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WATER SUPPLY SANITATION

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Disinfection of water used in fish culture is an effective means of disease control in fish culture situations (Burrows and Combs 1968; Vlasenko 1969; Bedell 1971; Sanders et al. 1972; Hoffman 1974; Conrad et al. 1975; Bullock and Stuckey 1977). Dupree (1981) has prepared an excellent review of the various methods of water disinfection for disease control. Several applications are listed below:

1. Disinfection can be used to permit the use of certain otherwise undesirable water supplies when alternate sources are unavailable or when fish pathogens are unavoidably present in the water supply. This reduces the risk of infecting fish or eggs in the culture station and helps avoid serious epizootics although disease may not be eliminated entirely (Vlasenko 1969; Sanders et al. 1972; Hoffman 1974). Signficant results may be achieved without disinfecting the entire hatchery water supply. In the case of IPN for example, disinfection of water used during early rearing will help prevent exposure to the pathogen until the fish are past the susceptible stage.
2. Disinfection can reduce the incidence of disease transmission between units if water must be reused (Conrad et al. 1975; Bullock and Stuckey 1977). If water is reused within the same unit, disinfection can help reduce the numbers of pathogens present in the system.
3. Disinfection can also be used to prevent dissemination of pathogens in effluents from fish culture operations. Disinfection (if not sterilization) would be valuable at a fish disease research facility, at a quarantine station for imports, or when protection of native fish populations or

method of disinfection for eradication of emergency diseases. The GLFDCC recommends a two hour exposure to 200 mg/l total chlorine. This is an extremely high concentration and is designed with a 'one time only' application in mind. Because chlorine is extremely toxic to aquatic organisms, even at very low concentrations, it must be completely neutralized before re-use or discharge. Obviously, the high concentration recommended by the GLFDCC poses serious problems for routine use, particularly with large water volumes.

Bedell (1971) and Sanders et al. (1972) were able to control (but not eliminate) ceratomyxosis by treating the water supply with 1 to 5 mg/I residual chlorine. Lower concentrations may also be equally effective in controlling other disease agents and the available data (Table 2) indicate this is the case. However, destruction of *Myxosoma cerebralis* spores requires exceptionally high chlorine concentrations for prolonged exposures (Hoffman and Putz 1969). Therefore, whirling disease control may not be practical by chlorination. For water supplies and reuse systems, residual chlorine concentrations of at least 1.0 mg/l for 10 minutes should effectively control most viral and bacterial fish pathogens. However, there are conflicting data on the efficacy of chlorine against IPN virus and caution is advised if this is the target pathogen.

OZONATION

Ozone gas (triatomic oxygen -0_3) has been used to disinfect fish culture water (Conrad et al. 1975; Spotte 1979) and the efficacy of ozone against certain fish pathogens has been determined (Table 3). Ozone, delivered at the rate of 90 mg/l with a minimum 20 min exposure time, should effectively control most bacterial and viral fish pathogens in water supplies and reuse systems although this level may not eliminate 100% of all pathogens. No data are available on control of *M. cerebralis* or C. *shasta* by ozonation.

Like chlorine, ozone is toxic to aquatic organisms. Furthermore, a break-down product of 0, is 0, and oxygen supersaturation may also occur. Ozonated water must be aerated in a holding tank prior to use or discharge.

An important consideration with ozonation is the energy inefficiency of current ozone generators (Spotte 1979). Therefore, ozonation is not a practical disinfection method for large water volumes.

ULTRAVIOLET (UV) IRRADIATION

UV light at wave lengths of about 254 nm has been shown to be an effective disinfectant for fish culture waters (Burrows and Combs 1968; Vlasenko 1969; Hoffman 1974; Spotte 1979). Hoffman (1974) reviewed the use

other fish culture facilities in/on downstream waters is desirable. This application is discussed below under **"Quarantine Facilities".**

Disinfection of effluent water is important because fish pathogens may reach high densities in the water. Desautels and MacKelvie (1975) detected significant levels of IPN virus in water taken from rearing troughs during an epizootic. *Aeromonas salmonicida* densities of 4,000 to 120,000 bacteria per ml were reported in the effluent from a tank of infected brown trout (Bullock and Stuckey 1977). Furthermore, some infectious pathogens can survive for a considerable time in the absence of a suitable host (Table 1). These high densities and long survival times suggest that the spread of pathogens from culture facilities can be significant.

