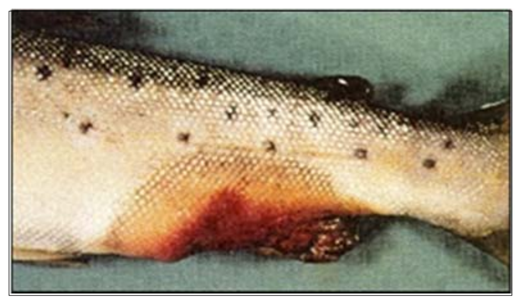
Biosecurity

Biosecurity – Fighting Disease Transmission

A fish's life-long battle against infectious diseases often starts while it is still an egg, as a fight against pathogenic fungi in heavily infested incubation trays. The struggle continues during the production process where factors such as other fish, equipment, and predators, threaten to introduce pathogenic organisms. At every stage fish are challenged with disease of one type or another, caused by the transmission of fungal (e.g. *Saprolegnia saprolegnia*), bacterial (e.g. Enteric Redmouth Disease; ERM), or even viral (e.g. Infectious Pancreatic Necrosis; IPN) infections.

Infection and Disease



Although the details of a specific disease may be quite complex, the basic concept of disease transmission is quite simple. It almost plays out like an episode from "Mission Impossible". The disease organism frantically seeks out a host fish, where in which it maneuvers through the host to an unsuspecting cell or group of cells. Once the target is found, the pathogen replicates, often killing cells and tissues in the process. When all available resources have been utilized, the organism must leave in search of another host. Unfortunately, by that time, the total number of infectious organisms has increased to unthinkable numbers (Table 1), creating an army of disease-causing agents. The best way to avoid such an invasion is to build barriers that will interrupt the disease transmission steps and reduce the likelihood of allowing any pathogen to become established or be transmitted onward.

Biosecurity

Table 1: *Aeromonas salmonicida* shedding rates from infected Atlantic salmon

Group	**No. Fish**	**Mean fish weight (g)**	**Bacterial shedding rate* (cfu/fish/h)**
Sea water			
Bath challenge (10^5 cfu/ml) dead	6	25.8	1.7×10^6 (1.7×10^5 – 1.1×10^7)
i.m. challenge (10^3 cfu) live	4	23.3	$1.3 \text{ X } 10^7$ (5.7×10^5 – 2.1×10^7)
i.m. challenge (10^5 cfu) dead	2	1200	$5.4 \text{ X } 10^7$ (9.0×10^6 – 6.4×10^8)
Fresh water			
Bath challenge (10^5 cfu/ml) dead	3	6.9	$4.1 \text{ X } 10^4$ (1.7×10^4 – 7.0×10^4)

*median values, range in brackets.
Reference: Rose AS, Ellis AE, Munro LS. Journal of Fish Disease. 1989. 12: 573-578.

Biosecurity Battles

Hatchery managers and their staff are on the front lines of the battle against disease transmission. Superior fish health programs include, vaccination, optimum husbandry, biosecurity, and treatment protocols. Although this article focuses primarily on biosecurity, other aspects of successful fish health programs are very important and should not be ignored.

Advanced biosecurity programs include detailed protocols for isolation, disinfection, monitoring and if necessary, destruction of the fish; activities that work best if they are specifically outlined in Standard Operating Procedures (SOP's).

Isolation

The first step in any well thought out biosecurity battle plan is to quarantine or isolate susceptible individuals. All fish should be isolated from other fish, people, and predators. Any vectors that can potentially transmit disease should be identified and dealt with on a case-by-case basis. Clear divisions should be made that separate fish by species, strain, year class, and health status, or any other division that may seem appropriate.

The movement of fish between areas of the hatchery or nursery should be kept at a minimum in order to isolate fish, or maintain secured zones. When it is necessary to move fish between zones, they should be moved only from high to low biosecurity priority. For example, older fish may be have been exposed to more pathogenic organisms than their younger cohorts. Therefore, to avoid disease transmission to younger fish, fish should only move from the incubation room to a first feeding room or a nursery, never the other way.

Biosecurity

Another way to isolate fish is to restrict the movements of people and equipment between zones. Hatchery staff should have designated working areas and whenever possible, each area should have designated equipment that remains in that area at all times.

If movement of people and equipment between zones is absolutely necessary, procedures, usually involving disinfection, should be in place to ensure that there is no cross contamination. Separate battle gear (raingear and boots) in each area can reduce the risk of transferring opportunistic diseases. If that is not possible, disinfection protocols, such as the use of footbaths and handwash stations, must be implemented.

Footbaths are a good way to ensure that biosecurity zones remain isolated. However, organic matter on the boots can severely reduce the efficacy of disinfectant solutions. For this reason, footbaths work best if they are laid out in a two-stage process. The first step is to remove any organic matter, such as mucus, mud, feces or otherwise. This may be done by simply brushing it away with a scrub brush, or by first rinsing the boot in water. The second step is to immerse the boot or shoe in disinfectant solution. A two-stage process will help ensure adequate disinfection and maintain isolation. Table 2 demonstrates how removing organic matter through brushing can increase the efficacy of footbaths.

Table 2: Post-treatment aerobic bacterial counts in cultures from 75-mm^2 (0.12-sq in) of boot sole.

