

## Herpesvirus in red-headed (common) agamas (*Agama agama*)

Gary L, Watson

Diseases induced by herpesviruses affect a broad spectrum of mammalian species, including humans, and have also been identified in birds, amphibians, and reptiles.<sup>1-4,7</sup> This case report describes a herpesvirus-induced disease in a group of red-headed (common) agamas.

Nine wild-caught red-headed agamas (*Agama agama*) were purchased from an animal dealer by the Detroit Zoological Park. The animals were quarantined prior to display, but all of the animals subsequently died; 8 over a period of 38 days and the ninth 6 months later. All of the lizards were necropsied by 1 of the zoo's veterinarians. The 2 agamas (agama nos. 1 and 2) that are the subjects of this report were found dead on the same day. Agama no. 1 had multiple pin-point white foci in the liver but no other evident gross lesions. Agama no. 2 had no gross lesions. Multiple tissues from both lizards were obtained at necropsy, fixed in 10% neutral buffered formalin, and submitted to the Animal Health Diagnostic Laboratory. The tissues were processed routinely and stained with hematoxylin and eosin (HE) for examination. Microbiologic and virologic examinations were not performed.

The liver and spleen of agama no. 1 contained multiple randomly distributed foci of necrosis. These regions had little associated inflammation, but at the periphery there were numerous eosinophilic to slightly amphophilic intranuclear inclusion bodies with peripheral halos (Fig. 1). The characteristics of these bodies were consistent with those of herpesvirus inclusion bodies. Within some of the necrotic hepatic foci

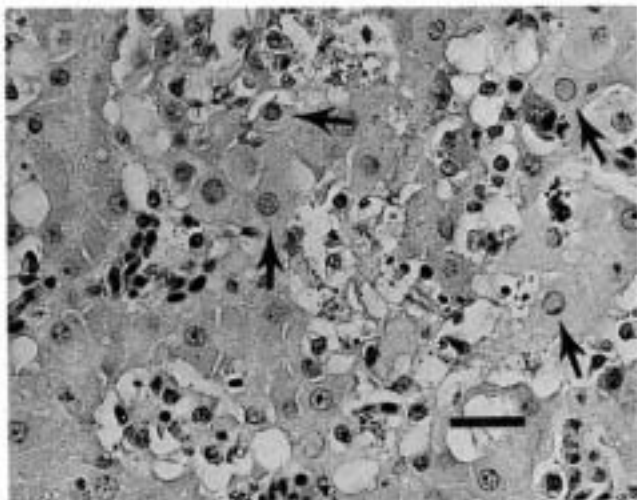
were clusters of gram-negative coccobacilli. The lung contained an occasional intranuclear inclusion body which totally filled the nuclei of airway epithelial cells. Agama no. 2 had a mild exudative pneumonia with similar inclusions in the lung but none in other tissues. With the exception of mild postmortem autolysis, there were no other significant lesions in the heart, stomach, kidney, small and large intestine, testicle, and trachea of either animal or in the spleen and adrenal gland of agama no. 2. Tissues were obtained from the liver blocks of agama no. 1 and from the lung blocks of both agamas. The tissues were deparaffinized, postfixed in osmium tetroxide, stained *en bloc* with uranyl acetate, and embedded in resin. Ultrathin sections were examined with a Philips 301 electron microscope.

Electron micrographs from agama no. 1 revealed intranuclear and intracytoplasmic virions of a size (approximately 100 nm) and morphology consistent with a herpesvirus (Fig. 2). In an electron micrograph from the lung of agama no. 2, several "naked" intranuclear viral particles were present, and others, present within an intracytoplasmic invagination or perhaps the endoplasmic reticulum, had outer envelopes (coats), most likely derived from the nuclear membrane<sup>6</sup> (Fig. 3). In 1 electron micrograph from the second agama, there were 2 viral particles budding from the nuclear membrane of a pulmonary epithelial cell (Fig. 4). The virion size, morphology, and mechanism of acquiring an outer coat, was most consistent with a herpesvirus and may help to explain the wide variability in sizes reported for herpesviruses (100-200 nm).<sup>5,6</sup>

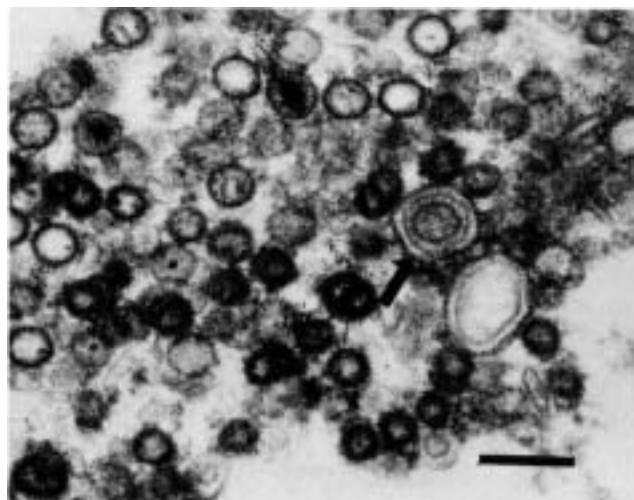
Of the 3 agamas examined, only nos. 1 and 2 had microscopic evidence of viral inclusion bodies. Other problems identified in the remaining 7 agamas included ectoparasitism,

From the Animal Health Diagnostic Laboratory, Michigan State University, East Lansing, MI 48909-7576.