METHODS OF DISINFECTION

The disinfection method(s) used must meet several criteria, including:

1. The method must not alter the physical-chemical properties of the water.
2. Treatment chemicals or treatment by-products must not be harmful to fish or other aquatic life or must be easily rendered safe. (The treatment must not damage any biofilters that may be in use in a re-circulating system.)
3. All equipment should be adaptable to flow through situations with minimal use of electrical or other forms of energy.
4. Every effort must be made to minimize the chance of a pathogen's escape by making the system as fail-safe as possible.
5. Most importantly, the method must be effective in eliminating the pathogenic organisms that are of concern in a particular situation.

Three methods meet some or all of these criteria. They are ozonation, ultra-violet (UV) irradiation, and chlorination. This report will not discuss designs for disinfection equipment. However, one important design feature must be used with each disinfection method. All three procedures are most effective and efficient following filtration of the water being treated. Burrows and Combs (1968) recommended UV irradiation following filtration through rapid flow sand filters to control diseases in closed or semi-closed systems. Bullock and Stuckey (1977) also found that filtration improved the effectiveness of UV radiation in destroying Gram-negative fish pathogens. Sanders et al. (1972) used a sand or Microfloc filter followed by chlorination or UV irradiation to control ceratomyxosis.

Filtration reduces the particulate organic material usually present in surface waters, re-circulation waters, and hatchery effluents. Organic material increases the amount of chlorine and ozone required to destroy pathogenic organisms. It also reduces the ability of UV radiation to penetrate the water being treated, thereby reducing its effectiveness.

Table 1. Survival time of several fish pathogens in water

Pathogen	Survival Time	Temp.	Remarks	Reference
IPN virus	24 wks.	4°C	residual infectivity remained	Desautels and MacKelvie (1975)
IPN virus	10 days	4°C		Tu, et al. (1975)
IPN virus	8 wks.	10°C	no loss of titer in hard or soft water	Wedemeyer, et al. (1978)
II-IN virus	7 wks.	10°C	hard or soft water	" "
Aeromonas salmonicida	2 wks.	20°C	soft lake water	Wedemeyer and Nelson (1977)
" "	2 wks.	20°C	hard lake water	" "
Yersinia ruckeri	20 days	20°C	survived over 20 days in hard and soft lake water	" "

Further discussions of factors affecting the use of disinfectants can be found in Dychdala (1968) for chlorination; in Hoffman (1974) and Spotte (1979) for ultraviolet irradiation; and in Farooq et al. (1977a, 1977b) and Spotte (1979) for ozonation.

There are no water disinfection standards for fish culture use. The remander of this report will review the available literature on the effectiveness of each method of disinfection in controlling fish pathogens and will recommend levels for control procedures.

CHLORINATION

Chlorination (the use of active chlorine compounds or liquids or gaseous chlorine) is used widely as a method of disinfection. Chlorination is recommended by the Great Lakes Fish Disease Control Committee (GLFDCC) as the

Table 2. Chlorine concentration and exposure time required to kill or inactivate certain fish pathogen

Pathogen	Concentration	Time	Water Quality	Reference
IPN virus	25 mg/l	30 min		Desautels and MacKelvie (1975)
" "	0.2 mg/l	10 min.	soft lake water(a)	Wedemeyer, et al (1978)
" "	0.7 mg/l	2min	hard lake water(b)	" "
IHN virus	0.5 mg/l residual	10 min	" "	" "
" "	" "	5 min	soft lake water	" "
" "	1.0 mg/l	30 sec.	hard lake water	" "
Aeromonas salmonicida	0.1 0.2 mg/l	30-60 sec.	hard or soft water	Wedemeyer and Nelson (1977)
Yersinia ruckeri	0.1 mg/l	2min	" "	" "
Ceratomyxa shas ta	uncertain (0.3 - 1.5 mg/l)	100 min.	lake water	Bedell (1971)
" "	2.2-5.3 mg/l residual		surface water	Sanders, et al. (1972)
Myxosoma cerebralis	1600 mg/l	24 hr.		Hoffman and Putz (1969)
	200 mg/l(c)	" "		" "

a) - Water hardness of 30 mg/l as $CaCO_3$; 10°C
b) - Water hardness of 120 mg/l as $CaCo_3$; 10°C
c) - Not proven 100% effective

Table 3. Ozone delivery rates and exposure times required to kill or inactivate certain fish pathogens

