

## MALARIA AND HEMOGREGARINES FROM LIZARDS OF THE WESTERN CARIBBEAN ISLANDS OF SAN ANDRÉS AND PROVIDENCIA

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### SUMMARY

*Plasmodium floridense* occurs in Iguanid lizards on San Andrés Island, throughout Middle America and the southeastern United States. It may have evolved as an offshoot of *P. tropiduri*, either in Middle America or on the larger Caribbean Islands. A single *Hepatozoon*-like hemogregarine infects lizards of the families Iguanidae, Gekkonidae, Scincidae and Teiidae on Providencia Island.

### INTRODUCTION

The regional distribution of lizard species has been modified considerably during Quaternary times in response to changing vegetation and climatic patterns. Lizard population models, in contrast to bird or mammal models, are characterized by non-migration and long-term association with specific geofloral complexes. Analyzing the distribution of infectious diseases provides insight into the historical zoogeography of their lizard hosts<sup>3</sup> or migration staging areas<sup>5</sup>, and helps clarify the evolution of the parasites themselves<sup>3, 10, 20</sup>. Relict animals and some of their diseases may survive on oceanic islands long after they have disappeared from the adjacent mainland.

This report concerns the finding of hemoparasites in lizards of San Andrés and Providencia-Santa Catalina islands. The islands are located about 230 km (110 miles) east of continental Nicaragua and are 60 to 90 km<sup>2</sup> in total area. Their herpetofauna includes 15 species, historically derived from Central, South America and Caribbean sources<sup>9</sup>. During May 23 through June 1, 1974, at the end of a four-month dry season, representative samples of 10 to 20 lizards were collected from most population groups of anole lizards on each island. Teiid, skink and gecko populations were sampled when time permitted.

### MATERIALS AND METHODS

DUNN & SAXE<sup>9</sup>, TAMSETT & VALDIVIESO<sup>19</sup> and CORN & DALBY<sup>8</sup> summarized the herpetofauna of San Andrés and Providencia. To make the sampling as extensive as possible, up to 4 or five lizards were collected from rural or semirural sites along trails, and subsequent collection sites were separated by several hundred meters. Only the anole populations were thoroughly sampled. The samples included: **(A) PROVIDENCIA ISLAND.** *Anolis pinchoti* (Iguanidae), 114 lizards from four general areas: Old Town 39, Fresh Water Bay 20, El Valle (Airport area) 21, and Santa Catalina Island 34. *Ameiva ameiva* (Teiidae), 14 lizards from three areas: Old Town 4, Fresh Water Bay 7, and El Valle 3. *Cnemidophorus lemniscatus* (Teiidae), 11 lizards from the valley surrounding Old Town. *Mabuya mabuya* (Scincidae), one lizard each from Old Town, Fresh Water Bay and Santa Catalina Island. *Aristelliger georgeensis* (Gekkonidae), one lizard each from Old Town and South Bay. **(B) SAN ANDRÉS ISLAND.** *Anolis concolor* (Iguanidae), 98 lizards from nine sites: Roca del Pescador 7, La Loma — El Cove highway (three highland sites) 13, 13 and 6, west shore highway (two lowland sites), Morgan's Cave 12, one km north of Morgan's Cave 12, east shore highway (three

lowland sites), three km north of San Luis 10, San Luis 10, and about four km south of San Luis 11. *Mabuya mabuya*, one lizard each from El Cove and Morgan's Cave.

Lizards were caught with a dental floss noose and kept in cloth bags until evening. Each was examined, its sex, size and site of capture recorded, and a single thin blood smear obtained from a clipped toe. The blood smears were air-dried, fixed in absolute methyl alcohol for two minutes and stained within two weeks with Giemsa's blood stain solution in distilled water buffered to pH 7.2. They were first studied for up to an hour to identify the positive slides, and these were then examined to describe the structure of the parasites found. Parasites were photographed and drawings made with a camera lucida. At least three or four hemogregarines were drawn and measured from each infected lizard, and a more detailed study of the parasite structure made of the more heavily infected individuals. Only thin blood smears were examined. Ticks removed from the Teiid lizards are deposited in the U. S. Public Health Service, Rocky Mountain Laboratory collection, Hamilton, Montana, U. S. A.