Treatment	**Mean bacterial count (n=5)**	**Standard deviation**
No boot bath	2.78×10^{8} [a]	6.77×10^{7}
Step through Virkon S	1.76×10^{8} [a]	6.06×10^{7}
Stand in Virkon S (2 min.)	2.59×10^{7} [a]	1.01×10^{7}
Scrub in water (30 sec.)	1.04×10^{5} [b]	7.09×10^{4}
Scrub in water (30 sec.) and then step through Virkon S	120 [c]	268
Scrub in Virkon S (30 sec)	20 [c]	45

[abc] Counts with different superscripts are different (P<0.0001).
Boot baths were rubber tubs containing 7.6 L of freshly prepared 1% Virkon S solution, or 7.58 L of water. Brushes used for scrubbing boots were dedicated to either water or Virkon.
Reference: Amass SF, Ragland D, Spicer P, Evaluation of the efficacy of a peroxygen compound, Virkon S as a boot bath disinfectant. J. Swine Health Prod. 2001; 9(3):121-123.

Handwash stations can also help isolate fish from outside invaders. A simple alcohol based hand disinfectant, like Purell's instant hand sanitizers, will easily remove 99.9% of most unwanted germs, and ensure that quarantine or isolation is not breached.

Disinfection

The second phase of the biosecurity battle plan should be disinfection. When total isolation is not possible, the only way to avoid introducing diseases is through cleaning and disinfection. In order to avoid cross contamination among different groups of fish, it is important that all incoming equipment is disinfected, and that each area,

including, floors, walls, and tanks is disinfected on a routine basis. Hard-to-reach areas must not be overlooked or neglected.

Selection of effective disinfection products is important. Chlorine products such as bleach (sodium hypochlorite), while very cheap, are adversely affected by organic matter, sunlight and evaporation. Chlorhexidene gluconates such as Novasan, Virosan, and Hibistat are relatively non-toxic but tend to be less stable and their effectiveness is lowered by organics. Gluteraldehydes like Sterol, Wavecide, and Sporicide on the other hand are extremely stable but may cause eye, skin, and respiratory irritations among the staff. Iodine solutions such as Ovadine, Povidone, and Wescodyne are relatively safe and effective to use but can discolour surfaces (walls tables, etc) and are unstable in light. Finally Quaternary Ammonium Compounds like Roccal, Parvosol, and Merquat, are not as susceptible to organics, however, they are not always effective against bacterial spores, *Pseudomonas*, fungi or mycobacteria, and hydrophilic viruses. In addition, they are tough to rinse away and may leave residues.

Newer families of peroxygen compound disinfectants such as Virkon S (Virkon in Canada) and hydrogen peroxide compound disinfectants such as PeroxiGard and Hyperox are user-friendly, fast acting, and tough on invading pathogens. Products like Virkon (Table 3) and Hyperox are specially formulated and are extremely effective against a wide range of fish pathogens. In addition they contain surfactants that help deal with organic matter.

Table 3: Virkon: effective dilutions for use against specific pathogens

Infectious Organism	**Fish Disease**	**Effective Dilution**
Bacteria		
Aeromonas hydrophila	Generally secondary invader	1:200
Aeromonas salmonicida	Furunculosis	1:1000 – 1:200
Yersinia ruckerii	Enteric Redmouth Disease (ERM)	1:50
Pseudomonas aeruginosa		1:100
Pseudomonas anguilliseptica		1:100
Renibacterium salmoninarum	Bacterial Kidney Disease (BKD)	1:100
Vibrio anguillarum	Vibriosis	1:100
Vibrio	Vibriosis in shrimp/prawns	1:500,000
Viruses		
ISAV	Infectious Salmon Anemia	1:200 – 1:100
Rhabdovirus	Infectious Hematopoietic Necrosis, Viral Haemorrhagic Septicaemia, Spring Viraemia of carp	1:500
Birnavirus	Infectious Pancreatic Necrosis (IPN)	1:100
White spotted red baculovirus	White spot syndrome in shrimp/prawns	1:111,111
Yellow headed virus		1:333,333

Biosecurity

Monitoring

When the biosecurity battle plan is established and isolation and disinfection protocols are in place, the next phase in the war against pathogens is routine surveillance or monitoring. Biosecurity protocols require taking culture samples on a routine basis. This will establish a baseline level of pathogen loading and show how the battle is going. It will also bring attention to any invaders that may have passed the front lines, indicating a breach in biosecurity.

The biosecurity plan itself may need auditing on a regular basis. Local staff can audit daily activities and make note of any obvious shortcomings. Fish health technicians can make routine visits and alert the local staff to any changes that may be needed. Finally, an outside fish health professional or veterinarian can do an independent third party check of the program. This three step monitoring system should ensure that nothing is overlooked, and that any needed change is implemented.

Destruction

If all else fails, and disease-causing organisms penetrate the biosecurity shield, the last option is to destroy the stock. Destruction is a last-ditch effort and must only be performed in the most serious of cases. When infectious disease threatens an entire industry, eradication procedures must be enforced. Eradication often requires the slaughter of all animals that are infected or have been in contact with infected animals. Once the infected animals are removed and properly disposed of, the site must be disinfected and/or fallowed to ensure there is no re-infection.

Conclusion

The battle against infectious diseases is complex and involves many factors from vaccination to fish husbandry to medication. Biosecurity is an important and sometimes overlooked factor in this fight. It involves: **isolation** from vectors that may transport disease; **disinfection** of anything, or anybody that may enter the unit; and routine **monitoring** of the system to ensure that pathogens are not getting through. The threat of disease is present at all stages of fish production. For this reason, biosecurity must be woven into hatchery procedures and become company policy. By lowering our guard we open doors to opportunistic organisms, starting as early as the ones that can infect our incubation trays.