

NOTE

Hepatic lesions in a redstriped rockfish (*Sebastes proriger*) suggestive of a herpesvirus infection

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ABSTRACT: Unusual lesions were observed in a redstriped rockfish (*Sebastes proriger*) collected during a survey of marine fishes off the coast of British Columbia, Canada. This particular fish exhibited prominent hepatomegaly, with large, coalescing, multiple hemorrhages. The affected liver exhibited remarkable histological changes that, taken together, strongly suggested infection by a virus of the herpesvirus group. Multiple, multinucleated giant cells or syncytia of hepatocytes occurred throughout the liver and were associated with massive, coalescing areas of coagulation necrosis, edema, congestion and cavernous hemorrhages (peliosis hepatis) with thrombosis. In addition, the liver showed multifocal inflammation, characterized by perivascular and peribiliary cuffing of mononuclear inflammatory cells. High magnification of the syncytia revealed that the nuclei were pleomorphic, hyperchromatic, and typically contained eosinophilic to densely amphophilic inclusion bodies of varying size, closely resembling Cowdry Type A inclusions. These inclusions stained red to purple in Feulgen's stain, indicating presence of DNA. Electron-lucent spheres (~100 nm diameter) were observed within hepatocyte nuclei by transmission electron microscopy, suggestive of herpesvirus capsids. To our knowledge, this is the first report of a putative or confirmed herpesvirus infection in any rockfish of the genus *Sebastes*.

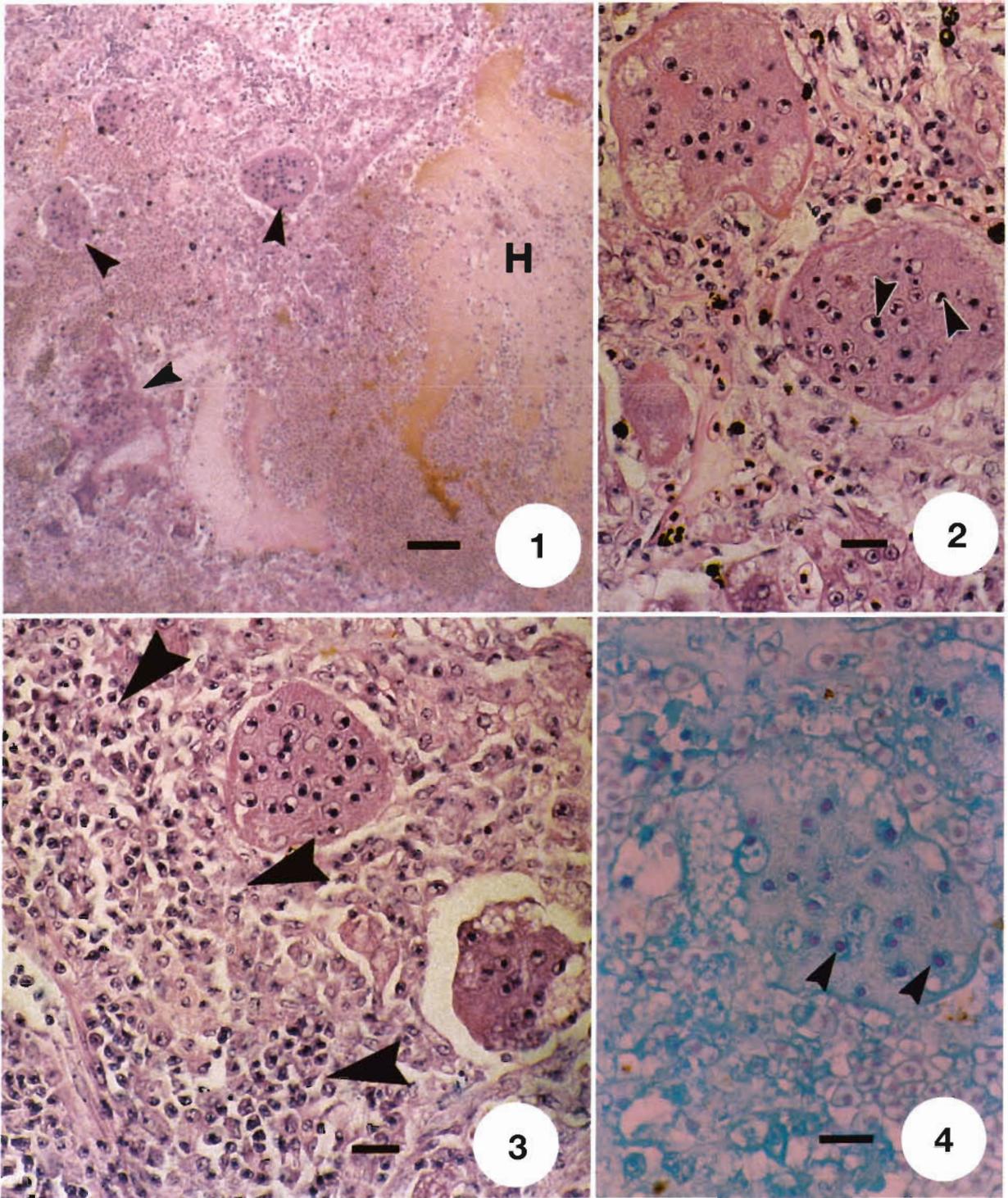
KEY WORDS: Herpesvirus · Rockfish · Liver lesions

