



HOSPITAL  
FOR  
**SPECIAL  
SURGERY**

# Non-Infectious Diseases

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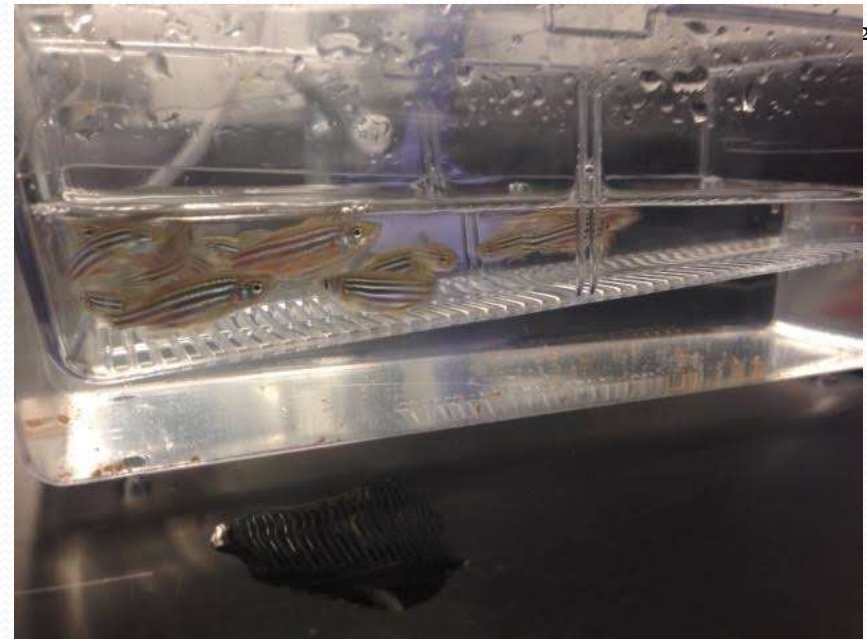
Memorial Sloan-Kettering Cancer Center & Weill Cornell Medical College

5th Annual International Zebrafish Husbandry Course

Buguggiate, Italy 2016

# Water Quality

- Lack of oxygen
  - Respiratory distress
- Inadequate hardness
  - Embryo/larval mortality



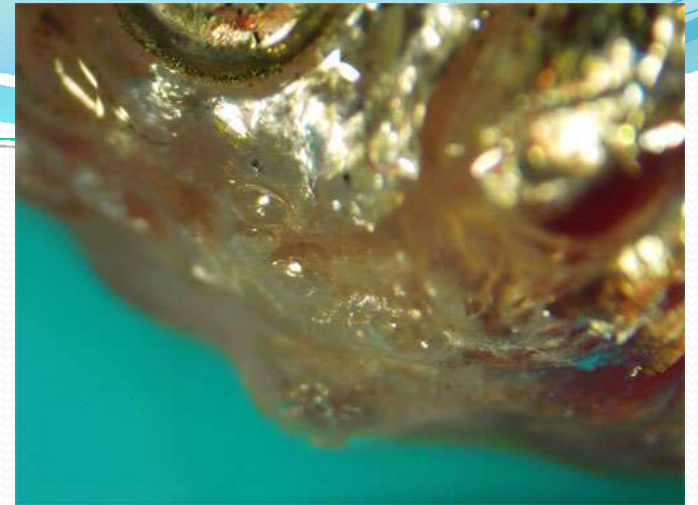


# Gas Supersaturation

- Partial pressure of dissolved gasses in balance with PP of same gases in air ( $N_2$ ,  $O_2$ , AR,  $CO_2$ )
- Air leak in piping suction side of pump\*
- Low water levels causing a vortex in sump
- **Air injection into the system**
- Gas dissolved in the water in excess of equilibrium results in supersaturation
  - Primarily  $N_2$
- **Excess gas will be released from solution**



# Gas Bubble Disease



- Fish tissues become supersaturated with gasses, which are released as bubbles causing tissue damage
- Bubbles
- Buoyancy problems
- Hemorrhage
- Death



