

## Ranaviral Disease Pathology



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## Fish

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## Species of fish-specific Ranavirus

- Three species recognized by the International Committee on Taxonomy of Viruses (ICTV)
  - Epizootic Hemorrhagic Necrosis Virus (EHNV; Australia)
  - European Catfish Virus (ECV; Europe)
  - Santee-Cooper Ranavirus (SCRV; primarily North American)

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....And some amphibian ranaviruses have been found to infect fish.

- FV3-
  - In wild fish: moribund threespine stickleback (*Gasterosteus aculeatus*) during a sympatric epizootic involving northern red-legged frogs (*Rana aurora* ; Mao et al. 1999a).
  - In various hatcheries (e.g., Waltzek et al. 2014)
- BIV –only a single outbreak in hatchery-reared Nile tilapia fry (*Oreochromis niloticus*) in Australia (Ariel and Owens 1997 ).

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## Ranaviruses in hatcheries

FV3 and SCRV have been detected in various hatchery reared freshwater fishes in the Americas and Asia (see: Woodland et al. 2002b ; Prasankok et al. 2005 ; Deng et al. 2011 ; George et al. 2014 ; Chinchar and Waltzek 2014 ; Waltzek et al. 2014)

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## Field and gross

- Loss of buoyancy
- Erratic swimming
- Anorexia
- Red swollen gills
- Hemorrhages (especially periorcular, fat bodies, swim bladder)
- Overinflated swim bladder
- Friable (necrotic) organs

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## Fish



Photo: Emilie Travis



Photos: Tom Waltzek



Photo: Ted Henry



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## EHN

- First ranavirus reported in mass die-off of vertebrate (Duffus et al. 2015)
- 1985, Australia, epizootic (see: Langdon et al. 1986, 1988; Langdon and Humphrey 1987); Unknown source of outbreak
- Redfin perch (*Perca fluviatilis*) and Rainbow trout (*Oncorhynchus mykiss*)
- Other species are susceptible based on laboratory challenges but no recent outbreaks (perhaps events aren't detected)

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## EHN-current status

- Endemic in wild Redfin perch in SE Australia
- Impact to aquaculture = farmed rainbow trout in SE Australia
- Redfin perch = highly susceptible; Rainbow trout = relatively resistant (see: Whittington and Reddacliff 1995)

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## EHNV –field and gross

- Melanosis (dark color)
- Anorexic (stopped eating)
- Ataxic
- Swollen abdomen
- Swollen spleen and kidney
- Multifocal pale foci (areas of necrosis) in the liver

See: Reddacliff and Whittington 1996

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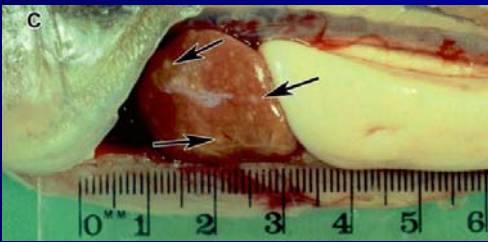
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## Gross lesions

- Multifocal hepatic necrosis and echymotic hemorrhage in the retroperitoneum; adult Redfin perch



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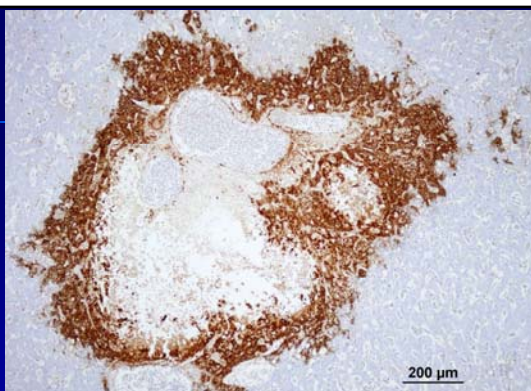
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Immunohistochemical staining in the areas of necrosis in the liver of a Redfin perch (*Perca fluviatilis*). Photo: Richard Whittington

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## ECV in European catfish

by Anna Toffan (DVM, PhD)  
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### Clinical signs

- Mortality 100%
- Melanosis (dark color)
- Exophthalmos (pop eye)
- Anorexia and lethargy
- Erratic swimming, gasping, «candle» position
- Petechial hemorrhages on skin

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### Clinical signs



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### Gross lesion

- Hemorrhages on external and internal organs (skin, fins, bladder, intracoelomic fat, liver)
- Anemic gill with petechiae and/or oedema
- Congestion and protrusion of the anus
- Congestion of the intestinal tract
- Necrotic foci in liver, spleen, kidney
- Spleen and liver enlargement