Herpesviruses (family Herpesviridae) are well recognised pathogens of captive and wild marine and freshwater fishes. To date, at least 20 herpesviruses have been described from a wide taxonomic host range of fishes (Wolf 1988, Hedrick & Sano 1989, Hetrick & Hedrick 1993). Many are epitheliotropic and are associated with skin lesions (e.g., Pacific cod herpesvirus, pike herpesvirus, walleye herpesvirus, shark

herpesvirus, sheatfish herpesvirus, smelt papilloma herpesvirus, turbot herpesvirus, *Herpesvirus cyprini*), whereas others cause systemic disease (e.g., channel catfish virus, *Herpesvirus salmonis*, *Oncorhynchus masou* virus) (Wolf 1988, Hetrick & Hedrick 1993). Cells infected with several viruses of this family exhibit typical, amphophilic to eosinophilic intranuclear inclusion bodies (Cowdry Type A inclusions), surrounded by an artifactual halo, that possess demonstrable DNA by the Feulgen stain (Roizman 1982, Von Lichtenberg 1984). Some, such as *Herpesvirus salmonis*, the closely-related steelhead herpesvirus, the turbot herpesvirus, and other herpesviruses infecting mammalian hosts, cause syncytia or multinucleated giant cell formation, along with coagulative necrosis, edema, and associated hemorrhage in internal organs such as the liver and pancreas (Strano 1976, Wolf & Smith 1981, Von Lichtenberg 1984, Wolf 1988, Eaton et al. 1989).

As part of an ongoing survey of diseases in wild marine fishes of British Columbia, Canada, we observed remarkable liver lesions in 1 of 36 redstriped rockfish (*Sebastes proriger*) collected off the coast of Vancouver Island lat. 51° 20.2' N, long. 129° 33.7' W on 24 August 1996. A variety of fish species were collected by otter trawl and necropsied on board the research vessel 'W.E. Ricker'. This particular fish exhibited prominent hepatomegaly, with large, coalescing, multiple hemorrhages, while other internal organs, gills and integument appeared normal. The liver, other internal organs, and gills were preserved in Davidson's fixative for histological analysis. Tissues were prepared using standard histological methods and sections were stained with either Harris' hematoxylin and eosin or Feulgen's nuclear stain for DNA (Preece 1972). Liver tissue that had been preserved in

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Figs. 1 to 4. *Sebastes proriger*. Severely necrotic and hemorrhagic liver infected with putative herpes infection. **Fig. 1.** Low magnification showing hepatocellular syncytia or multinucleated giant cells (arrowheads) in areas of liver exhibiting necrosis, edema, hemorrhage, and thrombosis. Note that the architecture of the liver is completely disrupted. H = hemorrhage. H&E stain. Scale bar = 50 μ m. **Fig. 2.** Hepatocellular syncytia or multinucleated giant cells with densely amphophilic intranuclear inclusions (arrowheads). H&E stain. Scale bar = 10 μ m. **Fig. 3.** Hepatocellular syncytia or multinucleated giant cells in area of liver also showing perivascular inflammation (demarcated by arrowheads). H&E stain. Scale bar = 20 μ m. **Fig. 4.** Feulgen-positive inclusions (arrowheads) in nuclei of hepatocellular syncytia or multinucleated giant cells. Feulgen nuclear stain for DNA. Scale bar = 20 μ m