DX: Fish on low or no water flow are ok  
Check TDG

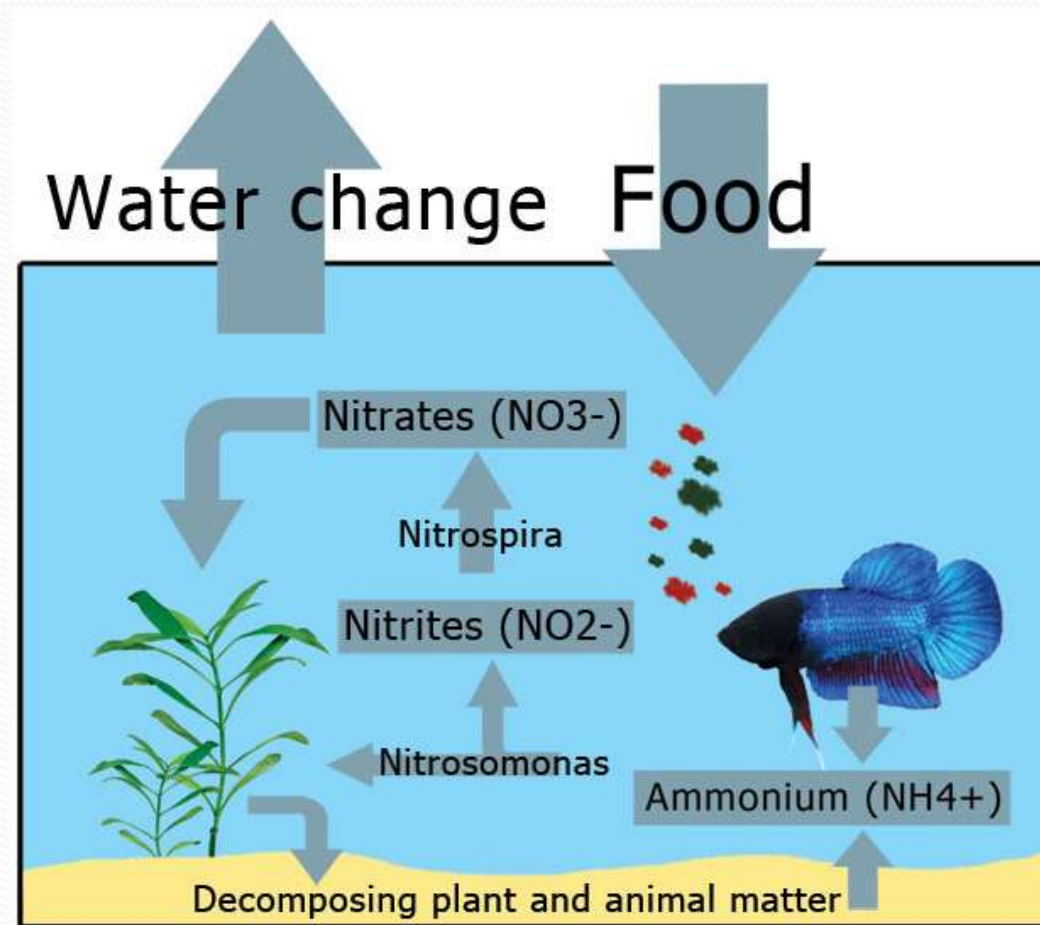
# GBD Treatment

- Stop the pumps!
  - Stop feeding
  - Turn off UV light
- Find the problem
- Maintain water quality without water flow
- Susceptible to secondary infections, negative impact on breeding\*





