Bacterial and viral diseases create serious problems in semi-intensive and intensive aquaculture. Use of surface water in flow-through systems represents a risk of contamination by introducing waterborne fish pathogenic microorganisms. Such contamination results in heavy losses in aquaculture worldwide, and has also limited the progress in commercial farming of new aquacultural species. Some commercial operations may be required to disinfect their discharge waters before release into the aquatic environment. Of serious concern are dependable means of controlling pathogens present in the inlet water. Disinfection by ozonation or UV-irradiation are two methods often applied in aquaculture. It is important to distinguish between disinfection of makeup waters (low organic loads) and recirculating aquaculture system (RAS) waters. Both applications are discussed in this section. Disinfection by ozonation and UV-irradiation are also used in other aquacultural applications, e.g., reducing or eliminating potential pathogens associated with live prey such as rotifers in marine larval production systems, and surface disinfection of fish eggs. Other methods of disinfection are also discussed at the end of this section.
Ozonation & Water Quality

- Used to improve water quality in ultra-intensive recirculating production systems.
  - Ozone can produce excellent water quality in recirculating systems without resorting to high daily water exchange rates.
  - Ozone can reduce fish disease problems.

Ozone has seen wide use in aquaculture because it has a rapid reaction rate, produces few harmful reaction by-products in freshwater and oxygen is produced as a reaction end-product.
Ozonation: +/-

- Advantages:
  - rapid reaction rate,
    - dissolved ozone half-life only 0-15 sec (Bullock et al., 1997);
  - few harmful reaction by-products in freshwater;
  - oxygen is produced as a reaction end-product.

- Disadvantages:
  - ozone is dangerous to humans and fish.

Ozone is an extremely reactive oxidant and a very effective bactericide and viricide. Ozone can also be used to achieve water quality improvements by microflocculating fine particulate matter (making particles that are easier to settle or filter) and oxidizing nonbiodegradable organic molecules (creating smaller and more biodegradable molecules), nitrite, and refractory organic molecules (reducing water color).
Ozone Supports Water Treatment

- directly oxidizes NO$_2^-$ to NO$_3^-$;
- helps remove color & dissolved organic matter:
  - breaks non-biodegradable compounds into smaller & more biodegradable compounds;
- helps remove dissolved & fine particulate matter
  - precipitates dissolved organic molecules,
  - micro-flocculates fine particulate matter,
  - improving solids removal by settling, filtration, or flotation.
Ozone Can Reduce Fish Disease

- Ozone is also added to recirculating systems to reduce fish disease, by:
  - improving water quality and reducing fish stress
  - disinfecting the water
    - large reductions in micro-organisms are possible, but
      - ozone’s rapid reaction with nitrite and organic matter limit C*;
      - requires much greater ozone doses than required for water quality control;
      - disinfection not commonly achieved in recirculating systems.
Ozone oxidation can kill microorganisms, but requires maintaining a certain dissolved ozone concentration in the water for a given contact time. Disinfecting efficiency depends upon the product of the ozone residual concentration and its contact time. An ozone contact vessel provides the time necessary for the ozone residual to react with and inactivate pathogenic microorganisms. Disinfecting waters may require maintaining a residual ozone concentration of 0.1–2.0 mg/L in a plug-flow type contact vessel for periods of 1–30 min, depending upon the target microorganism (Wedemeyer, 1996). In commercial aquaculture applications, it is extremely difficult to maintain residual concentrations above 1 mg/L; above 2 mg/L is almost impossible with conventionally available equipment.
Ozone Doses for Disinfection

- Must maintain a residual concentration (C) for a given time (t):

<table>
<thead>
<tr>
<th>Pathogen</th>
<th>C<em>t, mg</em>min/L</th>
</tr>
</thead>
<tbody>
<tr>
<td>ISAV</td>
<td>0.3</td>
</tr>
<tr>
<td>Aeromonas salmonicida</td>
<td>1.6</td>
</tr>
<tr>
<td>Yersinia ruckeri</td>
<td>0.45-0.6</td>
</tr>
<tr>
<td>Flavobacterium sp.</td>
<td>2.8</td>
</tr>
<tr>
<td>Flexibacter sp.</td>
<td>1.6</td>
</tr>
<tr>
<td>Streptococcus sp.</td>
<td>0.015</td>
</tr>
<tr>
<td>Vibrio salmonicidia</td>
<td>0.45-0.6</td>
</tr>
</tbody>
</table>

