INVESTIGATION OF THE ENTERIC BACTERIA OF THE TESTUDINATA — I: OCCURRENCE OF THE GENERA Arizona, Citrobacter, Edwardsiella and Salmonella

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An investigation of the enteric bacteria of wild and captive turtles was initiated early in 1968. Individuals belonging to 20 species, which represent 2 families and 10 genera have to date een found positive for the presence of H_2S positive, urease negative enteric bacteria of the family Enterobacteriaceae. The genera *Arizona, Citrobacter, Edwardsiella* and *Salmonella* have been isolated.

McNeil and Hinshaw (1946, Amer. J. Vet. Res. 7(22); 62-63) were the first to report *Salmonella* in turtles. At autopsy of two Galapagos tortoises in the San Diego Zoo, *Salmonella newport* and *S. sandiego* were found extensively throughout the internal organs. A number of additional reports of the occurrence of *Salmonella* in turtles have been made since that time. The most commonly investigated turtles have been members of the family Testudinidae, belonging to the genera *Pseudemys, Chrysemys* and *Testudo*. Kaufmann and Morrison (1966, Am. J. Epidem. 82(2): 364-370) summarized the literature with respect to *Salmonella* and *Arizona* in turtles, noting the public health significance. Kaplan (1957, Lab. Animal Care 7: 273-277) described a septicemia in turtles caused by *Citrobacter* (Bethesda-Ballerup Group). Jackson et al (1969, Assn. Southeastern Biologists Bull. 16 (2): 55) were the first to report *Edwardsiella tarda* in turtles. Rudat et al (1966, Pathol. Microbiol. 29(5): 623-629) and Lie (1968, Arch. Hyg. Bakteriol. 152(2): 139-155) reported *Salmonella* in zoo collections in Switzerland and Germany, respectively.

Methods

Samples were collected by inserting a sterile swab directly into the cloaca. The swab was then incubated in Selenite-F enrichment medium at 35°C for 18 to 24 hours. Routine procedures and culture techniques using Desoxycholate Agar plates and Kligler's Iron Agar slants were employed to isolate H_2S positive forms. Urease, ONPG (O-Nitrophenyl Beta-D-galactopyranoside), and indol tests were done on the isolates. H_2S positive, urease negative cultures were then serotyped.

Water samples were also occasionally tested from tanks holding turtles. 5 cc samples were collected with sterile pipettes and discharged into 5 cc double-strength Selenite-F enrichment medium. Turtles too small for a cloacal swab can be checked by collecting a fresh fecal pellet and discharging it into the Selenite-F medium.

Findings

Of 53 individuals designated as wild (having been in strict isolation in captivity for not more than 2 weeks and unfed), 8 (15%) were positive; of 123 captive individuals, 28 (23%) were positive. As is shown in Table 1, *Salmonella* (all forms) and *Citrobacter* (all forms) were the most commonly encountered bacteria, each being isolated from 9 species of turtles. *Arizona* was isolated from only a single turtle species. Five individuals were positive for two of the types of bacteria tested for.

It is of ecological interest that there were several additional bacterial species present, though only a tentative identification is available as yet. Of the first 40 turtles examined, 29 (70%) were positive for *Aeromonas hydrophila*. Members of the genera *Proteus, Aerobacter, Escherichia, Pseudomonas,* as well as streptococci were provisionally identified

by colonial morphology and other reactions. Further work with these associated species has been deferred until the primary objectives of the study have been achieved.

It is anticipated that the accumulating data will shed light on questions regarding prevalence of infection, degree of pathogenicity, and geographic distribution.

TABLE 1. *Turtles with their associated bacteria (C = captive; W = wild)*

Arizona
Chelydra serpentina (C)

Citrobacter
Geochelone elegans (C)

Citrobacter (Bethesda-Ballerup Group)
Chrysemys picta (C)
Chrysemys picta dorsalis (C)
Geochelone denticulata (C)
Gopherus polyphemus (C)
Kinosternon leucostomum (C)
(water sample)
Kinosternon subrubrum (W)
Pseudemys floridana (W)
Pseudemys scripta elegans (C)
Terrapene carolina bauri (C)

Edwardsiella tarda
Chrysemys picta (W)
Geoemyda annulata (C)
Pseudemys concinna suwanniensis (C)
(3 individuals)
Pseudemys nelsoni (C)
Pseudemys scripta (W & C)
Pseudemys scripta ornata (C)
Sternotherus minor minor (C)
Terrapene carolina major (C)
Terrapene ornata (C)

Salmonella bredeney
Geochelone elegans (C)

Salmonella infantis
Chelydra serpentina (W)

Salmonella livingstone
Terrapene carolina major (C)

Salmonella miami
Sternotherus minor minor (W)
(3 individuals)

Salmonella newport
Pseudemys scripta (C)

Salmonella thompson
Geoemyda annulata (C)
Macrochelys temmencki (C)
Pseudemys nelsoni (C)
Pseudemys scripta ornata (C)
Terrapene carolina bauri (C)

Salmonella Group C_1
(serotype undetermined)
Geochelone denticulata (C)
Geochelone elegans (C)

Salmonella Group E
(serotype undetermined)
Geochelone denticulata (C)

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