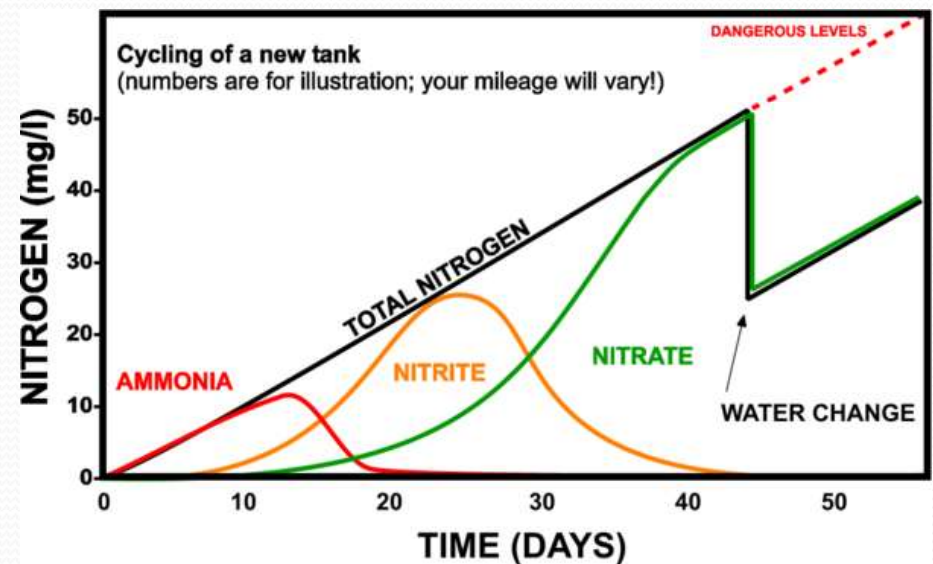
# Nitrogenous Waste Toxicity



<http://www.fnzas.org.nz/wpcontent/uploads/2011/05/NitrogenCycle3.jpg>

# Nitrogenous Waste Toxicity

- Ammonia ( $\text{NH}_3$ ): > 0.02 ppm (mg/L)
  - pH/temp impact
- Nitrite: > 0.5-1.0 ppm
- Nitrate: > 200 ppm
- Diagnosis: Water sample

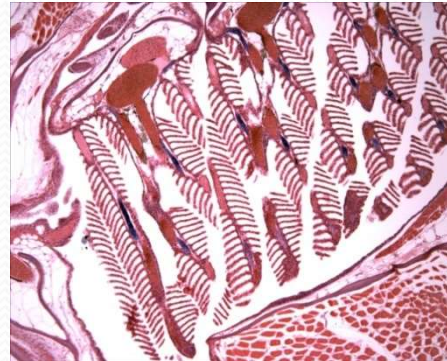




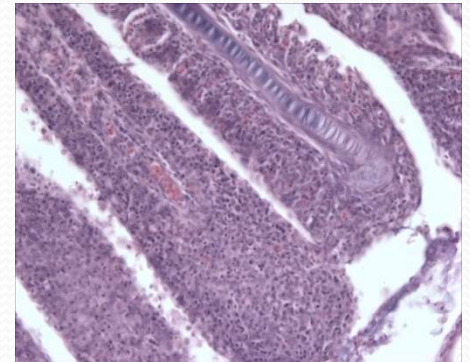
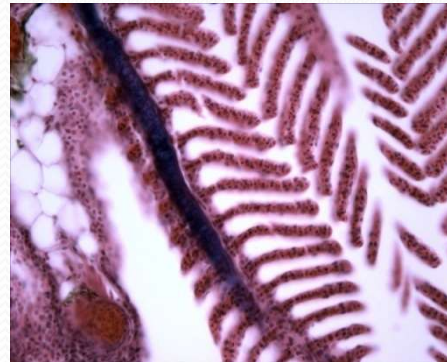
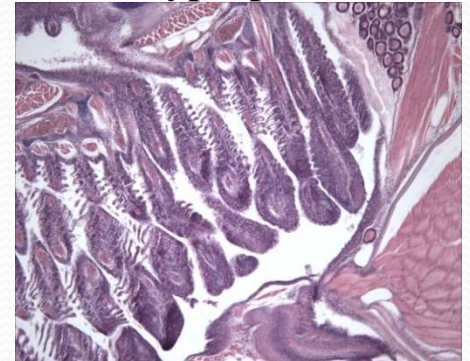
# Ammonia & Nitrite Toxicity