In general, bacteria and viruses pathogenic to salmonids are highly sensitive to residual ozone in water. Dose-response estimates of this sensitivity are fairly precise in demand-free water (inorganic buffers, distilled water), indicating 99.9% inactivation or more in the 0.01–0.10 mg/L residual concentration range. In batch experiments with natural water exerting an ozone demand, residual concentration tends to drop rapidly, making reliable dose-response estimates more complicated. As a rule, higher residual concentrations, i.e., 0.1–0.2 mg/L in natural sea-, brackish-, and freshwater, and 0.3–0.4 mg/L in fish farm effluents, seem necessary to obtain the legally required inactivation level.
Maintaining Ozone Residual

- Sometimes difficult to maintain ozone residual for a given contact time due to ozone demand of water.
  - dissolved ozone has a half-life of only 0-15 sec in recirc systems (Bullock et al., 1997);
  - Ozone demand of relatively clean surface water supplies can range from 2-10 mg/L!
  - Ozone demand of recirc water could be 20-100 mg/L (?????)!

In demand-free water, dissipation of ozone will still be observed due to the demand exerted by the added microorganisms. This demand will depend on type of organism, the preparation, and washing of the inoculum prior to supplementation, and the density of organisms in the ozonated suspension. In natural waters and in waters found within recirculating systems, additional ozone will be lost in reactions with organics and other compounds.
Ozone Dosing Rate

- Bullock et al. (1997); Summerfelt et al. (1997)
  - 0.025 kg O₃ per kg feed input
    - improved water quality and microscreen filter performance
    - reduced mortalities associated with Bacterial Gill Disease (BGD)
    - reduced chemical treatments required to control BGD
    - did not reduce bacteria counts by even 1 log₁₀
  - 0.036-0.039 kg O₃ per kg feed input
    - same type and magnitude of benefits of lower ozone dose
    - much more likely to kill fish

According to ozone demand tests on a high quality trout stream water that is being ozone disinfected at the US Fish and Wildlife Service Lamar National Fish Hatchery (Lamar, Pennsylvania), an ozone concentration of 2–4 mg/L must be transferred to maintain a 0.2 mg/L ozone residual concentration after 10 minutes (Steven Summerfelt, unpublished data). Cryer (1992) reported similar ozone demand results in tests on surface water supplies that were being disinfected at US Fish and Wildlife Service salmonid hatcheries in North America. All of the surface water supplies examined in these studies exhibit relatively high water quality with low concentrations of oxidizable organic material, iron, and manganese (Cryer, 1992; Steven Summerfelt, unpublished data), yet the ozone demand created still reduced the half-life of ozone to less than a few minutes. In comparison, the half-life of ozone dissolved in pure water at 20°C is 165 minutes (Rice et al. 1981).
The ozone demand of water within RAS, which contains much higher levels of organic material and nitrite, creates a short ozone half-life, e.g., less 15 seconds, and makes maintaining an ozone residual difficult (Bullock et al. 1997). For this reason, it is difficult to add enough ozone to achieve microbial inactivation in recirculating systems. In recirculation systems ozone is most often applied at doses that promote water quality improvement (Brazil, 1996; Bullock et al. 1997; Summerfelt and Hochheimer 1997; Summerfelt et al. 1997). Using ozone in recirculating systems can reduce fish disease simply by improving water quality, which reduces or eliminates environmental sources of stress (Bullock et al. 1997). These studies, as well as experience with ozone application at numerous commercial recirculating systems, indicates that both water quality and fish health can be improved by adding approximately 13–24 g ozone for every 1.0 kilogram of feed fed to a recirculating system.
Ozone & Water Quality

- Effect of O₃ on culture tank influent water quality:

<table>
<thead>
<tr>
<th></th>
<th>TSS (mg/L)</th>
<th>COD (mg/L)</th>
<th>DOC (mg/L)</th>
<th>Color (Pt-Co)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>6.3±1.1</td>
<td>44±4</td>
<td>7.1±0.4</td>
<td>17.7±1.2</td>
</tr>
<tr>
<td>Ozone trial 1</td>
<td>4.0±0.6</td>
<td>26±2</td>
<td>NA</td>
<td>5.3±0.9</td>
</tr>
<tr>
<td>Ozone trial 2</td>
<td>2.9±0.6</td>
<td>26±6</td>
<td>6.3±0.3</td>
<td>2.9±0.4</td>
</tr>
<tr>
<td>Ozone trial 3</td>
<td>5.6±0.5</td>
<td>37±2</td>
<td>6.0±0.3</td>
<td>2.1±0.5</td>
</tr>
<tr>
<td>Ozone trial 4</td>
<td>3.1±0.3</td>
<td>24±2</td>
<td>5.5±0.2</td>
<td>2.1±0.4</td>
</tr>
</tbody>
</table>

