from PNG. One of 13 (8%) white-bellied copper-striped skinks (*Emoia cyanura*), from Fiji, was passing another undescribed species of *Isospora* in its feces. Oocysts of *Isospora casei* n. sp. were elongate,  $31.8 \times 21.3$  ( $28\text{--}35 \times 18\text{--}24$ )  $\mu\text{m}$ , with a bilayered wall and a L/W index of 1.5. Micropyle, oocyst residuum, and polar granule were all absent. Sporocysts were ovoidal,  $15.3 \times 10.6$  ( $14\text{--}16 \times 10\text{--}12$ )  $\mu\text{m}$ , with a L/W index of 1.4. Stieda and sub-Stieda bodies were present, but para-Stieda bodies were absent. The sporocyst residuum consisted of scattered globules among sporozoites or as a cluster surrounding sporozoites. Sporozoites were elongate with spheroidal anterior and posterior refractile bodies. *Isospora casei* was also found in 1 of 2 (50%) Fiji slender treeskinks (*Emoia concolor*) from Fiji. This represents the first report of coccidia from *Emoia* spp. and, to our knowledge, the initial documentation of reptilian coccidia from herpetofauna from Papua New Guinea.

The Oceanian biogeographical realm and its East Melanesian and Papuan provinces, as defined by Udvardy (1975), are known to contain several globally important parasite biodiversity hotspots, including those on many of the islands of the South Pacific. Indeed, several recent papers have been published on new species of nematodes and malarial parasites of scincid lizards from the Independent State of Papua New Guinea (PNG) (see Bursey et al., 2005, 2006, 2011; Perkins and Austin, 2009). However, little is known about coccidian parasites of skinks in this region. Modrý and Jirků (2006) described 3 new species of coccidia from marble-throated skinks (*Marmorosphax tricolor*) from New Caledonia (Nouvelle Calédonie). However, to our knowledge, nothing has been published on coccidia of any skinks from PNG or the Republic of Fiji. Herein, we provide descriptions of 2 new isosporans from *Emoia* spp. from PNG and the Fiji Islands.

### MATERIALS AND METHODS

Between September and October 1991 and again during September 1992, 23 skinks (*Emoia* spp.) were collected by hand from various localities on Fiji and PNG and examined for coccidians. They included: 4 De Vis' emo skinks (*Emoia pallidiceps*) and 2 Pope's emo skinks (*Emoia popei*) from PNG, and 13 white-bellied copper-striped skinks (*Emoia cyanura*), 2 dark-bellied copper-striped skinks (*Emoia impar*), and 2 Fiji slender treeskinks (*Emoia concolor*) from Fiji. Fresh fecal samples were placed in individual vials containing 2.5% (w/v) aqueous potassium dichromate ( $\text{K}_2\text{Cr}_2\text{O}_7$ ) and examined for coccidia by light microscopy after flotation in Sheather's sugar solution (specific gravity = 1.30). Positive samples containing unsporulated oocysts were allowed up to 1 wk to sporulate. Measurements were taken on 25 oocysts using a calibrated ocular micrometer and reported in micrometers ( $\mu\text{m}$ ) as means followed by the ranges in parentheses; photographs were taken using Nomarski interfer-

ence-contrast optics. Oocysts were ~300 days old when measured and photographed. Descriptions of oocysts and sporocysts follow guidelines of Wilber et al. (1998) as follows: oocyst length (L) and width (W), their ranges and ratios (L/W), micropyle (M), oocyst residuum (OR), polar granules (PG), sporocyst length (L) and width (W), their ranges and ratios (L/W), sporocysts (SP), Stieda body (SB), sub-Stieda body (SSB), para-Stieda body (PSB), sporocyst residuum (SR), sporozoites (SZ) and sporozoites' anterior (ARB) and posterior (PRB) refractile bodies, and nucleus (N). Host vouchers were accessioned into the California Academy of Sciences (CAS), San Francisco, California. Photovouchers of sporulated oocysts were accessioned into the U.S. National Parasite Collection (USNPC), Beltsville, Maryland. Lizard taxonomy follows the reptile database (Uetz, 2011) and field guide to Pacific reptiles (Zug, 2013).

Only 4 of 25 (16%) *Emoia* skinks were infected, including 1 of 4 (25%) *E. pallidiceps* and 1 of 2 (50%) *E. popei* from PNG, which harbored an undescribed species of *Isospora* in its feces; 1 of 13 (8%) *E. cyanura*, 0 of 2 (0%) *E. impar*, and 1 of 2 (50%) *E. concolor* from Fiji were found to be infected with a different undescribed species of *Isospora*. The descriptions follow.

### DESCRIPTION

#### *Isospora grimbikpelapalai* n. sp.

(Figs. 1, 3)

**Description of sporulated oocyst:** Oocyst shape ellipsoidal to subspheroidal; bilayered wall, ~1.2 thick, smooth outer layer ~0.8, inner layer ~0.4; L  $\times$  W  $18.1 \times 14.9$  ( $17\text{--}20 \times 14\text{--}16$ ); L/W 1.2 (1.1–1.4). M, OR absent; prominent, irregularly shaped PG present, 2–3.