- Ammonia and Urea transporters heavily concentrated in gills
- Physiologic response is age dependent
  - Embryos and Larva more tolerant
- Toxicity influenced by other water parameters

Normal



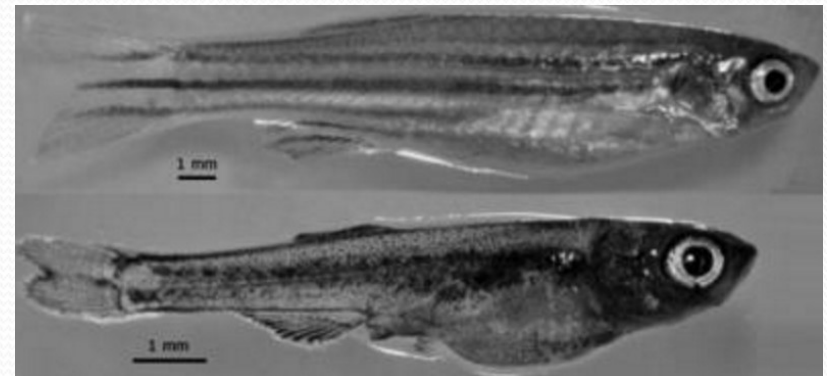
Hyperplastic





# Acute Toxicity

- Behavioral changes
- Respiratory distress
  - Gasping at water surface
  - Lethargy
  - Hemorrhage
- Death



Nitrate > 400ppm; Learmonth 2015

# Sub-acute/Chronic exposure

- Sub-lethal levels
- Reduced growth
- Decreased breeding success
- Stress
  - Immunosuppression
  - Disease (bacterial, fungal, etc.)
  - Death



Courtesy of George Sanders





# Treatment

- Treatment: Correct values **GRADUALLY!**
- Improve water flow- slowly! Watch pH!
- Reduce fish densities
- Stop/decrease feedings

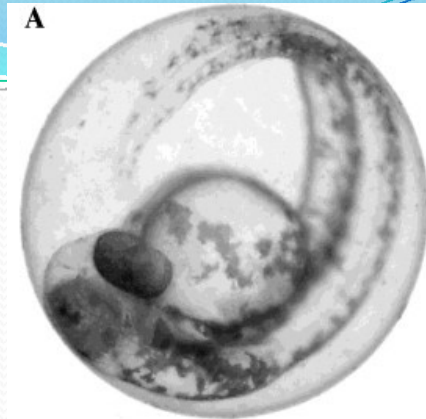
## PREVENT IT!

- Add fish to systems gradually
- Don't over feed
- Regular water exchange

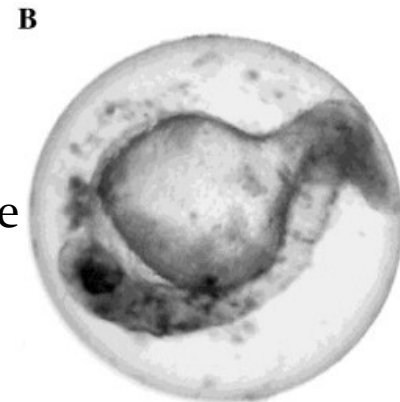
# Other Toxins

- Heavy Metals
  - Copper: Delayed hatching & neurologic signs
  - Lead: Neurotoxicity
- Medications
  - Sensitive- tox study model!