(Summerfelt et al., 1997)
Ozone & Microscreen Filtration

- Solids removal across the microscreen filter:
  - no ozone: 24% of feed fed removed
  - ozone: 33% of feed fed removed
- Total solids production in system was ~40% of feed fed.
- Increased solid removal was probably due to ozone:
  - precipitating dissolved organic molecules
  - microflocculating fine particulates

(Summerfelt et al., 1997)

Ozonation may enhance fine solids removal by changing particle size rather than separating particles from water. As an unstable reactive gas, ozone splits large organics into smaller biodegradable materials that can be more easily removed by heterotrophic bacteria. Conversely, ozone can polymerize metastable organics leading to enmeshment, direct precipitation, bridging, or adsorption (Reckhow et al. 1986). Ozone has been used sometimes with mixed success in a variety of aquaculture systems to remove color and turbidity (Colberg and Lingg, 1978; Williams et al. 1982; Paller and Lewis, 1988; Brazil, 1996; Summerfelt et al. 1997). Effects of ozonation upon particle size change in recirculating systems are still not clearly defined. The function of ozone is complicated; both qualitative and quantitative impacts of ozonation may be specific to a given system (Grasso and Weber, 1988). There are also concerns that even low ozone residuals may cause gill adhesions and mortality in fish exposed to freshly ozonated water (Rosenlund, 1975).
Ozone & Microscreen Filtration

- Microscreen filter improvements with ozone:
  - TSS removal increased 33%
  - Wash cycles reduced 35%
  - Sludge water production reduced 53%
  - Sludge water settled sludge volume reduced 77%

(Summerfelt et al., 1997)
Ozone & Solids Removal

- Also improves solids removal via
  - Foam fractionation
    - Sander & Rosenthal (1975)
    - Otte and Rosenthal (1979)
    - Williams et al. (1982)
  - Settling
    - Wilczak et al. (1992)
    - Reuter and Johnson (1995)
Ammonia and Ozone

- In freshwater systems:
  - Ozone does not oxidize significant NH$_3$ to NO$_3$ until pH > 9
Ammonia and Ozone

- In saltwater systems (if sufficient bromide is present), oxygen will react with bromide to produce hypobromous acid and this will react with ammonia to produce nitrogen gas while producing $\text{H}^+$ that consumes alkalinity.

$$\text{O}_3 + \text{Br}^- + \text{H}^+ \rightarrow \text{HOBr} + \text{O}_2$$

$$3\text{HOBr} + 2\text{NH}_3 \rightarrow \text{N}_2 + 3\text{Br}^- + 3\text{H}^+ + 3\text{H}_2\text{O}$$

$$\text{HCO}_3^- \rightarrow \text{CO}_2 + \text{H}_2\text{O}$$

(Haag and Hoigne, 1984)
Nitrite and Ozone

- Ozone stoichiometrically oxidizes nitrite to nitrate.

<table>
<thead>
<tr>
<th></th>
<th>NO2-N Influent (mg/L)</th>
<th>NO2-N Uptake Rate (kg/day)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0.28±0.01</td>
<td>0.76±0.02</td>
</tr>
<tr>
<td>Ozone trial 1</td>
<td>0.13±0.01</td>
<td>0.65±0.02</td>
</tr>
<tr>
<td>Ozone trial 2</td>
<td>0.15±0.01</td>
<td>0.51±0.02</td>
</tr>
<tr>
<td>Ozone trial 3</td>
<td>0.11±0.01</td>
<td>0.47±0.02</td>
</tr>
<tr>
<td>Ozone trial 4</td>
<td>0.10±0.01</td>
<td>0.41±0.02</td>
</tr>
</tbody>
</table>

(unpublished data from same experiment as Summerfelt et al., 1997)
Nitrite and Ozone

- Ozone oxidized nitrite to nitrate:
  - reduced nitrite concentration in water
  - reduced nitrite loading on biofilter
  - caused population of nitratifiers to decrease
  - reduced total nitrite removal capacity of biofilter
Nitrite and Ozone

- If ozonation is interrupted:
  - biofilter cannot remove all nitrite produced
  - rapid nitrite accumulation occurs
  - fish health is compromised
  - several weeks may be required for biofilter to adapt to additional nitrite loading
Ozone must be generated on-site. The most efficient method is by the electric corona discharge technique, which involves the passage of oxygen gas, or air, across a gap of narrowly spaced electrodes under high voltage.
Ozone Generation

- Corona discharge energy dissociates $O_2$ into oxygen radicals and produces $O_3$:

$$O_2 + \text{energy} \rightarrow O + O$$

$$O + O_2 \rightarrow O_3$$

- Ozone is produced when oxygen gas is exposed to electric current arcing between two parallel electrode surfaces.