Pathogen	Delivery Rate (a)	Exposure Time	Water Quality	Reference
Aeromonas salmonicida	90 mg/h/l	5 min	hard lake water (b)	Wedemeyer and Nelson (1977)
" "	" "	15 min	soft lake water (c)	"
" "	*20* mg/h/l	30 min	hard or soft	" "
Yersinia ruckeri	90 mg/h/l	10 min	" "	" "
	20 Mg/h/l	25 min	" "	" "
IPN virus	90 mg/h/l	10 min	hard lake water	Wedemeyer et al (1978)
" "	" "	*30 sec*	soft lake water	" "
IHN virus	*70* mg/h/l	10 min	hard and soft	" "

a) - rate of ozone delivery, not the residual ozone concentration
b) - water hardness of 120 mg/L as $CaCO_3$; 20°C (= 20°C)
c) - water hardness of 30 mg/L as $CaCO_3$; 20°C

of UV and its effectiveness against certain fish pathogens. Vlasenko (1969) reported on the effectiveness of UV against *Saprolegnia* and several protozoan parasites. Bullock and Stuckey (1977) found that a UV dosage of 13,100 uW/sec/cmz killed 99.99-100% of several species of Gram-negative bacterial pathogens. However, even 21,000 to 24,000 $uW/sec/cm^2$ did not consistently kill 100% of the test organisms (Bullock and Stuckey 1977). UV irradiation at 13,100 uW/ sec/cm^2 prevented the transmission of *A. salmonicida* from a tank of infected brown trout to Atlantic salmon fingerlings over a four week period (Bullock and Stuckey 1977).

Few data are available on the use of UV for the inactivation of fish pathogenic viruses. In a static system, MacKelvie and Desautels (1975) reported that 2000 uW/cm^2 reduced the concentration of infective IPNV particles over a six minute period but did not inactivate all viruses even after 15 min. The researchers

concluded that UV radiation could not effectively eliminate IPN virus from the water (MacKelvie and Desautels 1975).

Spotte (1979) recommended UV radiation at 35,100 uW/sec/cm2 for disinfection in re-use systems. However, 13100 uW/sec/cm2 may be sufficient to effect control of bacterial pathogens. In the case of protozoan parasites, such as Ichthyophthirius, 90,000 to 1,700,000 uW/sec/cm2 may be required for effective control (Vlasenko 1969). Many of the "effective" doses reported in the literature do not effect 100% kill consistently. However, 90-99% may be adequate for disease control in some circumstances. UV levels effective against viral fish pathogens are not known.

Ultraviolet systems require regular cleaning of the bulbs and annual bulb replacement. However, the systems are readily adapted to fish culture operations, no toxic products are produced, and the level of radiation can be controlled more easily than levels of chlorine or ozone.

UV irradiation appears to be the method of choice for disinfection of water supplies and reuse systems. Levels of at least 13,100 uW/sec/cm2 should be used to control bacterial diseases and increased to 35,000 uW/sec/cm2 if ***M. cerebralis*** or C. ***Shasta*** are of concern. Even higher levels are required to control other protozoan parasites. The final choice of disinfectant will depend, however, on such considerations as initial capital costs, annual maintenance costs, energy requirements and the pathogen(s) to be controlled.

QUARANTINE FACILITIES

The principle of quarantine facilities is to protect against the introduction of a new pathogen to an area (i.e. fish hatchery, watershed, political jurisdiction) through the introduction of a new stock of fish that may be infected. Quarantine facilities must be separated, either by distance or physical barrier, from other fish culture activities and all effluent water must be disinfected. The water supply sanitation procedures discussed above can be applied.

Inactivation of 90-99% of pathogens may produce effective results in sanitation of water supplies and re-use systems. However, in the case of pathogen containment and quarantine facilities, 99% or even 99.99% is not acceptable. For this reason, ozonation and ultraviolet irradiation are not recommended. Chlorination at 200 mg/I total chlorine for two hours is preferred. However, some facilities have had good results using lower chlorine concentrations, particularly if a longer exposure time is used. These levels are only recommended for small water volumes as use on large volumes of water could represent a serious environmental hazard.

Quarantine failities for fish culture are not in use outside of research facilities. However, systems are being developed in Alaska, U.S.A. and the Maritime Provinces of Canada. Suggested contacts for further information are:

1. Principal Pathologist
   Fish Pathology Section
   Alaska Department of Fish and Game
   333 Raspberry Rd.
   Anchorage, AK 99502

2. Connaught Research Institute
1755 Steeles Ave. W.
Wiiowdale, Ontario M2N 5T8

3. Fish Disease Laboratory
Department of Microbiology
Oregon State University
Corvallis, OR 97331

4. Fish Health Unit-Scotia-Fundy Region
Dept. of Fisheries and Oceans
PO. Box 550
Halifax, Nova Scotia B3J 2S7

5. National Fish Health Research Laboratory
Route 3, Box 700
Keameysville, WV 25430

A word of caution is in order. Although the principles behind the operation of quarantine facilities suggest that the potential for success is present, there has been insufficient experience to fully assess their potential.

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