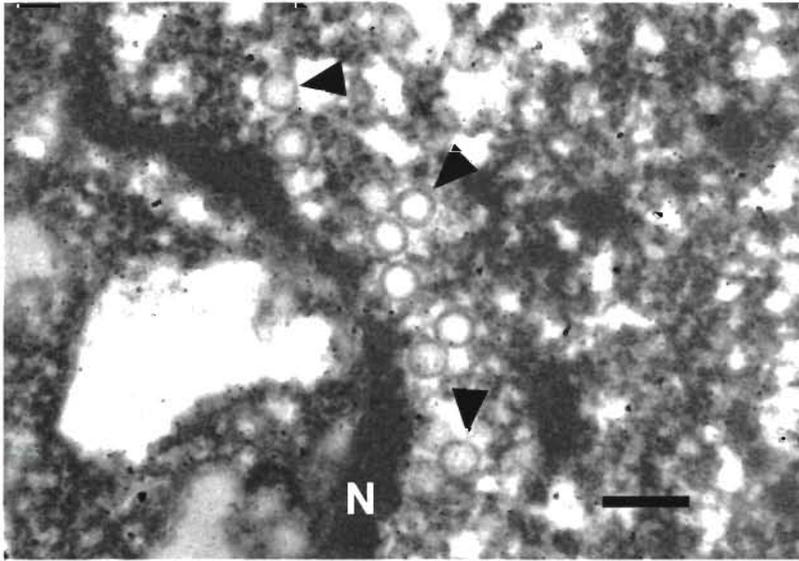


Fig. 5. *Sebastes proriger*. Transmission electron micrograph of hepatocyte nucleus containing putative herpesvirus capsids (arrowheads). N = nuclear membrane. Scale bar = 200 nm

Davidson's solution was also re-processed for transmission electron microscopy using standard methods.

The affected liver exhibited remarkable histological changes (Figs. 1 to 4) that, taken together, strongly suggested infection by a virus of the herpesvirus group. Multiple, multinucleated giant cells or syncytia of hepatocytes occurred throughout the liver and were associated with massive, coalescing areas of coagulation necrosis, edema, congestion and cavernous hemorrhages (peliosis hepatis) with thrombosis (Fig. 1). In addition, the liver showed multifocal inflammation, characterized by perivascular and peribiliary cuffing of mononuclear inflammatory cells (Fig. 3). High magnification of the multinucleate giant cells or syncytia (Figs. 2 to 4) revealed that the nuclei were pleomorphic, hyperchromatic, and typically contained eosinophilic to densely amphophilic inclusion bodies of varying size, closely resembling Cowdry Type A inclusions. These stained red to purple in Feulgen's stain, indicating presence of DNA (Fig. 4). The other internal organs examined (kidney, heart, intestinal tract, exocrine pancreas) and gill exhibited no significant histopathological changes.

The ultrastructural integrity of the liver sample processed for transmission electron microscopy was poor, typical of Davidson's-preserved tissues. However, electron-lucent spheres (~100 nm diameter) were observed within hepatocyte nuclei (Fig. 5).

The formation of syncytia or multinucleate giant cells are a hallmark of infections by some herpesviruses, and occur in both *in vivo* infections in vertebrates and in infected cell cultures (Strano 1976, Jawetz et al. 1982, Roizman 1982, Von Lichtenberg 1984, Wolf 1988). Therefore, hepatic lesions observed in the rockfish, especially the occurrence of hepatocellular syncytia or multinucleated giant cells with Feulgen-positive Cowdry Type A intranuclear inclusion bodies, are consistent with a herpesvirus infection. In addition, although the liver tissue examined by transmission electron microscopy was suboptimally preserved, we detected intranuclear particles consistent in size and shape with herpesvirus capsids. To our knowledge, this is the first report of a putative or confirmed herpesvirus infection in any rockfish of the genus *Sebastes*.

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