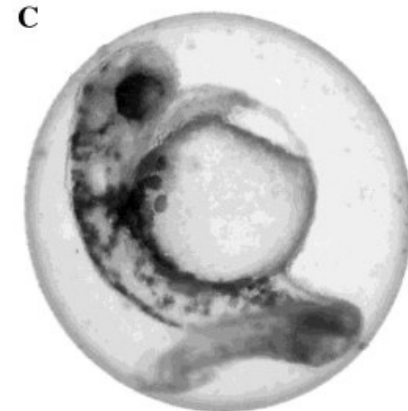
Control



Fenbendazole



Febantel



Carlssona, Gunnar, et al



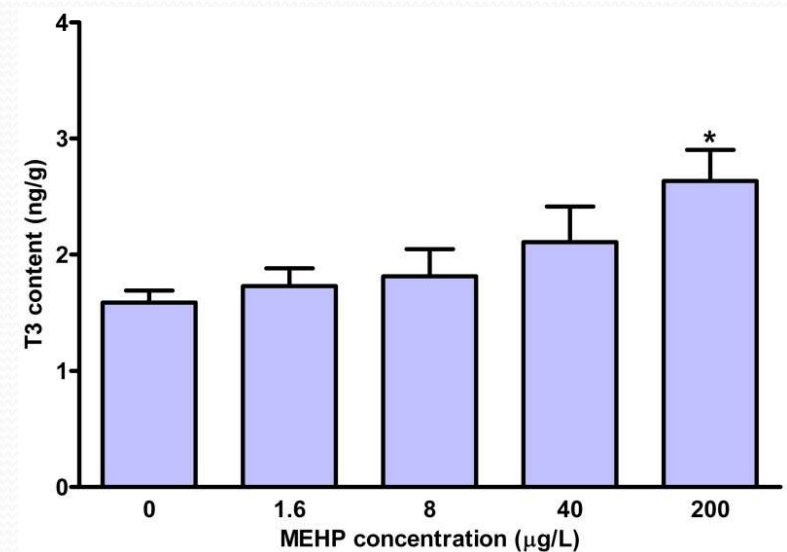
# Chemicals/Toxins

- Chlorine
  - Very sensitive
- Cleaning products
- Gloves
  - Toxicity to latex demonstrated
  - Avoid powdered gloves



# Plasticizers

- Plasticizers
  - Fry are much more sensitive
  - Caution with new systems and enrichment
  - Thyroid endocrine toxicity
  - Disturbance of spermatogenesis
  - Induce genetic aberrations