According to Masschelein (1998), effective ozone generation depends upon the composition of the feed gas, e.g., feed gas impurities, particulates, moisture, and pressure of the feed gas, efficiency of dielectric cooling, characteristics of the electrical current, concentration of the ozone produced, and dielectric design. Protecting the dielectrics within the corona discharge cells requires a feed gas supply that is dry, free from particulate matter and coalescible oil mists, and contains less than 15 ppm hydrocarbons (Dimitriou 1990; Masschelein, 1998).
Ozone Generation

- Must be generated onsite.
- Ozone generation requires 2-3 times less energy using enriched oxygen gas rather than air.
- Most economical ozone generation method takes advantage of the oxygen feed gas already required to increase carry capacity.

Also, some ozone generators may require some nitrogen gas impurity (>0.5% nitrogen) within the oxygen feed gas in order to achieve maximum ozone production efficiency. To meet this level of nitrogen contamination, some liquid oxygen supplies may require adding a small quantity of nitrogen to the feed gas before it enters the ozone generator.
Purified oxygen is already used to maximize carrying capacity within many intensive aquaculture systems. Corona discharge generators using purified oxygen feed gas require about 10 kWh of electricity to produce 1.0 kg of ozone (Masschelein, 1998). However, ozone production within an air feed gas is 2–3 times less energy efficient than using purified oxygen feed gas (Masschelein, 1998). Also, generating ozone in oxygen feed gas can produce a 10–15% (by weight) concentration of ozone, which nearly doubles the concentration of ozone that can be generated using air as the feed gas. The relatively high concentrations of ozone can be generated to reduce the overall mass of oxygen required to supply ozone. Yet, it is less energetically efficient to produce ozone concentrations of 10–15% (by weight) than to produce ozone concentrations of 4–6% (Carlins and Clark, 1982). Taking all of this into account, ozone production can be optimized according to the demands of the aquaculture system and economic considerations of feed gas cost and energy usage.
Ozone Generation

- Air cooled ozone generator

(Target Marine Hatchery)
Ozone Generation

- Water cooled ozone generator.

(Freshwater Institute)
Ozone Generation

- Safety interlocks shut-off generators in case of problems.

(Freshwater Institute)
Ozone generator fire also burned down Virginia Tech’s aquaculture laboratory in mid-1990’s.
Ozone Generation

- If it becomes necessary, cleaning dielectrics can be a hassle.

(Freshwater Institute)
Ozone Gas Transport

- Use stainless steel, teflon, viton, or kynar.

Ozone is an extremely corrosive material when used in any water application. Materials that ozonated water will come in contact with must be selected with appropriate resistance properties.
Ozone Gas Transport

- Use stainless steel, teflon, viton, or kynar.

(Freshwater Institute) (Nutreco’s Big Tree Creek Hatchery)

9th Annual Recirculating Aquaculture Systems Short Course
Ozone Monitoring in Gas Phase

(Freshwater Institute)

9th Annual Recirculating Aquaculture Systems Short Course
Ozone Transfer

- When adding ozone to a recirculating system, suggest taking advantage of the oxygen gas stream and oxygen transfer unit already used to increase the system’s carrying capacity.

Ozone is generated within either air or oxygen feed gas and this ozone/oxygen gas flow must be transferred into water for microbiological inactivation or other oxidative purpose. The ozone gas flow can be co-transferred into the water using any of the typical oxygen transfer devices (Summerfelt and Hochheimer, 1997), which have been discussed in Chapter 8 of this book. Effective transfer of ozone gas into water is important because the cost of producing ozone is not insignificant, especially if the ozone is carried within purified oxygen feed gas that was either purchased or produced on site.

The rate of ozone transfer and the subsequent rate of ozone decomposition depend upon the efficiency of the contacting system used and the rates that ozone reacts with constituents within the water. The rate that ozone reacts depends on the type and concentration of constituents within the water. Rapid reaction with oxidizable inorganics and organics will maintain a low apparent equilibrium concentration of ozone within the liquid film and increase the rate of ozone transfer. The driving force for ozone transfer is maximized when the ozone absorbed is rapidly consumed by reaction with constituents within water. In fact, when ozone reacts very fast, ozone decomposes at the gas surface and no molecular ozone is transferred into the water (Bablon et al. 1991).
Ozone transfer into RAS is sometimes accomplished using the same gas transfer unit that is used for oxygen supplementation. This can be done if the transfer unit is fabricated from ozone resistant material. In these situations, adding ozone to a recirculating system that is already using purified oxygen only requires installation of an ozone generator and the accompanying ozone distribution, monitoring, and control mechanisms. All of the other necessary equipment (oxygen supply and distribution system, gas transfer units, and control mechanisms) would already be in place.
Ozone Transfer in Recirc Systems

- Ozone added within LHO at WV Aqua LLC (Man, WV)

(system designed by PRAqua Tech.)