**Description of sporocyst and sporozoites:** SP ovoidal, ~0.4 thick, with a smooth single-layered wall; L  $\times$  W  $10.7 \times 7.6$  ( $10\text{--}11 \times 7\text{--}8$ ); L/W 1.4 (1.3–1.6); SB, SSB present, PSB absent; SR composed of large scattered globules dispersed between SZ. SZ elongate with spheroidal ARB and PRB; single N slightly posterior to midpoint of body.

### Taxonomic summary

**Type host:** *Emoia pallidiceps* de Vis, 1890, De Vis' emo skink (Sauria: Scincidae). Collected 18 September 1991. Symbiotype CAS 192830.

**Other hosts:** *Emoia popei* Brown, 1953, Pope's emo skink (Sauria: Scincidae). Collected 16 September 1991. CAS 192891.

**Type specimens:** Photosyntype deposited as USNPC 106379.

**Type locality:** Amron Village (W of Baitabag Mission), Madang Province, Papua New Guinea ( $05^{\circ}06'37.8259''\text{S}$ ,  $145^{\circ}45'43.7836''\text{E}$ ).

**Prevalence:** One of 4 (25%), *E. pallidiceps*; 1 of 2 (50%) *E. popei*.

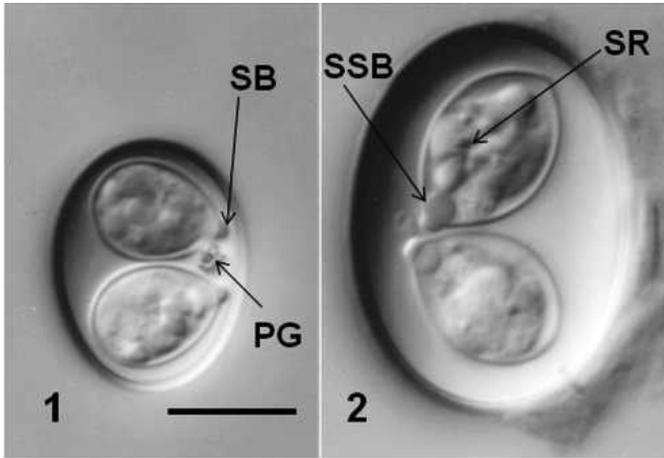
**Sporulation:** Exogenous. All oocysts were passed in the feces unsporulated and fully sporulated within 1 wk at ~23 C.

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FIGURES 1–2. Nomarski interference-contrast photomicrographs of oocysts of *Isospora grimbikpelapalai* n. sp. and *Isospora casei* n. sp. (1) Oocyst of *I. grimbikpelapalai* showing Stieda body (SB) and polar granule (PG). (2) Oocyst of *I. casei* showing sporocyst residuum (SR) and sub-Stieda body (SSB). Scale bars = 10  $\mu$ m.

*Prepatent and patent periods:* Unknown.

*Site of infection:* Unknown; oocysts recovered from feces.

*Endogenous stages:* Unknown.

*Etymology:* The specific epithet is given for “Green Dragon” in PNG Pidgin English, green is “grin,” big is “bikpela,” and dragon is “palai,” as that represents all lizards in PNG; there is no word for dragon independently in Pidgin English. The name is in honor of a B-25 Mitchell bomber (Green Dragon) crew from WWII that was shot down off of Madang, and 4 of the crew were taken prisoner of war to Amron (the type locality), where they were executed by the Japanese on 31 August 1943.

## Remarks

*Isospora grimbikpelapalai* can be easily differentiated from almost all isosporans reported from scincid lizards (Modrý and Jirků, 2006; Paperna, 2006). Only 1 species, *Isospora carliae* Paperna, 2006, from the blue-throated rainbow skink (*Carlia rhomboidalis*) is similar in size to *I. grimbikpelapalai*. Oocysts of *I. carliae* are  $18.0 \times 14.1$  (L/W = 1.3), SP are  $11.5 \times 7.8$  (L/W = 1.5), and the SP contain both SB and SSB as does *I. grimbikpelapalai*. However, *I. grimbikpelapalai* SZ possess both an ARB and PRB and a prominent PG, whereas *I. carliae* has a single RB and no PG.

### *Isospora casei* n. sp.

(Figs. 2, 4)

*Description of sporulated oocyst:* Oocyst shape elongate; bilayered wall,  $\sim 1.6$  thick, smooth outer layer  $\sim 1.0$ , inner layer  $\sim 0.6$ ; L  $\times$  W  $31.8 \times 21.3$  ( $28\text{--}35 \times 18\text{--}24$ ); L/W 1.5 (1.3–1.9). M, OR, PG absent.

*Description of sporocyst and sporozoites:* SP ovoidal; smooth single-layered wall  $\sim 0.6$  thick, L  $\times$  W  $15.3 \times 10.6$  ( $14\text{--}16 \times 10\text{--}12$ ); L/W 1.4 (1.3–1.5); SB, SSB present, PSB absent; SR scattered among SZ or as cluster surrounding SZ. SZ elongate with spherical ARB and PRB; single N slightly posterior to midpoint of body.