## RESULTS

### *Hemogregarines from Providencia-Santa Catalina*

Hepatozoon-like hemogregarines were found in erythrocytes of 32 lizards (Figs. 1-4), including 19 *A. pinchoti* (Old Town 6, Sta. Catalina Isl. 9, El Valle 4), one *A. georgeensis* (Old Town), one *Mabuya mabuya* (Old Town), five *A. ameiva* (Fresh Water Bay 2, El Valle 3), and six *C. lemniscatus*. The parasite's structure and effect on the host cell was so similar in every case and in each host species that it appeared a single parasite species was involved.

Infections in *A. pinchoti*, *M. mabuya*, and *A. georgeensis* ranged from light (1/1,000 cells infected) to scarcely detectable. In *A. ameiva* they were moderate, ranging from 1/500 to 1/1,000 cells infected, but in *C.*

*lemniscatus*, all infections were intense, with one parasite per each 30 to 400 erythrocytes.

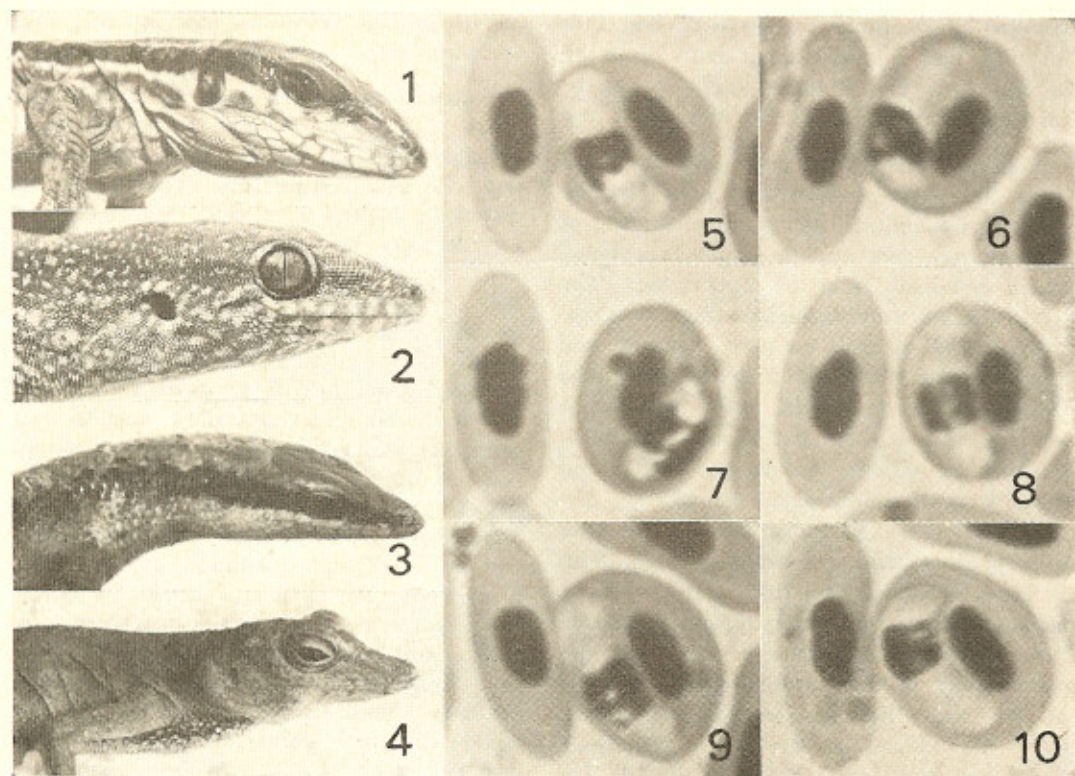
Parasites were seen only in erythrocytes (Figs. 5-10). Immature gametocytes were sausage-shaped or slightly tapered toward one end with a central nucleus. They caused no distortion of the host cell. Mature gametocytes were sausage-shaped, measuring 11 to 13.6 by 3 to 4  $\mu$ . They had a slight curve independent of their position relative to the host cell nucleus (although they usually curved around the nucleus), and they occupied about 1/2 of the available space in the host cell. The parasite's nucleus was off-center or subterminal; it appeared either compact or divided toward each side of the gametocyte. The cytoplasm stained a very light blue, perhaps because the surrounding capsule inhibited penetration of the stain. There were no obvious vacuoles in undistorted parasites. The capsule was frequently visible and often stained with a slight red tint. Infected erythrocytes stained normally but were almost always rounder (shorter and wider) than their uninfected neighbors. The host cell nucleus was undistorted and only slightly displaced by the gametocyte.