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Petechial hemorrhages

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### Exophthalmos and eye hemorrhages



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Intracoelomic (abdominal) hemorrhage

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Splenomegaly and petechial hemorrhages on liver



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Vascular congestion (especially on the stomach and intestines)



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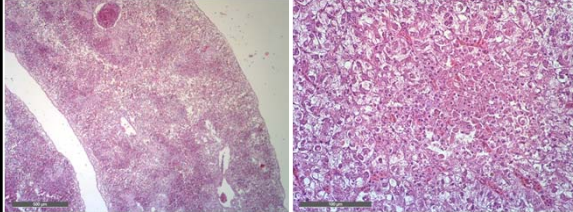
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## Histopathology



Spleen: depletion of lymphoid tissue  
pyknosis and cariorexis of white pulp

Liver: Necrotic foci with pyknosis of  
hepatocytes.

Picture by Tobia Pretto-IZSve

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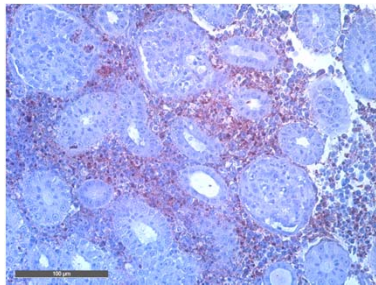
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## IHC

Kidney:  
Positive reaction in  
interstitial lymphoid  
tissue



Picture by Tobia Pretto-IZSve

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## SCRV

### Typical die-off event

- Fish die during summer
- Often only largemouth bass (*Micropterus salmoides*) >30 cm TL found
- Moribund fish at water surface
- External hemorrhages; however, there may be no external lesions unless there is another concurrent disease
- Swim bladder is over-inflated, reddened, or has yellow or brown exudate
- Fish-kill can last for 2-3 months

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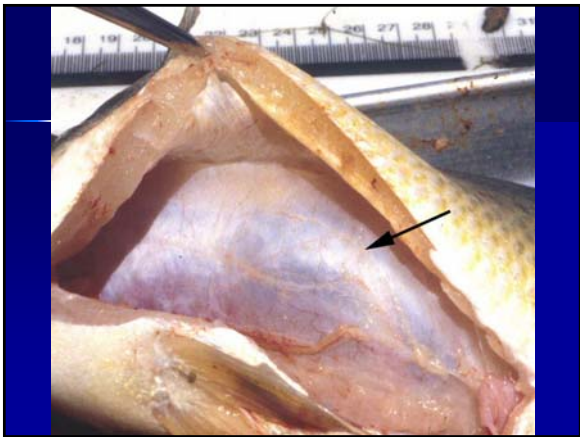
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## SCRV

- Survives in water for several days
- Occurs in fish mucus (sometimes)
- Isolated from trunk kidney and liver 1 hour after LMBV was added to the water in experimental studies

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## SCRV

- Can be found in many other bass species, as well as Crappie and Bluegill and few others

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## Gross lesions

- Necrosis of the epithelial lining of the gastrointestinal tract
- Necrosis of the gills
- Necrosis of the heart

See: Zilberg et al. 2000

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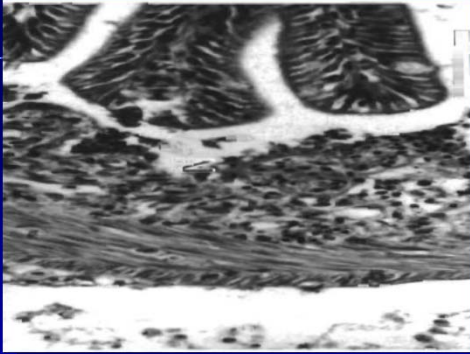
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Photo from Zilberg et al 2000



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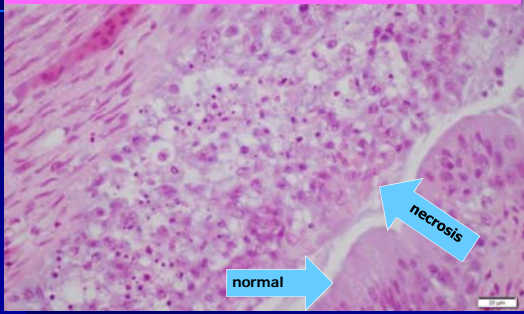
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NOTE: this is a turtle intestine but demonstrates the same lesion



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Ranaviral disease caused by amphibian ranaviruses

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## Gross pathology-FV3

- Pallid sturgeon (*Scaphirhynchus albus*) with cutaneous ecchymotic hemorrhage due to an FV3-like ranavirus. Photo by Thomas B Waltzek, University of Florida.