Justifications for ozone addition in the LHO with the oxygen feed gas used to increase carrying capacity:

1. Adding ozone in the LHO takes advantage of the oxygen gas stream and transfer unit already installed to supplement oxygen levels in the culture tank, which saves the fish farm both capital and operating dollars. If you add ozone at a different location in the recirculating system you will need another oxygen gas supply and another gas transfer unit (which are both new capital and operating costs).
(2) Adding ozone in the LHO before the culture tank allows some flocculation of the oxidized organic particles to occur in the fish culture tank (HRT = 30-60 min) before the water reaches the LHO. The flocculation will occur after the ozone has oxidized the surfaces of the organic particles, which in simplistic terms makes the particles sticky.
Ozone Transfer

- O₂ & O₃ transfer within low head oxygenators (LHOs)
  - Ozone transfer efficiency can approach 100% in a good oxygen transfer unit, because:
    - Ozone is 13 times more soluble than oxygen (Henry’s Law);
    - Gas residence time in LHO can be up to 45 min;
    - Nitrite & TOC rapidly react with ozone.

(Summerfelt & Hochheimer, 1997)

(3) Ozone demand of freshwater in recirc systems is so high that you can dose 13-20 g ozone per kg fish feed without much threat of ozone break through within the culture tank, as long as you continue to feed the fish (do not use demand feeders). I do suggest using a quality ORP controller to alert of elevated ORP levels that would indicate potential for ozone break-through in the culture tanks.
Ozone transfer units that have a continuous liquid-phase, i.e., units that disperse gas bubbles within a liquid, -- such as U-tubes, Speece cones, above, aspirators, bubble diffusers, and enclosed mechanical surface or subsurface mixers -- provide both ozone transfer and some reaction time. Ozone transfer units that have a continuous gas-phase, i.e., units that disperse liquid drops and films within a gas, such as spray columns, packed columns, and multi-stage low head oxygenators, provide efficient transfer but very little time for reaction. Continuous gas-phase transfer units are best suited for use in situations that normally require the transfer of the maximum amount of ozone in the shortest time for economical fixed and variable costs. On the other hand, continuous liquid-phase transfer units are usually selected for situations where reaction is rate limiting and an ozone residual must be maintained for a specific length of time.
Ozone transfer within continuous gas-phase units is not as common as within continuous liquid phase units (Bellamy et al. 1991). When ozone transfer has been reported within continuous gas-phase units, they are mostly packed columns. However, continuous gas-phase units can be designed to efficiently transfer ozone within relatively smaller vessels. Ozone transfer efficiency was 100% in the LHO (Low Head Oxygenator) units evaluated in the recirculating system at The Freshwater Institute. In this system, complete ozone transfer occurred because ozone is 13 times more soluble than oxygen in water according to Henry’s law; short circuiting in the gas phase within the LHO was prevented by breaking the chamber into eight separate compartments; gas residence times within the LHO chambers were about 45 min; and, there was nitrite and dissolved and suspended organic material in the water that rapidly reacted with the dissolved ozone.
Most ozone contactors rely on continuous liquid-phase units that bubble ozone into the liquid (Bellamy et al. 1991). High column bubble diffusers are frequently used for aquacultural applications and can achieve more than 85% ozone transfer to the liquid phase. These units are particularly well suited to situations where reaction is rate limiting and an ozone residual must be maintained for a specific length of time, such as during disinfection. Speece cones and U-tubes are also being used to efficiently and rapidly transfer ozone/oxygen feed gas within RAS, where oxidation of nitrite and organic matter are the primary goals of ozonation (not disinfection).
Ozone oxidation can kill microorganisms, but requires maintaining a certain dissolved ozone concentration in the water for a given contact time. Disinfecting efficiency depends upon the product of the ozone residual concentration and its contact time. An ozone contact vessel provides the time necessary for the ozone residual to react with and inactivate pathogenic microorganisms. Disinfecting waters may require maintaining a residual ozone concentration of 0.1–2.0 mg/L in a plug-flow type contact vessel for periods of 1–30 min, depending upon the target microorganism. In commercial aquaculture applications, it is extremely difficult to maintain residual concentrations above 1 mg/L; above 2 mg/L is almost impossible with conventionally available equipment. An example system that provides ozone contact time in a two reactor sequence vessel with residual removal is illustrated above.
Methods to Remove Dissolved Ozone

- Provide extended contact time & let ozone react away;
- Aerate to strip ozone into air;
  - G:L of 10:1 to 20:1
- Expose to high intensity UV light:
  - Wavelength of 250-260 nm;
  - 60-120 mW-s/cm² to remove 0.5 mg/L.
- React with hydrogen peroxide;
- Pass through an activated carbon bed or biofilter.