Biting culicine mosquitoes were abundant in most areas, especially along the more humid western coast. Ticks of the genus *Amblyomma* were removed from several *C. lemniscatus*.

Hemogregarines are reported from lizards of other Caribbean islands and surrounding countries, but they have been inadequately described, making specific identification of the Providencia parasite fruitless. Lack of information on the tissue or vector stages makes a species designation inadvisable.

### *Malaria in Anolis concolor from San Andrés*

Malaria was found in a single adult male *A. concolor* (Fig. 11), one of 11 captured near the extreme southeastern end of the island. The precise site was about 0.5 km inland from the ocean at the base of the first hill rising much above sea level, near one of the last of the remaining unfilled swampland habitats. Although very few hatchling *A. concolor* were seen anywhere on



Figs. 1-10 — Hemogregarines and hosts from Providencia Island: 1. *Ameiva ameiva* (Teiidae), 2. *Aristelliger georgeensis* (Gekkonidae), 3. *Mabuya mabuya* (Scincidae), 4. *Anolis pinchoti* (Iguanidae), 5-10. Hemogregarines from *Cnemidophorus lemniscatus* (Teiidae), similar in structure to those of the other four lizard host species.

the island, reddish lipoprotein coloration of the blood smears showed that most females were producing eggs<sup>6</sup>.

The infection state remains somewhat perplexing. Parasitemia was high with 9.5% of the erythrocytes infected. Asexual forms composed 72% of the total parasites present: 61% trophozoites, 5% two-nucleus schizonts, 5% advanced schizonts, 1% segmenters. Sexual stages included 6% immature gametocytes, 10% mature microgametocytes and 12% mature macrogametocytes. Despite the rather intense parasitemia, there was no evidence of anemia, and immature erythrocytes were virtually lacking. Almost all of the segmenters looked to be in a condition of arrested division ("crisis" forms of Huff), and about 1/4 of the gametocytes appeared ragged or senile.

The "crisis" segmenters, rounded-up advanced schizonts, senile gametocytes, high

proportion of mature gametocytes, and normal blood picture strongly suggest an advanced or chronic infection, but the intense parasitemia and the high number of trophozoites of all sizes suggest an active infection. Perhaps the parasite was undergoing a relapse of an older infection, or perhaps the infection was recent and acute, with the slide having been made a day or two after peak synchronized schizogony.

The parasite's structure coincides in nearly every aspect with *Plasmodium floridense* THOMPSON & HUFF 1944<sup>23</sup>, and is not re-described here. Apparently mature schizonts (Figs. 12-15) were nearly all fan-shaped, 6.4 to 8.8  $\mu$  across, and contained 6 to 14, average 8.6 nuclei ( $N = 25$ ); 25 consecutive segmenters contained the following numbers of merozoites: three had 12 nuclei, four had 10, two had 9, twelve had 8, one had 7, and three had 6 nuclei; 24/25 were in a