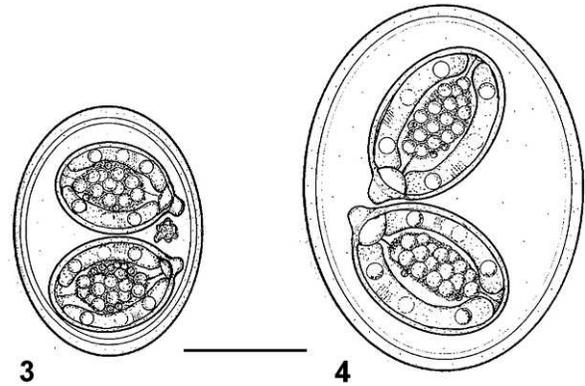
## Taxonomic summary

*Type host:* *Emoia cyanura* (Lesson, 1826), white-bellied copper-striped skink (Sauria: Scincidae). Collected 28 September 1992. Symbiotype CAS 252833.

*Other hosts:* *Emoia concolor* (Duméril, 1851), Fiji slender treeskink (Sauria: Scincidae). Collected 26 September 1992. CAS 252844.

*Type specimens:* Photosyntype deposited as USNPC 106380.

*Type locality:* Ovalau Holiday Resort, Ovalau Island, Lomaviti Province, Fiji ( $17^{\circ}39'01.67''$ S,  $178^{\circ}49'35.4''$ E).



FIGURES 3–4. Composite line drawings of oocysts of *Isospora grimbikpelapalai* n. sp. and *Isospora casei* n. sp. (3) *I. grimbikpelapalai* n. sp. (4) *I. casei* n. sp. Bar = 10  $\mu$ m.

*Prevalence:* One of 13 (8%) *E. cyanura*; 0/2 (0%) *E. impar*; 1/2 (50%) *E. concolor*.

*Sporulation:* Exogenous. All oocysts were passed in the feces unsporulated and fully sporulated within 1 wk at  $\sim 23$  C.

*Prepatent and patent periods:* Unknown.

*Site of infection:* Unknown; oocysts recovered from feces.

*Endogenous stages:* Unknown.

*Etymology:* The specific epithet is given in honor of Dr. Ted J. Case, University of California–San Diego, in recognition of his many contributions to the study of reptilian biology.

## Remarks

Oocysts of *Isospora casei* can be easily differentiated from all isosporans reported from scincid lizards (Modrý and Jirků, 2006; Paperna, 2006) based on the size of its oocysts and SP, with 1 exception: *Isospora arabica* Amoudi, 1989, from the ocellated bronze skink, *Chalcides ocellatus* from Saudi Arabia, possesses subspherical oocysts that are  $32.5 \times 25.0$  (L/W = 1.3) and ovoid SP that are  $19.0 \times 13.5$  (L/W = 1.4) (Amoudi, 1989). Oocysts of *I. casei* differ from *I. arabica* as follows: The oocyst L/W is larger (1.5), and SP are significantly smaller ( $15.3 \times 10.6$ ; L/W = 1.4). In addition, *I. arabica* does not have a SSB, while *I. casei* possesses a very prominent one (see Figs. 2, 4).

## DISCUSSION

The isolation of many South Pacific Islands and the extreme distances between them have most likely prevented scientists in the past from studying their coccidian parasites in a comparative context. However, Hanley et al. (1995) provided information on previously described eimerians from geckos (*Hemidactylus frenatus* and *Lepidodactylus lugubris*) from several island groups (see their Table 1) from across the Pacific, including Fiji. The first isosporans to be reported from Australian scincid lizards were described by Cannon (1967) from the snake-eyed skink, *Cryptoblepharus* (= *Ablepharus*) *boutonii*, and White’s rock skink, *Liopholis* (= *Egernia*) *whitii*. From that time to more recently, Modrý and Jirků (2006) provided a summary of the coccidia (*Isospora* spp.) of the family Scincidae that included a description of a new species of *Isospora* from *Marmorosphax tricolor* from New Caledonia. In addition, Paperna (2006) reported a new isosporan from the blue-throated rainbow skink (*Carlia rhomboidalis*) from Australia. Altogether, there are 14 species of *Isospora* from 11 species of skinks of the genera *Carlia*, *Chalcides*, *Cryptoblepharus*, *Eumeces* (= *Plestiodon*), *Liopholis*, *Marmoros-*

*phax*, *Scincella*, and *Trachylepis* (= *Mabuya*). Only 5 (36%) of these, namely *Isospora ablephari* Cannon, 1967, *Isospora bocagei* Modrý and Jirků, 2006, *Isospora carliae* Paperna, 2006, *Isospora cryptoblephari* Finkleman and Paperna, 1994, and *Isospora egeriae* Cannon, 1967, were reported from hosts collected from Australia and/or New Caledonia (Modrý and Jirků, 2006; Paperna, 2006). These South Pacific locales are found within the Australian biogeographical realm and are far removed, by many kilometers, from our study sites. Therefore, we provide descriptions of the first isosporans ever reported from skinks of the genus *Emoia* and from Fiji and PNG.

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