Creating an adequate level of residual ozone at the end of the contact chamber to ensure kill-off of bacteria will also necessitate that this same ozone be removed prior to the water reaching the aquatic organisms. Ozone residuals can be lethal to fish at ozone concentrations as low as 0.01 mg/L, but the actual concentration depends upon species and life stage. Due to the acute toxicity of residual ozone to aquatic animals, a de-ozonation unit has to be included. In many cases, residuals are eliminated by water retention within tanks immediately after ozonation, or by applying small doses of a reducing agent, e.g., sodium thiosulphate. Dissolved ozone can also be stripped into air when passed through a forced-ventilation packed aeration column. Air stripping will also remove dissolved oxygen concentrations that are in excess of saturation, which may or may not be desirable. Dissolved ozone can also be destroyed by passing the water through a biofilter or bed activated carbon, reaction with low levels of hydrogen peroxide, or contact with high intensity UV-light (catalyzing the conversion of O₃ to O₂).
UV Disinfection

- DNA is denatured by UV electromagnetic radiation at wavelengths of 100-400 nm,
  - kills or inactivates microorganisms,
  - 255-265 nm are most effective destroying DNA & RNA and are most lethal wavelengths,
  - 280 nm wavelength denatures proteins and enzymes

- The quantity of energy transmitted at a wavelength of 254 nm is the standard used to estimate UV inactivation potential.

Natural and artificial UV light (wavelength of 190–400 nm) may damage microorganisms by directly and indirectly altering nucleic acids. Direct damage is due to absorption of irradiation by DNA with resulting formation of photoproducts. DNA absorbance is high in the UV-C range (190–280 nm), but falls more than three orders of magnitude in the UV-B range (280–320 nm), and is negligible in the UV-A band (320–400 nm) (Miller et al. 1999). The DNA-damaging effect of UV-C is utilized in bactericidal lamps. The low-pressure mercury-vapor lamp emits approximately 85% of its energy output as monochromatic light at a wavelength of 253.7 nm, which is within the optimum wavelength range of 250 to 270 nm for bactericidal effects. Solar UV-B irradiation is responsible for both direct and indirect DNA damage, while UV-A produces only indirect damage. Only direct DNA damage is repairable by photoreactivation.

The UV-C damage often results from dimerization of two pyrimidine molecules. Cyclobutane pyrimidine dimers and pyrimidine-pyrimidone (6–4) photoproducts are the two major classes of photolesions formed by direct DNA absorption of UV-C irradiation (Friedberg et al. 1995). Once the pyrimidine residues are covalently bound together, replication of the nucleic acid is blocked or results in mutant daughter cells unable to multiply (Stover et al. 1986). The moderate energy level of UV irradiation leaves no toxic residuals in the treated water. Although chemical compounds can be altered by the radiation (Gjessing and Kallqvist, 1991; Lund and Hongve, 1994), the UV doses used for disinfection are too low to generate significant amounts of photoproducts (Oliver and Carey, 1976; de Veer et al. 1994). This non-toxicity is of crucial importance when UV is the method of choice for influent disinfection in aquacultural facilities.
Achieving UV disinfection requires maintaining a minimum UV dose:

$$\text{UV dose} = (\text{UV intensity}) \cdot (\text{exposure time})$$

$$= (\text{mW/cm}^2) \cdot (\text{sec})$$

$$= \text{mW \cdot sec/cm}^2$$

10-30 second contact times are typical (White, 1992).

UV irradiation units are commonly installed in smolt farms for sea and freshwater disinfection, and are also used for bacteriological control in recirculation systems (Rosenthal, 1981). However, before the UV dose can even reach the target organism, it must be able to transmit through the water. UV applications in highly turbid water as is often the case in RAS will be totally ineffective since the transmission into the water column is very minor, thus killing almost no organisms. Therefore, the lowest expected UV transmittance of the process water should be established and used to predict how much UV intensity must be generated to transmit the desired UV dose through the water between the target organism and the light source.
UV Dose

- Actual UV dose applied to water flow depends on:
  - Water flowrate (Q) and operating volume within UV vessel;
  - Lamp intensity (including losses at quartz sleeve);
  - UV transmittance of water (% Transm.).