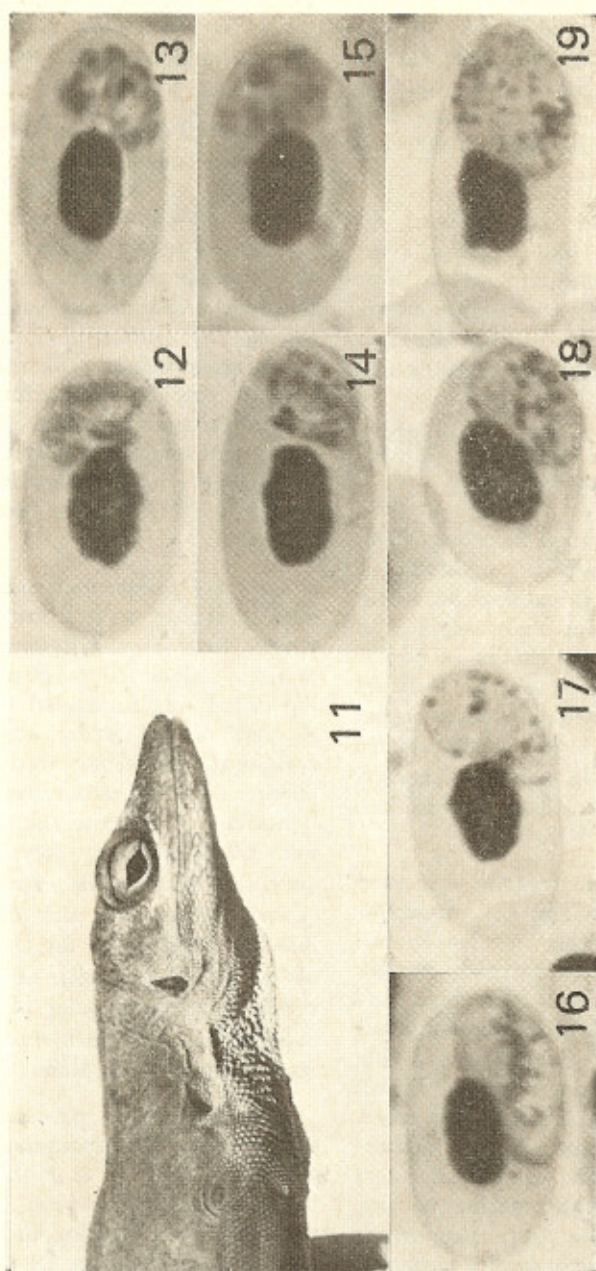


Fig. 11 — *Anolis concolor*, host of *Plasmodium floridense* on San Andrés Island. 12-15, *P. floridense* mature segments. 16-19 mature gametocytes.

polar position; no circulating exoerythrocytic schizonts were found. Mature gametocytes (Figs. 16-19) were 9.4 (5.5 to 16) by 5.8 (4 to 7.4)  $\mu$  ( $N = 25$ ), with a size (L x W) of 56.5, a length-width ratio (L/W) of 1.8; and a size relative to the host cell nucleus (gametocyte L x W / HC L x W) of 2.4; 19/25 were polar or subpolar in the host cell (round, 7 to 9  $\mu$  in diameter, or elliptical 5 to 9 by 4.2 to 5  $\mu$ ).

This parasite differed slightly from the Florida population<sup>23</sup> in that it had a lower merozoite number, segmenters which were nearly all fan-shaped, pigment golden-brown, and the host cells neither distorted nor their nuclei displaced by either segmenters or mature gametocytes.

#### DISCUSSION

The intense hemogregarine infections in *C. lemniscatus* from Providencia suggest that it may be the parasite's principle host. This large, blue lizard is exceptionally common on the island, overlapping the ranges of all the other lizard species in most areas. It is not surprising that a single hemogregarine species can infect lizards from such distantly related families as teiids, iguanids, skinks and geckos. Cross-family and even lizard-snake infections have been reported<sup>2, 4, 17, 18</sup>.

The more intense hemogregarine infections in *A. ameiva* and especially *C. lemniscatus* may be due to their eating infected lizards. I watched whiptails capture and swallow adult *A. pinchoti* twice in four days, and CORN & DALBY<sup>8</sup> mention similar observations. LANDAU et al.<sup>17</sup> have reported that snakes fed hemogregarine-infected lizards can directly acquire infections by this route. Its lizard-eating habit must expose *C. lemniscatus* frequently to acarine ectoparasites that could be harboring hemogregarine oocysts. The lower hemogregarine prevalence or intensity in the other lizard species probably reflects the scarcity of haematophagous arthropods in their diet.

Unfortunately, Teiid lizard populations were not sampled on San Andrés. *A. ameiva* is not known to occur there and *C. lemniscatus* may have been introduced from Pro-

videncia within the last 30 to 40 years<sup>9</sup>. Five *C. lemniscatus* adults brought from San Andrés to Cali in 1972 by Joyce Spain were uninfected. *A. concolor* and *C. lemniscatus* ranges do not closely coincide, and I found no hemogregarines in the 98 San Andrés anoles.