Zhai, 2014



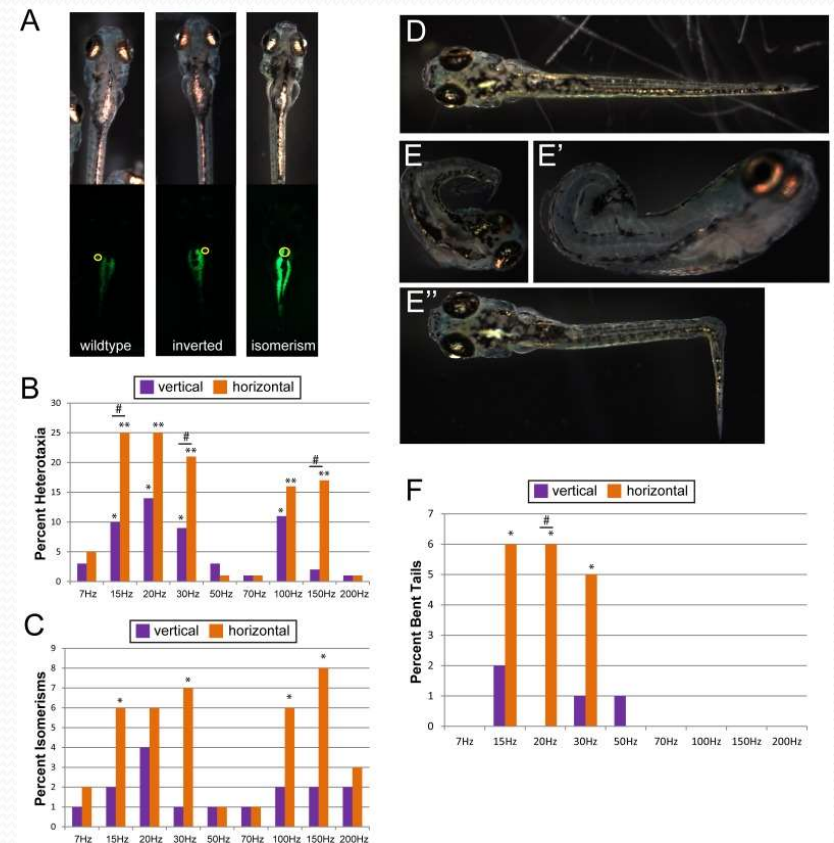
# Lighting

- Light
  - Altered rhythms impact activity and spawning
    - Routine monitoring of room lighting
- Exit signs
  - Control group spawned twice as many embryos compared to those housed in front of a red EXIT sign
  - Green EXIT sign showed no difference
- Other light producing equipment



# Vibrations

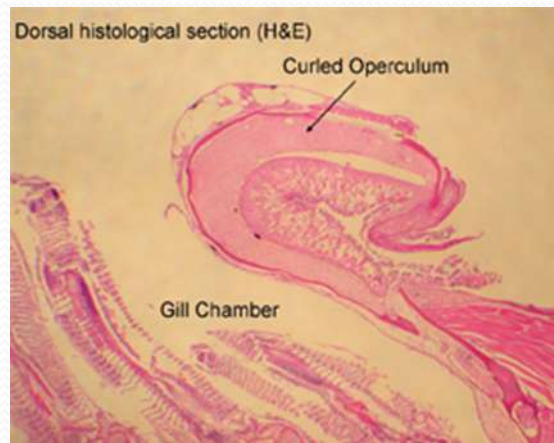
- Low level vibrations
  - Altered left-right patterning and tail morphogenesis
- Continuous vibrations
  - Lowered total activity levels
- Death in other aquatic species



Vandenberg, 2012



# Is this a problem?



ZIRC

# Iatrogenic

- Handling
  - Net catching
  - Egg stripping
    - Dermal abrasions
- Enrichment?
- Experimental
  - Compound administration
  - Behavioral testing devices





# Breeding Practices

- Eggs develop in females regardless of male presence
  - Food availability
  - Water quality



# Egg-Associated Inflammation (EAI)

- Likely caused by egg retention
  - Usually present with abdominal distention
  - Incidence seems to increase with age
- Sequela
  - Secondary Infections\*
  - Aggressive fibroplasia
  - Poor breeder