\[
\text{UV dose} = (\text{UV intensity}) \cdot (\text{exposure time}) \cdot (\text{transmittance factor})
\]

\[
\approx (\text{UV intensity}) \cdot \left(\frac{V_{\text{vessel}}}{Q}\right) \cdot a \cdot \exp(b \cdot \%\text{Transm})
\]

\[
= \# \text{ mW} \cdot \text{sec/cm}^2
\]
### UV Doses Required for Disinfection

- Dose to inactivate 99.9% of BACTERIA from Wedemeyer (1996) and Liltved (2001):

<table>
<thead>
<tr>
<th>Organism</th>
<th>mW-sec/cm²</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Aeromonas salmonicida</em></td>
<td>4</td>
</tr>
<tr>
<td><em>Aeromonas hydrophila</em></td>
<td>5</td>
</tr>
<tr>
<td><em>Vibrio anguillarum</em></td>
<td>4</td>
</tr>
<tr>
<td><em>Yersinia ruckeri</em></td>
<td>3</td>
</tr>
<tr>
<td><em>Pseudomonas fluorescens</em></td>
<td>5</td>
</tr>
</tbody>
</table>

Data on inactivation of fish pathogenic bacteria and viruses by UV irradiation are summarized in Tables 12.5 and 12.6 and above. The values show that UV doses of 2–6 mWs /cm² reduce the viable count of the studied species by 99.9% or more in laboratory batch experiments. However, under more realistic continuous flow experiments with particles present, considerably higher doses are required to obtain a high degree of inactivation (Bullock and Stuckey, 1977; Liltved and Cripps, 1999). Therefore, inactivation doses obtained in the laboratory should be used with caution in predicting dose requirements in actual situations.
### UV Doses Required for Disinfection

- **Dose to inactivate 99.9% of VIRUSES from Wedemeyer (1996) and Liltved (2001):**

<table>
<thead>
<tr>
<th>Virus</th>
<th>mW·sec/cm²</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>ISA</strong></td>
<td>4-10*</td>
</tr>
<tr>
<td><strong>IHN</strong></td>
<td>1-3</td>
</tr>
<tr>
<td><strong>IPN</strong></td>
<td>100-200</td>
</tr>
<tr>
<td><strong>Channel catfish virus</strong></td>
<td>2</td>
</tr>
<tr>
<td><strong>Herpesvirus salmonis</strong></td>
<td>2</td>
</tr>
<tr>
<td><strong>White spot syndrome baculovirus</strong></td>
<td>900*</td>
</tr>
</tbody>
</table>

*loss of infectivity

Studies by Torgersen (1998) indicate that the ISA virus is susceptible to UV light. Loss of infectivity was demonstrated when infected tissue homogenate was subjected to doses of 4–10 mWs cm-2. In contrast, the IPN virus, a non-occluded birnavirus responsible of infectious pancreas necrosis in Atlantic salmon, is UV resistant. A dose of 122 mWs cm-2 was required to obtain 99.9% reduction in virus titer in brackish water (Liltved et al. 1995). Doses in the same order of magnitude have been experienced in studies conducted by Japanese investigators (Sako and Sorimachi, 1985; Yoshimizu et al. 1986). In spite of the resistance of IPNV, UV irradiation has become the method of choice for disinfection of supply water in Norwegian smolt farms where the UV units are designed for bacterial inactivation. The IPNV will pass through these installations unaltered since the doses applied are far too low for inactivation. Due to the substantial losses from IPN outbreaks, preventive measures such as upgrading of existing UV units to IPNV inactivating capacity when using water from sources suspected to carry infective levels of IPNV should be considered. Such upgrading will require at least a 5-fold increase in UV dose compared with the present 25 mWs cm-2 requirement.
Even higher doses have been required to reduce the infectivity of the Asian shrimp baculoviruses to zero. The mid-gut glad necrosis virus (BMNV) and the WSBV were inactivated by doses of 410 and 900 mWs cm\textsuperscript{-2}, respectively (Momayama, 1989; Chang et al. 1998). Due to the extreme resistance of these viruses and the excessive water flows required, UV irradiation is not a feasible method for baculovirus inactivation in Asian grow-out shrimp farms. However, high efficiency UV units could be suitable for hatchery and nursery ponds with limited water use. There are other studies providing additional information on the UV levels needed to achieve disinfection (Hunter et al. 1998; Wedemeyer, 1996).
UV Doses Required for Disinfection

- Prefiltration through 50 μm screens can improve bacterial inactivation with UV by 3.0 log_{10} units.