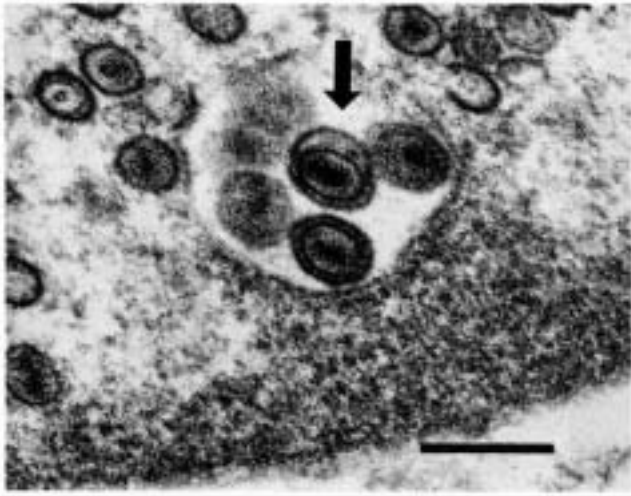
Received for publication November 3, 1992.



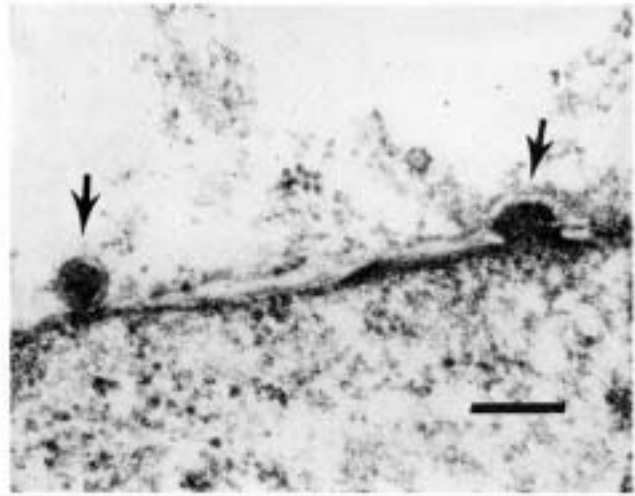
**Figure 1.** Photomicrograph of liver, agama no. 1. Note intranuclear inclusion bodies (solid arrows). HE. Bar = 32  $\mu$ m.



**Figure 2.** Electron micrograph of hepatocyte, agama no. 1. Note intracytoplasmic virions and solitary coated viral particle (solid arrow). Bar = 200 nm.



**Figure 3.** Electron micrograph of lung, agama no. 2. Note intranuclear virions and coated particles (arrows) in intranuclear invagination. Bar = 200 nm.



**Figure 4.** Electron micrograph of lung, agama no. 2. Two viral particles (arrows) are budding from nuclear membrane. Bar = 200 nm.

endoparasitism (trematodes, cestodes, nematodes, and microfilaria), and various degrees of postmortem autolysis. One animal had succumbed to a necrotizing enteritis of bacterial origin, but definitive causes of death were not apparent in the remaining 6 agamas. None of these 7 agamas had any evidence of viral disease or of inclusion bodies.

Several reports have described "herpes-like" viruses in reptilian species. These viruses induced necrosis in the liver of Pacific pond turtles;<sup>1</sup> the glottis, trachea, lung, spleen, and kidney of green sea turtles;<sup>2</sup> the glottis and pharynx of Argentine tortoises;<sup>2</sup> the liver of map turtles;<sup>4</sup> and the venom gland of Siamese cobras.<sup>7</sup> Virus particles were readily demonstrated via electron microscopy in all of these species. A virus that induced cytopathic effects, giant cell formation, and inclusions in green sea turtle (GST) kidney cell culture was isolated only from the green sea turtles.<sup>3</sup> Attempts at virus isolation from the other reptiles, using GST fibroblasts<sup>2</sup> and viper heart tissue culture cells,<sup>4,7</sup> were unsuccessful.

The failure to submit fresh or frozen tissues precluded attempts to culture or further characterize the virus in the 2 agamas. The tissue distribution (liver, lung, and spleen), tissue response (hepatic and splenic necrosis), the characteristic intranuclear inclusions, and supportive electron microscopic findings (virus size, morphology, and behavior)<sup>5,6</sup> support a diagnosis of herpesvirus disease in these 2 agamas.

*Acknowledgements.* I thank Drs. Dalen Agnew and Robyn Barbiere and the Detroit Zoological Park for submission of

the agama specimens, Carol Ayala for assistance with electron microscopy, and Ralph Common for assistance in photograph preparation.

### References

1. Frye FL, Oshiro LS, Dutra FR, Camey JD: 1977, Herpesvirus-like infection in two Pacific pond turtles. *J Am Vet Med Assoc* 171:882-884.
2. Jacobson ER, Clubb S, Gaskin JM, Gardiner C: 1985, Herpesvirus-like infection in Argentine tortoises. *J Am Vet Med Assoc* 187:1227-1229.
3. Jacobson ER, Gaskin JM, Roelke M, et al.: 1986, Conjunctivitis, tracheitis and pneumonia associated with herpesvirus infection in green sea turtles. *J Am Vet Med Assoc* 189:1020-1023.
4. Jacobson ER, Gaskin JM, Wahlquist H: 1982, Herpesvirus-like infection in map turtles. *J Am Vet Med Assoc* 181:1322-1324.
5. Katzenstein ALA, Askin FB: 1990, Infection. I. Unusual pneumonias. In: *Surgical pathology of non-neoplastic lung disease*, vol 13, Major problems in pathology, 2nd ed., pp. 323-334, WB Saunders Co., Philadelphia, PA.
6. Matthews REF: 1982, In: *Classification and Nomenclature of Viruses*, Fourth Report of the International Committee on Taxonomy of Viruses. Karger, Basel, Switzerland, and University of Auckland, Auckland, New Zealand.
7. Simpson CF, Jacobson ER, Gaskin JM: 1979, Herpesvirus-like infection of the venom gland of Siamese cobras. *J Am Vet Med Assoc* 175:941-943.