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## Gross pathology

- Hemorrhage in the fat bodies and spleen of a pallid sturgeon with FV3-like ranavirus. Photo by Thomas B Waltzek, University of Florida.



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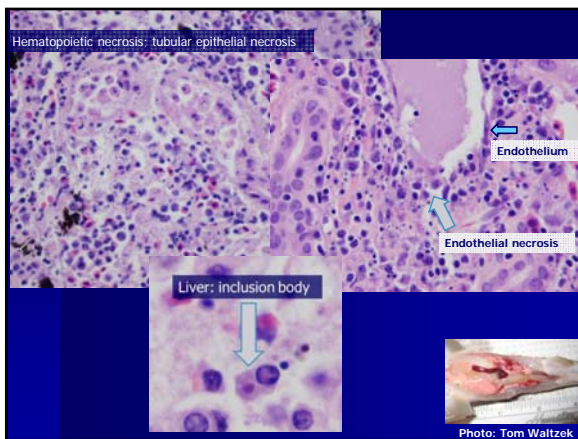
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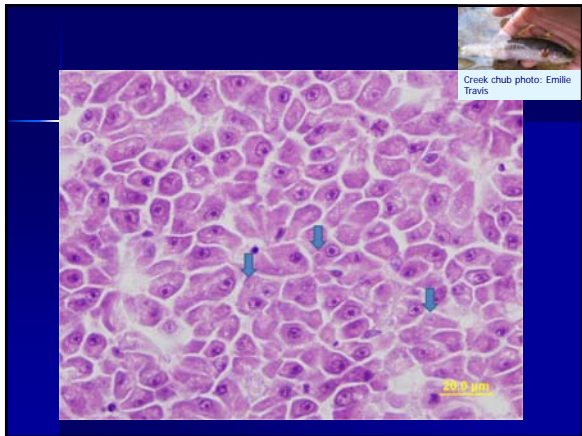
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## Conclusions

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**American bullfrog**  
*Lithobates catesbeiana*

**Eastern box turtle**  
*(Terrapene carolina carolina)*

Photo: Mark Ruder

**Red-eared slider**  
*(Trachemys scripta elegans)*

**Eastern spotted newt**  
*Notophthalmus viridescens*

Photo: Betsie B. Rothermel

**Creek chub**  
*Sanotilus atromaculatus*

Photo: Emille Travis

**Pallid sturgeon**  
*Scaphirhynchus albus*

Photo: Tom Waltzek

**CONCLUSIONS**

- Lesions can be similar across classes but present differently
- Multiple age groups are affected
- Multiple species (and classes) can be involved in a mortality event

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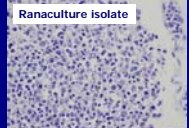
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### Conclusions

- Not only can the severity of lesions vary by host susceptibility, but the severity also can vary by ranavirus isolate




Pallid sturgeon isolate




Ranaculture isolate


- Host susceptibility varies (and thus community composition may matter in epizootics)



Wood frog (~ 100% for both)



Cope's Gray tree frog (~90% RI, ~40% FV3)



Bullfrog (~10% FV3)

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### Conclusions: Ectoparasites may play a role

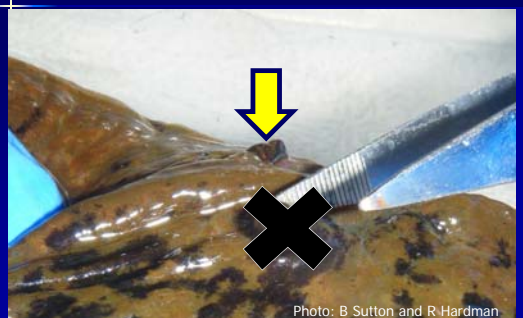


Photo: B Sutton and R Hardman

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### Acknowledgements (most listed throughout)

- Matt Gray
- Tom Waltzek
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- Jordan Chaney
- Richard Whittington
- Becky Hardman
- Ted Henry
- IZSVE
- Mark Ruder
- Betsie Rothermel
- Rolando Mazzoni
- All involved in these projects!

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# Questions?



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