  - Liltved and Cripps (1999)

Particle protection mechanism has been indicated for bacteria associated with Artemia fragments, due to lack of a dose dependent inactivation in the dose range of 10–22 mWs cm⁻². The results obtained suggest a possible transmission of fish pathogenic bacteria to land-based aquacultural installations, even if the influent water is disinfected by UV irradiation. It was further demonstrated that prefiltration improved bacterial removal. Mesh sizes of 50 μm resulted in more than 5 log_{10} removal efficiency, indicating that influent water to aquacultural systems should be filtered to remove crustaceal fragments and other particles capable of harboring bacteria before UV disinfection.
Absence of toxic residues and reaction products, low costs when treating high quality water, easy operation and maintenance, and minimal space requirements, are main advantages of UV irradiation. Fouling of the quartz sleeves must be dealt with on a regular basis, normally by mechanical or chemical cleaning. The major disadvantage of this method in aquacultural applications is the inefficiency against important fish pathogenic viruses. This might limit the use of UV irradiation in the future as virus control becomes a more important preventive measure in the fish and shrimp farming industry.
Types of UV Lamps

Mercury Lamps:

- Low pressure, low intensity (old technology)
- Low pressure, high intensity (newest technology)
- Medium pressure, high intensity (new technology)

UV light output strength continually deteriorates from the time you first turn on the machine. Assume in general a 3% per month decrease in output strength or 40% per year. UV output strength is also affected by air temperature. The UV output ratings are 100% at 100°F (38°C); at 32°F (0°C), the output strength is only 10% of the 100°F (38°C) rating. This problem can be addressed by using quartz sleeves that creates a warm air pocket around the UV bulb and this maintains output strength near its maximum potential. The quartz sleeve will reduce output by 5% just due to transmissivity of the sleeve. Bulb replacement in UV units can be of considerable expense, particularly when compared to other forms of sterilization and disinfection. As a rule of thumb, change UV bulbs at least once annually.
UV Systems Compared


<table>
<thead>
<tr>
<th></th>
<th>Low/Low</th>
<th>Low/High</th>
<th>Medium/High</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Lamp power</strong></td>
<td>15-70 W</td>
<td>120-260 W</td>
<td>4000 W</td>
</tr>
<tr>
<td><strong>Efficiency</strong></td>
<td>40%</td>
<td>35-40%</td>
<td>10-15%</td>
</tr>
<tr>
<td><strong>Lamp output @ 254 nm</strong></td>
<td>0.1-0.2 W/cm</td>
<td>0.5-1.0 W/cm</td>
<td>3 W/cm UVC tot</td>
</tr>
<tr>
<td><strong>Lamp temperature</strong></td>
<td>39°C</td>
<td>100°C</td>
<td>600°C</td>
</tr>
<tr>
<td><strong>Lamps needed for 0.8 m³/s flow</strong></td>
<td>~100</td>
<td>~30</td>
<td>~5</td>
</tr>
<tr>
<td><strong>Total power cons</strong></td>
<td>7.5 kW</td>
<td>7.8 kW</td>
<td>20 kW</td>
</tr>
</tbody>
</table>

9th Annual Recirculating Aquaculture Systems Short Course
Low-Pressure UV Output

(courtesy of Trojan Technologies)
Medium-Pressure UV Output

(courtesy of Trojan Technologies)
Medium-Pressure UV

- In addition to disinfection, medium-pressure UV can:
  - photo-oxidize organic carbon at 185 nm wavelengths,
    - produces hydroxy radicals (HO-) to break apart organic molecules
  - convert nitrate into nitrite if special quartz sleeve filtering is not applied.
Vessel Design

- Pressurized “Tube-and-Shell” design
  - Selected when water pressure needs to be conserved in a pipeline
  - Typically operates at > 0.2-1 m of water headloss
- Non-pressurized “Open Channel” Design
  - Selected in situations with little available water head
Horizontal Channel UV Filter

- Freshwater Institute’s Grow-out System

Courtesy PRAqua Technologies (BC)
9th Annual Recirculating Aquaculture Systems Short Course
Horizontal Channel UV Filter

- Courtesy of Trojan Technologies, Inc.
Vertical Channel UV Filters

- Three salmon smolt systems (~3000 gpm/system) at Nutreco’s Big Tree Creek Hatchery (BC)

(System designed by PRAqua Technologies)
Medium-Pressure UV Unit

- Tube-and-shell design

(courtesy of Trojan Technologies)
Medium-Pressure UV Unit

- Tube-and-shell design.

(courtesy of Ozonia)
Ozone Followed by UV Filtration

- Freshwater Institute’s Grow-out System.
Ozone Followed by UV Filtration

- Three salmon smolt systems (~3000 gpm/system) at Nutreco’s Big Tree Creek Hatchery (BC)

(system designed by PRAqua Technologies)

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