Structural differences between *P. floridense* from San Andrés and Florida are well within the limits of expected and explainable variation. THOMPSON<sup>21</sup> and JORDAN<sup>15</sup> showed that the average number of merozoites in *P. floridense* segmenters is significantly lower after an infection has passed its peak than during the period of initial acute rise, a phenomenon shared with many other malaria parasite species and attributed to the host's immune response<sup>13</sup>. Merozoite numbers also vary considerably in different strains of *P. floridense*<sup>15</sup>, in different lizards of the same species<sup>20, 21, 22</sup>, and in lizards of different species<sup>16, 20, 23</sup>.

The very presence of malaria in anoles on San Andrés at such a low level (one of 98 lizards from nine different areas) was surprising. About 10 years previously, most of the island was fumigated heavily with DDT as part of the *Aedes aegypti* eradication campaign. Residents remembered a complete absence of biting mosquitoes for several years following. As part of the campaign, swamps were filled and today they are almost absent, occurring only in the extreme southern end of the island, very near the site where the infected lizard was found. The remainder of the island has been almost entirely converted to coconut palm plantations. Lizard malaria may well have been common on San Andrés in previous times.

The low malaria prevalence and reported effectiveness of the mosquito control campaign tempts speculation that the parasite might be an avian *Plasmodium* species carried to the island in migrating birds. However, no similar parasites have been described from birds<sup>10</sup>, and it has not yet been demonstrated that any *Plasmodium* species from birds will infect lizards or *visa versa*, despite broad ultrastructural similarities.

More likely, the parasite represents a population of *P. floridense* that reached San

Andrés in late Cenozoic or Quaternary times and found there acceptable habitat and intermediate hosts. Like *P. mexicanum*<sup>3, 4, 12</sup>, *P. floridense* has a disjunct distribution, with isolated populations in both *Sceloporus* and *Anolis* species in the southeastern United States<sup>11, 16, 23</sup> and in Middle America including Panama, Costa Rica, Honduras, Belize and San Andrés Island<sup>14, 20</sup>. Several reptile genera have a similar disjunct distribution, the result of habitat changes associated with periods of glaciation during the Quaternary<sup>1, 7</sup>.

There may be two hypotheses of how *P. floridense* achieved its present distribution. It may be a Middle American offshoot of *P. tropiduri* as TELFORD<sup>20</sup> suggests. If so, it probably extended its distribution north and eastward to Florida along a moist circumferential Gulf Coast corridor<sup>1</sup> during the Quaternary. It is easy to visualize *Sceloporus* or *Anolis* species (or both) harboring *P. floridense* on the Pleistocene Gulf Coast.

Alternatively, *P. floridense* could have been widespread during the Quaternary on the larger Caribbean islands, especially Cuba and the now-submerged Bahama bank. *Anolis carolinensis*, its current anole host in the United States, was a Quaternary invador from Cuba. Anoles of the same Caribbean *A. carolinensis* species complex have successfully colonized the Bay Islands and Half Moon Cay, continental shelf islands off Honduras and Belize, and surely reached mainland Middle America many times (from Cuba?), although failing to establish permanent populations there<sup>24</sup>. *P. floridense* could thus have been introduced to both Florida and Middle America from the large Caribbean islands, and in this case it may still occur in anoles of Cuba, Hispaniola or Jamaica.

#### RESUMO

*Malária e hemogregarinas dos lagartos das ilhas Colombianas de San Andrés e Providencia, no Caribe Oriental*

*Plasmodium floridense* ocorre em Iguânidos na ilha colombiana de San Andrés, em toda a América Central e no sudeste dos Estados Unidos. Possivelmente evoluiu do *P.*

*tropiduri*, ou na América Central ou nas ilhas maiores do Caribe. Lagartos das famílias Iguanidae, Gekkonidae, Scincidae e Teiidae encontrados na ilha de Providencia são infectados pelo que parece ser uma só espécie de hemogregarina, aparentemente um *Hepatozoon*.

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