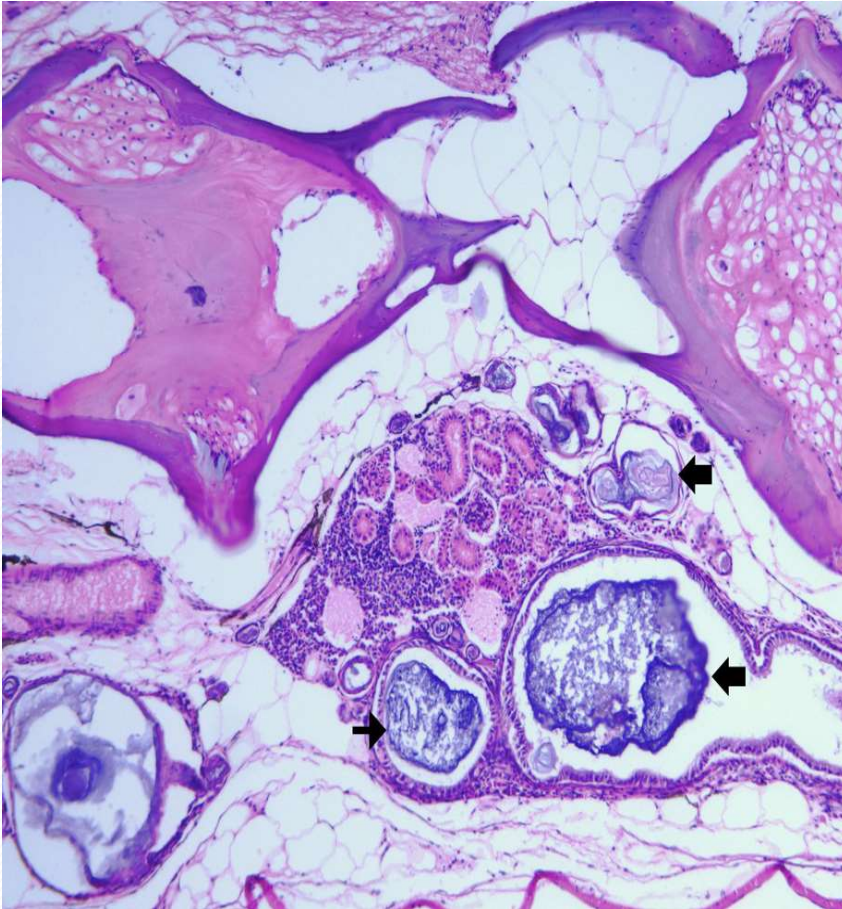
# Egg-Associated Inflammation

- Prevention:
  - Timely spawning of females
  - House males and females together
  - Retire fish appropriately





# Nephrocalcinosis



- Subclinical
- High CO<sub>2</sub>
  - Associated with crowding
- Use of calcium carbonate
  - Recommended buffering with sodium bicarbonate
- Excessive dietary Ca
- Dietary Magnesium
- Selenium toxicity



# Hepatic Megalocytosis

- Enlargement of hepatocyte nuclei and cytoplasm
- Not associated with progression to neoplasia
- Thought to be caused by hepatotoxicants
  - Dietary
  - Algal, etc.
  - Carcinogens
  - Unknown direct cause
- Cells are polyploid (failure of cell division)



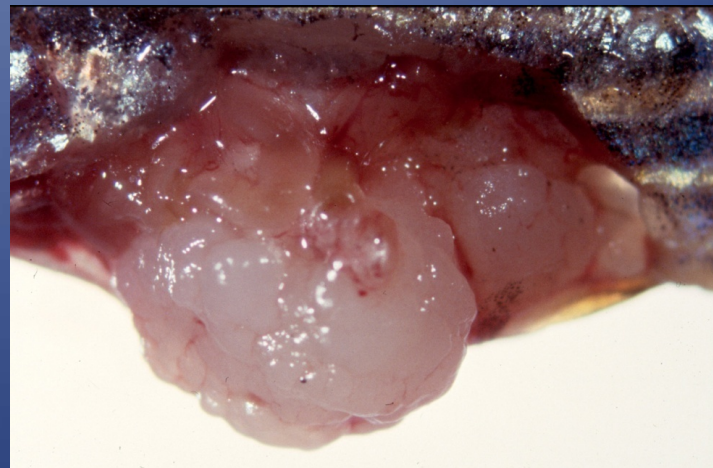


# Neoplastic Disease

- Reported in nearly every organ
  - Most cell types
- Liver most common target organ for carcinogens\*
  - Liver grows throughout life
- Reproductive organs
- Ultimobranchial neoplasia
- Brain, eye, spinal cord
- Not so common:
  - Epithelial
  - Melanoma
  - Liposarcoma
  - Hematopoietic

# Seminoma

- Most commonly reported spontaneous neoplasm of Zebrafish\*
- Can become extremely large (up to 50% of body mass)
- Most are extremely well-differentiated
- Not reported to metastasize





# Hepatic Neoplasia

- Hepatocellular adenoma or carcinoma have been the most commonly observed
  - +/- carcinogen exposure
- Cholangiocarcinoma and mixed also seen
- Soft white to tan masses that may be single or multiple
  - May bulge above the surface



2007 CL Davis Pathology of Genetically Engineered Rodents and Aquatic Species  
J. Spitsbergen Zebrafish III Neoplasia-Aging July 27, 2007  
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THE END

# References

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