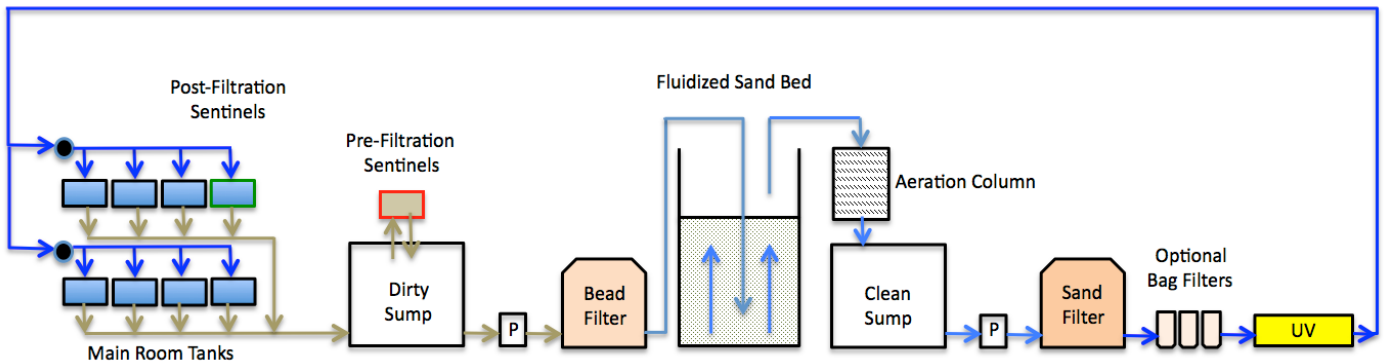




The Animal Health Report provides an overview of health monitoring, diagnostic sampling, and test results for zebrafish raised at ZIRC. The ZIRC raises zebrafish for in-house use and for shipment to customers. Some embryos shipped to customers are generated by in vitro fertilization using eggs from AB fish raised at ZIRC and cryopreserved sperm from males not raised at ZIRC. The health status of the males contributing the sperm was not evaluated and neither the embryos nor paternal stocks have been on the ZIRC water system. The ZIRC recommends use of strict quarantine practices for all imported fish, adults and embryos.

Location: ZIRC main fish room

Description of water system: Three recirculating water systems supply the main fish room. The room is divided into two sides. Water from two systems is intermingled and feeds side A. A separate water system feeds side B.



Water source is reverse osmosis treated municipal water with added salt and aragonite. 10-12% water exchanged per day.

Bead filter – mechanical and biological filtration

Fluidized sand bed – biological filtration

Sand filter – pressurized sand filter for fine particle filtration

UV sterilizer – minimum UV dose 132,000 $\mu\text{Wsec}/\text{cm}^2$

P = pump

New fish strains: Only surface-sanitized embryos enter the main fish room. The majority of new introductions are generated by in vitro fertilization using cryopreserved sperm and eggs from AB females. Occasionally adult fish in the quarantine room are spawned and their surface-sanitized embryos are moved to the main fish room.

Embryo surface sanitization: All embryos are surface sanitized by immersion in 30 ppm sodium hypochlorite for 10 minutes.

Diagnostic testing:

1. The majority of moribund fish are submitted for histopathology.
2. A subset of all 8-month wild-type stocks is submitted for histopathology or PCR for *P. neurophilia*.
3. A subset of retired stocks is submitted for histopathology or PCR for *P. neurophilia*.
4. A subset of fish from the sentinel source tank is screened for *P. neurophilia* by histology or PCR.



5. Pre and post-filtration sentinel fish are submitted quarterly for histopathology. Sentinel samples represent 6 months and 1 year of exposure to system parameters. One-year-exposure sentinels are sampled every 6 months.
6. *Mycobacterium* species is identified by qPCR on frozen fish.

Sentinel fish results:

Sample Date	July 2021	
	Pre-	Post-
Location of sentinel fish relative to filtration and UV lights	Pre-	Post-
Sample information	12 Fish	12 Fish
Time in sentinel tank	6 mos.	6 mos.
GROSS PATHOLOGY	1 red face	Normal
HISTOPATHOLOGY		
Cestode larvae	0	0
Encysted metacercariae (digenetic trematodes)	0	0
Fungal organisms	0	0
Gram-negative bacteria	0	0
<i>Edwardsiella ictaluri</i>	0	0
<i>Ichthyophthirius multifiliis</i>	0	0
<i>Mycobacterium</i> spp.*	0	0
<i>Myxidium streisingeri</i> n. sp. (myxozoa)	0	0
<i>Piscinoodinium</i> sp.	0	0
<i>Pseudocapillaria tomentosa</i> (nematode)	0	0
<i>Pleistophora hypheosobryconis</i> (microsporidia)	0	0
<i>Pseudoloma neurophilia</i> (microsporidia)	0	0
<i>Tetrahymena</i>	0	0

Other non-infectious lesions in sentinel fish: Hepatic megalocytosis, egg-associated inflammation, nephrocalcinosis, focally extensive inflammation involving distal gut wall extending into coelom, and hemorrhage at the tip of the ventral jaw.

****Mycobacterium* spp.**

A single species of *Mycobacterium*, *M. chelonae*, has been identified from zebrafish and biofilms sampled from the ZIRC aquaculture facility (Whipps et al., 2008). We continue to periodically test fish by qPCR for *Mycobacterium*. *M. chelonae* is the only species that has been identified in fish reared in the ZIRC main fish room.

Pathogens detected (all fish sampled):

In last 3 months: *Mycobacterium chelonae*, *Pseudoloma neurophilia*

In last 12 months: *Mycobacterium chelonae*, *Pseudoloma neurophilia*

In last 36 months: *Mycobacterium chelonae*, *Pseudoloma neurophilia*